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Botany

1

Diversity of Arbuscular Mycorrhizal Fungi and their Symbiotic Association with *Ocimum* species

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Abstract –*Ocimum spp.* is an important medicinal and essential oil-bearing plants of India. The present study was conducted at Vinayak Vidnyan Mahavidyalaya, Nandgaon kh. to know the symbiotic association of Arbuscular mycorrhizal (AM) fungi with different species of *Ocimum.* Microscopic analysis of the soil samples has revealed that all recovered spores were found to belong to the genus *Glomus* and *Acaulospora species*. Out of which *Glomus aggregatum, Glomus fasciculatum, Glomus geosporum, Gomus leptotichum, Glomus fecundisporum, Glomus reticulatum* and *Acaulospora* spp. were identified and quantified. *Glomus reticulatum, Glomus fecundisporum* were predominantly present and associated with all the *Ocimum* species. Root colonization studies revealed a maximum colonization (62.8%) in *Ocimum sanctum* (Indian basil) and a minimum colonization (35.2%) in *Ocimum gratissimum*. The application of AM fungi may improve the growth and productivity of *Ocimum* species.

Keywords**:** AM Fungi, *Ocimum*, medicinal plants.

Introduction:-

Tulsi, the Holy basil is a perennial shrub belonging to family *Lamiaceae.* It has been known for its curative properties and has been utilized as antimycotoxic, analgestic, antibacterial, antihaemorrhagic, antioxidant properties and it is considered as a good rejuvenator (Ghosh, 1995). Around 80% of plant species on the earth are known to be associated with arbuscular mycorrhizal fungi (AMF) (Kivlin *et al*. 2015). The AM fungi is a symbiotic association which develop special structures called as arbuscles and vesicles. The arbuscules help in the transfer of nutrients from the soil into the root system (Divya, 2015). In view of this effect of VAM and soil microorganisms is important thrust area in plant growth and development especially in medicinal plants. Lot of studies has been proved that arbuscular mycorrhizal fungi improve plant growth through phosphorous nutrition. In addition to phosphorous they also help in the uptake of another nutrient element. Nutrient absorption by fungal symbionts is due to external hyphae of the fungus proliferating beyond the nutrient depletion zone and reaching the source of nutrient. The improved plant growth promoting substances, tolerance to drought, salinity and transplantation shock, resistance to soil-borne pathogen and synergetic interaction with other beneficial microorganism (Sandhya *et al*., 1989). Therefore, in future AM fungi inoculation is one of the promising tools for the conservation and sustainable maintenance of medicinal herbs. Thus, prompted with the above-mentioned facts we undertook present study to understand how native AM fungi play their role in association with medicinal plants so that their bio-fertilizing potential can be exploited accordingly. The objective of this work was to study the diversity of AM fungi associated with different species of *Ocimum* plant.

Materials and Methods: –

Study area

Nandgaon Khandeshwar (Latitude: 20.80677 and Longitude: 77.3645) is one of the taluka of the Amravati district. It is a region with sufficient forest area and eighty percent of the people depend on agriculture.

Sample collection -The area of the botanical garden which is situated in the college campus was selected for the study. Three plant samples, root samples, and corresponding rhizospheric soil samples were collected. Soil and feeder root samples were collected from the three species of *Ocimum* plant. For this purpose, soil around the plant was dug up to 10-15 cm deep and rhizosphere soil and feeder roots were collected separately in polythene bags and tagged properly. Soil and root samples were then taken to the laboratory for analysis. Screening of roots was done by wet screening and decantation methods of Gerdemann and Nicolson (1963). The suspension prepared was allowed to pass through the sieves of different measurements. Spores were examined under the compound microscope. The spore population was determined as the number of spores per 10 grams of dried soil. Spores were identified based on morphological features i.e. spore colour, size, shape, thickness of the wall, lamination, and attachment pattern of hypae as described in the manual of Schenck and Perez (1988). Phillip and Hayman (1970) method were followed to determine the percent of AMF colonization. The method given by Gaur and Adholeya (1994) was used for counting AMF spores (Photoplate I). All the slides showing characteristics and features of AMF along with the isolated spores were photographed by using Carl Zeiss inverted compound microscope.

Processing of Roots:

The preserved root samples were used for further analysis by the process given by Phillips and Hayman (1970). The AM percent root colonization was calculated by using the Grid line intersect method (Giovannetti and Mosse, 1980).

III. Results and Discussions-

The result depicted in Table No. 1 presented the percent root colonization and spore population. The result shows that all three species of *Ocimum* were colonized by AM fungi. However, spore population and percent root colonization varied from plant to plant. Maximum percent root colonization was found in *Ocimum sanctum* (86%) followed by *Ocimum gratissimum* (75%) and *Ocimum basilicum* (67%) respectively. Both vesicles and arbuscules were found in the root segments of different plants. Vesicles were almost common in all the test plants but their frequency was higher in *Ocimum sanctum.*

Table No.1 - Number of spores/100g soil and percent of root colonized in rhizosphere of *Ocimum species.*

S.N.	Name of the species	No. of spores/100g soil	% root colonization	Presence of hyphae
Sample no. 1	*Ocimum sanctum*	271	86%	++++
Sample no. 2	*Ocimum gratissimum*	264	75%	+++
Sample no 3	*Ocimum basilicum*	212	67%	+++

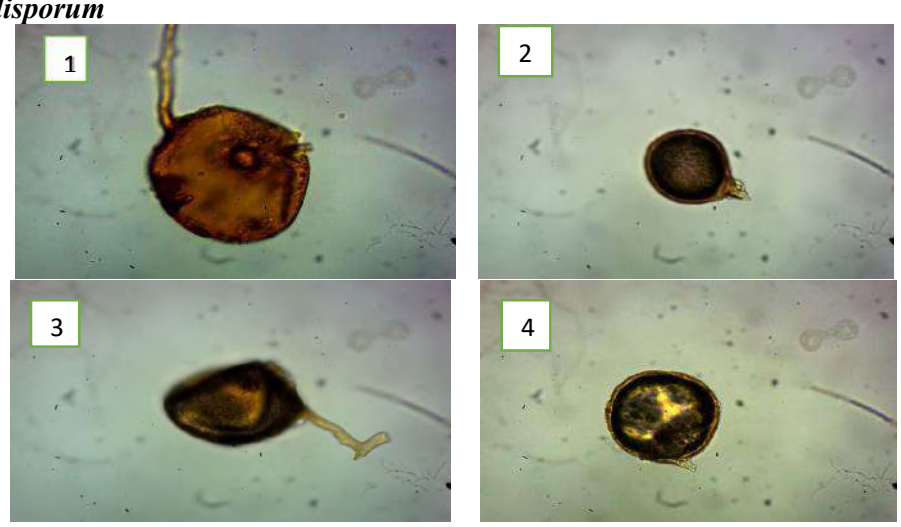
Maximum number of spores were found in the rhizospheric soil of *Ocimum sanctum* (271/100 g of soil) followed by *Ocimum gratissimum* (264/100g of soil), and minimum spore number was recorded in *Ocimum basilicum* (212 /100g of soil). Total numbers of 10 different AM fungal spp. were recorded from the rhizospheric soil from three different species of *Ocimum*. Out of which *Ocimum sanctum* shows maximum (10) AM fungi followed by *Ocimum gratissimum (*08) AM fungi each and *Ocimum basilicum (*08) AMF. The result obtained from the present study suggests that all the plants viz. *Ocimum sanctum, O.gratissmum and O. basilicum* show moderate root colonization. However, percent root colonization as well as

number of AM spores and AM fungal species varied from plant to plant. The variability in the spore population and AM species in different test medicinal plants may be interpreted on the basis of their respective root exudation (*Buee et al*., 2000; Tawarava *et al*., 1996).

Conclusion:-

In conclusion, increasing the demand of the *Ocimum* plants in the pharmacological, medical, and cosmetic industries arbuscular mycorrhiza AMF is a real avenue for increasing the quantity and quality of secondary metabolites in the *Ocimum p*lants.

Photoplate I- ***Fig1- G.fasciculatum 2. G.fecundisporum 3. G.retculatum, 4. G.fecundisporum***



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2

Preliminary phytochemical screening of secondary metabolites in leaves of *Schleichera oleosa* (Lour) Oken

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Abstract
In the present work leaves of *Schleichera oleosa* (Lour) Oken, screened for the detection of preliminary phytochemical and secondary metabolites. All parts of this tree are highly medicinal. *Schleichera oleosa* (Lour) Oken. belongs to Sapindaceae and is commonly known as kusum, lac tree, gum tree, or mokha tree. Plants are helpful in itching, hair growth, skin inflammation, etc.70-80% population believed in herbal medicine. Biodiversity of plants is the largest source of herbal medicine. From the seed of this tree oil is extracted and popularly known as 'kusum oil'. The present investigation of water, ethanol, acetone, and chloroform leaf extract revealed the existence of some secondary metabolites such as carbohydrates, protein, phenolic compound tannins, flavonoids, and alkaloids.

Keywords: *Schleichera oleosa*, leaf, phytochemical.

Introduction

Schleichera oleosa (Lour.) Oken belongs to Sapindaceae, commonly known as "Kusum" in the Indian Medicinal System since immemorial. The bark of plant is reported to have antioxidant properties (Srinivas *et al*, 2013). Phytochemical is the study of biochemical compound which is naturally occurs in plants. Preliminary phytochemical screening is the most important aspect for establishing a protocol for the isolation of extract for its chemical compounds. *Schleichera oleosa* (Lour) Oken. They were selected for the present study based on their traditional use for the treatment of various diseases such as dysenteries, wounds, colds, coughs, diarrhea, skin infections, etc (Gond Gopal, 2020). The parts of this tree are helpful in arthritis, malaria, headache, nostalgia, inflammations, and ulcers (Muthukrishnan *et al,* 2017). *S. oleosa* (Lour.) Oken bark and seed are traditionally used in rheumatic pain, hair problems, acne, itching, and skin problems. Seed, bark and leaves of this plant contain some phenolic compounds and seeds contain 40.3% oil with yellowish brown colour (Ram Krishna Sahu *et al* , 2021).

The preliminary phytochemical study of different seed extracts indicated the presence of carbohydrates, proteins, and secondary metabolites such as phenolic compounds tannins, saponins, terpenoids, glycosides, flavonoids, etc. Thus, seeds of *S. oleosa* can be used to cure various ailments also have great potential in pharmaceutical industries for the preparation of herbal drugs (Neha Tiwari *et al*, 2017). The plants contain tannin which has effective use as animal feed it may increase protein supply and also serves anti-inflammatory effects in the throat mouth, anti-diarrhoeal, anti-parasitic, and antimicrobial action (Westendarp, 2006). The plants produce chemicals that are called as phytochemicals. Phytochemicals are important for all living beings. They protect plants from diseases and pollution also phytochemicals are used as a medicine (Vishnu Balamurugan *et al*, 2019).

Materials and Methods

Collection and Identification of Materials:

Schleichera oleosa (Lour.) Oken. leaves were collected from Vanoja forest, district Washim during year 2015-2018. Herbarium of *S. oleosa* was prepared and authenticated from Department of Botany, Shri Shivaji College of Arts, Commerce and Science, Akola. The leaves

were washed and dried in shade for 10-15 days. Dried plant leaf materials were finely powdered with the help of an electric mixture grinder and then stored in air-tight container bottles at room temperature until used.

The solvent that used for plant extraction is important for determining the phytochemicals. Solvents have been less toxic, and easy to evaporate should preserve the compounds (Vishnu Balamurugan *et al*, 2019).10 gram of leaf powder was taken in a 50 ml of beaker and extract was prepared in using four different solvents such as ethanol, water, acetone, and chloroform. Then extract is filtered through Whatman filter paper and freshly prepared extract was used for phytochemical analyses.

Phytochemical Test:

- Carbohydrates: About 0.5 ml of extract was taken in a test tube in which 0.5 ml of Benedict's reagent was added. The mixture was heated for 3 minutes in a boiling water bath. The appearance of red PPT indicates the presence of carbohydrates.
- Proteins: About 2 ml of extract, and 2 ml of Millon's reagent were added. White PPT shows the presence of proteins.
- Amino acid: 2 ml of extract and, a few drops of nitric acid was added. The appearance of yellow color indicates the presence of protein and free amino acids.
- Oil: A small quantity of the extract was taken and pressed between two filter papers. The appearance of the spot indicates the presence of oil.
- Gums and mucilage: About 1 ml of extract, 1 ml of distilled water and 2 ml of absolute ethanol were added white precipitate indicates the presence of gum and mucilage.
- Carboxylic acids: About 1 ml of extract was taken, added a pinch of sodium bicarbonate. The formation of effervescence indicates the presence of carboxylic acid.
- Alkaloids: 2 ml of extract and, a few drops of Mayer`s reagents were added white precipitate indicating the presence of alkaloids.
- Glycosides: 2 ml of extract, and 3 ml of chloroform were added and shaken. The chloroform layer was separated, and then 10% ammonium solution was added. The formation of pink color shows the presence of glycosides.
- Phenol: About 5 ml of extract and 3 ml of 10% lead acetate solution were added. The formation of a bulky white precipitate shows the presence of phenol.
- Polyphenol: 1 ml of extract and a few drops of 5% lead acetate solution were added The formation of a yellow precipitate indicates the presence of polyphenol.
- Tannins: 5 ml of extract was taken and a few drops of 5% ferric chloride solution was added. The formation of a dark green color shows the presence of tannins.
- Flavonoids: 1 ml of extract was taken and 10%of lead acetate was added. The yellow precipitate indicates the presence of flavonoids.

Saponins: 0.5ml extract was vigorously shaken with a few ml of distilled water. The formation of frothing shows the presence of saponins (Vishnu Balamurugan *et al*, 2019; Sahira *et al*, 2015).

Observations and Results:

Preliminary phytochemical screening of *S. oleosa* leaves revealed the existence of various secondary metabolites. Ethanol, Chloroform, Acetone, and Water extract revealed the presence of Carbohydrates, Proteins, Amino acids, and Coumarins. All solvent extract showed the negative test for Fatty acids, Gums and mucilage, and Carboxylic acid. The water extract showed a negative test for Oil and the rest of the solvent showed a positive test. Ethanol and Acetone extract revealed the existence of glycoside whereas water and chloroform show absence. Chloroform extract showed a negative test whereas Ethanol, Acetone, and Water extract showed a positive test for Glycosides. Ethanol extract revealed the presence of Phenol, Polyphenols, Tannins, Flavonoids, Saponins, Steroids, and Terpenoids. Chloroform extract showed the detection of tannin. Whereas acetone extract revealed the presence of Phenol,

Glycosides, Flavonoids, and Saponins. Water extract showed the detection of Phenol, Polyphenol, Flavonoids, and terpenoids.

Table no. 1: Preliminary phytochemical analysis of *S. oleosa* leaves.

Sr. no.	Test	Ethanol	Chloroform	Acetone	Water
1.	Carbohydrates	+	+	+	+
2.	Proteins	+	+	+	+
3.	Amino acid	+	+	+	+
4.	Fatty acids	-	-	-	-
5.	Gums and mucilage	-	-	-	-
6.	Carboxylic acid	-	-	-	-
7.	Oils	+	+	+	-
8.	Glycosides	+	-	+	-
9.	Phenol	+	-	+	+
10.	Ploy phenol	+	-	-	+
11.	Tannins	-	+	-	-
12.	Flavonoids	+	-	+	+
13.	Saponins	+	-	+	-
14.	Steroids	+	-	-	-
15.	Terpenoids	-	-	-	+
16.	Coumarins	+	+	+	+

Present +, Absent -.

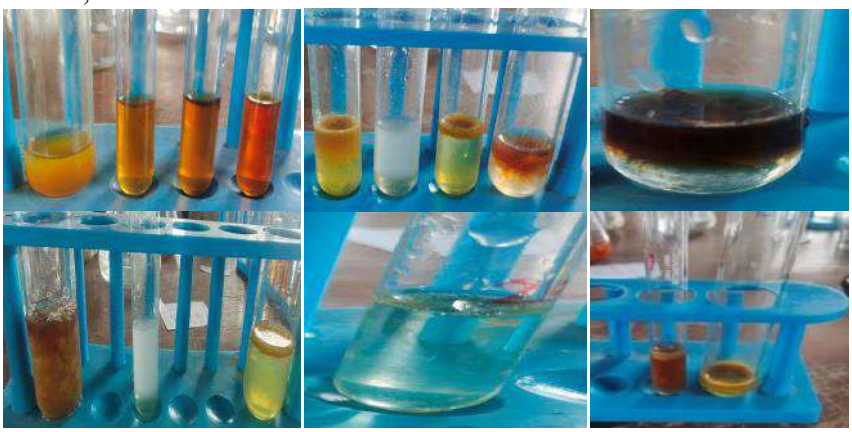

Fig. Photograph detection of various phytochemical

Conclusion

Thus from this present work, it is concluded that *S. oleosa* leaf extract produces various phytochemicals which are help to cure different diseases. *S. oleosa* leaf extract contains various secondary metabolites such as Carbohydrates, Proteins, Amino acid, Coumarins, alkaloids, flavonoids, tannins, triterpenoids, polyphenols, and Phenol. All plant parts are highly medicinal.

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3

Evaluation of phenolic compounds from leaves and stem of *Abutilon pannosum* (G.Forst) Schltdl

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Abstract
The present study was conducted to evaluate the antioxidant property depending on phenolic compounds content of *Abutilon pannosum* leaves and stem. Phenolic compounds are vital in defense responses including anti-inflammatory, anti-aging, antioxidant and antiproliferative activities also involve in defiance against UV radiation. Antioxidants have been reported to prevent oxidative damage caused by free radicals and can be used in the treatment of cardiovascular diseases. The main objective of this study is to evaluate the total phenol, ortho dihydric phenol, quinones, total flavonoids and tannins. For determination of phenolic compounds, 1g leaves and stem were used. The highest amount of total phenol, ortho dihydric phenol, quinones, total flavonoids and tannins were determined from the leaves while the least amount was determined from the stem of *Abutilon pannosum*.
Key words: *Abutilon pannosum*, Phenolic compounds, Spectrophotometer, Antioxidant.

INTRODUCTION

Medicinal plants play an important role in medicine both in modern as well as traditional views. Presence of various phytoconstituents such as flavonoids, alkaloid, tannins and phenolic compounds are responsible for medicinal property of plants they also are considered as pharmacologically active. Which is important component of the human diet due to their Antioxidant properties. It acts as antioxidants, structural polymers (lignin), attractants (flavonoids), signal compounds (salicylic acid, flavonoids) and defense response chemicals (tannins, phytoalexins). The phenolic compounds extracted from plants may help in the treatment of carbohydrate absorption, like diabetes by inhibiting the absorption of amylase. Plants synthesize phenolic compounds act as defense mechanisms against pathogens, parasites, and predators and also response to ecological and physiological conditions and mainly when they are attacked by pathogens and insects or exposed to UV radiation and wounding (Chung *et al.*, 2003; Crozier *et al.,* 2006; Diaz Napal *et al*., 2010; Kennedy and Wightman, 2011).

In biological system, cells can be damaging the DNA and lead to the oxidation of lipid and proteins by the reactive oxygen species (ROS) and reactive nitrogen species (RNS), excessive production of ROS and RNS induced by the exposure of cigarette, radiation, smoking, alcohol and environmental toxins. Naturally, human body can scavenge these radicals by antioxidant system occurring in human body, which helps to keep the balance between oxidation and anti-oxidation. Intake of exogenous antioxidants would improve the damage generated by oxidative stress through inhibiting the initiation or propagation of oxidative chain reaction. Acting as free radical scavengers as well as quenchers of singlet oxygen and reducing agents (Baiano and del Nobile; 2016).

Abutilon pannosum family (Malvaeae) commonly known as 'Kanghibunti', it is Perennial shrub. That grow in India, Arabia, China, Pakistan and Tropical Africa. The A. pannosum leaves were utilized in addition to medicines for stack problems. According to Bagi *et al*. (1985), the plant contains mucilage, tannins, gallic acid and sesquiterpenes. Historically, A. pannosum has been used to treat anemia, hemorrhoids, diabetes, urinary tract infections, and

wounds and ulcers (Ali *et al*, 2008). The root bark is applied topically to treat pyrexia, sedative, diuretic and in pulmonary disorders (Sadhu *et al*., 2016). Native people in the area utilize the plant's decoction to treat diarrhea, gonorrheal diseases, bladder inflammation, bronchitis, and high body temperature. Additionally, it is utilized for abrasion cleaning.) (Basu *et al*., 1991).

MATERIAL AND METHODS:

Plant material

The extensive survey, identification and collection of plant from Amravati region was carried out. Plant identification was carried out with the help of floras (Cook,1957; Dhore,1986;1998; Naik, 1998).

Preparation of plant material

Fresh leaves and stem were collected, dried under shade, finely powdered and stored in airtight container. All plant part were powdered separately. 1gm each of each plant part were taken for estimation of phenolic compounds gm/ µgm.

METHODS:

Estimation of Phenolics such as total phenol, Ortho-dihydric phenols, Quinones, and Tannins and Total flavonoid content were done according to the methods prescribed by Thimmaiah (1999), Saranya *et al*, (2017) which are given below.

Estimation of total Phenols

1gm of sample was grind with the help of mortar and pestle with 10 ml of 80% ethanol. And centrifuged at 10,000 rpm (20 minutes). Supernatant was collected and evaporated to dryness. after dryness residue was taken into a test tube and make up the volume with 5ml distilled water. 1 ml aliquot was Pipette out in test tube, and make up the volume up to 3 ml with distilled water. 0.5 ml of Folin- Ciocalteu reagent was added. After 3 minutes, into each tube 2 ml of 20% Na_2CO_3 solution was added. Mixed thoroughly and tubes was kept in boiling water for 1 minute, then allowed to cool and absorbance was measured at 650 nm against reagent blank. reagent blank was prepared similarly without the extract. Standard curve was prepared using different concentrations (0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, and 1 ml) of catechol. (Thimmaiah S. R. 1999).

Estimation of Ortho- dihydric phenols

1gm sample was grind with the help of mortar pestle with 10ml of 80% ethanol. And centrifuged it at 10,000 rpm (20 minutes) and supernatant was collected and evaporated to dryness. After drying residue was taken and volume made up to 5ml with distilled water. 1 ml of aliquot was pipette out in a test tube, in this test tube 1ml of 0.05 N HCL, 1 ml of Arnow's reagent, and 10 ml of distilled water and 2ml of 1N NaOH was added. Absorbance was measured at 515 nm against a reagent blank lacking only extract. Standard curve was prepared using different concentrations (0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, and 1 ml) of catechol. (Thimmaiah S. R. 1999).

Estimation of Quinones

1gm sample was grind with the help of mortar and pestle by using chilled phosphate buffer (5ml for each gm of tissue). The supernatant was collected after centrifugation for 30 minutes this was used as enzyme extract. 3ml of buffer, 3ml of standard catechol and 1.5 ml of enzyme extract was pipetted in a test tube. It was shaken gently and then placed in water bath for incubation. 4ml of TCA (Trichloro acetic acid) reagent (without ascorbic acid) to one and 4ml of TCA reagent (with ascorbic acid) was added. Precipitate was filtered. Absorbance was measured at 400 nm against a reagent bank lacking only extract. Standard

curve was prepared using different concentrations (0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, and 1 ml) of working standard catechol. (Thimmaiah S. R. 1999)

Estimation of Tannins

Vanillin hydrochloride method was used.

1 gm of sample was mixed in 10ml methanol after 20-28 hrs. centrifuged and supernatant was collected. 1ml of supernatant was pipette out into test tube and quickly added 5ml of vanillin hydrochloride reagent and mixed. After 20 min absorbance was read at 500nm. A reagent blank was prepared with vanillin hydrochloride reagent alone. A catechin standard graph was prepared from working standard (100µg/ml) of catechin and amount of tannins was calculated. (Thimmaiah S. R. 1999). A standard graph was obtained by plotting concentration on X- axis and the corresponding values of absorbance along Y- axis on a graph paper resulting straight line which passes through the origin and maximum points of standard reading. It is used to quantify the amount of a given compound present in an unknown sample whose absorbance value is matched against that of standard along Y-axis and a corresponding concentration could be read off along X-axis. (Thimmaiah S. R. 1999).

Estimation of total flavonoid content

The Aluminium chloride method was used to determine the total flavonoid content. In brief, 1gm sample was grind with the help of mortar and pestle with 10ml methanol. centrifuged and supernatant was collected. 0.5ml of supernatant was pipette out into test tube. 0.5 mL of each extract (1:10 g/mL) in methanol was separately mixed with 1.5 mL of methanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate, and 2.8 ml of distilled water was added. The reaction was left for completion for 30 min, and absorbance was measured at 415 nm against a methanolic blank (80% methanol). Standard curve was prepared using different concentrations (0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, and 1 ml) of catechol (Saranya *et al*, 2017).

RESULT AND DISCUSSION:

Sr. No	Parts used for Estimation	Total Phenol		Orth dihydric phenol		Quinone		Tannin		Total Flavonoid	
		Absorbance	µg /gm	Absorbance	µg / gm	Absorbance	µg/ gm	Absorbance	µg / gm	Absorbance	µg / gm
1	Leaves	0.80	1,200	0.10	140	0.114	1,460	0.023	850	0.110	220
2	Stem	0.58	765	0.07	95	0.110	1410	0.012	450	0.020	40

The results showed that the maximum amount of Total Phenol (1,200µgm/gm), ortho-dihydricphenol (140µgm/gm), Tannin (850µgm/gm) and Total Flavonoid (220µgm/gm) was observed in Leaves of *A. pannosum* while the least amount of Total Phenol (765µgm/gm), ortho-dihydricphenol (95µgm/gm), Tannin (450µgm/gm) and Total Flavonoid (40µgm/gm) was observed in stem. However, there is significant amount of quinone was found in Leaves (1,460µgm/gm) and stem (1,410µgm/gm).

In methanol extract of *A. pannosum* stem bark the total phenolic content determined was 55.485 ± 0.85 mg GAE/g dry extract while the total flavonoid content was 19.90 ± 0.58 mg Rutin/g dry extract (Khalil *et al*,2022). Only the methanolic extract of *A. pannosum* lacks flavonoids. Strong anticancer activity and potent water-soluble antioxidant characterize it (Asif and Khodadadi, 2013). It has been observed that flavonoids and phenolic compounds

have an antioxidative effect on biological systems by scavenging free radicals and singlet oxygen. (Rice-Evans, *et al*., 1997). It is well known that flavonoids and phenolic compounds have the ability to scavenge nitric oxide (Kim H. *et al.,* 2002). Many studies have been reported that coronary heart disease and cancer mortality reduced when dietary intake of natural phenolics increased, it also found effective in various health-related properties like anticancer, anti-inflammatory, antioxidant and antiviral activities (Ghafar, *et al*., 2010). Phenolic compounds especially flavonoids, phenolic acids and tannins, act as inhibiter of α-glucosidase and α-amylase, which are responsible for the digestion of dietary carbohydrates to glucose. Lin, *et al* (2016). tannins show the antimicrobial activity, tannins of unknown origin inhibited the growth of filamentous fungi Fomes annosus with a minimum inhibitory concentration (MIC) greater than 0.5 g/l (Haars et al.1981). Eyong, *et al* (2008) investigated that quinones exhibit antibacterial, antioxidant, ant plasmodial, neurological, antitumor, trypanocidal and anti-HIV activity.

Conclusion:

This study reveals that the leaves of *A. pannosum* contain highest amount of total phenol, ortho-dihydricphenol, tannin, quinone and total flavonoid justifies the medicinal use of the plant, good source of antioxidant activity, anti-aging, anti-inflammatory, anticancer, anti-diabetic, antimicrobial, ant plasmodial, neurological, antitumor, and anti-HIV activity and modulate immune responses. Even though *A. pannosum* is a common weed, it has not been thoroughly studied. As a result, the current study's findings may help the pharmaceutical industry to understand how this plant might be beneficial for human welfare. The use of natural antioxidants has been encouraged because of worries about the safety of synthetic drugs.

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4

Mitotic index and Chromosomal abbreviation in Gamma irradiated Wild Pea

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Abstract:
Cytogenetic studies are important for obtaining information regarding the role and effect of various mutagens and elucidating the response of various genotypes to a particular mutagen (Sharma *et al*., 2004). Though considerable work has been done on induced mutational studies in many plants mitotic abnormalities induced by physical mutagens have rarely been reported in wild *Pisum sativum* (L.)**.**
Keywords: *Pea, Physical mutagen, mitotic abnormalities,*

Introduction:
Mutagenic agents have been used to induce useful phenotypic variation in plant for more than 70 years**.** A large number of mutant lines have been isolated from many plants and these were used for plant research and crop breeding purposes (Evans, 1962; Gottschalk, 1983)**.** New techniques are needed for further improving crop cultivar apart from the traditional plant breeding. 'Mutational breeding' is therefore being promoted as a means to create additional variations. The application of ionizing radiation, chemical mutagen as well as somoclonal variation from tissue culture is quite common in the creation of genetic variations. Novel plant mutant like maize, alfalfa, potato, banana and barley among other and other cell lines of agricultural and industrial interest generated from tissues culture have also been quite popular**.** Physical mutagens like ionizing radiations (viz X-rays, gamma rays and neutrons) and UV light and also a series of chemical agents are the common example of mutagenesis technique that have a high efficiency at generating mutation in plants, animals as well as bacteria. In addition, the outcome of this treatment can at least be predicted to a certain extent.
The commonly used mutagenic agents cannot produce new genes but in fact they only alter those present in the treated genotype. Ionizing radiation, for instant generates chromosomal aberrations. Gene mutation is less frequent than **'chromosomal mutations**', which include translocation, inversion, deletion and deficiencies. Mutation in the narrow sense affected part or section of a gene either single bases pair or group of them. Exchange of base pairs or alteration of their sequences may change the primary gene produced and by way of a more or less complicated chain reaction of event ultimately leads to a modified phenotypic expression of one or several traits. Sometime the gene is affected in such a way limited by somatic effects such as reduced viability, growth abnormality and reduced fertility. Therefore, every mutagen has a most effective dose, which produced the maximum level of mutagenesis with minimal somatic effects. Apart from the choice of the proper mutagenic agent the dosage and the treatment conditions are important. Consideration must also be given to the plant material which is being treated. For example the stage at the life cycle of the plant or plant organs (seeds, pollen, vegetative meristems, etc.) and sensitivity of the plant species to the mutagenic agent. The possible genotypic difference in sensitivity is due to the mutagenic treatments.

Materials and methods:
The wild peas were collected from Melghat region and were treated with different doses of gamma radiation. The following cytological parameters were studied in control as well as in

gamma irradiation seed sets.

1. Germination of seeds

The healthy seeds from different radiation doses as well as from control were selected. Seeds were washed twice with distilled water. Seeds were soaked for 1-2 hour in sterile distilled water. Later on, they were transferred to Petri plate containing wet filter paper folds, to allow their germination in non-contaminated conditions. As the roots reached the length of 0.5cm – 1.0 cm they were cut down using sterile blades or forceps.

2. Fixation and preservation

The germinated roots were cut and immediately fixed in Cornoy's fluid 1 for a period of a minimum 12 hours. Different cutting periods were selected viz. 6.30 to 7.30 am; 8.30 to 9.30 am; 9.30 to 10.30 am and 10 .30 to 11.30 am. The best result in terms of maximum arrested active stages, was obtained, thus the final fixation was done between 11.30 to 12.00 am for 12 hrs. After fixation of root tips from corresponding doses, they were washed with distilled water thoroughly and finally, they were preserved in 70% alcohol for further use in the refrigerator.

3. Staining and squashing

To stain the chromosomal stages at specific loci, the general procedure is to over-stain it, followed by the excess stain- a process called differentiation. Thus the preserved root tips about 3 to 6 in number were washed in water and further hydrolyzed in 1N HCL at 60^{o} C for 15 to 20 minutes in hot air oven. After cooling down, a simple wash of water was given to them.

For staining the tips of the root were kept in 0.5% acetorcein for 10 to 15 minutes with slight heating on flame. Mordanting was done by iron needle, with very little pressure and also it needed further tapping with needle over cover glass. Cover glass was sealed with sealing wax thus temporary slides were prepared. The excess stain was removed by soaking it by filter paper, with much care. Slides of Belgian glass slides with smooth edges and a uniform thickness of 1mm are used with cover slip No. 1 18 mm thickness.

The observation was made under the light microscope for root tips from each radiation dose as well as for control. During the observations, the selection of a field having quite a good number of dividing cells was finalized. Total number of dividing cells, total number of abnormal cells, as well as cell with stickiness, breakages, anaphase bridges, scattered chromosomes, and micronucleus, were scored.

Photomicrographs of important plates were taken on an Olympus CK40 trinocular microscope instrument at 45 X and 10 X using the provided photographic attachment. Five slides for each dose along with control were taken. Observations were taken by focusing on the field having a maximum number of cells. Ten readings for each slide were taken in a zig-zag manner to cover the maximum surface of the slide. Various stages such as prophase, metaphase, anaphase, and telophase along with their abnormalities were scoured.

It was also found that the best result was obtained with 0.5% acetorcein instead of 0.2% acetocarmine. Thus the fresh slide from each treatment was observed for mitotic index and chromosomal irregularities.

A) Mitotic index

Mitotic index in term of the percentage frequency of dividing cells was taken into consideration. The active mitotic index was calculated by scoring only metaphase and anaphase from total dividing cells.

It was calculated as follows

$$\text{Active mitotic index} = \frac{\text{Number of metaphase + Number of anaphases}}{\text{Total Number of cells observed}} \times 100.$$

B) Chromosomal irregularities

The following chromosomal irregularities in mitosis were observed at the treatment of gamma radiation given to wild *Pisum* seeds.

a) An irregularity in prophase such as giant nuclei.

b) An irregularity in various Metaphases such as stickiness fragments disorganized

c) Metaphase change in polarity was scored.

d) Irregularities in various anaphases such as bridges, stickiness, fragments, laggards and disorganized anaphase etc.

e) The percentage of disturbed metaphase and anaphase was calculated using the following formula.

$$\text{Percentage of disturbed stages} = \frac{\text{Total no of disturbed stages}}{\text{Total no of dividing cells}} \times 100.$$

II) Materials required

For cytological preparations, material includes collection of seed irradiated with various gamma doses and control sets.

1. Fixative

Fixative may be defined as the process by which tissues or their components are fixed selectively at a particular stage to a desired extent. The purpose of fixation is to kill the tissues without causing any distortion of the components to be studied, as far as it is practicable (Sharma, 1990).

2. Carnoys fluid I (3:1)

Carnoys fluid I was prepared by adding 3 parts by volume of absolute alcohol to 1 part by volume of glacial acetic acid.

It is effective for all plant, animals, and human materials both for squash and block preparation, the period of fixation varying from 15 min to 14 h in cold or at room temperature.

3. Preservative

The 70% alcohol is found to be the best preservative for root tip. Material can be well maintained in this preservative at least for a period of two months.

4. Hydrolyzing agent

1N Hydrochloric acid – The 9.8 ml chemically concentrated hydrochloric acid (HCL) was dissolved in 10 ml of distilled water and volume was made to 100 ml with distilled water. To bring out good separation of chromosomes, and for clearing cytoplasm pretreatment was essential (Dnyansagar, 1986).

5. Nuclear stains

The two nuclear stains were used for the squash preparation. Acetocarmine 2% and 0.5% acetorcein. However, the best staining result was obtained in 0.5 % acetorcein stain.

a) Acetocarmine (2%)

First 45 ml of glacial acetic acid was diluted with distilled water to make 100ml. The 45% acetic acid thus prepared was boiled. Two grams of carmine powder was weighed and added to boiling acetic acid slowly. This solution was allowed to cool down up to 58^{o}C and then filtered, through Whattmans filter paper no 1. The filtered solution was labeled as 2% acetocarmine solution.

b) Acetorcein (0.5%)

Orcein was first employed as a chromosome satin by La Cour in 1941. The dye has a molecular weight of 500.488, the formula being C28H24N2O7. It is a very effective stain for the study of root tip and leaf tip chromosomes.

The 0.5 gm of orcein powder was added in 50 ml of 45% acetic acid which was boiled for 5

to 10 min, after condensation. Stain is filtering through Whattmans filter paper No. 1. The filtered purple color solution is labeled as 0.5% acetorcein solution (Sharma, 1990).

Statistical Analysis

Root tips 0. 5cm to 1.0cm long were cured and after 12 hours of treatment with Conroy's solution stored in 70 % alcohol. Before squashing, the root tips were treated with 1N HCl at 50◦ in oven for the softening of cell. Cytological studies were made from temporary acetorcein (0.5%) squash preparations. Different mitotic abnormalities were scored. The mitotic index was determined Bhalla *et al*., (1973).

The mitotic index was calculated by using the following formula

$$\text{MI (mitotic index)} = \frac{\text{Number of dividing cells}}{\text{Total number of cells}} \text{ x 1 00}$$

This was calculated for each treatment as a number of dividing cells/100 cells. Different types of aberration in each stage were scored and the percentage of each aberration was calculated by the formula,

$$\text{Percentage of chromosomal aberration} = \frac{\text{Number of abnormal cells}}{\text{Total number of cells}} \text{ x 100}$$

Standard Error

Ten aliquots of each treatment were used for recording observations. The data obtained was statistically analyzed as per the procedure given by Panse and Sukhatme (1978) using the formula as follows:

$$\overline{X} = \frac{\sum \overline{X}_i}{n}$$

Where, $\overline{X}$ = mean

$\sum \overline{X}_i$ = sum of 'i' observations

n = no. of observation

$$\text{S.D.} = \sqrt{\frac{\Sigma(\overline{X}_i - \overline{X})^2}{n-1}}$$

Where, Xi = Observations (i = 1.2---n)

$\overline{X}$ = mean

S.D. = Standard deviation

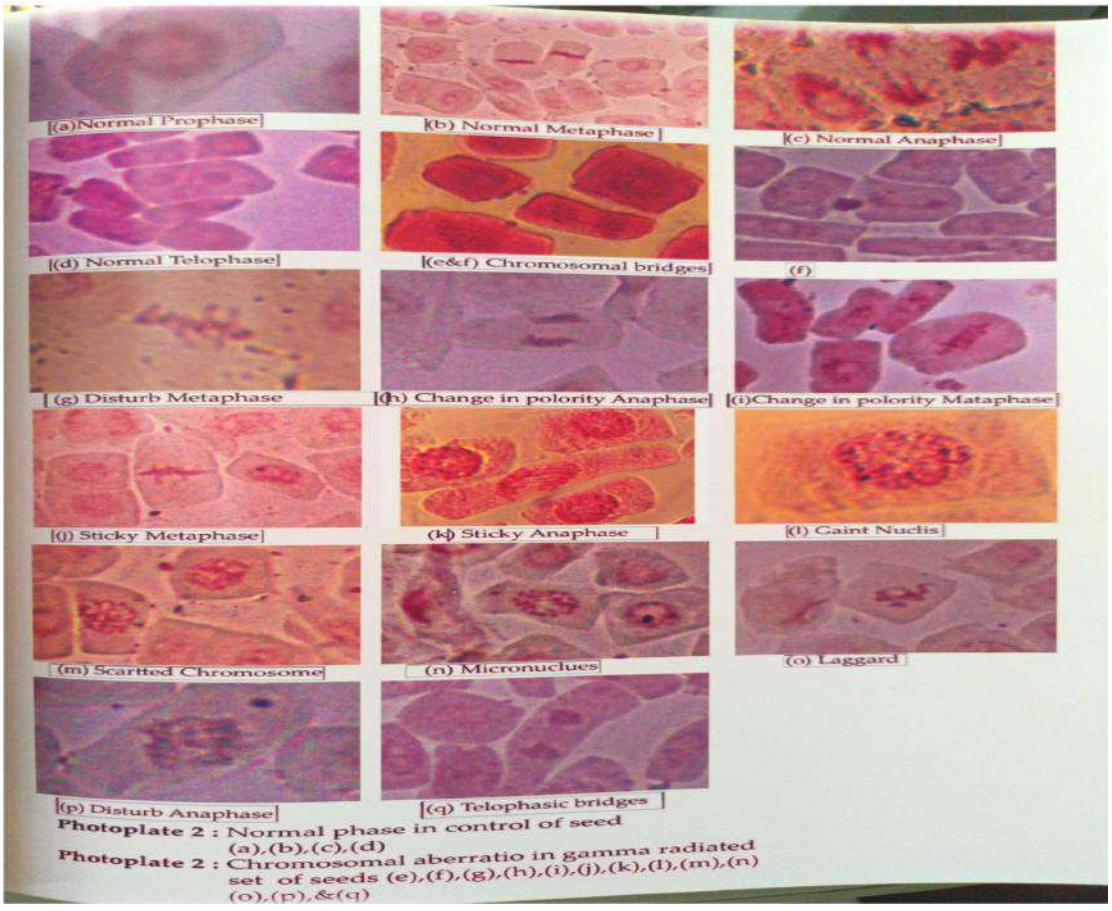
$$\text{S.E.} = \frac{\text{S.D.}}{\sqrt{n}}$$

Where S.E. = Standard error

Results and Discussion

In the present investigation the effect of different gamma radiation on the cytological study was carried out in control as well as irradiated sets of roots from germinated seeds from petriculture techniques. To study the effect of ionizing radiation on the plant material the quick and easy method is the analysis of the first mitotic cycle in the root tip cells of irradiated seeds. The result of the present investigation, shows the effect of six doses of gamma radiation from 5KR, 10KR, 15KR, 20KR, 25KR, and 30KR, along with control i.e. untreated seed. It was observed that, in control, no abnormalities were observed and well-spread metaphase plates showing 14 chromosomes could be easily seen while the treated seeds showed different types of mitotic abnormalities.

Mitotic index- However, in various mutagenic treatments a number of mitotic irregularities were induced. The number of cells showing various anomalies scored at different stages has been summarized. Mitotic index was found to be normal in the control set of seeds, which e shows the regular distribution of chromosomes at the mitotic metaphase stage. Normal formation of centromere with attached spindle fiber facing at opposite pole. All over meristematic cells of the untreated control root showed normal mitotic figures with normal prophase, metaphase, anaphases, and telophase. The treated seeds show various abnormal as well as normal stages represented in **Photoplate no 2a-d** and **Photoplate no 2e-n**. The graphical presentations of mitotic index (M.I) in control as well as in treated were presented in **Fig 1**.



Photoplate 2 : Normal phase in control of seed (a),(b),(c),(d)

Photoplate 2 : Chromosomal aberratio in gamma radiated set of seeds (e),(f),(g),(h),(i),(j),(k),(l),(m),(n) (o),(p),&(q)

Table 4- represents the mitotic index as well as standard error in control as well as gamma radiated seed of wild *Pisum sativum.* Similarly a percentage abnormality for different doses has been calculated by total number of aberrant dividing cell and number of cell scored. Whereas mitotic index had calculated by total number of dividing cell upon total number of cell scoured. Total no of dividing cell induced normal + disturbed cell in metaphase + anaphase + telophase respectively.

Table 4 -Frequency of different types of abnormalities at different stages of cell division after gamma radiation treated wild *Pisum*

Doses	**Control**	**5KR**	**10KR**	**15KR**	**20KR**	**25KR**	**30KR**
Total no of cells	4007	3150	3609	3653	3616	3371	3596
Total prophses %	0	14.07	11.61	11.5	18.94	20.85	30.2
Total abberant metaphase	0	20.16	22.0	25.56	30.9	24.4	22.19
Total abberant anaphase	0	8.55	10	5.69	16.62	15.7	18.73
Total abberrant telophase	0	4.1	5.9	0	2.9	0	0
Total percentage abnormalitiy	0	1.488	1.37	1.17	2.0	1.80	1.97
Mitotic index	11.63	7.9	9.76	10.2	7.43	7.066	6.530
mi±SE	11.63±1.62	7.93±0.57	9.79±1.094	10.2±1.21	7.43±0.41	7.06±0.26	6.53±0.15

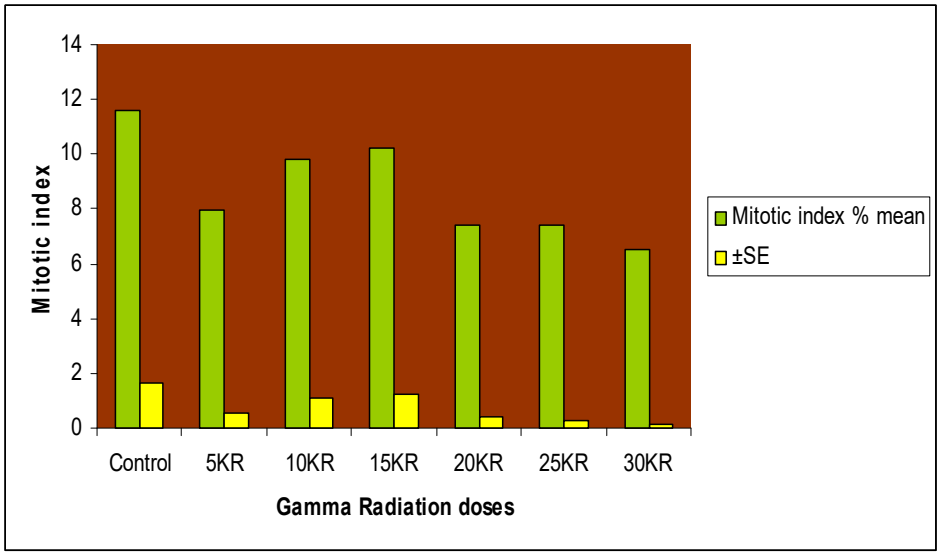
Table -5 represents chromosomal irregularities at different phases. In this percentage abnormality for different stages were calculated by scouring total number of particular abnormality upon total number of dividing cell multiplied by hundred. These were calculated for all doses of gamma radiation.

Table 5 – Gamma radiation induced chromosomal irregularities on % basis in mitosis of wild *Pisum* Variety

Irregularities	**5KR**	**10KR**	**15KR**	**20KR**	**25KR**	**30KR**
Aberrant Prophase						
Gaint nuclei	14.07	11.61	11.5	18.94	20.85	30.2
Aberrant Metaphase						
S M	4.7	8.3	5.4	13.9	12.7	9.86
C.P.M	3.66	5.3	7.69	10.3	2.89	3.46
S.C	5.70	6.19	10.2	6.66	5.77	6.87

D.M.	6.1	2.21	2.275	0	3.12	2.0
Total A.M.	20.16	22.0	25.56	30.9	24.4	22.19
Aberrant Anapha						
S A	2.24	1.60	3.41	4.51	6.35	6.41
A B	1.35	0.800	0.85	6.06	4.67	3.45
C.P	1.80	3.20	1.42	5.45	3.49	3.94
S.C	0	2.00	0	0	0	5.91
Laggard	0	0.79	0	0	0	0
D A	1.35	1.69	0	0.6	0.5	0
Total A.A	8.55	10	5.69	16.62	15.7	18.73
Aberrant Telopha						
M N	4.1	5.9	0	2.9	0	0

Fig 1: Showing column notation of total mitotic index of control as well as gamma-irradiated seeds.



However, various chromosomal abnormalities were recorded at various doses of gamma treatment. Although the spectrum of abnormalities was more or less the same for all doses there was a considerable difference in the frequency of specific anomalies. The results are shown in **Table -5**, which shows the frequency of different types of abnormalities at different stages of cell divisions after treating wild *Pisum* with different gamma doses.

The results reveal that the mitotic index showed corresponding fall, as the radiation doses increased. As in the present investigation, it was observed that the mitotic index for 5KR decreased form 7.93±0.57 to 6.530 ±0.15 for 30 KR which the same result also represented by Kumar *et al*. (1997). Considerable reduction in mitotic index indicates the potentiality of the gamma rays to arrest cell division at G1 phase or retardation during the S or G2 phase of the cell cycle. Increased concentration of the gamma dose interferes with the normal sequences of the cell cycle and reduces the number of cells to enter prophase and succeeding divisional stages. Such types of mitodepressive action of several chemicals/physical agents have been reported earlier (Omnakumari *et al*., 2007).

Although the Mitotic index for each dose were calculated and exhibits that it decreased considerably, whereas in 15KR, there was a slight increase from the rest of the doses as represented **Table- 4**. In the root tip cell of the irradiated seed it is observed, that due to

increasing doses of gamma radiation, the value of the mitotic index was decreased as compared to the control. This means 15 KR dose induces the cell cycle there by it permitting to increased span of mitosis (M- phase) and necessarily reads a biochemical environment for proper regulation of G1, G2 phases. Ultimately whole biochemical setup increases the DNA synthesis i.e. higher M.I was found to be at 15 KR. However the increase are in lower doses was more significant but after LD_{50}, the deleterious effect of radiation is more prominent (Jayabalan and Rao, 1987).

Prophase abnormality

In the Prophase phase, the percentage of aberrant prophase was found to be increased with that of increasing doses. In 5KR percentage frequency of giant nuclei was found to be 14.07% where as that of in 30 KR it was calculated to be 30.2%. This type of aberration arose due to breaking up of nucleus. As a result nucleus gradually disintegrated and disappeared from the cytoplasm. Nuclear lesions appeared in a number of cells. Bausch (1974) reported that nuclear lesion appeared as area of chromosome disintegration. According to him the chemical reacts with basic protein of the chromatin resulting in the breakdown of chromatin and subsequent disruption. Here the prophase nucleus showed light and dark regions. Changed nuclear morphology was appeared in a number of cells in all the treatments. Such abnormality was also noticed in treatment with *Cannabis* (Hashish) by Malallah and Kabarity, (1982).

Metaphase abnormality

Percentage of chromosomal abnormalities increased with increasing doses. Stickiness of chromosomes was the most common abnormality observed during the present investigation. Rate of sticky metaphase was found in lower doses of 5 KR, 10 KR, 15 KR, were as percentage frequency at 20KR and 25 KR was found to be higher by13.9% and 12.7%. Stickiness and clumping of chromosome may be suspected to be the primary effect of radiation caused by depolymerisation of DNA or disruption of bonds between protein and nucleic acid constituents of chromosomes (Cohn, 1979; Jaybalan and Rao, 1987) they also reported that stickiness was due to the disturbance in cyto-chemically balanced reaction by secondary effect of radiation. Stickiness could be due to depolymerisation of nuclei acid caused by mutagenic treatment or due to partial dissociations of the nucleoprotein and alterations in their pattern of organization (Sharma *et al*., 2003). Stickiness could be due to partial or complete clumping of chromosomes (Chaudary *et al*., 2004).

Disturbed metaphase was also more prominently found in aberrant metaphase whereas frequency was found to be higher 5 KR that is 6.1% whereas in 20 KR no such abnormality was found. Disturb metaphase may be due to the disturbance of spindle appearances.

Anaphase abnormality

Anaphasic separation in untreated material was quite normal. Abnormalities during anaphase as single, double or multiple chromatin bridges, one, two, or more laggards with or without an unequal distribution of chromosomes, and multipolar distribution were commonly observed with varying frequency in different treatments. Among these abnormalities, chromosomal bridges were more prominently found in higher doses of gamma rays. In our investigation percentage frequency of chromosomal bridges (Anaphasic bridges) was found to be higher at 20KR doses that are 6.06% where as in 25KR and 30KR it was found to be 4.67%and 3.45%. The formation of anaphasic bridge may be due to chromosome breakage and reunion. On other hand, bridges produced by gamma radiation may attributed to the stickiness of chromosomes, which makes their separation, and free movement complete and thus remain connected by bridge. The chromosomal bridges were observed in higher dose viz from 20 KR, 25KR, and 30KR, formation of chromosomal bridges (anaphase bridges) may be due to chromosome breakage and reunion. Due to an effect of higher doses of gamma radiation generally cause a chromosomal breakage.

Telophase

During further division in telophase these group of chromosome might have given rise to separate 2-3 nuclei into each daughter cell this inhibited the spindle activity of metaphase or anaphase have resulted into multinucleated or micronuclei cell after mitotic division. Bhattacharjee (1953) suggested that irregular distribution of acentrice fragment or laggards may result in the formation of micronuclei at telophase. Percentage of micronuclei was found to be higher in 10 KR doses that are 5.9%. Whereas 5 KR and 20 KR it was found to be 4.1% and 2.9% respectively.

Conclusion:

The present investigation thus envisages using chromosomal mutation for the development of some morphologically important mutants. Mutation breeding offer great prospect for crop improvement by incorporating of various micro –macro mutation. But, before any mutagen is selected for extensive usage, a preliminary screening of the effects of potential mutagens has to be done. Various workers have found different mutagens to be effective in different crops. The great mutagenic potentiality of mutagen can be judged by the percentage of abnormalities induced by it. Although many benefits of ionizing radiation have been discussed society does not uniformly embrace it. Many ill-founded, preconceived ideas and misconceptions have tainted the public's view of radiation. Science fiction movies and shows use the premise of some uncontrolled or diabolical disaster occurring when man tries to tinker with radiation. Since these shows are very popular with lay people who don't understand radiation, they become frightened by it. As a consequence, our society is working to better understand and use radiation to benefit all mankind. In order for the benefits of radiation to be used successfully, a major effort is needed to educate the public.

Following general conclusions can be drawn from the present investigation. Due to effect of gamma irradiation various types of chromosomal aberration were formed in test material. They were in form of sticky metaphase, scattered chromosomes, and non-oriented chromosomes multipolar or change in polarity and chromosomal bridges, giant nuclei add multinucleate or micronuclei formations.

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5

Pharmacognostic studies on *Phaseolus vulgaris* L. Analytical studies

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Abstract :-
Bush bean *Phaseolus vulgaris* L. (Fabaceae) is one such plant, having bean prescribe for Different extracts of Phaseolus vulgaris have been evaluated for pharmacological activities and have shown analgesic, antiobesity, antibacterial, anticancer, antidiabetic, Antifertility, anti-inflammatory, anti-oxidant, hepatoprotective, hypolipidemic ,Litholytic, trypsin and ,α-amylase inhibitor. This crude drug powder study was aimed to develop characteristics of powder crude methods in order to asses the quality of herbal drugs for therapeutic value. Sample subjected to various microscopical characteristics, physicochemical analysis and fluorescence test.
Key Words : physicochemical parameters, crude powder drug, Microscopy.

Introduction :-

Phaseolus vulgaris L. (Fabaceae), bean is an ancient legumes crop widely grown throughout the world for its vegetable or pulse for human consumption or as animal forage. Herbal medicine has been enjoying renaissance among the customers throughout the world. However, one of the impediments in the acceptance of the ayurvedic medicines is the lack standard quality control profiles. The quality of herbal medicines i.e. the profile of the constituents in the final product has implication inefficacy and safety. Due to the complex nature and inherent variability of the chemical constituents of plant based drugs, it is difficulty to establish quality control parameters. To overcome these problems modern analytical techniques are expected to help in circumventing this problem (Bagul et.al 2005). Between 1999 – 2001 the ayurvedic pharmacopeia of India was published in three volumes, which gave the botanical identity of plants , composition, analytical procedures etc. In spite the effort made for the standardization of ayurvedic medicines in actual use are believed to be at least 1000 with many regional variations (Anonymous, 1987). The absence post market surveillance and paucity of test laboratory facilities also make the qualities control of ayurvedic medicines exceedingly difficult at this time. Therefore an attempt has been made to analyse crude power of *Phaseolus vulgaris* L. used in has been reported to be a analgesic, antiobesity, antibacterial, anticancer, antidiabetic, Antifertility, anti-inflammatory, anti-xidant, hepatoprotective, hypolipidemic ,Litholytic, trypsin and ,α-amylase inhibitor (Shi John at.al, 2007).

Material & Methods: -

Plant material:-

Phaseolus vulgaris L. (seed) was collected from the local region of Akola (M.S.) and the plant material were authenticated by using flora of Maharashtra **.** Vaucher specimen of the same have been deposited in the laboratory for future reference.

Preparation of powder:-

Crude drug has taken and roasted in a stainless steel pan at a low temperature till it becomes free from moisture. The sample *Phaseolus vulgaris* L.(seed) was powdered in a

pulverizer and pass through sieve number 80#. It is packed in tightly closed container to protect from light and moisture.

Physicochemical Parameters :-

Physicochemical investigation of the drug were carried out and they include determination of moisture, extractive values and ash values . (Asokar et.al 1992).

Determination of foreign matter :-

Drugs should be free from moulds , insects, animal faecal matter and other contamination such as soil, stones and extraneous material.100g of the drug sample to be examined was weighed and spread out in thin layer. The foreign matter (Table 1) was detected by visual inspection, separated, weighed and the percentage present calculated (Pattnayak et.al 2010)

Determination loss on drying :-

It is important that the portion taken was large enough to be representative for the sample . About 10g of accurately weighed drug was dried at 105^0 C for 5 hours, and then weigh again. Percentage was calculated with reference to initial weight (Table 1).

Determination of total ash :-

The determination of total ash (Table 1) is a method to measure the amount of the inorganic residual substance when the drug sample is ignited (Mukhrjee, 2002) .Total ash determination constitutes detecting the physiological ash (ash derived from plant tissue) and nonphysiological ash (ash from extragenous matter, especially sand and soil adhering to the surface of the the drug). For its detection,2g of powdered material was placed in a suitable tared crucible of silica previously ignited and weighed. The powered drug was spread into an even layer and weighed accurately .The material was incinerated by gradually increased the heat, not exceeding 450^0 C until free from carbon, cooled in desiccator, weighed and percentage ash was calculated by taking in account the difference of empty weight of crucible & that of crucible with total ash.

Determination of acid in soluble ash :-

The ash obtained as above was boiled for 5min with 25ml of dilute hydrochloric acid; the insoluble matter was collected on an ashless filter paper, washed with hot water and ignited to constant weight. The percentage of acid in soluble ash (Table 1) with reference to the air – dried drug was calculated.

Determination of solvent extractive values

Alcohol soluble extractive :-

5 gm of coarsely powered air – dried drug was macerated with 100ml of alcohol in a closed flask for twenty four hours , shaking frequently during six hours and allowing to stand for eighteen hours . It was then filtered rapidly , taking precautions against loss of solvent 25 ml of the filtrate was evaporated to dryness in a tared flat – bottomed shallow dish at 105^0 C to constant weight and weighed. The percentage of alcohol – soluble extractive (Table 1) was calculated with reference to the air- dried drug & is represented as %. (Mukherjee2002).

Water soluble extractive:-

5 gm of coarsely powered air – dried drug was macerated with 100ml of water a closed flask for twenty four hours , shaking frequently during six hours and allowing to stand for eighteen hours . It was then filtered rapidly , taking precautions against loss of solvent 25 ml of the filtrate was evaporated to dryness in a tared flat – bottomed shallow dish at 105^0 C to constant weight and weighed. The percentage of alcohol – soluble extractive (Table 1) was calculated with reference to the air- dried drug & is represented as %. (Mukherjee2002).

Results and Discussion :-

In the present study, physicochemical studies were performed . The *Phaseolus vulgaris* L. (seeds) studies for the presence of foreign matter is mentioned in Table 1.

The percentage of moisture content in *Phaseolus vulgaris* was 7.13%, total ash 12.15%, acid soluble ash 2.6%, water soluble ash 3.25%, alcohol soluble extractive 20.1% and water

extractive 27.7%. However on the basis of polarity of solvents, the percentage of successive solvent extractive valued of extracts were in petroleum ether (1.9%), benzene (2.4%), chloroform (16.4%), acetone (18.6%), ethanol (17.4%), and water (21.83%) represented in (Table 1).

The foreign matter was removed and powered was prepared . A part of the pure powdered was kept aside to study the various parameters. Quality test for crude drug powered was performed for moisture content, ash content, water soluble extractive, methanol soluble extractive, acid insoluble ash and water insoluble ash were found to be standard range.

Table 1 – Analytical values of *Phaseolus vulgaris* (L.)

Sr.No.	Parameter studies	Value (% w/w)
1	Total ash value	12.15
2	Acid insoluble ash value	2.6
3	Water insoluble ash	3.25
4	Loss on drying (moisture content)	7.13
5	Solubility percentage in • Alcohol • Water	 20.1 25.7
6	Extractive values in • Petroleum ether • Benzene • Chloroform • Acetone • Ethanol • Water	 1.9 2.4 16.4 18.6 17.4 21.83

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6

Role of Biofertilizers in Sustainable Agriculture and an Environmental Development

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ABSTRACT
Producing healthy crops for the fulfilment of the demands of the world's growing population is fully dependent upon kind of of the fertilizers being used to provide the plants with all the important nutrients but more dependability on the chemical fertilizers is destroying the environmental ecology and negatively impacting the health of humans. Bio based fertilizers are basically the preprations of living cells or latent of efficient potential microbial strains that assist the plants in nutrient uptake by their associations in the rhizosphere region when supplied to the plants either through the seed or the soil .Thus using microbes as bio inoculants is believed to be the best substitute of chemical fertilizers as eco-friendly manner for plant growth and soil fertility.
KEYWORDS- Bio based fertilizers ; Environmental ; Microbial strains ; Soil fertility.

I-Introduction

The term Biofertilizers denotes nutrient inputs of plant growth which are biological origin .The Biofertilizer restore the soils natural nutrient cycle and build soil organic matter .The role of Biofertilizer in agriculture Production assumes Special significance particularly in the present context of expensive Chemical fertilizers. Moreever it can provide to the farmers a new strategy which is helpful for achieving the goal of increasing productivity. "Bio-Fertilizers refer to various inoculants or Cultures containing a specific microorganisms in concentrated form which are derived either from nodules of plant roots or from the soils of roots of Leguminous Plants or Non Symbiotically (free living) or to transfer native soil nutrients such as P,Zn, Cu, Fe,S etc. from the non usable (fixed) form to usable form through biological processes.".Biofertilizer is the need of modern agriculture since demand for safe and residue free food is increasing.Biofertilizer become popular to counter the negative impact of indiscriminate use of chemical fertilizers .Biofertilizers help in fixing atmoshpheric nitrogen, converting soil phosphate and potash into soluble forms to make them available to plants . Biofertilizers are selective microorganisms .They provide cost effective, eco-friendly and renewable source of nutrients.They improve the nutrient availability to the crops in which biological process is involved.

II- Scope and Importance of Biofertilizers

1.Permanent effect -Chemical fertilizers have temporary effect while Biofertilizers have permanent effect without any production problem.

2.Protection- Biofertilizers provide protection against drought and some soil born diseases.

3. **Cheap –** Biofertilizers are very cheap as compared to chemical fertilizers because raw material required for the growth of microorganism is very cheap. The infrastructure and equipment required for growth of microorganism is very cheap. Use of biofertilizers is economical with a high cost: benefit ratio, without risk.

4. **Simple methods –** The production method of Biofertilizer is very simple. It requires low investment, small space and less labour and equipment as compared to production method of chemical fertilizers. They can be manufactured in any simple microbiology laboratory.

5. **Natural –** Biofertilizers are natural. They are not foreign to the soil so they create no

pollution problem.

6. **Biocontrol –** Few microorganisms used as biofertilizers also controls plant pathogens either developing mechanical barrier for entry of plant pathogens e.g. Mycorrhiza or produce antibiotics killing plant pathogens e.g. *Streptomyces*.

7. **Supply nutrient** – They may supply other nutrients and increase fertility of soil. The Azotobacter added in the soil for nitrogen fixation may have amylolytic or proteolytic activity thus Azotobacter also helps in development of humus.

8. **Prevents soil erosion** – Biofertilizers may prevent soil erossion. Many microbial inoculant may produce extra cellular, capsular polysaccharide which is viscous in nature. This viscous substance adheres to the soil particle and prevents erossion of soil

9. **Supply hormones and vitamins –** They may supply vitamins and plant growth hormones. Many microorganisms secretes auxins, ethylene, abscisic acid, cytokinin, pantothenic acid, indol acetic acid and gibberellin like substances which promote plant growth.

10. **Mobilizes immobilized nutrients –** Biofertilizers convert immobilized chemical fertilizers into soluble forms. Soluble inorganic phosphates lost in the soil due to chemical reactions in insoluble inorganic phosphate us again converted soluble phosphate by biofertilizer. Thus biofertilizer can act as a renewable supplement to chemical fertilizers and organic manures.

. **11. Provides essential elements and enzymes-** Biofertilizer provide essential elements like nitrogen, potash phosphorous, sulphur etc. by directly supplying them or transforming them into soluble form; in addition, they also helps plants to uptake several micronutrients. They supplies some important enzymes, hormones and antibiotics that enhance crop growth and crop yields.

12. **Protects from adverse environment -**Some biofertilizers protects plants against drought, high . temperature shock, high salinity etc.

III- Microbes used as Biofertilizers

III-Types of Biofertilizers

Broadly Biofertilizers are divided into seven main categories, these are again divided in subtypes as follow

A. Nitrogen Fixers

The process of converting atmospheric nitrogen into ammonia by the diazotrophic microbes is known as biological nitrogen fixation (BNF). BNF allows the replenishment of total nitrogen content and the fixed nitrogen regulates the crop growth and yield. Chemical fertilizers cause increased nitrogen oxide emission, water eutrophication and soil acidification. Whereas, biologically fixed nitrogen is sustainable and is less available for leaching and volatilization. Nitrogen fixation is more or less limited to bacteria and archaea, which forms a large portion of diazotrophic organisms. Nitrogen-fixing groups include green sulphur bacteria, firmibacteria, actinomycetes, cyanobacteria and all subdivisions of the proteobacteria. However, only methanogens are able to fix nitrogen among archaea. Different bacterial strains are able to carry out nitrogen fixation with different physiologies including: aerobic (for example, *Azotobacter*), anaerobic (*Clostridium*), facultatively anaerobic (*Klebsiella*) or heterotrophs; an oxygenic (*Rhodobacter*) or oxygenic (*Anabaena*)

i)Symbiotic nitrogen fixers – (symbiotic nitrogen fixers live in association with other plant) *Rhizobium, Azolla*. etc.

ii)Non Symbiotic nitrogen fixer –(Non symbiotic nitrogen fixers are free living forms) *Azotobacter, Azospirillum, Anabaena,Nostoc,Oscillatoria*,*Bacillus*,etc.

B. Phosphate Suppliers

Phosphorous is a vital macronutrient required for the growth and development of a plant. Usually, phosphorous exists in the form of tricalcium, dicalcium phosphate and minerals. The process of solubilization and mineralization in soil i.e., conversion of organic form of

phosphate into inorganic form is carried out by phosphate-solubilizing bacteria . Mycorrhiza also play crucial role in phosphorus mobilization, nutrient cycling and enhancement of microbial biomass. Generally, indigenous arbuscular mycorrhizae (AM) are found in soil, which colonizes the plant roots and stimulate plant growth. Inoculation of low phosphorous soil with mycorrhiza causes a sudden increase in availability of phosphorous.

i) Phosphate solubilising microorganisms – *Bacillus, Aspergillus, Pseudomonas*

ii) Phosphate absorber – V.A. mycorrhiza (**VAM** fungi)

C. Sulphur Suppliers

Sulphur is generally regarded as trace element in majority of crop plants. But this is one of the major elements in oilseed crops, some important vegetables (onion, oat, cauliflower etc.) and inn some spices (ginger, garlic etc.) it is important element. Sulphur essential for biochemical synthesis of some important glycosides, pungent compound and disease resistant properties. Soil is composed of organic as well as inorganic sulphur and the process of conversion of organic sulphur into plant utilizable inorganic sulphur (i.e., SO_4^{2-}) form is carried out by sulphur-oxidizing bacteria (SOB) including *Xanthobacter, Alcaligenes, Bacillus, Pseudomonas, Thiobacillus.*Deficiency of sulphur in agricultural soil could be corrected using sulphur oxididizing bacteria as boifertilizer

D. Potash solubilizing bacteria

Potassium is ranked at third position as crucial plant nutrient after nitrogen and Potassium is available in plentiful amount in the soil but only a small fraction (1–2%) of it is available to plants. Hence, a system of continuous replenishment of potassium in soil solution is needed for its adequate availability to crop plants Like other nutrients, potassium also influences growth and development of plants. . In deficiency of potassium, root growth becomes slow and gets poorly developed, seeds will be of small size and disease susceptibility will be more leading to reduction in crop yield PGPRs present in the soil and rhizosphere convert the potassium present in insoluble form into soluble form. Some of the potassium solubilizing microbes (KSMs) are *Acidithiobacillus Arthobacter ,Enterobacter , Paenibacillus , Aminobacter, Pseudomonas, Paenibacillus,, Sphingomonas,, Bacillus,Klebsiella.a*

E. Zinc solubilising microbes-Among micronutrients, zinc deficiency is the most widespread nutrient deficiency. The alternative technology for providing zinc to the plant is to inoculate the crop with the zinc-solubilizing microorganisms. A major portion of zinc available to plant is provided by the microbial activity Microbes produce organic acids, which cause decline in pH and these organic acids act on zinc complexes in soil,thus cause sequestering the zinc cation.Prominent zinc-solubilizing microbes are *Pseudomonas protegens* RY2, *Rhizobium* spp., *Bacillus altitudinis , Thiobacillus thioxidans Azospirillum* and *Gluconacetobacter*.

F. Mycorrhiza -Fungal species like *Aphalosporra, Glomous, Jaigospora, Enterophosphora* etc penetrates roots of different crops (most commonly found in Litchi) and form specialized structures like Vesicles and Arbuscles within the cortex. For this reason they are popularly known as Vesicular Arbuscular Mycorrhiza or VAM. Almost 90% of plants, including the most important agricultural crops, are associated with VAM fungi. VAM fungi reported increases the uptake of water phosphorous and some other micronutrients like Cu, Zn, Mn, or Fe. Besides these, they possess synergistic interaction with beneficial soil microorganisms such as nitrogen fixing and PSMs. VAM fungi also supply some growth regulators to plants and protects crop plants from high temperature shock, drought and salinity and prevents different disease and nematode attack

G. Organic matter decomposer – Cellulolytic, Lignolytic, Proteolytic, or amylolytic

Cellulose decomposing inoculants Many soil borne fungal species like *Aspergillus, Penicillium, Tricoderma, Chaetomium* etc. acts as activator in the decomposition process of plant bodies containing cellulose or lignin. Plant bodies rich in cellulose and/or lignin are

resistant to microbial decomposition and therefore, takes long time before they could be used as organic source of nutrition. High quality compost could be prepared within a short time by applying the mentioned fungal species into organic waste material collected from farm or community

IV-Precaution for use of biofertilizer

1. Biofertilizer containing specific species of microorganism should be applied for specific crop.
2. Biofertilizer packet should not be exposed to direct sunlight for long time, the seeds treated with biofertilizer should be kept for 30 minutes in shady place.
3. For maximum result biofertilizer should always be mixed with bulky organic manures.
4. Biofertilizer should be used before its expiry date.
5. After treating the seeds with biofertilizers, seeds should not be treated with any kind of chemical fertilizer or pesticides.
6. Chemical fertilizers should not be applied one week before or after application of biofertilizer

V-Conclusion

Biofertilizers are one of the key factors in sustainable agriculture that can assist in solving the problems of feeding an increased world population at a time when agriculture is going through various environmental stresses.Hence research should be focussed on new aspects of Biofertilizers. In current agriculture practices , chemical fertilizers have reduced the fertility of soil, making it unsuited for raising crop plants. Additionally, the excessive use of these inputs has also led to severe health and environmental hazards such as soil erosion, water contamination, pesticide poisoning, falling ground water table, water logging and depletion of biodiversity. Biofertilizers spontaneously activates the microorganisms found in the soil in an effective and eco-friendly way, thereby gaining more importance for utilization in crop production, restoring the soils fertility and protecting it against drought, soil diseases and thus stimulate plant growth. Biofertilizers lead to soil enrichment and are suitable with long-term sustainability. Further, they pose no danger to the environment and can be substituted with chemical fertilizers. The application of bio-fertilizers can minimize the use of chemical fertilizers, decreasing environmental hazards, enhance soil structure and promote agriculture.

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7

Effect of Gibberellic Acid on Seed Germination and Metabolism in *Brassica juncea L.*

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Abstract:
Gibberellic Acid Increases Secondary Metabolite Production in *Brassica juncea* seeds.Gibberellic acid (GA3) is reported to have diverse effects on seed germination and growth of *Brassica juncea*. Therefore, the effects of GA3 on the growth,germination, primary metabolite (protein), and secondary metabolite (chlorophyll) production. Three concentrations of GA3, ranging from 25 ppmto 75ppm, as well as a control, were used for investigation. The moderate GA(3) concentration of 75ppm and the control, resulted in the highest concentrations of germination in both the 7 and 14-day treatments. High shoot and root lengths were observed in the 75ppm concentration of 14-day germinated seedlings. Additionally, carbohydrates,chlorophyll, and protein varied in 25ppm.50ppm,75ppm, and control groups. This study demonstrates that supplementation with GA(3) may be an excellent strategy to optimize the production of secondary metabolites from B.juncea. However, GA(3) is a critical factor.
Keywords: *B.juncea*, GA3, germination, chlorophyll, carbohydrates, and protein.

Introduction:

The plant hormones abscisic acid (ABA), gibberellins (GA), ethylene, brassinosteroids (BR), auxin, cytokinins, and other signaling molecules have profound effects on plant development at vanishingly low concentrations. They are chemical messengers for communication among cells, tissues, and organs of higher plants. Seeds of higher plants contain an embryo surrounded by covering layers and function to ensure the establishment of a new plant generation. Plant growth regulators play an important role in plant development, improving yield, and enhancing the quality of seeds.Low temperature is an abiotic environmental factor that affects plant growth, geographical distribution, and crop yields. Gibberellic acid (GA3) is a PGR that enhances seed germination, growth, stem elongation, photosynthesis, flowering, and cell expansion due to its phytohormonal function[1, 2].Studies have shown that gibberellic acid has the capability to improve growth, flowering, photosynthesis, nutrient transport, and yield of mustard [3, 4].GA3 has been reported to increase seed germination percentage and seedling growth in Cicer arietinum under PEG-induced water stress(Kaur S et al., 1998 and Kaur S et al., 2000). GA3 pre-treatment affected the germination rate, germination potential, hypocotyl length, and radicle length. With an increasing GA3 concentration, these indices first increased and then decreased. For seedling physiology characteristics in hemp, GA3 pretreatment significantly increased the osmotic regulating substances (soluble sugar and soluble protein contents) and the activities of antioxidant enzymes (SOD, superoxide dismutase, and POD, peroxidase), while sharply decreasing the lipid peroxidation (malondialdehyde, MDA) in seedlings grown under PEG-6000-induced drought stress (5).

Indian mustard (Brassica juncea L.) is one of the most important oil seed crop belonging to the family Brassicaceae and the genus Brassica. Among the edible oilseeds

cultivated, mustard ranks second after groundnut in India, and its contribution to the total oilseed acreage and production is 23.7% and 26.0%,respectively. The present study investigates the effect of GA3 on Brassica for studying different parameters, including Such as:

1. Seed germination %. 2. Root shoot length. 3. carbohydrate content. 4. Protein content. 5. Chlorophyll content.

Materials and Methods: For this study, a laboratory experiment was conducted using a single variety of Brassica and varying concentrations of the plant growth hormone, gibberellic acid. The seeds were treated with varying concentrations of 25, 50, 75, and 100 ppm of GA.The seeds of Brassica were soaked in varying concentrations of GA3, 50, 75 and 100ppm. for 18 hours, the seeds were transferred for germination. After germination, plantlets were collected from the soil at 7 and 14-day intervals to study the germination rate, percentage, and secondary metabolites.

OBSERVATION AND RESULT

Effect of GA3 concentrations on the germination of *B.juncea*: In the 7-day time period, both germination and vigor index showed the maximum value in the control group, with 50, and 75 ppm concentrations showing an increase. In the 14-day time period, the germination percentage was highest in the control group, but there was a change in the vigour index, with the 25 ppm having the highest value. Table No. 1. Showing germination and vigor index.

Table No. 1 Effect of GA3 on the germination percentage and vigor index of Brassica

Concentration in ppm	**Treatment duration**	**No. of seed sowed**	**No. of seed germinate**			
			After 7 days of sowing		After 14 days of Sowing	
			Germination	vigor	Germination	vigor
25	18	50	27.00%	3.20	50.00%	17.50
50		50	30.00%	8.81	20.00%	119.51
70		50	31.00%	7.93	31.00%	5.07
control		50	45.00%	17.40	80.00%	17.62

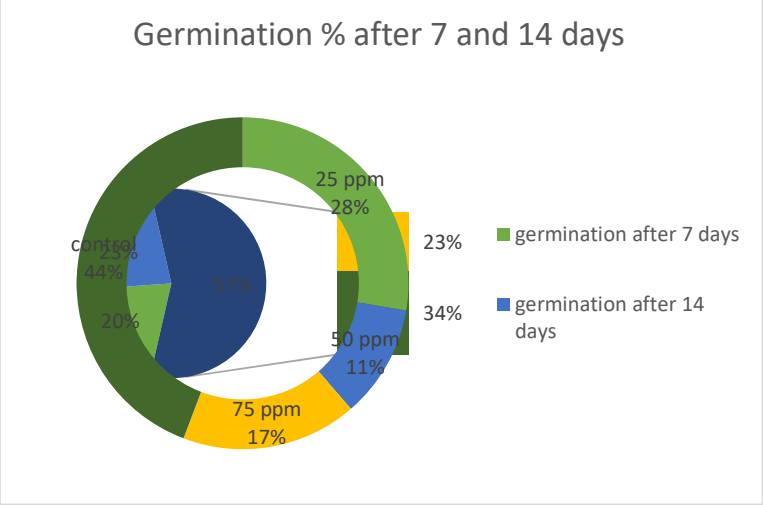

Fig1. Germination percentage of *B.juncea* seeds after 7 and 14 days

Shoot and root length of *B.juncea* after 7 and 14 Days :

Present study revealed that the shoot and root length of Brassica after 7 and 14 days varied. On 7 days germination, the highest root and shoot lengths were observed in 50ppm, which is 2.5cm and 4.2cm, respectively. after 14 days highest root length was observed in 50ppm, which is 4.8cm, and the highest shoot length was observed in 25ppm i.e. 2.1cm. Nearly similar results were observed by (Wareing et al., 1968).The increase in germination may be due to the antagonistic effect of gibberellic acid on germination, and endogenous gibberellins were reported to increase due to soaking (7). The minimum shoot and root lengths were observed in 75 ppm concentration on both the 7th and 14 days.

Table No.2 Shoot and Root lengthon 7 and 14 days

Concentration in PPM	7DAYS		14DAYS	
	Root	**Shoot**	**Root**	**Shoot**
25ppm	3.1cm	**3.5cm**	**2.2 cm**	**2.1cm**
50ppm	**2.5cm**	**4.2 cm**	**4.8 cm**	**5.1cm**
75ppm	**3 cm**	**4.6cm**	**5.4cm**	**5.7cm**

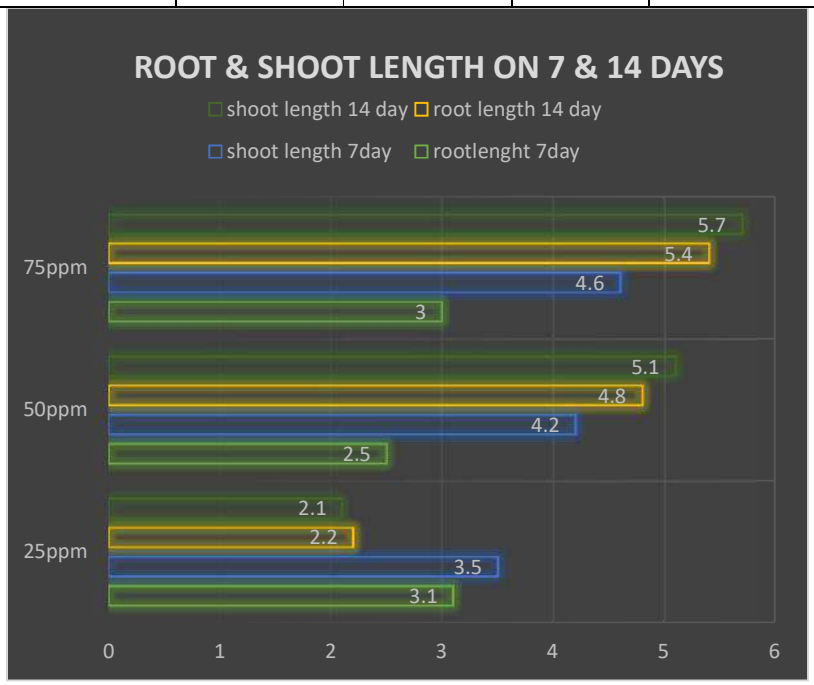

Figure 2: shoot and root lengths on the 7th and 14th days

Effect of GA3 concentration on chlorophyll content of *Brassica juncea* .

The seeds were treated with various concentrations of GA3 for 18 hours and then sown in soil after intervals of 7 and 14 days. Plantlets were collected, and several physiological tests were conducted, including chlorophyll analysis, carbohydrate, and protein tests (8). After 7 and 14 days intervals, chlorophyll estimation was observed at wavelengths 645 nm and 470 nm. And maximum absorbance recorded in 470nm i.e. 1.384nm in 50PPM concentration, which was recorded 1.384nm in control and 652nm on 7 days 0.881. The application of GA3 significantly increased the chlorophyll content compared to the corresponding treatments without GA3 application. The maximum increase in chlorophyll content was recorded by GA3P + GA3FS(Kashif Shahzad, Sadam Hussain *et.al.2021)*

Table No. 3: Estimation of chlorophyll on 7th and 14th days in *B.juncea*

Concentration in PPM	7 days Wavelength			14 days Wavelength		
	470nm	645nm	652nm	470nm	645nm	652nm
Blank	0.001	0.001	0.002	-0.004	-0.005	-0.005
25 PPM	0.836	0.339	0.881	0.678	0.658	0.673
50 PPM	1.384	0.732	1.252	0.674	0.694	0.852
75 PPM	1.165	0.379	1.276	0.924	0.949	0.956

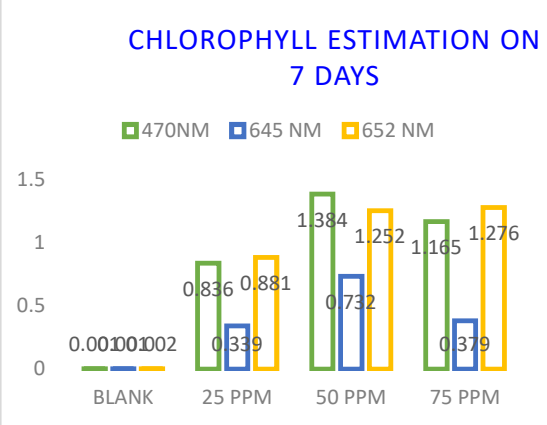

Fig.:3: Chlorophyll Estimation on Day 7 Days

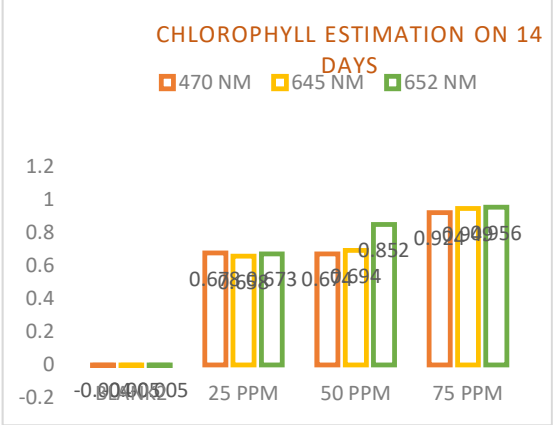

Fig.:4: Chlorophyll Estimation on Day 14 Days

Effect of GA3 on the protein content of Brassica:

Protein estimation was carried out using the standard method by Manickam and Sadashivam . On 7 days after seed germination protein estimation carried out which showing highest protein value in 25 PPM concentration at 645 nm i.e. 0.339then after in 50PPM that is 0.732 at 645nm and least value are observed in blank and 25 PPm and 75PPm i.e. 1.165and 0.379 Fayaz Asad1 , Naveen Dilawaret.al 2022 observed that Maximum proline content was observed for treatment 20ppm NaCl+20ppm GA3, while sugar records showed little variations between the treatments whereas higher value recorded for 10ppm NaCl+40ppm GA3. The lower value of Asad et al. 646 sugar content was recorded in 10ppm NaCl+40ppm GA3. After 14 days the maximum protein amount was found in 25 PPM and 75 PPM, with at 645 nm and 470 nm and absorbance are0.339and 1.165 and lowest readings were observed in blank(control) and 652nm 0.002 The total soluble protein significantly increased under GA3 application, with the maximum values recorded for GA3P + GA3FS indicating that the application of GA3 as seed priming and foliar spray effectively enhances soluble protein under salinity stress(Kashif Shahzad, Sadam Hussain *et.al. 2021).*

Table no.4: estimation of Chlorophyll from B.juncea on 7th and 14th days

Concentration in ppm	Treatment duration	Wave length of 7 days germinated seed				Wave length of 14days germinated seed			
		470 nm	645 nm	652 nm	663 nm	470 nm	645 nm	652 nm	663 nm
Blank	18 hours	0.001	0.001	0.001	0.002	-0.001	-0.001	-0.002	0.002
25 ppm		0.836	0.339	0.881	1.114	-0.001	-2.444	2.500	2.570
50 ppm		1.384	0.732	1.252	2.108	0.117	0.917	0.942	0.112
70 ppm		1.165	0.379	1.276	1.31	1.309	0.851	1.260	2.109

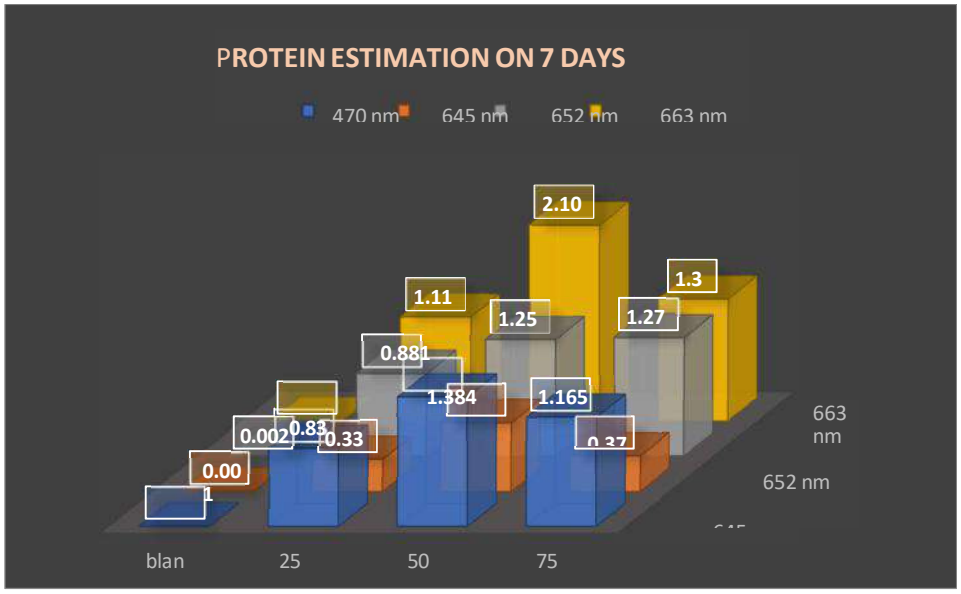

Fig:5 -Protein estimation on 7th day of germination

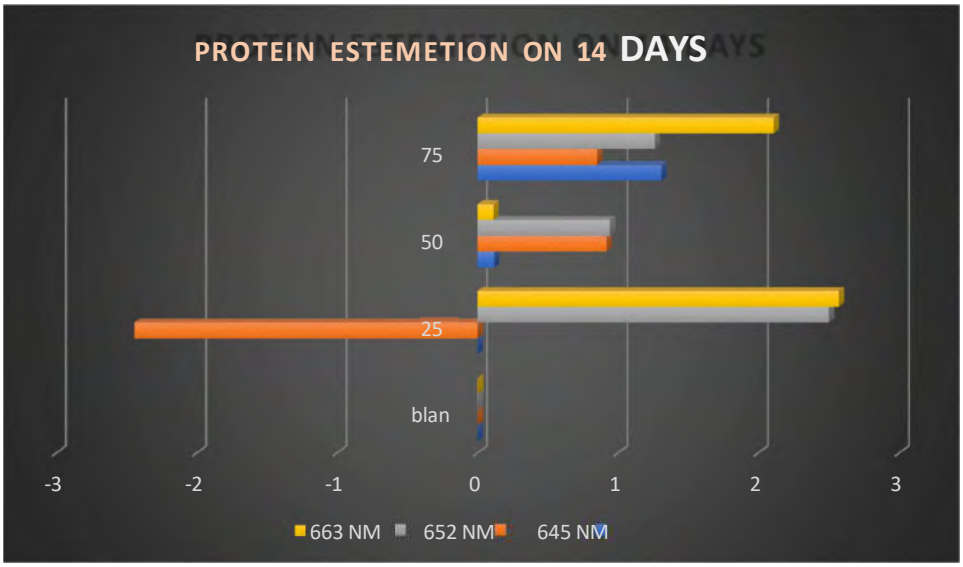

Fig: 6- Protein estimation on 14th day of germination

Effect of GA3 concentration on the carbohydrate content of *B.juncea*

In this investigation, seeds were soaked in GA3 for 18 hours. After germination, the estimation of carbohydrates was carried out. Absorbance was recorded at 630nm on the 7th and 14th days germinated seedlings by Manickam and Sadashivam. The maximum absorbance was recorded on concentration of 25 ppm, i.e. 0.954nm and the lowest absorbance was recorded in 75ppm i.e. 0.516nm,on 14 days germinated seed, the maximum absorbance was recorded in 50ppm concentration i.e. 0.517nm and the lowest absorbance was 0.419nm in 25ppm.(Abd El–Monem, A.A. (2007);El–Bassiouny, H. M. S.; Mostafa *et.al* 2008 and Hassanein R.A *et.al*. 2008) observed that The highest values of seeds mineral nutrients, carbohydrates, and total crude protein contents were gained by foliar spray with arginine at 300 ppm. These results could be supported by the results obtained by[8,9,10] who indicated that arginine is the most effective compound for increasing soluble carbohydrates, polysaccharides, total carbohydrates,

proline, total amino acid and protein contents of wheat plants and grains under normal or stressed conditions.

Table no.5 : Estimation of carbohydrates on 7th days and 14th days

Concentration in ppm	Treatment duration	Carbohydrate estimaton	
		On 7 days at 630nm	On 14 days at 630nm
blank	18 hr	-0.003nm	-0.001nm
25		0.954nm	0.419nm
50		0.692nm	0.517nm
75		0.634nm	0.516nm

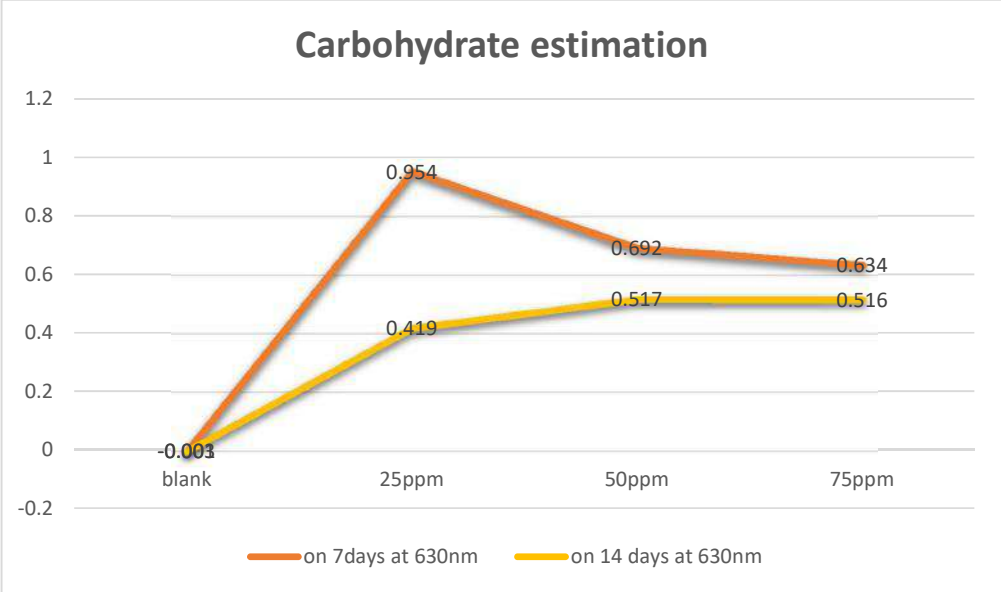

Fig: 7- Carbohydrate estimation on 7th and 14th day of germination.

Conclusion: Above investigation was caried out using phtohormones and observed therer effect on plant growth and phytocompound like carbohydrates, protein and chlorophyll and record was observed on 7 and 14 days interval period and it conclude that GA3 shows variation in growth and amount of metabolites.

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8

Identification of Agricultural Practices Related to Indigenous Grains in Tribals of Vidarbha Region

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Research Scholor – Home Science

Indigenous Technical Knowledge (ITK) is local specific knowledge and set of practices in agriculture, natural resource management, health and educational development generated and preserved by people and farmers and confirm for its stability over centuries. According to Michelle Warren (1993), indigenous knowledge is "Local Knowledge" that is unique to a given culture or society. It is information base facilitating communication among people coming from different backgrounds. Search knowledge is passed on from generation to generation in many societies by word of mouth. It is the acquired by local people through accumulation of experience, informal experiments and intimate understanding of environment in a given culture (Anonymous 1994).

The largest concentration of tribal is found in African continent. India accounts for nearly 2 to 4 % of the world's population. There are 314 tribal communities in India known as different names such as Adivasi (the original inhabitants.), Vannya Jati (caste of forest) , Adim jati (primitive people) etc. Total population of tribal in India, according to census 2011 which 7.59 crores and their proportion with total population of India was 8.01 per cent out of which 9.27 per cent of the tribal were in Maharashtra state. The eastern part of Vidharbha(Gondwana Region) inhibitates the major tribal population in the state. The area known as Melghat is comprises of mainly two tehsil of Amaravati district namely 1)Dharni tahsil 2) Chikhaldara.

Indigenous knowledge would help us to understand the concept the element of ustainability in integrated with the modern information system the efficient resource management. It can be used for the benefit of the community to provide information offer solution to common problem within the community. As it is seen that tribal, who's from sizable proportion of the farming population and the agriculture businesses on which they are mainly dependent for food on farming and forest. Efforts are being made worldwide to identify the Indigenous Technological knowledge. The indigenous agriculture practice is influence the current farming practice. The study would be helpful the planners and policy makers in Planning and implementing some scheme for the tribal farmer to improve upon their farming Status. It May divert the attention of decision maker and policy maker to the role that small scale agriculture producer can play significantly in achieving of national food self suffciency.

Sr. No	Indigenous Practices	Reasons for adoption of indigenous and agricultural practices.	Scientific reason
1.	Collecting and burning crop residues and farm waste the field crop	I .kill the microorganism harmful to the crop ii. destroy the weed seeds iii. ash acts as the good fertilizer	Farm waste is the source of various pest and diseases. Burning the farm waste destroy the harmful organism in the soil. Ash also contains potassium (K) and nitrogen (N) which improves the yield of crop. It is important organic manure supplement.
2.	Following the Rab method while raising the seedlings of paddy in nursery.	i. It adds more organic matter in the soil ii. It improves the soil fertility, productivity and water holding capacity	Dung of animals and farm waste is the source of egg. Pupae of insect pests. Burning the farm waste and dung kill the insect pest and ads more organic matter in the soil. It improves the quality and quantity of crop.
3.	For control of aphids	i. It is cheaply available	Ash acts as contact poison if used at heavy

	and jassids in the paddy field ash is used i.e. for controlling one acre area 15 to 20 kg of ash is	ii. ash is having certain characters which prevents the pests to cause damage	rate it controls the aphids/jassids
	dusted		
4.	Spraying mechanically the leaves extract of castor and mahua flower on paddy crop.	it is controlling aphids and jassids due to alcoholic extract	Mahua flowers contain alcohol which killed aphids and jassids, castor seeds contained ricine, a toxic alkaloid , which may kill aphids and jassids
5.	Beating drum in the standing crop of paddy for scaring birds	Prevents the birds to cause damage due to noise	
6.	Pelting stone with the help of gophan (A device made up of coir string for pelting the stone pieces with speed for scaring birds)	Birds are driven out from the fields and hence damage to crop is prevents	
7.	Fixing effigy in the centre of field for scaring animals	Prevents birds from causing damage	
8.	Making typical noise with the help of bamboo stick for scaring birds	This is traditional and having low cost practice	
9.	Harvesting matured crop and threshing by bullock cart	Keep the paddy fodder in good quality	
10.	Winnowing the threshold production in Morning or Evening by Facing north- south direction	Wind blow fast at morning and evening	Wind blows east to west of direction
11.	Storing paddy seeds in dholi or dhindwa and placing ash and neem leaves in the bin	i. it save the germination percentage of seed as such ii. ash is having certain characters which prevents the pest to cause damage iii. Neem extract is better in taste and also poisonous so that pest is prevented	A grain cellar i.e. dholi or dhindwa are the non cash structure available for the farmers which don not required repairs and give satisfactory storage of grain for longer length of time hence they are use. Ash cause physical injury to insect.

Major Indigenous crops

1. Jagni

Jagni is one of the most indigenous grains of tribal people.its rich in fibre also jagni oil has its own medicinal important value**.**

Sowing: Jagni is sown in mid of August to first week of September

-Method of sowing is by throwing with hands. **Maturity**: -

After sowing flower comes in one month. Then after 15-20 days from flowering plant is harvested. **Uses**

Uses were reported by the tribal farmers are listed below:

- Nourishment of child jagni oil is used
- Decreased cholesterol level
- Rich is fibre.
- 2-3 drops are used is eyes to removal of dust.

- Oil is used in nose and ear drops.
- Oil is used for cooking purpose.
- After crushing that grain material remains (bhukti) in that bhukti by adding salt + chill + coriander chutney is prepared, which is nutritious.

Yield:

Price:

In 1 acre 7.8 quintal is obtained. One kg oil is obtained from four kg of oil seeds.
Price of oil is 150 per litre and cost of grain is 200-250 Rs kg.

Storage:

Stored is kothi which is made up of soil.

2. Kutki

Kutki is use as staple food by many tribals. This grain has important value in ritual. Useful for pregnant woman

Sowing: sowing is done in last or ending of rainy season.

- Midnight of august to first week of September
- Sowing is done by broadcasting method.

Harvesting:

Harvesting is done by sickle manually. After cutting plant they are allowed to crush in Dawan (assembly in which ox is tied and grain is crushed under the legs of ox.) after harvesting the small grain they are taken into dalan kendra for removing of outer cover of grain.

Maturity: crop matures in 3 month after showing and ready to harvest. Height of plant is 1 to 2 feet.

Use:

Kutaki has its different used as stated below:

- Use as staple food like rice. This tribal consider as hot food hence Use to cure cold and cough. So people are avoiding eating it.
- Pej is given to pregnant women. Use to cure cold and cough.
- Kutki have its own ritual important value in ritual programmes especially in nawas the sweet porriage is prepared and offer as scarifies to God.

Yield:

Price:

In One acre four-five quintal grain is obtained.
Price is 20 to 25 Rs/kg.

Storage:

Stored is kothi which is made up of soil or in dholi.

3. SAWA

Sawa is one of best useful grain among all indigenous grains. Helps in digestion and it has important role in curing of cough, cold.

Sowing: Sown in august and first week of September.

Harvesting: Done by dawan method. Crop matured in 3 months. Harvesting is done manually by sickle with Labour.

Maturity: After sowing crop matured in 90 days. Maturity sometimes requires more water.

Use:

- Given to patients suffer from stomach problems.
- Used against digestion problem.
- Use to cure cough.

Yield:

In one acre four-five quintal grain is obtained.

Storage:
After harvesting grain is stored in field and covered it with grass to prevent it from rain. Then stored in kothi.

4. Kodo

Kodo is nutritious in value and one of the best use ot kodo is used for making chapatti. Useful as a source of multivitamins.

Sowing: sown in august and mid of September after sufficient rainfall.

Harvesting:
Done manually by labours. After fully maturity it seems like wheat.

Use:

- The grain is used for multipurpose in having high nutritious value given below:
- Hard to digest.
- For making nutritious chapatti, rich in iron, healthy to eating for all age groups.
- Do not give to patients suffering from diseases like loose motion, constipation, and dysentery

Yield:
From one acre ten quintal grain is obtained under best practices.

Storage:

- Stored in kothi.
- In farm optimum moisture content at storage should be 10 to 12 per cent.
- To prevent from rain garai structure is made.

Fig. 1. Grains storage structure (Dholi)

Fig. 2. Grains storage structure (Dhindwa)

Different reasons for adoption of indigenous agricultural practices were found above. The important reason behind adoption of indigenous practices Were mainly traditional and experienced based. The experiences transmitted from generation to generation have made the respondents to adopt indigenous agriculture practices. The Other reasons were are mainly low cost or no cost of the practices and it's easy availability in surrounding area.

The study suggests that the systematic effort on the path of extension agency are required to promote indigenous agricultural practices by imparting knowledge to them in the area under the guidance of subject matter specialists. This may be accomplished by organization campus and field visits to the farmers demonstration plot and field trials. The valuable indigenous knowledge may be provided through publication of department of Agriculture and University.

9

In Vitro Anti-diabetic Activity of Different Extracts of *Moringa oleifera* Leaves-Glucose Uptake by Yeast Cells Method

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Introduction:

Medicinal plants are frequently used as raw materials for extraction of active ingredients used in the synthesis of different drugs. There are many diseases for which herbal medicine are used to avoid side effect of modern medicine. Interestingly, it is estimated that more than 25% of modern medicines are directly or indirectly derived from plants (Singh Ayodhya, *et al.,* 2010).

Diabetes mellitus is a group of metabolic alterations characterized by hyperglycemia resulting from defects in insulin, secretion action or both. It has already been established that chronic hyperglycemia of diabetes is associated with long term damage, dysfunction and eventually the failure of organs, especially the eyes, kidneys, nerves, heart and blood vessels (Hung, T.H.W., *et al.,* 2005). India is also known as diabetes capital of the world and affects mainly rural and urban people. The frequency of diabetes in urban area is approximately 6 times more as compared to rural area (Verma S. *et al.,* 2018).

Moringa oleifera is a fast-growing, drought-resistant tree of the family Moringaceae, native to tropical and subtropical regions of South Asia. Common names include moringa, drumstick tree (due to long, slender, triangular seed-pods) and horseradish tree (due to the taste of the roots, which resembles horseradish).

The mechanism of glucose transport across the yeast cell membrane has been receiving attention as *in vitro* screening method for hypoglycemic effect of various compounds or medicinal plants. The main objective of present study is to evaluate the anti-diabetic activity of *Moringa oleifera* leaves by glucose uptake by yeast cell using different extracts.

Material and Method:

Step I: Preparation of plants leaves stock solution

10 mg of selected plant leaves extracts were taken and dissolved in 1 ml of Dimethylsulphoxide (DMSO), which was used as stock solution. From this stock solution, different concentration viz. 50, 100, 150, 200, 250 µg/ml were prepared (Chaudhari M.G. *et al.,* 2013).

Step II: *In-vitro* Antidiabetic Activity

Glucose uptake by Yeast cells

Yeast suspension was prepared by repeated washing, by centrifugation at 3,000×g for 5 min in distilled water until the supernatant fluids were clear (Cirillo V.P.,1962). 10% (v/v) suspension was prepared with the supernatant fluid. 1mL of glucose solution (5, 10 and 20 mM) was added to various concentrations of ethanolic and petroleum ether extracts (50,100,150,200 and 250 µg/ml) and incubated for 10min at 37 °C.

Reaction was started by adding 100 µl of yeast suspension, vortex and further incubated at 37 °C for 60 min. After 60 min, the reaction mixture was centrifuged (2,500×g, 5 min) and glucose was estimated in the supernatant. The content of glucose in the supernatant was estimated by DNSA method. The absorbance of the colour mixture was recorded at 540 nm.

Metronidazole was taken as standard drug. The percentage increase in glucose uptake by yeast cells was calculated using the following formula (Gupta D *et al.,* 2012).

$$\text{Increase in glucose uptake (\%)} = \frac{\text{Abs sample} - \text{Abs control}}{\text{Abs sample}} \times 100$$

Where, Abs control is the absorbance of the control reaction (containing all reagents except the test sample), and Abs sample is the absorbance of the test sample. All the experiments were carried out in triplicates.

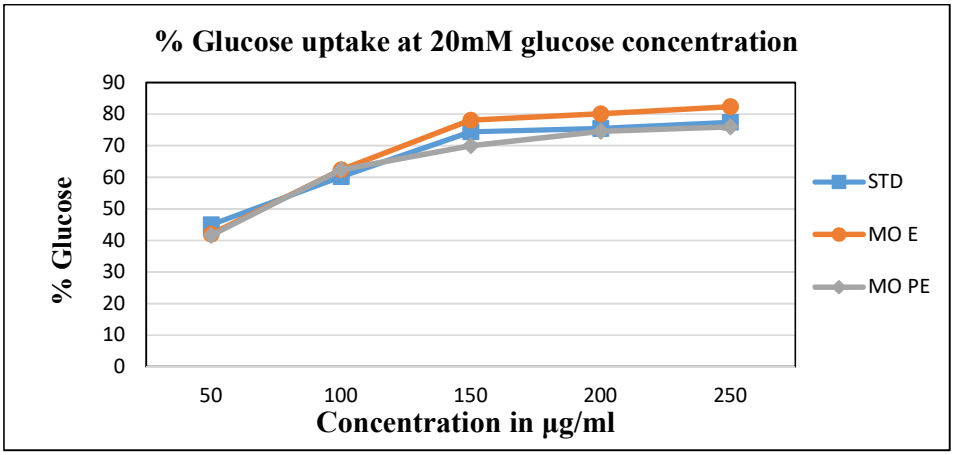
Result and Discussion:

Effect of ethanol and petroleum ether extracts of *Moringa oleifera* leaves on Glucose uptake by yeast cells activity were studied.

Table No.1: Effect of different extracts of *Moringa oleifera* on percent glucose uptake by yeast cells at 20mM glucose concentration

Concentration µg/ml	STD	MO E	MO PE
50	44.9±0.60	42.0±0.40	41.6±0.55
100	60.2±0.12	62.4±0.46	62.4±0.43
150	74.4±0.28	78.1±0.42	70.0±0.23
200	75.4±0.40	80.1±0.23	74.6±0.36
250	77.4±0.43	82.4±0.31	76.0±0.52

STD: Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves

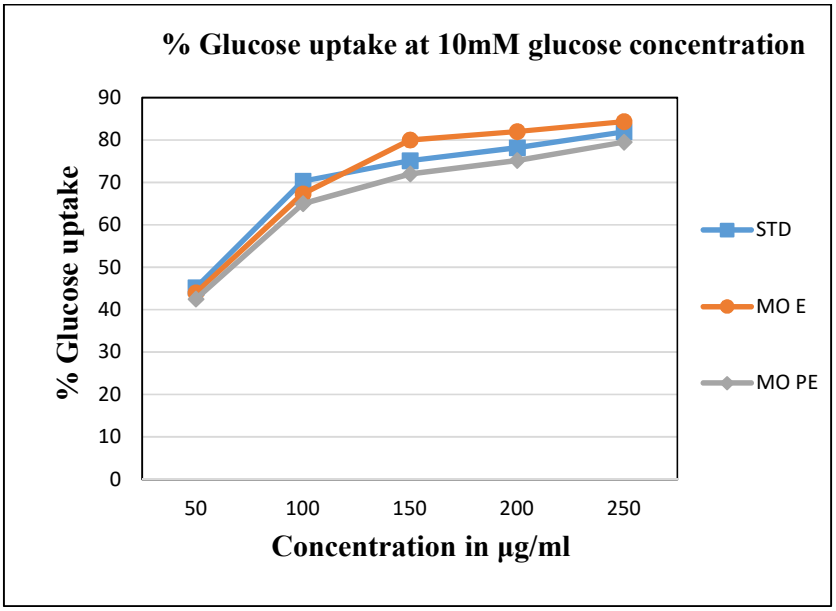


STD : Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves

Table No.2: Effect of different extracts of *Moringa oleifera* on percent glucose uptake by yeast cells at 10mM glucose concentration

Concentration µg/ml	STD	MO E	MO PE
50	45.2±0.26	44.0±0.22	42.5±0.51
100	70.3±0.49	67.3±0.40	65.0±0.34
150	75.1±0.20	80.0±0.72	72.0±0.38
200	78.2±0.26	82.0±0.40	75.2±0.34
250	81.9±0.46	84.3±0.37	79.5±0.25

STD : Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves

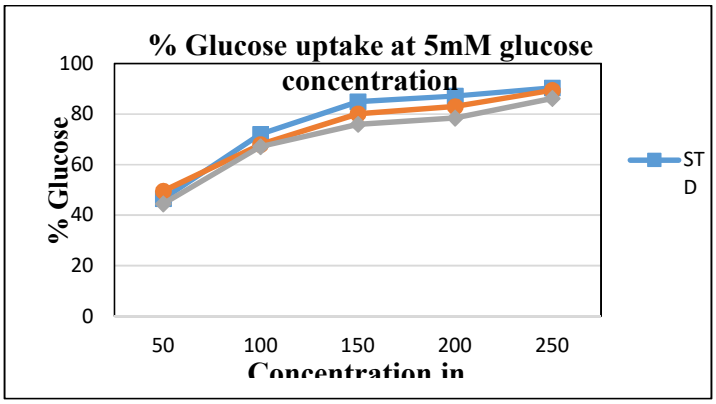


STD : Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves.

Table No.3: Effect of different extracts of *Moringa oleifera* on percent glucose uptake by yeast cells at 5mM glucose concentration.

Concentration µg/ml	STD	MO E	MO PE
50	46.6±0.23	49.5±0.42	44.5±0.46
100	72.0±0.11	68.0±0.52	67.2±0.40
150	84.9±0.08	80.1±0.27	76.0±0.29
200	87.2±0.23	83.0±0.31	78.5±0.31
250	90.4±0.42	89.5±0.28	86.2±0.39

STD: Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves



STD : Standard, MO (E): Ethanolic extract of *Moringa oleifera* leaves and MO (PE): Petroleum ether extract of *Moringa oleifera* leaves

The metabolizable and non-metabolizable sugars, and glycosides are transported across the cell membrane by facilitated diffusion (Cirillo VP, 1962). Facilitated carriers are specific carriers that transport solutes down the concentration gradient highlighting that the effective transport is only attained if there is removal of intracellular glucose. Hence, glucose transport occurs only if the intracellular glucose is effectively reduced or utilized (Revathi Pitchaipillai *et al.,* 2016, Teusink *et al*., 1998).

The present data, suggests that ethanol and petroleum ether extract of *Moringa oleifera* leaves were capable of enhancing glucose uptake effectively, which in turn suggests that they are capable of enhancing effective glucose utilization, thereby controlling blood glucose level. From Table No.1, 2 and 3 it was revealed that there is increase in uptake of glucose as the concentrations of extracts increases. However, an inverse relationship to the molar concentration of the glucose was observed among the 5mM, 10mM and 20mM for the same concentration of the extracts. Hence it can be said that lower the concentration of glucose in the solution higher the uptake of glucose.

Conclusion:

From present data, it can be said that ethanol and petroleum ether extracts of *Moringa oleifera* leaves were capable of enhancing glucose uptake effectively and enhancing effective glucose utilization, thereby controlling blood glucose level. However further comprehensive chemical and pharmacological investigation should be carried out to isolate the active compounds.

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Investigating the In Vitro Regeneration Potential of Commercial Cultivars of *Brassica*

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***Abstract*:**
In vitro regeneration is a pre-requisite for developing transgenic plants through tissue culture-based genetic engineering approaches. The genus *Brassica* necessitate the identification of a set of regeneration conditions for a genotype, which can be reliably used in transformation experiments. In this study, we evaluated the morphogenesis potential of four commercial cultivars. The explants namely cotyledons, hypocotyls, petioles and roots on three different *Brassica* regeneration protocols, BRP-I, -II and -III. The regeneration efficiency was observed in the range of 6–73%, 4–79.3%, 0–50.6%, and 0–42.6% from cotyledons, petioles, hypocotyls, and roots, respectively, whereas, the regeneration response in terms of average shoots per explant was found to be 0.76–10.9, 0.2–3.2, 0–3.4 and 0–2.7 from these explants. its regeneration frequency from cotyledons was up to 7.5-fold higher on BRP-I, while it produced up to 21.9-fold more shoots per explant. Our data show that the explant has strong influence on the regeneration response, ranging from 24% to 92%. While the growth of commercial cultivars was least elected by the regeneration conditions provided, the erect on Westar was twice that of the commercial cultivars. Inhibit the growth of untransformed cells for these cultivars. successfully grown to maturity within 16–18 weeks, with no altered phenotype noted and normal seed yields obtained. Therefore, the commercial variety, Aari canola, would be a good candidate for future genetic transformation studies.

Keywords: in vitro regeneration; tissue culture; commercial cultivars; Brassica Sp.

Introduction:

Brassica, from the family Brassicaceae, is an economically important genus. It includes several species that are often used as oilseed crops, vegetables, fodder crops as well as condiments. Brassica oilseed varieties producing oil low in anti-nutritive aliphatic glycosylates and acid as well as rich in unsaturated fatty acids are generally termed as ‘canola’ Conventionally, the term ‘canola’ was more often used for *B. napus* but now some canola quality varieties of *B. rapa* and *B. juncea* are also available. The development of stress tolerant *Brassica* is possible by transferring genes from the plant species that are adapted to harsh environmental conditions. These species present a rich reservoir of the traits that enable them to grow under stressful conditions. However, transferring these traits to salt or drought sensitive crops is only possible by genetic transformation, as they cannot be cross bred through conventional breeding approaches for example, a plant species must be responsive to in vitro regeneration protocols, and a robust regeneration system is one of the key pre-requisites for successful genetic transformation. Several indigenous Brassica varieties developed locally have canola characteristics. Being stress-sensitive, these varieties are unable to grow on marginal lands. Although transformation of *Brassica* species has been reported in several studies, several *Brassica* genotypes remain recalcitrant to

genetic transformation. Several factors including susceptibility to Agrobacterium infection, choice of explant and tissue culture conditions mainly responsible for these variations have been identified. These factors vary from genotype to genotype, indicating a strong genetic control on in vitro regeneration and transformation of Brassica genotypes. The information generated in this study will be useful for developing stress-resilient *Brassica* varieties by directly transforming the commercial cultivars.

Materials and Methods:

Plant Material and Growth Conditions Five different cultivars (four local and one model) belonging to *Brassica napus* and *B. juncea* were obtained from different

Table 1. List of cultivars with their sources used in this study.

Sr. No.	Cultivar Species	Source
1.	*Brassica juncea*	oil seeds are used
2.	*Brassica napus*	oil seeds are used
3.	*Brassica napus*	oil seeds are used
4.	*Brassica napus*	oil seed are used
5.	*Brassica napus*	oil seed are used
6.	*Brassica napus*	oil seed are used

- **Sterilization and Sowing**

Seeds were stratified at 4**°C** for 48 h before sowing to ensure uniform germination. Seeds were surface sterilized by immersing in 100% ethanol for 2 min followed by immersing in commercial bleach (sodium hypochlorite solution) containing 4–6% available chlorine plus 2–3 drops of 10% between 20 for 10 min. The seeds were then rinsed four times with sterile distilled water in a laminar flow and placed on sterile filter paper to dry and germinated on seed germination media (MS salts, 3% sucrose, 1 mg/L pyridoxine, 1 mg/L nicotinic acid, 10 mg/L thiamine-HCl, 100 mg/L Myo-inositol, 4 g/L phytagel, pH 5.7–5.8) at a density of 20 seeds per 90 30 mm petri dish in a growth room maintained at 23**°C** at 16 h light/8 h dark cycles under a light intensity of 50 mol m2 s1 provided by cool fluorescent bulbs.

- **Explant Isolation and Shoot Induction**

Four different explants intact cotyledons with approximately 2 mm of the petiole, hypocotyls (Approximately 3–4 mm), petioles (approximately 2 mm, with the 'leaf of the cotyledon removed', and roots (3–4 mm) sections were subjected to in vitro regeneration conditions. Four-day-old seedlings were used for explant isolation. A pair of long, sterile forceps was used to remove the seedlings from the germination media and placed in sterile petri dishes. Explants were cut and separated using a sharp scalpel blade and transferred to either shoot induction media (SIM) or callus induction media (CIM) (Table 2 for composition). Cotyledons were excised with a sharp scalpel blade with 2 mm petiole avoiding any part of the meristem. Explants were placed on the medium in such a way that the petiole was embedded in the media and the cotyledonary lamella was clear of the media. 10 explants were established on each plate and transferred to a growth room at 23**°C** under scattered light. Cultures were transferred to fresh medium every two weeks. The explants were subjected to regeneration under three different regeneration protocols termed Brassica Regeneration Protocols, BRP-I , BRP-II and BRP-III

Table 2. Composition of different mediums used in the study.

Reagent	BRP-I		BRP-II			BRP-III			
	SIM	GRM	CIM RIM		SIM	CIM	SIM	SOM	RI
MS salts (mg/L)	4.43		4.43	4.43	4.43	4.43	4.43	4.43	2.215
Gamborg's salts (mg/L)		3.1				4.43	4.43	4.43	2.215
Sucrose (mg/L)	30	10	30	30	30	20	20	20	10
Vitamin Stock (ml/L	1	1	1	1	1	1	1	1	1
Phytagel (g/L)	4	4	4	4	4	4	4	4	4
BAP (mg/L)	2			6	1	0.75		3	0.00125
NAA (mg/L)						0.1		0.2	0.2
IBA(mg/L)						1			
AgNO3 (mg/L)								5	5
GA3 (mg/L)								0.01	0.01
CaCl2 (mg/L)						435	435	435	
KI (mg/L)						05	0.75	0.75	0.75
Adenine hemisulphate (mg/L)						40			
PVP 40,000 (mg/L)						500			

BRP, Brassica regeneration protocol; SIM, shoot induction medium; GRM, Gamborg's rooting medium; CIM, Callus induction medium; RIM, Root induction medium; SOM, Shoot outgrowth medium.

Results and Discussion:

- **Shoot Regeneration from Cotyledons**

All the explants, including cotyledons, normally regenerated multiple shoots and occasionally a single shoot per explant. Regenerates formed from the cut end of the 2 mm petiole attached to the cotyledon. The highest regeneration efficiency was observed for Aari canola on all three protocols with 70.6% on BRP-I, 69.3% on BRP-II, and 73.3% on BRP-III Faisal canola showed the lowest regeneration efficiency on BRP-I (6.0%) which showed the lowest regeneration on BRP-II and BRP-III with 13.3% and 10.0% efficiency, respectively. The regeneration efficiency of on BRP-I, is 3.8-fold higher on BRP-II and 2.5-fold higher on BRP-III. In terms of total shoots regenerated, the highest shoot formation was observed. All the three protocols with 548 shoots on BRP-I (average 10.96 shoots/explant), 436.3 on BRP-II (8.72 shoots/explant) and 366.3 on BRP-III (7.3 shoots/explant) from a total of 50 explants. The least regeneration responsive was (25 shoots/50 explants; 0.50/explant) BRP-I, and BRP-II (38.3 shoots/50 explants; 0.76 shoots/explant) and BRP-III (25 shoots/50 explants; 0.50/explant). In terms of an average number of shoots per explant, it was in the range of 0.50–10.9, 0.76–8.7 and 0.5–7.3 from cotyledons on BRP-I BRP-II and BRP-III, respectively. Overall, the regeneration response of Aari canola, in terms of total shoots formation, was 21.9-fold higher than Westar on BRP-I, 7.0-fold higher on BRP-II and 2.3-fold higher on BRP-III.

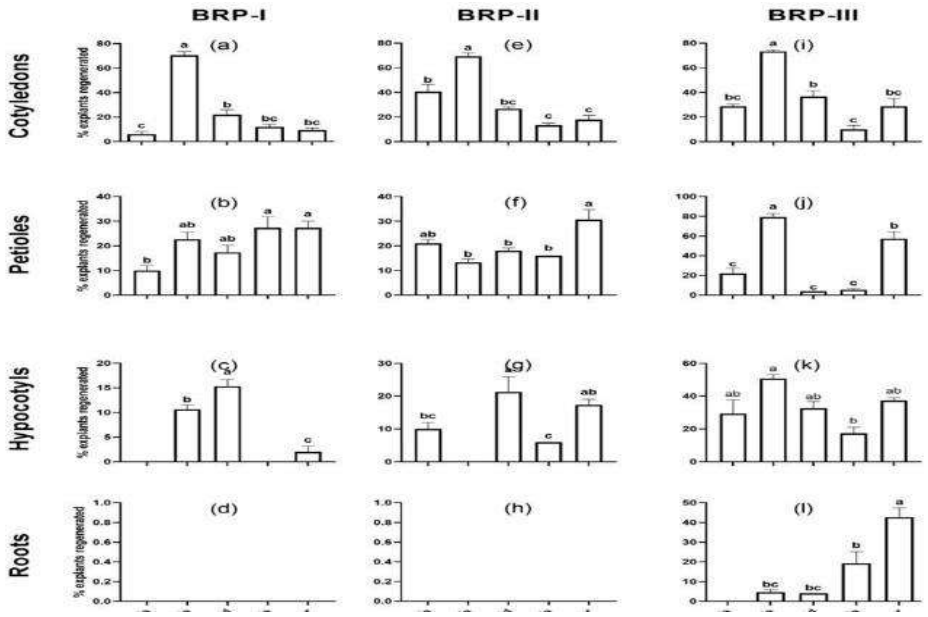

Figure 1. Regeneration efficiency of five Brassica cultivars

- **Shoot Regeneration from Detached Petioles**

Petiole explants regenerated shoots along the middle rib after 3 weeks on SIM. The highest regeneration efficiency from detached petioles was observed for on BRP-III (79.3%), BRP-I and BRP-II with a regeneration of 27.3% and 30.6%, respectively (Figure 1b,f,j). The lowest regeneration efficiency was observed for Faisal canola on BRP-I (10.0%), followed by BRP-II (13.3%), and Nifa Gold on BRP-III (4.0%). The regeneration efficiency and 2.3-fold lower on BRP I and BRP-II, respectively, while 1.3-fold higher on BRP-III. The highest number of shoots on BRP-I and BRP-II with 137 and 164.33 shoots from 50 explants the highest number of shoots (129 shoots) on BRP-II (Figure 2b,f,j) least number of shoots on BRP-I (14.3 shoots/50 explants) and BRP-III (15.6 shoots) while it was Westar, which showed the lowest response on BRP-II with 24.6 shoots from 50 explants. The average number of shoots per explant was in the range of 0.2–2.7, 0.4–2.5 and 0.3–3.2 on BRP-I, BRP-II and BRP-III, respectively. The number of shoots produced by Aari canola was 1.7-fold, 5.2-fold and 1.0-fold on BRP-I, BRP-II and BRP-III respectively.

- **Shoot Regeneration from Hypocotyls**

Regeneration from hypocotyl segments usually started after 3 weeks on SIM although callus formation started in the second week. The upper end of the hypocotyl cut 2 mm below the epicotyl region showed more swelling and calli formation, ultimately producing higher number of shoots as compared to the other end of the explant. Shoot regeneration was rarely observed from the middle rib portion. The highest regeneration efficiency from hypocotyls was observed on BRP-III (50.6%) followed by Brassica with 37.3% and 32.6%, respectively (Figure 1k). The highest regeneration efficiency on BRP-I and BRP-II was of Brassica with 15.3% and 21.3%, respectively, as compared to the other cultivars, suggesting the suitability of these protocols for Brassica for obtaining regeneration from hypocotyls sections. it was 5.3-fold higher on BRP-I, 17.3-fold lower on BRP-II and 1.3-fold higher on BRP-III. The highest number of shoots on BRP-I (60.3 shoots from 50 explants) and BRP-II (170 shoots/50 explants) while the highest shoots on BRP-III (121 shoots/50 explants) (Figure

2c, g, k). The hypocotyl segments of respond to BRP-II, BRP I. Regeneration on all three protocols, with highest shoot count on BRP-III (82 shoots/50 explants), followed by BRP-II with 61.6 shoots and BRP-I with 15 shoots. The average number of shoots generated from hypocotyls were in the range of 0–1.2, 0–3.4, and 0.4–2.4 per explant on BRP-I, BRP-II and BRP-III, respectively.

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- **Effect of Explant, Regeneration Conditions, and Their Interaction on In Vitro Regeneration**

The data obtained was analyzed for the significance and the degree of erect of the explant type, the growth regimes, as well as their interaction on the regeneration of all the cultivars. A standard analysis of variance was applied to analyze the regeneration data. Table 3 shows the summary of the analyses. The explant had a highly significant effect on regeneration (p _0.0001). The erect regeneration conditions were less significant. Other cultivars (p _ 0.001), whereas, it was highly significant (p _ 0.0001). The effect of explant regeneration conditions was statistically non-significant. The erect of replication was non-significant on all cultivars except. In terms of the percentage contribution of these erects to the regeneration efficiency, the erect of explant type was highest from all the other factors except (Table 4). It was highest in (91.87%) while lowest in (24.13%). The effect of regeneration conditions was much more pronounced on regeneration (57.21%) compared to that of the commercial varieties. It was highest for Brassica (27.12%) and Brassica (31.44%) while lowest for (4.95%).

Table 3. Mean square and significance levels from analysis of variance of data from the regeneration of five *Brassica* cultivars.

Source of Variation	DF	*Brassica* I	*Brassica* II	*Brassica* III	*Brassica* IV	*Brassica* V
Explant (E)	3	6552 ****	368,487 ****	26,280 ***	6644 ****	10,035 ****
Regeneration Conditions (RC)	2	8297 ***	11,116 ***	1026 NS	8261 ****	35,692 **
Interaction (ExRC)	6	2540 ****	9938****	1762 NS	3202 ****	2323 **
Replicate	8	115.9 NS	795.6 NS	891.1 NS	318.7 ***	302.9 NS
Residual Error	16	236.1	596.7	769.1	180.1	432.5

*, **, ***, **** Significant at p = 0.05, 0.01, 0.001 and 0.0001, respectively; NS,non-significant

Table 4. Percent contribution of explant type, growth conditions, and their interaction on the regeneration frequencies of five *Brassica* cultivars.

Variations	**Percent Contribution of a Treatment to the Total Experimental**				
Source of Variation	**Brassica I**	**Brassica II**	**Brassica III**	**Brassica IV**	**Brassica**
Explant Type (E	34.98	91.87	71.09	32.62	24.13

Regeneration Conditions (RC)	29.53	1.848	1.850	27.04	57.21
Interaction (E x RC)	27.12	4.956	9.533	31.44	11.17
Replicate	1.650	0.529	6.428	4.173	1.942
Residual Error	6.720	0.797	11.09	4.727	5.548
Total	100.0	100.0	100.0	100.0	100.0

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Study of Taxonomy *of Pistia stratiotes (*Water lettuce*)* and their Uses.

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Abstract :

Pistia stratiotes has a common name Water lettuce, is a small evergreen perennial plant with feathery roots. Free floating in lake, ponds etc, the plants forms rosettes of leaves up to 10 cm wide and 6cm tall. The plant can spread quickly especially in still water , to form quite extensive clumps . The plant is sometimes used locally for food , but only usually ,where nothing better is available.

Pistia stratiotes has a range of medicinal applications and is also used as a source of organic matter and to remove toxins from polluted water. It is sometimes grown as ornamental in the tropic and as an indoor aquatic ornamental in temperate regions.

Keywords: *Pistia stratiotes* , Taxonomic character, Uses.

Introduction:

Pistia stratiotes, also known as **"Jal Kumbhi "** or Water lettuce . It is a free floating aquatic plant of streams, of ponds and lakes . ***Pistia stratiotes*** has a stoloniferous nature so it is always found anchored to the hydrosoil when the water level recedes and in marshland conditions and love alkaline/ lime-rich water . It forms a dense mats on the surface of water bodies; as it is a floating weed ,it disrupting aquatic flora and found underneath and thus adversely affects the water ecosystem and hinders water flow, swimming ,boating , fishing water sports and navigation. (Attionu 1976,Halm et.al 1977,Sharma 1984) .It replace the native hydrophytes in ponds and other water reservoirs.(Marwat et al 2010).It lowers available oxygen and pH of water and thus damages rice crop when enters into paddy fields, develop roots in the soil and competes with crop under shallow water conditions (Hussain et.al.2000).

Taxonomy of *Pistia stratiotes*:

Pistia stratiotes is a genus of aquatic plants in the arum family , Araceae. The sole genus in tribe is ***Pistieae*** which reflects its systematic isolation within the family .The single species it comprises , ***Pistia stratiotes*** is often called Water cabbage ,Water lettuce, Nice cabbage or Shelf flower.

Pistia stratiotes is a perennial monocotyledone plants .

Leaves: The leaves are thick , soft leaves that from a rosette. These leaves can measure 2-15 cm long and light green, with parallel venations and wavy margins. The surface of the leaves is covered in short , white hairs which form basket , like structures that can trap air bubbles and increase the plants buoyancy. The spongy parenchyma with large intercellular spaces in the leaves also aids the plant in floating .

Roots: It floats on the surface of the surface of the water , it roots hanging submersed beneath floating leaves.

Flowers: The flowers are dioecious ,lake petals and are hidden in the middle of the plants amongst the leaves.

Inflorescence *: Pistia stratiotes* has a spadix inflorescence, containing one pistillate flower with one ovary and 2-8 staminate flowers with two stamens. The pistillate and carpellate

flowers are separated by folds in the spathe, where the male flowers are located above the female flowers oval, green berries with ovoid seeds from after successful fertilization .

Reproduction: The plant undergoes asexual reproduction by propagation through stolons , yet evidence of sexual reproduction has also been observed.

Pistia stratiotes are found in slow moving rivers, lakes and ponds . The species display optimal growth in the temperature of 22-30^0 , but can endure extreme temperature upto 35^{0}C.As a result ,***Pistia stratiotes*** do not grow in colder temperature. The species also require slightly acidic water in pH range of 6.5-7.2 for optimal growth.

.**Uses :**

Environmental remediation.

The high absorption property of ***Pistia stratiotes*** make it a great for biodegradable oil absorbents in marine oil spills. The leaves of ***Pistia stratiotes*** can efficiently absorb significant amount of hydrocarbons due to its large surface area and hydrophobicity.

As a hyper-accumulator, ***Pistia stratiotes*** has been studies as a potential candidate for waste water treatment plants . The roots and leaves of the plants have been found to absorb excess nutrients and heavy metals , such as zinc , chromium and cadmium in contaminated water.

Pistia stratiotes can be grown in water gardens to reduce harmful algal blooms and eutrophic condition . The plants is able to control the growth of algae by restricting light penetration in the water column and competing for nutrients , with significant uptake of phosphorus, ammonia and nitrogen.

Medicinal properties :

- **Anti- inflammatory properties :**-Extractions of the leaves of ***Pistia stratiotes*** reduces mast infiltration and degranulation in allergic reactions and present anti-inflammatory properties .The ethanolic extracts have also been positively correlated with a reduction in inflammatory disorders ,such as Arthritis and Fevers.
- **Anti fungal properties**:- With the popular use of ***Pistia stratiotes*** as a traditional treatment for ringworms, researchers have tested ***Pistia stratiotes*** methanolic extracts on dermatophyte fungi .The results of the studied depicted significant fungicidal activity on T-rubrum ,T-mentagrophytes , E-floccosum.

Medicinal treatment :

There are various medical uses of ***Pistia stratiotes*** throughout region in Asia .The dried leaves are prepared into powder from and are applied to wounds and sore for disinfection . A similar use in present Indian traditional treatment medicine, where the powdered leaf is applied to syphilitic eruption and skin infection .The leaf is infused in water to created an

eyewash to treat allergic conjuctivitis. The eyewash is known to have cooling and analgesic effect.

Therefore ,the plant is commonly called 'eye-pity' in Africa. In addition , the leaves of ***Pistia stratiotes*** can be burned into ash and the ash is used in treating ringworm infection of the scalp.

Consumption:

Pistia stratiotes is not palatable as it is rich in calcium oxalate crystals that are litters in taste. Nevertheless , there are records of the plant being utilized as famine food in India during the great famine of 1876-1878.

The Hausa people of Nigeria used the ash of the plant as a substitute for salt due to its high concentration of potassium chloride ,a mineral salt . This salt substitute , also called Zakankau , was of high importance , especially when imported salt was unavailable .

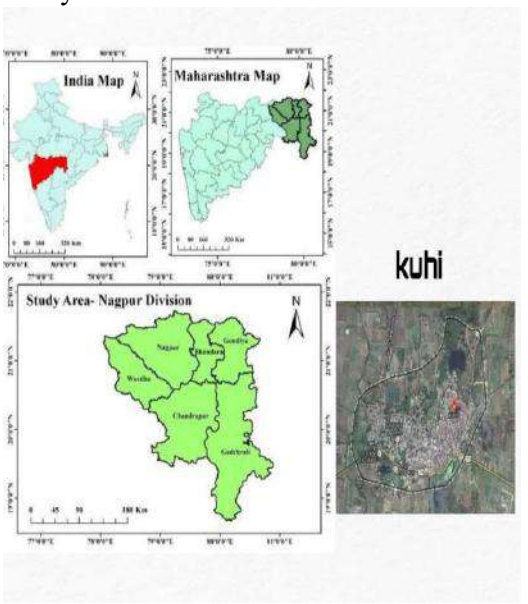
Pistia stratiotes, as the plant is a hyperaccumulator ,and can absorb and accumulate toxic heavy metals present in its environment .The presence of high concentration of calcium oxalate crystals can induce various health concerns, such as inhibited mineral absorption and kidney stones .

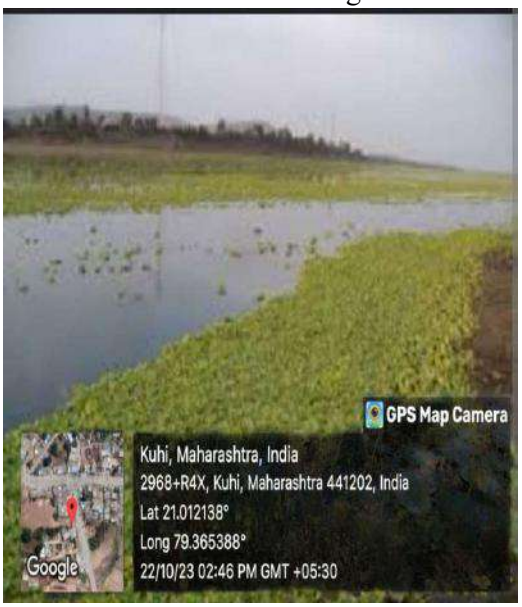
Pistia stratiotes is commonly grown as collected as animal feed for ducks and pigs . Water lettuce is also considered an alternative for poultry feed in Indonesia due to its high content of crude protein .

Conclusion:

Pistia stratiotes , commonly called Water lettuce . It is commonly used as an ornamental plant in water gardens . It produces rosettes ('4-6'acrass) of wedge -shaped , overlapping , fluted , soft green leaves covered with repellant hairs. This plant is mostly found in **Nagpur district** in **Kuhi Theshil** . The large amount of ***Pistia stratiotes*** plant is found in Kuhi lake, where the contaminated water is present..These plants effect the living micro-organisms present in the lakes. These affect the ecosystem of the lake. Biodiversity of the lake water changes .It most specifically changes the color of water. The water of the lakes become acidic in nature.

Pistia stratiotes weeds contain nutrients therefore this weeds can explored to possible use as fertilizer. Collection, processing and application at large scale might be difficult but kitchen gardeners can be motivated to collect this plant and use as fertilizers However, the heavy metal accommocation and the related ill effects need to be investigated.





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Review on Wild Members of Poaceae from Buldhana District, M.S. India

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Abstract
The family Poaceae is one of the most economically important group of Angiospermic plant, belonging to class monocotyledon. A plant specimens of this family commonly known as Grass. It is widely used for food, fodder, as well as making musical instrument like flute. Rice and Wheat are the major food crop belonging to this family. Flower of the grasses are small and minute, hence this group of plants are neglected. So it is necessary to study and explored this family.

Introduction

Grasses are most successful group of monocotyledonous plants. They occur on every type of soil, in all kinds of situations and under all climatic conditions. As grasses do not prefer shade, they are not usually abundant within the forest. But in open places they grow very well and some-time whole tracts become grasslands.

Robinson writes “Grass is King”; it rules and governs the world, without it the earth would be a barren waste. Grasses are important for entire ecosystem. Tiger is the King of forest ecosystem. If we want to save tiger, we have to save the grasses because tigers are indirectly dependent on grasses for their food.

In the early days when the population was much limited and when limited land was under cultivation, much of it was covered with plenty of green grasses. So farmers paid no attention to the grasses. But now population has increased, open land is much decreased and cattles have increased in number hence, farmers should pay more attention to grasses. Present destruction of grasses is mainly due to overgrazing, increasing agricultural practices, over use of herbicides, open field coal mines, construction of big dams, road widening, clean agricultural practices and trampling by men and cattles. Grazing lands need to be developed in areas around villages. Secondly use of herbicides is affecting the availability of fodder around agricultural fields and on bunds. We need reduce the use of herbicides.

Grasses play vital role in the life of human beings and animals. Family Poaceae is of major economic and ecological importance. There are about 10,000-11,000 species belongingto 700 genera in the world, Clayton and Renvoize (1989) and Watson and Dallwitz, (1994), in India there are more than 1200 species belonging to 268 genera Karthikeyan *et.al.(*1989), and Moulik (1997). In Maharashtra, there are 415 species belonging to 125 genera Potdar *et.al.(*2012).

Bamboos and grasses have played a vital role in developing civilizations. Many of thegrasses are known for their fodder value. They are the good soil binders and play important role in water percolation, soil conservation and retention of moisture. Some of them have a medicinal value and yield essential oils. Tender shoots of Bamboo are used as vegetable and also pickled by locals. Grasses are also used on large scale for paper manufacturing and for miscellaneous purposes such as thatching, matting, making ropes, furniture, stuffing for pillows, brooms and musical instruments like flute etc. Stem of *Phragmites vallatorious* was used for preparation of Boru (Tak-i.e. pen used for writing). The grains of grasses certainly provide a staple food supply for the human being *Oryza sativa, Triticum aestivum, zea mays,*

Avena sativa, Setaria italica, Eleusine coracana, Echinochloa colonum, and Sorghum species etc. Rice feeds more human being than any otherplant product. *Saccharum officinarum* is main source of sugar.

High proportion of the most fertile and productive soil were developed under the vegetative cover of grasses. Root, rhizome and other parts of grasses are good soil builders and effective soil stabilizers. Most of the birds and animals depend upon grassland habitat forfood, shelter, and normal completion of their life cycles (Gould,1968).

Grassland occupy about 25 percent of earth's vegetation. Grasses are found on all continents and in all climatic zones even they are found in the ice covered and Arctic regions.Generally high percentage of grasses found in hilly regions and open places.

SUMMARY

Despite at most importance of grasses to human being, the study on grasses continues to be a neglected subject. This is mainly because of the feeling that it is a difficult group for identification the leaves and branches of grasses are very much similar, small floral organs, special terminology and variation in the structure of spikelets and inflorescence. "*grasses of Burma, Ceylon, India and Pakistan"* studied by Bor (1960) is still main standard reference work on Indian grasses.

Hooker (1872-1896) was the first to carry out exhaustive floristic exploration of India along with independent countries like Nepal, Sikkim, Ceylon, and Malaysia. The work were published in seven volumes; the seventh being dedicated to Gramineae.

Necessity of regional and local floras resulted in publication of 'Flora of Bombay Presidency' by Cooke (1901-1908). This administrative division included Sindh, Karachi, Baroda, Belgaum, Karwar and Bombay region. It reported 223 species of grasses.

Blatter and McCann (1935) further studied grasses of Bombay. Bor (1960) published an account of grass species (excluding Bambusiae) growing in Burma, Ceylon, India and Pakistan (excluding Bambusiae) Both these work were inspired and published by Imperial Council of Agriculture Research.

Floristic surveys of different areas of Maharashtra have been compiled and published by Botanical Survey of India. This compilation reports, Sharma *et. al. (* 1996) 373 species of grasses. Almeida (1996) gave an account of 169 species belonging to 60 genera from Sawatwadi, 127 species belonging to 63 genera were reported from Nashik District, Lakshminarasimhan and Sharma (1991).

Yadav and sardesai (2002) gave an account of 212 grass species belonging to 87 genera occurring in Kolhapur district. Yadav and his associates have studied the grasses of Maharashtra in details. The best field guide for students entitled "*Know Your Grass Genera Through Hand Lens"* by Yadav (2010) provides key for two sub-families, 26 tribes and 115 genera of grasses occurring mainly in Maharashtra and Peninsular India, supported by life like photographs. Floristic survey of Baramati Taluka was made by Bhagat *et.al.*(2008). The survey reports 79 species of grasses. Potdar *et.al.*(2012) has published an account of grasses ofMaharashtra describing 415 species belonging to 125 genera.

Patunkar (1980) studied Grasses of Marathwada region and reported 167 species of grasses belonging to 76 genera. A thorough exploration of Marathwada has been done by Naik and his students. Outcome of this work is *Flora of Marathwada* (Naik 1998), which reported 203 species of grasses belonging to 81 genera.

Purekar (1985) studied Grasses of Nagpur district for Doctoral degree and reported 188 grasses. Just one year later Ugemuge (1986) report 134 species of grasses from Nagpur district. Patil (1991) reported 130 species of grasses from Chandrapur and Gadchiroli districts. Kahalkar (2009) noted 118 grasses from Gondia district for his Ph.D. thesis, Govekar (2016) recorded 220 species of grasses from Gadchiroli district.

Dhore (2002) reported 126 species of grasses from Amravati district. Pradhan and Kamble

(1988) reported 86 grasses belonging to 49 genera from Akola district. Till 1998 Washim was part of Akola district. From July 1998 Washim became separate district. Deore (2010) has conducted *"Floristic Survey of Washim District"* reported 63 species of family Poaceae. Masatkar (2018) during her research work she has reported 117 species of grass from Amravati district "*Morphotaxonomic studies of wild members of Poaceae from Amravati district, Maharashtra".* Tathod (2019) reported 178 species of grasses from Nagpur division *"Morphotaxonomic revision of family Poaceae with special reference to grasses from Nagpur division of Maharashtra". "Flora of Yavatmal District"* by Kartikeyen and Anandkumar (1993) reported 81 species of grasses from Yavatmal district, while Diwakar and Sharma (2000) reported 61 species of grasses belonging to 44 genera from Buldhana district.

Change has been a rule of nature, in due course of time; new species may arrive or disappear from any region or area. Unequal distribution of rainfall, and human invasion greatly affecting the Poaceae members of the study area. It was therefore, felt necessary to explore the wild members of Poaceae in the Buldhana district.

CONCLUSION

During just a few visits around the study area, some species not recorded in the "*Flora of Buldhana District"* could be collected. Flora of the Buldhana district was explored in 1989 and published in 2000. During this period some of the species might have been wiped out from the region. While some may have been added.

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Impacts of Water Pollution on Human Health and its Diseases

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Abstract:

As water is one of the most important compounds of the ecosystem. But due to increased human population, industrialization, use of the fertilizers in the agriculture, as manmade activities polluted the water. The natural aquatic resources are causing heavy and varied pollution in aquatic environment leading to pollute water quality and depletion of aquatic biota and ultimately affecting human health and sustainable social development. The water pollution causes diarrhea, skin diseases, malnutrition, and even cancer and other diseases related to water pollution. Consequently, research on the impacts of water contamination on human wellbeing especially the assortment of sickness is basic to stress the meaning of clean drinking water, which has huge hypothetical and pragmatic importance for acknowledging economic advancement objectives. This paper focuses on the impact of water pollution on human health and its disease.

Keywords: Water, Human health, Water pollution and Water quality.

Introduction:

Water is an essential component for life on earth. It has many distinct properties that are critical for the proliferation of life. Any known form of life requires water to exist. The water pollution is the contamination of water bodies, usually as a result of human activities, so that it negatively affects its uses. Water bodies include lakes, rivers, oceans, reservoirs and groundwater. There was water pollution, when contaminants mix with these water bodies. Contaminants can come from many sources such as sewage discharges, industrial activities, agricultural activities, and urban runoff including storm water. Water pollution is either surface water pollution or groundwater pollution. When people utilize dirty water for irrigation or drinking, it can lead to a number of issues, including the destruction of aquatic ecosystems and the spread of diseases. These contaminants discharged into the environment without any prior treatment, with adverse effects on human health and ecosystems. In the least developed nations, where there is a serious shortage of sanitation and wastewater treatment infrastructure, this share is higher.

The water pollution results from both human and natural factors. Various human activities will directly affect water quality, including urbanization, population growth, industrial production, climate change, and other factors and religious activities. The quality of drinking water is an important factor affecting human health. The poor quality of drinking water has led to the occurrence of water-borne diseases. According to the World Health Organization (WHO) survey, 80% of the world's diseases and 50% of the world's child deaths are related to poor drinking water quality, and there are more than 50 diseases caused by poor drinking water quality. The quality of drinking water in developing countries is a cause for concern. The negative health effects of water pollution continue to be a major cause of morbidity and mortality in developing countries. Different from the existing literature survey, this paper mainly studies the impact of water pollution on human health according to the heterogeneity of diseases.

Observations and Result:

The water pollution causes many diseases such as diarrhea, skin diseases, malnutrition, and even cancer and other diseases related to water pollution. Therefore, it is necessary to study the impact of water pollution on human health, especially the heterogeneity of diseases. Diarrhea is a common symptom of gastrointestinal diseases. The most common disease caused by water pollution such as diarrhea, is a leading cause of illness and death in young children in low-income countries. Skin diseases in swimmers can be caused by several pathogenic microorganisms. When examining the relationship between excess arsenic in drinking water caused by water pollution and skin diseases (mainly melanosis and keratosis), In addition, arsenic in drinking water is a possible cause of cancer in children. Children may develop goiter if their drinking water is contaminated with nitrates. The various water sources like arsenic, nitrate, chromium, etc. are highly associated with cancer. Ingestion of arsenic in drinking water can cause skin cancer and kidney and bladder cancer. Pollution caused by exposure to microbial infected water and food, and diarrhea in infants and young children can lead to malnutrition and reduced immune resistance.

Controls:

Managing water pollution requires adequate infrastructure and management plans and legislation. Technology solutions can include improving sanitation, sewage treatment, industrial wastewater treatment, agricultural wastewater treatment, erosion control, sediment control and control of urban runoff including storm water management. Also to aware the people to control the water pollution.

Conclusion:

In conclusion, water pollution is a significant cause of childhood diseases. Unfortunately, although several types of literature focus on water pollution and a specific disease, there is still a lack of scientific results that dissect the effect of water contamination on human wellbeing and the heterogeneity of sicknesses. This study is focusing on the impact of water pollution on human health and the heterogeneity of diseases from the perspective of different diseases, mechanism and influencing factors of water pollution and diseases. The developing countries, need to adopt corresponding water management policies to reduce the harm caused by water pollution to human health. Each society should prevent and control source pollution from production, consumption, and transportation. Also introduce environmental education, educate residents on sanitary water through newspapers, magazines, television, Internet and other media, and enhance public health awareness.

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The seasonal variation of physico-chemical characteristics of Mahan Dam, Mahan, District Akola (MS)

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Abstract:
Water is a necessary element for maintaining an ecosystem's high standard of living. India has a vast amount of both man-made and natural water bodies. The primary uses of these sources of water are agriculture and drinking. Present study deals with physico chemical study such as detection of pH, Total dissolved solids, temperature, conductivity, dissolve oxygen, Nitrate and phosphate of Mahan dam which is located at at latitude 20.49283and longitude 77.1358 in akola distict and it was found that water of this dam is safe for drinking and domestic use according to WHO.

Keywords: Mahan dam, water sample,P^H,TDS *etc.*

Introduction:
Water is a necessary element for maintaining an ecosystem's high standard of living. India has a vast amount of both man-made and natural water bodies. The primary uses of these sources of water are agriculture and drinking. Due to contemporary farming methods and the rising urbanization of the region, the quality of water bodies has lately declined. Because they tend to accumulate pollutants and have a limited potential for self-purification, lakes are said to have the most delicate ecosystems. Assessing the quality of the water is crucial for maintaining and safeguarding the natural habitat. The goal of the current project is to evaluate the Mahan dam's water quality in the Akola district of Maharashtra, with a particular focus on the dam's pollution level.

Materials and Methods:

Study Area

In the Akola district of Maharashtra (India), the Mahan Taluka is situated at latitude 20.49283and longitude 77.1358. The Mahan Dam is close to Barshtakali, an earthfill dam on the Katepurna River. It provides service to the suburbs and city of Akola. It supplies water to more than 80,000 residents of Akola and 69 neighboring settlements.

The height of the dam above its lowest foundation is 29.5 m (97 ft), while the length is 2,000 m (6,600 ft). Its volume is 693,000 m3 (24,500,000 cu ft), and its gross storage capacity is 97,670,000 m3 (3.449×109 cu ft).[1]

Sample Collection: For the present research work one liter water was collected from each sampling site throughout the year. Acquiring relevant data necessitates using proper sampling and preservation techniques. From June2021 to May 2022, water samples were taken all year round from the chosen locations. Periodically, during the first week of each month, samples were taken in the morning between 8.30 and 10.30. Samples were gathered in a five-liter plastic container that had been acid-washed.

Physico-chemical Analysis: Mahan's water bearing formation is basalt (Deccan trap) Physical parameters of water such as Colour, Odour, turbidity, pH, Electrical fractured, jointed and weathered basalt under phreatic conditions and the soil type is medium black cotton soil [2]. Conductivity, Total Dissolved Solids were analysed on sampling site and chemical parameters such as phosphate and nitrate were analysed within 24 hours by standard methods [3-4]. All

physico-chemical parameter were performed in triplicates and average was considered as the reading also data was analyzed statistically. [5].

Result and discussion: Physico-chemical properties as per seasonal variation of Mahan Dam, Dist. Akola. During year June 2021 to May 2022 is shown in table 01.

Parameter	Summer	Monsoon	Winter	Average
pH	7.58±0.10	6.86±0.03	7.34±0.10	7.36±
Total dissolved solids (mg/l)	566.30±0.65	619.50±0.84	344.40±2.25	510.07±
Temperature(^{0}C)	27.59±1.39	26.02±0.70	20.24±0.69	24.62±0.92
Conductivity(us/cm)	698.60±1.26	560.90±3.00	533.20±1.80	597.57±
Dissolved Oxygen	39.55±1.78	23.00±2.21	22.25±0.35	28.27±
Phosphate	0.21±0.007	0.32±0.002	0.16±0.002	0.23±
Nitrate	1.44±0.10	4.27±0.70	0.16±0.003	1.96±

±standard deviation.

All the samples were colourless and odourless at the time of collection slight acidic and basic condition is detected in all samples only water is basic in monsoon season with pH 6.86 and slight acidic in summer with pH 7.58. Water said to be alkaline when concentration of OH ion is more than H ions . The alkalinity of ground water is because of carbonates and bicarbonates [6]. Total dissolved solids ranges form 344 mg/L to 619 mg/L. and TDS is higher in monsoon season due to particulate matter runoff with water in dam. [7-8]. Higher temperature was recorded 27 ^{0}C in summer and lowest in winter 20.24^{0}C as usual. Conductivity is ranges form 698.60 to 533.20 us/cm. dissolved oxygen recorded 39.55 in summer 23.00 in monsoon and 22.25 in winter. Low DO in summer is due to insolubility of gases at high temperature [9]. Phosphate was recorded in very few quantity i.e 0.16 to0.32According to[10], the assimilation of phosphates by phytoplankton population reproduction is responsible for their growth and is responsible for decreasing the levels of phosphates in the winter months. Nitrate show great variation in ranges form 4.27 to 0.16.

Conclusion:

It is concluded from the present research work that water collected Mahan dam were found safe for drinking and domestic use according to WHO.

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Antibacterial and antifungal activity of*Woodfordia fruticosa*leaves in different solvents against selected pathogenic organisms

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ABSTRACT:
Plants have always been one of the most important sources of medicinal materials. The use of herbal medicine is growing in significance every day. The antibacterial activity of crude leaf extracts of *Woodfordia fruticosa* in four different solvents: acetone, petroleum ether, ethanol, and water is examined. The leaves extract of *Woodfordia fruticosa* exhibits antibacterial activity against *Bacillus subtilis* and *Escherichia coli.* Antifungal activity shown against *Candida albicans* at higher concentrations only. There is no antibacterial activity against *Staphylococcus aureus* and *Pseudomonas aeruginosa* also no antifungal activity observed *against Aspergillus niger.* Aqueous extracts dose not show antibacterial and antifungal activity.

INTRODUCTION -
Since ancient times, people have been treating a variety of ailments with plants as medicines, and these applications have shown to be quite successful. People have been studying the plants to find new medications, which has led to the usage of many medicinal plants to treat a wide range of illnesses (Verpoorte, 1998). Eighty percent of the population in underdeveloped countries, according to the World Health Organization (WHO, 2008), only receives primary healthcare from traditional medicine, the majority of which uses plant extracts (Sandhya et al., 2006).
Overuse of antibiotics has a negative impact on the environment, ecosystem, and health of people. Additionally, it might make drug-resistant infections more common (Jastaniah, 2014). Antibiotic resistance is a big global issue that is fast getting worse in hospitals and the general public when it comes to morbidity, mortality, and health care (Mill Robertson, 2015).
Multiple drug-resistant bacteria were the primary cause of the major failure in the treatment of infectious diseases because it has been discovered that almost all pathogenic bacteria are able to quickly acquire the resistance factor to the antimicrobial medications (Bisht, 2009). Therefore, in order to control resistant bacteria, it is required to investigate and develop alternate strategies. The strategic placement of bioactive phytochemicals with antibacterial properties is one approach that may be used (Silva, 2007). Researchers have looked into a wide range of secondary chemicals found in plants that may serve as a source for different antibacterial agents (Amer, 2007). Many structurally distinct bioactive chemicals found in those plants are good sources of naturally occurring medicinal medicines (Aly, 2004).
For the current investigation four pathogenic bacteria and two pathogenic fungi are selected to know the antibacterial and antifungal activity of *Woodfordia fruticosa* leaves extract in different solvents. The selected pathogenic organisms are *Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Aspergillus niger* and *Candida albicans.*
Woodfordia fruticosa is a wild deciduous aerial shrub of 2 - 3 m tall with spreading stems, branched, woody, branched, cylindrical, solid stem. Leaves are cauline and ramal subsessile, 4-11 x 2-4 cm, opposite, superposed, unicostate reticulate, ovate-lanceolate or lanceolate, sub-coriaceous, whitish velvety tomentose and finely orangish- or black-punctate beneath. Inflorescence is axillary cymes. Flowers are complete, bisexual, crimson, slightly zygomorphic, in 2-16-flowered; pedicels to 1 cm long. Calyx tube 1-1.5 cm long, tubular; lobes

6, short, more or less triangular, alternating with small callous appendages. Petals 6, red, 3-4 mm long, lanceolate-acuminate. Stamens 12, inserted near the bottom of the calyx tube, 0.5-1.5 cm long. Ovary 4-6 mm long, oblong, 2-celled; ovules many; style 0.7-1.5 cm long. Capsule 0.6-1 x 0.25-0.4 cm, ellipsoid, included in the calyx. Seeds numerous, trigonous-ovoid.

2. Materials and Methods

2.1 Collection and extraction of plant material:

The plant was collected from the roadsides in the Khatkali, Tq. Akot, Dist. Akola (M.S.). *Woodfordia fruticosa* plants was identified by using the flora of Amravati district with special reference to the distribution of tree species (Dhore, 1986). The fresh and matured leaves of *Woodfordia fruticosa*, wash with the running tap water to get rid of dust and other pollutants from the leaves. After being shade-dried for seven days, the leaves were finely ground, stored, and ready for use.

2.2 Preparation of plant extract for Phytochemical Analysis:

The leaf extract of *Woodfordia fruticosa* were prepared in acetone, petroleum ether, and ethanol using a Soxhlet extractor. Aqueous extract was made by boiling the powdered leaves in a solution. After 30 minutes at 50 to 60 º C, then filter the water through Whatman No. 01 filter paper. Following the extraction of every component, the solvent is evaporated to get the concentrated extract, which is then stored for later use.

2.3 Antibacterial and Antifungal Activity

The Antibacterial activity was checked by following Zone Inhibition Method. The MHA plates were inoculated by spreading with 100 µl of Bacterial and Fungal culture, followed by placing the disc containing 10 µl of different concentration (0 to 100 mg/ml). 10 % of the sample was taken and serially diluted to achieve the required amount to be loaded on the disc. One disc in each plate was loaded with solvent alone which served as vehicle control and Ciprofloxacin disc (10µg) for bacteria and Amphotericin B (50µg) for fungi were taken as positive control. The plates were incubated at 37 °C for 24 hrs. A clear zone created around the disc were measured and recorded.

RESULT AND DISCUSSION: The observations of antibacterial and antifungal activity of *Woodfordia fruticosa* are shown as following table –

Table 01 - Antibacterial and antifungal activity shown by leaves extract of *Woodfordia fruticosa* in different solvents.

Test Organisms	**Amount (µg/disk)** → / Solvents ↓	Zone of Inhibition						
		P.C.	**0**	**50**	**125**	**250**	**500**	**1000**
Bacillus subtilis	Acetone	23	0	6	7	8	9	10
	Petroleum Ether	31	0	14	15	16	16	17
	Ethanol	31	0	4	7	8	8	15
	Distilled Water	29	0	0	0	0	5	7
Staphylococcus aureus	Acetone	27	0	0	0	0	0	0
	Petroleum Ether	27	0	0	0	0	0	12
	Ethanol	27	0	0	0	0	0	10
	Distilled Water	27	0	0	0	0	0	0
Pseudomonas aeruginosa	Acetone	28	0	0	0	0	0	0
	Petroleum Ether	29	0	0	0	0	0	0
	Ethanol	28	0	0	0	0	0	0
	Dist. Water	28	0	0	0	0	0	0
Escherichia coli	Acetone	19	0	0	0	6	6	7
	Petroleum Ether	20	0	0	5	8	9	10

	Ethanol	20	0	0	6	7	8	11
	Distilled Water	18	0	0	0	0	0	0
Aspergillus niger	Acetone	18	0	0	0	0	0	0
	Petroleum Ether	22	0	0	0	0	0	0
	Ethanol	23	0	0	0	0	0	0
	Distilled Water	21	0	0	0	0	0	0
Candida albicans	Acetone	18	0	6	6	7	7	11
	Petroleum Ether	19	0	9	9	9	7	7
	Ethanol	18	0	6	6	6	7	14
	Distilled Water	17	0	0	0	0	0	0

CONCLUSION:

The antibacterial and antifungal activity of leaf extracts of *Woodfordia fruticosa* in various solvents like Acetone, Petroleum ether, Ethanol and Distilled Water leads to the conclusion that the plant leaves of *Woodfordia fruticosa* exhibit antibacterial activity against *Bacillus subtilis* and *Escherichia coli* in acetone, petroleum ether, and ethanol extracts. At increasing concentrations of acetone, petroleum ether and ethanolic extracts shows increasing antibacterial activity; the aqueous extract shows antibacterial activity against *Bacillus subtilis only at higher concentration in 500* µg *and 1000* µg . the zone of inhibition also grows in size with increasing concentrations, and the antifungal activity shown against *Candida albicans* only in Acetone, Petroleum ether and ethanolic extract of *Woodfordia fruticosa*. There is no antifungal activity shown against *Aspergillus niger.*

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Arbuscular mycorrhizal fungi diversity in coal mine soil of *Datura metal* at Chandrapur district

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ABSTRACT: An investigation was carried out on the species diversity of arbuscular mycorrhizal (AM) fungi in contaminated soils. Samples were analyzed for physicochemical parameters, including P^H, temperature, texture, sulfur, silica, and phosphorus, and it was concluded that AM fungi are known to enhance plant tolerance capacity in different stress conditions. A study was undertaken to assess the influence of coal mines on mycorrhizal colonization and revealed that mycorrhizal colonization and mycorrhizal spores are significantly positively correlated with various physiochemical properties in the contaminated soil. The results revealed that Glomus was the most dominant isolated mycorrhizal genus with 13 species, *Acualospora* having 06 species, *Scutellospora*-01 species, *Sclerocystis*-01 species, and *Entrophosphora*-01 species. This is because areas with heavy metallic elements in soil adversely affect the richness and diversity of AM fungal species.

Keywords: Contaminated rhizospheric soil, physiochemical analysis, AM isolation, AM spore species.

INTRODUCTION

AMF have played an important role in the evolution of land plants for more than four hundred million years [(1)] ,AMF can enhance tolerance of abiotic stresses such as drought and metal toxicity [(2)]. Therefore, it is evident that AMF are an important associate for crop plants in sustainable agriculture. Mycorrhizal fungi are known to affect the growth of most plant species in mine degraded areas, but in normal soil regions, plants, despite their ability to live independently, may increase nutrient uptake, growth, and reproductive success when associated with AM . Moreover, AMF improves soil quality [(3)] and enhances the ability of host plants to withstand abiotic stress and disease [(4)], thereby increasing plant performance [(5)] .Coal is the most abundant fossil fuel on Earth and accounts for about 75 % of the total fuel resources [(6)].These mine-degraded soils are a man-made habitat that presents a wide range of problems for establishing and maintaining a vegetation cover [(7)]. Mining activities exert lasting effects on ecosystems, both through structural changes and impacts on biodiversity. Mining activities produce wastes that may contain heavy metals as contaminants. These residues are generally deposited on the ground and often occupy large extensions. However, plants often have very limited development in contaminated areas. Due to their beneficial effects on plant growth under stressed conditions, AMF have been used to enhance the rehabilitation of contaminated soils through phytoremediation (8), and the beneficial role of vesicular arbuscular mycorrhizae in mine spoils is revegetation.

MATERIAL AND METHODS: The soil sample was collected from the coal mine spoil region at the Durgapur opencast coal mine site (E 20˚, N 79˚) at Chandrapur, Maharashtra State, India.

Fig.1- India, Fig.2-Maharashtra, Fig.3-Vidarbha, Fig.4-Chandrapur, Fig.5- Coal Mine Area

Fig.1

Fig.2

Fig.3

Fig.4

Fig.5

SAMPLING AND ANALYSIS:

The rhizosphere soil of *Datura metel*-contaminated region at chandrapur were collected in sterile polythene bags. The collection was carried out from the Durgapur opencast coal mine region of Chandrapur district. The collected soil sample had different soil textures like sandy, sandy loamy, loamy, clay, and clay loamy. The rhizosphere soil sample was packed in clean sterile polythene bags, dried, and stored at room temperature.

Fig:6 **A**-Durgapur opencast mine-Chandrapur, **B**-Contaminated soil sample, and **C**-Rhizosporic sample

QUALITATIVE AND QUANTITATIVE ESTIMATION OF VAM FUNGI:

Different methods are used for counting AM fungal spores. The procedure described by Gaur and Adholeya 1994[9] was used for counting AM spores as it is a simplified method for counting AM fungal spores. This method is used to count AM fungal spores in soil samples as follows. Various methods are used to isolate VAM spores from soil samples. In this study, the wet sieving and decanting technique was used [10]. Photographs illustrating the different characteristic structures of VAM fungal species, such as the wall layers arrangement of spores in sporocarp, loose sporocarp, and other details, were taken using an electron microscope. An electron microscope with an attached Pentax thousand camera (magnification 40x and 10x). The VAM fungi are identified using the manual of Schenck and Perez 1990 [11], keys of Morton and Benny 1990 [12] and the keys of Mehrotra and Baijal 1994 [13].

MINERAL TESTS FOR SOIL:-

In the present work, different mineral tests were also carried out using polluted soil from the mine region at Chandrapur, including the Sulphur test [14], Silica test [15], and Phosphorus test [16].

OBSERVATIONS AND RESULTS:

The soil samples collected had varying textures, identified through the sieving technique, and their pH was measured using a pH meter. The collected data are in Table No.1.

Table No.1-Soil analysis of rhizospheric soil.

Sr.No.	Site Name	P^H	Temperature	Soil Texture and			
1	Chandrapur-coal mine area	7.4	25ºC	Sandy	Loamy	Clay	Fine
				1mm	0.600mm	0.090mm	0.053mm

QUALITATIVE ANALYSIS:

AM fungi stimulate plant growth by aiding in nutrient absorption. In the present study, plant samples were collected from the mining region of Chandrapur area, and the rate of AM fungi for plant growth and development was measured. The following data was recorded in Table No. 2

Table No.2 Identified species with diameters in µm in the coal mine area at Chandrapur.

Sr. No.	Slide No.	Identified species	Diameter µm	Number of species
1.	S1	*Glomus aggregatum*	82.63 µm	*Glomus* – 1 species
		Scutellospora nigra	57.31 µm	*Scutellospora* – 1 species
2.	S2	*Glomus reticulatum*	220.14 µm	*Glomus*– 1 species
		Acaulospora foveata	178.50 µm	*Acaulospora* – 1 species
		Scutellospora nigra	65.41 µm	*Scutellospora* – 1 species
3.	S3	*Glomus globiferum*	80.80 µm	*Glomus* – 2 species
		Glomus reticulatum	195.21 µm	
4.	S4	*Glomus fragilistratum*	56.08 µm	*Glomus* – 1 species
		Scutellospora nigra	60.02 µm	*Scutellospora* – 1 species
5.	S5	*Acaulospora taiwania*	70.24 µm	*Acaulospora* – 1 species
6.	S6	*Glomus halon*	141.21 µm	*Glomus* – 1 species
7.	S7	*Glomus aggregatum*	227.02 µm	*Glomus*– 1 species
		Acaulospora mellea	157.18 µm	*Acaulospora*– 1 species
8.	S8	*Acaulospora taiwania*	297.05 µm	*Acaulospora* – 1 species
		Glomus aggregatum	97.24 µm	*Glomus*– 1 species
9.	S9	*Acaulospora longula*	180.68 µm	*Acaulospora* – 1 species
10.	S10	*Glomus tenerum*	73.01 µm	*Glomus*– 2 species
		Glomus fasciculatum	69.42 µm	
11.	S11	*Glomus gerdimannii*	82.54 µm	*Glomus*– 1 species
12.	S12	*Sclerocystis coremioides*	79.33 µm	*Sclerocystis* – 1 species
13.	S13	*Acaulospora longula*	180.68 µm	*Acaulospora*– 1 species
		Glomus albidum	97.23 µm	*Glomus* – 1 species
		Entrophospora	26.24 µm	*Entrophospora*– 1
14.	S14	*Scutellospora nigra*	121.44 µm	*Scutellospora* – 1 species
15.	S15	*Acaulospora foveata*	104.98 µm	*Acaulospora* – 1 species
16.	S16	*Acaulospora scrobiculata*	115.67 µm	*Acaulospora* – 1 species
		Glomus fecundisporum	92.25 µm	*Glomus* – 2 species
		Glomus diaphanum	88.47 µm	
17.	S17	*Glomus fasciculatum*	89.26 µm	*Glomus* – 2 species
		Glomus globiferum	78.56 µm	
18.	S18	*Glomus fasciculatum*	125.36 µm	*Glomus* – 1 species
19.	S19	*Glomus criticola*	59.23 µm	*Glomus* – 2 species
		Glomus globiferum	86.48 µm	
20.	S20	*Glomus globiferum*	94.35 µm	*Glomus* – 2 species
		Glomus aggregatum	85.26 µm	
21.	S21	*Acaulospora denticulatum*	76.94 µm	*Acaulospora* – 1 species
22.	S22	*Glomus clarum*	48.67 µm	*Glomus* – 2 species
		Glomus albidum	125.47 µm	
23.	S23	*Acaulosporadenticulatum*	52.87 µm	*Acaulospora*–1 species
		Glomus citricola	78.16 µm	*Glomus* – 1 species
24.	S24	*Glomus citricola*	68.15 µm	*Glomus* – 2 species
		Glomus fasciculatum	49.32 µm	
25.	S25	*Glomus fasciculatum*	73.45 µm	*Glomus* – 1 species

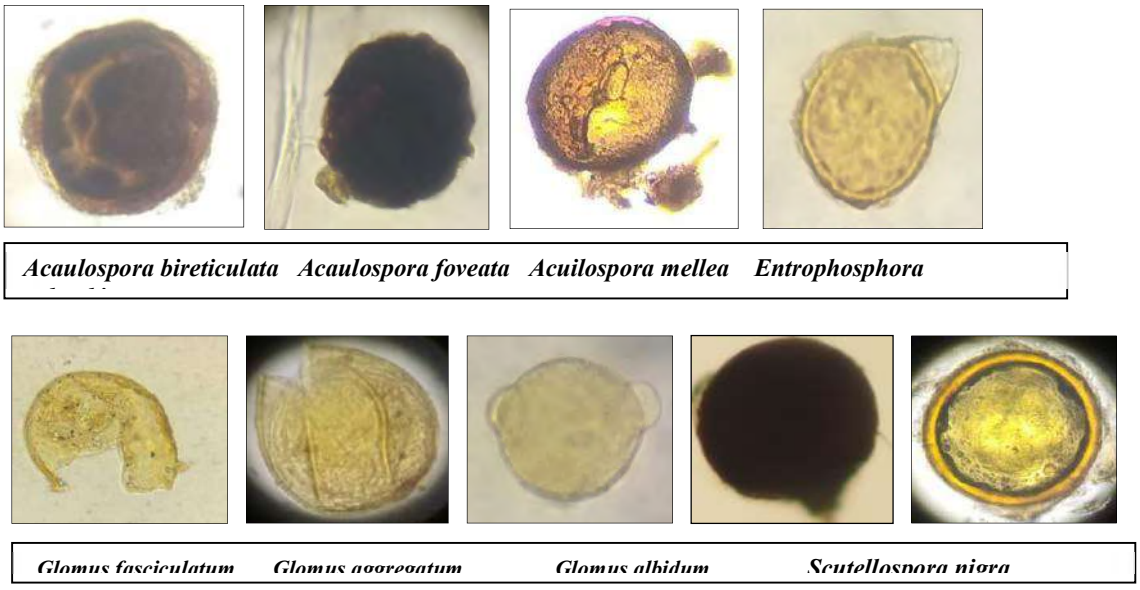

Fig 6: Observed and identified Am spores:

QUANTIFICATION:

Quantitative analysis revealed that the number of isolated spore species varies according to their tolerance capacity in polluted regions. This study, it was revealed that Glomus species were dominant in the mine region compared to others, as observed in Table No.3.

Table No.3 Quantitative analysis of identified species in coal mine soil

Sr. No.	Name of Species	Number of species in coal mine soil
1	*Glomus*	13
2	*Acaulospora*	06
3	*Scutellospora*	01
4	*Sclerocystis*	01
5	*Entrophosphora*	01

Fig 07: Quantification data of AM species in coal mine soil and normal soil.

MINERAL ANALYSIS:

Soil analysis is very important for plant growth and productivity, in the present work mining soil are used for various mineral test viz. Sulphur test, Silica Test and Phosphorous test for observing their amount in soil and growth rate of plant are as follow.

Table No.4 Mineral analysis of both collected soil samples

Sr.No.	Name of site	Mineral test Analysis		
		Sulphur Test	Silica test	Phosphorus Test
1	Coal Mine soil	21.77 mg/L	At 650 nm =0.011	33.045 ppm.

CONCLUSION:

Opencast mining, which is the main form of coal mining in India. In this area, plant growth is generally futile due to high temperatures and P^H, water shortages, and a lack of minerals. AM fungi play an important role in plant growth. Several environmental variables affect the occurrence and distribution of AM spore density in different soil profiles and their root infection status.

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17

Critical review of assessment of seasonal variation in water quality of Godavari river in Nashik city

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Abstract:
Study on Critical review on seasonal variation in water quality of Godavari river which flows in Nashik city. The standard methods have been used to analyze the physico-chemical biological parameters. Due to a variety of human activities, particularly home, industrial, and religious waste water connect without treatment in river quality of surface water is diminishing. The primary goal of the research is to analyze seasonal variation in water quality and develop corrective actions to lessen future degradation and its effects. To assess the quality of the water, the following factors were used Temperature, Color, Odor, Turbidity, pH, DO, BOD, COD, TSS, TDS, Total Hardness, Alkalinity, E-coli and Algae as well as Heavy Metals like Chromium, Iron, Lead, Cadmium, Nickel, Mercury etc. The selection of the water quality parameters, the creation of sub-indices for each parameter, the computation of the parameter weighting values, and the aggregation of sub-indices to calculate the overall water quality index are the four processes that WQI models typically involve assess the water quality of rivers, lakes, reservoirs, and estuaries. Accuracy problems with the models are also covered.

KEYWORD: Godavari River, Seasonal variation, Water Quality, Water quality parameters.

1. Introduction

water bodies are significantly impacted by human activity because of the rapid loss of vegetation in the area due to settlement expansion, industrialization, etc. Sludge and nutrient load rise as a result. Use of river water for defecation, sewage disposal, industrial waste water discharge, agricultural operations involving the use of chemical fertilizers, pesticides in large quantities, and organic input into water bodies.(Bagul & Kadam, n.d.)Water has an unpleasant odor coming from lakes, rivers, and beaches. A few indications of water pollution include the uncontrolled growth of aquatic plant species and the decline in fish populations. To understand chemical phenomena occurring in water bodies, general water quality parameters like pH, DO, BOD, and TDS are used. In natural water containing HCO3, CO3, and OH, pH is a measure of the concentration of H+ ions. Inorganic salts, mostly calcium, magnesium, potassium, sodium, bicarbonates, chlorides, and sulfates, make up total dissolved solids (TDS), along with some trace amounts of organic matter. TDS in water comes from sewage, urban runoff, industrial waste water and chemicals employed in the water treatment process, Dissolved oxygen (DO), We can learn a lot about water quality from DO. When organic matter decomposes, bacteria in the water oxidize the oxygen. Therefore, an abundance of organic material in rivers can result in eutrophic conditions, which are oxygen-deficient circumstances that can cause a water body to "die." We can better comprehend water body pollution by measuring a variety of additional indicators, such as BOD and Fecal coli. WQI serves as a summary of all of them.(Kharake & Raut, 2021) . The major objective of the current study is to create a simplified WQI in order to study the effects of human activities on the Godavari river's water quality in Nashik. Additional research will be useful to reduce behaviors that cause water contamination and to raise awareness among locals, farmers, business owners, etc (Uddin et al., 2021)

2. Material and Methods:

Due to ecological processes and human influence, there is an abundance of zooplankton in the Godavari river's water body throughout the summer, or the pre-monsoon season, as compared to the during and post-monsoon seasons. Due to human involvement including farming, home activities, and industrial discharges, the water at Odha was severely contaminated. Zooplanktons are excellent bio-indicators of the trophic state of the water as well as indicators of water system contamination. By feeding on them, they control the growth of algae and other parasitic organisms. (Bagul & Kadam, n.d.)

The variations in water parameters are caused by variations in fertilizer levels during the winter and summer seasons. It is evident that the Godavari river water in all of Nashik is unsafe for drinking when the observations are contrasted with the permitted limits established by standards (WHO and ISI). The results show that the Godavari River is polluted in various places, and that the pollution level rises over the summer. Due to the serious threat to public health posed by the situation, these river areas must implement strict pollution management and mitigation measures. (Popatrao Kaware & Manzoor, 2022)

At six sites, several factors, including physical, chemical, and biological data, are considered when determining the Narmada River's water quality. (S1–S6). To evaluate the quality of river water, eight water quality indicators—such as nitrate-nitrogen, pH, dissolved oxygen, phosphorus, biological oxygen demand, turbidity, total dissolved solids, and temperature—were considered. The results of the study show that the Narmada River's water quality was found to be suitable for human consumption during both the summer and winter months, with values of the parameters (pH 7.7-8.48, TDS 108-234 mg L1, turbidity 0.01-178.25 NTU, nitrate-nitrogen 0.03-3.14 mg L1, phosphate 0.01-0.52 mg L1, BOD 0.35-2.18 mg L1, and DO 2.4-7. (Gupta et al., 2017).

3. Discussion

Along the Narmada River, it was determined that the water quality was outstanding to good in the summer and winter and bad to unfit for human consumption in the monsoon season. Poor sanitation, turbulent flow, significant soil erosion and excessive anthropogenic activity were all factors in the decline in water quality during the monsoon season.

4. Conclusion

Water quality refers to the Physico- chemical, biological properties of water and heavy metals concentration in sediment usually in relation to whether or not it is suitable for a certain use. Water is used for a variety of purposes, including recreation, drinking, fishing, agriculture, and industry. Different defined chemical, physical, and biological standards are required to support each of these designated purposes. For instance, compared to water used in agriculture and industry, we demand greater standards for the water we drink and swim in. (Rahil Shaltami et al., n.d.)

5. Acknowledgement

Further work on creating more affordable to assess the Godavari river water quality may aid in the remediation of to suggest treatment process control water pollution. With thanks to all colleges for providing all the assistance to carry out this work.

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18

New Distribution Record of *Ophioglossum parvifolium* (Ophioglossaceae) from District Bhandara of Eastern Maharashtra

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ABSTRACT
The *Ophioglossum* L. is the most specialized and widely distributed, genus of the eusporangiate fern family Ophioglossaceae, also known as Adder's tongue or Snake tongue fern. Measurable diversity of *Ophioglossum* L. among the pteridophytic flora is reported in different states of India. Several members of the genus are reported from different regions of Maharashtra as well. In the present study, *Ophioglossum parvifolium* has been reported from the Bhandara district of Eastern Maharashtra. The species is entirely new to the Bhandara district of eastern Maharashtra, India.
***Keywords*:** Ophioglossum, Distribution, Eusporangiate, Ferns, Bhandara.

1. Introduction:

Eastern Maharashtra has a significant floristic diversity of different ecosystems. District Bhandara is situated in the Vidarbha region of Eastern Maharashtra,India. The district is rich in biodiversity, with moderate rainfall, forest cover, nutrient-rich soil, and availability of freshwater ecosystems in the form of rivers and wetlands making the region'sbiodiversity-rich. The *Ophioglossum* L. is the most specialized and widely distributed, genus of the eusporangiate fern family Ophioglossaceae, also known as Adder's tongue or Snake tongue fern.(1,2)Measurable diversity of *Ophioglossum* L. among the pteridophytic flora is reported in different states of India. Several members of the genus are reported from different regions of Maharashtra as well. The remarkable work on the distribution of *Ophioglossum* L. was done by Bhuskute, 1999 for the Bhandara district (Old), by reporting six species belonging to the genera *Ophioglossum* L. The reported species viz. *Ophioglossum costatum*R. Br. (*O. fibrossum Schum*.), *O. petiolatum*Hook., *O. reticulatum*L., *O. gramineum*Willd., *O. lustianicum*L., *O. nudicaule*L. and *O. Vulgatum* Gadpayaleand Chaturvedi, 2014. The present study'smorphological and other taxonomic investigation, distribution,and phenologyof *Ophioglossum* L . from the Bhandara districtwere recorded by frequent site visitsfrom June to September from the year 2018 to the year 2022.

2. Materials and Methods

The present study's morphological and other taxonomic investigation, distribution, and phenology of *Ophioglossum* L. from the Bhandara district was recorded by frequent site visitsfrom June to September from year 2019 to year 2022. Sample surveys regarding distribution in occurrence sites were sampled from each of these sites randomly. Final floristic accounts were prepared based on observations and investigations. Thebotanical identification was confirmed using floras, Monographs, and reviewing literature.The sites explore in the vicinity forest ranges of Khairlanji (21° 5' 28.65-79° 58 53.18), Sakoli, ofDistrict Bhandara, Maharashtra. The collected specimens were preserved at the Department of Botany D. D. Bhoyar College of Arts and Science, Mouda District Nagpur, MS India.

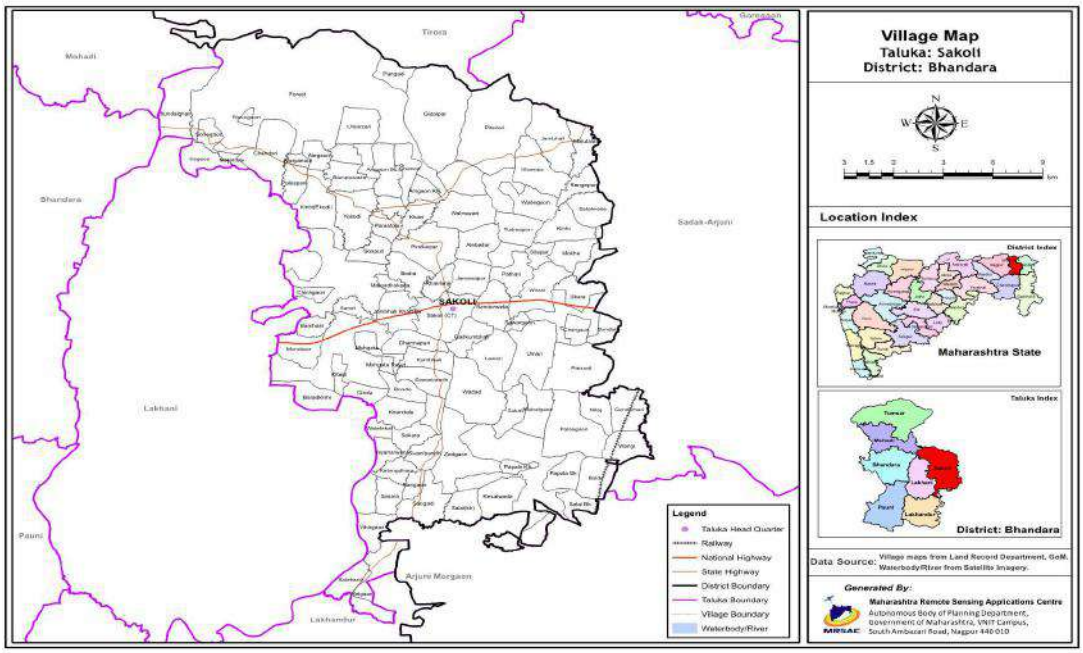

Fig. 1: Map of Sakoli Tehsil of District Bhandara, Maharashtra

Figure 2. *Ophioglossum parvifolium***(A)-(B)**. Entire plant in field**(C).**Trophophyll-position, size, shape fertile segment, and rhizoids.

Taxonomic treatment:

Terrestrial herb, 4.5-5.5 cm height; tropophylls 1-2, 0.5-1 cm long, 0.4-0.6 nm broad, ovate-lanceolate (Lindly), acute-apiculate, base cuneate, entire, veins obscure, four or five veinspassing, up through the stalk of the blade, nearly sessile, tropophylls found flat over the substratum forming an angle of 90^0 with the fertile segment. arrange parallelly with substratum soil surface; fertile segment 3.5 -5.5 cm long; strobili 0.5-1.3 long, 0.2-0.3 mm long broad, 4-8 pairs of sporangia are arranged into two alternate rows; spore 20-40 µ diameter; rhizome tuberous, erect, sometimes branched, rhizoids few, stoloniferous.

Distribution:
The worldwide distribution of *Ophioglossum parvifolium* Grev. & Hook is recorded from India, China, South America, Sumatra, Malaysia, and Thailand and distribution in India marked in Madhya Pradesh, Gujarat, Maharashtra,Karnataka, and West Bengal. However, *O. parvifolium* distribution in Maharashtra is first time recorded by Patil et al., 2014 from Panchgani, Ghanbi, Radhanagari, Dajipur, Ajara, Tilhari Nagar, Achirne, Fonda. The present work reveals the distribution from Khairlanji and Usagaon, Sakoli district Bhandara extends the distribution toward Eastern Maharashtra.

3. Results and Discussion

Sites studied during a survey of present work, in the vicinity forest of Khairlanji, (21° 5' 28.65-79° 58 53.18) and Usagaon, Sakoli,of District Bhandara, Maharashtra. The new population observed along with previously reported species of *Ophioglossum* L. Recorded observation at the in-situ occurrence site and other morpho-taxonomicalinvestigations of the specimen revealed that the species is *Ophioglossum parvifolium*Grev. & Hook, which is verified using available literature, (1–6,6)During the studies the different sites were investigated for the diversity of *Ophioglossum* L., Midst studied sites, two sites shows the sporadic, patchy, and clumped distribution of *O. parvifollium.* 70-100 sq. meter area.

4. Conclusion

Ophioglossum parvifolium has been reported in the present study from the Bhandara district of Eastern Maharashtra. The species is entirely new to the Bhandara district of eastern Maharashtra, India. This species was observed in open grasslands and clearings among trees, often found alongside other *Ophioglossum* L. species. The population of this species is likely concentrated in this region, but further comprehensive floristic explorations are necessary in similar ecological areasto precisely map its complete distribution range. The species might be distributed in similar ecological areas in the district range.

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19

Micromorphological Characterization of *Nelsonia canescence* (Lam.)Spreng: A Medicinal Herb

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Abstract
Plants are used from many years for the treatment of diseases throughout the world. Medicinal plants play an important role in the health of people living in rural societies to promote healing by traditional medical practitioners. Herbal medicines offer safe and effective treatment and are in great demand in the developed world for primary health care. *Nelsoniacanescens* (Lam.) Spreng (syn. *Justicia brunelloides* Lam.) is an important medicinal plant belonging to family Acanthaceae. It is commonly known as Blue pussyleaf (Eng), Sunga-pat (Garo) and Paramul (Bengali). This plant is common in moist and shady forest floors.The whole plant and all the parts of plant are used as medicine. To some extent, though an important medicinal plant there is no data available for drug characterization. Drug characterization is an important to understand the purity of drug. Here an attempt to study the macro and micro morphology of plant organ i.e. structure of root, stem, vessel elements of root and stem, leaf, leaf architecture and trichomes are studied.
Keywords *– Nelsonia canescens* (Lam.) Spreng*, medicinal plant, morphology, anatomy, leaf architechure, trichomes etc.*

Introduction

Medicinal plants play an important role in the health of people living in rural societies (Focho et al. 2009), where herbs have been used to promote healing by traditional medical practitioners. In Africa the use of medicinal plants has been the unique health care for 4000 years, long before the advent of orthodox medicine (Silverthorn 2010). The World Health Organization (WHO) has defined traditional medicine (including herbal drugs) as comprising therapeutic practices that have been in existence, for hundreds of years, before the development and spread of modern medicine are still in use today (Fahn,1989). Moreover, they reports that even today about 80% of people in developing countries depend on phytotherapy for their well-being (Mathias 2007). Herbal medicines offer safe and effective treatment and are in great demand in the developed world for primary health care. Now a days, they are also used as therapeutic drugs for age-related disorders such as memory loss, osteoporosis, immune disorders for which no modern medicine is available in the market (Kamboj, 2000).
Nelsoniacanescens (Lam.) Spreng (Acanthaceae) (syn. *Justicia brunelloides* Lam.) is an erect or diffused villious medicinal herbcommonly known as Blue pussyleaf (Eng), Sunga-pat (Garo) and Paramul (Bengali). This plant is common in moist and shady forest floors. It is useful in the treatment of pain, chickenpox, constipation and gastric ulcer. Take a full cup of the plant decoction at least three times a day or take after a meal instead of water. Pharmacologically it has an analgesic and anti-inflammatory activities (Owoyele et al.2005). The whole plant is used as a pest protectant for the storage of maize and sorghum by farmers of tropical African zone (Ngamo et al. 2007). Fresh juice of the whole plant is taken orally in difficult delivery (Focho et al. 2009). Decoction of the whole plant is used in wounds, diarrhoea, syphilis, gastric problems, blister and boils on tongue (Adhikari et al. 2010). Traditionally, the root is used by the traditional healers of Odisha (in the local name of

Badarasna-Rasna) for the treatment of various inflammatory and painful conditions, especially in arthritic conditions. (Brahman and Saxena (1990), Mohaddesi, 2013). It has been used for a long time in diverse contexts, i.e. as an ornamental plant, antioxidant (Sawadogo et al. 2006), antibacterial, anti-inflammatory, analgesic, purgative, anti- spasmodic, anti-ulcer (Owoyele et al. 2007), hepatoprotective (Dasgupta et al. 2012), anti- cancer and Ache inhibitor (Nabere et al. 2013).

MATERIAL AND METHODS

Plant material was collected from Amravati Dist. Maharashtra. Anatomy of root, stem and leaf was studied. For the anatomical studies freshly hand cut sections were taken and observed under microscope and sketches were made by camera lucida. Dried pieces of old root and stem were selected for maceration to observe vessel elements. Thin slices of roots and stems were treated with macerating fluid prepared by mixing 5% solution of HNO_3 and 5% solution of $K_2Cr_2O_7$ for 12 to 24 hours in cavity blocks. The macerate was then thoroughly washed with water and vessel elements were stained with 1% aqueous safranin and mounted in glycerin. Measurements were made by occular scale lens and camera lucida sketches were drawn. Classification of Radford et al. (1974) is followed for categorizing the vessel elements. Stomatal types described by following the Paliwal (1966a &b). Leaf constants such as stomatal frequency, stomatal index, palisade to spongy ratio (as seen in t.s.), PR value were determined (Kokate et al. 1996).

RESULTS AND DISCUSSION

Macromorphology

A diffuse herb, branching loosely; stem, short, hairy, decumbent spreading. Leaves chartaceous, opposite, elliptic-oblong, obtuse, entire, 1 - 1.5 x 0.5 - 0.8 cm long, shortly petiolate or subsessile, pubescent. Flowers in spikes; spike 3.5-5cm long; bract 8 x 5 mm, broadly ovate, obtuse, herbaceous, silky pubescent; bracteoles represent by 2 opposite silky hairs below the calyx. Calyx 4, partite, 0.5 - 0.7 x 0.2 cm long, sepals slightly unequal, lanceolate, very acute with strong parallel nerves. Corolla 1 - 1.5 cm, purplish. Capsule 5 x 3 mm ovoid, glabrous; seed rounded, brown, granular, hammer headed trichomes present on the margin of seed.

Micromorphology

Root: Root triarch. Pith absent (**Fig. 1a**). Endodermis distinct. Cortex narrow. Cambium produced normally. Behavior of cambium anomalous opposite protoxylem. It produces only phloem to outer side, no tissue produced towards inner side. Elsewhere secondary xylem is formed towards inner side and secondary phloem towards outer side normally. Vessels scattered, solitary or paired. Rays uniseriate. Vessel elements cylindrical as well as variously angled. Perforation plates mainly horizontal, few oblique. Tails on one side/both sides; short or long **(Fig. 1c).** It is extremely small (Class A)126 - 156 **x** 27 - 48 µm, very short (Class B) 189 - 249 **x** 15 - 36 µm, moderately short (Class C) 267 - 336 **x** 27 - 39 µm, medium sized (Class D) 354 - 555 **x** 33 - 42 µm. Phloem with scattered stone cells. Cortical cells large dividing tangentially to keep pace with growing girth. Cork cambium superficial; however, further development of cork not seen. **(Fig. 1b)**

Stem: Young stem cylindrical (**Fig. 2a**). Epidermis single layered, showing chlorophyllose bands with elevated stomata; stomata diacytic, monocyclic and non chlorophyllose bands; cells cutinised and cuticularised (**Fig. 2b**). Hypodermis collenchymatous, 1 - 2 layered interrupted by chlorenchyma below the stomata. Cortex parenchymatous; cells thin-walled with small intercellular spaces. Endodermis distinct; pericycle single layered. Vasculature uniform over the circumference. Outer and inner phloem with stone cells. On the lateral sides internal phloem is almost in the form of narrow continuous band interrupted at few places by 1or 2 tiers of

parenchyma. On dorsiventral sides internal phloem is in the form of small discrete patches. Pith small; cells parenchymatous, enclosing small intercellular spaces. **(Fig. 2c)**

Secondary growth normal in early stages. Phloem zone broad and irregular. During later stage of growth at places cambium ceases to form xylem. It produces only phloem to outer side thus making the zone of xylem irregular in outline. Simultaneously at few places pericycle divides to form patches of cambium that produce only parenchyma to the inner side, making vascular cylinder more irregular in outline. Vessels scattered, solitary, paired or in series. Vessel elementnarrow, mainly cylindrical. Tails long/short; on one side or both sides. Extremely small (Class A) 129 - 168 x21 - 27 µm, very short (Class B) 189 - 210 **x** 18 - 30 µm, ,moderately short (Class C) 255 - 288 x15 - 27 µm, medium sized (Class D) 465 - 800 **x** 36 µm **(Fig. 2f)**. Rays uniseriate. Internal phloem with stone cells. Cells of cortex divide tangentially to keep pace with growing girth. Large stone cells present in cortex. Basically stem becomes thick at node mainly by addition of cortical parenchyma. Towards node, growth becomes asymmetric, more on dorsiventral sides**(Fig. 2d & e)**

Node: Unilacunar single trace.In the node itself as trace departs to leaf it branches laterally and 3 traces enter the leaf base. Middle one in the form of large bundle and two lateral as small bundles.

Petiole: Upper surface flat with two lateral wings. Epidermis cutinised and cuticularised. Hypodermis collenchymatous, 1-3 layered. Ground tissue parenchymatous; cells thin-walled enclosing small intercellular spaces. Vasculature in the form of large central crescent with small bundles in wings. Vessels arranged in series separated by thin-walled cells. Phloem with stone cells forming interrupted layer outside xylem. Patches of internal phloem lie scattered towards protoxylem, with scattered stone cells. **(Fig. 3a & b)**

Lamina: Amphistomatous; epidermis cutinised and cuticularised, cells shallowly sinuous; cells almost polygonal in upper epidermis. Stomata slightly elevated, dicytic; bicyclic or hemibicyclic. **(Fig. 4a & b)**

Mesophyll with two distinct zones. Upper zone 4 - 5 layered; cells roughly rectangular, compactly placed, densely chlorophyllose. Lower zone 2 - 3 layered; cells isodiametric, comparatively larger, parenchymatous, with very few chloroplasts and each with a good amount of raphides and styloides. Vein bundle embedded in mesophyll. Raphide present in vein sheath also. **(Fig. 5a)** Palisade zone continuous upto margin. **(Fig. 5b)**

Midrib: Epidermis single layered, cutinised and cuticularised. Hypodermis collenchymatous; 1 - 3 layered. Ground tissue parenchymatous, cells thin walled enclosing small intercellular spaces with scattered large stone cells. Vasculature in the form of large central crescent. Outer phloem with scattered stone cells. Small patches of internal phloem lie scattered towards protoxylem side with scattered stone cells. **(Fig. 6a & b)**

Venation: -Eucamptodromous**(Fig. 7a).** Primary vein massive, straight, unbranched. 4-5 secondary veins along one side of midrib. Angle of divergence 45^0 - 50^0, acute moderate**,** Divergence angle nearly uniform. Relative thickness of secondary veins moderate, recurved, unbranched. Intersecondary veins composite. Tertiary veins**r**andom reticulate. Higher vein orderdistinct. Quaternary veins (4^0) normal. Highest vein order of leaf 4^0showing excurrent branching - 3^0. Marginal ultimate venation is looped **(Fig. 7b).** Veinlets **s**imple linear as well as branched once, simple linear more frequent. Areoles imperfect, irregular, random, medium.

Leaf constants:-

A. Epidermis	**Upper epidermis**	**Lower epidermis**
Epidermal cell dimensions	79.6 ± 1.642 **x** 36.6 ± 0.991 **x** 30µm	69.7 ± 1.184 **x** 26.2 ± 0.721 **x** 24 µm
Stomata dimensions	30 ± 0.350 **x** 14.8 ± 0.192 µm	28.5 ± 0.331 **x** 15.4 ± 0.219 µm

Stomatal frequency	28.4/mm^2	68.6/mm^2
Stomatal index	26.66 %	31.25 %

B. **Leaf dimensions** (in v.s. / t.s.) -
Thickness of lamina – 180 ± 1.192 µm
Height of palisade tissue – 81 ± 1.712 µm Height of spongy tissue – 45 ± 2.148 µm
Palisade : Spongy - 1 : 0.55
C. PR – 12.7
Trichomes - Simple as well as glandular from stem and leaf.
Simple trichomes: - Smooth, 2 – 5 celled 0.06µm to 0.4µm.
Glandular trichomes: - Sessile as well as stalked. Sessile 2- celled head. Stalk 2 - 3 celled; 0.06 µm to 0.07 µm head 2 - 3 celled. Head hammer like, cells arranged in a horizontal row.

Discussion

In most of the respect anatomy of *Nelsoniacanescens* (Lam.) Spreng is in confirmation with general anatomical feature of Acanthaceae (Metcalf and Chalk, 1950). However, many feature together characterised to the herb. These are- 1. Root Triarch. Behavior of cambium anomalous opposite the protoxylem, produces only phloem to outer side, no tissue produced towards inner side,stone cells present in outer phlom, 2. Stem cylindrical, vasculature uniform, outer and inner phloem with stone cells. On the lateral sides internal phloem is almost in the form of narrow continuous band interrupted at few places by 1or 2 tiers of parenchyma. On dorsiventral sides internal phloem in the form of small discrete patches. During later stage at places cambium ceases to form xylem and produces only phloem to outer side thus making the zone of xylem irregular in outline. Simultaneously at few places pericycle divides to form patches of cambium that produce only parenchyma to the inner side, making vascular cylinder more irregular in outline. Towards node, growth becomes asymmetric, more on dorsiventral sides,3. Class A, B, C and D types of vessel elements present in root and stem also. 4. Vasculature in the form of large central crescent present in petiole and midrib, 5. Stomata slightly elevated, dicytic; bicyclic or hemibicyclic, 6. Venation eucamptodromous, 7. Raphaids and styloids were present in lower zone of mesophyll, 8. Simple as well as glandular trichomes present in stem and leaf. All these microscopic characters reported in this paper could be used as diagnostic tool for the identification and standardisation of crude drug material.

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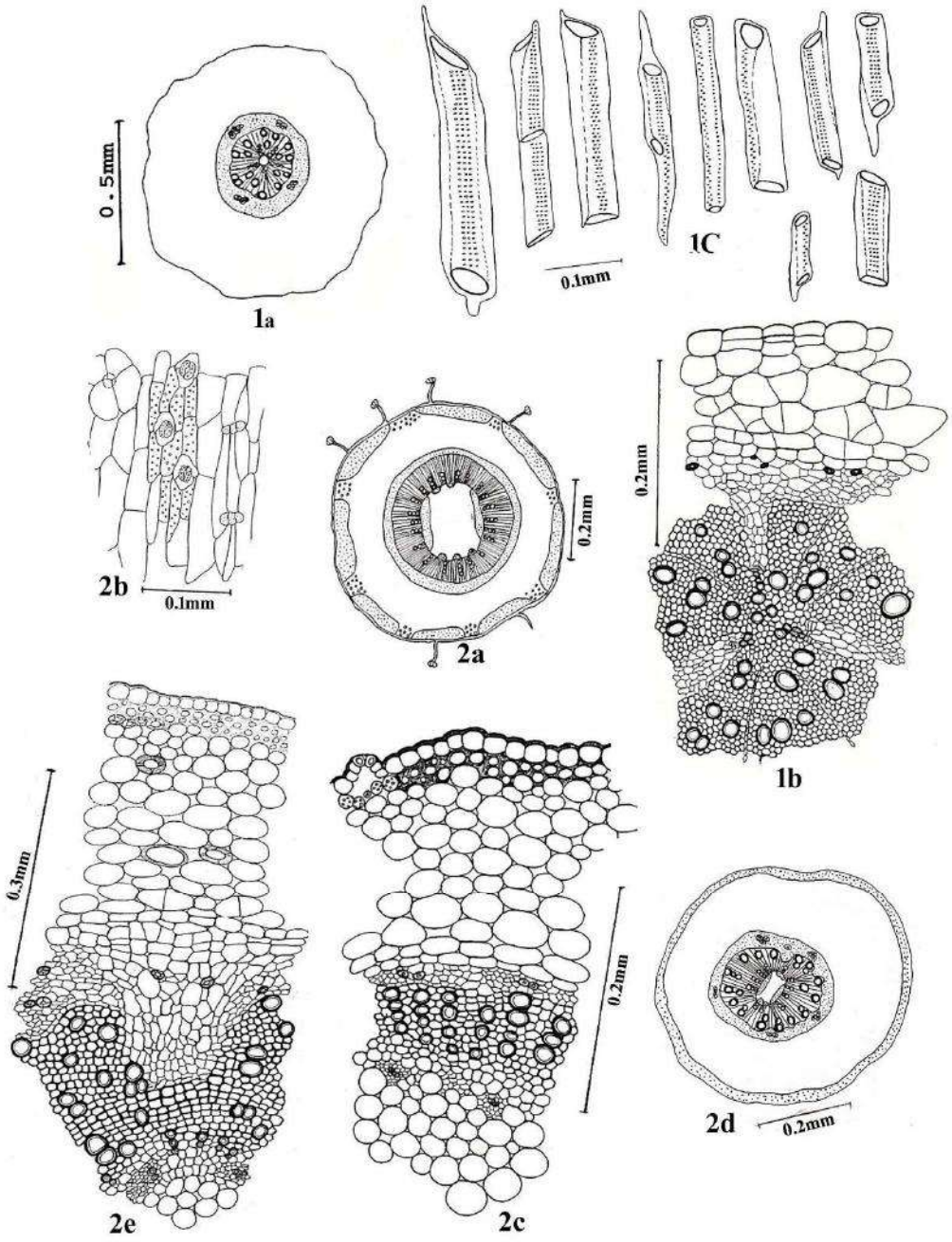

PLATE - I

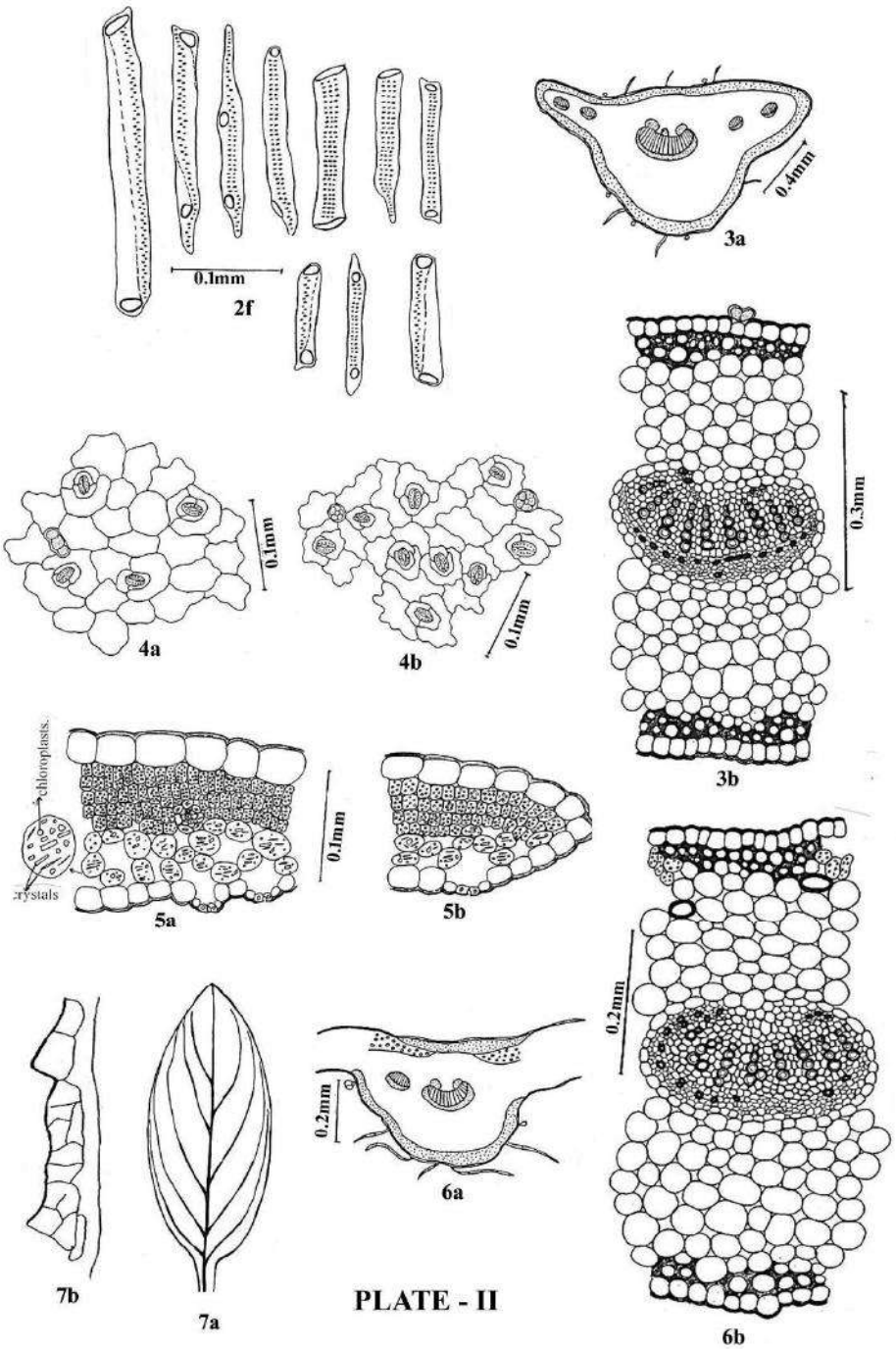
0.1mm
2f
0.4mm
3a
0.3mm
3b
0.1mm
4a
0.1mm
4b
chloroplasts
crystals
0.1mm
5a
5b
0.2mm
6a
0.2mm
6b
7b
7a

PLATE - II

20

Morphological and Anatomical Study of leaf of Ficus *krishnae* C. DC

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Abstract:
Current investigation was made on anatomy of leaf of Ficus krishnae C. DC. from Moraceae family. Many mythological stories are seen regarding the Ficus krishnae C. DC. The aim was to contribute with more information concerning the knowledge of anatomy. In this paper included study of microscopic (anatomical) characters of leaf. Ficus krishnae C. DC. belongs to the family Moraceae, less reports were available on anatomical studies, hence present efforts were undertaken to investigate the microscopic studies.
Keywords: *Ficus krishnae* C. DC, Anatomy, Crystals, tannin cells etc.

1. Introduction :

Genus Ficus is remarkable for the large variation in the habits of its species. Some species of Ficus shows remarkable development of aerial roots and they may grow as epiphytes. Moraceae is one of the largest family in dicotyledones, distributed all over India and various part of the world mostly in tropical region. *Ficus krishnae* C.DC is a very uncommon species. It shows presence of cup-shaped leaves. It is in large extent found in India, Sri Lanka and tropical Africa. It is large, fast growing, evergreen tree grows up to 30 m tall, with spreading branches and aerial roots. *Ficus krishnae* C.is worshiped in India. Many mythological stories are seen regarding the formation of cone-shaped leaves are associated with this species. In spite of having several morphological differences, *F. krishnae* C.DC is considered by some authors as a synonym of *F. benghalensis* L. which does not seem to be considerable. (Anand *et al*. 2016). *Ficus krishnae* C. DC is a perennial plant, it is used in number of folklore medicine, to treat various diseases like ulcer, vomiting, fever, diabetes, dysentery, inflammation, leprosy and cancer etc. (Kanjikar and Londonkar 2017). Ficus krishnae have an anti-diabetic effect in alloxan induced diabetic rats and their effect was equivalent to that of reference drug glibenclamide. (Mohana lakshmi et al., 2010). the leaves are also known as 'Makkhan Katori' (butter cup). God Krishna used to steal makkhan, once when his mother caught him stealing makkhan, he folded the leaves in the form of cone to hide makkhan, Then produces cone shaped leaves. This is mythology behind cone shaped leaves (Tiwari et al., 2015).

2. Materials and Methods :

The plant material of *Ficus krishnae* C.DC Were Collected from Nagarjun garden, Akola (dist.), Maharashtra, India. It was not easily available like other species of Ficus. The required specimen samples stem and leaves were cut and preserved in formalin water solution at room temperature. Hand cut sections had been taken and permanent slides were prepared. Slides were studied under different magnifications of light microscope, and anatomical feature were noted. Photomicrography of slides was taken in Department Of Botany of Govt. Vidarbha Institute of Science And Humanities, Amravati, Maharashtra. using trinocular microscope and A.S.A. 200 film..

3. Result and Discussion:

3.1 Morphology

Vernacular Names: English- Krishna fig, Krishna's butter cup, Hindi- Makkhann Katori, Marathi- Krishnavad

Medium size Tree, grows up to 20 m in height, solid, much branched, glabrous, dull whitish, leaf simple, form pouch at base cupuliform (peculiar leaves), alternate, 10-20 cm long and 5-10 cm broad, leaflet like appendages on the petiole. Petiole 3-10 cm long, ovate lanceolate, entire, acute or blunty acuminate, glabrous, reticulate unicostate venation, Inflorescence hypanthodium type. Fruit a syconium, globose, red coloured when riped.

Fruiting Period: Nov- Feb

Distribution: Not common, planted in gardens. (Nagarjun Garden Akola, Botanical garden of Dr. Babasaheb Ambedkar Marathwada University, Ch. Sambhajinagar)

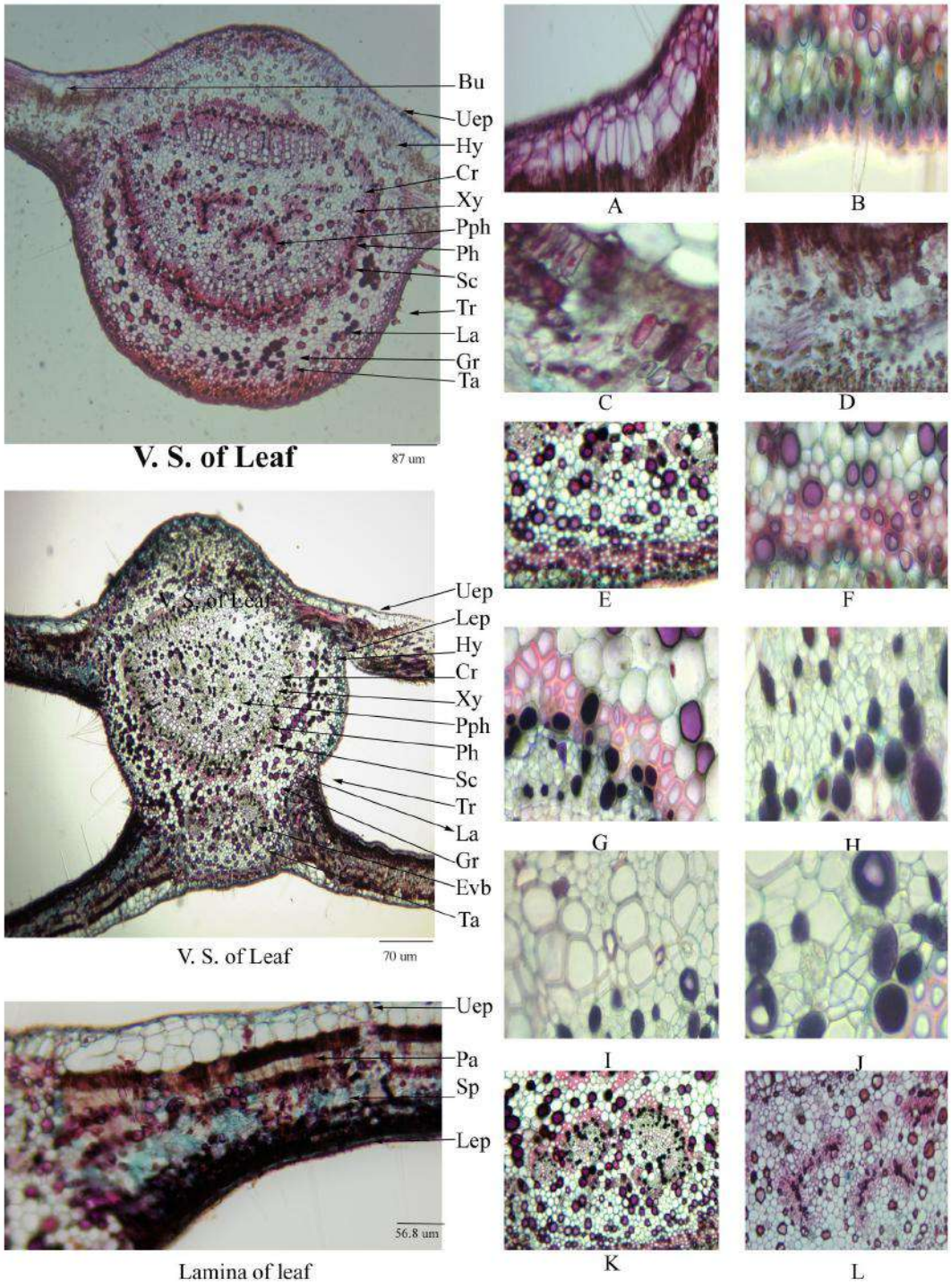
Microscopic Characters

Leaf of *Ficus Krishnae* C. DC. showed two and four lamina arm structure in two different sections of the same leaf. Leaf section through midrib showed two lamina arms but when the section had been taken through cup shape structure it showed four lamina arms structure, all four lamina arm had nearly same anatomical structure.

A vertical section of leaf showed dorsiventral structure. Unicellular, uniserriate trichomes were abundant, pointed at apex and broader at base. Stomata sunken, present on lower epidermal surface and absent on upper epidermal surface. Cuticle very thick with wavy outline. Upper epidermis 1-3 layered, cells thin walled, loosely arranged; rectangular, squarish and polygonal in shape. Bulliform cells present on upper epidermis. Recorded average 23±11.5×26.8±17.5 µm and range 11.5-34.5×11.5-46 µm. Lower epidermis single layered with rectangular, squarish and oval in shape; cells smaller in size as compare to upper epidermal cells. Recorded average 9.96±3.51×9.2±2.3 µm and range 6.9-9.2×6.9-11.5 µm.

Mesophyll differentiated into spongy and palisade tissues. Palisade mesophyll 2-3 layered, elongated in shape, filled with chloroplasts. Recorded average 16.8±1.32×5.36±1.32 µm and range 16.1-18.4×4.6-6.9 µm. Below the palisade tissue present spongy mesophyll; cells irregular in shape with intercellular spaces. Recorded average 9.2±4.6×10.7±7.02 µm and range 4.6-13.8×4.6-18.4 µm. Presence of numerous laticifers were recorded.

Midrib region showed semi-circular shape. Hypodermis composed of thick walled lacunar collenchyma; 2-5 layered; cells oval, circular and hexagonal in shape. Recorded average 9.2±4.6×9.2±4.6 µm and range 4.6-13.8×4.6-13.8 µm. Below the hypodermis, 7-8 layered parenchymatous ground tissues present. Recorded average 19.9±9.57×21.46±11.5 µm and range 9.2-27.6×9.2-32.2 µm. Number of vascular bundles arranged in circular strcture. Sclereids and sclerenchyma formed cap like structure on vascular bundles; 2-4 layered, compactly arranged, circular, oval, polygonal and irregular in shape. Recorded average 8.05±3.04×8.43±3.51 µm and range 4.6-10.35×4.6-11.5 µm. In the section of four arms two extra vascular bundles were notes above the lower epidermis and between two lamina arms of pouch. Phloem 4-6 layered; cells squarish, rectangular and oval in shape. Recorded average 4.6±1.15×4.21±0.663 µm and range 3.45-5.75×3.45-4.6 µm. Cambium not observed. Xylem vessels circular to oval in outline, arranged in rows, in radial multiple of 4-6. Recorded average 22.2±12.6×24.53±16.1 µm and range 9.2-34.5×9.2-41.4 µm. Perimedullay phloem bundles present in pith region along with some xylem patches. Sphaeraphides crystals, solitary crystals, tannin cells, laticifers scattered abundantly.



V. S. of Leaf

V. S. of Leaf

Lamina of leaf

V. S. of Leaf: (M-4X) Tr- Trichome, Uep- Upper epidermis, Lep- Lower epidermis, Pa- Palisade tissue, Sp- Spongy tissue, Bu-Bulliform cells, Hy- Hypodermis, Gr- Ground tissue, Sc- Sclerenchyma fibres, Ph- Phloem, Ca- Cambium, Xy- Xylem, Pph- Perimedullary phloem patche, La- Laticifers, Cr- Crystals Evb- Extra vascular bundles, Ta- Tannin cells

A- Upper epidermis with some bulliform cells, B- Lower epidermis, C- Palisade tissue, D- Spongy tissue, E- Hypodermis and ground tissue, F- Lacunar collenchyma tissue, G- Sclerenchyma fibres, H- Phloem, I- Xylem, J- Crstals, K- Extra vascular bundles, L- Perimedullary phloem patches

4. Conclusion:

Anatomical information of leaf of *Ficus Krishnae* C. DC. will be very useful for researchers and students of Botany. Laticifers, tannin cells and rhomboidal crystals present in cortex. Endodermis not prominently observed. Laticifers observed in phloem region. Starch grains observed in parenchyma all the tissues. Tannin cells and solitary crystals present abundantly in peripheral region and scattered in all the tissues.

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Zoology

1

3D Printing Technology for development of Transdermal Drug Delivery Systems

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Abstract:
The innovative approach of three-dimensional printing enables the on-demand production of transdermal drug delivery systems. This technology, already applied in dentistry, orthopedics, and pharmaceuticals, particularly stands out in the latter field. It facilitates the printing of medical devices and diverse formulations of active pharmaceutical ingredients, featuring controlled-release characteristics and varied geometries. This study provides an overview of these pharmaceutical applications, focusing on the 3D printing of transdermal patches. The discussion encompasses different printing technologies and material systems known for their customization capabilities, generating intricate geometries with precise characteristics crucial for transdermal systems, thereby enhancing bioavailability. The study includes case studies, explores the advantages and limitations of the technology, and forecasts industry growth, projecting a value exceeding USD 8 billion by 2025. Despite this potential, the conservative nature of the pharmaceutical industry leans toward cost-effective methods for large-scale production. Nevertheless, 3D printing has the potential to revolutionize the current 'one size fits all' manufacturing approach, becoming an integral part of the drug development timeline.
Keywords: 3D Printing, Inkjet Printing, Microneedles, Patches, Transdermal Delivery, Pharmaceutics

Introduction

The historical application of therapeutic substances, including herbal ointments and various drugs (such as scopolamine, estradiol, fentanyl, and rivastigmine), on the human skin serves both medical and cosmetic purposes. Over the past decades, the skin has proven to be an accessible surface for drug administration, making systematic therapy through percutaneous drug absorption feasible (Prausnitz and Langer, 2008; Alkilani et al., 2015; Pastore et al., 2015). The transdermal route emerges as an attractive alternative to traditional methods like oral administration or hypodermic injections. Oral administration may face issues of partial drug absorption, complications related to gastrointestinal metabolism, and slow onset, making it impractical for emergency cases. Hypodermic injections, while effective, are invasive, pose infection risks, require skilled administration, and generate medical waste (Awodele et al., 2016).

Contrarily, transdermal systems offer advantages such as bypassing metabolic systems, ensuring higher bioavailability, and promoting sustained and controlled drug release. Additionally, transdermal drug delivery (TDD) holds promise for vaccinations due to the abundance of dendritic cells in the skin. This patient-friendly approach is noninvasive, contributes to psychological well-being, and provides independence as it doesn't require professional care for repositioning, removal, or replacement.

However, TDD faces limitations, primarily stemming from the skin barrier's nature. The stratum corneum, the outermost skin layer, acts as a significant barrier due to its density and low hydration (15–20%). Overcoming this impermeable barrier has been the focus of TDD research, presenting both challenges and opportunities for future progress.

In the current global trend towards personalized patient care, traditional mass-production methods of drug delivery systems are being questioned for their ability to tailor dosages cost-effectively. New technologies, particularly additive manufacturing (AM), such as 3D printing,

are being investigated for their potential in pharmaceutical technology. Initially introduced in the 1980s, 3D printing has gained attention across various industries, contributing to the production of complex structures beyond the capabilities of conventional techniques.

The application of 3D printing in pharmaceuticals is relatively recent, aiming to produce targeted-release and customized drug delivery systems (Goole and Amighi, 2016). In the field of TDD, although studies are limited, they demonstrate the transformative potential of 3D printing. This review explores the existing research on 2D and 3D printing as direct or indirect fabrication methods for TDD systems. The materials and drugs associated with 3D printing in TDD systems are also examined in this context.

3D-printing techniques for optimized transdermal drug delivery systems

Additive Manufacturing (AM), commonly referred to as 3D printing or Solid Freeform Fabrication (SFF), encompasses various techniques that utilize a virtual Computer Aided Design (CAD) model to construct a physical object by depositing consecutive layers. Introduced in the 1980s, 3D printing has revolutionized industrial and scientific sectors, offering fast and precise production of intricate structures beyond the capabilities of traditional methods. The medical field quickly recognized the transformative potential of 3D printing, leading to the creation of customized implants and prosthetics and ongoing investigations into live tissue printing (Chia and Wu, 2015).

The application of 3D printing in drug delivery has recently gained attention with the FDA approval of Spritam, the first 3D-printed oral administration tablet. This has given rise to the term 'pharmacoprinting' (Prasad and Smyth, 2015; Jacob et al., 2014; Goyanes et al., 2015; Di Prima et al., 2016). While its impact on oral drug delivery is well established, 3D printing's potential in transdermal drug delivery (TDD) is currently under exploration, with a growing body of relevant studies.

Inkjet Printing

Inkjet printing, involving the controlled deposition of small droplets, has seen successful applications in medicine but is yet to be extensively explored for TDD. Studies have used inkjet printing for coating microneedles with various agents, demonstrating its potential for controlled and selective deposition on suitable substrates (Boehm et al., 2011, 2013, 2014; Ross et al., 2015; Uddin et al., 2015). While its application for building complex three-dimensional TDD structures remains unexplored, inkjet printing's high resolution and selective deposition make it promising for microneedle coating, enabling personalized dosages with high reproducibility.

Photopolymerization-based Technologies

A significant group of 3D printing technologies relies on selective polymerization of photo-sensitive polymers through laser emissions or light projections. Techniques like stereolithography (SLA) and digital light processing (DLP) enable layer-wise polymerization of UV-sensitive polymers. These technologies offer versatility in geometric complexity and resolution, making them suitable for TDD applications. Studies have utilized micro-stereolithography (DLP) to create microneedle arrays indirectly, contributing to the customization of therapeutic approaches (Boehm et al., 2014, 2011, 2012). Photopolymerization-based 3D printing has proven applications in fabricating TDD systems, offering high resolution and flexibility.

Fused Deposition Modeling (FDM)

Fused Deposition Modeling (FDM), based on the melt-extrusion process, is a versatile 3D printing technique with potential applications in TDD. While FDM's limitations in resolution and sensitivity to process parameters are acknowledged, its ability to produce structures through extrusion without solvents makes it a compelling choice for certain pharmaceutical applications. The combination of FDM with hot-melt extrusion (HME) processes holds promise for producing drug/polymer blends for cost-effective TDD system fabrication.

Materials:

In the contemporary landscape, 3D printing technologies possess the capability to manipulate a diverse array of materials, ranging from ceramics and metals to polymers. The categorization of these techniques implies that the choice of a specific technology inherently limits the materials compatible with the corresponding printing apparatus. For instance, SLA or DLP printers exclusively handle photo-cured polymers, while FDM printers utilize thermoplastic filaments. This limitation poses challenges for the widespread adoption of 3D printing as a direct manufacturing technique for transdermal drug delivery (TDD) systems, as the material must meet specific criteria for integration into such systems. (Sharma et al., 2011) Essential parameters include stability, biodegradability without toxic by-products, mechanical strength, and non-reactivity with the drug. Material biocompatibility is a critical consideration, as evidenced by studies involving Gantrez, a biocompatible copolymer used in TDD applications. While there's evidence of manufacturing Gantrez biocompatible microneedles using 3D-printed molds, the multi-step nature of this approach may hinder mass production scalability. (Boehm et al., 2014, 2011, 2012; Donnelly et al., 2012).

Numerous polymers with biomedical and pharmaceutical applications show promise for integration into 3D-printed TDD systems. Polyvinyl alcohol (PVA) and polylactic acid (PLA) are examples. However, challenges exist, such as PVA's limited biodegradability and PLA's slow degradation rates and poor mechanical properties when employed in FDM technology. Biopolymers, like chitosan and collagen, exhibit favorable attributes for TDD. Bioprinting advancements further expand the possibilities. Yet, ongoing research on materials remains vital for the evolving field of 3D-printed TDD. (Economidou et al. 2018)

3D Printed Transdermal Drug Delivery Systems: Future Challenges and Expected Impact:

Transdermal Drug Delivery (TDD) systems, facilitated by 3D printing, hold potential as a user-friendly, personalized pharmaceutical therapy. The layer-by-layer fabrication inherent in 3D printing aligns well with TDD requirements. This technology enables the creation of systems with varying drug concentrations across layers, catering to individual needs. Customization possibilities enhance TDD efficiency by addressing factors like skin hydration and thickness variations among patients. In vaccination, microneedles offer promise, particularly in regions facing challenges with traditional administration methods. 3D printing's role in reducing costs and providing needle-free solutions is crucial for global health initiatives. However, challenges such as limited biomaterial options, dosing constraints, and drug degradation characteristics need resolution. The development of 3D printable materials and improvements in existing technologies could drive the evolution of TDD systems. (Economidou et al. 2018)

Regulatory Considerations:

For the commercialization of 3D-printed TDD systems, adherence to regulatory requirements is essential. Despite the FDA's approval of the first 3D-printed oral tablet, TDD systems face unique regulatory challenges. Microneedle patches, viewed as medical devices, must adhere to Good Manufacturing Practice (GMP) guidelines. The FDA emphasizes technical considerations, including the impact of printing parameters, in-situ quality control, design validation, sterilization, and post-process cleaning. A 2017 guidance document provides recommendations for 3D-printed medical devices, addressing issues like patient-matched devices and data protection. Sterilization, a regulatory requirement, presents challenges for microneedles. Future success depends on addressing material limitations, improving technology, and establishing specific regulatory frameworks for 3D-printed TDD systems. (Economidou et al. 2018)

Conclusion:

Since its inception, 3D printing has revolutionized fabrication methods, with potential applications in medicine and pharmaceutics. Despite being in the early stages, advancements in 3D-printed transdermal drug delivery (TDD) systems show promise. Inkjet printing, photopolymerization-based technologies, and FDM have been explored, with inkjet printing successfully depositing films on microneedle surfaces and commercializing 3D inkjet-printed microneedles. The integration of elaborate microneedle array systems with precise 3D printing techniques has the potential to reshape modern drug administration. Overcoming engineering, chemistry, and material challenges through interdisciplinary research is crucial. Regulatory considerations, addressed by the FDA, are imperative to ensure the safety and effectiveness of 3D-printed TDD systems. Success hinges on resolving material limitations, advancing technology, and establishing specific regulatory frameworks.

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2

Parasitic infections in freshwater ornamental fish

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ABSTRACT:

A total of 1520 ornamental fish of 13 species out of were collected from 26 export farms in Sri Lanka between October 1999 and March 2000 and examined for parasites. The fish species studied were Guppy Poecilia reticulata, Goldfish Carassius auratus, Platy Xiphophorus maculatus, Molly Poe cilia sphenops, Angel Pterophyllum scalare, Swordtail Xiphophorus helleri, and Tetras Hyphessobrycon sp. karass. nus carpio, destroyers Betta spe lendens and others (Brachydanio and Astronotus spp.). Nine species of monogenean trematodes(Dactylogyrus extensus, Dactylogyrus cf. extensus, D. vastator, Dactylogyrus cf. host Dactylo gyrus spp., Gyrodactylus turnbulli, G. katherineri, Gyrodactylus cf. katherineri, Gyrodactylus spp.), 7444 species of protozoa (Trichodina nigra, Trichodina spp., Tetrahymena corlissi, T. pyriformis, Ichthyo phthirius multifiliis, Ichthyobodo necator, L. Piscinoodinium spp.), Erpocypos4, Erpocypos4 gasilus ceylonensis, Argulus foliaceus), 1 metacercaria digenean trematode (Centrocestus spp.) and 1 nematode (Capillaria spp.). Parasites were found in fish from 23 out of 26 farms, the total fish prevalence was 45.3%. In the farm, the difference in prevalence between different parasites was significant (p and lt; 0.01). Fish infection rates form on ogenean trematodes, protozoa, crustaceans, digean trematodes, and nematodes were 28.3, 18.4, 4.8, 0.8, and 0.4%, respectively. Overall, 50 of 590 guppies (50/590) had Tetrahymena infections compared to 13/930 for all other species, a statistically significant result of (p and lt; 0.01). Argulus foliaceus and Lernaea cyprinacea had 13/44 and 18/44 carp, respectively, while all other species had a total of 7/1476 and 15/1476 (p andlt; 0.01). Capillaria spp. found only in guppies (4/590) and angelfish (3/92), while Centrocestus spp. was found only in goldfish (12/153).

Keywords: Parasite, Prevalence, Ornamental fish, Tropical regions.

INTRODUCTION:

Over the last two decades, the export of ornamental fish has become an important activity generating foreign exchange for Sri Lanka, with more than 300 million Sri Lankan rupees(~US$35,000) earned each year (Weer akoon 1998). Sri Lanka has a reputation as an exporter of quality marine ornamental fish. A sim porting countries have introduced regulations to prevent imports of wild-caught ornamental fish due to resource depletion and extinction (Andrews 1990), the demand for farmed freshwater ornamental fish is increasing. Although Sri Lanka's freshwater ornamental fish industry has enormous potential for development, one identified weakness is the lack of understanding of disease conditions (Mee 1993).

Parasitism is one of the most influential problems in farmed fish (Scholz 1999). Fish in intensive farming are constantly affected by environmental fluctuations and management practices such as handling, overcrowding, transport, drug treatment, malnutrition, changing temperatures and poor water quality. All these factors can significantly stress the homeostatic mechanisms of fish, making them susceptible to various parasites (Suba singhe 1997).

In addition to the direct losses caused by mortality, parasites can significantly affect fish growth and behaviour (Scholz 1999) and thus reduce farm efficiency and production, which in turn increases costs and reduces profits and externalities. Exchange income.

This study was conducted to estimate the prevalence of selected important parasites associated with ornamental fish production in Sri Lanka.

MATERIALS AND METHODS:

The study was conducted in the western and north western provinces of the country, where almost all ornamental fish export farms are located. Some of these exporters are partially dependent on small farms in nearby rural areas for a continuous supply of ornamental fish, which helps the keep costs down. In 1999, there were 30 registered ornamental fish exporters in the country. The area of freshwater ornamental fish farms varied from 0.5 to10 hectares and the monthly production varied from40, 000 to 500,000 fish.

Ornamental fish are reared in cement, glass, or fiberglass tanks and mud tanks, and exporters usually maintain their own breeding stock. More than 75% of exports consist of 4,444 guppy varieties. Guppies live and 1-day-oldlarvae are collected and processed in separate tanks. After about 3 weeks, the chicks are sexed and divided into growth units. At the age of 2-3 months, depending on the length of the fish, the guppies are ready for marketing and are quarantined for 1 week before export. The remaining 4,444 fish species are farmed longer before being exported. Nutritional programs vary by growth stage. Feed contains shrimp Artemia Salina L., tubifex worms and formulated fish meal. Most farmers use methylene blue (2 ppm) and salt as a routine hygiene measure before fish are taken to rearing units and at the beginning of the pre-export quarantine period. Before export, representative fish samples are reviewed by Animal Husbandry Department and Health Department Officer. Only whole fish are recommended for export.

Between October 1999 and March 2000, 26 registered ornamental fish export farms out of were visited in the Western and North Western Provinces of Sri Lanka. These farms accounted for 87 percent of registered exporting companies, and 95 percent of all exporting companies. Total export of ornamental fish in 1999. At each farm, tanks/ponds were numbered and tanks/ponds were randomly selected (Fowler and Cohen 1994) from which fish were selected for study. In this way, a total of1520 fish from 13 species were selected (Table 1). The number of fish selected varied from 16 to 195 fish per farm. Species and growth stage are subject to availability on collection day. The smallest fish collected were guppies (length 1-3 cm), while carp was the largest (3-15 cm). Live ornamental fish were brought to the laboratory in polyethylene bags filled with oxygen-rich pond water. Each farm was visited only once, and information on farm governments was also recorded.

In the laboratory, 4,444 fish were initially examined for spots or lesions visible to the naked eye. Next, wet scrapings (body surface mucus from pectoral fin adjacent to dorsal fin and excision, cut gills, lesions and intestines) from freshly killed fish were examined using a light microscope. With ×10 and ×40 magnification. The use of fresh specimens facilitates visualization of mobile parasites (Post 1987, Southgate 1994, Wild Goose 1998). Specimens were then preserved in 10%buffered formalin and 70% ethanol for preservation prior to further identification using keys described by Yam agouti (1963), Kirti singhe (1964), Fernando and Hanek (1971, 1973), Elliot (1971) . , Cheng (1986), Li guo et al. (1991), Cone (1995), Dickerson and Dawe (1995), Lom(1995) and Hoffman (1999). Type specimens are deposited in the Department of Zoology, National Museum, and Colombo, Sri Lanka.

Prevalence rates were calculated for of each parasite genus recovered. If was a sufficient number, chi-square tests were used to compare prevalence using the computerized statistical package Minitab release 10.1 (Minitab).

RESULTS:

Identification of parasites:

The list of parasites identified from different places of different fish species is given in Table 2.18 parasite species were found, 9 of the monogenean parasites, 7 protozoan species, 3 species of toothed arthropods, 1 rev. worm and 1 metacercarial stage in a digenean trematode. Of these, 15 were identified at the species level and the remaining were identified only at the genus level (Table 2).

Table 1. Species and number of ornamental fish examined, number of different parasites recovered in different species of fish and the prevalence of parasites and number of farms infected.

Species of Ornamental fish	**No Examined**	**No infected**	**Parasite Species**											
			Dactylogyrus	Gyrodactylus	Trichodina	Tetrahymena	Ichthyophthiri	Ichthyobodo	Piscinoodinium	Lernaea	Ergasilus	Argulus	Capillaria	Centrocestus
Guppy Poecilia reticulata	590	262	91	63	42	50	0	13	0	0	14	0	4	0
Goldfish Carassius auratus	153	94	47	35	7	3	0	0	9	7	2	4	0	12
Platy Xiphophorus	143	35	15	7	11	0	5	0	0	0	2	3	0	0
Molly Poecilia sphenops	106	65	33	9	22	5	0	5	0	6	0	0	0	0
Barbs Capeota and Puntis spp.	95	36	11	7	12	1	7	3	0	0	0	0	0	0
Angel pterophyllum scalare	92	71	36	2	26	1	5	0	0	0	0	0	3	0
Fighters Betta splendens	84	30	15	4	6	0	6	0	4	0	0	0	0	0
Tetras Hyphenssobrycon sp.	75	28	12	8	10	0	0	0	0	0	0	0	0	0
Swordtail Xiphophorus helleri	66	11	05	2	2	0	0	0	0	0	2	0	0	0
Gourami Colisa sp	64	28	9	10	3	0	0	0	4	2	0	0	0	0
Carp Cyprinus carpio	44	21	6	4	0	3	0	5	2	18	0	13	0	0
Other (Brachydanio and Astronotus spp.)	08	0	0	0	0	0	0	0	0	0	0	0	0	0
Total fish	1520	689	689	151	141	63	30	26	19	33	20	20	7	12
Prevalence (%) of infected fish		45.3	45.3	9.9	9.3	4.1	2.0	1.7	1.3	2.2	1.3	1.3	0.5	0.8
Number of infected farms	26	23	23	21	13	9	4	3	5	10	5	3	4	3

Detection of Dactylogyrus extensus in goldfish and carp confirmed its large size, with body length ranging from 1120 to 1280 µm and width from 150 to 160 µm. The hamuli were relatively long (75 - 85 um) (museum registration number PS 2002-6-1). Similar specimens of A ngel fish were identified as Dactylogyrus cf. extension The identification of Dactylogyrus vatator and goldfish from plate was confirmed by its relatively small body size, 320-380 µm long and80 µm wide. Their hamules were relatively small (38-40 µm). (Museum identification numberPS-2002-6-2). Molly and guppy fish specimens with similar morphology were identified as Dactylogyrus cf. the opposite.

Specimens of Gyrodactylus katherineri collected from goldfish were identified by their large body length, which varied from 800 to 1100 µm, and hamulus length (80to 108 µm). (Museum identification number PS-2002-6-4). Similar specimens taken from angel and lava fish were identified as Gyrodactylus cf. katherine.

Specimens of Trichodina nigra collected from guppy, barbel, discus and goldfish were identified by the diameter (38-60 µm) of a sticky disc with a dark center and a tooth-shaped ring (20-35 µm) (Museum) , accession no. -2002- 6-5).

The identification of Tetrahymena Corliss from guppies was confirmed by its egg-shaped body (40 × 60 µm) and tail cilium (Museum ID number PS-2002-6-6), during the identification of. Most of T. pyriformis was dated to due to its pyriform body (50 ×30 µm) and lack of caudal cilia (Museum, accession number PS-2002-6-7).

Specimens of Ichthyophthirius multifiliis were identified by their oval to rounded body,, 500 to 800 µm in diameter, and the horseshoe-shaped macronucleus from trophodes (museum accession number PS-2002-6-8).

Specimens of the free-swimming forms of Ichthyobodo necator were identified by their oval body (10 × 5 µm), containing 2 free irregular flagella on the abdomen, and a centrally located nucleus (Museum accession numberPS-4-6 -200 PS-4). -6). 9).

individuals of Lernaea cyprinacea were identified by their length (6.5-6.8 mm, width approx. 0.6 mm), and egg sac length (between 1.4-x 1.5 mm, width approx. 0.25 mm). mm) (museum accession number PS-2002-6-10).

The identification of Ergasilus ceylonensis was confirmed based on its size (1.1-1.3 mm), barrel-shaped genital segment, and the typical random placement of large numbers of eggs in egg sacs (museum accession number PS-2002-6-11).

Table 2. Parasites recovered from different body locations of ornamental fish

Parasite	Host fish Species	Body Location
Dactylogyrus extensus	Goldfish, carp	Gill lamellae
Dactylogyrus cf. extensus	Angel	
Dactylogyrus vastator	Goldfish, Platy	Gill lamellae
Dactylogyrus cf. vastator	Guppy, molly	
Dactylogyrus spp.	Tetras, gourami, swordtail, molly, goldfish, barbs, fighters	Gill lamellae
Gyrodactylus turnbulli	Guppy, molly	Body surface, fins
Gyrodactylus katherineri	Goldfish	Body surface, fins
Gyrodactylus cf. katherineri	Angel, platy	
Gyrodactylus spp.	Tetras, gourami, swordtail, carp, goldfish, barbs, fighters	Body surface, fins
Trichodina nigra	Goldfish, guppy, barbs, platy	Body surface
Trichodina spp.	Angel, tetras, gourami, swordtail, molly, fighters	Body surface, gills
Tetrahymena corlissi	Guppy	Body surface, gills, muscles
Tetrahymena pyroformis	Guppy, Goldfish, molly, carp, angel, barbs	Body surface, gills
Ichtyophthirius Multifiliis	Angel, platy, barbs,fighters	Body surface
Ichthyobodo necator	Guppy, molly, barbs, carp	Body surface
Piscinoodinium spp.	Goldfish, fighters, carp, gourami	Body surface
Lernaea cyprinacea	Carp, gourami, molly, goldfish	Skin close to caudal fin
Ergasilus ceylonensis	Guppy, goldfdish, platy, swordtail	Gills
Argulus foliaceus	Goldfish, platy, carp	Body surface
Capillaria spp.	Guppy, angel	Gut
Centrocestus spp.	Goldfish	Gills

Specimens of Argulus foliaceus were identified by their cephalothorax size (9 mm length) with distinct thoracic regions and posterior lobes not reaching the base of the abdomen with lobes separated by a clear slit. (Museum Extension No. PS-2002-6-12).

The overall prevalence of parasitism in of 1520 fish was 45.3% (95% CI 42.8 to 47.9%). The genus Dactylogyrus (18.4% of infected fish) was the most common and Capillaria was the least common (0.4%).

Farm infection rates:

Parasitic fish were found in 26 of fish farms visited in 23 of them. The difference in national prevalence among the parasites was significant (p and lt; 0.01). Most of the farms (23/26) had fish infected with Dactylogrid; 4,444 rooms infested with gyrodactylids were also common (21/26). The lowest farm prevalence (3/26) was recorded for Ichthyobodo necator, Argulus foliaceus and Ceroces tus spp. (Table 1).

Fish infection rates:

In those parasite genera for which sufficient information was available, infection rates varied significantly among fish species (p and lt; 0.01). 50 of 590 guppies were Tetrahymena spp. -infected compared to 13/930 for all other species (p and lt; 0.01). Argulus foliaceus and Lernaea cyprinacea were infected in 13/44and 18/44 carp respectively, for all other fish species a total of 7/1476 and15/1476 (p and lt; 0.01). Dactylopyrids, gyrodactylids and trichodinids occurred on 12.11., 11.12. and 10.12. on the fish species tested, suggesting that they were not specific to any particular ornamental fish species.

DISCUSSION:

As Sri Lanka is a tropical country, one would expect a wider spectrum of parasites in the ornamental fish than in this study. Although 4,444 parasitological examinations were performed on all fish collected, the failure to find certain groups of parasites could be due to several reasons. First, most of the farms had independent water supplies to the ponds or tanks to avoid cross-contamination, and the ponds and tanks were emptied and cleaned after each production cycle. Second, the ponds and tanks in study farm were covered with wire mesh to prevent access to the area by birds and other animals that could act as definitive hosts of digenean trematodes. Finally, fish were collected from export farms with common antiparasitic compounds such as formalin, malachite green, acriflavine and methylene blue. However, the general prevalence of parasitic fish in these farms was high (45.3%), although most infections were monospecific (37.9% of fish). However, the presence of detectable parasites at all levels is significant because the fish studied were from populations intended for export and any rejections during quarantine certification could negatively affect the generation of courses.

Dactylopyrids (Dactylogyrus cf. extensus) isolated from angelfish were morphologically similar to carp and goldfish D. extensus, while dactylogyrus (Dactylogyrus cf. vastator) from fish were morphologically both. D. to the opposite market. Gyrodactylids (Gyrodactylus cf.katherineri) found in angelfish and discus fish were morphologically similar to G. katherineri found in goldfish.

Monogeenne trematodiit (Dactylogyrus extensus, Dactylogyrus cf. extensus Dactylogyrus vastator, Dactylogyrus cf. vastator, Dactylogyrus spp., Gyrodactylus turnbulli, G. katherineri, Gyrodactylus. They also had the highest infection rate of fish, suggesting that they were the most common parasites of ornamental fish species produced for export from Sri Lanka. The high prevalence of these infections can be explained, first, by the fact that monogenic trematodes have a high reproductive rate and effective transmission increases under poor management conditions (Soulsby 1982, Cone 1995). Second, viable Gyrodactylus and oviparous Dactylogyrus have very short direct life cycles that do not require medium strains, allowing them to reproduce rapidly to dangerous levels under the management conditions prevailing in ornamental fisheries (Citino 1996).

In addition to monogenean trematodes, protozoan parasites were frequently found in this study, especially Trichodina nigra and Trichodina spp. These obligate ectoparasites can survive without fish forhours, possibly days, and can be temporarily supported by several host species other than fish (Lom 1995). This may explain their frequent occurrence and relatively higher prevalence.

Most of the farmers in this study were aware of the risk of mortality due to infection with Tetrahymena. In Sri Lanka, when fish are injured in tanks or ponds, it is common practice

to kill fish in tanks or ponds. This may explain the low prevalence (4.1%) of T.corliss and T. pyriformis infection in this study, which is much lower than expected.

This study identified Lernaea cyprinacea, Ergasiluscey lonensis, and Argulus foliaceus, and all 3 species were recorded from freshwater fish (Fernando and Hanek1973, Fernando 1999). The prevalence of these parasites was also low (2.2, 1.3 and 1.3%). The life cycle of arthropod parasites can be extended to 3 months depending on temperature (Soulsby 1982). The most common export fish species are farmed for only 2-3 months before being introduced to the market. Thus, the possibility of such fish being exposed to parasitic infection is reduced if pond sand tanks are properly drained and cleaned after each production cycle. Since arthropods, especially A. foliaceus and L. cyprinacea, are visible with the naked eye, farmers can also implement early control measures, which may also be the reason for the observed low incidence. Centrocestus spp. needs an intermediate snail host; therefore, infections can be controlled with good treatment (Citino 1996). This may explain the Centrocestus spp. the very low prevalence of the metacercarial stage and the absence of other metacercarial stages during the study.

The lowest rate of infection of fish was in his paw pajo. This was expected because nematodes are smaller in fish than in terrestrial vertebrates (Anderson 1996).23 of the 26 farms had one or other parasites. Parasitism in fish occurs as a result of interactions between the parasite, the fish and the environment (Wildgoose 1998). Fish in intensive culture are constantly affected by environmental fluctuations and management practices such as handling, slaughter, transportation, drug treatment, malnutrition, fluctuating temperatures and poor water quality (Subasinghe 1997, Wildgoose 1998). A small change in the treatment, which is a common treatment by any standard, puts significant stress on the homeostatic mechanisms of the fish, making them susceptible to multiple parasitic infections (Ling et al. 1996, Scholz 1999). However, the proportion of parasitic fish detected in this study varied among the 4,444 farms. This could be a true difference, or the could be explained by a difference in the number of fish, as fish (16-195) were sampled from each farm, resulting indifferent detection opportunities.

Fifty of 590 guppies were infected with Tetrahy mena, compared to 13/930 for all other fish species, for a total of, a statistically significant result of (p and lt; 0.01) and indicating that this parasite is more common among guppies. . . This finding can also be explained by the lack of disease resistance in Guppycultivars due to the development of new cultivars for better appearance and color but few other factors. Breeding of new species is often practiced with guppy, which forms the largest part of Sri Lanka's export market (4,444). Besides the Ergasilus ceylonensis strain, other arthropods were observed mainly in carp and goldfish. Unlike other commonly caught fish species, these fish require up to a 12-month growth period before being released into the market, resulting in a greater likelihood of infection with odontopods, which have a relatively long life cycle.

n conclusion, of the 12 genera (21 species) recovered, Gyrodactylus, Dactylogyrus and Trichodina were the most common, and the presence of parasites in ornamental fish export farms was widespread. Parasites such as Tetrahymena were more common in guppies, while Argulus and Lernaea were more common in carp, suggesting that they were common to such fish species.

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3

Diversity of Spiders in Agriculture fields of Patur Dist,Akola

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Abstact:
Spiders belongs to order Araneaeare air-breathing arthropods that have eight limbs, chelicerae with fangs generally able to inject venom, and spinnerets that extrude silk. They are the largest order of arachnids and rank seventh in total species diversity among all orders of organisms. Spiders are found worldwide on every continent except for Antarctica, and have become established in nearly every land habitat. As of November 2023, 51,673 spider species in 136 families have been recorded by taxonomists. Spiders are beneficial predators that have been reported to prey on a variety of insect pests in agro ecosystems including aphids, caterpillars and beetles, forming an important component of biological pest control(Clough etal.,2005;Menalled et al., 2007) present study was carried out in the agriculture field of patur in the month of may-june 2023.during the presnt study survey we have reported 81 species of spiders belonging to 18 families and 63 genera. Spiders of families Araneidae,Clubionidae, Eresidae, Gnaphosidae, Lycosidae, Miturgidae, Oxyopidae, Philodromidae, Salticidae, Scytodidae, Tetragnathidae, Theridiidae, Thomisidae,Uloboridae,Hersilidae,Sparassidae,Oecobidae,Corinnidae were recorded during the investigation.

Key Words: Spider, Agriculture

Introduction:

Spiders are an important but generally poorly studied group of arthropods that play a significant role in the regulation of insect pests and other invertebrate populations in most ecosystems. Spiders play an important role in insect pest control without any harm to agro-ecosystem. Recently in agricultural fields reduced pesticide use and ecological sustainability have lead to increased interest in spiders as potential biological pest control agents. Spiders act as natural biological control agent in agro-ecosystem. Considerably insect populations increase when release from predations by spiders. Regularly use of pesticides in agricultural fields which decreases the spider populations.

Material and Method:

Spider survey was carried out in Agricultural Fields of Patur District Akola. Spiders were collected from in the month of May-June & Nov-Dec 2023. Spiderspecimens were collected from different areas of Agricultural Fields of Patur region. For collection of spiders visual searching, Pit fall trapping, Insect nets, beating steak with umbrellas,litter sampling were used. The Spiders Specimens were put in 75% alcohol, labeled and identified according to Kaston spider book and Tikader 1962. Before preservation the photographs were taken in different views with the help of Canon 60 D-Macro lense, to get the clear eye position, leg pattern and shades and size of cephalothorax, abdomen, spines and hairs pattern of the body stereozoom binocular microscope with photographic attachment (Zeiss Make) used.Spiders were observed under the microscope in the laboratorywith the help of identification keys. All the specimens were initially separated from other to the family level using the taxonomic keys for Indian spiders given by Tikader.

Result and Discussion

During the present study we have reported 135 species of Spiders belonging to 18 Families and 63genera. Spiders of Families Araneidae, Clubionidae, Eresidae, Gnaphosidae, Hersilidae, Lycosidae, Miturgidae, Oxyopidae, Philodromidae, Salticidae, Scytodidae, Sparassidae, Tetragnathidae, Theridiidae, Thomisidae, Uloboridae, Hersilidae, Sparassidae, Oecobiade, Corinnidaewere recorded duringpresent investigation. We have seen that the abundance of nine family species were more. The Orb waver spiders of Family Araneidae and Jumping spiders of Family Salticidae are widely distributed. The Orb waver spiders of Family Araneidae form web and the insect pest entangled in webspiders feeds on them. The members of Salticidae family i.e. jumping Spiders they directly capture the insect pest and feeds on it. Araneidae>Salticidae>Lycosidae>Oxyopidae>Thomisidae>Gnaphosidae>Theridiidae>Corinn idae> Uloboridae

TABLE: Showing number Spiders observed in the Agricultural region.

Sr. No.	Family	Genera	Species
1.	Araneidae	10	32
2	Clubionidae	02	03
3	Eresidae	01	02
4	Gnaphosidae	04	08
5	Hersilidae	02	02
6	Lycosidae	10	20
7	Miturgidae	01	02
8	Oxyopidae	04	13
9	Philodromidae	04	02
10	Salticidae	12	26
11	Scytodidae	02	01
12	Sparassidae	02	02
13	Tetragnathidae	01	03
14	Theridiidae	01	02
15	Thomisidae	05	11
16	Uloboridae	01	02
17	Oecobidae	01	02
18	Corinnidae	02	02

Conclusion:

A total18 Spider Families with 63 genera & 135 species. With the present results and discussion, it has been observed that Spiders are very much important creature and act as good Pest controller. If the farmers avoid the regular use of pesticides in agricultural fields the spider may be use as a natural pest killer in agroecosystem which leads to high yield. Spiders are beneficial bio-control agent of insect pest in agro-ecosystem.

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4

Diversity of Earthworm in Amravati region'

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Abstract:
Biodiversity is responsible for maintaining the ecosystem service, in the soil earthworm represent largest component of biomass of soil ecosystem. It is also important to quantify the spatial distribution of earthworm of different agro ecosystem in order to understand the effect of abiotic soil process and to link earthworm abundance to the spatial distribution of macrospore in the soil. The information collected on the type of earthworm species and their abundance of different habitat may also provide useful information on the efficiency and strength of that ecosystem. The review aims to assist people involved all aspects of management of agro ecosystem and importance and increases nutrient fertility of soil to obtain a broad knowledge of ecosystem service.

Keyword:
Earthworm diversity, soil variable species, Abundance

INTRODUCTION:

Animal evolution began in ocean over 600 million years ago with formation of numerous biomolecules. Since then animals have evolved into a highly diverse kingdom. Although over one million extant species of animals have been identified, Scientist is continually discovering more species as they explore ecosystem around the world.

The animal classification system characterizes animals based on their anatomy, morphology, evolutionary history, features of embryological development, and genetic makeup. This classification scheme is constantly developing as new information about species arises understanding and classifying the great variety of living species help us better understand how to conserve the diversity of life on earth.

Biodiversity generally refers to the variety and variability of life on earth. According to the United Nations Environment Programme (UNEP), biodiversity typically measure variation at the genetic, the species and the ecosystem level. Terrestrial biodiversity tends to be greater near the equator (Gaston et.al; 2000). Which seems to be greater the result of the warm climate and high primary productivity (Field et.al; 2009) Biodiversity is not distributed evenly on earth, and is richest in the tropics. These tropical forest ecosystems cover less than 10 percent of earth's surface, and contain 90% of the world's species (Young 2003).

"Biodiversity" is most commonly used to replace the more clearly defined and long established terms, species diversity and species richness. Davis (2011) defines biodiversity as the "totality of genes, species and ecosystems of a region". An explicit definition consistent with this interpretation was first given in paper by Bruce A. Wilcox (1982) commissioned by the International Union for the conservation of Nature and Natural Resources (IUCN) for the 1982 world National Parks conference. Wilcox's 1982 definition was "Biological diversity is the variety of life forms at all levels of biological system." In 1992 United National earth Summit defined "biological diversity" as the variability among living organisms from all sources, including inter alia, terrestrial, marine and other aquatic ecosystem.

Factors affecting Biodiversity:

During the last century, decreases in biodiversity have been increasingly observed. In 2007, German Federal Environment Minister Sigmar Gabriel cited estimated that up to 30% of all species will be extinct by 2050. The main cause of the loss of biodiversity can be attributed

to the world's ecosystem, in fact human beings have deeply altered the environment, and modified the territory, exploiting the species directly, for example by fishing and hunting, changing the biogeochemical cycles and transferring species from one area to another of the planet. Factors contributing to habitat loss are: Overconsumption, Overpopulation, Land use change, deforestation (Hogan 2010), pollution (air pollution, water pollution, soil contamination) and global warming or climate change.

Invertebrate Fauna: Invertebrates are the most successful and prolific animals on the planet. They have been around for over 400 million years and dominate the animal kingdom in terms of numbers of species and numbers of individuals. Invertebrates have also adapted to occupy practically every ecological niche. The number of invertebrate species is staggering and new species are being discovered all the time. To date scientists have only documented 1.7 million invertebrate species but they estimate numbers could range from 5 - 30 million. At this rate it will take scientists over a thousand years to identify all invertebrate species. Unfortunately, species numbers are declining faster than we can record their existence.

Annelids: The annelids (Annelida, from Latin anellus, "little ring") (Mclntosh1878) also known as the ringed worms or segmented worms, are a large phylum, with over 22,000 extant species including rag worms, earthworms, and leeches. There are over 22,000 living annelid species (Rouse 2002).

Diversity of Earthworm

Earthworms are important soil invertebrates belonging to the Phylum Annelida and Class Oligochaeta. Since long earthworms have been known as "Farmer's friend", "Nature's best fertilizers" and "Intestine of earth. Earthworms are one of the most important fauna of agro-ecosystems and they dominate the biomass of invertebrates in many soils of temperate and tropical regions of the world. More than hundred years ago, Darwin (1881) realized the value of earthworms as a major contributor to the formation of stable structural aggregates in soil. Many farmers, organic gardener and researchers have recognized earthworms as important organisms contributing to healthy soil. They are useful in land reclamation, soil improvement and organic waste management.

Earthworms are very diverse and very versatile and found in nearly all terrestrial ecosystems. During last three decades various efforts have been made to explore the taxonomy, biology, population dynamics, behavior, ecology, physiology, ecotoxicology and biotechnology of earthworm. The composition of different species of earthworms in different soils has been studies by number of workers (Satchell, 1983, Singh, 1997). Over 3500 earthworm species have been described worldwide and it is estimated that further surveys will reveal this number to be much larger(Reynolds , et al.;1994)Earthworms are among the most important components of soil biota in terms of soil formation, maintenance of soil structure and fertility (Bhadauria& Saxena;2010). Although not numerically dominant, their size makes them one of the major contributors to invertebrate biomass in soils (Edwards, 2004).Approximately 4,400 different species of earthworms have been identified worldwide (Sinha, 2009).The composition of different species of earthworms in different soils has been studied by number of workers (Satchell, 1983, Singh, 1997).Earthworms in the soil act as aerators grinders, crushers, chemical degraders and biological stimulators (Edwards &Bohlan, 1996). Earthworms belong to the class Oligochaeta. However there is much controversy with the classification of these organisms. Many scientists have developed their own classification schemes and theses have been further revised and developed over the years. Some have placed them in to the class Clitellata making Oligochaeta the subclass.

Classification

Kingdom – Animalia
Phylum - Annelida
Class - Clitellata

Subclass – Oligochaeta

Kingdom: **Animalia**

Earthworm belongs to the Animalia kingdom.They are multicellular organisms that are also eukaryotic; this means that their cells have nuclei.

Phylum: Annelida

Annelids are segmented worms. Earthworms belong to this phylum because their bodies are sectioned, creating the ridged or ringed appearance that give the "ring worms" of this phylum their name.

Class: Clitellata

The name of the class of earthworms owes itself to their clitellum: the collar that serves as a reproductive center during the adult phase of the earthworm's life this collar secretes clitella or cocoon during reproduction.

Subclass: Oliogochaeta

Earthworms have setae or bristles on the body, which helps them to attach to the surface during movement. They lack lateral appendages or parapodia, which is a characteristic feature of the subclass polychaeta

Order: Haplotaxida

The common earthworm is categorized under Haplotaxida, which is one of the two orders of Oligochaeta.

Families:

- Acanthodrilidae
- Ailoscolecidae
- Almidae
- Benhamiinae
- Criodrilidae
- Diplocardiinae
- Eudrilidae
- Exxidae
- Glossoscolecidae
- Hormogastridae
- Kynotidae
- Lumbricidae
- Lutodrilidae
- Megascolecidae
- Microchaetidae
- Moniligastridae
- Ocnerodrilidae
- Octochaetidae
- Sparganophilidae
- Tumakida

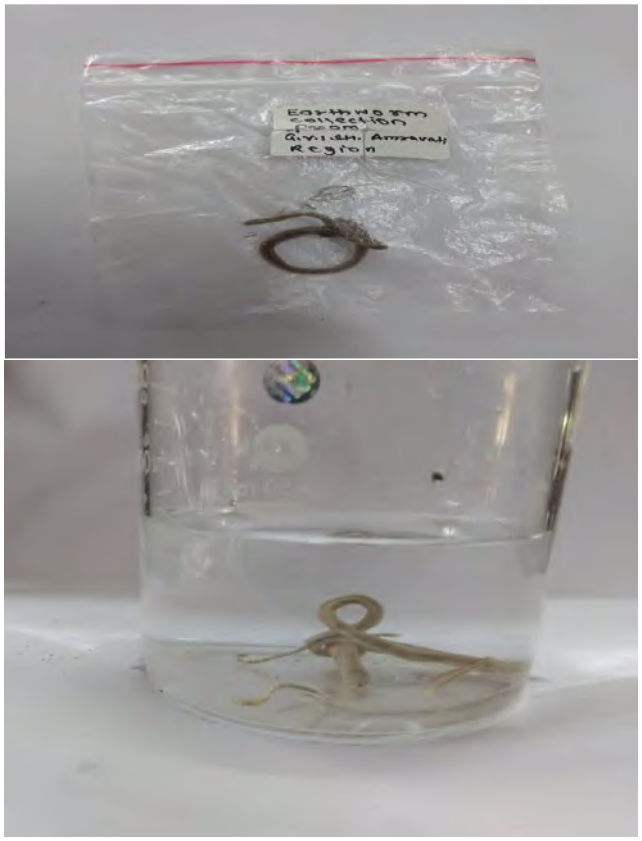
MATERIAL AND METHODS

Amravati city is a municipal council located in Amravati district of Maharashtra, India. The region Latitude is 20^{o} 55' 33" North and Longitude is 77^{o} 45' 53" East. The city is located in Satpuda range of hills in central India, hence rich in biodiversity. This region is well known for cotton, orange, sorghum, wheat, pulses, oil seeds crops. It has a pleasant climate. The region has tropical wet and dry climate with hot, dry summers and mild to cool winters. The temperature ranges from as low as 5 ^{o}C in winters and upto as high as 49 ^{o}C in summers. The average rainfall in this region is around 700-800 mm. with average daytime humidity around 51%. The main rivers in this region are Purna and Pedhi.

The study was carried out during the monsoon season of in the year 2021 for Six months. An extensive survey was conducted in between June 2021 to November 2021 in the diverse habitat of Amravati and surrounding agriculture area where earthworm can be found.
The places selected in the region for study were G.V.I.S.H campus, Wadali Lake, Chatri Talao, Bamboo Garden and a few villages around the city. The location selected as possible earthworm habitats were those where were often visibly moist. The Handpicking method was used for the collection of earthworm. Samples were taken from compost soil, garden soil, lawn soil, and agricultural land, waste water ponds, under stones, uppermost soil strata, rotton wood and other kind of plant debris. The photographs were taken in natural habitat of earthworm. The soil was dug up to depth of 20 cm and the earthworms were extracted by sieving and carefully hand sorting. The sampled earthworms were thoroughly washed in fresh water and collected in zip lock poly bags, with labeling of name of places from where the sample was taken.

Preservation

After collecting the samples of worm, they were bought to the laboratory and immediately these worms were transferred to the suitable size vials or bottles filled with 70% ethyl alcohol or 5-10% formalin and label with locality name, date and collectors name was added to each vial. The collected earthworms then identified with their size, colour, shape, body texture and other morphological character which are also matched with photographs of the earthworms on net.



RESULT AND DISCUSSION

In this study total six species of earthworms were recorded this six species are *Eisenia fetida, Pheretima posthuma, Lumbricus rubellus, Lumbricus terrestris, Aporrectodea coliginosa.* Belonging to five genera and three families of Oligochaeta were identified from various

habitats of the study areas of Amravati. The present study revealed that family Lumbricidae are dominating earthworm fauna in this region.

Sr. No	Family	Species	Taxonomical Characters				
			Body length	**Colour**	**Body Shape**	**No. of segment**	**Shape of clittelum**
1	Lumbricidae	*E.Fetida*	7-10cm	Red darker and lighter band	Cylindrical	100-120	Annular
		L.Terrestris	20-25cm	Reddish	Cylindrical	105	Annular
		A Calignosa	6cm	Grey	Cylindrical	100-180	Annular
		L.Rubellus	2.5-10.5cm	Reddish Brown	Cylindrical	95-105	Annular
2	Eurilidae	*E.Eugeniae*	10-12cm	Brown Red or dark violet	Cylindrical	145-190	Annular
3	Megascolecidae	*P.Posthuma*	15-30cm	Dark Brown	Cylindrical	100-120	Annular

The following species were identified from the study area.

Identified species

1. Eisenia fetida

It belongs to the Order: Haplotaxida, Family: Lumbricidae, Genus: Eisenia and Species: *E. fetida*

Commonly called as red wiggler, red worm, they are Smaller in size at arount 7-10cm. long for an adult worm. They have alternating bands of darker and lighter red. They have a slightly flat bottom with rest of the body being round. Tail is sometimes a lighter in color often with yellow tip. This worm is commonly used for vermin composting of both domestic and industrial organic waste

2. Pheretima posthuma

It belongs to the Order – Haplotaxida, Family – Megascolecidae, Genus – Pheretima, Species -*P.posthuma*

An adult worm measures about 15-30 cm in length, Dark brown in colour due to presence of pigments. The body is made of 100 to 120 segments, of which the first segment is divided into an anterior **prostomium** and posterior ring-like **peristomium.** Segments 14-16 form a girdle-like thick band of glandular tissue called **clitellum** that secretes mucus. The Earthworms live in a moist soil rich in dead organic matter or humus. They are abundantly found in old pastures, lawns and garden.

3. Eudriluseugeniae

It belongs to the Order – Haplotaxida, Family –Eurilidae, Genus – Eudrilus, Species -*E. eugeniae*

The common name of Eudriluseugeniae is night crawler. Eudriluseugeniae species shows faster growth rate and is the second most widely used earthworm for vermicomposting. The live worms are brown and red to dark violet on surface .The length of worm ranges from 10 to

12cm. and the diameter is 5 to 8mm. The total number of body segments range from 145 to 190.

4. Lumbricus terrestris

It belongs to the Order – Haplotaxida, Family – Lumbricidae, Genus – Lumbricus, Species - L.terrestris

It is commonly called as night crawler. The night crawler may be up to 20-25cm.long. The body is divided into 150 ring-like segments. The mouth is at the tapering front end, which is usually slightly darker than the rest of the body; the tail end tends to be more flattened than the head and lighter in color. It is commonly found in lawns, parks and gardens, but less common in woodlands and arable soils.

4. Aporrectodea calignosa

It belongs to the Order – Haplotaxida, Family – Lumbricidae, Genus – Aporrectode, Species - A.calignosa

It is also called Grey worm. It is recognizable by the three distinct shades of colour at its front end. Length is 6 cm. when not moving.100-180 segment are present on body surface. The worm mostly lies in non-permanent horizontal burrows in topsoil, and is rarely found in leaf litter. Like most worms, its diet consists only of soil.

6. Lumbricus rubellus

It belongs to Order – Haplotaxida, Family – Lumbricidae, Genus – *Lumbricus*, Species – *L. rubellus*

It is usually reddish brown or reddish violet, iridescent dorsally, and pale yellow ventrally. *Lumbricusrubellus*, or the "**red earthworm**", ranges from 2.5cm. to 10.5cm. in length and has smooth, reddish, semi-transparent, flexible skin segmented into circular sections. Each segment contains four pairs of setae, or bristles, and the total number of segments per matured organism ranges from 95–105. *Lumbricusrubellus* naturally lives in soils high in organic matter, preferably dung and feces.

DISCUSSION

Based on the survey which was carried out in the present study from June 2021 to November 2021 from different sites of Amravati city, total 6 species belonging to 5 genera and 3 family were identified. The most dominating family is *Lumbricidae*. Similar study was carried out in Hariyana, India where total nine species of earthworm belonging to six genera and three families were identified from various habitats of the surveyed areas of Transgangetic plains of Haryana. (Sharma and Bhardwaj; 2014) and 8 species of earthworms belonging to 5 genera, three families were found in Kashmir Valley, India (Ishtiyaq Ahmed Najar &Anisa B. Khan; 2011). Another study was carried out in Uttar Pradesh state of India where four genera with nine species of earthworms from three Oligochaeta families were identified (Om prakash 2009). In Dakshina Kannada District, South West Coast, Karnataka Earthworm survey was conducted in 5 different taluks of Dakshina Kannada District revealed occurrence of 5 genera and 11 species belonging to family Octochaetidae (Mussaiah Siddaraju,Kanale S. Sreepada, Krishna M P 2013). In total 22 species of earthworms belonging to 6 families were found from different pedo-ecosystems of Varanasi, Mirzapur, Allahabad, Ghazipur and Ballia districts of eastern Uttar Pradesh (S.N. Rai 2017). A total mean number of 2458.59 earthworm specimens were collected from various habitats of district Narowal Pakistan during the study period, representing five families, 12 genera, 20 species of earthworms (Abdul Gafoor et.al; 2008).

CONCLUSION

While studying the diversity of Earthworm from different region of Amravati city, a total 6 species belonging to 5 genera and 3 families were recorded in the present study. The most dominating family is *Lumbricidae* and the remaining are *Eurilidae* and *Megascolecidae.*

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5

Check List of Beetles (Order-Coleoptera) around Region of Khamgaon Dist. Buldana 444303 (India)

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Abstract:

Present study reported that 36 species were from the area of Khamgaon region Dist. Buldana. two sub –order, Adephaga and polyphaga and Eight Families.form Order-Coleoptera Sub-order Adephaga includes family Carabidae which represent 10 species. Sub-order Polyphaga includes seven (7) families namely Scarabaeidae, Coccinellidae, Lampyridae, Chrysomelidae, Meloidae, Elateridae and Cerambycidae. Sub-order Polyphaga represents 26 species out of which family Scarabaeidae contains 16 species, Coccinellidae includes 3 species, Lampyridae contains 2 species, Chrysomelidae 2 species, Meloidae, Elateridae and Cerambycidae represents only one species each. Most diverse family is the Scarabaeidae containing 16 species list diverse families are Meloidae, Elateridae and Cerambycidae including only one species each. Out of two sub-order studied, the dominant sub-order is Polyphaga (26 species) as compared to the sub –order Adephaga (10 species). Study was conducted in the month of July to October-2022 Collection was done at morning and night timing. Study will provide useful information about diversity of beetles in the said area as well as provides baseline data for upcoming researchers and gives wide scope for further study in entomology and biodiversity.

Keyword: Beetles, , Coleoptera, Biodiversity, khamgaon

Introduction

Biological diversity is the variability among the living organisms from all sources including aquatic and terrestrial ecosystems (Harper and Hawksworth, 1994). This includes variety within species, between species, and of ecosystems. Roughly 30 million species are found in the world, only 1.4 million species have been briefly described out of which, about 750,000 species are insects, which shows the percentage of insects in the world. They are either harmful or beneficial, and play an important role in the ecology of majority of ecosystem. Khamgaon area located in the Buldana Districts. Coleoptera (Linnaeus, 1758) is an extremely diverse order of class insecta. They are cosmopolitan and are modified for every possible habitat on our planet, except marine and Polar Regions (Pawara, *et al*., 2014). The beetle shows diversity; they show a great deal of ecological significance. Order coleoptera are very large and dominant order of animal kingdom. Several of them are specialized feeder of animal and plant waste (Banerjee, 2014), about 40% (4, 00,000) of all described insect species are beetles (Hammond, 1992) and approximately 15,088 species were recorded India (Kazmi, 2004) while some are not. Many of species are harmful by eating on some plant parts like flowers, fruits and seeds, which eventually damage our economy. Some beetles are useful, by controlling the populations of serious pests of agricultural plants, examples ladybird feed on aphid colonies, thrips, scale insects and mealy bugs that damage crops (Pawara, *et al*, 2014). In different part of Maharashtra collection of beetles was as done by (Dabhade *et al.* 2012) studied 25 beetles species belonging to the 8 super families and 11 families from Mangrulpir Tahsil, Dist. Washim, Maharashtra. Researcher (Thakare and Zade 2012) noticed 10 species belonging to 6 different families viz. Gyrinidae, Tenebrionidae, Carabidae, Scarabaeidae, Meloidae and

Buprestidae of beetles from various habitats at Melghat Tiger Reserve, Maharashtra were collected (Thakare *et al*.2012)

Material and Methods:

I. Study area:

Study area in the vicinity of around the Khamgaon taluka in the Buldana Distrcts. The entire survey was thoroughly so as to select the suitable sampling sites. Sampling and Collection was done during early rainy season in the month of July to October 2022 Survey was carried out by Collection, visual sighting and photo documentation. In some cases when identifications became difficult by photography or by visual sighting then collection was done by manually by pitfall trap & hand-picking, light trap method. Collection was done morning and night time. Ground beetle is collected with the help of pith fall trap.

II. **Collection and Identification of Beetles.**

Pitfall trap was used for the collecting ground beetles. Small Plastic tuffs filled with combination of 70% Ethyl alcohol and Glycerin was hidden up to the rim in the ground so that passing insects may fall. Light trap was used to collection of nocturnal beetles by using sources of white light Hand Picking method was used for some Beetle are elusive or have concealed habits other can be hand picked off their feeding or resting place. Many Beetles can be picked off flower and foliage. Dung beetle family Sacarabaeidae were collected from the dung with the help of forceps. The collected beetles were processed and stored in the Zoology lab for identification. Identification of unidentified species was done by using the standard keys, published articles and reference books. (Bousquet-1991).

Results and Discussions:

Present study reported that the 36 species were found in the region of Khamgaon two sub –order, Adephaga and polyphaga and Eight Families. Sub-order Adephaga includes family Carabidae which represent 10 species. Sub-order Polyphaga includes seven (7) families namely Scarabaeidae, Coccinellidae, Lampyridae, Chrysomelidae, Meloidae, Elateridae and Cerambycidae. Sub-order Polyphaga represents 26 species out of which family Scarabaeidae contains 16 species, Coccinellidae includes 3 species, Lampyridae contains 2 species, Chrysomelidae 2 species, Meloidae, Elateridae and Cerambycidae represents only one species each. Most diverse family is the Scarabaeidae containing 16 species; list diverse families are Meloidae, Elateridae and Cerambycidae including only one species each. Out of two sub-order studied, the dominant sub-order is Polyphaga (26 species) as compared to the sub –order Adephaga (10 species). List of all species reported are listed in the following table with the different families. Family Coccinellidea, Cerambycidae, Chrysomelidaen are the serious pest on the agriculture, crop its damage the small crop and plant in agriculture ecosystem (Ghate H.V 2012., Jadhav. S.S(2012.). Beetles in Family Carbide commonly called as the ground beetles are important for the biological control agent for Argo-ecosystem.

Table 1: list of Families with species from two sub-orders Adephaga and Polyphaga.

S.N.	Sub-order	Family	Species
1.	Adephaga	1. Carabidae	***1.*** *Pterostichus melanarius*
2.			***2.*** *Broscous cephalotes(Linnaeus)*
3.			***3.*** *Scarites vicinus(Fabricius)*
4.			***4.*** *Scarites subterraneus(Fabricius)*
5.			***5.*** *Anthia sexguttata(Fabricius)*
6.			***6.*** *Thermophilum homoplatum(Fabricius)*
7.			***7.*** *Eudema angulatus(Fabricius)*
8.			***8.*** *Bembidion quadrimaculatus(Latreille)*
9.			***9.*** *Scaritinaes subterraneus(Fabricius)*
10.			***10.*** *Chlaenius sericeus(Fabricius)*
11.	Polyphaga		***1.*** *Clinteria kluge(hope)*
12.			***2.*** *Catharsius molossus (linnaeus)*
13.			***3.*** *Oniticellus (onticellus) cinctus*

14.		2. **Scarabaeidae**	***4.*** *Onthophagus pactolus(fabricius)*
15.			***5.*** *Onthophagus gazella(fabricius)*
16.			***6.*** *Onthophagus spinifex(Fabricius)*
17.			***7.*** *Onthophagus aff.anthracinus(Harold)*
18.			***8.*** *Onthophagus striatulus(Fabricius)*
19.			***9.*** *Gymnopleurus gemmatus (harold)*
20.			***10.*** *Gymnopleurus miliaris(fabricius)*
21.			***11.*** *Heliocopris bucephalus(fabricius)*
22.			***12.*** *Garreta nitens(olivier)*
23.			***13.*** *Catharsius sgax(quenstedt)*
24.			***14.*** *Heliocopris bucephalus(Fabricius)*
25.			***15.*** *Callistethus marginatus(Fabricius)*
26.			***16.*** *Heteronycha sarator(fabricius).*
27.		3. **Coccinellidae**	***1.*** *Coccinella septempunctata(linnaeus)*
28.			***2.*** *Cheilomenes sexmaculata(Fabricius)*
29.			***3.*** *Adalia punctata.(Fabricious)*
30.		4. **Lampyridae**	***1.*** *Pteroptyx tener(Olivier)*
31.			***2.*** *Pyractomena punctiventri*
32.		5. **Chrysomelidae**	***1.*** *Cassida rubiginosa (Muller)*
33.			***2.*** *Cassida flaveola (Thunberg)*
34.		6. **Meloidae**	***1.*** *Mylabris phalerata (Pallas)*
35.		7. **Elateridae**	*1. Melanotus pertinax(Say-1839)*
36.		2. **Cerambycidae**	***1.*** *Apriona marcusiana*

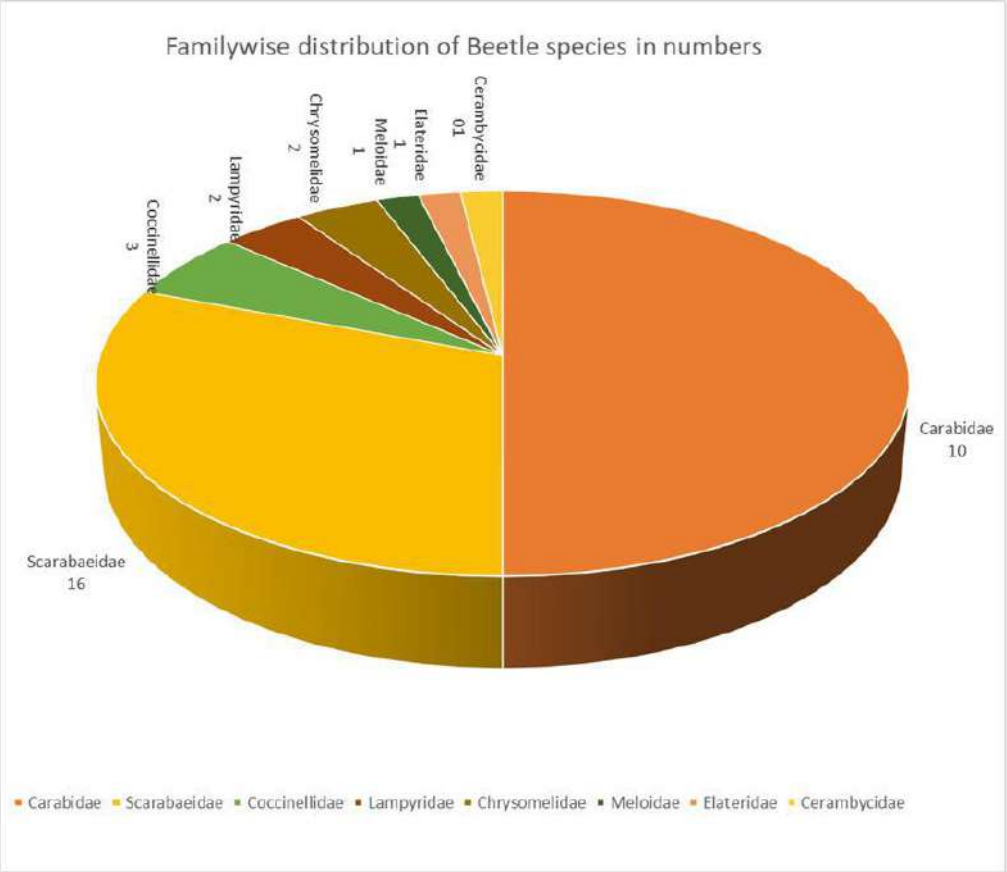

Figure 2: Pia Chart Showing the Beetles Families.

Conclusion:

Family Scarabaeidae are most dominant due to the noticeable and site due to their large size, bright color, often with elaborate ornamental, it called as dung beetle for their dung consumptions and relocation activity, for helping the series of ecological function such as nutrient dispersal, nutrient cycling and bioturbation. Elateridae also called as the click beetle, they are generalist feeding on a large variety of crop resulting in damage seed, roots stems, Elateridae are less reported less as compared to other family of Beetles. Lampyridae Meloidae. Above result its collection of four month. This diversity may be due to high diversity of flora in the present study area with favorable environmental conditions. Above collection and Check list provides useful information about Bio-diversity of beetles in the said area as well as provides baseline data for upcoming researchers and gives wide scope for further study in entomology and biodiversity.

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6

Influence of steroidal and non-steroidal contraceptive pills on ascorbic acid alterations in the reproductive organ of wistar female *albino rats.*

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Abstract

The effects of steroidal and non-steroidal contraceptive oral pill on ascorbic acid alterations in the reproductive organ of wistar female albino rats for 30 days. The steroidal combined oral contraceptive pill (norgestrel + ethinylestradiol) was diluted to 0.14mg/ ml (Low Dose) , 0.21 mg/ml (dose as per literature), and 0.43 mg/ ml (high dose). The non-steroidal oral contraceptive pill (Centchroman) was diluted to 0.29 mg/ ml (Low Dose) , 0.43 mg/ml (dose as per literature) and 0.87 mg/ml (high dose) . The optimum level of ascorbic acid (Luck *et al*., 1995) and proteins are necessary for maturation and fertilization of ova and all these are regulated by androgens. Therefore, the decreased level of these biochemical constituents in the ovary of Centchroman fed rats as well as steroid fed rats (normal and high doses) suggests an anti-androgenic effect.

Keywords: Steroidal and non-steroidal contraceptive oral pill, ascorbic acid, female reproductive organ, histology, female wistar albino rat.

Introduction

Oral contraceptives (OCs) are oral contraceptives that work by obstructing ovulation as their primary method of prevention. These are one form of birth control and come in two varieties: the progestogen-only pill and the combined oral contraceptive pill (COC), which contains both progesterone and oestrogen. These days, the market is filled with a variety of oral tablets, both steroidal and non-steroidal, but their outcomes differ significantly.

To get a better understanding of how OCs affect ascorbic acid status, scientists looked at how much ascorbic acid is present in the plasma leukocytes, platelets and whole blood entities. According to some reports, using OCs—especially those containing estrogen, which is assumed to speed up ascorbic acid metabolism—lowers the levels of ascorbic acid in platelets and leukocytes (Veninga, 1984). The change in blood levels has been suggested to be due to changes in tissue uptake patterns resulting in changes in vitamin distribution (Thorp, 1980). Women with luteal phase defects may see higher serum progesterone concentrations after taking ascorbic acid supplements (Henmiet.al., 2003). Moreover, lower concentrations of ascorbic acid were observed among women with recurrent spontaneous abortions linked to a luteal phase defect than among women with better reproductive outcomes (Verulet.al., 2003). Because of its critical function in hormone secretion, ascorbic acid has been closely linked to fertility. (Franceschi, 1992). It also improves fertility and aids in iron absorption for women who have luteal phase defects, gamete production, and gonad tissue remodelling (Franceschi, 1992). A deficiency of this vitamin results in a range of clinical conditions, including: scurvy wound damage vasomotor instability connective tissue disorders. The aim of this study was to evaluate the alterations in ascorbic acid in the reproductive organ of female albino rats fed with steroidal and non-steroidal contraceptive pills for 30 days.

Materials and Methods

Experimental Animal Models:-

The present study was carried out in wistar female albino rats weighing about 125g ± 2 g. The animals were procured from National Institute of Nutrition (NIN), Hyderabad. Animal experiments were conducted according to "INSA – Ethical guidelines for use of animals for scientific research after getting permission from ethical committee". The animals were kept in vivarium throughout the period of experiment. They were regularly fed on standard pellet diet provided by National Institute of Nutrition, Hyderabad and water *ad-libitum*. The remaining food and waste matter was removed from the cages on the next day and proper care was taken to avoid any infection. Only healthy rats were used for the present experiments.

Pills:

The experimental female albino rats were given selected non-steroidal contraceptive oral pills in calculated doses.Non-steroidal oral contraceptive pill with Brand name, Saheli. Each Tablet Contain Centchroman - 30 mg (Manufactured by: - Hindustan Latex Limited).

Doses:

Dilutions of pills were made by using double distilled water (DDW). The non-steroidal oral contraceptive pill (Centchroman) was diluted to 0.29 mg/ ml (Low Dose), 0.43 mg/ml (dose as per literature) and 0.87 mg/ml (high dose). The doses of both drugs were calculated as per body weight of rats considering the human consumption and available literature.

Experimental set up:-

Experiments were carried out by dividing female albino rats into three groups:

Group I:-Control female albino rats administered orally with 1ml DDW/ rat / day upto 30 days DDW being used as vehicle .

Group II: - Group of rats were administered orally with Centchroman.This group was divided into three sub groups.

Subgroup I:-Experimental female albino rats administered orally with1ml Centchroman / rat / day upto 30 days. 1 ml dose contains 0.29 mg Centchroman.

Subgroup II:-Experimental female albino rats administered orally with1mlCentchroman / rat / day upto 30 days. 1 ml dose contains 0.43 mg Centchroman.

Subgroup III:-Experimental female albino rats administered orally with 1 ml Centchroman / rat / day upto 30 days. 1 ml dose contains 0.87 mg Centchroman .

Histological studies:

The histological studies were carried out for adrenal gland of the control and experimental female albino rats. The tissues were washed with saline water to remove adhering particals and blood stains and then fixed in Bouin's fixative for 24 hrs. . Then the tissues were washed thoroughly with water, dehydrated with graded series of alcohols and embeded in paraffin wax and sections were cut at 4 to 5 microns. The sections were processed and stained with Haematoxylin - Eosin by standard methods as described by Weissman (1978).

Estimation of ascorbic acid:

Ascorbic acid in the tissue (ovary / fallopian tube / uterus) of the control and experimental female albino rats was estimated by the method of Roe andKuether's (1943) .

Observation and Result:

Significant decrease in ascorbic acid was obtained in ovary and uterus of steroidal and non-steroidal contraceptive pills fed rats after 30 days of treatment (Table-1). In fallopian tubes of steroidal pill fed rats ascorbic acid was significantly increased however,in excess dose of non-steroidal pill fed rats, the fallopian tubes exhibitted rise in ascorbic acid mg/gm tissue after 30 days. In normal and subnormal dose condition ascorbic acid as significantly reduced in fallopian tubes.

Table1:-Alterations in Ascorbic Acid (mg / gm of tissue) of female albino rats fed with steroidal and non - steroidal contraceptive pills for 30 days.

Sr. No.	Tissue	Control	Steroidal Pill			Non-steroidal Pill		
			0.14mg/ml / rat/day	0.21mg/ml / rat/day	0.43mg/ml / rat/day	0.29mg/ml / rat/day	0.43mg/ml / rat/day	0.87mg/ml / rat/day
1	Ovary	3.01 ± 0.06	5.35 ± 0.16 (+77.74)	4.31 ± 0.18 (+43.18)	1.94 ± 0.02 (-35.54)	3.64 ± 0.17 (+20.93)	1.54 ± 0.05 (-48.83)	2.67 ± 0.10 (-11.27)
2	Fallopian tube	2.94 ± 0.05	3.65 ± 0.11 (+24.14)	3.99 ± 0.22 (+35.71)	3.83 ± 0.03 (+30.27)	1.19 ± 0.06 (-59.52)	0.83 ± 0.02 (-71.76)	1.42 ± 0.16 (-51.70)
3	Uterus	3.73 ± 0.08	1.48 ± 0.26 (-60.32)	2.57 ± 0.09 (-31.09)	2.25 ± 0.11 (-39.67)	6.24 ± 0.21 (+67.29)	2.77 ± 0.09 (-25.73)	1.9 ± 0.11 (-49.06)

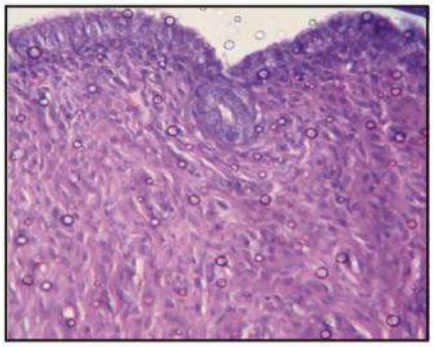

Fig. A T. S. of fallopian tube 3 of female albino rat fed with 0.29mg/day non-steroidal contraceptive pill for 30 days. Haematoxylin-eosin, X400

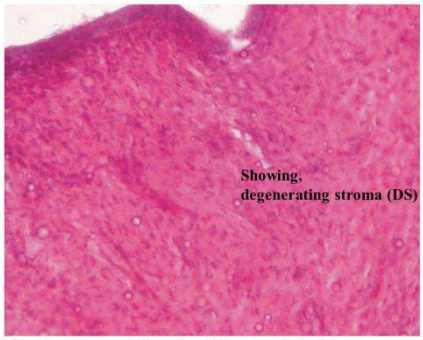

Fig. B T. S. of uterus of female albino rat fed with 0.29 mg/day non-steroidal contraceptive pill for 30 days. Haematoxylin-eosin, X400

Discussion and Conclusion

Ascorbic acid is an important biologically active reductant which plays a significant role in reproductive process. Ascorbic acid accurs in free and bound forms (ascorbogen) in reproductive tissue (Chinoy, 1972). The ascorbogen content of reproductive tissue, vary considerably. Ascorbic acid is bound to macromolecules by charge transfer complex formation (Chinoy and Sanjeevan1978). The storage, tissue distribution and also synthesis of ascorbic acid are under the control of sex hormones in rats (Chinoy*et al*., 1979). Actively growing and regenerating tissues also possess higher concentrations of vitamin-C. Ascorbic acid inhibits the activity of phosphodiesterase (Lewin, 1976) and thereby indirectly helps in increasing the level of c-AMP, which is the important "second messenger" in mechanism of hormone action. In the present investigation ascorbic acid is found to be elevated in fallopian tube of steroidal pill treated rats indicating healthy condition and normal function of fallopian tube, however in rats treated with Centchroman, ascorbic acid was drastically declined indicating disturbed metabolism in it (Fig. A). Thus, Centchroman also has a secondary impact on ascorbic acid. Further in steroid fed rats, ascorbic acid was found to be declined significantly (Fig. B) in uterus exhibiting adverse action.

The optimum level of ascorbic acid (Luck *et al*., 1995) and proteins are necessary for maturation and fertilization of ova and all these are regulated by androgens. Therefore, the decreased level of these biochemical constituents in the ovary of Centchroman fed rats as well as steroid fed rats (normal and high doses) suggests an anti-androgenic effect. No doubt, it has anti-estrogenic action, it prevents the ova to pass through fallopian tubes and anti-implantation activity however it also shows some adverse effect on the ovary. More research is needed to calculate the perfect dose which will not lead to any adverse effect on the ovary.

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7

Exploring the Green Synthesis and Characterization of Zinc Oxide Nanoparticles by Freshwater*Hydrilla*spp.

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Abstact

'Algae' are important members of aquatic photosynthesizing microorganisms in water bodies. Chlorophytes, charophytes and glaucocystophytes constitutes fresh water green algae. Macroalgae (seaweeds) and microalgae (phytoplanktons) are natural feed for freshwater fishes. Freshwater algae like spirulina are nutritionally important as they are a rich source of proteins. They contain high levels of B-complex and vitamin E, beta carotenes, manganese, iron, copper, zinc, and selenium, and essential fatty acids eg. Gamma linoleic acid. This huge range of biomolecules and metabolomes makes them a suitable moiety for the synthesis of various metal based nanoparticles like silver, copper oxide and zinc oxide nanoparticles. In this study, we report the synthesis of zinc oxide nanoparticles (ZnONPs) by *Hydrilla* spp. Synthesized ZnONPs were confirmed by a formation of white precipitate with absorbance at 302 nm. Nanoparticle tracking analysis confirmed AgNPs with an average size of 63 +/- 30.2 nm (concentration $8x10^9$ particles/ml) and zeta potential as -13 mV. FTIR showed the presence of transmittance peaks for various metabolites stabilizing ZnONPs. XRD and EDX confirmed face centered cubic crystal. FESEM elucidated irregular shaped ZnONPs. It can be concluded that green method for synthesis of ZnONPs by an ecofriendly approach was developed.

Introduction

Nanotechnology is continuously advancing towards the development of new and novel techniques for the synthesis of various nanoparticles. It is intended for ease of synthesis, lowering cost, imparting desired biocatalytic activity, shape and size, etc., [7]. Various plant materials and their byproducts are explored for the fabrication of nanoparticles using green method with desired physico-chemical properties. Despite of limiting challenges the diverse methodologies have been developed to benefit the requirement [2]. In the present study the zinc oxide nanoparticles (ZnONPs) were synthesized using the fresh water Hydrilla or waterthyme i.e. *Hydrillaverticilliata*extract. The crude extract of Hydrilla was resulted in the pale white colored precipitate of ZnONPs. Synthesized ZnONPs were further characterized using various techniques for the confirmation of physical and biochemical properties.

Materials and methods

Hydrilla plants were collected from the nearby fresh water river. Plants were further oven dried at 60^{o}C for 24 hrs followed by grinding and resulting powder (**Figure 1a.**) was utilized for the synthesis of ZnONPs. 1gm of powder was weighed and mixed in distilled water and boiled for 20 min to get the extract, which was filtered through multilayered muslin cloth to remove any particulate matter (**Figure 1b.**). ZnONPs were synthesized by simple mixing of 50mM zinc sulfate with diluted Hydrilla extract, resulting in pale white precipitate of NPs. ZNONPs were subjected to UV-visible spectrophotometric analysis to determine absorbance maximum. Further, size and zeta potential was determined by NTA (Nanoparticle Tracking and Analysis) and zetasizer. FTIR (Fourier Transform Infrared) spectrometry and XRD (X-ray diffractometry) depicted the presence of various functional groups on the surface and structure of crystal lattice respectively. FESEM was performed to confirm the structure of ZnONPs crystals.

Observation and Results

The extract was prepared from ground Hydrilla material and was used for the synthesis of ZnONPs. White precipitate formation confirmed the synthesis of ZnONPs (**Figure 1c.**).

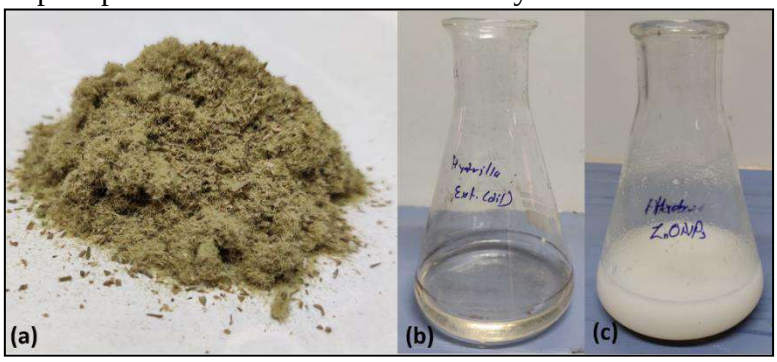

Figure 1. Synthesis of Hydrilla mediate ZnONPs, (a) powdered hydrilla material, (b) aqueous extract of Hydrilla and (c) white precipitate of ZnONPs

UV-visible spectrum of ZnONPs showed the absorbance peak at 302 nm (**Figure 2a.**). NTA analysis showed mean size as 63 nm $\pm$ 30 nm and particle concentration of 8.6 x 10^9 particles per ml (**Figure 2b.**). Zeta potential was observed as -13.0 mV (**Figure 2c.**). **Figure 2d**, represents the distribution of ZnONPs in 2D.

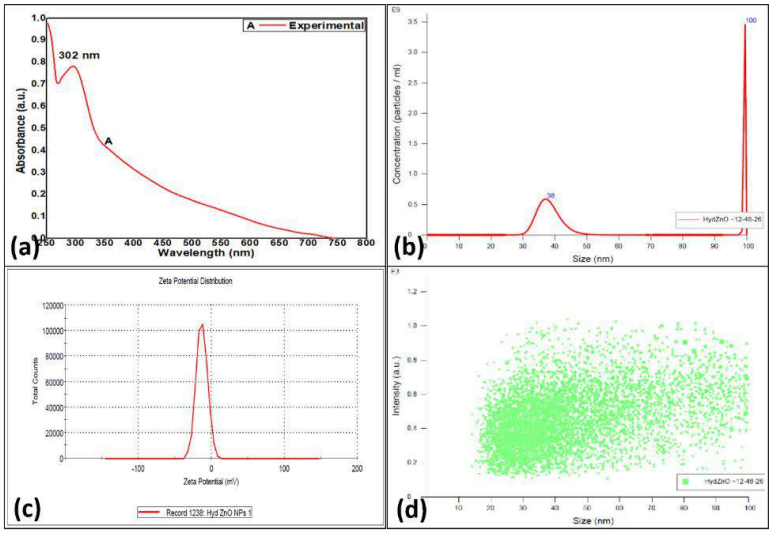

Figure 2. Characterization of Hydrilla mediated ZnONPs, (a) UV-visible spectrum, (b) NTA analysis (c) Zeta poterntial measurements and (d) particle distribution in 2D

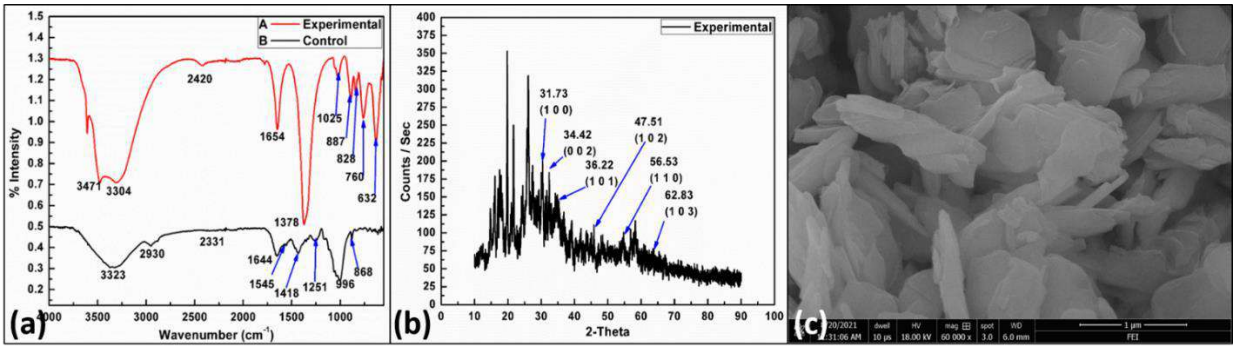

Figure 3. Characterization of Hydrilla mediated ZnONPs, (a) FTIR spectrum, (b) X-ray diffraction pattern and (c) FESEM image

Table 1. FTIR analysis of Hydrilla mediated ZnONPs

Sr. No.	Spectrum range	Observed Wavenumber (cm^{-1})	Functional group assigned	Compound present

	(cm^{-1})	**Hydrilla extract**	**ZnONPs**		
1	3570-3200	3323	3471, 3304	-OH	Carboxylic acid, alcohol, water, phenol
2	2935-2915	2930, 2331	2420	C-H streching	Alkanes
3	1650-1550	1644	1654	=N-H bend	Secondary amine
4	1560-1540	1545, 1418	1378	-N=O stretch	Aliphatic nitro compound
5	1350-1250	1251	-	-P=O stretch	Organic phosphate
6	1050-990	-	1025	C-O group	Alcohol, acid or esters
7	995-850	996, 868	887, 828	C-H plane bend	Vinyl compounds

FTIR spectrum (**Figure 3a.**) represented the presence of various peaks that are assigned to various functional groups from the biomolecules present in the extract. The peaks indicated the presence of primary and secondary amine groups, hydroxyl stretch, ethereal compounds and aliphatic hydrocarbons present in the capping layer of ZnONPs (**Table 1**). XRD spectrum elucidated the face cubic centered structure of ZnONPs crystal (**Figure 3b.**). FESEM images represented the formation of flakes of ZnONPs (**Figure 3c.**).

Discussion

Hydrillaverticillata (L.f.) is a widely distributed aquatic weed having prolific growth habits and a wide range of habitat. It can tolerate a wide range of physical and chemical conditions of water and can survive in low nutrient supply. *Hydrilla* is a potential accumulator of several metals and metalloids. The accumulation potential of *Hydrilla* plants is attributable to induced synthesis of thiol (-SH) containing ligands like glutathione and phytochelatins. Huge quantities of biomass can be generated in very short time. It is an excellent source of a secondary metabolites [8]. These properties makes *Hydrilla* a promising subject for the synthesis of various types of nanomaterial's. Fabrication of silver nanoparticles (AgNPs) using *H. verticillata* has been reported by Sable et al. [6]. ZnONPs synthesized from *Hydrilla* showed peculiar peak wavelength of absorbance as reported previously for biogenic ZnONPs. Spectral detection followed by visible color change from transparent to white, thus confirmed the formation of nanoparticles [3]. NTA analysis and zeta potential determination depicted formation of stable ZnONPs with the size below 100 nm and higher particle distribution below and near to 100 nm [3, 6]. Similarly, the FTIR spectrum showed various peaks of different intensities which were assigned to respective functional groups from the secondary metabolites from *Hydrilla*extract [1, 4]. FESEM images agglomerated particles and XRD spectrum confirmed the crystalline nature and FCC (Face centered cubic) crystal lattice of ZnONPs indexed with JCPDS No. 89-510 (Joint Committee on Powder Diffraction Standards 01-079-0207) [5].

Conclusion

It can be thus concluded that fresh water Hydrilla can be used as source of secondary metabolites which can be easily extracted and explored for the phytofabrication of the ZnONPs. The plant material can be easily grown even in limited nutrient supplies. Stable ZnONPs can be synthesized with various functional characteristics based upon the nature of their capping layer. These Hydrilla mediated ZnONPs can thus be applied in various fields as desired and with prior evaluation *in vitro* and *in vivo*.

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8

Preliminary Study of Dipteran Flies of Buldhana District in Maharashtra

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Abstract
The order Diptera, comprising of two-winged or true flies, is one of the most commonly recognized and ubiquitous insects all over the world. The flies are arguably the most important, if only because the kill millions of people a year by transmitting most devastating diseases, but this enormous impact is due to only a few dozen species and Dipteran damage to forest, crop and stored product is similarly due to mere dozens of different fly genera. Vastly more are beneficial, contributing to the pollination of plants, biological control of pest insects and disposal of the dung, carrion and other organic matter. The present study provides a preliminary survey of Dipteran flies. A comprehensive survey was made during 6th November 2021 to 6th September 2023 to study biodiversity of Dipteran flies in Buldhana District of Maharashtra, India. About 15 families of flies were found during the survey, which comprises 25 genera. A camera trap method was employed for the study purpose.
Keywords: Diptera, Buldhana district, true flies, beneficial flies.

Introduction:

Diptera is an order of insects commonly referred to as true flies. The name Diptera, coined by the Greek philosopher Aristotle more than 2,000 years ago. Diptera stands for two-winged insects (di = two; petra = wing) because the first pair of wing is primarily used for flying and the second pair is modified to form a small, club-shaped structure called halters which helps in flight and balance. Over 1,60,000 known species the number of Dipteran species in the order is currently expanding by about 1% per year (Marshall 2012). The Diptera is an important group that represents about 10% of the world's biodiversity, besides being rich in species, flies are extremely important to humans, from a negative standpoint e.g., as vectors of disease and from a positive standpoint e.g., pollinators of various plants, biological control agents, and important decomposers in many habitats (Brown 2005).

The first pioneering work on the study of Diptera in India was undertaken by Brunetti (1912, 1920, 1923) and White *et al*. (1940). Contribution from the Zoological Survey of India and other institutions have led to a better understanding of the diversity of this order across the country. Saha *et al*. (2012) and Sharma (2012) have created a checklist of Diptera of the state of Maharashtra as a part of State Fauna Series (Dhamorikar 2017). The diversity of Dipteran flies remain untouched in Buldhana district. So this Ph.D work was carried out to survey the diversity of Dipteran flies from this district.

Materials and Methods:

a) Study area:

The present study was carried out in Buldhana district which is located in the Amravati division of Maharashtra, India. It is surrounded by Madhya Pradesh in the North, Akola, Washim, and Amravati districts on the East, Jalna district on the South, and Jalgaon and Aurangabad districts on the West. The Latitudes are 19.51° to 21.17° N and the longitudes are 75.57° to 76.59° E. The average annual temperature is 25.5ºC | 77.9ºF in Buldhana while annual rainfall is 983mm | 38.7 inch.The district has a rich vegetation of forests

like Botha Forest, Ambabarva Forest, Ajanta forest etc. About 840.66 Sq KM of area is under forest which constituted 8.70% of the total area. Forest resources contribute significantly to the economy of the district. The district has its unique identification of Lonar Crater ecosystem. As of 2010, the district of Buldhana comprises thirteen Talukas viz. Buldhana, Chikhli, Deulgaon Raja, Malkapur, Motala, Nandura, Mehkar, Sindkhed Raja, Lonar, Khamgaon, Shegaon, Jalgaon Jamod and Sangrampur. (Buldhana district gazetteer)

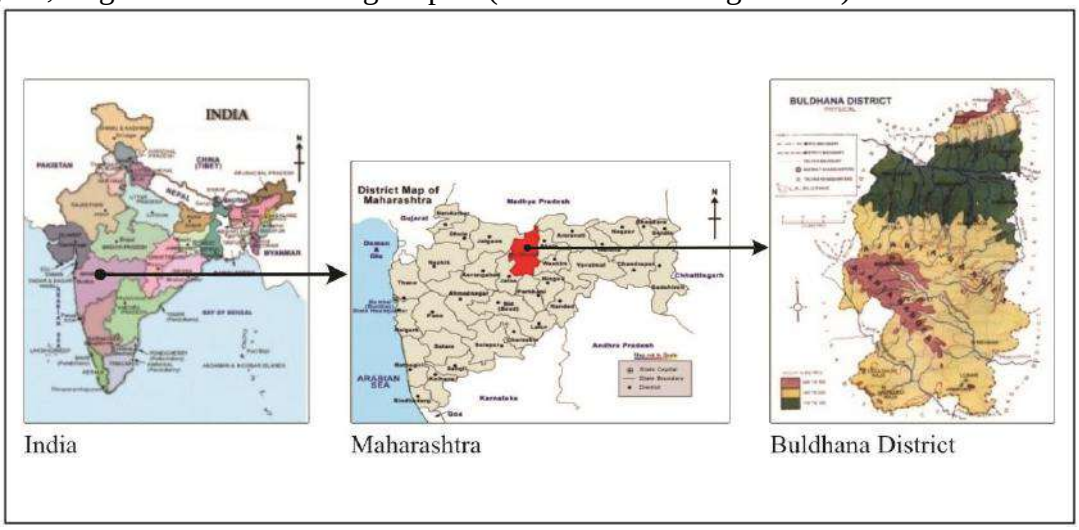


Fig 1: Geographical Location of the Research Area (Source: Buldana District Gazetteer)

b) Methods:

In the present study the flies were identified upto genus level (Image 1-25) using identification keys provided by Marshall (2012). Flies were photograph through various angles to obtaine morphological details such as wing venation, head, leg features etc. wherever possible, specimen were sampled under this study. During this survey all thirteen talukas visited every season, and the nature of survey was unstructured.

Observation and Result:

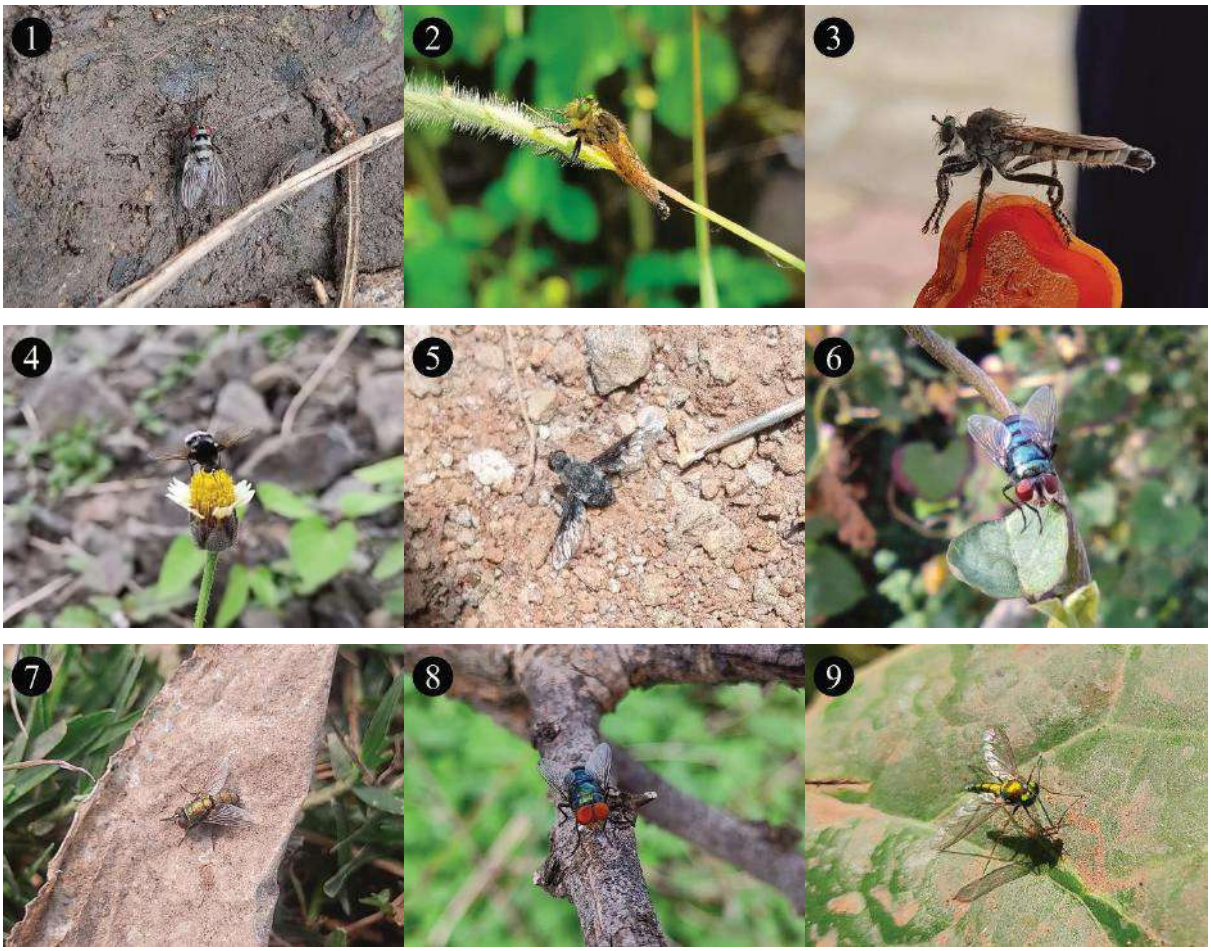
Since there are more than 10,000 genera of flies, it is a very critical job for person to recognize all of them. However, it is extremely useful to know what genus a fly belong to because generic identification can unlock a wealth of literature about biology, distribution, relationship and diversity. Due to lack of good recent taxanomic revision, flies are often unidentifiable to the species level and will remain so until the taxanomic work is done (Marshall 2012). So the currentstudy revealed the diversity of Dipteran flies from Buldhana district up to genus level, which comprises 15 families distributed in 25 genus as shown in table 1.

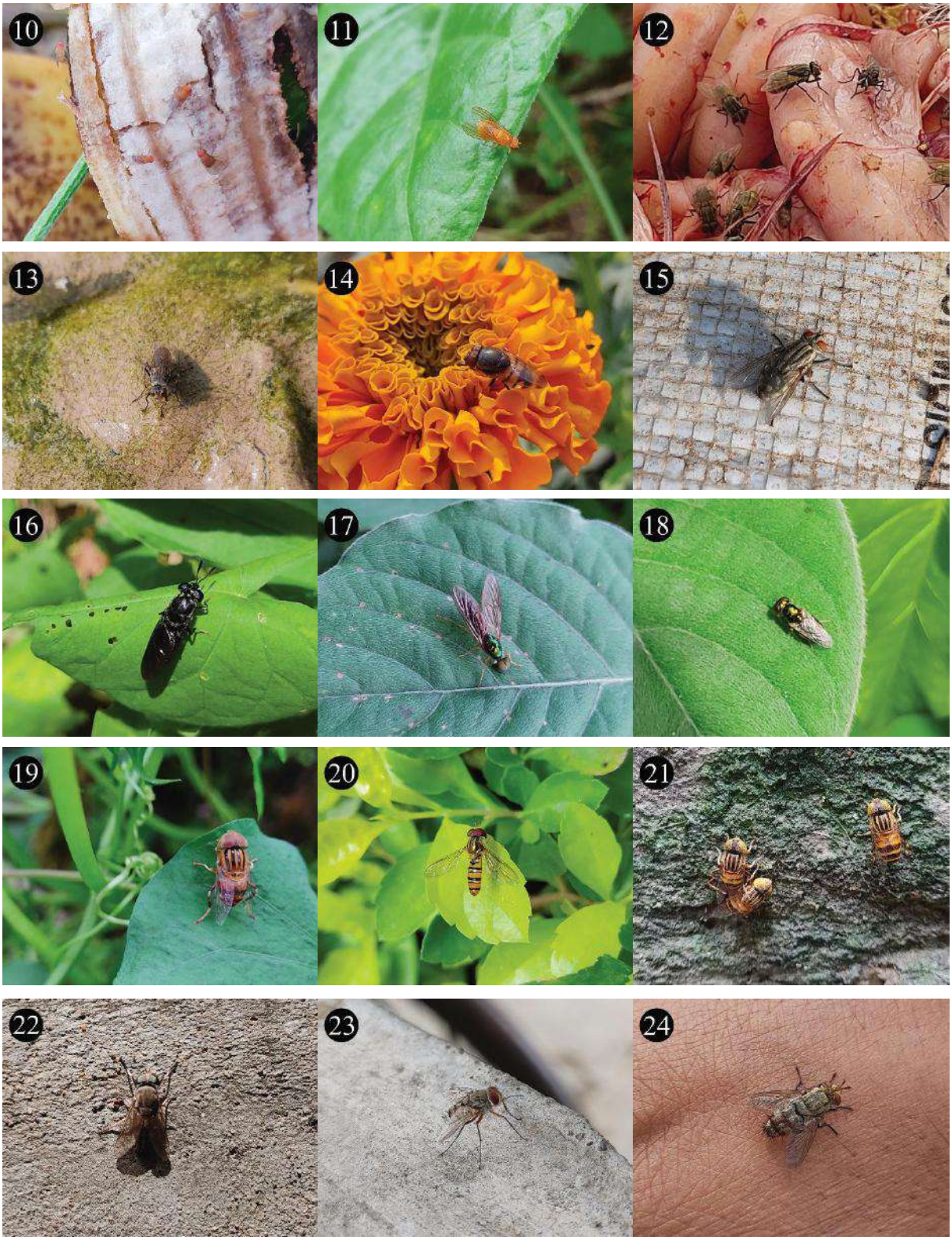
Family	Image No.	Genus	Common Name
Anthomyiidae	1	*Anthomyia oculifera*	Root maggot fly
Asilidae	2	*Unidentified*	Robber Fly
	3	*Unidentified*	
Bombyliidae	4	*Euchariomyia dives*	Bee Fly
	5	*Anthrax*	
Calliphoridae	6	*Calliphora*	Blow Fly
	7	*Lucilia*	
	8	*Chrysomya megacephala*	
Dolichopodidae	9	*Chrysosoma*	Long legged fly
Drosophilidae	10	*Drosophila*	Fruit Fly
Lauxaniidae	11	*Sapromyza*	Lauxanid Fly
Muscidae	12	*Musca domestica*	

	13	*Lispe*	House Fly
Rhiniidae	14	*Unidentified*	Rhiniid Fly
Sarcophagidae	15	*Wohlfahetia*	Flesh Fly
Stratiomyidae	16	*Hermetia*	Solider Fly
	17	*Sargus*	
	18	*Microchrysa*	
Syrphidae	19	*Eristalinus megacephalus*	Hover Fly
	20	*Episyrphus*	
	21	*Eristalinus taeniops*	
Tabanidae	22	*Tabanus*	Horse Fly
Tachinidae	23	*Prosena*	Tachinid Fly
	24	*Exorista*	
Ulidiidae	25	*Physiphora*	Picture-winged Fly

Table 1: Diversity of Dipteran flies observed from Buldhana District during November 2021 to September 2023

Plate 1: Photographs of different Dipteran flies obtained during November 2021 to September 2023.

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Discussion:

The comprehensive survey was made during 6th Nov 2021 to 6th Sep 2023 to study biodiversity of Dipteran flies from buldhana district of Maharashtra. India it is represented by 87 families containing well over 6,000 species (Alfred *et al*. 1998).The Zoological Survey of India has identified 37 families in the state of Maharashtra (Saha *et al*. 2012; Sharma 2012a,b). from Mumbai Metropolitan Region 18 families were recorded taking the number of Diptera families of Maharashtra to 55 (Damorikar 2017). This study add genus such as *Anthomyia oculifera* (family Anthomyiidae), *Anthrax* (family Bombyliidae), *Microchrysa* (family Stratiomyidae), *Eristalinus taeniops* (Family Syrphidae) and *Exorista* (family Tachinidae) from Buldhana district.

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9

Review of Distribution and Diversity of Butterflies in and around Bhatkuli Region Dist. Amravati

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Abstract:-

Butter flies are important bio-indicators which should be protected to converse the biodiversity and environment. The survey was prepared a checklist of butter flies in Bhatkuli region. Regular survey were conducted from July 2020 to Dec 2020 by visual observation. This short term survey recorded 20 species of butter fly be longing to five families, 6 species of butterfly belonging to pirridae family, 5species belonging to papilinidae, 4 species belonging to Nymphalidae, 3 species of Lycaenidae and Hesperidae showed lowest species with 2. This study will enlighten information regarding the diversity of butterflies and forms a base line data for future butterfly studies.

Keywords-Butterflies, Bhatkuli, Diversity, Specimen, Family, Species, Checklist

Introduction :-

The Butterflies are the most attractive element of the biological diversity of the universe.(Losey and Vaugham 2006). They are beautifully colored ecologically important insects belongs to order Lepidoptera of class Insecta Butterflies show Co-evolutionary relationship with the plants and performs prominent roles in pollination(Tipleet,al 2006).As pollinators butter flies are Valuable creatures in maintaining the pollution dynamics of feral composition of natural and man-made ecosystems (Klein et,al 2008)have estimated that 35% of food used by human contributed from crop pollinated by insects majority by butterflies. As an integral part of prey-predator system they play major role in maintaining ecological balance in any type of ecosystem. As a biological indicators butterflies are useful in monitoring the ecological imbalance due to pollution. Uncontrolled exploitation of natural resources illegal encroachment and significant in studying the
Impact of rapid urbanization of ecology in developing countries like India (Khoirunnisas et,al 2015).

Butterflies are one of the most conspicuous species of earth's biodiversity andare extremely responsive to any change in their environment such as temperature, humidity, light and rainfall patterns (Murphy D.D and weiss S.B) They have differenthabitat types for muting breading and nectaring there are 16,823 species recordedfrom all over the world among them 1501 species of butterflies are recorded in India (Ganorkar 1996) of the varies butterflies habitats found in India. The western Ghat is one of the most diversified areas containing a wide variety of species of butterflies due to typical co-climatic and geographic features.

Butterflies are seasonal in their occurrence. They are common only for few months and rare or absent in others the seasons when they are rare or not active theyare either caterpillars or pupae. When they are active called as flight period distinct flight periods naturally imply seasonality of the early stage of butterfly as well. (Kunte 2000). In this paper an attempt is made to study diversity of butterflies in Bhatkuli Region.

Material and Method:-

The survey of butterfly was done using pollard walk method (Pollard 1977).Butterflies were observed, captured identified and released immediately at the spot ofcapture. Butterfly net was used for this purpose the dead specimen, many of them not in very good condition

were kept in butterfly collection boxes. Collecting live specimen was avoided during the study.

The butterflies were observed from study site for short period of six months from July 2022 to December 2022. During the survey an efficient protocol was adapted. The study area is visited twice a month fromI am to5.Pm.

Determination of abundance: -The species were further divided into 4 categories very common (V.C), common not rare (NK) rare (K), on their count from study area.Any specimen with count less than 10 times were placed in rare category. Count between 10 to 15 were placed in not rare category. Count between 15-20 were categorized as common. While specimen with count more than 20 times was placed in very common category.

Identification of butterflies: -The key characters used for identification were color pattern using span and mode of flight. Photographic images and collected specimens were examined carefully and identified using various references Grey et, al.(1992) Harbal(1992) and internet references (http/www butterflies all scientific names follow Varshney (1979) and classification with common English names are after winter-Blyth(1957)

Observation:-

List of butterflies recorded from survey site together with status.

Sr. No.	Family	Common Name	Scientific Name	Status
I	Popilionidae	1. Common mormon 2. CommonJay 3. ToiledJay 4. Bluemormon 5. Limebutterfly	Popilio Polytes Linnaeus Graphium doson Graphium Agamemnon Papilio Polymnestor cormer Papiliodemolecs Linnaeus	
II	Pieridae	6. Common grass yellow 7. Common Emigrant 8. Pioneer 9. Spotlessgrassyellow 10. Commongull 11. Orangetipsmall	Furenaheca becatopsilia pomona Anaphaeisqurota Euremalaetace poranerissa colotisetrida Junonia Lemonias Junonia Orithya Dunois Chrysippus Euploea care	CC V.cC
III	Nympholidae	12Lemonpansy 13. Blue pansy14Stripedtiger 15.Common Indian crow	Junonia Lemonias Junonia Orithya Dunois Chrysippus Euploeacare	CCC C
IV	Lycaenidae	14. Rounded Pierrot 15. Tinygrassblue 16. Plainscupid	Tarucus nara Zirula hylasechilades Pandava	U
V	Hesperidae	17. Riceswift 18. Small brandeds wift	Borbocinnara Wollaee Polo pidasmathias	V.c

Based on the result obtained from the study on the butterfly diversity in thestudy area peridae family was found maximum number of species of butterfly amongall the families followed by palilianadae Nymphalidae, Lycaenidae and Hesperidae therefore it is concluded that the study are rich in butterfly diversity and further

Research could be conducted to obtain details and documentation on butterfly diversity for the conservation in Bhatkuli

Result and Discussion:-

The present study is small effort to investigate the butterfly diversity in Bhatkuli district Amravati. In the present investigation a total five families were recorded over a period of six months fromJuly 2022 to December2022. The total butterflies species observed is tabulated in table I results showed that total 20 species of butterfly belonging to five families were recorded

in the study area pieridae family comprised six species of butterfly followed by Pqpilinidae with five species. Family followed by Nympholidae with 4 species of butterfly family lycaenidae followed with three species. Family Hesperidae showed lowest with 2species of butterfly.

Based on family wise composition of checklist of the species of butterfly observed in the studyarea. Papilionidae family showed high number of the species of butterfly among the other families with may be due to adaption and habitat preference of the species similar studies pre-parted by Singh and clib (2014). Onpreliminary checklist of butterflies recorded 125 species of butterfly from 78 genera belongs to 5 families.

The diversity is niche time stability dependent meaning if a large number of nichesare available higher diversity is found in general homogenous condition yield lower diversity while heterogeneous condition yield higher diversity (Sanders 1968, Gray1980).

As the work is restricted to very short period is 6months and the occurrence of the butterfly show seasonal prevalence and within short period of 6 months.

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10

POLYMORPHISM OF ERYTHROCYTES IN THALLASEMIC PATIENTS FROM AKOLA CITY

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ABSTRACT
Thalassemia is the most common inherited blood condition in the world. This condition is caused by changes to the genes for hemoglobin. Hemoglobin is a protein in red blood cells that carries oxygen around the body. Changes affecting hemoglobin result in severe anemia. Thalassemia is usually diagnosed within the first six months of life and can be fatal in early childhood without treatment. There are two different types of thalassemia: alpha (a) and beta (b). Alpha-thalassaemia involves genetic changes in two genes (HBA1 and HBA2). Beta-thalassemia involves changes in one gene (HBB). Alpha-thalassaemia is more common in countries in Africa, Asia, and the Middle East. Beta-thalassaemia is more common in Mediterranean countries.
Keywords: thalassemia, RBC, hemoglobin.

INTRODUCTION
In thalassaemia, the genetic defect results in reduced rate of synthesis of one of the globin chains that make up hemoglobin. Reduced synthesis of one of the globin chains can cause the formation of abnormal hemoglobin molecules, and this in turn causes the anemia which is the characteristic presenting symptom of thalassemias.

The disease is particularly prevalent among Mediterranean peoples, and this geographical association was responsible for its naming: Thalassa is Greek for the sea, and Haema is Greek for blood. InEurope, the highest concentrations of the disease are found in Greece, including the Greek islands; in parts of Italy, in particular, Southern Italy and the lower Po valley; and in the Italian islands. Sicily, Sardinia (islands located at the Italian peninsula), Malta, Corsica (French island and Cyprus and Crete (Greek islands) are heavily affected in particular. Other Mediterranean people, as well as those in the vicinity of theMediterranean, also have high rated of thalassemia, including Middle Easterners, North Africans, and South Asians. The highest concentration of carries (18% of the population) is in the Maldives.

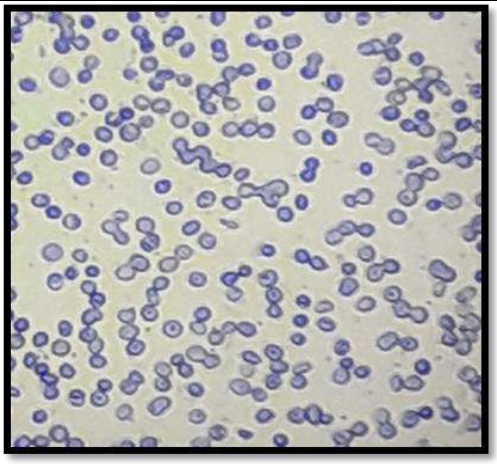
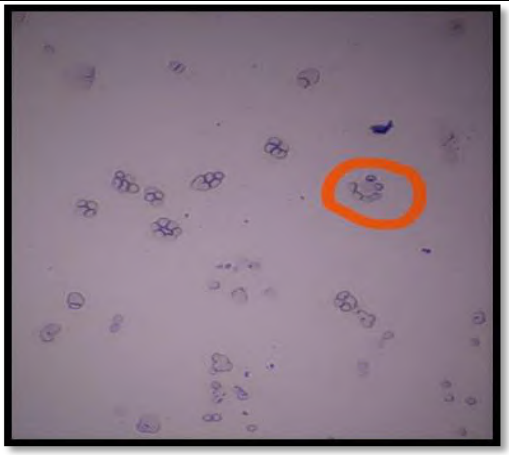
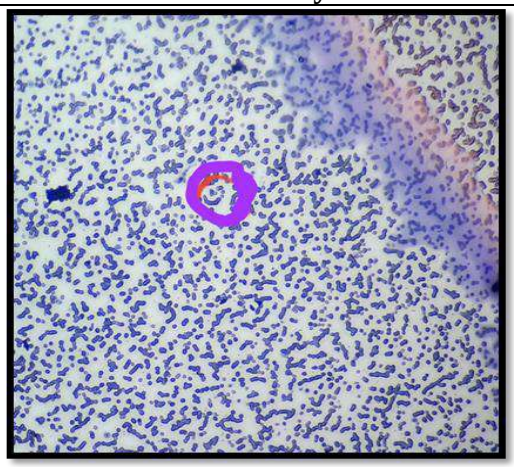
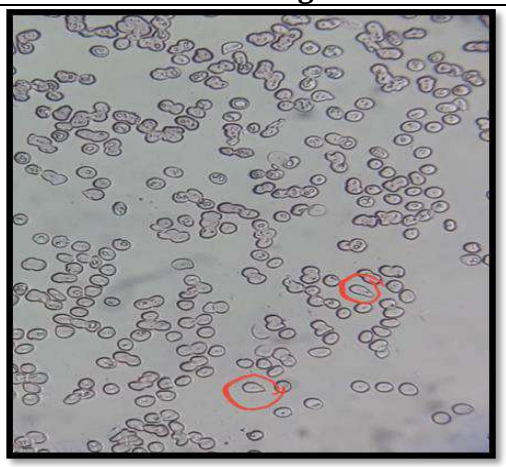
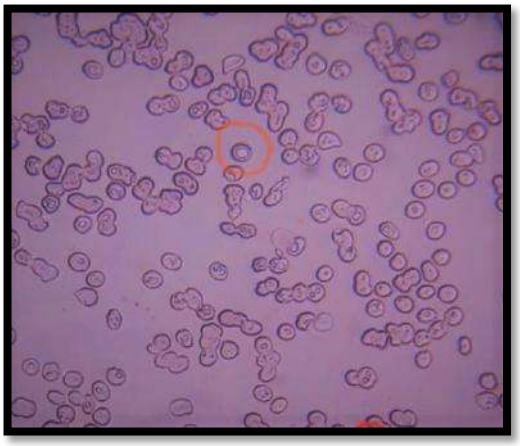
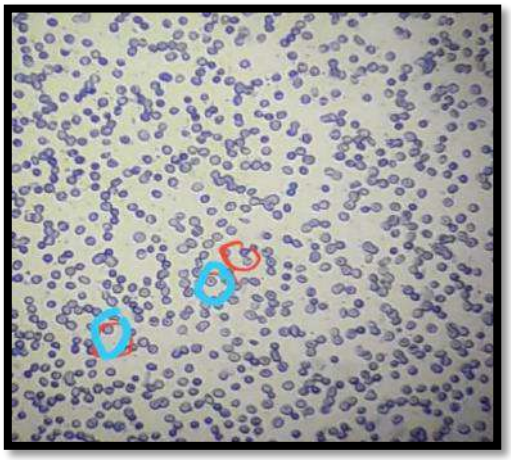
MATERIAL AND METHOD
Survey:- Akola is a city in the state of Maharashtra, located in the Vidarbha region of central India. Akola district has an area of about 54.31 square kilometres and a population of 6,629,000. Major hospitals include civil hospitals and government hospitals. From October 2016 to March 2017, a survey of patients with thalassemia major was conducted in Akola City. Twenty-eight thalassaemic children were enrolled as study subjects who are transfusion dependent from different hospitals as well as blood bank[21].

OBSERVATIONS&RESULT
3.1.Survey Some salient features observed in patients with thalassemia were:

1) Gender bias with a M:F ratio of 4:1
2) Mean age at initial diagnosis was 4±12 months and
3) The mean number of transfusions in the preceding year was 17.7±3.9 units.
4) After blood group detection, it was found that most of the patients are B+ve and AB+ve. One patient found to be A-ve.

3.2 Blood Collection: - 5ml blood from transfusion dependent patients with thalassemia major collected in a vial with anticoagulant.

3.3: Peripheral blood smear showing abnormal erythrocytes (Photo-Plate 1)

	
Coanocytes	**The Target cell**
	
The Sickle cell shaped RBC	**The Tear Drop Cell**
	
Anulocyte	**The Target Erythrocytes**

CONCLUSION

Different populations are ignorant about the medical, social, and financial burden of the disease. This further compound the problem. Primary prevention of thalassemia major is possible through carrier detection and the availability of antenatal diagnosis. In preventing this disease, the attitudes of the population at large and those of families directly involved in

the care of affected children are of primacy. Cyprus, Greece, Italy, and Sardinia are excellent examples demonstrating the effectiveness of control programs for preventing the birth of thalassemia-major children. In the Indian context, prevention of thalassemia is also possible only by bringing about sensitization at individual, social, and state levels.

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11

Determination of heavy metals in spinach by Atomic Absorption Photometric Method

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Abstract
Heavy metals were determined from the leaf of spinach from various market places of Amravati through atomic absorption photometric method. The result showed that maximum concentration of cadmium was about 21.53 ppm and minimum concentration was found 5.8 ppm. Present study showed higher conc. of Cadmium in spinach leaf powder. Highest concentration of calcium was about 125.08 ppm and least concentration was 92.32 ppm, Maximum concentration of sodium was 28.14 ppm and minimum concentration was about 20.91 ppm, highest concentration of lead was about 0.24 ppm and least concentration was 0.02 ppm. Obtained values were compared with available literature.
Keywords: Heavy metals, atomic absorption spectrophotometer,

Introduction

Eating vegetables regularly in diet can have many health benefits by reducing diseases and used to convert fats and carbohydrates into energy **(Mercola, 2014).** Vegetables are important components of the human diet since they provide essential nutrients that are required for most of the reactions occurring in the body **(Gilden, *et al*., 2010).** Rapidly increasing urbanization and emission of heavy metal contaminated fumes from the industries and vehicles have contribution to agriculture soils and consequently in food chain by deposited on the vegetable surfaces during their production, transport and marketing. Application of wastewater to irrigate agricultural lands is one of familiar practice in suburban and industrial areas in many parts of the world **(Gupta, *et al*., 2008).** According to chemical properties, heavy metals are elements that reveal metallic properties and are defined based on density, atomic number or atomic weight, chemical properties or toxicity **(Sanchez, 2008)**. Recent reports indicated that heavy metals take driver's seat among the chief contaminants of leafy vegetables. Dietary ingestion of heavy metals infected vegetables may pose serious hazard to human health. Plants can receive these metals from soil by their roots, transport them upwards to their shoots, and finally collect them inside their tissues, although there are large variations among different plant species in terms of metal gathering capacity **(luo, *et al*., 2011)**. It has been reported that almost half of the means of intake of lead, copper and chromium through food is due to plant origin (fruit, vegetables and cereals) and it sometimes in more than permissible limits within urban areas **(Yebppella, *et al*., 2011)**. Conversely, nutritional value and consumer reception must be taken into contemplation when vegetables are being considered as food, because vegetables can contain both essential and nonessential elements over a wide range of concentrations **(Chien, *et al*., 2002).**

Diverse agro climatic conditions of the country permit growing of several vegetables round the year. Uncontrolled irrigation of crops with sewage water leads to the accumulation of some potentially toxic metals in agricultural soil and have very adverse effects on the growth of the plants (Muhammad, *et al.,* 2013). Waste water contains few important plants growth nutrients like potassium (K), zinc (Zn), phosphorus (P), nitrogen (N), and organic solids (Gibbs, *et al.*, 2006). Whereas it is also the prime source of potentially hazardous organic and

inorganic toxic materials. Heavy metals like Iron (Fe), Copper (Cu), Ni and other trace elements are important for proper functional of biological systems and their deficiency or excess could lead to a number of disorders. Food chain contamination by heavy metals has become a burning issue in recent years because of their potential accumulation in bio-systems through contaminated water, soil and air (Lokeshwani and Chandrappa, 2006). Many studies have shown that sewage water irrigation has elevated the levels of toxic heavy metals such as nickel (Ni), Zn, cobalt (Co), manganese (Mn), and Fe in receiving soil (Ali, *et al.,* 1996; Singh, *et al.,* 2004; Mapanda, *et al.,* 2005), and after that vegetables take up metals by absorbing them from that contaminated soils, as well as from deposits on different parts of the vegetables exposed to the air from polluted environments (Oluwole, *et al.,* 2013).

One of the major constraints in vegetable production is pest problem and crop losses in the country due to various pests range from 10 to 30 per cent each year depending upon the severity of pest attack, pest control by use of chemicals has been playing a vital role in sustainable increase in the vegetable production. Farmers use pesticides as first line of defence for the management of pests and frequently resort to indiscriminate and non-judicious use of pesticides, which leads to several problems such as resistant development in insects / pathogens, resurgence of pests due to destruction of natural enemies, toxic hazards due to pesticide residues on the edible products and deficient pollination due to destruction of pollinators resulting in non-setting of fruits and low yields (Kodandaram, *et al*., 2013). The problem of environmental pollution due to toxic metals has begun to cause concern now in most major metropolitan cities. The toxic heavy metals entering the ecosystem may lead to geoaccumulation and bioaccumulation (Lokeshwari and Chandrappa, 2006).

India is the second largest producer of vegetables in the world and accounts for about 12 per cent of world production of vegetables with the productivity of 15 tonnes per ha which is quite low compared to other countries. The current production level is over 87.5 million tonnes and total area under vegetable cultivation is around 6.2mha which is about 2.8 per cent of the total area under cultivation in the country (Sharma, 2008). Vegetable crops provide an important source of income for the small and marginal farmers of India. The increase in population, urbanization and the rising incomes have given great impetus to the cultivation of vegetable crops which form an important source of minerals, particularly calcium, magnesium and iron, vitamins like A, B-complex and C and fibres. Vegetables are largely required in the vegetarian diet of our people and the demand for vegetables is increasing (Bose and Som, 1986). Moreover vegetables add variety and taste to Indian diets. At present it is recommended that 5 servings of different vegetables and fruits should be consumed per day to keep healthy (Srilaxmi, 2009). Spinach is just like other vegetable plant used as food and constitute an important part of human diet because is rich in carbohydrate, protein, as well as vitamins, minerals and trace elements (Oluwole, *et al.,* 2013).Leafy vegetables 'Spinach' is said to be the 'Prince of vegetables' (Caleras, *et al.,* 1982). They are also made up of chiefly cellulose, hemi-cellulose and pectin substances that give them their texture and firmness (Sobukola and Dairo, 2007). The food value of spinach is very high as it is a source of high grade iron. The composition of iron in leafy vegetables per 100 g edible portion of Spinach beet (Palak) is 16.2 mg, Spinach - 10.9 mg. The other chemical constituents of spinach are essential amino acids, Vitamin A and ascorbic acid (Singh, *et al.,* 2001). Eating vegetables is one of the most important pathways for the human body to absorb dietary minerals necessary for healthy development, but unfortunately harmful elements such as heavy metals may lead to intoxication due to prolong accumulation found in these vegetables (Elsevier, 2008). Some of these metals after accumulating in the soil are transferred to food chain which can cause serious health hazards to human beings and animals. Besides, metals induce deficiency of other nutrients like Cu, Fe and Mn by inhibiting plant uptake of Zn, possibly because of competition for the same carrier site in soil-water system (Hafiz, et al., 2013). Metals like Fe, Mn, Co, Cu,

and Ni are essential nutrients but their permissible limits are quite low in living organism (Qadir, *et al.,* 2010). Heavy metals are considered one of the main sources of pollution for the environment because they have an important effect on its ecological quality (Sastre, *et al.*, 2002) and are substances with a specific gravity of greater than 4.0 or 5.0g/cm3 Heavy metals refer to any metallic element that has a relatively high density and is toxic or poisonous at high concentration. They include mercury, cadmium, Arsenic, Chromium, Thallium and lead. Heavy metals are natural components of the earth's crust; they cannot be destroyed or degraded. To a small extent they enter our bodies via food, drinking water and air **(Harrison, 1981)**. Vegetable plants growing on heavy metal contamination medium can accumulate high concentration of trace elements and cause health risk to consumers **(Long, *et al.*, 2005).** Cadmium is an acutely toxic metal and therefore needs to be frequently monitored in the food chain because it is readily taken up and translocated to different parts of plants **(Al-Alawi and Mandiwana, 2007).** It has been well established that exposure to an excess of cadmium produces adverse health effects for humans. Cadmium can be incorporated into blood by adsorption in the stomach or the intestine after food or water ingestion or by absorption from the lungs after inhalation. It mainly accumulates in the kidney, and at high levels it can lead to serious kidney failure **(Nezio, *et al*., 2005).**

Food safety is an important issue that attracts all of us. While the consumers are concerned about the safety of what they eat, the governments on their side are concerned with finding ways to reduce food related risks and illnesses. From a viewpoint of food safety, the consumers would prefer no or as little pesticide residues in the food as possible. Safe food indirectly contributes to health and productivity thereby providing effective platform for development and poverty alleviation **(WHO, 2002)**. As human activities increases, especially with the application of modern technologies, pollution and contamination of the human food chain has become inevitable. Heavy metal uptake by plants grown on polluted soils has been studied by many researchers (Wong, *et al.*, 2003). Therefore, it is essential to determine elemental contents of food items and to estimate their daily dietary intake **(Jansen, *et al.*, 1990).**

The present study was undertaken to detect heavy metals in vegetables by atomic absorption spectrophotometric method at Amravati city, Maharashtra state.

Material and Method

Apparatus

Test tubes, Test tube stand, beakers, Funnel, Whatman filter paper, Atomic absorption spectrophotometric unit.

Reagents

Nitric acid, Perchloric acid, Hydrochloric acid, Deionised distilled water and Methanol.

Sample preparation for analysis

To analyse metal elements by AAS, it requires pre-treatment to the sample. The acid decomposition of the sample is done by Odoh and Kolawole (2011) method. The powdered sample (2 g each) accurately weighed and placed in the conical beaker. Then sample mixed with water and 25 ml of nitric acid is added, mixed it and set aside. Now gently heating started to a sample reaction. After cooling, 10 ml of perchloric acid is added. Midway if the contained material becomes dark, add 2-3 ml portion of nitric acid and continue heating. When the contained material turns yellowish or colourless indicates that decomposition is complete. After cooling 2 ml of hydrochloric acid is added and distilled water is used to prepare the fixed volumes of solutions.

Instrument

Atomic Absorption Spectrophotometer (Model - AA 7000, Shimadzu, Japan) with oxygen air-acetylene flame analysis technique was used to analyse the digested solutions. The wavelength range works in between 185 and 900 nm. Flame temperature ambient to 3000°C, which makes it possible to choose the best atomization temperature for different elements. Hollow cathode

lamps were used as sources of radiation, background correction was provided by a deuterium lamp and fuel was air acetylene. Standard operating parameters were set and given in table no. 1 to table no. 5

The present study "Determination of heavy metals in spinach by Atomic Absorption Photometric Method" focused on detection of heavy metals from vegetable i.e. Spinach. The levels of the heavy metals were analysed by using Atomic Absorption Spectrophotometer (Model - AA 7000, Shimadzu, Japan). The organic solvents like methanol, and deionised distilled water were purchased from Labline sales, Amravati. The working standard solutions were prepared from stock solution by dilution using deionized water. All the used reagents were of analytical grade. Vegetable samples were purchased from different areas of Amravati city (Maharashtra state) during the month of October 2019 to February 2020. The collected samples were washed thoroughly and under running tap water to remove adhered particles and dust. Then samples dried in a shadow, not in direct sun light and dried spinach leaves were then grinded to prepare fine powder with the help of mixer and the transferred to the bottles with proper labelling. To make test solution, methanol was added to the powder and solution was filtered through funnel with the help of Whatman filter paper and transferred to the test tubes, the mouth of the test tube was tightly closed by cork to avoid the evaporation of solvents and volatile chemicals from the samples and was kept in refrigerator. Detection of metals into them was analysed at CIC (Central Instrumentation Cell), from Shri. Shivaji Science College, Amravati.

Observation

Table 1. Cadmium (Cd) analysis in Spinach leaves.

Sr. No.	**Parameter**	**Amt. – 1***	**Amt. – 2***	**Amt. – 3***	**Amt. – 4***	**Amt. – 5***
1	Conc. (ppm)	19.1333	5.8000	7.4000	5.8000	21.5333
2	Absorbance	0.0070	0.0020	0.0026	0.0020	0.0079

*Represents different collection area of Amravati city.

Table 2. Calcium (Ca) analysis in Spinach leaves.

Sr. No.	**Parameter**	**Amt. – 1***	**Amt. – 2***	**Amt. – 3***	**Amt. – 4***	**Amt. – 5***
1	Conc. (ppm)	92.3291	106.9458	112.0000	116.8557	125.0856
2	Absorbance	1.3025	1.5087	1.5800	1.6485	1.7646

*Represents different collection area of Amravati city.

Table 3. Sodium (Na) analysis in Spinach leaves.

Sr. No.	**Parameter**	**Amt. – 1***	**Amt. – 2***	**Amt. – 3***	**Amt. – 4***	**Amt. – 5***
1	Conc. (ppm)	20.9142	28.1498	22.2846	27.3311	25.7610
2	Absorbance	3.2203	3.5862	3.2896	3.5448	3.4654

*Represents different collection area of Amravati city.

Table 4. Lead (Pb) analysis in Spinach leaves.

Sr. No.	Parameter	Amt. – 1*	Amt. – 2*	Amt. – 3*	Amt. – 4*	Amt. – 5*
1	Conc. (ppm)	0.2494	0.0690	0.2388	0.0265	0.1380
2	Absorbance	0.0047	0.0013	0.0045	0.0005	0.0026

*Represents different collection area of Amravati city.

Table 5. Metal analysis in Spinach leaves.

Sr. No.	Element	Wavelength (nm)	Slit Width (nm)	Conc. (ppm)	Absorbance
1	Cadmium (Cd)	228.8	0.7	11.933 ± 3.472	0.004 ± 0.001
2	Calcium (Ca)	422.7	0.7	110.643 ± 5.483	1.560 ± 0.077
3	Sodium (Na)	589.0	0.2	24.888 ± 1.417	3.421 ± 0.071
4	Lead (Pb)	283.3	0.7	0.144 ± 0.044	0.002 ± 0.0008

Values represents Mean (n=5) ± SE

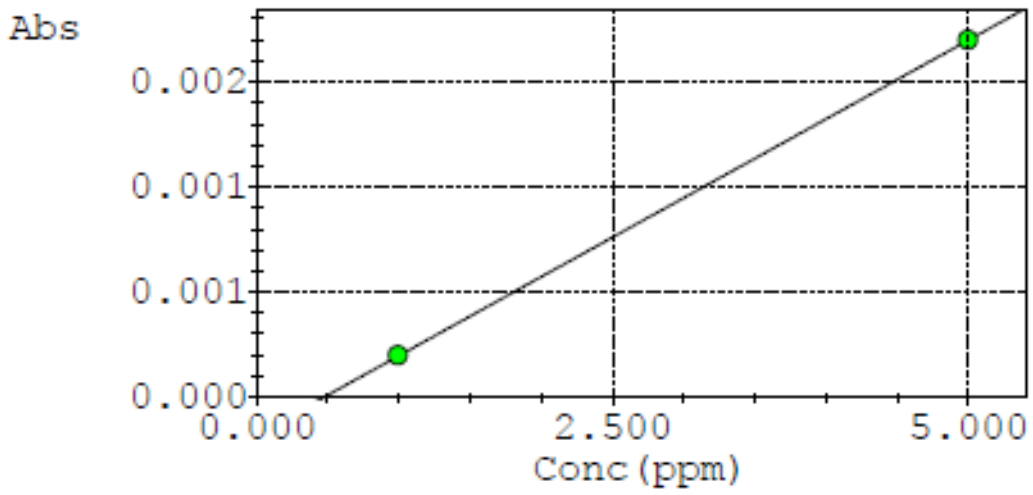

Figure 1: The calibration curve of standard Cadmium (Cd) solutions by the AAS technique.

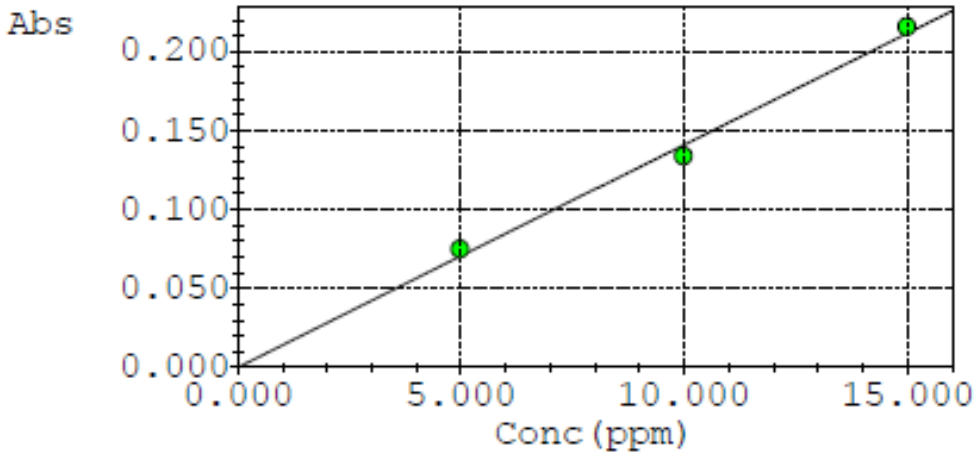

Figure 2: The calibration curve of standard Calcium (Ca) solutions by the AAS technique.

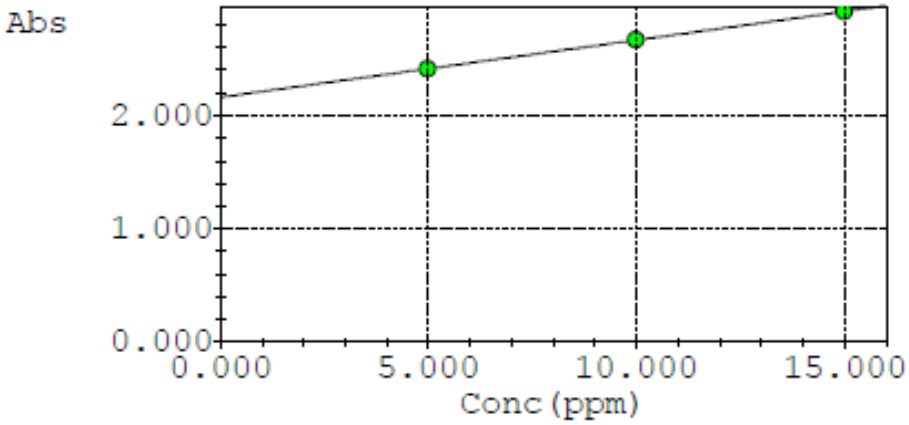

Figure 3: The calibration curve of standard Sodium (Na) solutions by the AAS technique.

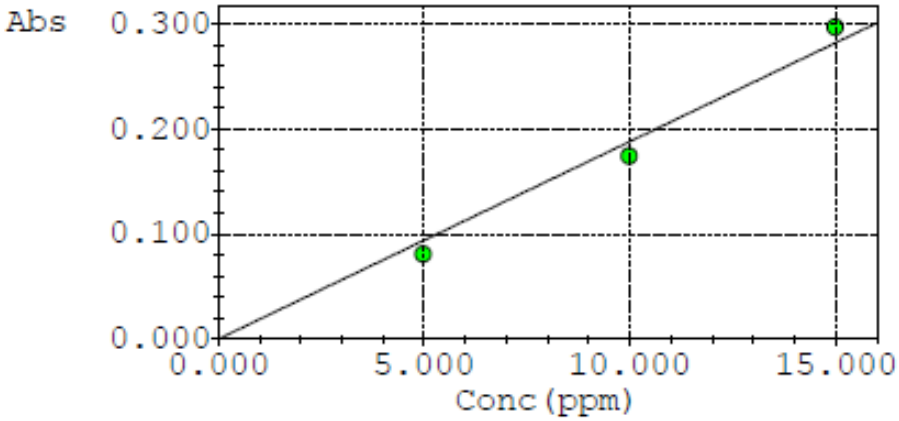

Figure 4: The calibration curve of standard Lead (Pb) solutions by the AAS technique.

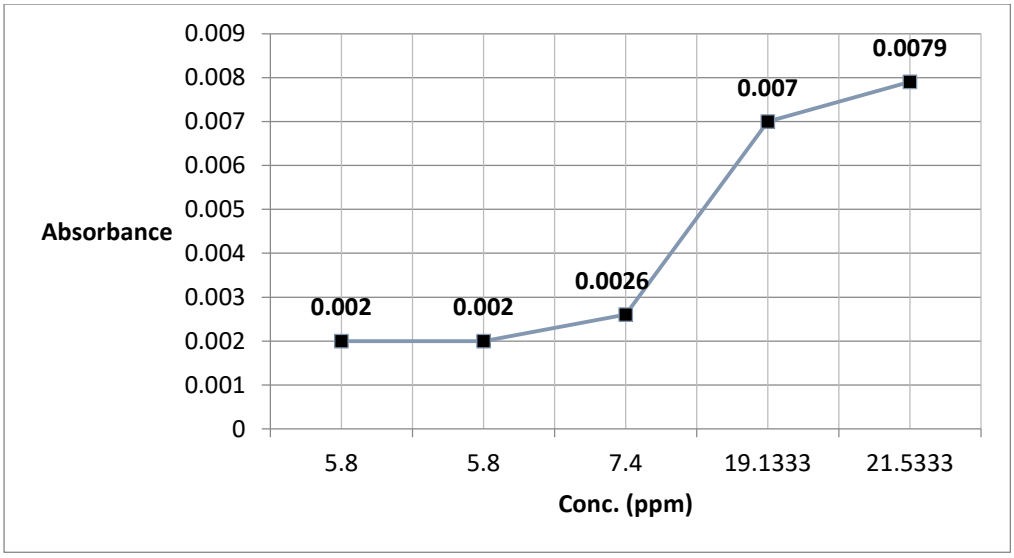

Figure 5: Cadmium (Cd) content in extract of Spinach leaves.

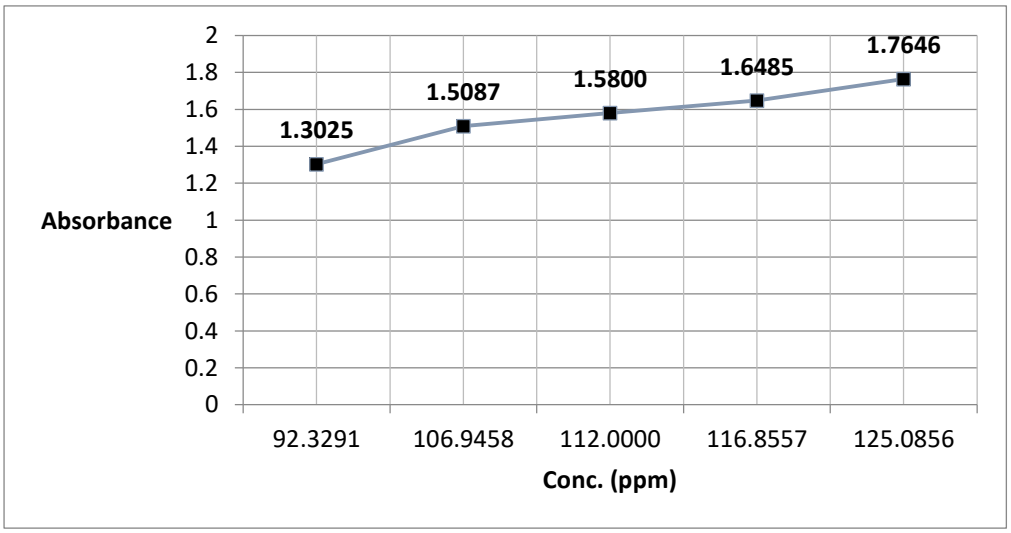

Figure 6: Calcium (Ca) content in extract of Spinach leaves.

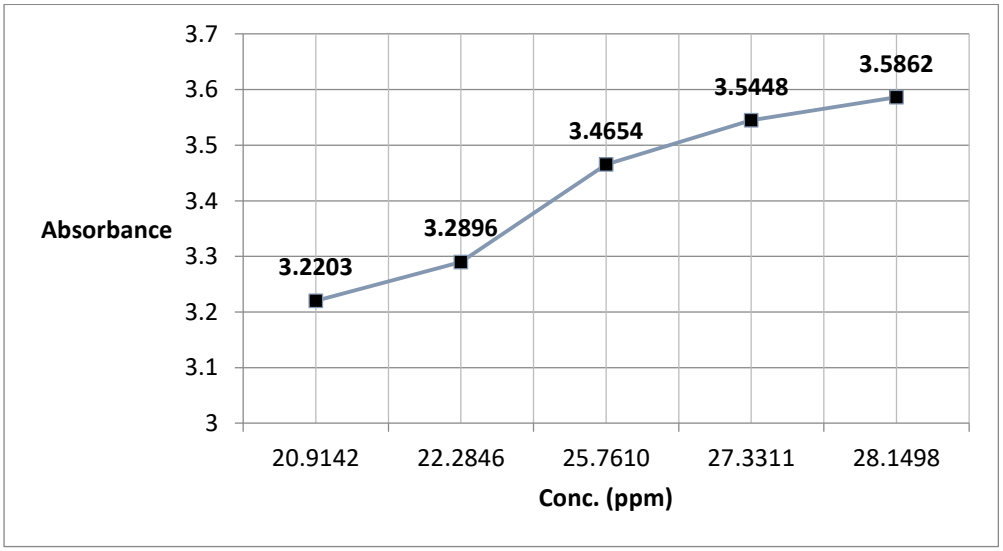

Figure 7: Sodium (Na) content in extract of Spinach leaves.

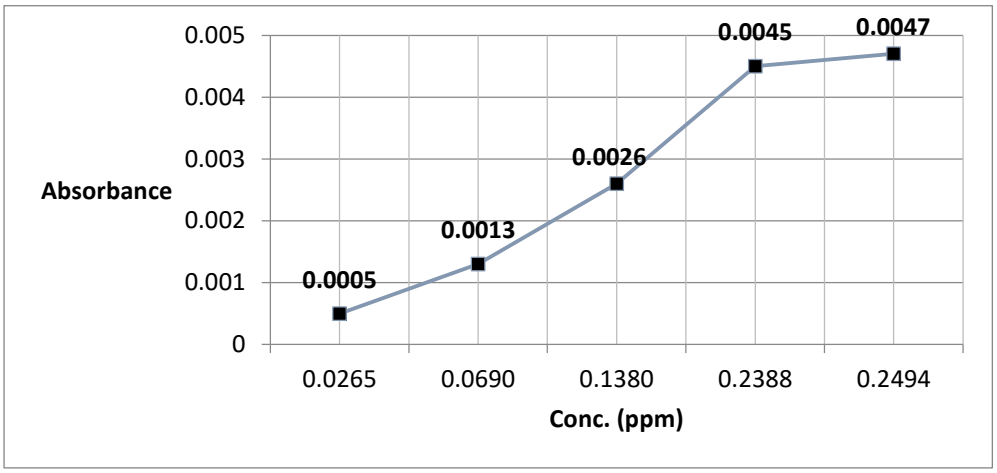

Figure 8: Lead (Pb) content in extract of Spinach leaves.

Result and Discussion

Present study was conducted to determine the heavy metals by atomic absorption photometric method in spinach from different areas of Amravati. Samples were analysed and the data so obtained was studied to draw meaningful conclusions. Findings of the study are presented and discussed in this chapter. To analyse samples by the AAS techniques, the calibration curve of each element must be determined first with good linear regression **(Altwaiq, *et al.*, 2019).** Table 1 to 5 shows analysis of Cd, Ca, Na and Pb in spinach leaves respectively. Figures 1 to 4 shows the obtained calibration curves by AAS measurements of the interested metals. These elements include cadmium, calcium, sodium and lead. Figures 5 to 8 shows content of Cd, Ca, Na and Pb in spinach leaves respectively. Maximum concentration of cadmium was found in spinach leaves which contain about 21.53 ppm and minimum concentration was found 5.8 ppm while the Mean and SE of 5 samples was 11.933 ± 3.472 respectively. **Oymak, *et al.*, (2009)** reported the conc. of Cd in spinach leaves was 0.08 ± 0.01 µg g^{-1}. So in present research we found higher conc. of Cd in spinach. Cadmium cause adverse health effects in humans and their widespread presence in the human environment comes from anthropogenic activities (Vinas, Pardo-Martinez, and Hernandez-Cordoba, 2000). The maximum limits allowed for cadmium (Cd) in vegetable edible leaves is 0.2 mg kg^{-1} (http://www.puntofocal.gov.ar/doc/r_gmc_12-11.pdf). Highest concentration of calcium was found in spinach leaves which contain about 125.08 ppm and least concentration was 92.32 ppm while the Mean and SE of 5 samples was 110.643 ± 5.483 respectively. **Siong, *et al.*, (1989)** observed 116.2 mg/100 gm in spinach. Maximum concentration of sodium was found in spinach leaves was 28.14 ppm and minimum concentration was found 20.91 ppm while the Mean and SE of 5 samples was 24.888 ± 1.417 respectively. Nerdy (2018) observed that conc. of Na in broccoli and cauliflower 118.213 ± 0.557 and 101.213 ± 0.542 mg/100g respectively. Highest concentration of lead was found in spinach leaves which contain about 0.24 ppm and least concentration was 0.02 ppm while the Mean and SE of 5 samples was 0.144 ± 0.044 respectively. **Parvin *et al.*, (2014)** observed 0.17±0.018 concentration of lead in spinach sample which was collected at industrial area. The conc. of lead was found lower than the safe limit because high lead concentration leads to health risk in human beings **Demirezen and Ahmet (2006).** Toxicity by the heavy metals to human is well documented **(Nordberg, *et al.*, 2007).** Cd and Pb are extremely toxic to humans; which accumulate in internal organs over time **(Zanini *et al.*, 1992, Singh, *et al.*, 2004, Chen, *et al.*, 2005, Turkey, *et al.*, 2017).** It is reported that human skin absorb lead and within six hours urine, blood and sweat showed increased concentration (Stauber, *et al.*, 1994).

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12

Histopathology of *Indian Major Carps* infected with Trichodinid parasites

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Abstract
The present study was undertaken to study the histopathological effect of Trichodinid parasites on Indian major carps, *Labeo rohita, Catla catla and Cirrhina mrigala. T*he results of the present study indicate heavy infection of Trichodinid parasites namely *Trichodinella epizootica, Tripartiella obtusa, Tripartiella copiosa, Tripartiella bulbosa* with strongest effect noticed on target organ i.e. on gill surface. Histopathology reveals the parasite induced intensive proliferative changes on the epithelium of the gill filaments and the proliferation appeared extending along the whole gill filaments.
Key words: Histopathology, Trichodinid parasites, *Indian major carps*, Freshwater fish

Introduction

Histopathology is an important disease diagnostic tool that detects early signs of disease not easily recognized on gross examination and helps in etiology and prevention of disease **(Meyers and Hendricks, 1982).** Furthermore, the alterations found in these organs are normally easier to identify than functional ones **(Fanta *et al.*, 2003),** and serve as warning signs of damage to animal health **(Hinton and Laurén, 1990).**

Pathological effect of these parasitic species is rare studied area hence the present investigation is carried out to study the pathological effect of of Trichodinid ectoparasites on gill surface of Indian major carps with the help of histopathology tool. The present study is also aimed in order for public understanding and support to make the people aware of the value of fish by improving protective measures against parasitic diseases with the help of histopathology tool.

Materials and Methods

The host fish for the present study were collected from various dams and the local fish market in Washim. The collected fish were brought to the laboratory for necropsy procedures for examination of Trichodinid parasites. After confirming the presence of Trichodinid in fish, the histopathological analysis was carried out.

For histopathological analysis, the infected gill tissues were fixed in 10% buffered formalin for 24 hours, embedded in paraffin, cut into 4 µm thick sections, and stained with hematoxylin and eosin. The photographs of the histopathology slides were taken with a digital camera and microscope. Identification of Trichodinid parasites in tissue sections was done using standard literature **(Lom and Dykova, 1992; Purivirojkul, 2012).**

Results and Discussion

The present study reported four species of Trichodinid ectoparasites: Trichodinella epizootica, Tripartiella obtusa, Tripartiella copiosa, and Tripartiella bulbosa, with the strongest effect noticed on the target organ, i.e., the gill surface.

The infected fish were very slimy, and the gills appeared pale in color with excessive mucous production. The phase contrast microscopic examination of the gills of the carp revealed a moderately altered structure when invaded by Trichodinids. (Fig.1-4). Trichodinids were located between secondary lamellae, on the tip of the (sometimes shortened) secondary lamellae. It was observed that the parasite induced intensive proliferative changes on the epithelium of the gill filaments, and the proliferation appeared to extend along the whole gill filament. As a result, the secondary lamellae fused together and appeared as one unit, and in

many cases, the proliferated epithelial cells displayed degeneration and desquamation in most of the superficial cells of different degrees, either vacuolar and/or complete (Figures 5 and 8).

Although most gills examined were functionally normal, certain changes in gill structure were regularly noticed, e.g., subepithelial oedema of the secondary epithelium, focal hyperplasia between secondary lamellae, curling of the pillar cell system, and mild circulatory changes, together with hypertrophy and hyperplasia of gill tissues **(Somatkar and Dabhade, 2015).** In addition to these lesions, an excessive accumulation of mucus on the gills of infested fish with Trichodinid infection caused serious pathological lesions in the gills, such as hyperplasia of the epithelial cells and clubbing and fusion of the gill filaments, which have been previously reported by many authors **(Padnos and Nigrelli, 1942). Davis (1947)** reported hyperplasia and necrosis in the gill tissue of fish infected by Trichodinid ciliophorans. **Sarig (1971)** observed Trichodina sp., Tripartialla sp., and Glassatella sp. to be so abundant on gills and skin that they destroy the normal structure of the epithelium of host fishes. **Lom (1973)** extensively studied the mechanism of the injury of host cells by Trichodinid ciliophorans. Extensive damage to gills, i.e., hypertrophy, vacuolar degeneration, and desquamation of epithelial cells, is due to the presence of trichodinids. This was also reported by **Ahmed (1976), Mcardle (1984), Eisa et al. (1985), Das and Pal (1987), Hassan (1999), Acharya and Dutta (2005), and Vera et al. (2003).**

It was observed that the Trichodinid ciliophorans are pathogenic to host fishes if present in large numbers **(Lom, 1973).** The histopathological observations indicate that the infestation with Trichodinid ciliophorans result in a wide range of deleterious changes ranging from hypertrophy and hyperplasia of gill epithelium. The histological damages in the gills inhibit the normal physiological functions of the gills**. Hughes (1972)** observed that proliferation and swelling of gill epithelium significantly reduced the oxygen uptake capacity of gills. Heavy mucous production by fish gills as observed implicates hypoxic condition **(Gardener, 1975). Lom (1973)** reported the presence of stimulating substance of fish which helped Trichodinids to proliferate massively. The presence of high amount of mucous might have provided a congenital environment for ciliates. The massive mucus production on infested fishes is a defense mechanism produced by the host to eliminate the parasite. It is likely that those moderate gill alterations were not induced exclusively by trichodinids, since they coincided with unfavorable environmental conditions in the fish-ponds, i.e., low dissolved oxygen concentration, high water temperature, high stock density, etc., which could also lead to increased mortality of fish fry in the rearing ponds **(Bykovskaya- Pavlovskaya, 1964; Kabata, 1985).**

Conclusion

Histopathological analysis of *Indian Major Carps* revealed four species of Trichodinids namely *Trichodinella epizootica, Tripartiella obtusa, Tripartiella copiosa, Tripartiella bulbosa* seriously damage the epithelial or epidermal cells by their constant ectoparasitic attachment and movement. Under these circumstances the trichodinids behave like serious ectoparasites, feeding on disruptive surface and associated bacterial growth. These may even penetrate into gills or skin tissues and feed on it and multiply in large numbers. The damage of gills due to presence of this parasite might be the cause for mortality of carps often encountered. So in order to prevent the tissue damage of fish due to Trichodinid ectoparasites, it is needed to disrupt the steps of parasitic transfer from one host to another by avoiding overcrowding of fishes and transfer from one water body to another with suitable and effective preventive and control measures.

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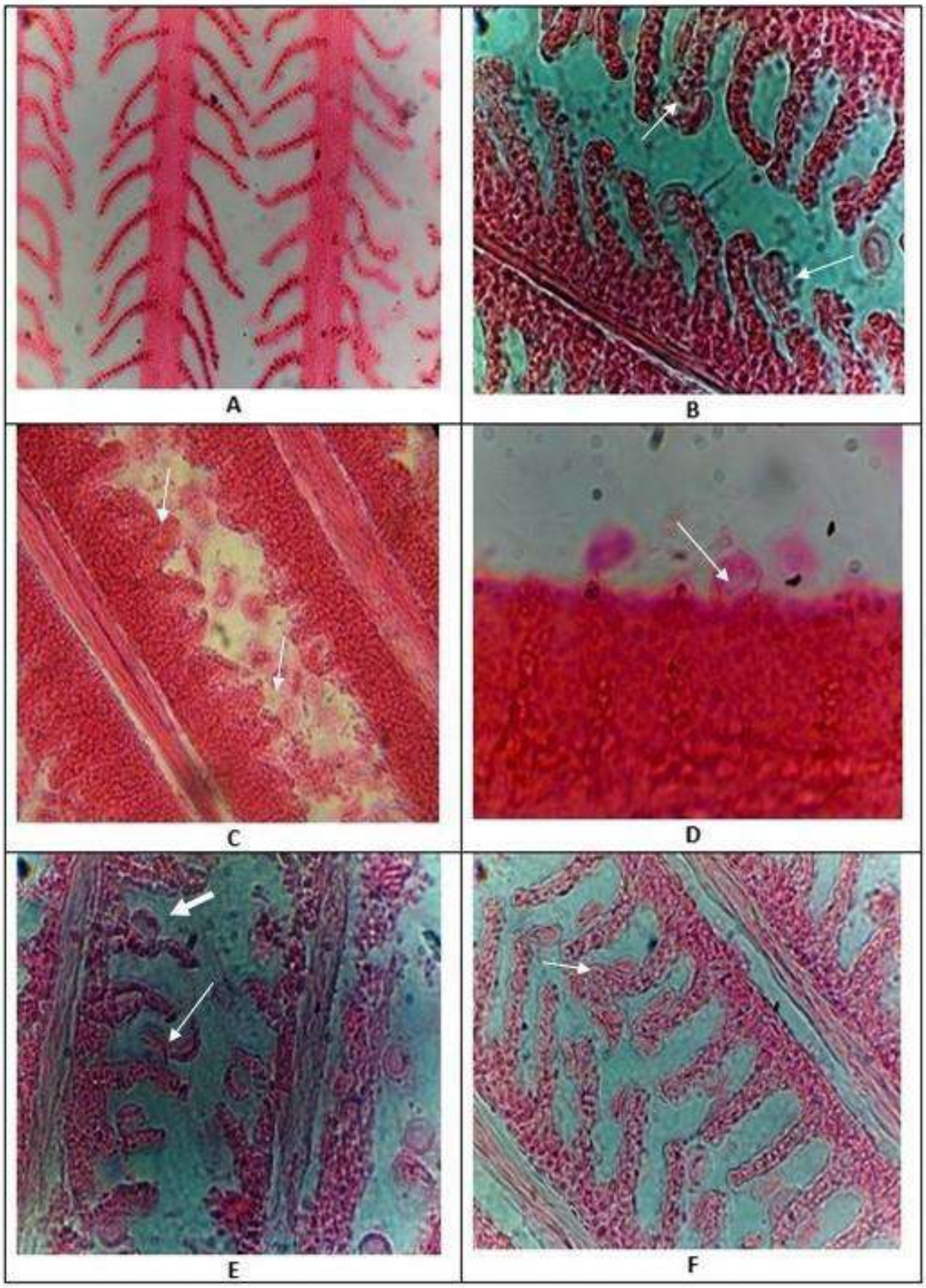
Photoplate I: Histopathological effect of Trichodinid ectoparasites on gill tissues of Indian major carps. ***A: l.S. Of Gills showing healthy gill tissues. B-F: Infected gills of Indian major Carps showing heavy load of Trichodinid parasites.***



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Growth promoter effect of garlic (Allium sativum) oil on catfish *Clarias Batrachus* (Linnaeus, 1758)

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Abstract -
The research aimed to examine the effect of garlic on growth performance of catfish *Clarias Batrachus* **(Linnaeus, 1758)** This fish found in stagnant, slowly moving water like ponds, swamps, streams, rivers, paddy fields, and ephemeral pools. For experiment, the specimens were Brought from fishmarket of the local 'Wadali Lake' near Amravati. The fishes measuring about 20±0.5 cm length and weighing ranges from 50±05g in weight were selected for the experimental study. After acclimatization for two weeks and maintained in glass aquarium. The specimens were fed on formulated diet. During the experiment, fish was fed with pellet containing 0.5, 1and 2 ml garlic oil per kg pellet at 5% of body weight per day, twice a day at 08.00 am and 16.00 pm. Feeding period was four consecutive weeks. Growth of fish was measured at the end of feeding period. The result showed that feeding the fish with garlic oil - supplemented pellet for four weeks had significant effect ($p<0.05$) on fish growth as compared to that of control fish. The highest weight gain was achieved in fish fed pellet supplemented with 1 ml garlic per kg pellet. As conclusion, incorporation of garlic oil into fish feed improved the growth performance of fish.
Keywords: garlic, Allium sativum, *Clarias Batrachus* , weight gain**.**

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Evaluation of the growth and survival of fingerlings of the freshwater fish *Labeo rohita* after feeding them with extract of *Ocimum sanctum*.

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Abstract:
The studies were carried out using feed supplemented with *Ocimum sanctum* extract to determine the influence on growth performance and survival rate in *Labeo rohita* fingerlings. Fingerlings were supplied in experimental tanks at a density of 13 per tank. Experiment diets were designed with varying concentrations of O. sanctum extract, which were 200mg, 400mg, and 600mg. In addition to these three experimental tanks, one control tank was set up with no extract in their diet. Results after 60 days revealed that tank 3 with 600 mg of extract had considerably greater mean weight gain, specific growth rate, and survival. Tank 3 had a greater survival rate, but there was no appreciable difference between the other treatments. The current study's findings showed that adding Tulsi to the diet of *Labeo rohita* fingerlings had a major impact on the fish's mean weight increase, SGR, and survival rate.
Keywords: *Ocimum sanctum*, *Labeo rohita*, growth, weight, SGR

Introduction:

Fish farming in India, also known as pisciculture, is the practice of raising fish in tanks or ponds for commercial purposes. It plays a crucial role in meeting the increasing demand for fish as a source of protein in the country. India has a long history of fish farming, with traditional practices like backyard ponds and village tanks. However, in recent years, modern fish farming techniques have gained popularity. Fish farmers are now adopting advanced methods like the construction of concrete tanks, raceways are commonly used in fish farming as they provide a sturdy and durable structure for containing fish. These tanks can be designed to accommodate specific fish species, allowing for optimal water flow and oxygenation.

About 85% of all fish produced in India's freshwater aquaculture industry are big carps **(Sundaray JK et. al, 2017)**, and this percentage will not change until other species that can be widely adopted are available. In 2016, global freshwater aquaculture production reached 47.9 million tons, with carps accounting for 59.7%. With 4.2 million tons of carps, India is the world's third largest aquaculture producer, accounting for around 73.7% of total India aquaculture production in 2016 **(FAO, 2018)**. Freshwater fish of economic significance, *Labeo rohita* is extensively farmed throughout Asia and the Indian subcontinent **(Iqbal et. al, 2014; Ranjan et. al, 2017)**. According to **(Bharathi et. al. 2013)**, it is a highly desirable and flavorful fish species. It takes a year for it to reach a marketable size of 800–1,000 gram that is valued at between 200 and 300 rupees per kilogram. **(Musharraf and Khan, 2018)**.

Medicinal plants are the primary source of many secondary metabolites and essential oils, and are regarded as one of the most significant sources of medicine and pharmaceuticals in use today **(Singh, et. al, 2010)**. Since the very beginning of civilization, medicinal plants have been utilised to treat a variety of human health conditions all throughout the world **(Upadhayay et. al, 2013)**. India is known for its rich tradition of herbal medicine, and the country has a wide variety of medicinal plants. *Ocimum sanctum*, commonly known as holy basil or tulsi, is a medicinal plant native to India and Southeast Asia. It has been used for centuries in traditional Ayurvedic medicine for its various health benefits. The chemical components of the two types of *Ocimum sanctum*—black (Krushna Tulsi) and green (Rama Tulsi)—are comparable. Additionally, both types share similar therapeutic qualities **(Das S K**

and Vasudevan D M 2006). Medicinal herb extracts are sometimes used in animal growth to promote health and improve growth rates. These extracts are derived from various plants that possess medicinal properties, such as antimicrobial, antioxidant, immune-enhancing, or anti-inflammatory effects. They can be administered orally, added to feed, or used topically. It's important to note that the use of medicinal herb extracts in animal growth should be done under appropriate recommended dosage guidelines. Additionally, it is vital to ensure that any herb extracts used are safe and suitable for the specific animal species being treated. With this knowledge, a methodical assessment of the impact of *Ocimum sanctum* extract on the development and survival of *Labeo rohita* fingerlings was carried out in this investigation.

Materials and Methods:

The experiment was carried out at the Department of Zoology's Research Laboratory. Fish were obtained as fingerlings from the Fish Seed Production center.

Fish collection and management: Fingerlings of *Labeo rohita* were obtained from the Fish Seed Production Centre. The fishes were divided into four glass tanks for varying experimental feed dosages of *Ocimum sanctum* extract after 10 to 15 days.

The diet's preparation and composition: The tulsi leaves were collected, cleaned, and crushed into a paste using a mixture grinder. The tulsi leaves paste was then thoroughly combined with the fish meal in multiple doses. Tulsi extract was added separately to other ingredients, and diets were combined to produce a paste of each diet. The amounts of *Ocimum sanctum* extract given here are 200 mg in the first tank, 400 mg in the second tank, and 600 mg in the third tank. Lastly, the tank that receives no experimental food is regarded as the control tank. The necessary quantities of components were collected and precisely weighed in accordance with the feed formula, as given in the table.

Ingredients	Experimental Treatment			
	Expt. A	**Expt. B**	**Expt. C**	**Control (C)**
Mustard oil cake powder	28 gm	28 gm	28 gm	28 gm
Groundnut oil cake powder	20 gm	20 gm	20 gm	20 gm
Wheat Bran	12 gm	12 gm	12 gm	12 gm
Wheat Flour	14.8 gm	14.6 gm	14.4 gm	15 gm
Rice Flour	22 gm	22 gm	22 gm	22 gm
Vitamin	1 gm	1 gm	1 gm	1 gm
Salt	0.5 gm	0.5 gm	0.5 gm	0.5 gm
Starch (Binder)	0.5 gm	0.5 gm	0.5 gm	0.5 gm
Lime stone	1 gm	1 gm	1 gm	1 gm
***Ocimum sanctum* extract**	**0.2 gm** (for 200 mg concentration)	**0.4 gm** (for 400 mg concentration)	**0.6 gm** (for 600 mg concentration)	Nil

Duration of the experiment and the actions taken: After acclimatization, fish were divided into four tanks and kept there for a duration of sixty days. The experiment was carried out in 40-liter rectangular glass aquarium tanks. Up to thirty liters of fresh water were placed within aquarium tanks.

Growth parameter:

1. **Mean weight and Length:**
2. **Specific Growth Rate (SGR):**

3. **Statastical analysis:** ANOVA, or one-way analysis of variance, was used to analyse the data. To investigate treatment differences in terms of growth and survival, multiple comparisons were made using Tukey's test.

Result:

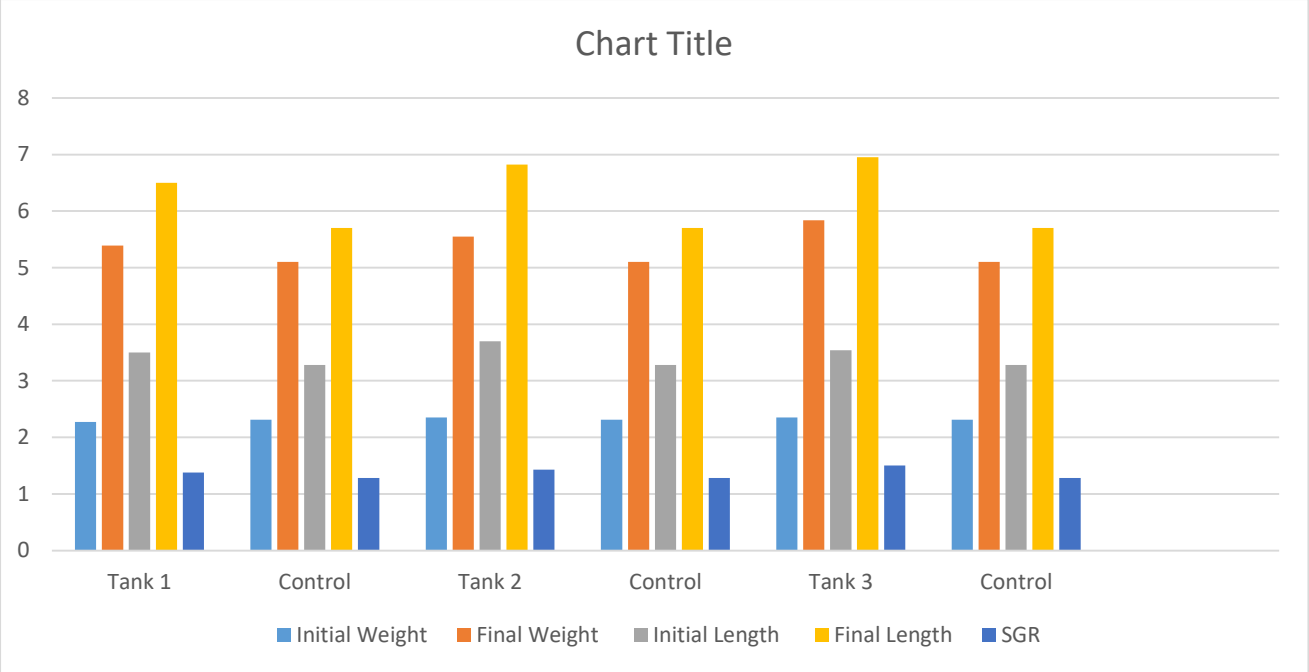
All of the fish fed actively and appeared to be in good health. The growth responses of fish given *O. sanctum* extract meal showed improvements. The fish fed the control diet showed the slowest growth, whereas the fish given 600 mg of *O. sanctum* extract meal showed the strongest growth response. The growth performance did not change significantly between the various *O. sanctum* extract concentrations. The table displays information about the fish's growth performance during the experiment.

Water's physico-chemical characteristics: Weekly analyses of the water quality parameters, including temperature, pH, dissolved oxygen (DO), and total hardness, were conducted during this trial. Every week, the temperature of the water in several test tanks was noted. The water's temperature varied between 21 and 26°C over the course of the experiment. Similarly, weekly pH readings were taken from the water in each experimental tank. Water had a pH of between 7 and 8 for the duration of the experiment. For the whole trial, the temperature and pH remained in the ideal range. Total hardness ranged from 220 to 325 ppm, while dissolvesd oxygen varied between 5 and 7 ppm during the experimental duration period.

1. **Weight gain:** The rohu fingerlings were initially measured for length and weight before being placed in the tanks. Every tank's mean weight was determined at every two-week interval. A fingerling's initial measurements included a length of 5–6 cm and a mean weight of 4–5 g. Upon completion of the 60-day experiment, the fingerlings in the first 200 mg tank weighed 5.38 gm and measured 6.4 cm in length; in the second 400 mg tank, they weighed 5.57 gm and measured 6.8 cm; in the third 600 mg tank, they weighed 5.84 gm and measured 7 cm in length; and in the control tank, they weighed 5.12 gm and measured 6 cm in length. Tank 3 had the highest final mean weight ($p < 0.05$) while the control treatment had the lowest. The table that follows shows the initial and ultimate mean weight of the *L. rohita* fingerlings after 60 days in culture.

	Initial Weight	Final Weight	Initial Length	Final Length	SGR (in %) 60 days
Tank 1 (200mg)	2.27 ± 0.03	5.39 ± 0.01**	3.5 ± 0.15	6.5 ± 0.15**	1.38 ± 0.02**
Control	2.31 ± 0.02	5.1 ± 0.06	3.28 ± 0.19	5.7 ± 0.24	1.28 ± 0.02
Tank 2 (400mg)	2.35 ± 0.02	5.55 ± 0.02**	3.7 ± 0.15	6.82 ± 0.12**	1.43 ± 0.01**
Control	2.31 ± 0.02	5.1 ± 0.06	3.28 ± 0.19	5.7 ± 0.24	1.28 ± 0.02
Tank 3 (600mg)	2.35 ± 0.02	5.84 ± 0.03**	3.54 ± 0.30	6.95 ± 0.09**	1.50 ± 0.02**
Control	2.31 ± 0.02	5.1 ± 0.06	3.28 ± 0.19	5.7 ± 0.24	1.28 ± 0.02

Mean weight (g) of *L. rohita* fed with *tulsi* supplemented diets during experimental period (n=5 fish, Mean ± SD). Mean values with different superscripts in the same row are significantly different ($P<0.05$), where values are significant at * means moderate and ** means highly significant. Specific growth rate (%) of *L. rohita* fed with *tulsi* supplemented diets at the end of experimental period (Mean ± SD).



Mean weight (gm) and length (cm) and specific growth rate (%) of *L. rohita* fed with *tulsi* supplemented diets at the end of experimental period in comparison with control fish L. rohita.

2. **Specific Growth Rate (SGR):** A certain growth rate's value is utilized to compare growth on a daily basis. A nutritional supplement plays a substantial impact in growth performance, as seen by the significantly greater growth rate and specific growth rate. After 60 days of the investigation, third tank (diet with 600 mg of *O. sanctum* extract supplement) had the greatest SGR ($p < 0.05$), which was substantially different from treatment T1, treatment T2, and control treatment. According to Table below, the treatments mean showed that tank 3 (1.50%) had the best SGR, followed by tank 2 which is of 400 mg extract, tank 1 of 200 mg, and forth tank of control.

The current study's findings showed that tank 3 of 600mg concentration of *Ocimum sanctum* extract supplemented had considerably greater mean weight increase and SGR ($p < 0.05$). It was discovered that tank 3 (600 mg) had a better survival rate. According to these findings, it is advisable to add 0.6 gram of tulsi powder per 100 grams of *Labeo rohita's* fingerling diet in order to increase the fingerlings' mean weight growth, SGR, and survival rate.

Discussion:

The *Ocimum sanctum*, or tulsi, is regarded as the queen of herbs and has extensive medical properties that are well-documented in Hindu mythology. However, there are many more herbal plants in the world. The fish in this experiment that were fed a diet mixed with tulsi extract grew noticeably faster than the control group. As the concentration of the tulsi extract supplement increased, the growth rate also increased. The Nile tilapia *(Oreochromis niloticus)*, when fed diets enriched with tulsi, exhibited a marked increase in weight gain, length, SGR and FCR **(Panprommin et al., 2015)** in contrast to the fish given a control diet. In the current investigation, fish fed a meal supplemented with tulsi exhibited considerably faster growth, increased in SGR and FCR than fish fed a control group. Similar results were also found by (**Immanuel et al. 2009)** after supplementing *Oreochromis mossambicus* fish with extracts from four medicinal herbs. As **(Bhavan et al. 2011)** investigated the impact of a meal supplemented with tulsi powder on *M. rosenbergii*, they found that the prawn fed the tulsi powder diet had the best survival rate (84%), as compared to the prawn fed the control diet.

These findings were consistent with the current investigation. As the amount of tulsi in the diet grew, so did the survival rate.

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Physicochemical characteristics of Kapsi lake, Kapsi Dist. Akola (Maharashtra) India

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Abstract :
The physicochemical characteristics and features of the water in Kapsi Lake, Kapsi, were studied from December 2022 to November 2023. Monthly water samples were collected from selected 8 stations of Kapsi Lake with the help of a Ruttner water sampler from specific depths. Colour, odour, turbidity, transparency, pH, conductivity, T.D.S. Dissolved oxygen, free C02, total hardness, calcium hardness, magnesium, P. alkalinity, total alkalinity, chloride, sulphate, and nitrate of water were tudied. Colour of water is apparent and seston Theodor of the water is earthy. Water temperatures of 28-29 degrees Celsius were recorded. Monthly variation of physicochemical characteristics features of water were studied, and stationwise variation in physicochemical characteristics was recorded. Minimum value of nitrate 0.010 mg/lit.at station no.1 were recorded. The maximum value of physicochemical characteristics recorded at station no. 8 is magnesium. The coefficient of co-relation of physicochemicals was also studied.
***Keywords**:* Kapsi lake, Kapsi, Physicochemical characteristics, coefficient of corelation.

Introduction

Water is one of the ubiquitous elements in the world that support vitality in every ecosystem, whether it is aquatic, terrestrial, or otherwise. The water quality of any aquatic ecosystem can be better understood through information about its physical, chemical, and biological regime. Aquatic ecosystem is the most varied ecosystem hence it's hydrobiology vary one to another water body. The physicochemical properties of water and the co-related dependence of all vital activities make it mandatory to take account of its hydrobiological investigation to understand the ecology of a reservoir. The present study deals with an evaluation of water quality in terms of the physicochemical characteristics of Kapsi Reservoir, a tropical reservoir meant for fisheries activity. It was constructed near Akola: Kapsi village in Akola District of Maharashtra. It is surrounded by slum areas. Rivers and reservoirs play a major role in agriculture and fisheries, along with the use of water for drinking purposes. Several factors that determine the water quality of a reservoir include seasonal climatic changes (Chapman, 1996; Barik et al., 2010), seasonal precipitation, wind action, the geologic origin of the catchment basin, and the pattern of hydrological cycle prevalent in the lake (Tundisi & Straskraba, 1999).

The study of lakes as a science started as early as 1887, when Micros described the lake as a 'Microcosm'—a little world within itself. Forel (1901), for the first time, designated these studies as 'limnology' and wrote an inspiring book, 'The Science of Lakes, which provided an impetus for investigations on freshwaters, and many workers entered into this new field. Among the pioneer workers, the contributions of Naumann, Welch, Birge, Juday, Thienemann, Ohle, Pearsall, Brehm, and Ruttner laid firm foundations. An epoch-making monographic contribution by G.E. Hutchinson, 'A Treatise on Limnology, appeared a little later. The global consciousness towards the freshwater systems arose by this time, and ultimately the International Biological Programme and International Man and Biosphere Programme (No. 5) were floated to generate information on the structure and function of inland aquatic environments, their productivity, and the impact of human interference on them, realising that

freshwater resources are finite and may limit the growth and development of mankind on this planet.

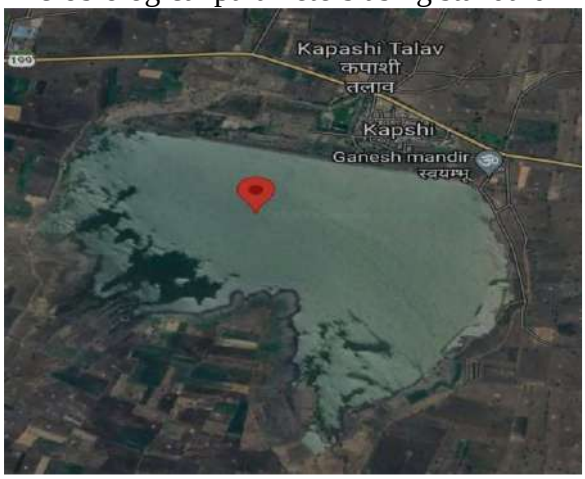
The freshwater systems of India received scientific attention rather late, and the pioneering works of Prashad, Pruthi, Ganapati, Chacko, Zafar, Krishnamury, and Sreenivasan, mostly on South Indian waters, are the milestones in the history of Indian limnology. The Republic of India rightly perceived the conclusions drawn by the International Environmental Conference (1972) and focused the attention of the entire scientific community on the non-availability of good-quality water in sufficient quantities and the problems of its management, realising that the fast-depleting water resources.

Material and Method

Study Area: - The village of Kapshi Lake is located in Akola Tahsil of Akola District in the State of Maharashtra, India. It comes under the Akola community development block. The nearest town is Akola, which is about 20 kilometres away from Kapshi Lake. The total area of the village is 836.67 hectares. Kapshi Lake is a village panchayat located in the Akola district of Maharashtra, India. The latitude 20.5876787 and longitude 76.960249 are the geocoordinates of Kapsi Lake.

Present studies were made on the physicochemical characteristics and features of Kapsi Lake Kapsi from December 2022 to November 2023. The physicochemical parameters were analysed using APHA (1998). In this study, monthly water samples were collected from eight selected stations by using a Ruttner water sampler at a specific depth of water**.**

Sample collection - Water samples from eight different stations were collected from the Kapsi Lake Reservoir region for a period of one year (December 2022–December 2023) for the evaluation of physicochemical parameters and the enumeration of microbial populations. Samples were collected during the first week of every month in sterile plastic containers and stored at 40 °C until analyzed. They were then tested for various physicochemical and microbiological parameters using standard methods.



Observations and results

The present study is made on Kapsi Lake Kapsi. Monthly samples were collected from eight selected stations of a lake. Physicochemical parameters were analysed from December 2022 to November 2023 (Table). Monthly variations were also observed during the present study. Station-wise variation was observed during the present study. The average minimum and maximum values of physicochemical characters were also determined; this value is depicted in Table 1**.**

Table 1: Physicochemical characteristic features of Kapsi Lakes Kapsi District Akola During December 2022 to November 2023

Physicochemical Parameter	Dec	Jan	Feb	Mar	Apr	May	June	July	Aug	Sept	Octo	Nov	Ave-rage	Mini	Maxi
Colour	Apparent	apparent	Seston	seston	apparent	apparent	apparent	sestoon	sestoon	sestoon	apparent	apparent	6.03	2.27	12.51
Odour	Petrichor	petrichor	Petrichor	petrichor	petrichor	petrichor	petrichor	earthy	earthy	earthy	petrichor	earthy	-	-	-
Turbidity (NTU)	4.64	4.10	3.89	4.10	2.25	2.25	12.55	11.05	9.02	9.16	4.46	5.25	6.42	2.25	12.55
Sechi disc Transparency (meter)	0.122	0.087	0.083	0.082	0.084	0.078	0.054	0.084	0.082	0.101	0.114	0.130	0.090	0.054	0.130
pH	9.34	8.53	8.28	8.33	8.83	8.77	8.28	8.25	8.25	8.33	8.23	8.38	8.51	8.23	9.34
Water Temp (°C)	28.1	28.75	28.92	29.0	29.13	24.34	13.00	13.25	13.66	13.80	14.00	14.10	21.33	13.00	29.13
Conductivity (u mhos/cm)	0.68	0.67	0.64	0.84	0.94	0.66	0.77	0.68	0.72	0.74	0.85	0.86	0.74	0.64	0.94
TDS (mg/lit)	0.547	0.891	0.584	0.555	0.845	0.609	0.611	0.547	0.891	0.584	0.555	0.845	0.672	0.547	0.891
Dissolved O2 (mg/lit)	5.6	6.4	6.9	5.9	6.2	6.24	5.84	5.6	6.4	6.9	5.9	6.2	6.04	5.6	6.9
Free CO2 (mg/lit)	9.6	11.4	7.6	4.4	4.4	7.8	9.6	9.6	10.8	6.0	2.4	4.4	7.55	2.4	11.4
Total Hardness (mg/lit)	256.0	450.0	230.0	240.0	230.0	315.0	272.6	256.0	450.0	230.0	240.0	230.0	281.22	230.0	450.0
Calcium Hardness (mg/lit)	132.3	157.5	136.7	147.0	157.0	158.9	141.7	132.3	157.5	136.7	147.0	157.0	148.08	132.3	158.9
Magnesium (mg/lit)	223.0	417.0	192.0	243.0	250.0	240.8	242	248	260	243	190	255	308.07	192.0	417.0
p. Alkalinity (mg/l)	0.2	0.1	0.1	0.1	0.2	0.2	0.1	0.2	0.2	0.1	0.2	0.1	0.15	0.1	9.2
T. Alkalinity (mg/l)	10.5	9.5	10	11.5	10.8	10	10	10.5	9.5	10	9.5	9.5	10.25	9.5	11.5
Chloride (mg/l)	130.94	130.96	130.96	130.99	130.92	130.92	130.94	130.91	130.94	130.91	130.94	130.96	130.96	130.91	130.99
Sulphate (mg/l)	0.63	0.59	0.60	0.47	0.60	0.47	0.60	0.64	0.51	0.63	0.59	0.60	0.52	0.04	0.63
Nitrate (mg/l)	0.040	0.050	0.020	0.032	0.050	0.056	0.056	0.060	0.058	0.052	0.044	0.036	0.045	0.020	0.060
Phosphate (mg/l)	0.091	0.091	0.092	0.083	0.083	0.087	0.088	0.092	0.083	0.091	0.083	0.087	0.882	0.83	0.92
Silicate (mg/l)	0.05	0.04	0.04	0.06	0.02	0.01	0.05	0.02	0.05	0.04	0.05	0.04	0.057	0.02	0.1

Discussion –

The temperature of water is basically important because it affects biochemical reactions in aquatic organisms. A rise in the temperature of water leads to the speeding up of chemical reactions in water, reduces the solubility of gases, and amplifies the taste and odour. The temperature of water, found in the range of 13.97 to 17.69, regulates most of the biological

processes and biochemical reactions. In a balanced ecosystem, pH is maintained within the range of 8.83 to 9.14 (Chandrasekhar et al., 2003). Due to diurnal variations in the water temperature of a system, the pH of a water body is a diurnally variable property (Ojha and Mandloi, 2004). In this study, pH values were found in the range of 8.83 to 9.14 in the water samples. All the locations of the sampling points are within the desirable limits. The oxygen content of a water body is important for the direct needs of many organisms. Further Oxygen is also known to affect the solubility and availability of many nutrients, and hence, it is one of the most significant parameters affecting the productivity of aquatic systems (Wetzel, 1983). The factors affecting oxygen content in natural waters include input from the atmosphere, photosynthesis, and output from respiration, decomposition, and mineralization of organic matter, as well as losses to the atmosphere. In this study, dissolved oxygen was found to be 15.91 to 18.14 mg/l.

Apart from this, the decomposition of organic matter and the respiration of aquatic plants and animals also contribute to the release of carbon dioxide. In this study, free cos is found in the range of 28.42 to 33.53 mg/l. High alkalinity is a function of ion exchange, in which calcium ions are replaced by sodium ions and later contribute to alkalinity (Sharma and John, 2009). Alkalinity is imperative for fish and aquatic life as it buffers against rapid changes in pH (Sheeja and Ebanasar, 2006).

Conclusion- Preliminary physicochemical parameters, viz., water temperature, pH, conductivity, Free CO2, dissolved oxygen, Total dissolved solids, phenolphthalein alkalinity, total alkalinity, total hardness, calcium hardness, magnesium content, sulphate, phosphate, nitrate, and silicate of Kapsi Lake were studied for twelve months, from December 2022 to November 2023. Samples were collected monthly from the eight selected stations of the Kapshi Lake.

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- Birge: One of the most prominent limnologists associated with this name is Chancey Juday, who collaborated with Edward A. Birge. Their work together includes numerous papers on the limnology of lakes, especially in the Wisconsin region of the United States.
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- Hutchinson "A Treatise on Limnology" John Wiley & Sons Publication 1957 Iran Journal of Environmental Biology, 29(5), Page 715-720.
- Juday: Chancey Juday is known for his collaboration with Edward A. Birge,
- Naumann: One prominent figure is Ernst Naumann, who made contributions to limnology, particularly in Germany in the late 19th and early 20th centuries. One of his

notable works is "Die BinnengewässerMitteleuropas" (The Inland Waters of Central Europe).

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The Survey of Road Kill Animals in and Around Karanja (Lad)

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Abstract:
The study area is located in Washim district. Karanja is located 64 km towards North from District Washim, 63 km to Amravati division. Total 61 road kills animals were recorded during December 2022- March 2023. Of these 13 kills were of amphibians. The reptiles accounted for 10 numbers and mammals 34 numbers. Bird kills were 4. The 61 kills belong to 13 species, which include 1 species of amphibians, 5 species of reptiles, 2 species of birds and 5 species of mammals. Of the species recorded as road kills, reptiles formed about 16%, mammals 56%, amphibians 22% and birds about 6%. The monthly distribution of animal kills observed during the period is given in fig, which indicates the peak in February 2023.

Introduction:-
In India, many highway roads bisect reserve forests and many protected areas. In recent times, the impact of highway construction on forest areas has been recognized. For this reason, the construction of new roads and the widening of the existing roads are opposed by the forest department and non-governmental organizations. These roads have been identified as the source of disturbance to animals, both directly and indirectly (Deepan et al.,njum et al., 2019). Animals are killed while crossing roads in different vehicles, e.g., bikes, trucks, motor vehicles, and buses. The reasons behind crossing the road are logical. Few of them crossed roads in search of grazing ground. Reptiles may habitually cross roads as they hunt, scatter from their natal sites, migrate among seasonal habitats, and move for mating purposes. fades are nocturnal, devoid of limbs, hence slow in locomotion; they become victims of road accidents. Domestic animals like pigs and dogs live in rural areas; their presence on the road is universal. Monsoon and winter are the seasons when more dogs are victimised compared to summer. (D. Solanke et al., 2017).
The majority of wildlife habitats are fragmented by linear structures; the occurrence of unwanted and dangerous encounters that happen on roads worldwide is common. Roadkill is among the most significant threats to wildlife and human safety. In particular, road structures that interfere with the behaviour of wildlife, such as road crossings, could be a critical factor in determining the spatial pattern of road kills. When animals are already on the road, other types of barriers can prevent them from crossing the road. For example, the median barriers reduce the permeability of roads to wildlife, and the barrier effect increases road kill risk.

Material & Method
Study Area:
Reserve forests and other protected areas are divided by several highways in India. The effect that highway building has on forested regions has come to light recently. For this reason, non-governmental organizations and the forest department oppose building new highways and extending existing ones. It has been determined that these roadways are the direct and indirect cause of animal disturbance (Deepan et al., 2019). When crossing highways in various vehicles, such as bikes, trucks, cars, and buses, animals are killed. There are reasonable justifications for crossing the street. Few of them went over roadways in quest of pasture. When hunting, migrating between seasonal habitats, or scattering from their natal places, reptiles may

frequently cross highways.

Map of Karanja (Lad)

Selection of study roads:-

Present study based on survey on road-kill animal. Road-kill animal survey was recorded in and around Karanja. The observation is specially made for the taxa amiphibians, reptiles , aves and mammals. From the period December 2022 to March of 2023, the stretch of state highway between Nagpur-Aurangabad and Yavatmal highway was driven four in month, totaling 120 days. Each sampling trip totaled 54.4 km. The route was driven during the day between 7.00 and 11.00 and during the evening between 16.00 and 18.00, the survey is alternatively conducted to acquire the finest result. On each slighting of road-kill ,location types of road and climatic condition are recorded. The dead animal were identified up to its species level, and removed from the road to avoid recounting and if unidentified. And we count vehicular traffic to study the pressureof vehicles on animals.

Observation & Result:

Common name	**Scientific name**	**Number of road kills**
Amphibians		
Common Indian Toad	*Duttaphrymus melanostictus*	13
Reptiles		
Common Garden Lizard	*Calotes versicolor*	7
Banded kukri snake	*Oligodon arnesis*	3
Mammals		
Common domestic cat	*Felis catus*	6
Indian local dog	*Canis lupus familiaris*	8
Domestic cow	*Bos taurus*	1
Domestic goat	*Capra hircus*	3
House mouse	*Mus musculus*	6
Pig	*Sus scrofa cristatus*	10
Birds		
Common chicken	*Gallus gallus domesticus*	2
Greater coucal	*Centropus sinensis*	2

Table 1. The details of animal kills observed during December 2022-March 2023 in & around KaranjaLad.A total of 61 road kills were recorded during December 2022–March 2023. Of these 13 kills, 13 were by amphibians. Reptiles accounted for 10 numbers, and mammals 34 numbers. Bird kills were 4. The 61 kills belong to 13 species, which include 1 species of amphibians, 5 species of reptiles, 2 species of birds, and 5 species of mammals. Of

the species recorded as road kills, reptiles formed about 16%, mammals 56%, amphibians 22%, and birds about 6%. The monthly distribution of animal kills observed during the period is given in Fig. 1, which indicates the peak in February 2023.

Among the amphibian kills recorded, the Common Indian Toad, Duttaphrynus melanostictus, had 13 numbers. The highest number of amphibian deaths on the road was recorded between December and January. Among the reptile kills recorded, the Common Garden Lizard Calotes versicolor had 7 numbers, and the Banded Kukri Snake Oligodon arnesis had 3 numbers. The highest number of reptile deaths on the road was recorded between February and March. Among the mammals killed recorded were the common domestic cat Felis catus (6 numbers), the Indian local dog Canis lupus familiaris (8 numbers), the domestic cow Bos taurus (1 number), the domestic goat capra hircus (3 numbers), the house mouse Mus musculus (6 numbers), and the pig Sus scrofa cristatus (10 numbers). The highest number of mammals killed on the road was recorded between February and March. Among the bird kills recorded, the Common Chicken Gallus gallus domesticus 2 number and the Greater Coucal Centropus sinensis 2 number. The highest number of bird deaths on the road was recorded between December and March.

Discussion:-

In the present study, a total record of 60 roadkill incidents is found, in which the most affected taxa is Mammals, Amphibian, Reptile and Aves. During the survey period, the most affected taxa were mammals. Among them, the present survey recorded that the common domestic pig was the most affected species by roadkill. During the study, mammals were affected more in number. Here, most of the mammalian road kills recorded in the present study are dog, cat, rat, and goat species that could have been killed while crossing the roads as they got blinded by the vehicle's headlights. The speed of the traffic, the size of the species, and its dispersal behaviour are also cited as important factors when assessing the barrier effect of a road. Wide roads with high traffic densities restrict animal movement most effectively.

Roads are dangerous to small mammals, reptiles, and amphibians that are affected by their habitat fragmentation. The ecological characteristics and the requirements of the species in question are important factors. Due to these factors, the probability of detecting the presence of species based on traffic reports varies among cells. Considering the topography and the regional distribution of human settlements, this methodology seems to be most reliable in two situations (S.J. Peris 2016). The actual rate of mortality per day and the seasonal variability of road kills could not work out. Research depicts the results of the road kill surveys during the study period and discusses the implications for wild-life management in protected areas and the increasing pressure of external development (A. Purohit 2019).

Conclusion:-

The importance of life is given equally to animals as well as to humans.

Humans have destroyed the living space of animals by damaging nature, including opening roads and developing cities to pursue the convenience of life. It is important to remember that an environment where animals cannot live will soon become an environment where humans cannot live. Now, humans should stop being selfish and try to create a world that can co-exist with animals.

The present study shows that highways have a severe impact on wildlife. This survey is a short-term study; it is recommended that long-term studies be conducted to suggest the various impacts highways have on animals. Earlier records indicate that pigs, snakes, frogs, cats, dogs, garden lizards, hens, cows, and rats have been killed by vehicular traffic. The impact of roads on animals may be attributed to anthropogenic activities and vehicular accidents. The need for speed breakers in wildlife habitats and improvised forest ranges can minimise the road killing of all types of animals.

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Birds of Kumbhari Dam Akola - A Checklist

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Abstract:
As significant bio-indicators of the environment, birds have ecological importance and are an essential part of the natural system. Additionally, they are crucial for the food chain, insect management, and seed dispersal. The current study was conducted to ascertain the diversity and richness of bird fauna at the Kumbhari Reservoir in Akola,Maharashtra State, India, because there is a dearth of information regarding avian diversity and its abundance in this reservoirs. Each of the five stations that were chosen had data collected from it. Data was collected using the point count method, with each site being visited four times per month during 2023.There were 12 order,47 families111 species observed. Of these, 15 species (representing 10 families) were connected with wetlands, and 8 species (representing 13 families) were specific to wetlands. The study focused on diversity indicators, species evenness, relative density, and abundance. It was also discovered that a large amount of species richness and bird diversity can be found in freshwater reservoirs.
Keywords: Bird Diversity, Kumbharilake , Akola

Introduction:

Birds are environmental indicators, a slight change in environmental conditions can impact their behavior pattern, population, reproduction and migration. (Harisha and Hosetti, 2009). Ecologically birds are very important creatures because they help in pollination and perform crucial role in seed dispersal (Bibi and Ali, 2013). Therefore, it is crucial to comprehend the diversity and structure of birds in order to describe the local landscape. Species are becoming extinct at an alarming rate, their conservation has arisen as one of the most significant issue today (Hu et al 2011). Kumbharireservoir inhabits several local and migratory bird. The biodiversity in the wetland is not studied in depth. Comparative research on the composition of avian communities in wetlands, habitats linked with wetlands, forests, grasslands, and even urban and suburban areas might help us understand the broad patterns and processes that define distinct bird species and communities. Birds that depend on wetland and wetland associated vegetation have experienced a greater decline than any other habitats, habitat loss and degradation of winter foraging and breeding ground were observed as leading causes of this decline. (Mankadan 2014, West, 2016, Johnson et al., 2019).

Materials and Methods

Observation of avifaunal diversity of Kumbhari Dam having coordinates 20.66923570044009, 77.07776784812778was carried out during year 2023 on the occasion of bird week, Wetlands Day and as apartof MOU made with forest department in collaboration with twoNGOs namely Nisargakatta Akola and Aadhar Foundation . Around 18 volunteers were present during the site visits. One of the member of each is equipped with e-Bird an android application developed by Cornell Lab of ornithology and checklist were submitted to database successfully. https://ebird.org/checklist/S97383123During the study, birds were ascertain by direct sighting and by their calls (for very few species). Bird survey in their active hours were carried out by adopting the line-transect method (Burnham et al.1980). Birds were observed with Nikon A211 10 X 50 binoculars, Comet, Zeiss.

Observations and Results:

There were 12 order,47 families 111 species observed. Of these, 15 species (representing 10 families) were connected with wetlands, and 8 species (representing 13 families) were specific

to wetlands. The study focused on diversity indicators, species evenness, relative density, and abundance. It was also discovered that a large amount of species richness and bird diversity can be found in freshwater reservoirs.

Sr. No.	Common Name	Scientific name	Family	Order	R/ M	Status
1	Bar Headed Goose	*Anserindicus*	Anatidae	Anseriformes	W	LC
2	Ruddy Shelduck	*Tadornaferruginea*	Anatidae	Anseriformes	W	LC
3	Comb Duck	*Sarkidiornissylviola*	Anatidae	Anseriformes	R	LC
4	Indian Spot-Billed Duck	*Anaspoecilorhyncha*	Anatidae	Anseriformes	R	LC
5	Green-Winged Teal	*Anascrecca*	Anatidae	Anseriformes	W	LC
6	Northern Pintail	*Anasacuta*	Anatidae	Anseriformes	W	LC
7	Northern Shoveler	*Spatula clypeata*	Anatidae	Anseriformes	W	LC
8	Common Pochard	*Aythyaferina*	Anatidae	Anseriformes	W	LC
9	Gadwall	*Marecastrepera*	Anatidae	Anseriformes	W	LC
10	Little Ring Plover	*Charadriusdubius*	Charadriidae	Charadriiformes	R	LC
11	Yellow-Wattled Lapwing	*Vanellusmalabaricus*	Charadriidae	Charadriiformes	R	LC
12	Red-Wattled Lapwing	*Vanellusindicus*	Charadriidae	Charadriiformes	R	LC
13	Black Tailed Godwit	*Limosalimosa*	Scolopacidae	Charadriiformes	W	NT
14	Common Snipe	*Gallinagogallinago*	Scolopacidae	Charadriiformes	W	LC
15	Common Sandpiper	*Actitishypoleucos*	Scolopacidae	Charadriiformes	W	LC
16	Common Red Shank	*Tringatotanus*	Scolopacidae	Charadriiformes	W	LC
17	Little Stint	*Calidrisminuta*	Scolopacidae	Charadriiformes	W	LC
18	Black Winged Stilt	*Himantopushimantopus*	Recurvirostridae	Charadriiformes	R	LC
19	Small Pratincole	*Glareolalactea*	Glareolidae	Charadriiformes	R	LC
20	River Tern	*Stemaaurantia*	Laridae	Charadriiformes	R	NT (U)
21	Wood Sandpiper	*Tringaglareola*	Scolopacidae	Charadriiformes	W	LC
22	Barred Buttonquail	*Turnixsuscitator*	Turnicidae	Charadriiformes	R	LC
23	Black Headed Gull	*Chroicocephalusridibundus*	Laridae	Charadriiformes	W	LC
24	Little Egret	*Egrettagarzetta*	Ardeidae	Ciconiiformes	R	LC
25	Cattle Egret	*Bubulcus ibis*	Ardeidae	Ciconiiformes	R	LC
26	Gray Heron	*Ardeacinerea*	Ardeidae	Ciconiiformes	W	LC
27	Purple Heron	*Ardeapurpurea*	Ardeidae	Ciconiiformes	R	LC
28	Indian Pond Heron	*Ardeolagrayii*	Ardeidae	Ciconiiformes	R	LC
29	Asian Open Bill	*Anastomusoscitans*	Ciconiidae	Ciconiiformes	R	LC
30	Asian Wooly Necked Stork	*Ciconiaepiscopus*	Ciconiidae	Ciconiiformes	R	LC
31	Black Headed Ibis	*Threskiornismelanocephalus*	Threskiornithidae	Ciconiiformes	R	NT
32	Red-Naped Ibis	*Pseudibispapillosa*	Threskiornithidae	Ciconiiformes	R	LC

33	Eurasian Spoonbill	*Platalealeucorodia*	Threskiornithidae	Ciconiiformes	R	LC
34	Great Egret	*Ardea alba*	Ardeidae	Ciconiiformes	R	LC
35	Intermediate Egret	*Ardeaintermedia*	Ardeidae	Ciconiiformes	R	LC
36	Rock Pigeon	*Columba livia*	Columbidae	Columbiformes	R	LC
37	Eurasian Collared Dove	*Streptopeliadecaocto*	Columbidae	Columbiformes	R	LC
38	Spotted Dove	*Spilopeliachinensis*	Columbidae	Columbiformes	R	LC
39	Laughing Dove	*Spilopeliasenegalensis*	Columbidae	Columbiformes	R	LC
40	White-Throated Kingfisher	*Halcyon smyrnensis*	Alcedinidae	Coraciiformes	R	LC
41	Pied Kingfisher	*Cerylerudis*	Alcedinidae	Coraciiformes	R	LC
42	Asian Green Bee-Eater	*Meropsorientalis*	Meropidae	Coraciiformes	R	LC
43	Indian Roller	*Coraciasbenghalensis*	Coraciidae	Coraciiformes	R	LC
44	Eurasian Hoopoe	*Upupaepops*	Upupidae	Coraciiformes	R	LC
45	Indian Gray Hornbill	*Ocycerosbirostris*	Bucerotidae	Coraciiformes	R	LC
46	Greater Coucal	*Centropussinensis*	Cuculidae	Cuculiformes	R	LC
47	Asian Koel	*Eudynamysscolopacea*	Cuculidae	Cuculiformes	R	LC
48	Common Hawk Cuckoo	*Hierococcyxvarius*	Cuculidae	Cuculiformes	M	LC
49	Black Kite	*Milvusmigrans*	Accipitridae	Falconiformes	R	LC
50	Shikra	*Accipiter badius*	Accipitridae	Falconiformes	R	LC
51	Gray headed Swamphen	*Porphyrioporphyrio*	Rallidae	Gruiformes	R	LC
52	Eurasian Coot	*Fulicaatra*	Rallidae	Gruiformes	R	LC
53	Eurasian Moorhen	*Gallinulachloropus*	Rallidae	Gruiformes	R	LC
54	Crested Lark	*Galeridacristata*	Alaudidae	Passeriformes	R	LC
55	Wire Tailed Swallow	*Hirundosmithii*	Hirundinidae	Passeriformes	R	LC
56	Red Rumped Swallow	*Cecropiisdaurica*	Hirundinidae	Passeriformes	R	LC
57	Grey Wagtail	*Motacillacinerea*	Motacillidae	Passeriformes	W	LC
58	Small Minivet	*Pericrocotuscinnamomeus*	Campephagidae	Passeriformes	R	LC
59	Red Vented Bulbul	*Pycnonotuscafer*	Pycnonotidae	Passeriformes	R	LC
60	Common Iora	*Aegithinatiphia*	Irenidae	Passeriformes	R	LC
61	Bay Backed Shrike	*Laniusvittatus*	Laniidae	Passeriformes	R	LC
62	Rufous-Backed Shrike	*Laniusschach*	Laniidae	Passeriformes	R	C
63	Black Drongo	*Dicrurusmacrocercus*	Dicruridae	Passeriformes	R	LC
64	White Bellied Drongo	*Dicruruscaerulescens*	Dicruridae	Passeriformes	R	LC
65	RufousTreepie	*Dendrocittavagabunda*	Corvidae	Passeriformes	R	LC
66	House Crow	*Corvussplendens*	Corvidae	Passeriformes	R	LC
67	Eurasian Golden Oriole	*Oriolusoriolus*	Oriolidae	Passeriformes	R	LC
68	Brahminy Starling	*Sturnuspagodarum*	Sturnidae	Passeriformes	R	LC
69	Rosy Starling	*Sturnusroseus*	Sturnidae	Passeriformes	R	LC
70	Common Myna	*Acridotherestristis*	Sturnidae	Passeriformes	R	LC
71	Common Babbler	*Argyacaudata*	Leiothrichidae	Passeriformes	R	LC
72	Common Tailorbird	*Orthotomussutorius*	Muscicapidae	Passeriformes	R	LC
73	Ashy Prinia	*Priniasocialis*	Muscicapidae	Passeriformes	R	LC
74	Great Tit	*Parus major*	Paridae	Passeriformes	R	LC
75	Purple Sunbird	*Cinnyrisasiatica*	Nectarinidae	Passeriformes	R	LC
76	Paddy Field Pipit	*Anthusrufulus*	Estrildidae	Passeriformes	R	LC
77	House Sparrow	*Passer domesticus*	Passeridae	Passeriformes	R	LC
78	Baya Weaver	*Ploceusphilippinus*	Passeridae	Passeriformes	R	LC

79	White - Bellied Minivet	*Pericrocotuserythropygius*	Campephagidae	Passeriformes	R	LC
80	Common Woodshrike	*Tephrodornispondicerianus*	Vangidae	Passeriformes	R	LC
81	Brown Shrike	*Laniuscristatus*	Laniidae	Passeriformes	W	LC
82	Indian Bushlark	*Mirafraerythroptera*	Alaudidae	Passeriformes	R	LC
83	Plain Prinia	*Priniainornata*	Cisticolidae	Passeriformes	R	LC
84	Gray-Breasted Prinia	*Priniahodgsonii*	Cisticolidae	Passeriformes	R	LC
85	ZittingCisticola	*Cisticolajuncidis*	Cisticolidae	Passeriformes	R	LC
86	Booted Warbler	*Idunacaligata*	Acrocephalidae	Passeriformes	W	LC
87	Blyth's Reed Warbler	*Acrocephalusdumetorum*	Acrocephalidae	Passeriformes	W	LC
88	Barn Swallow	*Hirundorustica*	Hirundinidae	Passeriformes	W	LC
89	Wire-Tailed Swallow	*Hirundosmithii*	Hirundinidae	Passeriformes	R	LC
90	Red-Rumped Swallow	*Cecropisdaurica*	Hirundinidae	Passeriformes	R	LC
91	Streak-Throated Swallow	*Petrochelidonfluvicola*	Hirundinidae	Passeriformes	R	LC
92	Sulphur-Bellied Warbler	*Phylloscopusgriseolus*	Phylloscopidae	Passeriformes	W	LC
93	Greenish Warbler	*Phylloscopustrochiloides*	Phylloscopidae	Passeriformes	W	LC
94	Lesser Whitethroat	*Currucacurruca*	Sylviidae	Passeriformes	W	LC
95	Jungle Babbler	*Turdoidesstriata*	Leothrichidae	Passeriformes	R	LC
96	Large Grey Babbler	*Turdoidesmalcolmi*	Leothrichidae	Passeriformes	R	LC
97	Tickell's Blue Flycatcher	*Cyornistickelliae*	Muscicapidae	Passeriformes	R	LC
98	Siberian Stonechat	*Saxicolamaurus*	Muscicapidae	Passeriformes	R	LC
99	Pied Bushchat	*Saxicolacaprata*	Muscicapidae	Passeriformes	R	LC
100	Yellow-Throated Sparrow	*Gymnorisxanthocollis*	Passiridae	Passeriformes	R	LC
101	Tawny Pipit	*Anthuscampestris*	Estrildidae	Passeriformes	R	LC
102	Little Cormorant	*Microcarboniger*	Phalacrocoracidae	Pelecaniformes	R	LC
103	Great Cormorant	*Phalacrocoraxcarbo*	Phalacrocoracidae	Pelecaniformes	R	LC
104	Oriental Darter	*Anhinga melanogaster*	Anhingidae	Pelecaniformes	R	NT
105	Indian Cormorant	*Phalacrocoraxfuscicollis*	Phalacrocoracidae	Pelecaniformes	W	LC
106	Coppersmith Barbet	*Psilopogonhaemacephalus*	Megalaimidae	Piciformes	R	LC
107	Little Grebe	*Tachybaptusruficollis*	Podicipedidae	Podicipediformes	R	LC
108	Rose Ring Parkeet	*Psittaculakrameri*	Psittacidae	Psittaciformes	R	LC
109	Plum Headed Parkeet	*Psittaculacyanocephala*	Psittacidae	Psittaciformes	R	LC
110	Alexandrine Parkeet	*Psittaculaeupatria*	Psittacidae	Psittaciformes	R	NT (U)
111	Spotted Owlet	*Athenebrama*	Strigidae	Strigiformes	R	LC

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Study of web pattern in spiders of Akola region Maharashtra

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Abstract:-
Spiders secrete silk for many purposes: to protect their young ones, catch food, make houses, and move around. Spider silk is an elastic and sometimes adhesive material. Silk is secreted as a fluid, which hardens as it oozes out of the silk spinning organs or spinnerets, which are mobile finger-like projections. Webs (a characteristic feature of spiders) built out of silk are used to catch insects, making web-building spiders efficient predators and even biological control agents. It is concluded that variability in web pattern is related to different families of web-building spiders. These observations illustrate how web patterns enable the identification of otherwise taxonomically ambigious specimens, such as juveniles or whatever the condition of the specimens may be.
Keyword: Spider, Web, Juvenile, Silk.

Introduction:-

Spiders secrete silk for many purposes: to protect their young ones, catch food, make houses, and move around. Spider silk is an elastic and sometimes adhesive material. Silk is secreted as a fluid, which hardens as it oozes out of the silk spinning organs or spinnerets, which are mobile finger-like projections. Webs (a characteristic feature of spiders) built out of silk are used to catch insects, making web-building spiders efficient predators and even biological control agents (Riechert, 1999; Symondson et al., 2002). In a typical field survey, the majority of spiders collected cannot be identified by species level because they are juveniles (Brennan et al., 2004). Web-building spider communities can be assessed and identified on the basis of the characteristics of their web architecture (Gollan et al., 2009). The evolution of the web itself has been extensively studied (Benjamin and Zschokke, 2004; Gan et al., 2015). A number of efforts have been made to trace the history and relationship of species by means of web resemblances, under the assumption that a more "primitive," simple, and irregular web was the forerunner of the more elaborate pattern.

Material and methods:-

Web building is very sensitive to disturbance, especially during the early stages of web building (Zschokke, 1996); because of this, we selected particularly non-disturbing sites in the experimental region to study and observe the webs. A web study was undertaken in Patan (Sikar, Rajasthan) and caves and rocky areas around the Katali River in Khandella (Sikar, Rajasthan). Since spider web-threads are very thin (0.5 to 5 µm), taking pictures of webs requires the clear visibility of threads. To increase the thread visibility, we used a highly resolving camera (Nikon). The record of every photograph was maintained in a laboratory notebook.

The main requirements to capture highly clear photographs of spider webs are bright light from the sides and a very dark background. In this order, we applied a black sheet of rigid paper as a dark background, putting it just behind the web threads. During web observations, a few egg sacs and spiderlings were also found entangled in the webs of some spider species. These egg sacs and spiderlings were also collected in plastic jars separately and were subjected to being reared in a laboratory to study the biology and feeding efficacy of these species. The webs observed in the study area were examined, identified, and discussed on the basis of matching the architecture and photographs of webs with the descriptions of webs given by Witt

et al. (1968) and Sebestian and Peter (2009).

Results and Discussion:-

Orb- webs :

The characteristic feature of an orb web is that the central portion consists of a series of radiating lines of dry and inelastic silk that support a thread of viscid and elastic silk. Orb webs vary in structure, shape, and size according to the families and genera of the spiders. In the more symmetrical types of orb webs, the viscid lines extend throughout the majority of their length as a spiral line. The webs of the families Araneidae, Tetragnathidae, and Uloboridae were found to be good examples of orb webs (Fig. 1).

Fig. 1 : Orb-webs

Sheet web :

this type of web, the principal part of the web consists of a closely woven sheet extended in a single plane, but the threads are extended in all directions. Spiders of the families Linyphidae and Pholcidae construct this type of web (Fig. 2).

Fig.2 Sheet Web

(Zig-Zag)-web :

Popularly known as the “signature spider." This spider shows a unique decoration of stabilimentum, which is called a signature. Argiope changes its stabiliser shape from zigzag

to round. The species that was collected changes its stabilimentum design after 2–3 days (Fig. 3).

Fig. 3 Zig –Zag Web

Funnel webs :

The common grass spiders of the genera Agelena (Agelenidae) and Hippasa (Lycosidae) were found building this type of web. The principal part of a funnel web is sheet-like in structure, but webs of this type differ from true sheet-like webs in having a tube extending from one edge. This tube leads to the retreat of the spiders. Usually, a very loose, irregular net is spun above the sheet of a funnel web that obstructs the flight of insects and causes them to fall on the sheet, where the spider charging from its retreat can capture them (Fig. 4).

Fig4. Funnel Web

The triangular webs :

This type of web, shaped like a triangle, had been observed in some members of the family Uloboridae (Fig. 5a and b).

(a)
(b)

Irregular web: This type of web thread extends in all directions as an irregular shape.

Most members of the family Theridiidae were found spinning irregular webs (Fig. 6).

Fig. 6 irregular web

The single line thread web :

The web of *Uloborus* sp. (Uloboridae) was found as a single horizontal line, generally attached at both ends to branches that stretch about four feet across open spaces in the forest. These spiders have developed a marvellous trapping device for catching prey with the help of a single-line web (Fig. 7).

Fig. 7 The single line thread web

Conclusion:- It is concluded that variability in web pattern is related to different families of web-building spiders. These observations illustrate how the web pattern enables the identification of otherwise taxonomically ambigious specimens, such as juveniles, or whatever the condition of the specimen may be.

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Spider Diversity in Amravati District, Maharashtra: A Comprehensive Review

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Abstract:
This review offers a comprehensive exploration of spider diversity in Amravati District, Maharashtra, emphasising ecological intricacies, behavioural patterns, and existing threats. Through a meticulous synthesis of field studies and scholarly works, we categorise spider species based on families, genera, and species prevalent in the region. The ecological discourse unveils preferred habitats, seasonal variations, and interactions, with particular attention to spiders' vital role in pest control and ecosystem services.
Delving into behavioural studies, the paper synthesises existing knowledge on hunting strategies, mating behaviours, and web-building patterns, showcasing the adaptability of spiders to local environmental conditions. Despite substantial progress, research gaps persist, necessitating detailed taxonomic studies, long-term monitoring projects, and the incorporation of molecular techniques. Future directions advocate for citizen science involvement and the utilisation of advanced technologies, offering a roadmap for a holistic approach to spider conservation in Amravati.
Keywords: spider diversity, Maharashtra, Amravati District, ecology, behaviour, threats, conservation, research gaps

Introduction:
Spiders, as integral components of ecosystems, hold a pivotal role in sustaining ecological equilibrium and preserving biodiversity. Their intricate interactions within the web of life contribute significantly to the natural balance. As such, comprehending the diversity of spiders in specific geographic regions becomes imperative for devising effective conservation strategies. This review focuses on elucidating the multifaceted aspects of spider diversity in Amravati District, Maharashtra. By delving into the ecological intricacies, behavioural patterns, potential threats, and conservation needs, this paper seeks to provide a comprehensive and nuanced understanding of the arachnid community in this particular locale.
Amravati District, situated in the heart of Maharashtra, represents a unique ecological niche where spiders play an often understated yet critical role. The intricacies of spider diversity go beyond mere species enumeration, encompassing their ecological functions, behavioural adaptations, and the challenges they face in their habitat. Through a thorough exploration of these dimensions, this review aims to shed light on the ecological tapestry woven by spiders in Amravati District. Furthermore, understanding their behavioural nuances and the threats posed to their existence is vital for formulating conservation measures that are not only region-specific but also aligned with broader biodiversity preservation goals.
Against the backdrop of increasing anthropogenic pressures and environmental changes, Amravati District stands as a microcosm that demands meticulous scrutiny. This review endeavours to consolidate existing knowledge, drawing from both field studies and scholarly works, to present a cohesive narrative on spider diversity in the region. By highlighting the interconnectedness of ecological, behavioural, and conservation aspects, the ensuing sections of this paper will unfold a comprehensive overview. As we embark on this exploration, it

becomes evident that unravelling the intricacies of spider diversity in Amravati District is not just a scientific pursuit but a crucial step towards fostering sustainable coexistence between human activities and the delicate arachnid ecosystems.

Spider Diversity in Amravati District, Maharashtra:

This segment of the review delves into a comprehensive exploration of the spider kingdom within Amravati District, Maharashtra. The region proves to be a captivating canvas, adorned with a rich tapestry of arachnid fauna. Drawing upon a synthesis of data derived from meticulous field studies and extensive literature reviews, our investigation seeks to categorise and showcase the diverse array of spider species thriving in this locale.

Amravati District, situated amidst the diverse landscapes of Maharashtra, hosts a myriad of spider families, genera, and species. Through systematic analysis, this review brings forth a nuanced understanding of the intricate relationships and ecological roles played by various spider taxa. By categorising them based on their taxonomic classifications, including families and genera, we aim to provide readers with a vivid picture of the biodiversity that graces the district. This categorization, built upon a foundation of both field observations and scholarly insights, contributes to a comprehensive repository of knowledge regarding the spiders inhabiting Amravati.

In synthesising the available data, we not only highlight the sheer variety of spiders but also strive to uncover patterns in their distribution and abundance. This section sets the stage for subsequent discussions on the ecological nuances, behavioural characteristics, and conservation considerations of the diverse spider community in Amravati District. As we unravel the taxonomic intricacies, a deeper appreciation emerges for the intricate web of life woven by these fascinating arachnids within the distinct ecological milieu of Amravati.

Ecology of Spiders in Maharashtra:

This section ventures into a thorough examination of the ecological dimensions that govern the lives of spiders across the diverse landscapes of Maharashtra. By unravelling their preferred habitats, seasonal dynamics, and intricate interactions within the local ecosystems, we aim to paint a holistic picture of the ecological tapestry that spiders contribute to in this region.

Maharashtra, with its varied topography and ecosystems, serves as a dynamic stage where spiders actively participate in shaping ecological dynamics. Through an exploration of their preferred habitats, ranging from lush forests to urban environments, we seek to discern the factors influencing their distribution and abundance. Insights into the seasonal variations in spider populations further enrich our understanding, shedding light on the adaptability of these arachnids to the ever-changing environmental conditions prevalent in Maharashtra.

This segment also delves into the intricate web of interactions spiders engage in with other species within their habitat. From intricate predator-prey relationships to potential symbiotic alliances, we aim to uncover the interconnectedness that defines the ecological roles of spiders in Maharashtra. A particular emphasis is placed on their role in pest control, underscoring their contribution to maintaining a delicate balance within the ecosystem. As natural predators, spiders play a vital role in curbing insect populations, thereby contributing to the overall health and stability of Maharashtra's ecosystems. Through this exploration of ecological intricacies, we lay the groundwork for a deeper appreciation of the indispensable role spiders play in sustaining the biodiversity and ecosystem services of Maharashtra.

Behavioural Studies:

In this section, we embark on a comprehensive exploration of the behavioural intricacies that define the lives of spiders inhabiting the diverse landscapes of Amravati District. By synthesising existing knowledge and observations, we delve into the fascinating realms of their hunting strategies, mating behaviours, and web-building patterns. Additionally, a keen focus is placed on unraveling the adaptability exhibited by spiders in response to the specific environmental conditions prevailing in Amravati.

The hunting strategies employed by spiders reveal a complex interplay of instinct and adaptation. From ambush tactics to intricate web-building techniques, this section aims to dissect the various approaches spiders utilise to secure their prey. By drawing upon field studies and documented behaviours, we illuminate the nuanced intricacies of their predatory mechanisms, shedding light on the diversity that characterises spider hunting strategies within Amravati District.

Mating behaviors represent a crucial aspect of spider life cycles, often marked by elaborate courtship rituals and mating displays. Through a synthesis of existing research, this section provides insights into the diverse mating behaviours exhibited by spider populations in Amravati. The examination encompasses the spectrum of reproductive strategies, shedding light on the adaptations that enhance their reproductive success within this specific geographical context.

Web-building patterns stand as an iconic representation of spider behavior. This section explores the diverse architectures woven by spiders in Amravati District, emphasising the species-specific intricacies that define their silk-based creations. The discussion encompasses variations in web designs, functions, and the adaptability of spiders to the local environmental conditions that influence their weaving patterns.

Moreover, a closer look at the adaptability of spiders within Amravati District unravels the mechanisms through which these arachnids navigate and thrive in their specific habitats. Insights into their behavioral plasticity, response to environmental cues, and ability to adjust hunting and mating strategies in varying conditions contribute to a nuanced understanding of the behavioral repertoire of spiders within this distinct locale. As we navigate through these behavioural studies, a vivid portrait emerges, portraying the dynamic and intricate lives of spiders in Amravati District.

Research Gaps and Future Directions:

While considerable progress has been made in understanding the intricacies of spider populations in Amravati District, this section highlights persistent research gaps that warrant focused attention. Identifying these gaps is crucial for steering future investigations toward a more comprehensive understanding of the arachnid community in this specific locale.

One notable research gap pertains to the need for more detailed taxonomic studies. Despite strides in cataloging spider species, a finer resolution is required to discern subtle variations within taxa. Enhanced taxonomic clarity would not only contribute to our understanding of species diversity but also aid in more targeted conservation efforts. This section advocates for investments in systematic, in-depth taxonomic studies to unravel the finer nuances of the spider fauna in Amravati.

Long-term monitoring projects emerge as another essential avenue for future research. Sustained observation over extended periods allows for the detection of temporal trends, fluctuations in population dynamics, and responses to environmental changes. Establishing such projects in Amravati District would provide invaluable insights into the long-term resilience and adaptation of spider communities, enabling more informed conservation strategies.

Incorporating molecular techniques into spider research represents a frontier that holds immense potential. Genetic analyses can unravel hidden diversity, identify cryptic species, and illuminate evolutionary patterns within spider populations. This section advocates for the integration of molecular methodologies to complement traditional morphological studies, offering a more holistic understanding of the genetic diversity and evolutionary dynamics of spiders in Amravati.

As we envision the future of spider research in Amravati, proposals for research directions emerge. The integration of citizen science stands out as a collaborative approach, involving the local community in data collection and monitoring efforts. Engaging citizens not only broadens

the scope of research but also fosters a sense of environmental stewardship. Moreover, the adoption of advanced technologies, such as remote sensing and DNA barcoding, holds promise for efficient data collection and species identification.

In conclusion, addressing these research gaps and pursuing the proposed future directions will not only enrich our understanding of spider ecology in Amravati District but also contribute to broader advancements in arachnological research. The collaboration of researchers, citizens, and technological innovations is poised to shape the trajectory of spider studies, paving the way for more informed conservation practices and sustainable coexistence with these fascinating arachnids.

Conclusion:

In summary, this comprehensive review illuminates the intricate world of spider diversity in Amravati District, Maharashtra. Through a meticulous exploration of ecological intricacies, behavioural patterns, and prevailing threats, the significance of continuous research and dedicated conservation efforts becomes evident. The synthesis of knowledge presented herein underscores the need for a holistic approach to spider conservation in this distinct region.

The multifaceted nature of spider ecology in Amravati, as unveiled in this review, accentuates the interconnectedness of various components within the arachnid community. The interplay of ecological nuances and behavioural adaptations not only enriches our understanding of local biodiversity but also underscores the delicate balance that spiders contribute to the broader ecosystem.

Crucially, the identification of existing research gaps serves as a call to action for the scientific community. Bridging these gaps, including the need for detailed taxonomic studies, long-term monitoring projects, and the integration of molecular techniques, is paramount for fostering a more nuanced comprehension of spider diversity. The proposed future directions, encompassing citizen science and advanced technologies, present a roadmap for sustainable research initiatives that align with conservation goals.

As we navigate the intricate web of spider diversity in Amravati, the importance of a collective effort to safeguard these vital components of the ecosystem becomes clear. Ongoing research endeavours, coupled with strategic conservation measures informed by the insights presented in this review, hold the key to preserving the rich tapestry of spider life in Amravati District. In essence, this review not only encapsulates the current state of knowledge but also advocates for a dynamic and collaborative approach towards fostering the coexistence of humans and spiders in this unique ecological setting.

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Seed Production of Major Carps and hatchery management in Akola district.

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Abstract:
Fish are an important source of protein, vitamins, and omega-3 fatty acids. They are also filled with important nutrients that keep our hearts and brains healthy, so they are considered the healthiest food on the planet. Fish produce a number of products used in medicine and other products, contributing to economic growth and nutritious food.
Carps do not breed in limited water due to natural environmental factors. For the increasing demand for carp and good-quality fish seed, it is necessary to breed in confined water through induced breeding. The development of fish farming depends on the quality of fish seed. The present study is based on the current status of fish seed production using new-generation drugs.
Keywords: Hatchery, Induced breeding, Seed production, Major carps.

Introduction:
Taking into consideration the importance of fresh water fish farming, the Department of Assistance Commissioner of Fisheries is taking efforts to produce fish seed and encourage fresh water fish farming in Akola district through the induced breeding method by creating artificial conditions such as running water, continuous showering, etc. required for the breeding of fish. Also, using various new generation drugs available in the market that are substituted for pituitary extract, these agents are needed for fruitful reproduction of major carp.

Material and Methods:
During the present study, the Mahan-Fish Seed Production Centre has been undertaken in the academic year 2023–24 for the study of seed production of major carps and hatchery management in Akola district.
Fish Seed Production Centre at Mahan, District Akola, Government of Maharashtra, was established in 1977–1978. The farm was established for the seed production of major carps and common carps, which are not breed in confined water. Good-quality fish seed of these species, such as Rohu, Catla, Mrigal, and Cyprinus, is distributed throughout Akola district to the fish farmer.

Result and Discussion:
Now a days in artificially breeding techniques, various types of synthetic drugs have been introduced as an alternative to the pituitary extract, Ovaprim, Ovatide Gonopro etc. and obtain good results. Earlier workers 1-15 performed experiments on induced breeding by using various inducing agents on Indian major carps and non-cyprinoid fishes in chine circular hatchery.

Chinese Circular hatchery for Induced breeding of Major Carps
In Akola district, Maharashtra, a Chinese circular hatchery is located near the Mahan reservoir, which is the largest in Akola district and highly satisfactory for the huge fish diversity and remarkable quality seed of the Indian major carps. One operation of hatching required four days. Chinese circular hatchery for successful breeding of fish and incubation require constant water supply with gravity to reproduce carp.

Components of Chinese Circular Hatchery:

1) **Overhead Tank**

The Mahan fish seed production centre depends on the Mahan reservoir pipeline and filtered chambers arranged to supply water to the overhead tank. The floor of the tank is 20 feet above ground level, with a capacity of 50,000 litres to supply adequate water for the breeding and spawning tank of the Chinese circular hatchery.

2) **Breeding /spawning tank**

A breeding pond is circular shape made of concrete with 6 to 12 m. and depth 1.0 to 2.0m. The base of breeding tank is sloped towards the centre, there is outlet. Periphery of the pond fixed to the nozzle mouth flush at the bottom so as to provide water current and false riverine environment, continuous showering maintain water temperature and increase the dissolved oxygen which results satisfactory spawning and hatching. For the induced breeding of carps, clean the breeding tank, fill up it with clean water from overhead tank and released injected brooders for spawning. After breeding, remove the brooders and transfer them to the previously prepared post-spawning tank. Collect the fertilised eggs and transfer them into the incubation tank. This pond requires a 0.2–03 m/sec water current.

3) **Incubation /Hatching tank:**

Hatching tanks are circular water flow systems from overhead tanks; the water current is about 0.2–0.3 m/sec. constructed with cement with a 3.6-metre internal diametre and 70,000 million eggs holding capacity provided with aeration. There are two chambers in the incubation pond, each with a depth of one metre. The innermost chamber is provided with a 10cm diametre to the upright outlets with holes at distinct heights for releasing a lot of water. Another circular wall via a fixed nylon partition is supplied at 0.76 m from the outer wall. Unidirectional outlets locked with 7.5-cm-diameter spawns together with water movement from these tanks to the hatch collection chamber, lower level, to the hatching pond where spawns can be collected

Incubation /Hatching tank

4)Hatchling receiving pond:

Is a rectangular pond constructed with cement water supply from the overhead tank and placed beneath the incubation pond so as to pump out water from it downward force. Spawn was taken away from incubation ponds with 7.5-cm diameter pipes. For the collection of spawn, two opposite sides of the pond walls are connected to the net with the help of hooks.

Hatchlings

Functioning of the Chinese Circular hatchery:

Fully matured fish are stored in the breeding pond for around 4–5 hours for acclimatisation. Both sexes are injected with new-generation drugs in single doses (Ovaprim, Ovatide, etc.), depending upon the weight of the fish. After the injection generates water current, a shower jet is started to create a natural environmental condition. After 5 to 8 hours, breeding takes place. The fertilised eggs are collected from the bottom of the breeding pond and shifted into the incubation ponds. During the breeding season, from July to August, hatching operations are carried out. It's easy to operate with less effort and manpower.

This year, 2023–24, the Mahan fish seed production centre produced 322 lakh spawns.

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Seasonal Prevalence of Gastrointestinal *Trichuris species* in Sheep and Goat of Amravati District, Maharashtra , India.

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Abstract:
Trichuris Ovis commonly called as whipworm of sheep and goat found in caecum and colon region of large intestine. A Study on seasonal prevalence of gastrointestinal Trichuris species in Sheep and Goats were carried during 2022 to 2023. Total 720 fecal samples were collected from various Talukas of Amravati district of Maharashtra. The highest prevalence of Trichuris species were recorded in the winter i.e. 63.39%, whereas the prevalence was lowest in the summer season i.e.32.75%. The median range of prevalence was noted in the monsoon i.e. 44.69%. In the above study it was observed that the prevalence of Trichuris species were recorded highest in the winter season and lowest in the summer season. Age and sex wise Trichuris species infection was more prevalent in female hosts (43.75%), followed by male hosts (29.16% likewise, there were no more differences; they were about similar, i.e., in kids (41.66%, young (41.96%), and adults (41.69%), because of a lack of cleanliness, poor management practices, and less awareness about deworming.
Keywords: seasonal, Trichuris ovis, gastrointestinal, parasite, sheep, goat.
Introduction: The goat acts as a multi-purpose animal that plays an important role in the economy and nutrition of landless and marginal farmers. The estimated population of sheep and goats in India has been 47.26 million and 148.88 million, respectively, whereas the North-Western Himalayan state of Himachal Pradesh has 0.79 million sheep and 1.1 million goats as per the 20th livestock census (20th livestock census, 2019). India's livestock sector is one of the highest in the world, accounting for 26.40% of goats, which play an important role in the Indian economy (Anon, 2012). Trichuris is a widespread gastrointestinal parasite that can occur in a broad range of hosts. Its life cycle is direct, where orally ingested embryonated eggs hatch in the small intestine and discharge larvae shelter inside the intestinal wall of the caecum and colon, where they develop into mature worms (Jenkins, 1970; Beer, 1973). Gastrointestinal parasites cause economic losses in different ways, like lower fertility, reduced work capacity, slow food intake capacity, and slow weight gain treatment and cost in massively parasitized animals. The nematode parasite causes the host-parasite relationship, which results in large-scale damage at the site of attachment and consequently economic loss (Padwal et al., 2011). Trichuriasis is found in small ruminants, i.e., sheep and goats, caused by trichuris ovis. Gastrointestinal parasitic diseases are the main issue that affects the productivity of the goat industry in India and worldwide (Pathak and Pal 2008). Seasonal prevalence of species is done to find out the time at which infection with larvae starts, rises too high and low, and so treatment can be timed to avoid the development of massive infection. Age-wise prevalence has been most important to find out which age group is more susceptible to Trichuris species and which is less susceptible. In Trichuris specie, sexual dimorphism occurs in males 50–80 mm in length, of which a narrow and filamentous anterior end constitutes three quarters of length (Soulsby 1982), females are 37–70 mm long, the anterior end is narrow, and filamentous forms two-thirds to four-fifths (Urquhart GM). The spicule is fully evaginated, 5–6 mm long, with a sheath that bears a swelling a short distance from its distal side, which is covered by a spine that is smaller in size towards the distal side. Trichuris eggs are brown, barrel-shaped, or lemon-shaped at both ends; transparent, conspicuous ends are present and have a length of 70–80 by

30-42 cm with a plug inside the unsegmented embryo when laid. High worm burdens lead to severe anaemia and dehydration, and jaundice may lead to the death of the animal (Soulsby, 1982; Bowman, 2002; Taylor et l., 2007). The eggs of Trichuris species were identified on the basis of morphological characters (Soulsby Helminths, Arthropods, and Protozoa of Domesticated Animals, CLBS &Bailliere Tinda, London, 1982).

Material and Method:

Study area: The study was conducted at different talukas in Amravati District, Maharashtra, India, from December 2022 to December 2023. The study was done on various breeds of sheep and goats. The sheep breeds Deccani and Madgyal, and the goat breeds Beetal, Osmanabadi, Sirohi, Nondescriptive (Deshi Breed), Barbari. The age of both hosts was considered to be 6 months to 6 years for both sexes.

Collection of faecal sample: During the three years of study, a total of 720 fresh faecal samples were collected in the morning directly from the rectum of each animal by using sterile disposable gloves and collected in plastic zip-lock bags. The sample collection date, sex, age, breed, and place label are placed on the zip-lock bag. The samples were transferred directly on the same day of collection to the laboratory of the zoology department of the Government Vidarbha Institute of Science and Humanities, Amravati, Maharashtra, and then stored at 40 °C for one month.

Faecal sample examination: Each faecal sample was examined by the smear method, as adopted by Soulsby (1986). Identification of eggs and larvae on the basis of morphological characteristics, as per Urquhart et al. (1996), was observed under the compound microscope at 10X and 40X magnification, and photographs were taken. Age-wise category: 6–8 months (kids), 9–36 months (young), and above 3 years (adults).Seasonal variation was studied in the three seasons: winter (October to January), summer (February to May), and monsoon (June to September).

Result and Discussion:

Month-wise prevalence of Trichuris species: The highest prevalence of Trichuris species was recorded in the months of January 2022 and February 2023, i.e., 82% and 86%, respectively, whereas prevalence was lowest in the months of April and May 2023, i.e., 34.72% and 35.41%. However, the monthly prevalence and parasitic abundance shown in the current finding are in close association with the report of Lone et al. (2011), who reported that the highest prevalence was recorded during the month of January 2012 (66.6%), whereas the lowest prevalence was recorded in the month of August 2012 (20%). These results did not correlate with our study.

Seasonal prevalence of *Trichuris species:* According to our research, as Table No. 1 illustrates, the greatest occurrence occurred in the winter (63.39%) and the lowest in the summer (32.75%). These findings bear careful comparison to those of other studies (Padwal et al., 2011). According to Umar (2005), the late high wave of infection that occurred in the winter could have originated from the eggs laid in late October and early September by both young and older sheep grazing on grassland. Saha et al. (1996) found similar findings, suggesting that favourable climatic circumstances and the availability of food throughout their growth might account for the increased prevalence of parasite infection during the winter.

Table 1: Seasonal prevalence *Trichuris species*

Season	No. examined	No. positive	Prevalence %
Winter	224	142	66.39
Summer	232	76	32.75
Monsoon	264	118	44.69

Sex wise Prevalence's of *Trichuris species:* *Our data indicates that the prevalence of females is higher (43.75%) than that of males (29.16%). Our results were in good agreement with those of other observers (Patel et al., 2001; Raza et al., 2007; Asanji and Williams, 1987; Pal and*

Qayyum, 1992; Saiful Islam KBM and Taimur MJFA, 2008). The high incidence in females could be brought on by unique physiological features. The aforementioned conditions are stressors, which lower their immunity to infections. In addition, because women are weak from breastfeeding, they are more vulnerable to infections for unknown reasons (Kuchai et al. 2011).

Table 2: Sex wise Prevalence of *Trichuris species*

Sex	Examined	Infected	Prevalence %
Female	480	210	43.75
Male	240	70	29.16

Age wise Prevalence of *Trichuris species:* In the present study, age-wise prevalence is near to similar, i.e., in kids (41.66), young (41.96), and adults (41.69), because of a lack of cleanliness, poor management practices, and awareness about deworming. This study was completely different from Islam M.K.'s (1989) finding that a high occurrence of Trichuris species was observed in middle-aged animals. This result is also different from Tariq et al. (2008). The low level of parasitism reported in adult animals is due to the immunity of the host. Previous infections and the age of the host provide effective protection against re-infection. The low level of immunity in adults is initially low but increases with the intensity and duration of exposure to infection. During this study, goats were found to be more susceptible to trichuris infection than sheep. It could be assumed from the fact that sheep do have a higher immunological response to gastrointestinal parasites compared with goats (Urquhart et al., 1996). According to Talukdar (1996), age-wise, a higher incidence of infection in young animals as compared to adults was observed in the goats of Assam, and the same type of results were reported by Pundlikrao (2009) in the goats of Nagpur, Maharashtra. The highest prevalence rate of parasites was recorded in the younger age group, while the lowest prevalence was in the older age group. A previous study by Priyanka (2019) observed the highest infection of Trichuris in young animals as compared to adults recorded in the Aeolion Plains of Haryana.

Table 3: Age wise Prevalence of *Trichuris species*

Age	Examined	Infected	Prevalence %
Kid	96	40	41.66
Young	305	128	41.96
Adult	319	133	41.69

Conclusion: The highest prevalence of Trichuris species was recorded in the winter, i.e., 63.39%, whereas the prevalence was lowest in the summer season, i.e., 32.75%. The median range of prevalence was noted in the monsoon, i.e., 44.69%. In the above study, it was observed that the prevalence of Trichuris species was highest in the winter season and lowest in the summer season. Age and sex wise, of the Trichuris species infections examined, female hosts had a higher prevalence (43.75%) than male hosts (29.16% Likewise, age-wise, there were no more differences; they were about similar, i.e., in kids (41.66%, young (41.96%), and adults (41.69%), because of a lack of cleanliness, poor waste management practices, and less awareness about deworming. In the present study, it is observed that the highest prevalence of Trichuris species is observed in the winter season and reaches its minimum level in the summer. This type of result, due to environmental factors and feeding habitats for goats and sheep, increases the chance of seasonality of parasitic infection, either directly or indirectly. But the age-wise prevalence of Trichuris species is similar to each other because of poor management practices, a lack of cleanliness, and less awareness about deworming in grazing livestock. Parasitism is of supreme importance in many agro-ecological areas and is still a serious threat to the livestock economy worldwide. Sheep and goats are known to suffer from various gastrointestinal parasites, which are of great importance. We observed that in the Amravati

district of Maharashtra, goat and sheep farmers are now very well aware of deworming practices.

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Role of Spiders in Ecosystem

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ABSTRACT:
Spiders are ubiquitous and being terrestrial predators that show high degree of diversity in ecosystem according to prey capture strategies. Despite the fact that many people have an innate fear of them, they exhibit a wide range of behaviors that contribute to the overall health of the ecosystems. They are skilled hunters, preying on insects and other small invertebrates, which helps control populations and prevent outbreaks of pests. Additionally, by eating decomposers like flies and beetles, spiders contribute significantly to the cycling of nutrients. Additionally, their webs catch flying insects. Because they disseminate seeds and act as pollinators, spiders are essential to the survival of biodiversity. Spiders serve as population control agents in most environments, preventing insect populations from becoming excessive.
Keywords: Spiders, Predator-prey, biological control, ecosystem.

INTRODUCTION:

Spiders are abundant and ubiquitous predators in terrestrial ecosystems. In fact, spiders play a vital role in the environment and without them; the earth would be a very different place. In most ecosystems spiders act as agents of population control, ensuring insect numbers are not overwhelming. Identified as generalist consumers, they also prey on a variety of organisms beyond insects, regulating the density of the species they go after. In this regard, they also benefit humans, as they eat pests such as mosquitoes, which can be deadly if carrying diseases. By eating agricultural insects like grasshoppers and beetles that feed on the crops being gathered for food, they also indirectly assist mankind. Spiders reduce the number of insect pests in agro-ecosystems, which will reduce crop damage from pests and increase crop yield, thus act as a biocontrol agents. They play an important role in the overall ecological system. Not only do they assist the environment by controlling insect populations but they also assist humans directly by stopping other pesky pests from reaching us. Some species of spiders are terrestrial that is they live on the ground, while others are arboreal, meaning they live in trees. In addition to this characteristic, they have shown a preference for inhabiting unusual environments, such as tropical woods and freezing caverns. Certain specialised species endure a variety of harsh environments. Some families, like as the orb weavers, use their distinctive webs for passive hunting. Others do this by active hunting, such as wolf spiders. Due to the fact that many species overwinter, they can aid in the early reduction of prey populations, providing farmers, horticulturists, and gardeners with an advantage over the rest of the season. It also kills other arachnids and spiders even those of the same species which helps keep their own numbers in check. Furthermore, a number of birds, lizards, wasps, and mammals—particularly those found in desert regions—use spiders as a major source of food. Bats may be preyed upon by some species of tarantulas (Theraphosidae), huntsman spiders (Sparassidae), orb weavers (Araneidae), and Nephilidae family members. In certain cases, spiders have been seen feeding on birds, and it has also been reported that birds can get entangled in their webs. These reports have led scientists to propose that flying vertebrates may be an important source of prey for certain species of spiders. *In spite of our innate dread of them, spiders are mostly benign organisms. It might surprise you to learn how important of a part spiders play in the*

ecology. Continue reading to discover the remarkable functions of certain frightening spiders as well as the part that they play in the ecology. It could make you less afraid of spiders and alter the way you perceive them.

SPIDERS MANAGE THE FOOD CHAIN:

One of the important reasons why spiders are essential predators is that they control the population of insects and other invertebrates, which could otherwise cause significant damage to crops and other vegetation. In this way, the presence of spiders in ecosystem could help to reduce pest problems associated with common spider prey, acting as a natural insect killer. On the other hand, spiders are also helpful to the ecosystem as prey. Numerous other creatures, including frogs, lizards, and birds, eat spiders. This indicates that in addition to helping to reduce pest-related problems, spiders can also support the populations of beneficial species. Spiders regulate the food chain, but they also serve as sensitive environmental health indicators because of their sensitivity to changes in their habitat and their presence or absence. can indicate the quality of the ecosystem.

SPIDERS ARE POLLINATORS:

As bee populations decreases, spiders might be the next solution to the pollination process. Spiders indirectly aid in pollination, although they do not actively engage in the process like bees or butterflies do. Spiders are known to build their webs in areas with high plant density, and their sticky webs catch a variety of flying insects, including bees and other pollinators. Spiders make sure that the pollinators that remain visit more flowers by lowering their population, which raises the likelihood of pollination. Additionally, spiders' webs can trap pollen grains, which can be transferred to other flowers when the spider moves around. Therefore, spiders play a crucial role in the interdependence of plants and animals, ensuring the health and survival of both.

SPIDERS CONTROL THE SPREAD OF INFECTIONS:

Spiders can help to kill insects that carry diseases, such as mosquitoes and moths. Typhoid, cholera, and other illnesses can be transmitted by fleas, flies, and cockroaches. Additionally, illnesses like malaria that claim more lives every year are spread by mosquitoes. The webs of spiders are highly effective at ensnaring little insects and halting the spread of illness.

SPIDERS IN CONTROLLING PEST POPULATIONS:

Spiders are natural and effective solution to reduce pest populations. Spiders play a crucial role in controlling pests, especially in agriculture where they can prevent significant damage to crops. Spiders are natural predators that feed on insects like aphids, caterpillars, and beetles, which are known to cause extensive harm to plants. By preying on these pests, spiders help maintain the balance of the ecosystem and protect agricultural yields without the need for harmful pesticides. Their ability to control pest populations naturally makes them valuable allies for farmers and gardeners alike.

SPIDERS ROLE IN NUTRIENT CYCLING:

Spiders contribute to nutrient cycling is by breaking down organic matter through their feeding habits. They are voracious predators, preying on a wide range of insects and other small creatures. When they catch and consume their prey, they not only obtain energy for themselves but also help in the decomposition process. As spiders break down the organic matter through digestion, they release nutrients back into the ecosystem. This decomposition activity performed by spiders plays a crucial role in maintaining soil fertility. The nutrients released from decomposing prey become available for plants and other organisms, ensuring a healthy balance within the ecosystem.

SPIDER VENOM:

Spiders are of great importance in the medical field and have been used for various research and product development. Venom by its name is believed to be toxic and harmful, but it can also be quite beneficial. Spider venom has the potential to treat several ailments. It’s a safe

painkiller which can be productively used to treat muscular dystrophy and strokes. Also, the venom itself is excellent anti-venom and is used to treat bites from toxic spiders.

SPIDER SILK:

They use silk to spin their webs, and the silk they produce has innumerable benefits. Spider silk is one of the most remarkable materials in the world. It is lightweight, flexible, stretchy, almost transparent, and practically stronger than steel. Its strength can be used in making strong building materials, and even in making bullet proof vests. The flexibility of it makes it practical to be used in flexible suspensions. Also, it has already been used in optical measuring devices, and for producing cross-hairs in telescopes.

CONCLUSION:

In conclusion, spiders play a crucial role in biodiversity. They act as predators, controlling pest populations and maintaining ecosystem balance. Spiders also serve as indicators of ecosystem health, reflecting the overall well-being of an environment. Their diversity is essential for maintaining a rich and thriving ecosystem. Moreover, spiders contribute to pollination and seed dispersal, while also being an important part of food webs and nutrient cycling.

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Nutritional Analysis of Different Eggs Sources in Amravati City, Maharashtra

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ABSTRACT: The present study was to determine the quality of different eggs and their nutritional composition available in Amravati City. A sample of different breeds of eggs from *Gallus gallus domesticus* was collected randomly from the local market. Among them were Kadaknath eggs, Busra eggs, and White Leghorn (Boiler) eggs. Moisture, ash, crude protein, and crude fat levels of the meat samples were compared between egg sources. A high amount of protein and ash content was observed in Kadaknath chicken eggs, which are a good source of food. A high amount of moisture content was also observed in Busra eggs. *Gallus gallus domesticus* (Busra) can be recommended as a good source of protein in food, is also a good source of nutrition, and can be beneficial for human health.
Keywords: nutrition value, egg, amravati.

INTRODUCTION

Eggs, which are main in our diet, is composed by the most of the living population. It has been widely held that the egg is an excellent food (Naber, 1979). Eggs have a great nutritional value as a food, not only because of the quantity and ratios of nutrients they contain, but also because of their bioavailability and chemical properties that respond well to man's nutritional needs. Having multifunctional properties, eggs are perceived as a nutraceutical food product (Kiczorowska et al., 2015). The potential of chicken eggs as a nutritionally complete protein and source of key micronutrients has been progressively recognised across the globe, particularly in resource-poor settings. Eggs have a high nutrient-to-energy density and contain a wide array of complete proteins, fats, vitamins, and minerals (Wallace *et al.*, 2023).

The high content of highly bioavailable proteins has provided a surplus. However, in most industrialised countries, the supply of protein from eggs does not play an important role because of the high total protein intake. In addition, eggs are recognised as a major source of vitamins (A, E, B2, B6, B12, and folic acid) and minerals (iodine, iron, selenium, and zinc) in the diet (Seuss-Baum, 2005). The mineral contents of eggs, such as Ca, P, Cu, Fe, Mn, and Zn, belong to the most influential and basic microelements that are essential for human and chicken nutrition (Attia *et al.*, 2014).

Protein is necessary to supply the materials in the animal body, which are composed of various amino acids. In the case of all animals of all ages, the need is first for maintenance (Heuser, 1941). Since chicken meat is rich in protein with low fat content, it is an indispensable food source for patients with cardiovascular systems and also for people undergoing obesity treatment (Yaranoğlu et al., 2023). Egg yolk contains specific antioxidant nutrients that support eye function (Ruxton *et al.*, 2010).

This study was aimed at determining the content of basic nutrients and selected macro- and microelements in the albumin and yolks of eggs produced in commercial poultry farms as well as organic red courtyard farms. As proportions of protein and water in the yolk vary between species, one cannot reconstruct the chemical composition of an egg solely from the proportions of albumen and yolk, together with the chemical composition of the albumen and yolk in eggs of domesticated pre-social species. In addition, caloric values cannot be used to calculate the proportions of protein and water in the egg.

The present study is therefore proposed to evaluate the nutritional composition of some species of eggs that are found in Amravati city, Maharashtra, assessing protein estimation, moisture content, and ash.

Material and Methods

The details of the experiments, materials used, and techniques adopted during the course of the investigation are briefly presented in this chapter. Different eggs were collected from markets in Amravati city. Among them were Kadaknath eggs, Busra eggs, and White Leghorn (Boiler) eggs. The identification of eggs was done according to the chicken species that was observed in Amravati City.

Collection of eggs:

Different eggs were collected from the local market in the Amravati district. Some eggs were collected from the poultry. Each egg, weighing about 50 g to 70 g, was collected.

Extraction of yolk:

The yolk part of the egg was extracted with the help of equipment. This yolky part of the egg is then kept aside and used for the experiments, which proceed with the estimation of total moisture (APHA, 1998), ash content (AOAC, 1995), and total protein by Lowry's method (1951).

The yellowish egg part contains most proteins; that's why egg yolk is used in place of egg white for the calculations.

Observation and Result: The nutritional composition of the three different types of eggs, i.e., Busra, Kadaknath, and White Leghorn (Boiler), found in Amravati city market was analysed, and the results are shown in Table 1. The moisture content among the different eggs was found to be highest in Busra, i.e., 97% moisture. It has been recorded that protein content among the eggs was slightly similar, ranging from 90.66 ± 2.7 mg/100 g to 181 ± 3.8 mg/100 g. Ash content is also found in a range of 9.00% to 9.777%.

The protein content was found to be highest in Kadaknath eggs and lowest in White Leghorn (Boiler) eggs. As the yolk contains more protein than the egg white, the protein quantity is higher in the yolk.

Nutritional composition generally refers to the percentage composition of basic constituents such as water, protein, carbohydrate, lipid, and ash. The measurement of some nutritive composition profiles, such as protein contents, carbohydrates, lipids, and moisture contents, is often necessary to ensure that they meet the requirements of food regulations and commercial specifications.

Table no. 1 Total Protein in different eggs of chicken

Sr.No	Different eggs of chicken	Total Protein mg/100gm
1	Busra	119 ± 1.9
2	White Leghorn (Boiler)	90.66 ± 2.7
3	Kadaknath	181 ± 3.8

Table no. 2 Ash and Moisture content of different eggs

Sr. No	Different eggs	Total Ash %	Total Moisture %
1.	Busra	9.099	97
2.	White Leghorn (Boiler)	9.578	86.5
3.	Kadaknath	9.750	88.5

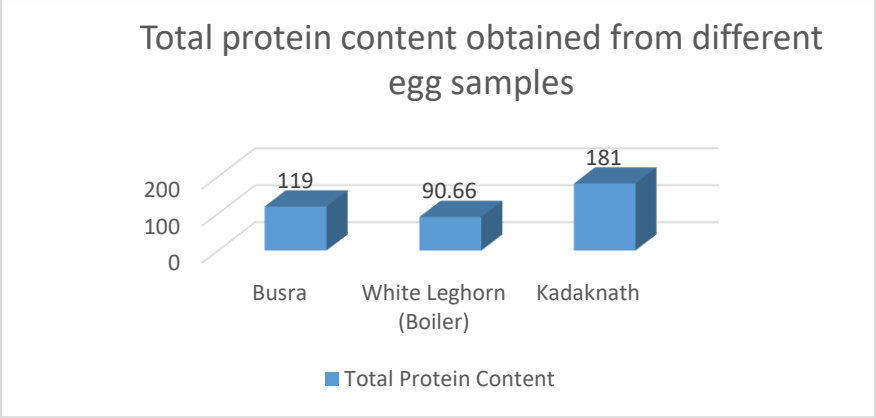

Fig. No. 1 – Total Protein content in different eggs.
The results are the mean ± S.D of the samples taken in triplets.

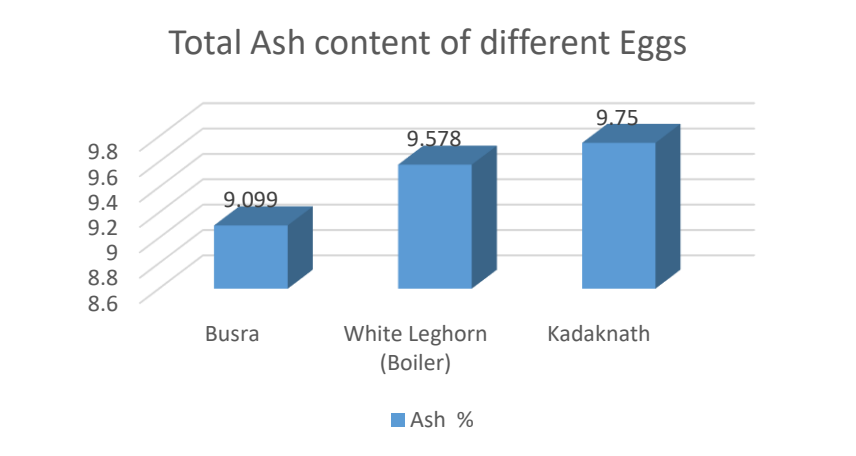

Fig No. 2 –Ash content in different Eggs

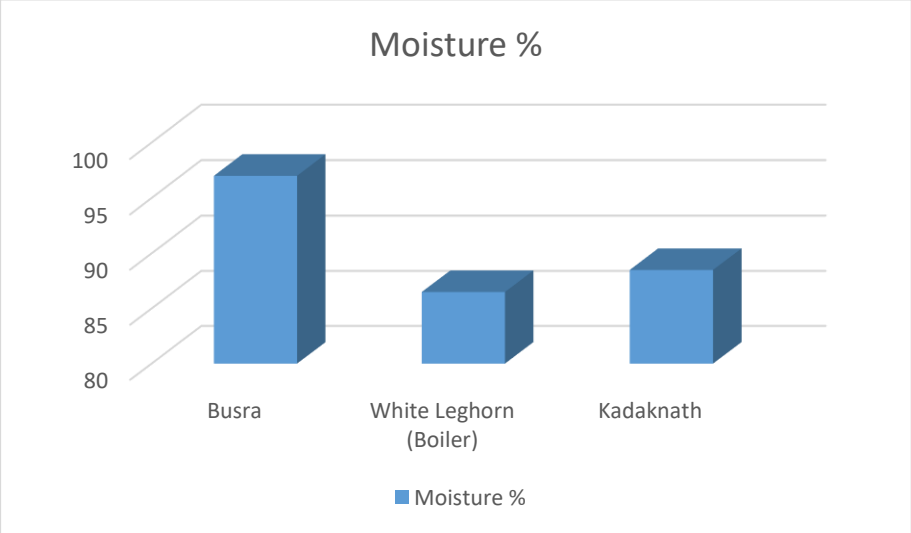

Fig No. 3 Moisture content in different Eggs.
The Samples are mean ± S.D. of the samples taken in triplets

Discussion

The different sources of eggs constitute one of the major sources of cheap nutrition for the rural population. The nutritional value of different egg sources depends on their biochemical composition, like protein, moisture content, ash content, etc. The protein concentration in the eggs of the chickens, i.e., Busra, Kadaknath, and White Leghorn (Boiler), was studied and has served the people of rural as well as urban areas of the Amravati region as a good source of protein nutrition and could be exploited commercially. The egg proteins were observed in this study. The yolk contains a large number of structural proteins with a low turnover rate, whereas the yolk is highly active metabolic tissue rich in functional proteins with higher turnover rates. Hence, this yolk had a high concentration of protein. Taking into consideration the observed high levels of protein, moisture, and ash from different egg sources, it may be suggested that consumption of these eggs' sources could be developed as a source of protein nutrition for the people of the Amravati region.

Conclusion

Based on the above results of the present research, it can be concluded that the nutritional composition of selected egg sources, including nutrients, is within the nutritional ranges required by human beings in their diet. The percentages of protein content ranged between 90.0 and 181 mg/100 g, which is quite high. From the present study, it is clear that the different egg sources are good sources of quality protein, moisture, ash, and essential minerals in the habitats of Amravati, and each egg source has its own nutritional value parameters due to their different food preferences and ecological conditions. The range of ash content (88% to 97%) gave an indication that each egg sample is a good source of minerals such as calcium, potassium, zinc, iron, and magnesium.

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Spider Diversity of Katepurna Sanctuary District Akola (MS) India

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ABSTRACT
The present study was made on the preliminary spider diversity of the Katepurna sanctuary. The Katepurna Sanctuary, located in the Akola and Washim districts of Maharashtra, is an exotic sanctuary dotted with an abundance of flora and fauna. The sanctuary lies in close proximity to the catchment area of Katepurna reservoir (Mahan Dam). Its area is geographically located at 20°25'0.54"N and 77°10'50.14"E. The land vegetation at Katepurna Sanctuary is a southern tropical dry deciduous forest. There are over 115 species of plants in this sanctuary. During the spider survey, 92 species of spiders were recorded, belonging to 19 families. Most species of spiders found belonged to the families Araneidae and Salticidae. Out of the total spider species recorded, about 48% were found to be web builders, and 52% were ground wanderers. The patterns of web-building egg laying, egg sac, feeding, and reproduction were noticed for different species and properly recorded. Araneidae (25 species) and Salticidae (9 species) Oxipidae(7 species) Gnaphosidae (6 species) Theridiidae (6 species) Philodromidae (5species) Thomisidae (4 species) Lycosidae(3 speices) Hersillidae (2 species) and Uloboridae (4 species) were recorded from different areas of Katepurna wild life. Sanctuary family, generic, and species diversity were observed. Katepurna Sanctuary has a good diversity of spiders. There is a need for the conservation of this creature because it is part of the food chain in the protected area.
Keywords: spider diversity Katepurna sanctuary salticidae, Araneidae, thomisidae, and lycosidae.
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Introduction :
Spiders are air-breathing and belong to the order Araneae of the class Arachnida and phylum Arthropoda spiders are the largest order of arachnids and rank seventh in total species diversity among all orders of organisms. Spiders are found in every habitat in the world: temperate forests, scrubland, deserts, mountains, tropical areas, and the exception of polar regions.
Spiders have eight legs, chelicerae with fangs generally able to inject venom, and spinnerts that extrude silk; their front four legs are generally larger than the hind legs. The legs of a jumping spider allow it to jump 25 times their body size, which lets them jump far and capture their prey.
Spiders have two main body parts from front to back: the cephalothorax and the abdomen. The first of these is the fused sections of the head and thorax, which include the brain, mouth, venom system, and stomach. It's also where the limbs attach. The abdominal organs include the lungs, heart, reproductive system, spinnerets, and digestive tract. Spiders have an open circulatory system. Katepurna Sanctuary is a southern tropical dry deciduous forest. There are over 115 species of plants in this sanctuary. During the spider survey, 92 species of spiders were recorded, belonging to 19 families. Most species of spiders found belonged to the families Araneidae and Salticidae. Out of the total spider species recorded, about 48% were found to be web builders, and 52% were ground wanderers. The patterns of web-building egg laying, egg sac, feeding, and reproduction were noticed for different species and properly recorded. Araneidae(25species) Salticidae (9 species) Oxipidae(7 species) Gnaphosidae (6 species) Theridiidae (6 species) Philodromidae (5species) Thomisidae (4species) Lycosidae(3 speices) Hersillidae (2 species) and Uloboridae (4 species) were recorded from different areas of

Katepurna wild life. Sanctuary family, genera, and species diversity were observed. Katepurna Sanctuary has a good diversity of spiders. There is a need for the conservation of this creature because it is part of the food chain in the protected area.

Material and Methods:

Katepurna Wildlife Sanctuary is known for its four-horned antelopes and barking deer. Other animals that can be seen at the sanctuary include black buck, leopard, wolf, wild boar, hyaena, hare, nilgai, jungle cat, and monkeys. Spider surveys are carried out for ground spiders and spiders along slow-flowing shallow streams, spiders from decaying barks of trees, shrubs, and crevices of rocks. Well-established sample techniques for spider collection are used in several designated sampling areas. The detailed descriptions of the collection techniques are as follows:

(i) Sweep Netting: This sampling method is applied to collect the foliage spiders from low-level vegetation of shrubs (up to 2 m in height). The sweep net consists of a 90-cm handle and a 40-cm ring, and the collection is poured on white canvas. The net was emptied at regular intervals to avoid loss and destruction of the specimen. During sampling time, the sweep net was moved back and forth to cover all ground-layer herbs and shrubs until all vegetation in the sampling plots was swept thoroughly.

(ii) Ground Hand Collecting: Ground Hand collection entailed collecting spider samples from the ground up to knee level. This method of sampling is used to collect the spiders, which are found to be visible in the ground, litter, broken logs, rocks, etc.

(iii) Aerial Hand Collecting: Aerial hand collection involved the collection of spiders.
samples from knee level to arm length level. This method accesses web-building and free-living spiders on the foliage and stems of living or dead shrubs, high herbs, tree trunks, etc.

(iv) Vegetation Beating: The method is employed to access spiders living in shrubs, high herb vegetation, bushes, and small trees and branches. The spiders are collected by beating the vegetation with a stick and collecting the samples on a cloth (1m by 1.2 m).

(v) Litter sampling: Litter, i.e., deciduous from the ground, was collected by hand and put in a big tray. Litter sampling involved sorting spiders from the litter collection tray. With the above methods of collection, the spiders were collected and observed under a stereo-zoom binocular microscope (for small or tiny spiders) wherever necessary in the field itself. Later, all the spiders were photographed by a Canon 60D with a macro lens in their natural habitat.

Observation and results:

During the study, 92 species were recorded, belonging to 19 families that represent 30.64% of the total families reported from India. Most of the species of spiders found belonged to the families Salticidae and Araenidae. *Neoscona* was found to be the most abundant species in this region, followed by *Oxyopes, Theridion, Plexippus paykulli*, Sp., etc. Out of the total spider species recorded, about 48% were found to be web builders, and 52% were ground wanderers.

Table- Spider families and species recorded from Katepurna wild life sanctury.

Sr.No	Family	Species	Guild1
1	Araneidae	*1Araneus ellipticus* Tikader & Bal, 1981 Female 2 *Argiope aemula* Walckenaer, 1842 Female 3 *Chorizopes bengalensis* Tikader, 1975 Female 4 *Cyclosa bifida* Doleschall, 1859 Male, Female 5 *Cyclosa hexatuberculata* Tikader, 1982 Female 6 *Cyclosa insulana* (Costa 1834) 7 *Cyrtophora cicatrosa* Stoliczka, 1869 Female	Orb weaves

		8 Cyrtophora citricola (Forsskål, 1775) 9 *Eriovixia excelsa* Simon 1889 Female 10*Eriovixia laglaizei* Simon, 1877 Female 11*Larinia chloris* Audoin, 1825 Female 12 *Larinia argiopiformis* *13 Larinia lineata* 14 *Neoscona adianta* 15 *Neoscona bengalensis* 16 *Neoscona crucifera* 17 *Neoscona mukerjei* Tikader, 1980 Female 18 *Neoscona nautica* L. Koch, 1875 Female 19 *Neoscona punctigera*, Male , Female 20*Neoscona subfusca (C. L. Koch, 1837)* 21*Neoscona theisi* Walckenaer, 1842 Female 22*Neoscona vigilans* Blackwall, 1865 Female, Male 23*Poltys nagpurensis* Tikader, 1982 Female 24*Poltys illepidus C. L. Koch, 1843* 25 *Zygiella indica* Tikader & Bal, 1980 Male, Female	
2	Clubionidae	*1.Clubiona abbotii* L. Koch, 1866 *2.Clubiona filicata* O. P.-Cambridge, 1874 Female *3.Cheiracanthium inclusum*	Foliage
3	Eresidae	*1.Stegodyphus sarasinorum* Karsch, 1891 Female *2.Stegodyphus lineatus, Latreille, 181*	Orbweb weavers
4	Gnaphosidae	*1.Agroeca pratensis* Emerton, 1890 Female *2.Litopyllus temporaries* Chamberlin, 1922 Female *3.Micaria longipes* Emerton, 1890 Female *4.Talanites echinus* Chamberlin, 1922 Female *5.Zelotes fratris* Chamberlin, 1920 Female *6.,Zelotes sp.* Female	
5	Hersiliidae	*1,Hersilia savignyi* Lucas, 1836 Female *2,Hersilia sp*	Foliage hunters
6	Lycosidae	*1.Hippasa* greenalliae (Blackwall,1867) Female *2.Lycosa poonaensis* Tikader & Malhotra,1980 Female . *3.Pardosa pseudoannulata* (Bösenberg & Strand,1906) Female, Male	Ground runners
7	Eutichuridae	*Cheiracanthium inornatum* O. P.-Cambridge,1874 Female, Male	
8	Oecobiidae	*Oecobius putus* O. P.-Cambridge,1876 Female	
9	*Oxyopidae*	*1.Oxyopes bharatae Female, Male* *2Oxyopes pankaji Gajbe & Gajbe,2000 Female .* *3.Oxyopes javanus Thorell, 1887 .* *4.Oxyopes macilentus Female, Male* *5.oxyopes ramosus Female, Male* *6.Peucetia viridana (Stoliczka, 1869).* *7Peucetia albescens*	Stalkers
10	Philodromidae	*1.Philodromus kuttanadensis* *2.Philodromus rufus*	

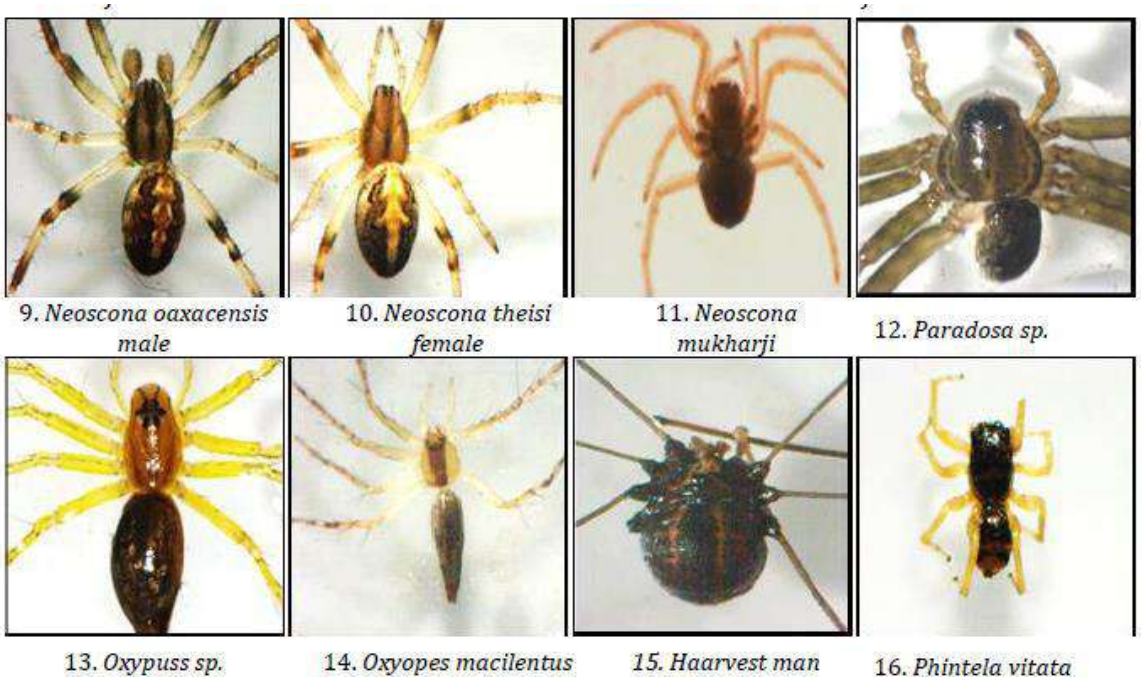
		3.Philodromus sp. *4Philodromus sp.* *5Tibellus oblongus*	
11	Pholcidae	*1.Artema atlanta* Walckenaer, 1837 Female *2.Crossopriza lyoni* (Blackwall, 1867) Female *3.Pholcus phalangioides* (Fuesslin, 1775) Female, Male	
12	Pisauridae	*1.Dolomedes sp.* *2.Thalassius albocinctus*	
13	Salticidae	*1.Cosmophasis thalassina* . *2.Hasarius adansoni* (Audouin,1826) Female *3.Hyllus semicupreus* (Simon,1885) Male *4Menemerus bivittatus* *5.Phintella vittata* (C. L. Koch,1846) Female, Male *6.Plexippus pkulli* (Audouin,1826) Female, Male *7.Rhene flavigera* (C. L. Koch,1846) Male *8.Telamonia dimidiata* (Simon,1899) Female, Male 9.*Thyene imperialis* (Rossi,1846) Female, Male	Stalkers
14	Scytodidae	*1.Scytodes fusca* Walckenaer, 1837 Male and Female *2.Scytodes pallida,* Male and Female *Scytodes univittata*	Foliage hunters
15	Sparassidae	● *Heteropoda cervina* ● *Heteropoda venatoria* ● *Micrommata virescens* ● *Olios argelasius*	
16	Tetragnathidae	*1.Leucauge decorata* (Blackwall,1864) Female *2.Tetragnatha mandibulata* Walckenaer,1841Female	Orbweb weavers
17	Theridiidae	*1.Ariamnes colubrinus* female 2.. *Argyrodes argentatus* O. P.-Cambridge,1880 Female, Male *3Parasteatoda mundula* (L. Koch,1872) Female . *4.Theridula gonygaster* (Simon,1873) Female *5Nesticodes rufipes* (Lucas,1846) Female, Male *6Theridion sp.* Female	Scattered line weavers
18	Thomisidae	*Indoxysticus minutus* (Tikader, 1960) Female, Male . *2.Thomisus okinawensis* Strand,1907 Female *3.Tmarus angulatus* 4. *Xysticus cristatus* Female	Ambushers
19	Uloboridae	*1.Octonoba sinensis* *2.Uloborus plumipes* *3.Uloborus walckenaerius* Latreille,1806 Female, Male *4.Zosis geniculata*	Orbweb weavers

Table2 :

Sr No	Family	No of species
1	Araneidae	25
2	Clubionidae	3
3	Eresidae	2
4	Gnaphosidae	6
5	Hersiliidae	2
6	Lycosidae	3
7	Eutichuridae	1
8	Oecobiidae	1
9	Oxyopidae	7
10	Philodromidae	5
11	Pholcidae	3
12	pisauridae	2
13	Salticidae	9
14	Scytodidae	3
15	Sparassidae	4
16	Tetragnathidae	2
17	Theridiidae	6
18	Thomisidae	4
19	Uloboridae	4

1. *Araaneus sp.* 2. *Cyclosa sp* 3. *Eriovixia sp.* 4. *Neoascona theisi sp*

5. *Neoscona oaxacensis female* 6.*Neoscona female* 7. *Neoscona male* 8. *Neoscona oaxacensis female*



9. *Neoscona oaxacensis male* 10. *Neoscona theisi female* 11. *Neoscona mukharji* 12. *Paradosa sp.*

13. *Oxypuss sp.* 14. *Oxyopes macilentus* 15. *Haarvest man* 16. *Phintela vitata*

Summary and Conclusions: The spider fauna of India is represented by 438 genera and 1685 species (Keswani et. al. 2012). The present study represents 19 families, 63 genera and 92 species arranged according to their foraging behaviour in the field. The distribution of some families was found to be continuous (Araenidae, Hersiliidae, Salticidae, Tetragnathidae, etc.), while others had a very discontinuous distribution.

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Impact of soymeal on gut morphology of freshwater fish *Labeo rohita*

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Abstract: Nowadays, various types of ingredients are used as food in aquaculture. The choice of these foods depends on the amount of nutrition present in them and how much nutrition they can provide to fish. Considering all these factors, a 30-day experimental trial was performed to check the impact of soy meal on fish. Soy is a high source of protein and can be easily used as a fish meal as it is easily available at a low price. However, it is found that the presence of some content, such as high carbohydrate and saponins, lectins, and phytates, can have a negative impact on the gut health of the fish. An experiment performed in the lab showed that a high level of dietary soybean meal can cause intestinal inflammation called enteritis in fish (Labeo rohita), which can lead to reduced nutrient absorption, high mucus secretion, and low digestion. We can conclude that high use of soy products can affect human health, as well as that content like saponin and tannin become incompatible with human gut health.
Keywords: aquaculture, fish feed, saponins, tannins, gut health.

Introduction

Fish food plays a very important role in cultivating fish. Fish meal, with its nutritional quality, is a major component of feed for most cultivable fish species. The food materials that are more easily and conveniently available and are cheaper to produce than fish meal are plant proteins that have been used in combination with fish meal. Among all the plant protein sources that are available, soybean is being highly used for fish because of its rich protein content, which can help to balance the amino acid profile. When used as a dietary supplement (Aslaksen et al., 2007). It can help provide the necessary nutrients for fish growth and development. Soybean meal is also often used as a cost-effective alternative to traditional fishmeal in fish feeds. This can help reduce the overall cost of aquaculture production, making it more economically viable and sustainable. Using soybean meal in fish feeds can contribute to sustainability in aquaculture. By reducing the reliance on fishmeal, which is often made from wild-caught fish, soybean meal can help conserve marine resources. In many areas, groundnut (peanut) is also used as a fish meal and is also a good source of protein in fish feed. Even though groundnut oil cake is highly used and is easily palatable as it has better binding properties than soybean oil cake meal (Lovell 1989), its use as fish meal is low because it has low lysine and methionine contents (Robinson & Wilson 1985; Lovell 1989).

Using soybeans can reduce the overall production costs of fish farming. As the demand for fishmeal increases due to the growing aquaculture industry, there is a need for sustainable protein sources. Soybean is a renewable resource and can help reduce the pressure on wild fish stocks that are used to produce fishmeal. Soybean is not only a good source of protein but also provides essential nutrients, including vitamins, minerals, and fatty acids.

which can contribute to fish health and growth. In freshwater aquaculture, carp like Labeo rohita are commonly used (ICLARM, 2001). L. rohita is a major carp, widely cultured throughout India owing to its high commercial value. Rohu is often raised in aquaculture systems and has become a significant species for fish farming due to its rapid growth and adaptability to various water conditions. It's an important source of protein for the local populations and contributes to the fishing and aquaculture industries in South Asia. Growth rate is one of the most important parameters determining the economic efficiency of commercial fish culture, which is influenced by several biotic and abiotic factors (Brett and

Groves, 1979).In many fish species, fishmeal protein can be replaced by soybean meal protein, which also enhances growth. It is also found that high dietary soy protein can result in low food intake, reduced weight, some morphological changes in the intestinal epithelium, and some abnormal illnesses (Baeverfjord G; Krogdahl Å 1996). The challenges behind using soya meal as a protein are the low level of the amino acid methionine and the presence of a high carbohydrate level, which may negatively affect fish. The present study mainly focuses on the impacts of different soymeals as proteins on the gut health of Labio rohita. There are many studies that have investigated relevant topics and found many surprising results (Carter et al., 2013; Garamszegi et al., 2013).

Materials and Methods

Preparation of experimental tanks:The experiment was carried out in lab conditions. The fish were obtained from the nearby fish farm, and the healthy fish were acclimated for about a week. The fish were kept in glass tanks with a capacity of about 100 capacity.The tanks were categorised as controlled and experimental and were filled with normal well water with proper aeration.

Stocking of fish: Each tank was stocked with fingerlings (10 fish per tank). The fish were stocked in equal numbers. The water was changed every day, and dechlorinating liquid was added to avoid any fungal infection. All other parameters, such as pH, dissolved oxygen, temperature, total alkalinity, total hardness, and free carbon dioxide, were recorded following standard methods (APHA, 1998). The initial body weight and overall body composition of the fish were determined before starting the experiment. The fish, Labeo rohita, was fed twice a day. The remaining food and excretory waste were removed every second day by using the syphoning method. Before stocking, the weights of the fish were recorded.

Preparation of an experimental diet: Fish were fed a normal diet in the form of a palette prepared by using rice bran, groundnut oilcake, fish meal,soybean oilcake, wheat bran, multivitamins,multivitamin etc., as these additional supplemental foods can enhance growth (Rahman et al., 2006). Control diet
was prepared without soy meal, while the experimental diets contained a supplement of soy meal. i In the experimental diet, the soya content was 2 g, which was gradually increased to 4 g and 6 g. All the dietary ingredients were finely ground into powder and sieved properly. By using distilled water, dough was prepared, and small palates were prepared by using a palletizer. The feeding intensity was observed by visual estimation (Pillay, 1953) based on the enlargement of the gut and the amount of food contained in it.

Histological study: A histological study was performed to analyse the morphological changes in the gut, where tissue was collected and slides were prepared by the microtome method. Further, the slides were stained and observed. The histological parameters were observed with increasing dietary soybean meal.

Results and discussions:

After giving soymeal for 30 days, it was found that high levels of soy protein in fishmeal may disturb the fish's intestinal functions or damage the gastrointestinal tract. Soybean meal contains antinutritional factors such as trypsin inhibitors, lectins, phytates, and tannins. These compounds can interfere with the digestion and absorption of nutrients such as protein, calcium, and zinc in the fish's gut, leading to reduced nutrient utilisation and growth. Excessive use of soybean meal in fish diets can lead to gut inflammation. Which may cause allergic reactions: These allergic reactions may be due to the saponin content of soybeans. Some fish species can be sensitive to the saponin content, which may cause hemolysis and create a soap-like sticky substance that can obstruct fish intestines from absorbing specific components in soybean meal. These sensitivities can manifest as allergic reactions, inflammation, and gut health issues.

Imbalance of Amino Acids: Soybean meal may not provide a complete profile of the essential amino acids required by all fish species. Inadequate amino acid composition causes imbalances in the diet, negatively impacting gut health and overall growth (Chen et al., 2011).
Reduced Digestibility: The digestibility of soybean meal was found to be difficult for the fish species. In these cases, soybeans were found to be less digestible, leading to reduced nutrient absorption and potential gut health issues.
Changes in Gut Microbiota: It is very well known that all fish have a microbiota in the GI tract (Ringø E, Zhou Z, 2016). The inclusion of soybean meal in fish diets can alter the composition of the gut microbiota. An imbalance in the gut microbiota can affect overall gut health and digestion.

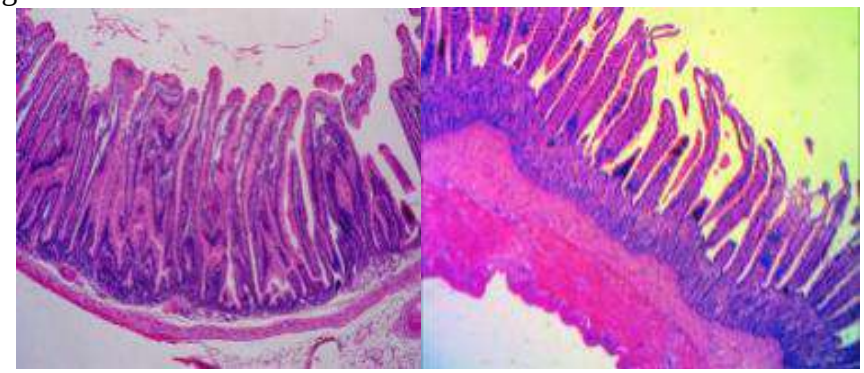

Fig: (A) Fig: (B)

(A) Histology of the intestine with the impact of the control diet showing a normal intestinal condition. (B) Histology of the intestine with the impact of soymeal, showing some disintegration in the intestinal lining and inflammation in the mucosal lining.
Enteritis: It has been found that excessive use of soybean meal in fish feed can lead to enteritis, which is inflammation of the intestine. Laporte and Trushenski (2012) Enteritis can disrupt the digestive process, cause diarrhoea, and impact overall fish health.
Impaired Nutrient Utilisation: The antinutritional factors in soybean meal can interfere with the proper utilisation of nutrients, leading to suboptimal growth and overall health in fish (Wilson & Poe, 1985; Olli et al., 1994), which can be possible causes of death.
The presence of soy in water also showed some changes in water quality parameters such as PH, alkalinity, hardness, temperature, dissolved oxygen, etc.

Parameters	Normal water	Water with soymeal
PH	7.9	8.7
Temperature	25-27 ° c	25-27° c
alkalinity	144mg/l	140mg/l
Acidity	0	0
hardness	112 mg/l	103 mg/dl
Dissolved oxygen	6.7mg/l	6.3mg/dl

Soybean meal is commonly used as a plant-based meal for fish. However, a proper diet can enhance the growth performance of fish, but as the amount increases, it can significantly affect the gut health of fish (Krogdahl et al., 2003). After some days of feeding trials, results indicated that dietary soybean meal can significantly affect the gut health of fish. This research suggests that one content of soya, known as soya saponins, is not able to be absorbed properly in the GI

tract and is poorly metabolised in the colon. The study of gut analysis shows various results about the impact of the soya diet on fish (Ekpo et al., 2014).

Conclusion

Being a good source of protein, having high availability, and being cheap, soybean meal is very commonly used as a fish meal in aquaculture. However, an appropriate amount of soy meal can be a good source of protein and may act as a good alternative to commercial food. But plant-based products can have both advantages and disadvantages (Barrows et al., 2008). But the high inclusion of soy as a protein source in aquafeed can cause damage to the intestinal lining and gastrointestinal tract and impair fish immunity. There are many products made from soya that are also being used by humans, as legumes are a good source of protein and many micro- and macronutrients. But the study suggests that the saponin content in soya is not easily digestible and is poorly absorbed and metabolised in the GI tract (Bach-Knudsen, 1997). Therefore, these findings can be important for further understanding the bioactivity of these compounds in the human body.

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26

Fluttering Diversity: Butterfly Species Around Sant Gadge Baba Amravati University Campus, Amravati Region.

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ABSTRACT:
Located in the centre of Amravati, SantGadge Baba Amravati University is home to a wide variety of butterflies, adding to the great biodiversity of the area. The diverse terrain of the institution, which includes both woodland and meadow areas, provides the perfect habitat for these sensitive animals. This investigation explores the fascinating realm of butterflies, emphasising their distinctive traits and functions within the environment. "True butterflies" refers to a family of butterflies that includes swallowtails, brush-foots, gossamer-wings, whites and sulphurs, and metalmarks, all of which contribute to the colourful nature's fabric.
Keywords: butterfly, diversity, SantGadge Baba Amravati University campus, Amravati.

INTRODUCTION

According to **Whitaker and Capatin (2008),** one of the most fascinating aspects of the world is its remarkable diversity, which includes roughly 10 million species. Biodiversity is generally considered the "Umbrella term," referring to organisms found within the living world. In the heart of Amravati, the SantGadge Baba Amravati University campus is more than just an academic hub; it's a thriving habitat for a captivating variety of butterflies. The university's diverse landscape, ranging from vibrant meadows to wooded areas, provides a perfect home for these delicate creatures. This invites you to discover the enchanting world of butterflies that add a splash of colour to the university's surroundings, showcasing the fascinating connection between nature and education. As you stroll through the university grounds, you'll encounter a mesmerising display of butterfly species, each contributing to the vibrant tapestry of biodiversity. The geographical mix of the Amravati region creates different habitats, making it an ideal environment for butterflies to not only survive but also thrive.

The second-biggest order of arthropods, Lepidoptera (butterflies and moths), are particularly useful for biodiversity surveys since they are the easiest to identify **(Erhardt, 1985; Kremen, 1994; Inouye, 2001; Tiple and Arun, 2009).** One of the most exquisite creatures in the kingdom Animalia's largest phylum, Arthropoda, are butterflies. Due to their enormous taxonomic diversity and quick response to environmental changes, arthropods are excellent markers of the biodiversity of their ecosystems (Zamre**, S., & Pradnya Kadam, V. S. T., 2020).** The foundation for ecosystem sustainability is this diversity. Features that enable the species to function and produce commodities and services necessary for human well-being. Pieridae family (which includes white, sulfurur, Jezebel, and cabbage butterflies; around 1100 species divided into four subfamilies).

Butterflies belong to about six families. The term "true butterflies" refers to the first five families: swallowtails, brush-foots, whites and sulphurs, gossamer-wings, and metalmarks. Sometimes the last group—the skippers—is taken into consideration independently. The term "swallowtail" (Family Papilionidae) refers to the appendages on the hindwings of numerous species in this family that resemble tails. Since not every member of the Papilionidae family has a tail, a butterfly without one may still be a swallowtail. Additionally, the colours and patterns of swallowtails' wings make it simple to identify the species. The largest family of butterflies is the brush-footed family, which has around 6,000 species described globally, despite the fact that there are only about 600 Papilionidae species that are found worldwide.

With the wings of a gossip The gossamer-winged butterflies are a group of hairstreak, blue, and copper butterflies belonging to the family Lycaenidae of butterflies. Since most are rather small, they can be challenging to recognise, hard to capture, and challenging to photograph. The term "gossamer-winged" describes the wings' sheer appearance, which is frequently speckled with vibrant colors. In the sunlight, they flash. The tropics are home to hairstreaks, whereas temperate zones are typically home to blues and coppers.

Metalmarks, belonging to the Riodinidae family, are mainly found in tropical regions and range in size from small to medium. The metallic-looking patches that frequently cover their wings give rise to the moniker "metalmarks."

It is simple to distinguish skippers (Family Hesperiidae) from other types of butterflies. A skipper's sturdy thorax, in contrast to most other butterflies, may give the impression that it is a moth. In addition, skippers' antennae differ from those of other butterflies. Skippers have antennae that terminate in a hook, as opposed to butterflies' "clubbed" antennae. The term "skippers" refers to the way they travel, as they quickly hop from flower to flower. Skippers are often bland-coloured birds, despite their spectacular flying display. Most have orange or white markings and are brown or grey in colour.

Materials and Methods

Study Area: The Amravati University campus is spread over an area of about 470 acres and is situated at about 4 k.m. north-east of the city of Amravati in the Pohara forest range. In terms of biodiversity, SantGadge Baba Amravati University Campus, Amravati, is a compact, diversified block located in the Indian state of Maharashtra. The butterflies were listed by various methods. That is, by actual observations or photographic methods. The observations were done at a distance of 1-3 metres and identified by using physical features. Portable cameras for recording the diversity of butterflies. During field trips, we used our smartphones to take pictures of butterflies in various habitats. The ease of usage and minimal disturbance were made possible by the simplicity of mobile photography.

OBSERVATION:

Fig. 1 – *Danauschrysippus*

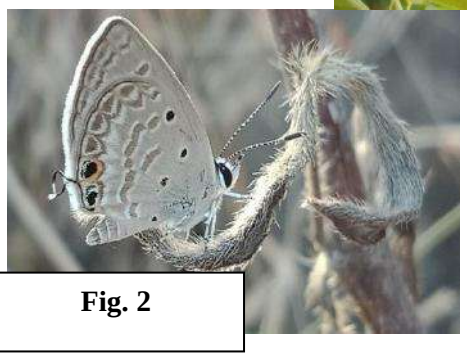
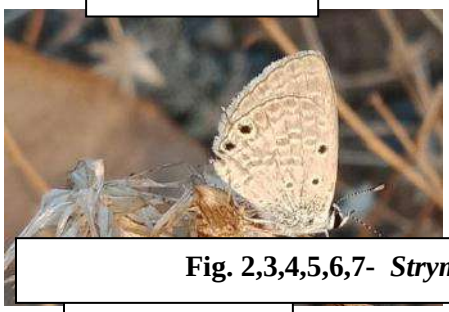


Fig. 2

Fig. 3

Fig. 4

Fig. 5



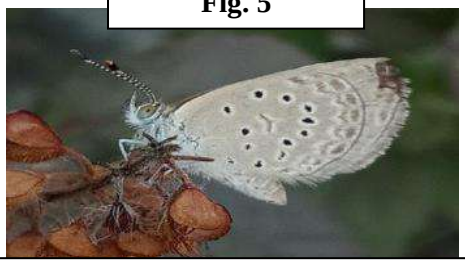


Fig. 2,3,4,5,6,7- *Strymonmelinus*(family- Lycaenidae)

Fig. 6

Fig. 7

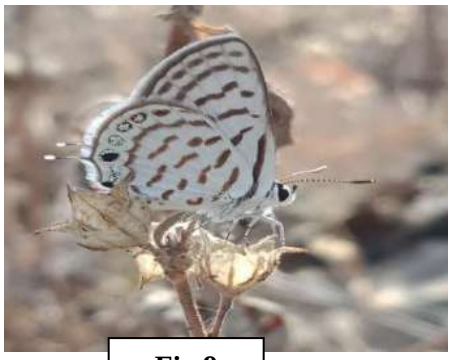


Fig. 8

Fig.9

Fig. 8,9- *Tarucus*(Family – Lycaenidae)

Fig. 10 – *Catopsilapyranthe*
Family Pieridae

Table 1. Obeserved butterfly species-

Sr. No.	Name of Butterfly Species	Family
1.	*Danuschrysippus*	Nymphalidae
2.	*Strymonmelinus*	Lycaenidae
3.	*Papilodemoleus*	Papilionoidae
4.	*Tarucus*	Lycaenidae
5.	*Euremahecabe*	Pieridae
6.	*Graphiumagamemnon*	Papilionoidae
7.	*Junoniaorithya*	Nymphalidae
8.	*Chaladespandava*	Lycaenidae
9.	*Papiliopolytes*	Papilionoidae
10.	*Catopsilapyranthe*	Pieridae

DISCUSSION: Because diversity is based on niche time stability, more diversity is seen in niches when there are many available niches. While varied settings produce increased diversity,general homogeneity conditions reduce diversity. (Grey, 1980; **Sanders, 1968).** The purpose of the current study is to look at the variety of butterflies on the SantGadge Baba Amravati University Campus, Amravaty, Maharashtra. A total of 10 butterfly species, representing 4 families, were identified during the current survey.

Table 1 lists all of the butterfly species that have been observed. Of the total number of butterfly species given, two are members of the Pieridae family, three of the Lycaenidae family, two of the Nymphalidae family, and three of the Papilionidae family. The majority of the identified species are members of the Licaenidae family.

The listed species belong to diverse butterfly families, showcasing the rich biodiversity within the order Lepidoptera. *Danuschrysippus, Junoniaorithya, and Papiliopolytes* are members of the Nymphalidae and Papilionoidae families, while *Strymonmelinus*, *Tarucus*, and Chaladespandava belong*ito thepyranthe and Euremahecabe are* classified under Pieridae, contributing to the family's vibrant colors. Papilodemoleus*gamemnon is* another Papilionoidae species. This collection exemplifies the intricate patterns and ecological significance found in butterflies across various habitats, underscoring their role in pollination and ecosystem dynamics.

Conclusion: In conclusion, SantGadge Baba Amravati University Campus is home to a delightful variety of butterflies, bringing natural beauty to the campus. Families like Papilionidae, Nymphalidae, and Pieridae showcase the diverse colours and patterns of these winged creatures. From the majestic Common Mormon to the intricate designs of the Blue Tiger, these butterflies create a picturesque environment for students and faculty. The images provided capture the charm of these fluttering inhabitants, highlighting the harmonious blend of academia and nature at the university.

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27

Amino Acid Modulation In Fresh Water Fish *Ophiocephalus Striatus* Exposed To Cypermethrin

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Absract: Cypermethrin, a synthetic pyrethroid, showed non-target effects on the freshwater fish, *Ophiocephalus striatus*. The present study showed the impact of a sublethal concentration (0.0007 µ/lit) of cypermethrin on the concentration of amino acids in two tissues, such as the liver and muscle, of the freshwater fish *Ophiocephalis striatus* at different time intervals. There was an observed increase in amino acid levels in both the muscle and liver tissues of *Ophiocephalus striatus* at different time intervals.

Key Words: Cypermethrin, liver, muscle, amino acid.

Introduction

Water is the main component of the environment, which is a dynamic entity. It has an effective means of transferring and transporting waste and other materials. Soil, on the other hand, is a stationary entity that is indirectly affected by the contaminated water. Toxic waste dumped on the soil may cause harm to animals and, indirectly, to humans. The structure and function of lotic ecosystems changed due to pollutants. [1] The toxic effect of contaminated water on non-target organisms is observed by 2, 3. One of the environmental problems is the lack of proper management of domestic and industrial wastes, which release hazardous chemicals. There is no doubt that these excessive levels of pollutants are causing a lot of damage to human and animal health. The organic pollutants may cause declines, deformities, and the death of autistic people, which in turn cause disease in humans. [4-6]. The aquatic environment is very important because it is a storehouse for a variety of fishery resources. Presently, aquatic pollution is a serious problem around the world. It has been estimated that about 70,000 man-made chemicals are used every day. These chemicals have contributed a lot to the green revolution, but their deleterious effects on various ecosystems cannot be ignored.

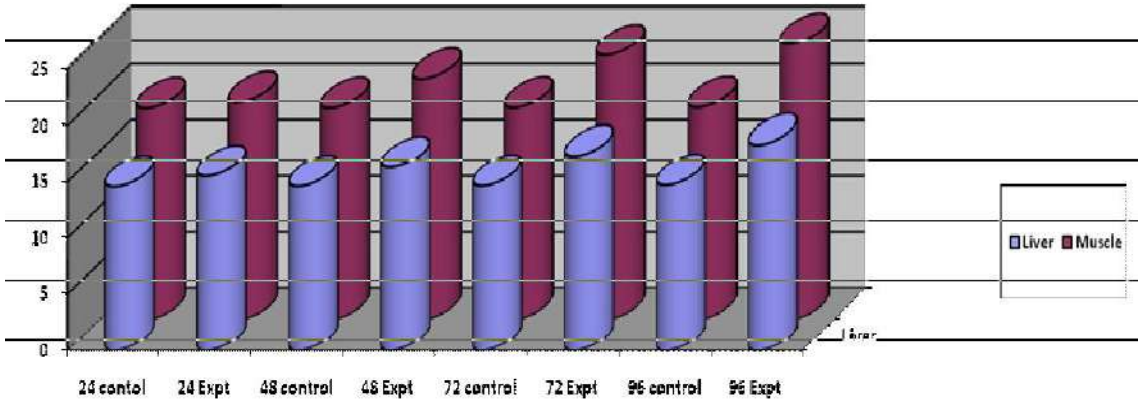
Material and Method

The freshwater fish, *Ophiocephalus striatus*, was collected from Wadali Lake around Amravati, India. The fish were acclimatised to laboratory conditions for one week. The LC_{50} value was calculated by probity analysis method 8. The LC_{50} value is 0.0007 µ/lit at 72 h. The acclimatised fish were exposed to sublethal concentrations for 24 h, 48 h, 72 h, and 96 h; simultaneously, a control group of healthy fish were maintained under identical conditions. The fish were sacrificed at the end of the exposure period, and the liver and muscle were processed for biochemical estimation. Amino acid was estimated by the method of Moor and Stein[10].

Result

The amino acid contents in the liver and muscle of the freshwater fish *Ophiocephalus striatus were* exposed to sublethal concentrations of cypermethrin at different time intervals, and they showed a higher trend as compared to the control value. Raised amino acid levels were the result of the breakdown of proteins for energy and the impaired association of amino acids in protein synthesis.

Fig: Changed in the Liver and muscle amino acid of the freshwater fish *Ophiocephalus striatus* exposed to sublethal concentration of cypermethrin at different time interval



Discussion

In the present study, an increase in the amino acid level was observed under the toxic effect of a synthetic pyrethroid, cypermethrin, exposed to the freshwater fish *Ophiocephalus striatus*. Amino acids are considered one of the most reliable techniques for the detection of changes in protein synthesis in cells, and therefore, the protein pattern can be used as a criterion for the differentiation between several organs exposed to some pollutants.

A similar study was also given by 10 on the freshwater fish *Cirrhinus mrigala.* The toxic effect of cypermethrin also showed an increased trend in *Cirrihinus mrigala11; it was* observed that there was an increase in the amino acid level in the tissues of *Labeo rohita* and *Cirrhinus mrigala* exposed to fenvelerate. A similar increase in amino acids in *Labeo rohita* exposed to endosalfan was observed by 12. The freshwater fish *Clarias batrachus* exposed to cypermethrin, showed increase level of amino acid in muscle and kidney of fish13, 14, observed that piscicidal activities of an aqueous extract of *Euphorbia tirucalli* on freshwater fish *Chanaa punctatus* altered the level of amino acids. [15] reported the effect of cypermethrin on the gill, liver, and muscle of the freshwater fish *Tilapia mossambica* that increased in amino acids.A similar study was also given by16, who observed an increase in the amino acid level in the liver of *Channa maruius* when exposed to dimethoate and monocil.

It was concluded that protein decreased because of increased utilisation due to pesticidal stress, which also breakdown protein released amino acids in the tissue that was used for a prolonged period.

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28

Study of Diversity of Moths In Urban Areas of Khamgaon City, Buldhana(M.S.)

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Abstract:
Moths belong to the order Lepidoptera, and this type of fauna is easily affected by the slightest change in environment. Keeping this in mind, moths could be used to check the minute change in environment, which can be called bioindicators of environment. Taking a chance to investigate environmental health, all possible efforts were carried out in this work to list and unfold this hidden fauna of urban areas of Khamgaon city and some surrounding areas. The collection of moths was carried out from June 2022 to August 2022. A total of 13 moth specimens were collected by using the simple light trapping method operated from dusk to dusk and by using the photographed method. The moths were identified by family level. The families Sphingidae, Geomtridae, Erebidae, Noctuidae, and Geometridae were recorded. Family Erebidae dominated among all 5 families in diversity and abundance.
Keywords: lepidoptera, bioindicators, simple light trapping, erebidae, urban Urban Areas.

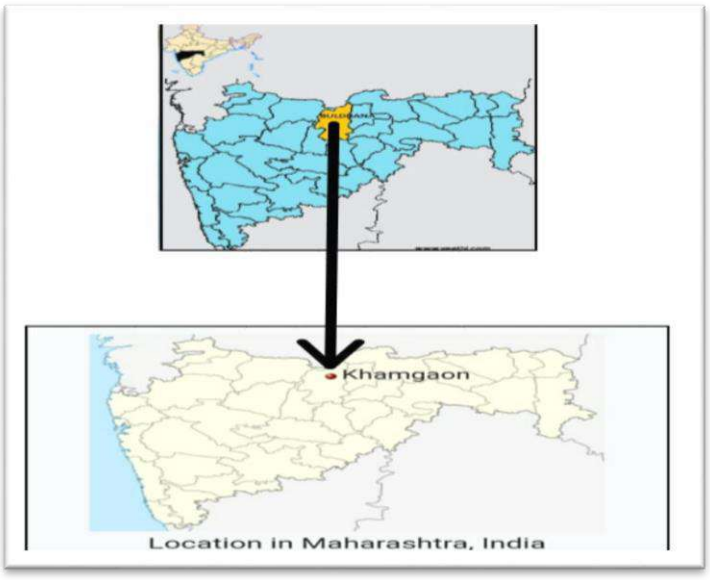
Introduction:
Biodiversity and natural resources form the root of all living systems. Insects comprise more than half of the world's known animal species (Wilson, 1992), of which the second largest and most diverse order is the Lepidoptera of the class Insecta (Benton, 1995). Insects, especially moths, play an important role in earth ecosystems and have an effect on the environment. A recent recorded report shows over 1,27,000 species of moths found all over the world (Alfred et. al., 1998) and over 12,000 species found in India. (Chandra and Nema, 2007). *Lepidoptera* is probably one of the most suitable groups for most quantitative comparisons between insect faunas to be valid, for the many reasons elaborated by Hollway (1980, 1984, and 1985), especially their abundance, species richness, response to vegetation and climate, ease of sampling using light traps, and relatively advanced taxonomy. Moths are the cousins of butterflies. Moths also play a vital role in telling us about the health of our environment, like canaries in the coalmine. Documenting the diversity of moth fauna can help lead to new evolutionary insights and be a first step in developing conservation goals for lepidopteran insects. Hence, in the present study, an attempt has been made to study the diversity of moths in and around Khamgaon city, Maharashtra, which is still not investigated. The main objective was to study the moth fauna, collect them, identify the moth diversity, and determine their occurrence. The study was carried out from June 2022 to August 2022.

Study Area: The present study was carried out in an attempt to understand and measure the status of moth diversity in and around Khamgaon city. Khamgaon, the urban as well as largest industrial area and taluka place, is situated in Buldhana district of Maharashtra state and lies in the biogeographic zone of the **Deccan Peninsula**. The state of Maharashtra is located in the Deccan region of India. This area is located between Longitude 200 34'07 N and Latitude 760 23'21" Khamgaon is situated 50 km from Buldhana. Moths were collected from in and around Khamgaon city. The survey was conducted by visiting the areas of the residential area: Rekha Plot, Khamgaon, Januna Lake, the college campus, and the botanical garden. The latitude of Rekha Plot is 20.711622 and the longitude is 76.566132, and the DMS latitude is 20°42'41.8392" N and the DMS longitude is 76°33'58.0752" E. Januna Lake's latitude is 20.4785° N and longitude is 77.0399° E. The study was conducted from January 2020 to March 2020 in an area around Khamgaon.

Geographical Location of the Study Area

Material and Methods

Collection of Moths: Most of the moths were attracted by the light trap technique, which



involved using actinic tubes and mercury bulbs of about 20 to 125 watts. Baiting techniques such as sugaring and the use of fruit pulp are also successful. But the most suitable method is the sheet method. The white cloth sheet was used to attract the moth along with a bright light source. A light trap was also set during the 6–9 pm time period using a 160 watt mercury vapour bulb over a 3×3m (square) white cloth sheet that was hung between two vertical poles.

The moths collected were killed by ethyl acetate and later pinned to an insect stretching board. All specimens were preserved in an airtight insect box with naphthalene balls as fuming moths. Each specimen was provided with a label indicating the locality and date of collection.

Moths were photographed, and colour images were created by using a Canon digital camera (Power Shot, SX160IS, 16x, 42x optical zoom) and a Nikon^{TM} D300 with a 105 mm macro lens or a Nikon^{TM} D60 with an 18-55mm lens.

Identification of Moth

The available literature was used to identify the moths, including Moore (1880–1840), Hampson (1891–1896), Bell and Scott (1937), Holloway (1983–2011), Kendrik (2002), and Kirti and Singh (2015–2016). The classification system used by van Nieukerken et al. (2011) was followed**.**

Result and Obervation

A comprehensive survey was carried out in various habitats in the region to study the diversity and distribution of moths. This survey was carried out from June 2020 to August 2022 in and around the study area while studying the biodiversity of moth fauna in Khamgaon city and its surrounding area. A total of 13 species belonging to 5 families like Crambidae, Sphingidae, Erbidae, Noctuidae, and Geometridae were recorded in the present work. Among the members of the family Erebidae, they were predominant in the collection and had high species richness. All the identified species are listed in Table 1. Diversity indices were calculated using Past3 software, which showed Fisher's alpha, Shannon index, evenness, and species richness of family. The monthwise distribution of species is given in Table 2, and the percentage of species distribution in different families is given in Table 3.

Graphs plotted with the help of MS-Ecxel, in which Grap 1 shows the monthwise distribution of species, Graph 2 shows species richness in families, and Graph 3 shows species percentage

in families. Photographs of all 13 identified moth species are also provided, along with the scientific name, by using a smartphone camera.

Table 1: Family wise distribution and list of Moth General and identified species recorded from study area.

Sr. No.	Species	Location	Month
1.	Hymenia perspectalis	College campus	June
2.	Diaphnia Indica	Residential area	June
3.	Lascoria ambigualis	College campus	June
4.	Sphingomorpha chlorea	Januna Lake	June
5.	Orvasca subnotata	College campus	July
6.	Pleuroprucha Insularia	College campus	July
7.	Daphnis nerii	Residential area	July
8.	Achea Janata	Januna Lake	July
9.	Olepa ricini	Residential area	July
10.	Leuconycta Diphteroids	Residential area	August
11.	Iridopsis species	College campus	August
12.	Hyphantria Caunea	Residential area	August
13.	Mythimna unipuncta	Residential area	August

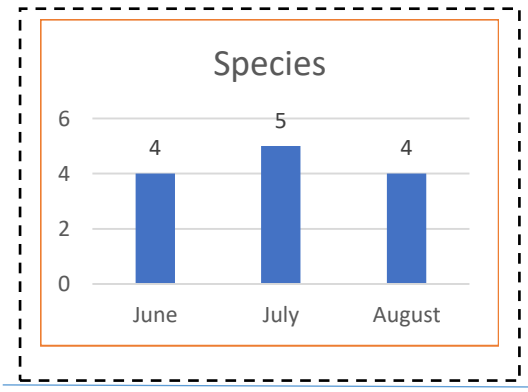
Table 2: Showing month wise species distribution.

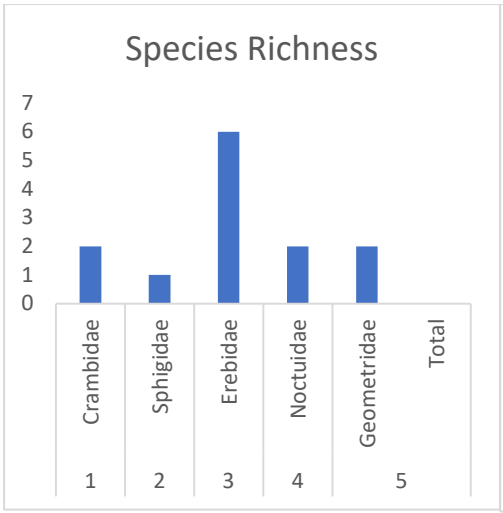
Sr. no.	Name of species	Family	Subfamily	Genus
1	Hymenia perspectails	Crambide	Spliomelinae	Hymenia
2	Diaphania indica	Crambide	Spliomelinae	Daphania
3	Daphnis nerri	Sphingidae	Macro glossinae	Daphnis
4	Lascoria ambigualis	Erebidae	Herminiiae	Lascoria
5	Sphingomorpha cholorea	Erebidae	Erbinae	Sphingomorpha
6	Achaea janata	Erbidae	Arctiinae	Achaea
7	Olepa ricini	Erbidae	Arctiinae	Olepa
8	Orvasca subnotata	Erbidae	Arctiinae	Orvasca
9	Hyphantria caunea	Erbidae	Noctuoidea	Hyphantria
10	Mythimna unipuncta	Noctuoidea	Noctuoidea	Mythimna
11	Leucpnycta diphteroids	Noctuoidea	Condicinae	Leucpnycta
12	Iridopsis	Geometridae	Ennominae	Iridopsis
13	Pleuroprucha insularia	Geometridae	Sterrhinae	Pleuroprucha

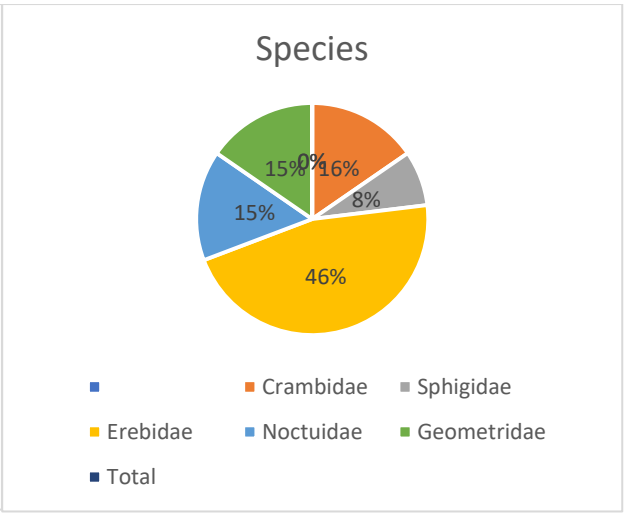
Table 3: Percentage of species distribution in different family.

Sr.no	Family	Species Richness	Percentage
1	Crambidae	2	15.38%
2	Sphigidae	1	7.69%
3	Erebidae	6	46.15%
4	Noctuidae	2	15.38%
5	Geometridae	2	15.38%
	Total		100

Graph 1 : Monthwise Distribution of Species







Photograph of 13 Identified Moth Species

Hymeniaperspectalispers

Sphingomorpha chlorea

Olepa ricini

Diaphania indica

Orvasca subnotata

Achaea

Hyphantria caunea

Mythimna unipuncta

Leuconycta

dominant family, which was represented by six species. This family was followed by Crambidae 02 species, followed by Sphingidae 01 species. Followed by Noctuidae and Geometridae 02 species each.

Conclusion

Reference

In this study, we have attempted to study the diversity of moths in Khamgaon City, Buldana Region. This work adds to the inventory of moths in this region, which could be utilised for future studies.

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Chemical Composition of Different Meat sources from Amravati City, Maharashtra

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Abstract:
The present study aimed to investigate a comparative analysis of the chemical composition of different meat sources available as food sources in Amravati city. As for the study, 50 meat samples were used, 10 from each of the *Capra aegagrus hircus, Dendrobranchiata, Coturnix coturnix, Gallus gallus domesticus, and Gallus gallus domesticus (Kadaknath)* species, provided that each of them belonged to a different animal. Moisture, ash, and crude protein levels of the meat samples were compared between meat sources. The higher values of protein and moisture were determined in the *Gallus gallus domesticus (Kadaknath)* species ($P<0.001$). Also, Gallus *gallus domesticus* had the highest ash ratio ($P<0.001$). *Gallus gallus domesticus* (Kadaknath) meat was advised to be included in the diets of patients suffering from obesity or cardio-vascular diseases because of its high protein and lower fat content.
Keywords: moisture, ash, crude protein, meat.

Introduction:
As the global population continues to grow, there is a corresponding increase in the demand for food, and this puts pressure on agricultural systems to produce more food to meet the nutritional needs of a larger population. Meeting the nutritional needs of a growing population is not just about producing more food but also about producing diverse and nutrient-rich foods. Adequate nutrition is crucial for physical and cognitive development, especially in children, as well as for maintaining overall health throughout life.
Population growth can pose challenges to food security, defined as having access to sufficient, safe, and nutritious food to meet dietary needs. This requires not only increased food production but also equitable distribution, reduced food waste, and improved access to food for vulnerable populations. To overcome this lacuna, animal meat is widely used all over the world as a source of nutritional food. Animal products are highly demandable in nutrition and the livestock sector. Consumers' access to these products at lower costs and the amount of animal protein consumption in diets are important parameters that give information about the development level of countries **(Frunza *et al.*, 2023**). The Food Standards Australia New Zealand (FSANZ) Food Standards Code defines meat as 'the whole or part of the carcass of any buffalo, camel, cattle, deer, goat, hare, pig, poultry, rabbit, or sheep, slaughtered other than in a wild state, but does not include eggs or foetuses (New Zealand Gazzatte, 2003).
According to the OECD-FAO data for 2021, while meat consumption per capita was 35.2 kg in the world, this rate was 69.5 kg in developed countries and 27.6 kg in developing countries. The nutritional composition of meat can vary depending on the type of meat and its cut. However, in general, meat is a good source of high-quality protein and contains various essential nutrients. In recent years, the quality of the yields obtained from animals has gained importance, as has the quantity. A lump of quality meat should be soft, high in moisture, contain more muscle fibres than connective tissue, be pink in colour, and have a suitable aroma **(Kumar *et al.*, 2023; Nutautaite *et al.*, 2023).**
Moisture, crude protein, crude fat, and ash ratios constitute the chemical composition of meat. It varies according to the species of animal, genotype, sex, age, body condition score,

nutritional status, and muscle structure of the animal **(Ketoon *et al.*, 2014).** Henceforth, chicken meat is widely used due to its richness in protein and the availability of its sources.

The present investigation aimed to examine the chemical composition of different meat sources originating from goat, prawn, common quail, broiler chicken, and kadaknath chicken consumed in Amravati city in terms of nutrient content and to reveal their superior aspects compared to each other.

Material and Methods:

In the present study, a total of 50 meat samples, 10 from each of the *Capra aegagrus hircus*, *Dendrobranchiata*, *Coturnix coturnix*, *Gallus gallus domesticus, and Gallus gallus domestics* (Kadaknath) species, were used, provided that each of them belonged to a different animal. The tissue samples were collected from the weekly one-day market in Amravati city. The muscle and liver tissue were taken into the mortar and crushed with the help of a pestle. The ground muscle and liver tissue were then subjected to analysis of moisture **(APHA, 1998),** ash **(AOAC, 920.153),** and crude protein in the tissue **(the Kjeldahl method, AOAC, 928.08).**

Fig. No. 1: Collection of different meat samples for investigation of Moisture, Ash, and Protein

Statistical Analysis:

The findings of the research were studied with the SPSS 25.0 package program. A one-way analysis of variance was used to determine significant differences between the groups, and the significance level was determined to be 0.05.

Result and Discussion:

The chemical composition ratio of different meat sources from different species in Amravati City was analysed in the present investigation, given in Table No. 1.

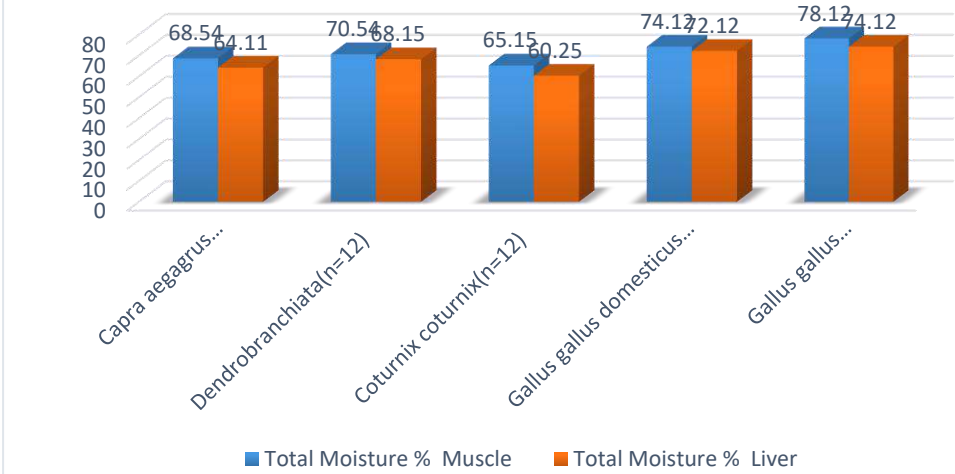
Table 1. Chemical composition values of meat samples obtained from different species of animals

Meat Sample	Total Moisture %		Total ash%		Total Protein	
	Muscle	Liver	Muscle	Liver	Muscle	Liver
Capra aegagrus hircus(n=12)	68.54± 0.56a	64.11± 0.52a	1.60± 0.08a	1.34± 0.06a	24.68± 0.52a	26.54± 0.52a
Dendrobranchiata(n=12)	70.54±0.58b	68.15±0.52b	1.30± 0.07b	1.80± 0.11b	26.14± 0.48b	27.24± 0.62b
Coturnix coturnix(n=12)	65.15±0.52b	60.25±0.50b	1.68± 0.6b	1.90± 0.9b	27.26± 0.68ab	28.15± 0.65ab
Gallus gallus domesticus (Broiler) (n=12)	74.12± 0.66c	72.12± 0.62c	2.40± 2.8ac	3.48± 0.03ac	26.80± 0.70c	28.26± 0.56c
Gallus gallus domesticus(Kadaknath) (n=12)	78.12± 0.69b	74.12± 0.65b	2.23± 0.20b	3.20± 0.32b	28.67± 0.65c	29.25± 0.57c
P	***	***	***	***	***	***

a, b, c: Values within a column with different superscripts differ significantly at P<0.05, ***: P<0.001

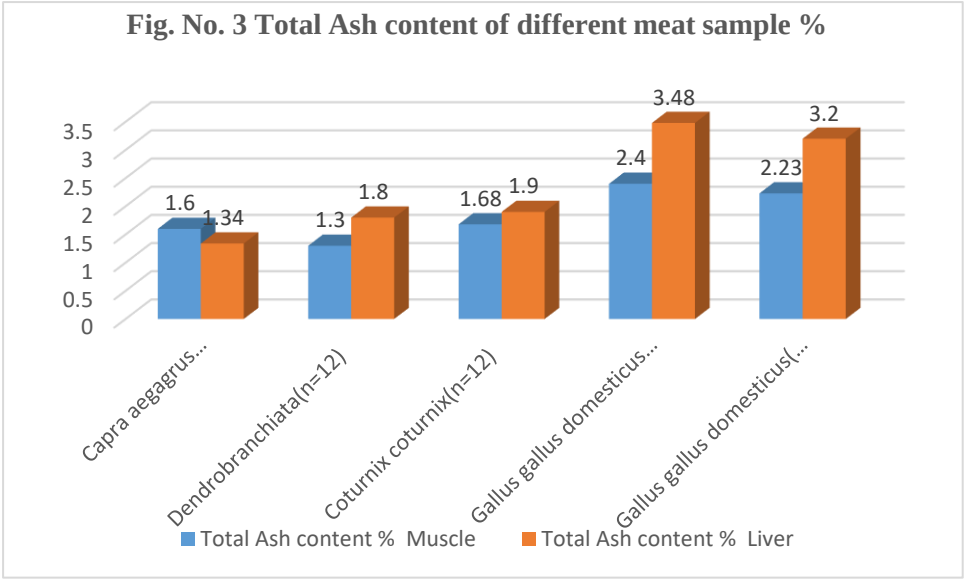
In the present analysis, the highest moisture content observed in ***Gallus gallus domesticus(Kadaknath)*** (P<0.001) as compared with other species of meat samples was lower in *Capra aegagrus hircus, Dendrobranchiata, Coturnix coturnix, and Gallus gallus domesticus,* and no significant difference was found between them (Fig. 2). Water is an important thermoregulatory medium and solvent, as well as playing important roles in cell and organ metabolism and the transport of metabolites and wastes (**Ketoon *et al., 2014)***. The water-holding capacity of the meat is an important factor affecting the appearance, colour, tenderness, taste, and aroma of the meat (**Apple and Yancey, 2013**). Water is essential for the retention of nutrients in meat. Proper hydration helps preserve the nutritional value of the meat, ensuring that essential vitamins and minerals are retained. Meat that does not lose its water depending on the cooking methods and can keep its content is evaluated in the quality meat category **(Belichovska et al., 2017; Lima et al., 2022**).

Fig. No. 2 Moisture content of Different meat Samples %



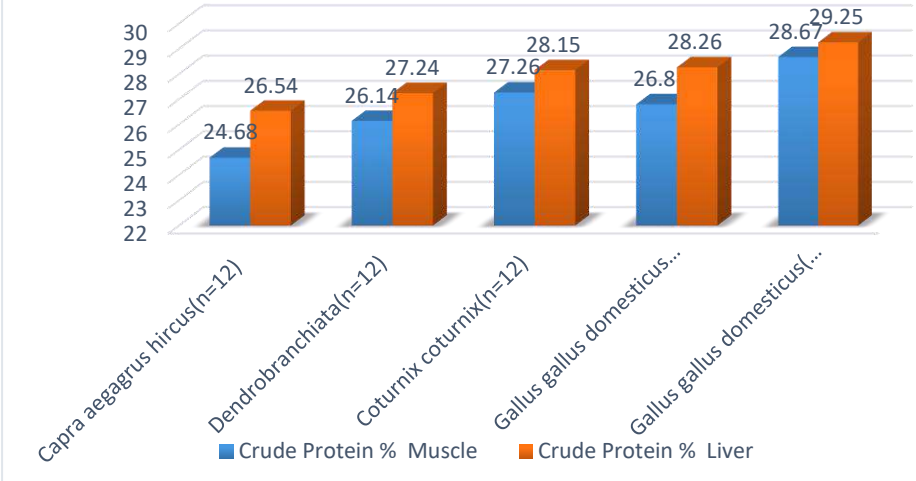
During the investigation of the ash ratio in the studied meat samples, the highest value was determined for *Gallus gallus domesticus* (Figure 2); the *Coturnix coturnix* ash ratio was found to be lower than *Gallus gallus domesticus(Kadaknath)* meat (P<0.001). There was no significant difference between *Coturnix coturni* and *Gallus gallus domesticus (Kadaknath)* meat in terms of ash content, as shown in Table 1. Ash in meat consists of minerals such as calcium, phosphorus, potassium, magnesium, sodium, and trace elements like iron and zinc. These minerals are essential for various physiological functions in the human body, including bone health, nerve transmission, and enzymatic processes. There are also mineral substances stored in body fluids (iron, sodium, and potassium), enzymes (zinc), and nucleotides (phosphorus) (**Ketoon et al., 2014).** Because mineral deficiencies cause significant discomfort in the body, it is very important to take them from food sources (**Williams, 2007; Soriano-Santos, 2010; Pereira and Vicente, 2013; Romero-Bernal et al., 2017**).

Fig. No. 3 Total Ash content of different meat sample %



While during the study of crude protein, the highest protein content was found in the *Gallus gallus domesticus (Kadaknath)* species in muscle and liver tissue as compared with other species of meat samples taken for the study ($P<0.001$). Protein is an essential macronutrient that the body needs for various functions, including tissue repair, muscle building, and enzyme production. Consuming meat, a rich source of high-quality protein, helps meet the body's protein requirements. Protein content in meat is influenced by many factors, such as species, genotype, age, gender, and ration composition. Animals with the same genotype may have different nutrient contents. (Pereira and Vicente, 2013; Marangoni et al., 2015). In human nutrition, proteins of animal origin have an important place in terms of essential amino acids and fatty acids. It is generally desirable to have a higher protein content in meat products (**Akçapınar and Ozbeyaz, 2021).** The rates found for chicken meat were higher than the reported values **(Cullere and Dalle Zotte, 2018; Fathi et al., 2023**).

Fig. No. 4 Crude Protein of differnt meat sample %



Discussion:

The meat sources constitute one of the major sources of nutrition for the rural population. The nutritional value of different meat sources depends on their biochemical composition, like protein, moisture content, ash, minerals, vitamins, etc. Protein is an essential macronutrient that

the body needs for various functions, including tissue repair, muscle building, and enzyme production. Consuming meat, a rich source of high-quality protein, helps meet the body's protein requirements.

As a result of the present investigation, the different meats of various species vary in moisture, ash, and protein levels. Taking into consideration the identification of higher levels of protein, moisture, and ash in different meats helps suggest the consumption of nutritional food by the people of the Amravati region.

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Diversity of nocturnal insects in some villages (Khalar, Akoli ruprao, Selapur) of Amravati District, Maharashtra

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Abstract:
In the present study the diversity of nocturnal insects in selected sites of rural areas (Khalar, Akoli Ruprao, Selapur) of Amravati district of Maharashtra was observed. The specimen collections were carried out between October 2022 to November 2022. The specimens were trapped once a week between 7:00 pm to 6:00 am. Light traps and manual methods were used for the collection. In the present findings total of 24 species were collected which belonged to 9 orders from all sites.

Kyewords: Diversity, Nocturnal insects, Light trap.

Introduction:

Insects are a diverse and highly successful group of invertebrate animals belonging to the class Insecta within the phylum Arthropoda. They represent the largest and most diverse group of organisms on Earth, with over a million described species and an estimated 10 million total species, making up about 80% of all known animal species. The number of actually described insect species in the world stands at 965,431–1,015,897 **(Chapman, 2009).** Insects are not only fascinating from a biological standpoint but also have significant ecological, economic, and cultural importance. Insects are the dominant component of biodiversity in terrestrial ecosystems and play important roles in ecosystem processes **(Weisser and Siemann, 2004).**

The world of insects is incredibly diverse, and this diversity manifests in numerous ways. Here are some key variations in insects across various aspects such as seasonal variability, Size, dietary diversity, mobility, strategy, and habitats. Insect communities constitute an integral part of terrestrial ecosystems because of the diversity of both the species and life forms **(Adjaloo *et al.*, 2012).** The total number of extant species is estimated at between six to ten million, potentially over 90% of the animal life forms on earth are insects. Insects dominate many food webs and food chain lengths **(Sugihara *et al.*, 1997)** and have great importance because of their diversity, ecological roles, and influence on agriculture, natural resources, and human health **(Foottit and Adler, 2009).** The class includes 30 orders with a variable number of species: the less diverse include the Mantophasmatodea (gladiators, 24 spp.) (**Zompro *et al, 2002*: Damgaard *et al.*, 2008**), Grylloblattodea (ice crawlers, 32 spp.) (**Wipfler *et al.*, 2014**), and Zoraptera (angel insects, 30 spp.) (**Mashimo *et al.*, 2014**); whereas the more diverse comprise the Dipteran (flies, 100,000 spp.). Hemiptera (bugs, 100,000 spp.), Hymenoptera (wasps, bees, ants, and sawflies; 120,000 spp.) Lepidoptera (butterflies and moths, 150,000 spp.) and of course, the Coleopteran (beetles, 370,000 spp.) (**Capinera 2008**).

So the present study focuses on investigating the varieties of insects from different orders at some selected villages (Khalar, Akoli Ruprao, Selapur) of Amravati District Maharashtra.

Material and Methods:

In the present exploration, the light trap method is used for the collection of insects. Light trapping is an effective means of assessing species composition and relative abundances, but sampling has to be carried out all night to maximize catch size and avoid biases due to different flight times of species **(Beck and Linsenmair, 2006**) With the help of light source

and the polythene sheet which was hanging on the pole, the insects are attracted towards the light source and captured on the sheet. The cotton swab dipped in benzene was put in the killing bottle where insects are collected by beating tray aspirator, and forceps. Insects were captured in the following months from November, December, and January at different intervals of time.

All the collected insects were anesthetized in a killing bottle and then proceeded to dry preservation overnight by using of 170w mercury bright light. The average temperature ranged from 25–30 °C maximum and 15–20°C minimum during the collection period. The collected insects were then labelled, and sorted order-wise. The labelled dried specimens were kept in the storage box with a small amount of flake naphthalene so that they were protected from other insects.

Observation and Results:

During the present investigation the total 9 orders of insects are found *Coleoptera, Orthoptera, Mantodea, Hemiotera, Lepidoptera, Diptera, Blattodea (Acheta, Domesticus, Branchinus Crepitans, Teleogryllus commodus, Scarbaeian, Dytiscus latissimus, G. Portentosa, Heterochychus aerator, Mantis religiosa, Platymeris biguttatus, Platymeris biguttatus, S. americana, T. infestans, S. granarious, N. hexadae, G. Campestris, C. hyaline, Chalenius grosser, M.domestica, N. appendicullata.).* The highest number of *Coleoptera* were collected from November to December 2022. Many *Coleoptera* species are highly specific to certain host plants where they lay their eggs, and their larvae feed. The availability of these specific plants directly influences the presence and abundance of particular Lepidoptera species in a habitat.

Mainly collected insects are identified and classified at genus level and the rest are identified at a family level below under classifies as follows

Table No. 1 Collected insects Classification and identification.

Classification	**Figure 1**	**Fig. 2**	**Fig. 3**	**Fig. 4**	**Fig. 5**	**Fig. 6**	**Fig 7**	**Fig. 8**	**Fig. 9**	**Fig. 10**
Order	*Orthoptera*	*Hemiptera*	*Coleoptera*	*Orthoptera*	*Orthoptera*	*Orthoptera*	*Coleoptera*	*Diptera*	*Diptera*	*Orthoptera*
Family	*Acrididae*	*Reduviidae*	*Carculionidae*	*Gryllotalpidae*	*Gryllioidea Laicharting*	*Acrididae*	*Carabidae*	*Muscidae*	*Tipulidae*	*Gryllidae*
Genus	*Schistocerca*	*Triatoma*	*Sitophilus*	*Neocurtilla*	*Gryllus*		*Chlaenius*		*Nephrotoma*	*Teleogryllus*
Species	*S.americana*	*T.infestans*	*S.granarious*	*N.hexadae*	*G.campestris*	*C. hyalina*	*Chlainius Grosseri*	*M.domestica*	*N. appendiculata*	*Teleogryllus comfortable*

Table No. 2 Collected insects Classification and identification.

Classification	**Fig. 11**	**Fig.12**	**Fig. 13**	**Fig. 14**	**Fig.15**	**Fig.16**	**Fig. 17**	**Fig.18**	**Fig.19**	**Fig.20**
Order	*Coleoptera*	*Coleoptera*	*Coleoptera*	*Blattidae*	*Blattodea*	*Coleoptera*	*Orthoptera*	*Coleoptera*	*Coleoptera*	*Mantodae*
Family	*Scarbaeiformia*	*Dytiscidae*	*Tenebrionidae*		*Blaberidae*	*Carabidae*	*Tenebrionidae*	*Crabidae*	*Scarabacidae*	*Mantidae*
Genus	*Ochicanthan*	*Dytiscus*	*Alphitobius diaperinus*	*Blattaria*	*Gromphadorhina*	*Branchinus*	*Tenebrio*	*Harealus*	*Hetero vcnus*	*Religiosa*
Species	*Scarbaeian*	*Dytiscus*	*Alphitobius*	*Gregarious*	*G. portentosa*	*Crepitans*	*Alphitobius*	*H.pensylvanicus*	*Arator*	*Mantis*

Table No. 3 Collected insects Classification and identification

Classification	Fig. 21	Fig.22	Fig. 23	Fig. 24
Order	*Hemiotera*	*Lepidoptera*	*Lepidoptera*	*Lepidoptera*
Family	*Reduviidae*	*Sphingidae*	*Arebidae*	*Geometridae*
Genus	*Platymaris*	*Daphnis*	*Chionarctia*	*Earophila*
Species	*P. biguttatus*	*D. nerii*	*C. nivea*	*A. Badiata*

Fig.1

Fig. 2

Fig.3

Fig. 4

Fig. 5

Fig.6

Fig. 7

Fig.8

Fig.9

Fig.10

Fig.11

Fig. 12

Fig.13

Fig.14

Fig. 15

Fig.16

Fig.17

Fig. 18

Fig. 19

Fig. 20

Fig.21

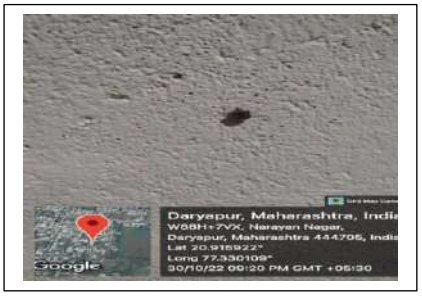

Fig.21

Fig.22

Fig. 23

Discussion:

In the present investigation based on the survey are total of 24 species of 12 orders are found. Mainly the species are collected in November 2022. Species diversity and seasonal abundance of fruit-piercing moths were carried out from different localities in Tamil Nadu. They observed five species of fruit-piercing moth belonging to two genera **(Ramkumar 2010).**

Comprehensible surveys of moth diversity have been done in Hawaii **(Zimmerman 1948**) and on larger continental islands in New Zealand (**Hudson 1928**), and (**Holloway 1976).**

Insects represent an incredibly diverse group of organisms, with estimates suggesting there are millions of insect species, and possibly many more yet to be discovered. This diversity plays a crucial role in various ecological processes and has significant implications for the health of ecosystems and even human well-being. Studying insects helps to understand biodiversity and ecological processes, and can contribute to pest control strategies and conservation efforts. The diversity of insects is essential for the health and functioning of ecosystems, and understanding and conserving this diversity is crucial for the well-being of the planet.

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Diversity of Spiders in Agricultural Fields of Tahsil Morshi, District Amravati. (M. S.)

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ABSTRACT

Spiders are one of the most diverse animal groups in the World. Spiders are widespread and diverse predators that are part of terrestrial Arthropod assemblages. Spiders play an important role as stabilizing agents or regulators of insect populations in agro and other terrestrial ecosystems. Thus their presence in an ecosystem may well influence the population dynamics of other arthropods present. Spiders play an important role in insect pest control without any harm to agricultural fields. Recently in agricultural fields reduced pesticide use and ecological sustainability have lead to increased interest in spiders as potential natural biological pest control agents. Insect populations rise significantly when they are freed from spider predation. insecticides are often used in agricultural areas, which lowers the number of spiders.

Spider species abundance in agro-ecosystem can be high as undisturbed natural ecosystem. Spiders act as pest control creature, which feeds on crop destructive insects. Spiders are beneficial bio-control agent of insect pest in agro-ecosystem. A survey of Spiders was carried out in Agricultural Fields of Morshi, District Amravati during July 2023 – December 2023. During the present study I have reported 138 species of Spiders belonging to 14 Families and 60 genera. Spiders of Families Araneidae, Eresidae, Gnaphosidae, Hersilidae, Lycosidae, Oxyopidae, Philodromidae, Saltisidae, Scytodidae, Sparassidae, Tetragnathidae, Theridiidae, Thomisidae, and Uloboridae were recorded during the investigation. This article presents a study on the distribution and current status of spider families in these agricultural fields of Morshi, district Amravati.

Keywords: Diversity, Agricultural Fields, Spiders, Morshi.

Introduction:

Spiders are among the most abundant insectivorous predators of Terrestrial ecosystem. Spiders are one of the most diverse animal groups in the World. Spiders are carnivorous creature. Spider plays an important role in regulating insect pests in the Agricultural Ecosystem. They mostly feed on insects, even though they may also feed on various other kinds of prey. There are 43,426 spider species are found all over the world in almost every kind of habitat. They mainly prey on insects, even though they may also feed on various other kinds of prey.

Spiders are beneficial to human beings in the sense that they feed not only on the pests of agro ecosystem but also the pest of man such as cockroaches, flies, Mosquitoes. In households, a particular spider as the giant crab spider has been known as an effective in controlling cockroaches and other insect pests found in the domestic environment.

They have usually been treated as an important biological control agent, because there is ecological role of spiders in pest control. Use of chemical pesticides has killed natural predators in the agro ecosystems and also disturbing the natural fauna. Several toxic insecticides and pesticides are recommended to control pests in agricultural field. These chemicals insecticides and pesticides are destroying the vegetation.

The constant use of a wide range of pesticides has caused many side effects, like loss of biodiversity, the problem of secondary pests, insecticide resistance, residual toxicity, the

recovery of insect pests and Environmental Pollution. Spiders consume a large number of small creatures and do not injure vegetation

Material and Method:

A survey of Spiders was carried out in Agricultural Fields of Morshi Tahsil, District Amravati during July 2023 to December 2023. Spiders were collected from different areas of Agricultural Fields. For collection and studying of spiders direct searching, collected by Insect nets, Pit fall trapping, beating steak and umbrellas were used. The Spiders Specimens were identified according to Kaston spider book. The photographs were taken in different views, to get the clear eye position, pattern and shades of cephalothorax and abdomen, spines and hairs pattern.

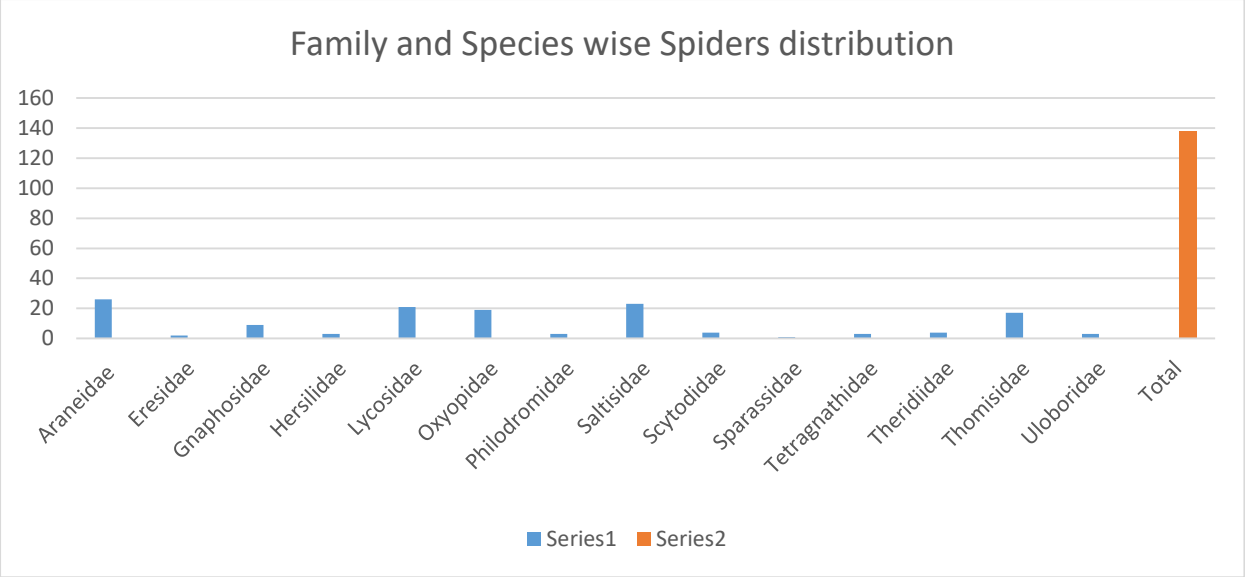
Observation and Result:

During the present study I have reported **138 Species** belonging to **14 Families** and **60 Genera of Spiders in Agricultural fields of Morshi Tahsil, District Amravati, Maharashtra State.** Spiders of Families Araneidae, Eresidae, Gnaphosidae, Hersilidae, Lycosidae, Oxyopidae, Philodromidae, Saltisidae, Scytodidae, Sparassidae, Tetragnathidae, Theridiidae, Thomisidae, and Uloboridae were recorded during the investigation. For details I have arranging the data in a Table Format of systematic way.

In my investigation I have seen that the abundance of Five Family Spiders species were more. The Orb waver spiders of Family Araneidae and Jumping spiders of Family Salticidae are widely distributed. The Orb waver spiders of Family Araneidae form web and the insect pest entangled in web spiders feeds on them. The Members of Salticidae directly feeds on insect Pest. **Araneidae>Salticidae>Lycosidae>Oxyopidae>Thomisidae**

Sr. No.	Family	Genera	Species
01	Araneidae	13	26
02	Eresidae	01	02
03	Gnaphosidae	04	09
04	Hersilidae	01	03
05	Lycosidae	09	21
06	Oxyopidae	08	19
07	Philodromidae	02	03
08	Saltisidae	10	23
09	Scytodidae	02	04
10	Sparassidae	01	01
11	Tetragnathidae	01	03
12	Theridiidae	02	04
13	Thomisidae	05	17
14	Uloboridae	01	03
Total		**60**	**138**

Table 1: Genus and Family wise distribution of Spiders in Agricultural Fields of Morshi, District Amravati.



Graph 1: Family and Species wise distribution of Spiders in Agricultural fields of Morshi, district Amravati.

Conclusion:

Spider's predatory capacity can have an effect in decreasing densities of insect pests, when they are used to balance the effect of insecticides and Pesticides. Some spiders are among the most effective predators of leafhoppers, caterpillars, and other pests. Aphids are rarely important pests of Cotton. Some Spiders and Spider lings are main control agents of aphids. Due to destroying the pest or insects, spiders are friends of farmer. Most spiders feeds on insects that's why productivity of crop gets increased, hence spiders are important Pests control agents. The present work includes the Taxonomic position and list of diversified species of spiders. The major families abundant in these agricultural fields are **ARANEIDAE 26, SALTICIDAE 23, LYCOSIDAE 21, OXYOPIDAE 19 and THOMISIDAE 17.**

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Water Quality Study: Physico-Chemical Characteristics of Takli Lake, Yavatmal (M.S.), India

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ABSTRACT
In the present study, the physico-chemical characteristics of lake water were studied. The duration of the study was from October 2023 to December 2023. Two sampling sites were selected for the study. Water samples were collected from two different locations in Takli Lake. This study involves the determination of the physical and chemical parameters of lake water. The physical parameters studied were TDS, temperature, electrical conductivity, and chemical parameters like pH, total alkalinity, chloride, and dissolved oxygen.
Key words: physico-chemical, pH, conductivity, TDS, alkalinity

INTRODUCTION
Water is life; thus, life cannot be imagined without water. Water is one of the most important natural resources available to all living organisms and supports human development and economic diversity. Water is used for drinking, agriculture, domestic use, and many other purposes. The demand for water is increasing with the increasing population. In India, the percentage of rainfall is decreasing; hence, the good quality of water is becoming essential for mankind.

Water is known to contain a large number of chemical elements. The interaction of both the physical and chemical properties of water plays a significant role in the composition, distribution, and abundance of aquatic organisms. Various water-borne diseases are making life difficult. Water pollution is an acute problem all over India; hence, it is essential to study the quality of this lake water.

Yavatmal is one of the eleven districts of the Vidarbha region of Maharashtra. It is bounded on the east by Chandrapur district, on the south by Andhra Pradesh state and Nanded district, on the west by Washim and Hingoli districts, and on the north by Amravati and Wardha districts. Yavatmal district belongs to the Balaghat range. It is bounded by the main rivers, Wardha and Penganga.

Materials and Methods
Water samples were collected in previously sterilized polythene bottles. All the water samples were collected during October-2023 to December-2023 from two sites for Takli Lake in the morning (9:00am to 11:00am). Temperature, electrical conductivity, total dissolved solids, and pH of the water samples were measured immediately after the collection of the samples because the values to be measured may change significantly in a matter of minutes. Temperature, pH, TDS, and electrical conductivity are measured with the help of a digital thermometer, conductometer, TDS meter, and pH meter. Other physico-chemical parameters were analyzed in the laboratory.

Results and Discussion
Temperature: Temperature is mainly related to the atmosphere and weather conditions. Temperature effects certain chemical and biological activities in aquatic organisms. The temperature is in the range of 27.8 degrees Celsius to 30.7 degrees Celsius.

pH: The pH values range from 7.00 to 8.12. This range of pH indicates the basic nature of water.

Electrical Conductivity (E.C.): Electrical conductivity varied from 567 to 878 microsiemens/cm. It measures the dissolved electrolytes.

Dissolved Oxygen: Dissolved oxygen is one of the most important parameters for the assessment of water quality, as it indicates the physical and biological processes prevailing in the water. Non-polluted water is generally more saturated with dissolved oxygen. Dissolved oxygen was observed from 4.57 to 5.37 mg/L.

Chloride: Chloride is practically found in all natural water. Chloride is one of the most common inorganic anion present in water. Man and animals excrete a high quantity of chloride. Salts that are present in the soil are also the source of chloride. Chlorides in the water samples were found to be 47.3 to 49.7 mg/L.

Total Dissolved Solids (TDS): The total dissolved solids of water are expressed by the weight of residue left when a water sample has been evaporated to dryness. The mean value was observed at 390 mg/L to 2390 mg/L.

Total Alkalinity: maximum value recorded at site 1 and minimum value recorded at site 2; both values are above the desirable ranges as prescribed by the BIS drinking water standards.

CONCLUSION

The results show a comparison of the present study parameter values within the permissible limits as prescribed by the BIA (Bureau of Indian Standards), but some parameters give alarm for the protection of water from pollution. For drinking purposes, it should be filtered and then used. For agriculture and industry purposes, it can be used directly.

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Impact of Invasive Species on Fish Fauna in and Around Morshi Taluka, Amravati District, Maharashtra, India

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Abstract:
Invasive species have become a significant threat to ecosystems worldwide, including aquatic environments. This research paper investigates the effects of invasive species on the fish fauna in and around Morshi Taluka, located in the Amravati District of Maharashtra, India. Through a comprehensive literature review and field studies, this paper examines the ecological impacts of invasive species, their mechanisms of invasion, and the repercussions on native fish populations. Additionally, it explores management strategies and recommendations to mitigate the adverse effects of invasive species on the aquatic ecosystem in the region.

Introduction: Invasive species are non-native organisms that establish and proliferate in ecosystems beyond their natural range, causing detrimental impacts on native biodiversity, ecosystem functions, and human activities. The aquatic ecosystems of Morshi Taluka, situated in the Amravati District of Maharashtra, are vulnerable to the introduction of invasive species due to various anthropogenic activities such as aquaculture, trade, and recreational activities. Understanding the effects of invasive species on fish fauna is crucial for devising effective management strategies to conserve native biodiversity and maintain ecosystem integrity in the region.

Amravati District in Maharashtra, India, is known for its rich biodiversity and diverse aquatic ecosystems, including numerous lakes and water bodies that support a variety of fish species. However, in recent years, the region has been facing significant challenges due to the introduction and spread of invasive species of fish fauna. Among the affected areas is Morshi Taluka, which has witnessed profound ecological disruptions attributed to these invasive species. The introduction of invasive fish species into the water bodies of Morshi Taluka has triggered a cascade of detrimental effects on the native aquatic fauna, habitats, and overall ecosystem health. These invasive species often outcompete native fish for resources such as food, shelter, and breeding grounds, leading to a decline in native fish populations. Moreover, they can alter the structure and function of aquatic ecosystems, causing disruptions in nutrient cycling, water quality, and habitat availability. Invasive fish species are known to have profound socio-economic impacts as well, affecting local fisheries, aquaculture practices, and traditional fishing communities that depend on native fish species for their livelihoods. The invasion of these species may also have repercussions on recreational activities such as angling and ecotourism, which contribute to the local economy. Furthermore, invasive fish species can pose significant threats to human health by harboring parasites and pathogens or by altering the transmission dynamics of diseases within aquatic ecosystems. Additionally, some invasive species may exhibit aggressive behavior or have venomous spines, posing risks to human safety during recreational activities or fish handling. In light of these challenges, understanding the dynamics of invasive fish species in and around Morshi Taluka is crucial for developing effective management strategies to mitigate their impacts. This requires interdisciplinary research efforts encompassing ecology, fisheries science, conservation biology, and socio-economic studies to assess the extent of invasion, identify key drivers of invasion, and devise appropriate management and control measures. Therefore, this study aims to provide a

comprehensive analysis of the effects of invasive fish species on the aquatic ecosystems of Morshi Taluka, highlighting their ecological, socio-economic, and public health implications. By shedding light on the magnitude of the problem and exploring potential management strategies, this research endeavors to inform policy-making and conservation efforts aimed at preserving the biodiversity and ecological integrity of the region's aquatic ecosystems.

Invasive Species:

Concepts and Characteristics: Invasive species possess certain characteristics that enable them to outcompete native species and thrive in new environments. Factors such as rapid reproduction, high dispersal ability, phenotypic plasticity, and a lack of natural predators contribute to their invasive success. Mechanisms of invasion include introduction through trade pathways, accidental release, deliberate introduction for biological control, and range expansion due to climate change. Fish Fauna in Morshi Taluka: Morshi Taluka supports diverse fish species, including both indigenous and exotic varieties. Native fish fauna play essential roles in ecosystem functioning, including nutrient cycling, prey-predator dynamics, and the maintenance of ecological balance. However, these populations face threats from habitat degradation, pollution, overfishing, and the introduction of invasive species. **Invasive Species in Morshi Taluka:** Several invasive species have been documented in the water bodies of Morshi Taluka, posing significant threats to native fish populations. Common invasive species include African catfish (Clarias gariepinus), common carp (Cyprinus carpio), and Nile tilapia (Oreochromis niloticus). These species often outcompete native fish for food and habitat resources, leading to declines in native fish abundance and diversity.

Ecological Impacts of Invasive Species: The establishment of invasive species can disrupt ecological processes and alter the structure and functioning of aquatic ecosystems. Competition for resources such as food and spawning sites can result in reduced fitness and reproductive success for native fish species. Predation by invasive predators can lead to population declines and changes in community composition. Habitat alteration by invasive species, such as vegetation removal and sediment disturbance, can further degrade ecosystem integrity.

Case Studies and Field Observations: Field surveys and observations conducted in Morshi Taluka provide empirical evidence of the impacts of invasive species on native fish populations. Comparative analysis of invaded and uninvaded sites reveals significant differences in fish community composition, with invaded sites exhibiting lower native species richness and abundance.

Management Strategies: Effective management of invasive species requires a combination of prevention, control, and eradication measures. Preventative measures include regulating the trade of potentially invasive species, implementing biosecurity protocols, and raising public awareness. Control and eradication efforts may involve mechanical removal, chemical treatments, biological control agents, and habitat restoration techniques.

Socio-Economic Implications: The presence of invasive species can have significant socio-economic implications for local communities dependent on fisheries and aquaculture for their livelihoods. Reductions in native fish stocks can lead to decreased income and food security for fisherfolk and aquaculture farmers. Furthermore, the costs associated with invasive species management and control efforts impose economic burdens on government agencies and stakeholders.

Ecological Disruption: Invasive species can disrupt the natural balance of ecosystems by outcompeting native species for resources such as food and habitat. This disruption can lead to a decline in native fish populations, affecting the overall biodiversity and ecological health of the region.

Economic Losses for Fisheries: If invasive species outcompete native fish species, it can result in reduced catches for local fishermen who rely on these fish for their livelihoods. This

decline in fish populations can lead to economic losses for the fishing industry, affecting both fishermen and related businesses in the supply chain.

Impact on Agriculture: Some invasive fish species may also have detrimental effects on agricultural activities. For example, certain species may disrupt irrigation systems or compete with native species for water resources, potentially impacting agricultural productivity in the region.

Health Risks: Invasive species can sometimes carry diseases or parasites that can affect native fish populations or even pose risks to human health if consumed. This can have implications for public health and food safety in communities that rely on fish as a dietary staple.

Cultural and Social Impacts: Fishing often plays a significant role in the culture and social fabric of communities, providing not only livelihoods but also recreational opportunities and traditional practices. The decline of native fish populations due to invasive species can disrupt these cultural traditions and social connections, leading to a loss of cultural identity and community cohesion.

Management Costs: Controlling and managing invasive species requires resources and funding, which can place a burden on local governments and conservation agencies. These costs may include measures such as monitoring, eradication efforts, and restoration projects aimed at mitigating the impacts of invasive species on the local ecosystem.

Tourism and Recreation: If invasive species negatively impact the natural beauty and biodiversity of the region, it could deter tourists and outdoor enthusiasts who visit the area for activities such as fishing, birdwatching, or ecotourism. This could result in losses for local businesses reliant on tourism revenue.

Addressing the socio-economic implications of invasive species requires coordinated efforts involving policymakers, scientists, local communities, and stakeholders. Strategies may include implementing invasive species management plans, promoting sustainable fishing practices, raising awareness about the impacts of invasive species, and supporting alternative livelihoods for affected communities.

Community Engagement and Awareness: Engaging local communities and stakeholders in invasive species management is crucial for fostering a sense of stewardship and promoting sustainable practices. Education and outreach programs aimed at raising awareness about the impacts of invasive species can empower communities to participate in monitoring and control efforts. Citizen science initiatives enable local residents to contribute valuable data on invasive species distributions and impacts.

Policy and Legislative Framework: Existing policies and regulations governing the management of invasive species in Maharashtra provide a basis for conservation efforts.

Conclusion: The proliferation of invasive species poses significant challenges to the conservation of native fish fauna and the integrity of aquatic ecosystems in Morshi Taluka, Amravati District, Maharashtra, India. Addressing these challenges requires concerted efforts from government agencies, researchers, local communities, and other stakeholders. By implementing effective management strategies and fostering community engagement, it is possible to mitigate the impacts of invasive species and safeguard the ecological and socio-economic sustainability of the region's aquatic resources.

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These references cover various aspects of the impact of invasive species on fish fauna in and around Morshi Taluka, providing valuable insights into the ecological dynamics and management implications in the region.

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Traditional Medicinal Plants Used For The Treatment Of Male Sexual Dysfunction – A Review

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ABSTRACT
Modern lifestyles and certain environmental exposures have resulted in male infertility, which is increasing in almost every part of the world. Modern medicines provide nutritional, physiological, and psychopharmacological treatments; however, many of them have a negative impact on physiological processes. Aphrodisiacs are substances that are used to increase libido and promote fertility. Herbal aphrodisiac provides a safer way to counteract various problems associated with male infertility. This review discusses the plant's biological name, common name, family, and the plant parts used to study that are useful for the researcher to formulate the aphrodisiac potential of the plant. Ethnobotanical surveys have indicated a large number of plants as aphrodisiacs. [16] The paper reviews the recent scientific validation of traditionally used medicinal plants as aphrodisiac herbs for the management of erectile dysfunction and other sexual disorders. The plants mentioned in this review have provided scientific evidence for their aphrodisiac activity.

Keywords: male infertility, aphrodisiacs, sexual disorders, erectile dysfunction

INTRODUCTION
Many plants growing locally are used as sources of medicines throughout the world. Some of these plants have pharmacological properties. Most of these plants have occupied an important place in traditional as well as modern medicine systems. These medicinal systems have great importance these days due to the side effects caused by synthetic drugs. **[2]** Many people rely on traditional medicine, mostly plant drugs, for their primary health care needs. India officially recognises over 3000 plants for their medicinal value. The products from these plants are used to cure many diseases. Of these, erectile dysfunction is one of the serious medical and social symptoms that occur in men. The problem of erectile dysfunction can be overcome by the use of substances known as aphrodisiacs. **[4]**

Male sexual dysfunction
Sexual relationships are among the most important social and biological relationships in human life. Sexual dysfunction in men is a serious medical and social symptom. It is the repeated inability to achieve normal sexual intercourse that may contribute to infertility. Male sexual dysfunction (MSD) affects not only sexual relationships but also the overall quality of life. ED and premature ejaculation (PE) are the two most prevalent male sexual complaints. Erectile dysfunction (ED), also called male impotence, is a common medical condition that affects the sexual life of men. Erectile dysfunction is the repeated inability to get or maintain a firm enough erection to allow sexual intercourse.

The normal male sexual response cycle can be functionally divided into five interrelated events that occur in a defined sequence: libido, erection, ejaculation, orgasm, and detumescence**.** **[4]**

- **Libido:** Libido is defined as the biological need for sexual activity (the sex drive) and frequently is expressed as sex-seeking behaviour.
- **Erection:** Erection is the enlarged and rigid state of the sexually aroused penis sufficient for vaginal penetration.

- **Ejaculation:** Ejaculation is the act of ejecting semen. It is a reflex action that occurs as a result of sexual stimulation.
- **Orgasm:** This is the climax of sexual excitement. The entire period of emission and ejaculation is known as the male orgasm.
- **Detumescence:** This is the subsidence of an erect penis after ejaculation.

Males with sexual dysfunction cannot attain the above events required for normal sexual intercourse, which subsequently leads to infertility.

Infertility Risk Factors [14]

1. **Cigarette smoke**: Sperm counts of smokers are on average 13–17% lower than those of non-smokers.
2. **Pesticides:** Exposure to pesticides results in a reduced sperm count and an increase in abnormally shaped sperm.
3. **Air pollution**: Men living in industrial and polluted towns have six times more abnormal sperm than those living in clean areas.
4. **Chemicals:** Sperm count drops in men exposed to chemicals like DDT, PCB's (polychlorinated biphenyls), dioxins, and some petroleum by-products.
5. **Food additives**: Food additives like monosodium glutamate (MSG) cause infertility.
6. **Anaesthesia:** Animals exposed to the anaesthesia enflurane show a 50% higher sperm damage rate than those not exposed to enflurane.
7. **Occupational exposure:** Men who work in the aircraft industry, textiles, plastic, welding, chemical solvents, or even antibiotics are more at risk of having abnormal sperm.
8. **Ozone affects:** As the level of ozone in ambient air increases, the sperm concentration goes down. Ozone, once inhaled, gets rapidly metabolised, triggering an inflammatory reaction that could adversely affect the sperm.
9. **Zinc deficiency:** Zinc is involved in every aspect of male reproduction, including hormone metabolism, sperm formation, and sperm motility. Zinc deficiencies are characterised by decreased testosterone levels and sperm counts.

Taking into consideration the above-mentioned circumstances, we reviewed some of the plants that can be used as aphrodisiacs.

Sr. No.	Scientific Name	Common Name	Family	Part Used	Reference
1.	*Aloe barbadensis*	Aloe vera	Asphodelaceae	Roots	11
2.	*Asparagus racemosus*	Shatavari	Liliaceae	Roots	1
3.	*Anethum graveolens*	Dill	Apiaceae		13
4.	*Aquilaria malaccensis*	Karas	Thymelaeaceae	Leaves	17
5.	*Blepharis sindica*	Bhangari	Acanthaceae	Seeds	9
6.	*Borassus aethiopum*	African fan palm	Arecaceae	Hypocotyl	8
7.	*Capsicum annuum*	Capsicum	Solanaceae	Seed	3
8.	*Carthamus tinctorius*	Safflower	Asteraceae		13
9.	*Chlorophytum tuberosum*	Safed Musli	Liliaceae	Whole Plant	3
10.	*Citrus aurantium*	Bitter orange	Rutaceae		13
11.	*Cocos nucifera*	Coconut palm	Arecaceae	Endosperm	13
12.	*Cordia dichotoma*	Indian cherry	Boraginaceae	Fruits	10
13.	*Coriandrum sativum*	Coriander	Apiaceae	Leaf	3
14.	*Crocus sativus*	Saffron	Iridaceae	Stigma	3
15.	*Cucurbita pepo*	Pumpkin	Cucurbetaceae	Seeds	3
16.	*Cymbopogon citrates*	Lemongrass	Poaceae	Whole Plant	3
17.	*Eriodendron anfractuosum*	White silk cotton tree	Bombaceae	Whole Plant	3
18.	*Erythroxylum*	Catuaba	Erythroxylaceae	Bark	5

	vacciniifolium				
19.	*Ficus carica*	Fig	Moraceae	Fruits	18
20.	*Glycyrrhiza glabra*	Liquorice	Fabaceae		13
21.	*Heinsia crinata*	Bush apple	Rubiaceae	Root Bark	6
22.	*Hibiscus rosa-sinesis*	China rose	Malvaceae	Leaf	3
23.	*Lagenaria vulgaris*	Bottle Gourd	Cucurbitaceae	Fruit	3
24.	*Mangifera indica*	Mango	Anacardiaceae	Bark	3
25.	*Melissa officinalis*	Lemon balm	Lamiaceae		13
26.	*Myristica fragrans*	Nutmeg	Myristicaceae	Seed	4
27.	*Nerium indicum*	Kaner	Apocynaceae	Roots	4
28.	*Nigella arvensis*	Wild fennel flower	Ranunculaceae		13
29.	*Phoenix dactylifera*	Date Palm	Arecaceae	Pollen	4
30.	*Pinus pinea*	Stone pine	Pinaceae		13
31.	*Prunus mahaleb*	Mahaleb cherry	Rosaceae		13
32.	*Psoralea corylifolia*	Babchi	Fabaceae	Seeds	7
33.	*Rhaphiostylis beninensis*		Icacinaceae	Roots	12
34.	*Ricinus communis*	Castor	Euphorbiaceae	Seeds	3
35.	*Rosa damascena*	Rose	Rosaceae	Petals	4
36.	*Santalum album*	Sandalwood	Santalaceae	Heart Wood	4
37.	*Sesamum indicum*	Til	Pedaliaceae	Seeds	4
38.	*Solanum melongena*	Brinjal	Solanaceae	Unripe Fruit	4
39.	*Zea mays*	Purple corn	Poaceae	Crude Extract	15
40.	*Zingiber officinale*	Ginger	Zingiberaceae	Rhizome	3

CONCLUSION

A number of herbs have been used to improve sexual performance. Thus, people are seeking herbal remedies and drugs instead of synthetic medicine to treat their sexual dysfunction. It can be stated that the use of herbal medicine and safer herbal products for improving sexual disabilities could provide better effects on male sexual dysfunction. The information recorded in this review is the plant's scientific name, common name, family, part used for the aphrodisiac activity, and reference. The plants mentioned in this review have significant aphrodisiac activity and can be effective with no or fewer side effects.

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The Study of Osmoregulation Mechanism in Fresh Water Fish

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Abstract: -

Fish are most important content of aquatic ecosystem not only source but also, they provide link between ecosystem and source of income and employment to many regions. It is best source of Omega-3 fatty acids osmoregulation imbalance is often caused by poorly throughout ant destructive fish activity. Osmoregulation is very important biochemical process that help fish maintain in their bodies salt and water balance and without mortality and increase survivability of fish because fresh water fish tolerate in water with salt concentration of less than 1.05% fresh water having low sale concentration. If increase the sale concentration direct affect the fish metabolism and imbalance osmoregulation. Main source of increase salt concentration is soil and rock of earth crust, precipitation cycle, show falling into water and evaporation salt are compound like Nacl, Mgso4, KNO3-, NaHCO3 which dissolve into ion (water). Calcium and magnesium require the hardness of water calcium can use are increase fish face the premature batching and calcification of larval skeletal oxygen consumption rate of fish was maximum at 2.5% in salt which gradually decrease with increase salt concentration from the result of the study. It can be estimated that increase sale concentration the survival rate, growth and tissue, decreased acid level intake and reproduction level decreased with increased in salt concentration. These results clearly indicate *Labeo rohita*, Calta, Mrigal, Tilapia fresh water fishes are not proper grow and survive to higher salt concentration.

Keywords: - Osmoregulation, Salt Concentration, Fish, Survive, Growth, Ecosystem, Fresh water

Introduction: -

Fish is often referred to as "Rich food for poor people" fish content high quality protein, low fat with omega-3 fatty acid and vitamin. Fish is a rich source of mineral like calcium, phosphorus, iron, iodine, magnesium and potassium fish contain good quality of protein, about 18-20% and all eight essential amino acid fat for fish contain the polyunsaturated fatty acid (PUFAS) namely EPA (eicosqpentaenoic acid and DHA doco sabesaqenoic acid which are essential for proper growth of children brain development vitamin D present in fish liver and oil is crucial for bone growth since it is essential for the absorption and metabolism of calcium. Fish is also called "Brain food" help development and function of brain and "Heart food as it contributes to lower risk of heart attack and strokes. Fish to make sure the level of water and salt do not become dilute or concentrate according to environment condition is an internal balancing process of fish is called osmoregulation.

Right concentration of solute and amount of water must be maintaining fresh water fish hence osmoregulation body become neither dilute nor become concentrated. When taking about osmoregulation 3 terms are important.

1) **Diffusion-** It is random movement of molecules along the concentration gradient from the region of high concentration to region of low concentration.

2) **Osmosis –** In osmosis selective diffusion of solvent is driven by internal energy of solvent molecule we can also say that osmosis it specialized case of diffusion that involve passive transport of solvent from its region of higher concentration to its region of lower concentration through a semipermeable membrane, water is naturally attracted toward the more sale.
3) **Osmotic pressure –** It is defined as pressure required to completely stop the entry of solvent of osmotically active solution on the basis of above describe termed and living cell can be present in three condition with respect to its external environment
4) **Isotonic Solution-** When concentration of ion of Cu is equal to concentration in external environment or in solution hypotonic solution when concentration of salt (ion) in cell is more than to concentration external environment
5) **Hypertonic solution –** When concentration of ion in a cell is less than concentration in external environment.

Osmoregulation process keep their internal water salt content stable. This stable content known as osmotic concentration (osmolarity) of body fluids, salinity is an important factor in determining many aspects of the chemistry of natural waters and of biological processes. Salinity is an ecological factor of considerable important, influencing the type of organisms that live in body of water. Sale are expensive to remove from water and content in an important factor in water use, factoring into portability and suitability of irrigation increase in salt have been in lakes and river in the united states due to common road sale and other salt decrease in run off salt within are selectively absorb through and internal membrane. Only some ion minerals and water can pass this internal barrier is called as semipermeable membrane. The fish internal body fluid is stronger solution than surrounding environmental solution. Mechanism of osmoregulation in fishes take place in kidney. Kidney is the main osmoregulatory organ in fishes but grill membrane also help in osmoregulation grill are an example of semipermeable membrane reduction of skin permeability to water and ions by secreting a thicker layer of mucus **(Shephard 1994).**

Process of osmoregulation in fresh water fishes. These fish are Rohu, Catla, Mahseer, Magur, Ritha Rani, Mystus, Tengara, Tilapia, Palasa fish, Hilra, Prawn fish. These fishes live in hypertonic environment their body have more salt concentration as compared to the surrounding water. (Fresh water may produce the equivalent of 30% of its total body weight in urine every day.) So, this fish faces the following problem.

Fluids through skin and gills membrane (endo-osmosis) ions (Na^+ & Cl^-) through diffused out through skin and gill's membrane fresh water fish produced large quantity of dilute urine mean lot of water and salt not in urine presence of large glomeruli in nephron which filter out surplus water salt reabsorption mechanism take place in kidney tubules. These fish do not drink water active uptake of salt loss of salt is compansatly by taking up Na^+ & Cl^- ion from surrounding water into their body by an active transport with the help of special cell. The ionorite and chloride cell are present in gill lamellae. Osmoregulation depend on active transport cross the skin and gills and concluded that the different ion composition of the extracellular or intracellular body fluids of freshwater fish and their natural surrounding not true equilibrium but maintain steady state against a passive diffusion and requiring expenditure of energy. The water that fish live in is not a pure water. It is the nature of water for salt dissolve in it. The salt dissolve in body of water give its ionic balance of our personal water at level that is optimum for our biochemistry. For most species internal balance is not osmoregulation is the fish maintain internal ionic balance that is different water bodies they are living.

Most animals are stenohaline unable to tolerate large external osmolarity fluctuations Euryhaline species, like salmon can change osmoregulatiory status when salmon migrate from fresh water to the ocean. They undergo physiological changes, such as producing more cortisol to grow salt secreting cell. Fresh water fish (teleost) at fresh water teleost internal environment

is more concentrated that their surrounding environment. They are continually water gain by diffusion through gills and skin the water eliminates kidney large volume of dilute urine. The main difference between earyhaline and stenohaline is that euryhaline organism can adopt to a wide range of salinities, whereas stenohaline organism can only adapt to a narrow range of salinities.

The kidney of fresh water teleosts are very large in relation to their body size to permit formation of large amount of glomerular ultra-filtration and later to the formation of a copious amount of urine many fresh water teleosts have developed a urinary bladder for storage due to cupious secretion of urine. Urine contain little salts. The cupious flous cause a significant amount of salt to be lost. Gill tissue lost salt by diffusion. The lost and balance salt in food and by active absorption through the gills special cells in gills lamellae contain sodium and chloride pumps. These pumps are special enzymes that use energy to move the ions up their concentration gradient to maintain their higher concentration in the body some substances are reabsorbed at specific location as the fluid possess down the kidney tubul salt are reabsorbed in distal tubul and glucose is reabsorbed by proximal tubul, urinary bladder function as osmoregulation. The wall is permeable to ion and impermeable to water. This paper updates the knowledge base on how salt affect biotic and physical components of fresh water and ecosystem. Explore the need for information and now effect of fresh water change in salt concentration osmoregulation can have great impact on type of organism that live in body of water salt are compound like sodium chloride, magnesium sulfate, potassium nitrate and sodium bicarbonate which dissolve into ions. If increase salt concentration direct effect on metabolities, survival growth, feed intake and distribution. Reproduction and survival of young fish observed that a hardness of is-ISO Mgl-1 is optimum for fish over > 300 mg-1 of CaCO3 is lethal to fishes and hardness under 20 mg-1 stress to fish due to an availability of nutrients in water osmotic imbalance when salt concentration increases fish face osmotic problem. To overcome their excretory system work harder (disturb) more energy for excretion less energy for (assimilation) which result poor growth oxygen consumption rate 2.5% if salt is increase decrease rate. Survival rate increased decrease survival rate growth increase salt decreased the growth rate of fish. The control of urine producing processes is affected by pressure change by pituitary hormone e g (orginine vasotocin) m corticosteroids mediate many body functions in vertebrates including osmoregulation in fishes contricosteroids are responsible for controlling among other function metabolism immunology as more regulation stress responses.

Various metabolic activities control by water salt concentration and when fish exposed to high acidic and high alkaline water, decrease in ionic balance of gills is observed which eventually resulted in high mortality. Effect of salt concentration have been studied in several fish species but detailed work on effect of salt on growth survive, biochemical response in case of Labeo rohita, cutia, cirrhiuus mrigala, tilapia fresh water fish species are limited.

Review Of Literature: -

Evolutionary origin of Nacl uptake mechanism in Fresh water fishes13.

August Krogh was Danish contemporary of Smith & Marshall, & the post-doctoral advisor of Ancel keys. His notable contribution to the study of fish osmoregulation was the hypothesis that fresh water fishes extract needed Nacl from the environmental via parallel $Na^+/NH4^+$ and $Cl^-/HCO3^-$ exchangers **(Krogh, 1937-1938)**

Thus, living in dilute environment poses great challenges for acquiring essential ions against steep concentration gradients between body fluids and the environments **(Beyenbach, 2001: Morris 2001; Tsai and Lin, 2007; Lee *et al*; 2012).**

Fresh water animal can-not survive without maintaining elevated extracellular fluid.

Renfro (1959, 1960) suggested that the presence of Freshwater fishes in saline habitats of the Aransas River, Texas, was generally short term he observed that many fresh water fish collected were transients, because they were found in salinities greater than they

could tolerate for more than limited time period fresh water fishes cold move into low salinity areas.

Methods and Materials: -

Healthy *Labeo rohita* were collected from the fresh water fish seed. The fish were acclimatized to the laboratory condition for period of days separately to the experimental trials. *Labio rohita* also known as Rohu is a species of fish carp family. Fresh water was collected from river, pond for experimental 60 fish were taken for study taken body weight per day. Standard pelleted feed with protein level was used for feeding fishes. Uneaten food and faucal mater were removed on daily basis and complete water exchange was done once in a weak and study was conducted for a period of 60 days.

Put 60 fishes in different 3 salinity tank and observe the effect of growth and weight of *Labeo rohita* observe the initial and final weight of fish percentage of mortality was 12, 24, 36 intervals dead fishes were immediately removing from respective tank following formula are used for monthly sampling was carried out to certain weight gain.

$$\text{Weight gain \%} = \frac{\text{Final weigh} - \text{Initial weight}}{\text{Initial weight}} \text{ x } 100$$

Result and Discussion: -

The study was conducted to investigate the effect of salinity level on survival and growth of *Labeo rohita* increase salinity level reduce the growth rate survival, hemoglobin and protein concentration in *Labeo rohita*. According to study have reported that an influence of water salinity on fish development and growth in most species egg fertilization and incubation Yolk sac reabsorption early embryogenesis, swim bladder in function larval growth also depend on salinity. Specific growth rate (SGR) decrease as the salinity increase highest SGR was 0% salinity and lowest SGR was 8% salinity. Normal growth was observed at salt concentration 0 to 4%. In case of *Labeo rohita* hyper activeness or erratic behavior shown at 8 to 12%.

Conclusion: -

An excessively high salinity level can be fatal to fresh water fish but low salinity can positively affect the growth and survival of fish.

Acknowledgement: -

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A Detail Overview Of Some Soil Invertebrates From Ghatanji Region, Yavatmal District, (M.S.), India

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ABSTRACT

A soil Invertebrates is an invertebrate that spends all or much of its life in the soil. Soil Invertebrates perform a wide range of functions that contribute to ecosystem health, through the maintenance of nutrients cycling, water storage and primary productivity. Small animal living in soils, called soil invertebrates, represent a very diverse group of soil inhabitants. They include Earthworm, Nematode, and Annelida, Arthropods, Coleoptera, Mites, Snails and some insect's etc. soil invertebrates feed on dead plants, on fungi & bacteria or on other soil invertebrates. Members of each of these groupings of invertebrates exhibit characteristic that improve the health of the soil. Due to various factor the diversity of soil invertebrates is under threat so, the present study was undertaken to study this soil invertebrates around Ghatanji region. The sampling sites for the present study were chosen randomly in an around the Ghatanji city. The study was carried out in six month in which photography and identification up to possible lower taxa were done.

Keywords: - Soil invertebrates, Diversity, Annelids, Arthropods, Ghatanji region.

INTRODUCTION

In nature the soil is one of the integral elements that enable life on earth. Soils are essential sources of a wide diversity of eco-system services defined as the goods and ecosystem functions that provide benefit to human populations. Soil invertebrates are essential part of soil and they regulate productivity and health of soil **(Alagesab, P. and Ramanathan, B. (2013))**. Soil invertebrates are organisms present in soil which play a key role in nutrients cycling, decomposition and energy flow in the terrestrial habitat. Soil invertebrates comprises all larval and adult stages livings temporarily or permanently in soil and sometimes on surface **(Choudhari, C.R, Dumbare, Y.K. and Theurkar, S.V. 2014).** The presence of soil invertebrates alter the physical and chemical structure of the soil rate and extent of soil process. Soil represents one of the most important reservoirs of biodiversity. It reflects ecosystem metabolism since all the bio geo-chemical processes of the different ecosystem components are combined within it. Soil invertebrates make up diverse communities living in soil pores and on the soil surface, digging burrows and tunnels, processing organic matter and interacting with microbes **(Nuria, R., Jerome, M., Leonide, C., Christine, R., Gerard, H., Etienne, I., & Patrick, L. 2011).** Soil invertebrates have complex relationships with the surrounding environment and are affected by variations in the microhabitat and fluctuations in the types and quality of resources **(Ferlian, O., Eisenhauer, N., Aguirrebengoa, M., Camara, M., Ramirez-Rojas, I., Santos, F., & Thakur, M. P. 2018).** Soil fauna plays a key role in many soil functions, such as organic matter decomposition, humus formation, and nutrient release, modifying soil structure, and improving its fertility. Soil invertebrates play key roles in determining soil suitability for agricultural production and realizing sustainable farming systems **(Ali A., G.A Bhat and M. Ali (2012))** They include an enormous diversity of arthropods, nematodes, and earthworms. However, this fauna suffers from the impact of agricultural activities with implications for the capacity of soil to maintain its fertility and provide ecosystem services.

MATERIAL AND METHOD

Study Area: - The study area Ghatanji Taluka is located at the Yavatmal District in the State Maharashtra, India. The longitude of the study area is 78°18' 42. 00 48" E and latitude is 20° 8' 37.27 68" N is neighborhood of the Yavatmal District. Yavatmal district lies in the Vidarbha region of the state and is popularly known as the cotton city because in this area farmer produces a fine quality of cotton.

Collection of Samples: - In the present study sampling sites are randomly selected, such as grass lands, agricultural fields, garden and grounds. Soil invertebrates were collected from selected sampling sites. The Specimen were collected by hand picking method and general observation with photographic collection of specimen was done at sampling sites and then these animal were released in their habitat. Identification is done by using Standard Invertebrates keys and by internet facility. **(Rombke, J,J. Bernard, and F. Martin-Laurent 2018)**

RESULT AND DISCUSSION

The main groups of soil invertebrates are flatworms (Platyhelminthes), Earthworms (Annelida), nematodes (Nematoda), crustaceans, arachnids and insects (Arthropoda). Snails and slugs (Mollusca).

Flatworms are most diverse in tropical regions and are usually found under rocks or in leaf litter where conditions are humid and moist. They do not have the ability to retain water on their own and will desiccate without an external water source. They are predators and typically eat other invertebrates, including arthropods, earthworms, snails, and slugs. Invasive flatworms can have a damaging effect on an ecosystem when they consume native species. This is especially concerning when native earthworm populations are affected, as earthworms are important soil fauna.

Snails and slugs are typically found in leaf litter in forests, but can also be found in gardens, fields, river banks, and urban areas. Most are considered decomposers and feed on plants, fungi, and algae. Some species are carnivorous and will eat nematodes and other snails and slugs. They are also important sources of food for many predators such as beetles, millipedes, small mammals, reptiles, and birds. Snails are important for calcium cycling as they uptake calcium from their food and use it to create their shells, and then are eaten by predators. Calcium availability, soil moisture, and land use strongly affect snail populations.

Although annelids include leeches and ragworms, The most ecologically important type that occurs in soil are earthworms. Earthworms decompose dead organic matter and are major drivers of nutrient and water cycling, plant growth, and changes to soil structure. Some species live at the very surface of soil and within leaf litter, while others live in the upper layer of the soil. Some types are deep burrowers and create permanent burrows several meters long that they use to pull organic matter from the surface down into the soil. Earthworms also consume fungi and bacteria, and are commonly preyed upon by mammals and birds. Since earthworms majorly affect organic matter and microbial populations in soil, they indirectly influence the amount and distribution of other soil fauna.

Invasive species of earthworms can also be extremely damaging to ecosystems. Invasive earthworms negatively affect abundance and diversity of both microorganism and microorganisms in the soil and cause changes to the physical and chemical properties of the soil. These effects can in turn negatively affect overall ecosystem functioning and services Nematodes are small, non-segmented worms that are found in forest, grassland, and agricultural soils. Food sources differ between species, with some nematodes feeding on fungi, others on bacteria, and some on protozoa or other nematodes. They are typically found where their food sources are concentrated. Nematodes are preyed upon by insects and predatory nematodes, and are parasitized by bacteria and fungi. A few species cause diseases in plants, but beneficial nematodes are important for nutrient cycling and dispersing microbes within the soil.

Arthropods are an extremely diverse and prolific group of soil fauna, comprising up to 85% of the species present in soil. They include crustaceans, arachnids, insects, myriapods (centipedes and millipedes), and scorpions. Due to their multitude and diversity, arthropods carry out a wide range of functions and processes within soil. Some arthropods feed on fungi, which also contributes to nutrient cycling. Arthropods can also be herbivores or predators, and many act as a bio control to crop pests or as crop pests themselves. Although some are pests, most arthropods are beneficial to soil ecosystems. The majority inhabit the top few inches of soil, with abundance and species diversity diminishing with depth. Because of their diversity and abundance, soil invertebrates play many roles in the ecosystem. As part of the food web, invertebrates both consume and are a source of food for other organisms, altering the composition and abundance of species of both plants and animals in a community.

Many invertebrates are also drivers of nutrient and water cycling. By breaking down decaying plant matter and contributing to decomposition, nutrients such as nitrogen, phosphorus, and carbon become available for plants to uptake, which in turn stimulates plant growth. They can also be used as bio indicators of soil quality and health. Invertebrate populations are affected by soil contamination, ecosystem clearing, and land management practices. The presence of indicator species and diversity are indicative of higher soil quality.

Sr. No	Phylum	Class	Order	Family	Genus	*Species*
1	Annelida	Oligochaeta	Opisthopora	Megascolecidae	Pheretima	*posthuma*
2	Annelida	Hirudinea	Gnathobdellida	Cylicobdellidae	Hirudinaria	*granulosa*
3	Annelida	Clitellata	Haplotaxida	Lumbricidae	Aporrectedea	*calignosa*
4	Arthropoda	Chilopoda	Scolopendromorpha	Scolopendridae	Scolopendra	*morsitans*
5	Arthropoda	Diplopoda	Polydesmida	Xystodesmidae	Harpaphe	*H. haydeniana*
6	Arthropoda	Diplopoda	Spirostreptida	Spirosdtreptidae	Orthoporus	*O. ornatus*
7	Arthropda	Insecta	Hymenoptera	Formicidae	Camponotus	*pennsylvanicus*
8	Arthropoda	Arachnida	Araneae	Lycosidae	Pardosa	*amentata*
9	Artropoda	Malacostraca	Decapoda	Cancridae	Metacarcinus	*magister*
10	Arthropoda	Insecta	Hymenoptera	Formicidae	Camponotus	*ocreatus*

Pheretima posthuma

Hirudinaria granulosa

Aporrectedea calignosa

Scolopendra morsitans

Camponotus pennsylvanicus

Pardosa amentata

H. haydeniana

Orthoporus ornatus

Metacarcinus magister

camponotus ocreatus

CONCLUSION

The present research work is deals with the overview of soil invertebrates in the study area Ghatanji. It is a qualitative survey of soil invertebrates carried out over period of 6 month from August-2022 to January-2023. The random sites were selected for the present study at Ghatanji city. Sampling sites were visited frequently during the present study period and the photographic collection were done at the sampling sites. Total 10 Species of soil invertebrates were recorded during the present study.

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The Present Scenario Of Gastro-Intestinal Helminthic Infection In *Capra Hircus* From Yavatmal Region, District: Yavatmal (M.S.) India

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ABSTRACT

The present communication deals with the current scenario of gastro-intestinal helminthic infection in *Capra hircus* from Yavatmal region, district Yavatmal. During January 2023 to December 2023. Out of **262** samples, 158 samples (60.30%) were positive for helminthic infection. The incidence of helminthic infection is slightly more in adult *Capra hircus* i.e. (210/62.03%) than Kid (78/52.17%). The seasonal variation of helminthic infection shows Adult & Kid the higher Prevalence of cestode& nematode occurs in winter C- 115, N-78 followed by Monsoon C- 108, N-54 and summer C- 85, N-22 respectively the present study indicates that the higher infection occurs in cestode parasites in all Seasons than nematode parasites.

[C- Cestode, N- Nematode]

KEY WORDS: Gastro-intestinal, *Capra hircus*, Kid, Cestode, Nematode.

INTRODUCTION

Gastro-intestinal parasitism are harmful to animal life, they cause low productivity and occasional death in farm animals. The present work has been undertaken to study the Prevalence and seasonal variation of gastro-intestinal parasitic infection in *Capra hircus* and their Kid, at Ghatanji. Dist.: Yavatmal (M.S.) Goat (Capra hircus) is one of the important domestic livestock as a source of dairy, meat and manure. Female goats are referred to as does or nannies, intact males as bucks or billies their offspring are kids. Goat meat from younger animals is called kid or cabrito, and from older animals is called chevon or "mutton" in some areas **(www.wikipedia.org/goat)**. Gestation length is approximately 145-155 days. Twins are the usual result with single and tripled births are also common **(www.bva.awf.org.uk).** Capra hircus is an herbivorous. Goats are primarily reared for meat and manure and regarded as the second important animal species (first being buffalo) for generating their cash income by farmers **(Gatenby *et al*. 1990).**

Goats are supposed to be the first farm animals domesticated **(Zenuer 1963 and Devendra 1998)** and is believed that domestication Resource poor farmers of the hills, who cannot invest large sums of money in cattle and buffalo, prefer sheep and goat husbandry which has no social, religious or cultural taboos, or caste restrictions **(Ghimire 1992).** Traditionally, in Nepal meat and meat products of all domestic animals are consumed except cattle. Animal slaughter is a common practice not only for consumption but also for religious sacrifices and traditional ceremonies. **(Kennedy C. R. 1977 (a).)**

Parasites are classified as endoparasites and ectoparasites on the basis where they live inside or on the body cavity. Endoparasites are those organisms living in their hosts, in the gut, body cavity, liver, lungs, gall bladder and blood and within the intestinal, tissues or cell of the host. Among different parasitic infections, gastrointestinal diseases are most varied and of common occurrence. Different grades of infections with fluke, tapeworms and roundworms, are responsible for marked deleterious effects that tend to lower overall production both by the way of morbidity and mortality.

REVIVE OF LITERATURE

Research on the present scenario of gastro-intestinal helminthes infection in *Capra hircus*, which is the scientific name for goats, however, there have been studies conducted on helminthic infection in goat specially mention the cestode and nematode

Gastro-intestinal helminthes infection are a common problem in goat, they can causes significant economic losses for farmer. The most common helminthes found in goat include round (nematodes) such as Haemonchus contortus, Trichostrongylus spp., Teladorsagia cicumcincta, and Strongyloides Spp. Additionally, tapeworms (Cestode) and flukes (trematodes) can also infect goats.

These helminthic infections can lead to reduced productivity, including poor weight gain, reduced milk production, and lower reproductive efficiency. They can also lead anemia, diarrhea and gastrointestinal Issues,

To manage and control gastro-intestinal helminthic infections in goat, various methods are employed. These include the use of anthelmintic drug (dewormers), pasture management practices, and genetic selection for resistance to helminthic. **(K.M. Shaikh, M.S. Nirmale, D.B. Bhure and H.S. Chaudhari.2010.)**

It is essential for goat farmers and researchers to continuously monitor and study the prevalence and impact of gastro-intestinal helminthic infection in Capra hircus to develop effective control measures and minimize the Economic losses associated with these infection.

MATERIAL AND METHODS

A total of 262 samples from *Capra hircus* of different age groups collected from Ghatanji , Yavatmal district (M. S.) , during January 2023 to December 2023 and were processed by Standard method**(Souls by, 1982)**. The infected material which was collected that Preserved in 4% formalin, 10% glycerol; Viz. Cestodes and Nematodes respectively. Borax caramine and Haematoxylin stain are used for permanent slides and ready for cestode identification where the nematode parasites are fixed in glycerin jelly for identification.

RESULTS AND DISCUSSION

Out of the 262 samples subjected to examination 158 samples were positive for Gastro-intestinal parasites (60.30%). The overall infection with Strongyloides Nematode was (55.06%) Trichuris (44.93%) compares to other parasites. Moniezia infection was (37.5%), Stilesia (34.02%), Alizia (18.75%) and Avitellina (9.72%). Highly infection of Strongyloides sp. Occurs in adult *Capra hircus* and Kid. The Moniezia, Stilesia, Alizia, Avitellina and Trichuris spp. were observed in Kid and adult. Trematodes infection did not show any age group. The seasonal variation of helminthic infection shows Adult & Kid the higher Prevalence of cestode& nematode occurs in winter C- 115, N-78 followed by Monsoon C- 108, N-54 and summer C- 85, N-22 respectively the present study indicates that the higher infection occurs in cestode parasites in all Seasons than nematode parasites.

The prevalence of Strongyloides and Trichuris was more in winter month which was similar the finding of **Vasdevan and Basuthakar (1986)** and **Katoch *et. al* (1999**). Who reported highest? Strongly infections in Rainy and winter month respectively. The present study indicates that the occurrences of parasites are dependent on Suitable environment which they require in its development. Hence winter Seasons are Favorable to parasitic development.

TABLE-A

PREVALENCE SHOWING THE INFECTION BETWEEN HOST AND PARASITES

C. hircus	**Total no. of sample Examined**	**No. Of Infected Sample**	**No. of Cestode Collected**	**Prevalence (%) (2&3)**	**No. of Nematode Collected**	**Prevalence (%)**
Adult	216	134	210	62.03%	128	59.25%
Kid	46	24	78	52.17%	30	65.21%
Total	262	158	288	60.30%	158	59.62%

TABLE-B

PREVALENCE SHOWING THE CESTODE PARASITES IN HOST (ADULT & KID)

Name of Cestode	No. of Parasite	Prevalence (%)
Moniezia	108	37.5%
Stilesia	98	34.02%
Alizia	54	18.75%
Avitellina	28	9.72%
Total	288	9.72%

TABLE- C

PREVALENCE SHOWING THE NEMATODE PARASITES IN HOST (ADULT & KID)

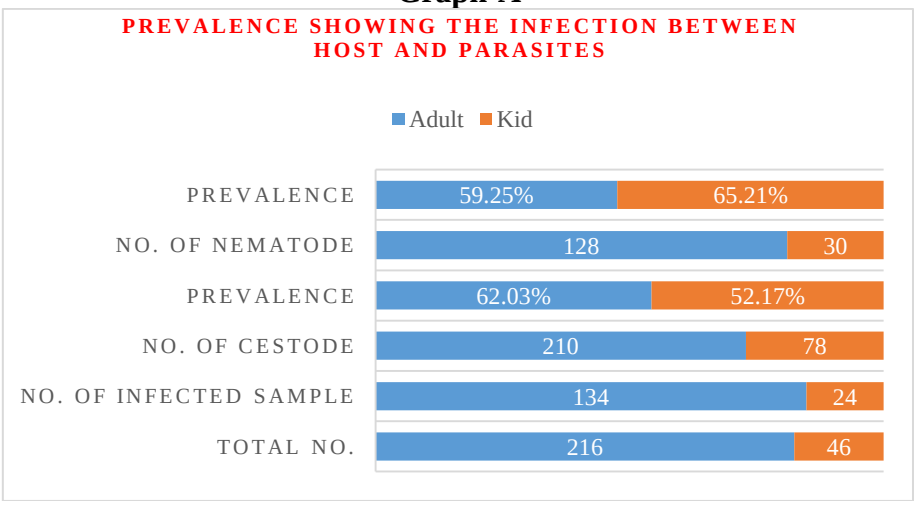
Name of Nematode	No. of Parasite	Prevalence (%)
Strongyloides	87	55.06%
Trichuris	71	44.93%
Total	158	60.30%

TABLE-D

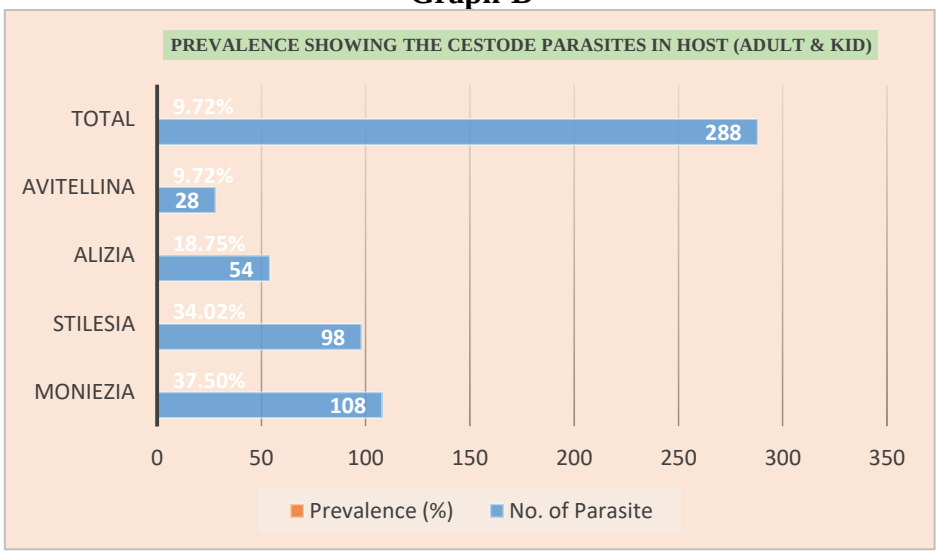
SHOEING THE INFECTION BETWEEN SEASON AND HELMINTHIC INFECTIONS

SEASONS	TOTAL NO.OF SAMPLE EXAMINED	Name of parasites	NO. OF INFECTED SAMPLE
	Adults / Kids		Adult /Kid
Winter	87	Cestode	115
		Nematode	78
Summer	88	Cestode	85
		Nematode	22
Man soon	87	Cestode	108
		Nematode	54
	[134+24]=262		[134 + 24]= 158 Cestode=288 & Nematode= 158

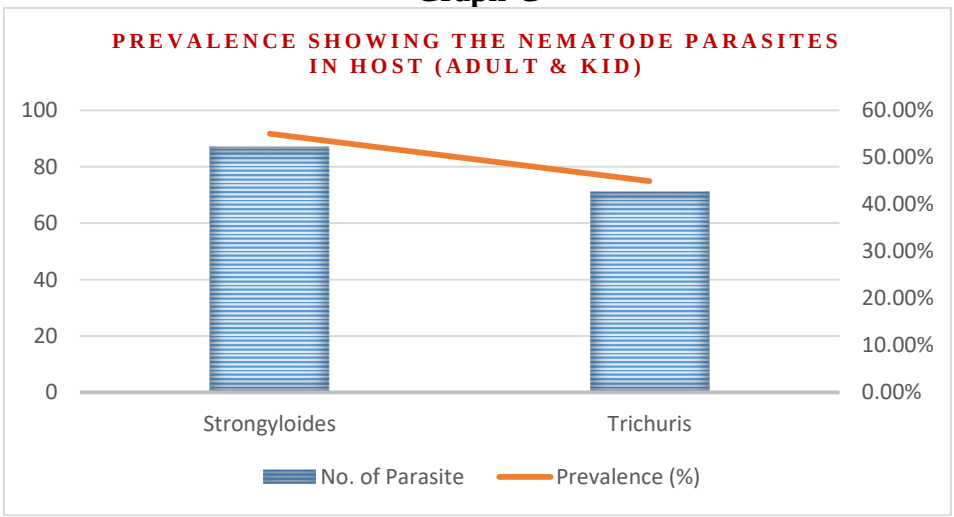
Graph-A



Graph-B



Graph-C



CONCLUSION:

The present study indicates that the higher infection occurs in the host *Capra hircus* were cestode parasites in all Seasons than nematode parasites. Gastro-intestinal parasites are harmful to animal life, they cause low productivity and occasional death in farm animals. The present work has been undertaken to study the Prevalence and seasonal variation of gastro-intestinal parasitic infection in *Capra hircus* and their Kid, at Ghatanji. Dist.: Yavatmal (M.S.) In conclusion, various gastrointestinal parasites have been found in cattle in the study area. Hence, the high prevalence rate of helminthiasis in livestock needs to be checked periodically. Regular control measures should be practiced and farmers educated in the proper use of antihelminthiasis. Epidemiological facts suggest that high standard of sanitation in modern animal husbandry will prevent exposure of livestock to graze in deteriorated and environmentally polluted range lands will be effective in controlling disease.

RECOMMENDATION

The following recommendations will help in the prevention or reduction of helminth infections in the study area.(i)There should be legislative control over slaughtering of goats and their distribution; the abattoir workers should be properly trained on meat handling and zoonotic infections.(ii)Animals should be restricted to special areas of land provided by the government for grazing.(iii)The public should be enlightened on proper cooking of animal parts especially the intestine.(iv)A comprehensive approach should be adopted to ensure all inclusive meat inspection in the abattoirs before distribution to the public for consumption.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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Review: Methodological Approaches to investigate different neuronal groups of brain in *Mus musculus*

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Abstract:
This paper explores the diverse methodologies employed in neuroscience to unravel the complexities of distinct neuronal groups within the brain's architecture. Beginning with early anatomical studies and histological staining, the evolutionary progression is traced through observational techniques, electrical recordings, neurochemistry, neuropharmacology, molecular biology, imaging techniques, functional imaging, and advanced methods like optogenetics and chemogenetics. The focus extends to specific applications in studying nitric oxide, encompassing electrophysiological techniques, electrochemical sensors, chemical detection, molecular biology, immunohistochemistry, and behavioral studies. The evolution of neuroscience methodologies reflects technological advancements, from basic observational tools to cutting-edge approaches, showcasing the relentless pursuit of knowledge in understanding the intricate web of communication within the nervous system. This multidisciplinary journey promises continued innovation and breakthroughs, shaping the future of neuroscience exploration.
Keywords: Chemogenetics, Neuron, Electrocardiograph, Fluorescent, PCR etc.

Introduction:

The intricate web of communication within the nervous system has long been a subject of fascination and exploration in the field of neuroscience. In the enigmatic realm of neuroscience, the investigation of distinct neuronal groups within the intricate architecture of the brain stands as a critical endeavor. As we strive to unlock the secrets of cognitive function and behavior, understanding the diversity and functional specialization of these neural ensembles becomes paramount. This paper delves into the methodological approaches employed by researchers to explore the nuanced intricacies of different neuronal groups. From advanced imaging techniques to cutting-edge molecular analyses, the tools at our disposal shape the landscape of neuroscientific inquiry. Join us on a journey through the methodologies that illuminate the pathways to deciphering the unique contributions of various neuronal populations, as we endeavor to unravel the profound complexities woven into the fabric of the brain's neural tapestry.

The evolutionary history of methodologies: In neuroscience, from simpler to more complex approaches, reflects the progression of technological advancements and the deepening understanding of the nervous system.

Observational Techniques

Early Anatomical Studies: In the early days, anatomists relied on simple observational techniques, such as dissections, to study the gross structure of the nervous system. This laid the foundation for understanding basic neuroanatomy. Herophilus first differentiated nerves, blood vessels and tendons.Ghosh, S. K., & Narayan, R. K. (2020).

Histological Staining

Golgi Staining: Introduced by Camillo Golgi, this method allowed for the visualization of individual neurons in their entirety. It was a major breakthrough in understanding neuronal morphology and connectivity. It gives simple tree dimensional neuron morphology based on

formation of opaque intracellular deposit of silver chromate by reacting potassium dichromate and silver nitrate.Torres-Fernández O. (2006).

Electrical Recordings

Electroencephalography (EEG): The recording of electrical activity from the brain surface provided early insights into overall brain function. EEG remains a relatively simple and non-invasive technique to study large-scale brain activity. The technique is based on electrodes that are attached to scalp to communicate via electrical impulses and measures electrical activity all the time and it shows as wavy lines on EEG recording. I help to diagnose epilepsy, sleep disorder and brain tumor. Fuzik et.al (2016) and Sauseng(2010).

Electrophysiology

Single-Cell Recordings: Techniques like intracellular and extracellular recordings enabled researchers to study the electrical properties of individual neurons and their responses to stimuli. It provides brain images under 40-60 cells at a single recording but the main role of these method is highest resolution among all present brain imaging methodology that based on fluctuation of flow of electrons or voltage in neuron which operates by microelectrodes over scalp or skull. Biasiucci, A., Franceschiello, B., & Murray, M. M. (2019)

Above figure depict the investigation in different neuronal group of *Mus musculus*

Neurochemistry

Neurotransmitter Identification: Advancements in chemical analysis allowed researchers to identify and characterize neurotransmitters, providing insights into chemical signaling in the nervous system. Several techniques are reported to observed neurotransmission among different neuronal groups. 1. Positron emission tomography (PET): it is noninvasive method used to measure molecular or metabolic activity indirectly help to measure neuronal activity, by using isotope with short life whose disintegration gives positron emission. 2. Single photon Emission computed tomography (SPECT): radioactive technique which is acquired by the emission of gamma radiation and these radio tracing is injected in patient bloodstream and chemical properties are taken in account to which receptors of tissue which are the subject of tomographic imaging technique on nuclear scale. It is recorded by gamma camera and reconstructed by computer algorithms which gives spatial visualization of different neurotransmitter. 3. Fluorescence: the technique is based on fluorescence and wavelength fluctuation for e.g. Some neurotransmitter such as Dopamine, Serotonin, and nor Adrenalin is detected by simple difference in emission of wavelength incident and emitted longer wavelength and incident lower wavelength. Brady, S., & Siegel, G. (2011) and Sawamoto (2001)

Neuropharmacology

Drug Interventions: Pharmacological approaches involving the administration of drugs allowed researchers to manipulate neurotransmitter systems, leading to the understanding of the role of specific neurotransmitters in behavior and cognition. Cooper, J. R., Bloom, F. E., & Roth, R. H. (2003).

Molecular Biology and Genetics

PCR and Gene Expression Studies: The advent of molecular biology techniques facilitated the study of gene expression in the nervous system, providing insights into the molecular basis of neuronal function. It gives immense information to understand the working mechanism of targeted region in brain by using RT PCR and DNA chip technology. Roth et.al.,(2006).

Imaging Techniques

Computed Tomography (CT) and Magnetic Resonance Imaging (MRI): These non-invasive imaging techniques allowed for the visualization of brain structures, aiding in the diagnosis of neurological disorders. Bootz et.al.,(1992).

Functional Imaging

Functional Magnetic Resonance Imaging (fMRI): This technique measures changes in blood flow and has become a powerful tool for studying brain function during various tasks, providing a non-invasive way to investigate regional brain activity.Buxton, R. B. (2013).

Optogenetics and Chemogenetics

Precise Neural Manipulation: Optogenetics and chemogenetics involve the use of light or chemicals to manipulate specific neuronal populations, allowing researchers to causally link neural activity to behavior.Vlasov (2004).

Electrophysiological Techniques

Patch-Clamp Electrophysiology: This technique is crucial for studying the effects of nitric oxide on membrane potential and synaptic transmission. Patch-clamp recordings allow researchers to monitor changes in neuronal activity and evaluate the impact of nitric oxide on ion channels and synaptic currents. Bucher, E. S., & Wightman, R. M. (2015) and Tao et.al (2015).

Electrochemical Sensors: Amperometric and voltammetric sensors provide a means to measure nitric oxide release in specific brain regions. These sensors enable high temporal resolution measurements of nitric oxide concentrations.Neher, E., & Sakmann, B. (1992).

Chemical Detection and Measurement

Fluorescent Probes: Utilizing fluorescent indicators, researchers can visualize and quantify nitric oxide levels in real-time within neuronal cells. This includes dyes such as DAF-FM (4-amino-5-methylamino-2',7'-difluorofluorescein), which becomes fluorescent upon reaction with nitric oxide.Zhao et.al (2004)

Immunohistochemistry and Imaging

Immunofluorescence Staining: By using specific antibodies against neuronal nitric oxide synthase (nNOS), researchers can visualize the distribution of nitric oxide-producing neurons in the brain. This helps in identifying regions where nitric oxide may function as a neurotransmitter.

Confocal Microscopy: High-resolution imaging techniques help in studying the subcellular localization of nitric oxide synthase and its association with synaptic structures.R. M. (2015).

Molecular Biology Techniques

PCR and RT-PCR: These techniques are employed to analyze the expression of nitric oxide synthase genes in different brain regions. This provides insights into the transcriptional regulation of nitric oxide production.

Western Blotting: Protein analysis using Western blotting allows for the quantification of nitric oxide synthase protein levels in specific brain tissues. Wen et.al.,(1998).

Neurochemical Assays

Microdialysis: In vivo microdialysis allows researchers to collect extracellular fluid from specific brain regions. This technique enables the measurement of nitric oxide levels, providing information about its release dynamics in response to various stimuli.Mantyh P. W. (1982).

Behavioral Studies

Behavioral Pharmacology: Investigating the behavioral consequences of manipulating nitric oxide levels provides insights into its functional significance. This includes assessing the effects of nitric oxide modulators on learning, memory, and other cognitive functions.Lau et.al., (2024)

Genetic Manipulations

Transgenic and Knockout Models: Creating genetically modified animal models, such as transgenic mice overexpressing or lacking specific nitric oxide synthase isoforms, helps in elucidating the in vivo roles of nitric oxide in neurotransmission. J. L., & Somogyi (1998)

Conclusion

The evolution of neuroscience methodologies is a testament to human ingenuity and relentless pursuit of knowledge. From humble beginnings, the field has evolved into a multidisciplinary

science, integrating techniques from anatomy to genetics, from electrophysiology to computational modeling. This journey not only deepens our understanding of the nervous system but also paves the way for innovations and breakthroughs with profound implications for neuroscience and beyond. As technology continues to advance, the future promises even more sophisticated methodologies, opening new frontiers in the exploration of the brain and its complexities. On the basis of present information, I will try to reveal midbrain region of mice with three immunoreactive positive neuronal group: Accumbence and Accumbence shell, Median forebrain bundle (MFB) and paraventricular nucleus of the hypothalamus (PVN).

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Distribution of ICT Tools Teaching in Science & Technology Dist-Amravati -Maharashtra

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Abstract:

The study that Education has been described as means to modify the behaviour. Biological science deals with study of living organism, their living environment and various interactions between living things and environment. The traditional content of biology has been replaced by modern discoveries in the fields of cell biology, basic genetics, bioengineering and biotechnology. Biology as a subject has both theory and practical components. ICT has opened new avenues like, online learning, e-learning, virtual university, ecoaching, e-education, e-journal, etc. It has provided opportunity for the learner to use maximum senses to get the information. . Learning of biology can be made easier and more comfortable by integrating ICT tools in instructional stratagies for teaching biology. The students of biology can make use of ICT for easy understanding. ICT can change traditional classroom into smart classroom and improve teaching-learning process in biology.

Keywords: Teacher, Lecturer ,Professor, Research scholar, ICT; Biology; e-learning; e-coaching; e-education; e-journal ,students

Introduction-

Information Technology was limited only to the textual mode of transmission ofinformation with ease and fast. But theinformation not only in textual form but inaudio, video or any other media is also tobe transmitted to the users. It has openednew avenues like, online learning, elearning, virtual university, e-coaching, eeducation, e-journal, etc. The ICT bringsmore rich material in the classrooms andlibraries for the teachers and students. Ithas provided opportunity for the learner touse maximum senses to get theinformation. It has broken the monotonyand provided variety in the teaching –learning process (Agashe, L, 1995).

Radioactive compounds are especially useful in biochemical studies involving metabolic pathways of synthesis and degradation. Radioactive compounds are incorporated into cells in the same way as their nonradioactive counterparts. These compounds provide information on the sites of specific metabolic activities within cells and insights into the fates of these compounds in both organisms and the ecosystem.Animal-related industries produce food (meats and dairy products), hides, furs, wool, organic fertilizers, and miscellaneous chemical by products. There has been a dramatic increase in the productivity of animal husbandry since the 1870s, largely as a consequence of selective breeding and improved animal nutrition. The purpose of selective breeding is to develop livestock whose desirable traits have strong heritable components and can therefore be propagated. Heritable components are distinguished from environmental factors by determining the coefficient of heritability, which is defined as the ratio of variance in a gene-controlled character to total variance. (https://www.britannica.com/science/zoology/Methods-in-zoology)

Material Method-

Monitoring on digital culture and digital literacy: Computer technologies and other aspects of digital culture have changed the ways people live, work, play, and learn, impacting the construction and distribution of knowledge and power around the world.(50)Graduates who

are less familiar with digital culture are increasingly at a disadvantage in the national and global economy. Digital literacy—the skills of searching for, discerning, and producing information, as well as the critical use of new media for full participation in society—has thus become an important consideration for curriculum frameworks.

Result Discussion –

In many countries, digital literacy is being built through the incorporation of information and communication technology (ICT) into schools. Some common educational applications of ICT include:

- *One laptop per child:* Less expensive laptops have been designed for use in school on a 1:1 basis with features like lower power consumption, a low cost operating system, and special re-programming and mesh network functions.
- *Tablets:* Tablets are small personal computers with a touch screen, allowing input without a keyboard or mouse. Inexpensive learning software ("apps") can be downloaded onto tablets, making them a versatile tool for learning.(7)(The most effective apps develop higher order thinking skills and provide creative and individualized options for students to express their understandings.
- *Interactive White Boards or Smart Boards*: Interactive white boards allow projected computer images to be displayed, manipulated, dragged, clicked, or copied. Simultaneously, handwritten notes can be taken on the board and saved for later use. Interactive white boards are associated with whole-class instruction rather than student-centred activities.(Student engagement is generally higher when ICT is available for student use throughout the classroom.(10)
- *E-readers*: E-readers are electronic devices that can hold hundreds of books in digital form, and they are increasingly utilized in the delivery of reading material.(10) Students both skilled readers and reluctant readers—have had positive responses to the use of e-readers for independent reading.(10) Features of e-readers that can contribute to positive use include their portability and long battery life, response to text, and the ability to define unknown words. Additionally, many classic book titles are in e-book form..
- *Flipped Classrooms:* The flipped classroom model, involving lecture and practice at home via computer-guided instruction and interactive learning activities in class, can

- allow for an expanded curriculum. There is little investigation on the student learning outcomes of flipped classrooms.(5) Student perceptions about flipped classrooms are mixed, but generally positive, as they prefer the cooperative learning activities in class over lecture.

Table No -1-Data Monitoring of 20 students in class.

Sr. No	***Name Of Students***	***One laptop per child:***	***Tablets***	***Interactive White Boards or Smart Boards***	***E-readers:***	***Flipped Classrooms:***	***Mobile Phone***
1	Akash D. Shelke	-	-	-	-	+	+
2	Ashiwani P. More	-	-	-	-	+	+
3	Bharti R. Ruchake	-	-	+	-	+	+
4	Dipika S. Bhashkar	-	-	+	-	+	+
5	Arshiya F. M. Ali	-	-	+	-	+	+
6	Divyani G. Nage	-	-	+	-	-	+
7	Kiran U. Adhau	-	-	+	-	-	+
8	Mediha T. Baig	-	-	+	-	-	+

9	Pornima D. Bhonde	-	+	+	-	-	+
10	Prashant A. Adhau	-	+	+	-	-	+
11	Pratiksha S. Patel	-	+	+	+	-	+
12	Purva B. Umbarkar	-	+	+	+	-	+
13	Renuka M. Alkari	-	+	-	+	-	+
14	Shradha R. Gavhale	-	+	-	+	-	+
15	Shivani S. Bhise	-	+	-	+	-	+
16	Sonu R. Gaur	-	+	-	+	-	+
17	Vikas G. Chaudhari	-	+	-	+	-	+
18	Mohd. ArshidM.Baig	-	+	-	+	-	+
19	DivyaniA.Aswar	-	-	-	+	-	+
20	Mohini S. Muratkar	-	-	-	+	-	+

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Study On Physicochemical Condition And Cladocera Diversity Of Popatkhed Reservoir From Akola District, Maharashtra (India)

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Abstract

The study was conducted during Winter 2022–23 to analyse the physicochemical status of Popatkhed reservoir with reference to Cladoceran diversity. The study revealed the rich Cladoceran diversity with eight identified species. The observed status of Cladoceran diversity was mostly related to the studied physicochemical parameters. The observed results were correlated and significantly different at $p<0.05$. The results show a positive relationship between these physicochemical parameters and observed Cladoceran diversity. But compared with the BIS drinking water standard, the water at these sites is not that suitable for drinking purposes but can be used for drinking after proper processing and filtration. But the water in the reservoir is suitable for irrigation and fish culture.

Key words: Cladocera, Diversity, Physicochemical, Popatkhed Reservoir, Zooplankton.

INTRODUCTION

Water is probably the only natural resource to touch all aspects of human civilization, from agricultural and industrial development to cultural and religious values. Today, this freshwater has become the fastest-depleting natural resource globally. Only a small percentage of water exists as freshwater, and the portion accessible to humans is again a negligible part of its global stock of surface water bodies such as rivers and lakes. However, knowingly or unknowingly, it is this rarest resource that we abuse severely. Ignorant, irresponsible, and careless management has brought the water of the world to serious depletion. The available freshwater is not accessible to all people due to differences in geographical, geological, climate, and demographic reasons. A global literature survey reveals that 70% of the earth's surface is covered by water. Although it is surprising that there is a shortage of pure freshwater because more than 97% of the water is marine and only 3% of the fresh, soft water is suitable for human consumption and other uses (Brönmark and Hansson, 2005; O'Sullivan and Reynolds, 2005),

Among the entire aquatic biota, the zooplankton are one of the important biological indicators that represent the health of the water body. Zooplankton are heterotrophic plankton. Plankton are organisms drifting in oceans, seas, and bodies of fresh water. Individual zooplanktons are usually microscopic, but some are larger and visible to the naked eye. Zooplankton is a classification spanning a range of organism sizes, including small protozoans and large metazoans. It includes holoplanktonic organisms whose complete life cycle lies within the plankton, as well as meroplanktonic organisms that spend part of their lives in the plankton before graduating to either the nekton or a sessile, benthic existence. Although zooplankton are primarily transported by ambient water currents, many have locomotion, which is used to avoid predators or increase prey encounter rates (Joshi, 2011; Barskar and Jawalkar, 2022).

In this regard, the study was conducted during Winter 2022–23 to analyse the physicochemical status of the reservoir with reference to zooplankton diversity.

MATERIALS AND METHODS

Study Area: Popatkhed Dam is an earthen-type reservoir located between 21.20°N and 77.08°E. It is an earthfill dam on a river near Akot, Akola district, in the state of Maharashtra, India. The height of the dam above the lowest foundation is 42.6 m (140 ft). The gross storage capacity is 12,192.00 km3 (2,925.02 cu mi). During monsoons, reservoirs get

enough water, but in the postmonsoon period, particularly in March and April, water levels are very much reduced. The reservoir is surrounded by red laterite soil and black cotton soil. The inland reservoir is fed by seasonal drainage to its periphery and nearby local streams and springs (Akola Gazetteer, 2021).

Water Sampling and Physicochemical Analysis: Water samples were collected from the lake early in the morning. From different stations. Sample collection and analysis were performed as per routine methods (Trivedy and Goel, 1986; IAAB, 1999) without any modification, and the samples were compared with the drinking water standards of BIS (2014).

Zooplankton Collection and Identification: Water samples were collected separately for the study of all the zooplankton. The zooplanktons were identified using methodologies developed by APHA (1998) and IAAB (1999).

Statistical Analysis: Results were recorded as mean ± standard deviation (SD). Data were collected, organised, and analysed using Microsoft Excel program.

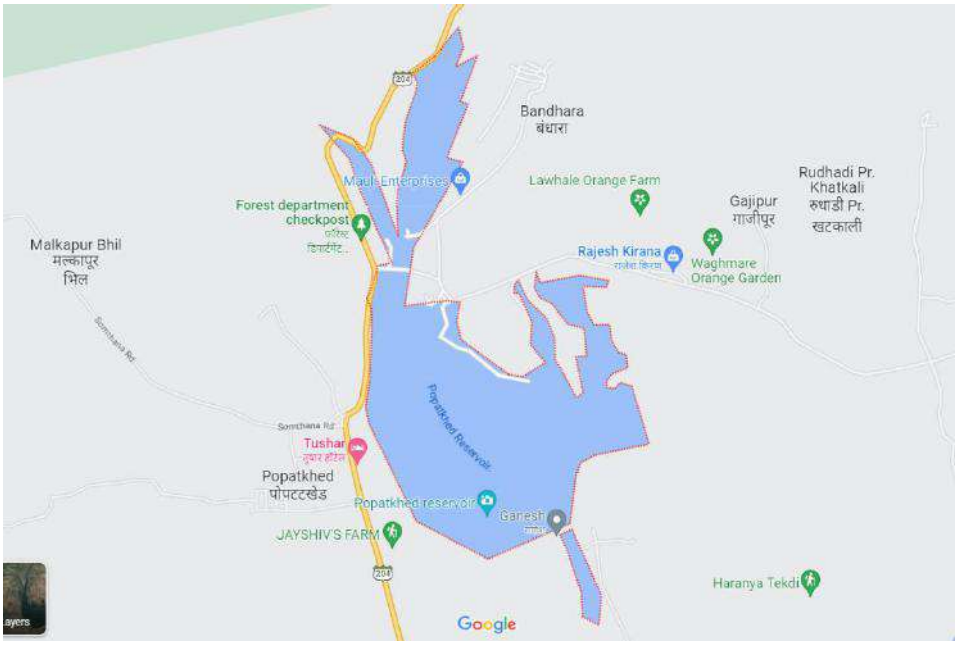

Figure 1: Map of Popatkhed Reservoir

RESULTS AND DISCUSSION

The study was conducted during Winter 2022–23 to analyse the physicochemical status of the reservoir with reference to zooplankton diversity. The following Table 1 represents the physicochemical status and zooplankton (Cladocera) abundance in the reservoir during winter 2022–23. During the period of investigation, about eight Cladoceran species, namely Bosmina longirostris, Ceriodaphnia laticaudata, Chydorus sphaericus, Dadaya sp., Daphnia laevis, Leydigia acanthocercoides, Macrothrix sp., and Moina brachiata, were identified. Species showed an important ecological role as a biotic indicator of the health of an aquatic ecosystem. Figure 2 represents the zooplankton abundance at different studied spots in Popatkhed.

Figure 2 represents the zooplankton abundance at different studied spots in Popatkhed reservoir.

Tabel 1: The physicochemical status of reservoir

Parameter	A	B	C	D	E	Mean	+ SD
Temperature	23.54	25.14	24.94	24.64	25.74	24.80	0.811
pH	7.68	7.58	7.88	7.68	7.98	7.76	0.164
Transparency	96.40	94.34	96.43	104.43	97.43	97.83	3.847
TDS	259.55	259.55	269.75	270.15	259.55	263.71	5.698
Tot. Hardness	126.12	124.54	130.68	132.27	133.28	129.38	3.850
Tot. alkalinity	182	184	184	185	187	186.4	3.362
DO	11.3	11.0	11.0	11.2	11.2	11.14	0.134
Free CO_2	3.90	3.80	5.60	6.10	6.30	5.14	1.205

The study revealed the rich zooplankton diversity. The observed status of zooplankton diversity was mostly related to the studied physicochemical parameters. The observed results were correlated and significantly different at $p<0.05$. The results show a positive relationship between these physicochemical parameters, such as water temperature, pH, total dissolved solids, total hardness, and total alkalinity, and observed zooplankton diversity. But compared with the BIS drinking water standard, the water at these sites is not that suitable for drinking purposes but can be used for drinking after proper processing and filtration. But the water in the reservoir is suitable for irrigation and fish culture. The presented observations are in good agreement with previous studies by Saxena and Saxena (2014), Bera *et al.* (2014), Valentina *et al.* (2015), and a few others.

The reservoir represented rich zooplankton diversity. Biodiversity generally refers to the variety and variability of life on Earth. Biodiversity typically measures variation at the genetic, species, and ecosystem levels. Terrestrial biodiversity tends to be greater near the equator, which seems to be the result of the warm climate and high primary productivity. Biodiversity is not evenly distributed; rather, it varies greatly across the globe as well as within regions. Among other factors, the diversity of all living organisms depends on temperature, precipitation, altitude, soils, geography, and the presence of other species (Mathur *et al.,* 2008; Kulkarni, 2009).

CONCLUSION

According to physicochemical status and zooplankton diversity, the reservoir seems to be productive but moderately polluted due to agricultural run-off and domestic sewage, which indirectly suggests the beginning of eutophication. The present study also provides an insight into the distribution, abundance, diversity, and ecology of zooplankton in reservoirs. The investigation generated some important baseline data on the water quality and zooplankton plankton community structure of the reservoir. These data would be helpful in planning future policy decisions on using the reservoir as a drinking water source as well as in the better conservation and management of the precious wildlife in the world-famous sanctuary. Though the water body under investigation is not severely polluted, it requires careful monitoring in the future to maintain the quality of the water by proper means. The supervision of experts and remedial measures are essential for rehabilitation and conservation for a long period of duration.

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Cosmetic Technology

1

Cosmetofood: Aam Panna Brightening And Hydrating Serum

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ABSTRACT:

The present study was conducted to formulate and evaluate dark spots on the face by using water lily extract & niacinamide in the formulation. This serum is formulated on the ideology of Cosmetofood. The serum was prepared with natural active agents. The concentration of active agents was kept in a range of 0.5%, 0.8 %, and 1%, each was incorporated and three combinations of each were prepared.

The moisturizing activity was determined by using a corneometer. The moisture content of the skin increases with the continuous use of the product. In the present work, the de-pigmentation formulation gave satisfactory good moisturizing and antioxidant properties and this is achieved by the use of natural actives like water lily extract. The serum was prepared by the conventional procedure and all the factors, and parameters such as pH, viscosity, stability, and microbial analysis were determined. It was also kept accelerated by stability testing for 30 days. The moisturizing property is determined by the coreometer and conductivity method. Then the product i.e. serum was applied on human volunteers and a progressive effective result was found. Thus photographs of before application and after application were taken out from Photographs: Thus the formulation F3 of serum containing 1% water lily extract and 0.5% niacinamide was found stable and gave the most effective results.

KEYWORDS:

Water lily extract,niacinamide,cosmetofood,serum

INTRODUCTION:

SERUM

"Serums are thin-viscosity topical products that contain concentrated amounts of active ingredients," Skin Serum is a skincare product you can apply to your skin after cleansing but before moisturizing with the intent of delivering important constituents directly into the skin. The serum is particularly suited to this task because it's made up of lower motes that can access deeply into the skin and deliver veritably high attention to active constituents. This makes them a great tool for targeting specific skincare enterprises, like saturation, and signs of ageing.

What Is a Face Serum?

A face serum is a formula that contains a high volume of active constituents. Serums can specifically target one concern or a combination of them. Your skin will reap the prices of a concentrated formula that will give your complexion the boost it needs. It's recommended to add a facial serum to your skincare

routine when you're in your 20's, it's impeccably fine to begin using a serum in your 20s if you feel it's necessary.

Different Types of Face Serums

Whether you want to target fine lines, dark spots, or dry skin, a duly formulated serum can deliver results with regular use. The following are the most common types of facial serums you'll find.

1. Anti-Aging Serums: Anti-aging serums can help or lessen the appearance of fine lines & wrinkles. These generally contain actives similar to Niacinamide & Retinol.
2. Brightening Serums: Brightening serums are designed to target hyperpigmentation and reduce abrasion. C composites are easier for the skin to absorb than pure vitamin C. In addition

to that, niacinamide also brightens skin-defined in the study as " reflection of light from skin's face." This leads to a more radiant, vibrant complexion.

3. Hydrating Serums: Hydrating serums give a redundant boost of humidity for your skin. They are used in confluence with regular moisturizers. Hyaluronic acid is the most common component set up in hydrating serums. Hyaluronic acid's appeal lies in its capability to hold,000 times its weight in water.
4. Exfoliating Serums: To help your skin from falling victim to dullness and flight, slipping regularly is encouraged. Exfoliation comes in two forms: physical and chemical. A physical exfoliant is a coarse mite that can be used to manually remove dead skin cells. A chemical exfoliant, meanwhile, is a less abrasive volition that's generally produced in serum form. Exfoliating serums contain alpha-hydroxy acids(AHAs), beta-hydroxy acids(BHAs), or poly-hydroxy acids(PHAs).
5. Firming Serums: Firming serums target sagging skin. A drop in collagen, elastin, and ceramide(humidity) product in your more mature times is frequently the cause behind loose, coarse skin. Therefore, incorporate a serum that's formulated with constituents that will promote collagen product (similar as retinol and niacinamide) and boost hydration(like hyaluronic acid).

COSMETOFOOD:

Cosmetofood delivers tastable organic, vegan free and skin-friendly cosmetics. Cosmetofood is a concept with the medical virtuousness of ECOCERT and COSMOS-certified constituents. Cosmetofood has always believed in the testament of sustainability with a vision to transfigure the beauty industry., Cosmetofood distinguishes itself with an authentic handwrought skincare line that elevates glorifying and remedial goods to the skin texture without any side effects. These products contain vegetable extracts, herbs, fresh fruits and essential oil without dangerous constituents. Cosmetofood enables you to find the proper ritual for indefectible skin. Crafting non-toxic products made from organic constituents with recyclable packaging; is the Cosmetofood approach in helping us to live an authentically green life.

MATERIALS AND METHODS

Aam panna hydrating and lightening serum is prepared by using following ingredients and equipment.

Materials:

List of Ingredients

1. Water
2. Disodium Edta-Finar
3. Glycerine-Oleon
4. Dub Diol-St.Dubois
5. Aquepec HV 701 EDR
6. Sepinov EMT'10-Seppic
7. Euxyl k 830-Ashland
8. Naoh-Finar
9. Water lily Extract-Seppic
10. Niacinamide-Lasons
11. Aam Panna FLV 8071
12. Yellow+Green [Nileekon]

List of Equipment

1. Precision balance: CA series contech
2. Mechanical stirrer : Shettal Scientific industry Pvt It.,Mumbai
3. pH meter: Digital Model 111E-E-1 Electronic india

4. Brook field Viscometer : S.M.S Scientific Industry Pvt.Ltd. Mumbai (DV-E-version1, B-34/03)

Method:

Aam Panna Hydrating and Lightening Serum

1:Preparation of serum base

All formulations of batches were prepared according to the Table 1.Entire procedure is Cold Process Method. The desired concentration of Disodium EDTA were weight and dispersed in water.On continuous stirring Glycerin and Dub Diol were added into the water.In this same mixture Aquepec HV 701 EDR was sprinkled over it.Stirred the mixture until it dissolved properly.Then Sepinov EMT 10 was added in continuous stirring..Euxyl k 830 is a preservative which was added in last.pH is balanced out by using NaOH.Prepared formulation were filled in suitable container and labeled accordingly.These preparation is further evaluated.

Table 1:Base formulation of serum

Sr No	*Ingredients*	*BF1 [100%]*	*BF2 [100%]*	*BF3 [100%]*
1.	Water	93.9	94.6	95.1
2.	Disodium EDTA	0.1	0.1	0.1
3.	Glycerin	3	2	1.5
4.	Dub Diol	1	1	1
5.	Aquepec HV 701 EDR	0.3	0.2	0.3
6.	Sepinov EMT 10	0.3	0.4	0.5
7.	Euxyl k 830	1	1	1
8.	NaOH	0.4	0.4	0.4

[BF =BASE FORMULATION]

PROCEDURE

All ingredients belong to a single phase an are water soluble.

1. Mix disodium EDTA & glycerine & dub diol in water under stirring.
2. Sprinkle Agupec HV 701 EDR to mixture
3. Stir until dispersed properly.
4. Add Sepinov EMT 10 into the mixture & mix well
5. Add Euxyl k 830 & Adjust pH with Naoh. **2:Preparation of serum with water lily extract** Entire procedure is the same as Table:1.

Water Lily Extract were added before addition of preservative and pH balancer.Prepared formulation were filled in suitable container and labeled accordingly.These preparation is further evaluated.

Table 2:Base formulation with active[water lily extract]

Sr No	*Ingredients*	*A1F1 [100%]*	*A1F2 [100%]*	*A1F3 [100%]*
1.	Water	95.1	95.1	95.1
2.	Disodium EDTA	0.1	0.1	0.1
3.	Glycerin	1.5	1.5	1.5
4.	Dub Diol	1	1	1
5.	Aquepec HV 701 EDR	0.3	0.3	0.3
6.	Sepinov EMT 10	0.5	0.5	0.5
7.	Euxyl k 830	1	1	1
8.	NaOH	0.4	0.4	0.4
9.	Water Lily Extract	0.5	0.8	1

[A1F=formulation with water lily extract]

PROCEDURE

All ingredients belong to a single phase and are water soluble.

1. Mix disodium EDTA & glycerine & dub diol in water under stirring.
2. Sprinkle Agupec HV 701 EDR to the mixture.
3. Stir until dispersed properly.
4. Add Sepinov EMT 10 into the mixture & mix well
5. Add Water Lily Extract and blend it properly
6. Add Euxyl k 830 & Adjust pH with Naoh..

3.Preparation of serum with niacinamide:

Entire procedure is the same as Table:2.

Niacinamide were added before addition of preservative and pH balancer.Prepared formulation was filled in suitable container and labeled accordingly.These preparation is further evaluated.

Table 3:Base formulation with active[niacinamide]

Sr No	*Ingredients*	*A2F1 [100%]*	*A2F2 [100%]*	*A2F3 [100%]*
1.	Water	93.9	95.1	94.1
2.	Disodium EDTA	0.1	0.1	0.1
3.	Glycerine	1.5	1.5	1.5
4.	Dub Diol	1	1	1
5.	Aquepec HV 701 EDR	0.3	0.3	0.3
6.	Sepinov EMT 10	0.5	0.5	0.5
7.	Euxyl k 830	1	1	1
8.	NaOH	0.4	0.4	0.4
9.	Water Lily Extract	1	1	1
10.	Niacinamide	0.2	0.5	1

[A2F=formulation with niacinamide]

PROCEDURE

All ingredients belong to a single phase,all ingredients are water soluble.

1. Mix disodium EDTA & glycerine & dub diol in water under stirring.
2. Sprinkle Agupec HV 701 EDR to the mixture.
3. Stir until dispersed properly.
4. Add Sepinov EMT 10 into the mixture & mix well.
5. Add Water Lily Extract and blend it properly.
6. Then add Niacinamide and mix it properly.
7. Add Euxyl k 830 & Adjust pH with Naoh.

4:Preparation of final serum

Entire procedure is the same as Table 3.

Flavor and color were added in the last.Prepared formulation was filled in a suitable container and labeled accordingly.These preparation is further evaluated.

TABLE 4:Final formulation of serum

Sr No	*Ingredients*	*F1 [100%]*	*F2 [100%]*	*F3 [100%]*
1.	Water	93.9	95.1	94.1
2.	Disodium EDTA	0.1	0.1	0.1
3.	Glycerine	1.5	1.5	1.5
4.	Dub Diol	1	1	1
5.	Aquepec HV 701 EDR	0.3	0.3	0.3

6.	Sepinov EMT 10	0.5	0.5	0.5
7.	Euxyl k 830	1	1	1
8.	NaOH	0.4	0.4	0.4
9.	Water Lily Extract	1	1	1
10.	Niacinamide	0.2	0.5	1
11.	Aam Panna FLV 8071	0.1	0.1	0.1
12.	Yellow+Green	Q.S	Q.S	Q.S

[F=Final Formulation]

EVALUATION OF SERUM [F1,F2,F3]

1] IN VIVO EVALUATION OF PARAMETERS

2] IN VITRO EVALUATION

A] DETERMINATION OF PHYSICAL PARAMETER:

Appearance: Visually appearance of the formulation observed.

Color: Color of the formulation check visually.

Consistency: Consistency was checked whether it's satisfactory or poor or good.

Table 5:Determination of physical parameter

Sr No.	***Parameter***	***F1***	***F2***	***F3***
1.	Appearance	++	+	+++
2.	Colour	+	++	++
3.	Consistency	+	++	+++
4.	Odour	++	++	+++

GOOD = +,BETTER = ++,BEST = +++

B] DETERMINATION OF pH

Principle: Serums are used for topical application so their pH should be similar to that of with the skin. The skin these acidic mantle and the pH of the face wash as per standard should be in the range of 4-5.9 **Apparatus:** pH meter

Procedure: Take 1 gm of sample in beaker. Then with the help of pH meter reading was taken.

Table 6:Determination of pH of F1,F2,F3

Sr.No	**No. of days**	**Ph Value**		
		F1	**F2**	**F3**
1	1 st day	5.5	5.6	5.6
2	15 th day	5.6	5.7	5.7
3	30 th	5.7	5.8	5.8

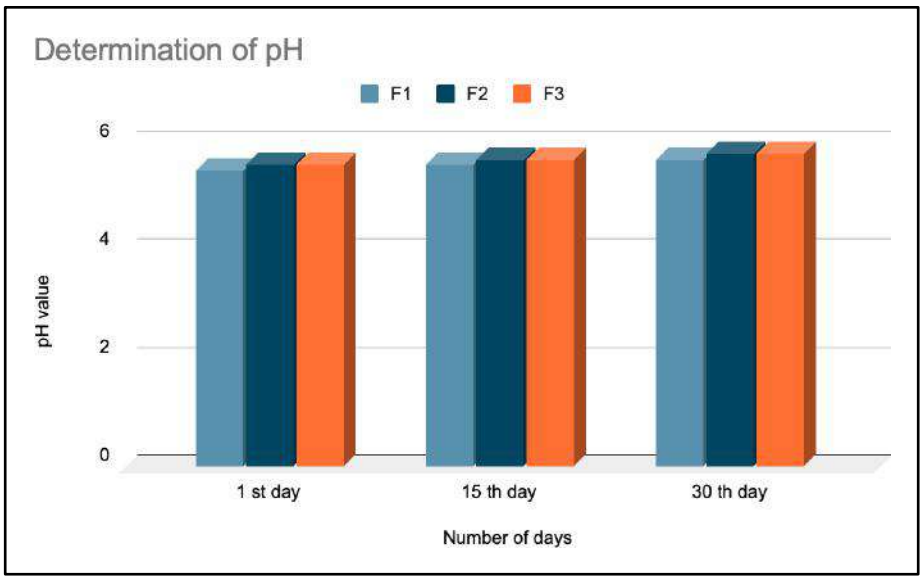


Figure 1: Graphical representation of pH of F1,F2,F3

C] DETERMINATION OF VISCOSITY

Apparatus: Brookfield viscometer

Principle: The viscosity is the most important parameter in the evaluation of cosmetic products. Viscosity governs the many properties such as spreadability, pourability of the product from the container.

As viscosity is affected by many factors such as change in temperature, change in manufacturing condition, quality of the raw material. Hence it is very important to measure the viscosity of the product. **Proceduce:** The viscosity of serum was determined by using spindle no.4 using a brook field viscometer then all the operating conditions were set up. Then five readings were taken at different rom and average of there will be the final reading. Viscosity was measured at 6 rpm in ops.

Table 7:Determination of Viscosity of F1,F2,F3

Sr.No	No. of days	Viscosity		
		F1	F2	F3
1	1 st day	14360	13730	13466
2	15 th day	14375	13759	13466
3	30 th	14375	13759	13567

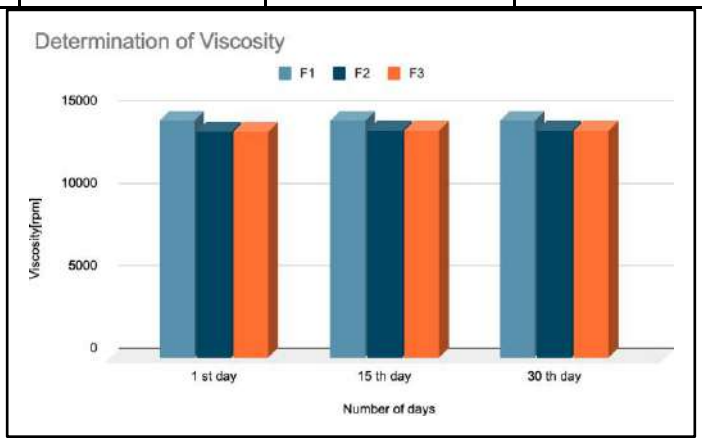


Figure 2: Graphical representation of viscosity of F1,F2,F3

D] DETERMINATION OF SPREADABILITY TIME

Principle :It is very important for any cosmetic product that after application the product must be easily spread over the skin. Spreadability is affected by many factors such as viscosity, temperature etc. The spreading time must be very less.

Apparatus:The apparatus consists of a wooden block, with a movable glass slide with one end tied to a weighted pan rolled on pulley.

Procedure: 2 Gm of serum sample was placed on a surface. A slide was attached to a pan to which 20 gm weight was added. The time (seconds) required to separate the upper slide from the surface was taken as a measure of spreadability.

T**able 8:Determination of Spreadability Time of F1,F2,F3**

Sr.No	No. of days	Surface Area[cm]			Weight	Time
		F1	F2	F3		
1	1 st day	8.1 cm	7.9 cm	8 cm	20 gm	2.1 sec
2	15 th day	8.7 cm	8.3 cm	8.5 cm	20 gm	2.1 sec
3	30 th day	9.1 cm	8.7 cm	8.9 cm	20 gm	2.1 sec

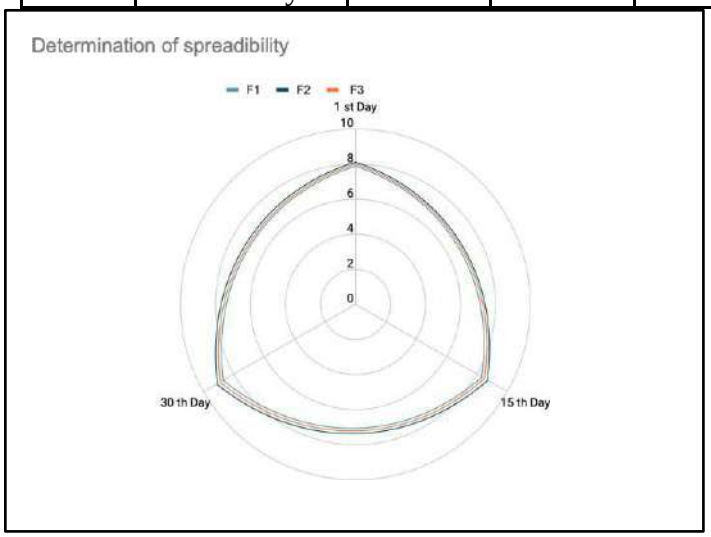


Figure 3:Graphical representation of Spreadability Time of F1,F2,F3

A]DETERMINATION OF MELANIN ACTIVITY

Principle: Mexameter is a spectrometer measurement technique,based on light reflection and absorption.The probe emits three wavelengths of light, chosen to correspond to the different absorption rates of melanin and hemoglobin.

Apparatus: Mexameter equipped with probe.

Procedure: First, a probe is placed on a sample and emits three light wavelengths. Then, a receiver measures the amount of light the skin reflects back.A body part with more light absorption has higher melanin levels and is darker in color compared to one with more reflection.The results are shown in 1 second as index numbers between 0 and 999. The probe allows the measurement to be made quickly (1 second). The probe head is spring loaded so that a constant pressure is provided.

Table 9:Determination of Melanin Activity of F1,F2,F3 by using Mexameter.

Sr.No	No. of days	Melanin Deposition[nm]
1	Before App	397 nm
2	1 st day	345 nm

3	5 th day	339 nm
4	10 th day	330 nm
5	15 th day	325 nm
6	20 th day	323 nm
7	25 st day	321 nm
8	30 th day	218 nm

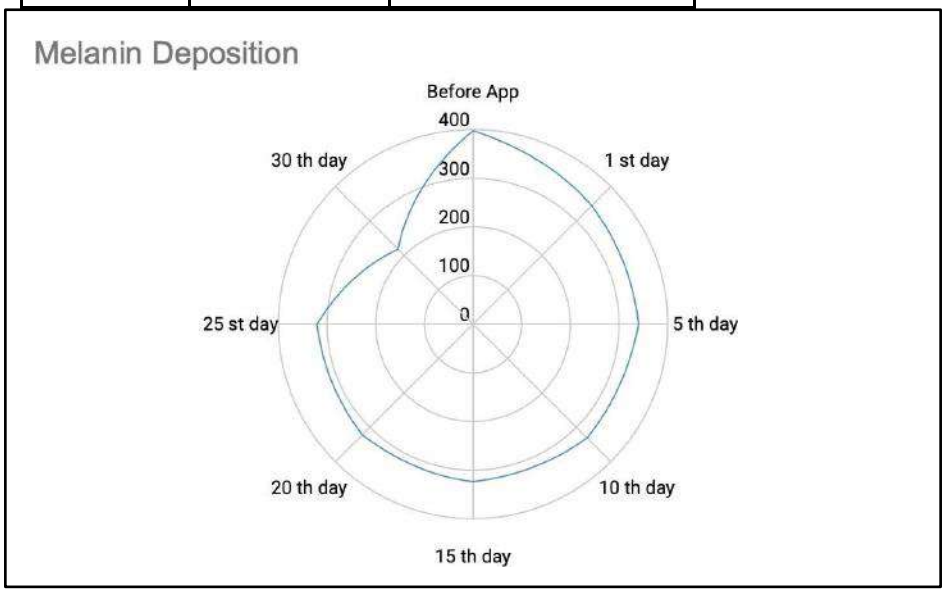


Figure 4:Graphical representation of Melanin Activity of Serum.

B]DETERMINATION OF MOISTURE CONTENT

Principle: Corneometer is device which is equipped with a moisture sensitive probe which determines the accurate moisture content of stratum corneum. Hence it plays an important role in Altering the moisturizing activity of the product on the stratum corneum after its application on skin.

Apparatus: Corneometer equipped with a probe.

Procedure: The volunteers were selected and the probe of the corneometer was applied onto the selected part of skin before application of the product and the reading was recorded. Then the selected part of skin was rinsed with product allowed to dry properly and again the probe was applied onto the skin and reading was recorded. The voluntree were allowed to wash the selected area of skin with the product twice a Day and then the same procedure was followed for 14 days.

Table 10:Determination of Moisture Content of F1,F2,F3 by using Corneometer.

Sr.No	No. of days	Moisture Activity
1	EFORE APP	101
2	1 st day	102
3	2 nd day	102.3
4	3 rd day	103
5	4 th day	103.5

6	5 th day	103.6
7	6 th day	104.2
8	7 th day	105.2

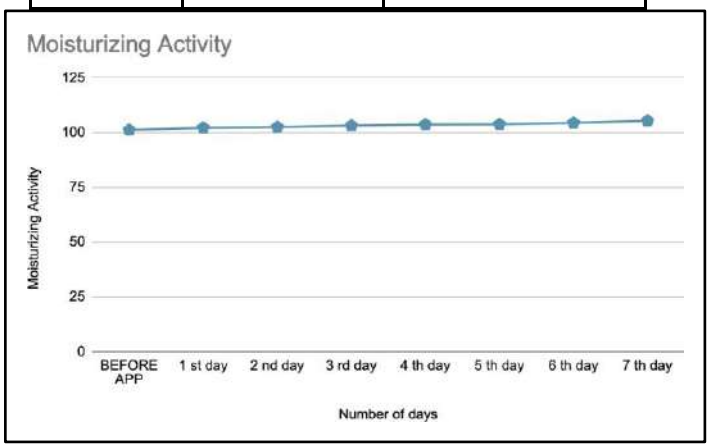


Figure 5:Graphical representation of Moisture Content of F1,F2,F3

C]SKIN IRRITATION

The skin irritation was carried out on human volunteers. For formulated Serum, five volunteers were selected and 1.0 gm of formulated serum was applied on an area two square inches to the back of the hand. The volunteers were observed for lesions of irritation.

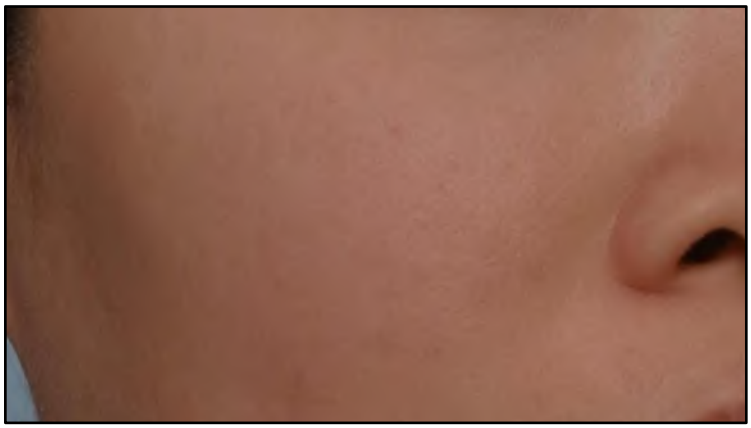
Table 11:Determination of Skin Irritation Test of F1,F2,F3

Sr.No	PARAMETER	SKIN IRRITATION TEST
1	F1	No Irritation
2	F2	No Irritation
3	F3	No Irritation

D] Photographic evaluation:

Photographic evaluation is carried to see the effect of the product visually. In case of determination of hydrating & lightning activity photographic evaluation was adopted. In this method the Photograph of skin before and after application for 6 weeks of skin were taken out and the effect of the product was determined.

FIGURE 6:The Skin Before Application of the SERUM.



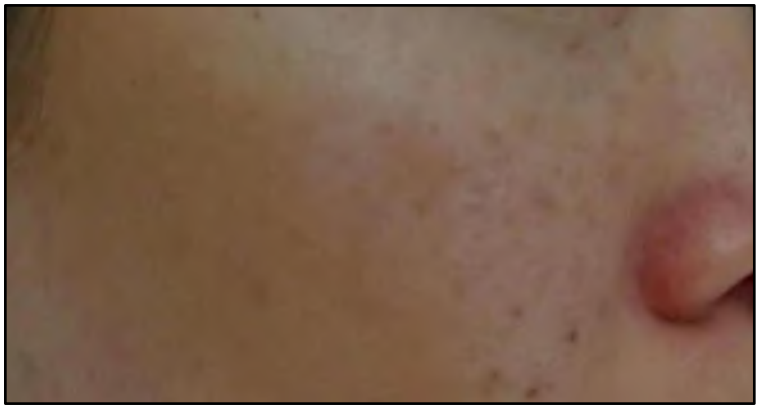


Figure 7:The Skin After Application of the Serum.

RESULT: The above photograph shows that the dark spots minimized within 15 days of application of the product.

CONCLUSION

This study aimed at preparing a stable formula Serum, rich with Water Lily Extract that contains eco friendly ingredients to reduce the risk of chemicals. Results showed that all ingredients used to formulate the serum were found to be safe and the physiochemical evaluation showed ideal results.

Stability studies showed a stable homogeneous appearance during six months of storage at different temperatures (4-8°C, 40°C and at ambient temperature. However, formula 3 (F3) gave optimum stability, especially the stability of olive leaves extract. Further research is required to improve its quality especially on the conditioning performance.

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2

Formulation and Evaluation of Layered Lipstick: Innovating Cosmetic Design with Multiple shade

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Abstract
In the realm of cosmetics, innovation is the key to meeting the diverse needs and desires of consumers. Layered Lipstick stands out as a promising innovation in cosmetics, introducing a harmonious blend of multiple shades within a single stick. Beyond merely imparting colour, it serves as a canvas for self-expression. This research delves into the formulation and development of Layered Lipstick, promising to revolutionize lip makeup application and enhance confidence and style through its seamless fusion of colours and natural radiance. In response to persistent challenges in achieving seamless lipstick shade gradation, this paper presents the Layering Lipstick Series, a groundbreaking approach characterized by a unique design, advanced formula, and effortless shade gradation.

Key words
Layered Lipstick, multiple shades, innovation, cosmetics.

Introduction

In the dynamic landscape of cosmetics, innovation continually reshapes beauty routines, offering novel ways for individuals to express themselves. Among these innovations, Layered Lipstick emerges as a promising advancement, redefining traditional lip makeup with its seamless blend of multiple shades in a single application. This research paper delves into the formulation and development of Layered Lipstick, exploring its transformative impact on lip aesthetics and consumer experiences.

Layered Lipstick transcends conventional makeup norms by introducing a harmonious blend of multiple shades within a single stick. Its allure lies not just in its ability to adorn lips with vibrant hues but in its capacity to transform the way individuals perceive and engage with lip colour. With the Layering Lipstick series, the mundane act of applying lipstick evolves into an art form, promising a seamless fusion of colours and a natural radiance that amplifies confidence and style.

The core objective of this research is to explore the formulation and development processes behind Layered Lipstick, unravelling the science and artistry that culminate in its exquisite blendability and rich pigmentation. Through meticulous analysis, we aim to elucidate the benefits of Layered Lipstick, showcasing its ability to impart plumpness, volume, and natural luminosity to lips, thereby enhancing facial features and empowering individuals to express their unique identities with confidence.

The burgeoning popularity of cosmetics, coupled with evolving consumer preferences, underscores the immense market potential of Layered Lipstick. Recent market research indicates significant growth in the colour cosmetics segment, with lipstick emerging as a cornerstone product in the makeup arsenals of women worldwide. As consumer demand for versatile and innovative beauty solutions continues to soar, Layered Lipstick occupies a pivotal niche, offering a transformative experience that resonates with individuals across diverse demographics and lifestyles.

Moreover, insights into consumer behaviour reveal a significant appetite for lipstick, with a substantial majority of women incorporating it into their daily beauty routines. This

underscores not just the market viability of Layered Lipstick but also its intrinsic appeal as a staple product that transcends fleeting trends, catering to the enduring desire for self-expression and enhancement.

In conclusion, the formulation and development of Layered Lipstick represent a convergence of science, art, and consumer-centric innovation. As we embark on this research journey, we endeavour to unravel the mysteries of Layered Lipstick, uncovering its secrets and unlocking its transformative potential in the realm of beauty and self-care.

Objectives

This research aims to explore the formulation and development of Layered Lipstick, focusing on the innovation, versatility, and user experience offered by the Layering Lipstick series. The objective is to analyse the effectiveness of this lipstick line in delivering a rich colour gradation with just one swipe, while also enhancing lip volume and providing a natural finish.

Specifically, the research seeks to:

1. Investigate the formulation process of the Layering Lipstick series, including the selection of colour shades and the blending techniques employed to achieve seamless colour transitions.
2. Evaluate the user experience of applying the Layering Lipstick, assessing factors such as ease of application, blendability of colours, and overall satisfaction with the product.
3. Examine the visual impact of the Layering Lipstick on lip appearance, particularly its ability to create plump, voluminous lips with a natural finish.
4. Explore the versatility of the Layering Lipstick series in catering to a wide range of style preferences, from subtle elegance to bold statements, and its compatibility with different skin tones.
5. Assess the market perception and consumer acceptance of Layered Lipstick formulations, focusing on the Layering Lipstick series as a case study.By addressing these objectives, the research aims to contribute to a deeper understanding of the formulation and develop layered lipstick.

Sr. No.	Phase	Ingredients	Quantity for 100% (in gm/layer)	Function
1	A	Cocoa butter	8	Texture
2		Octyl dodecanol, Euphorbia Cerifera (Candelilla) Wax, Isostearyl Isostearate, Stearyl Behenate, Polyhydroxystearic acid, Olea Europaea (Olive) Fruit Oil, Stearyl Alcohol, Behenic Acid, Stearyl Stearate, Cetyl Stearate	6	Moisturizer
3		Avocado oil	6	Oil
4		Wheat germ oil	6	Oil
5		Hydrogenated Vegetable Oil	5	Oil thickener
6		Bees wax	4	Thickener
7		Ozokerite wax	3	Thickener
8		Carnauba wax	3	Thickener
9		Candelilla wax	3	Thickener
10	B	Pigment	10	Colourant

11		Castor oil	5	Oil
12		Dioctyldodecyl Dimer Dilinoleate (and) Propanediol	3.5	Film former
13	C	Trimethylsiloxysilicate (and) Isododecane (and) Alcohol (and) Hydrogenated Rosin	14	Emollient
14		Hydrogenated Polydecene	8	Emollient
15		Pentaerythrityl Tetraisostearate	7	Emollient
16		Hydrogenated Polyisobutene	6	Emollient
17		Vanilla	1.5	Flavour
18		Euxyl k 830	1	Preservative

Formulation and moulding techniques

The essence of the Layered Lipstick design centres on a rectangular lip bar housing multiple layers, each showcasing unique shades. The formulation process entailed blending a diverse range of waxes, including beeswax, ozokerite wax, carnauba wax and candelilla wax.

Additionally, the incorporation of cocoa butter and oils such as avocado oil, wheat germ oil, and castor oil played a pivotal role in establishing the ideal foundation for the Layered Lipstick.

Through the integration of specialty ingredients, the formulation has attained exceptional quality and attributes, including water resistance, extended durability, effortless spreading, moisturizing properties, sun protection, pigment dispersion, flawless application, and the versatility to achieve either a matte or glossy finish.

A) Formulation Process:

Weigh phase B ingredients in a dish or bowl, ensuring uniform pigment distribution. Heat phase A ingredients in a heat-resistant beaker until waxes are melted. Combine phase B with phase A, stirring thoroughly, and remove from heat. Prepare phase C ingredients in a separate beaker, then add to phase AB mixture, stirring well to ensure homogeneity.

B) Moulding Process:

Grease the lipstick mould with oil. Pour the first layer (phase ABC) into the mould and allow it to cool briefly. Add the second layer (Phase AB'C), where 'B' represents a different shade, ensuring seamless integration.

Evaluation Methods

The evaluation of the Layered Lipsticks were carried out by the methodology of IS 9875 (1990).

Colour and Texture:

Formulated lipsticks were checked for colour, glossy and smooth texture.

pH:

The pH of formulated Layered Lipsticks was determined using digital pH meter (Systronics,802).

Determination of Melting Point:

Determination of melting point is an important parameter for lipstick formulation as it is an indication of the limit of safe storage. The melting point of formulated lipstick was determined by capillary tube method. Approximately 50 mg of lipstick sample

was taken and melted and filled into glass capillary tube opened at both ends. Capillary was cooled with ice for 2h and fastened with thermometer. Thermometer with capillary was deep in the beaker containing full of water which was placed on heating plate with magnetic stirrer. Heating and stirring was started slowly at fixed speed. The temperature at which material moves along the capillary tube was considered as melting point.

Breaking Point:

This test was carried out to find out the value of maximum load that lipstick can withstand before it breaks. This test gives strength of lipstick. Prepared Layered Lipstick was held horizontally in a socket inch away from the edge of support. The weight was gradually increased by a specific value (10 gm) at specific interval of 30 second and weight at which breaks was considered as the breaking point.

Softening Point:

Lipstick should be able to withstand range of conditions to which it will be subjected in the consumer's handbag. It should be resistant to varying temperature conditions and be just as easy to apply in hot and as in cold weather. Softening point of lipstick was determined by Ring and Ball method.

Ring and Ball method:

A ring or support orifice is taken, and prepared Layered Lipstick was inserted into it. Extra mass above and below the orifice was removed using a sharp blade leaving a tablet of lipstick fitted into the ring. This was placed in refrigerator (6°C) for about 10 min. Ring was tied onto a stand. A beaker containing 500 mL water at room temperature is placed on a hot plate with magnetic stirrer. A steel ball was delicately placed on the lipstick tablet. The bar with support was then inserted into the beaker till it submerged into it. Heating and slow agitation was then begun. Temperature was monitored using a thermometer. The temperature at which the lipstick mass and steel balls were loosed and falls to the bottom of the beaker was noted as softening point of lipstick. Surface anomalies: This was studied by the surface defects, such as formation of crystals on surface, contamination by moulds, fungi, formation of wrinkles, exudation of liquid substances and of solid fatty substances, etc.

Aging stability:

Prepared Layered Lipsticks were stored at refrigerator temperature (4oC), room temperature (20-25oC) and high temperature (30-40oC) for 1h. Various parameters such as bleeding, streaking, catering, and blooming were observed.

Perfume stability:

The prepared Layered Lipsticks were tested after 10 days, to record fragrance.

Result and Discussion

A) Formulation and Objective

The formulation and evolution of Layered Lipstick, as demonstrated by the innovative Layering Lipstick series, signify a significant advancement in cosmetic design. Through a meticulous blend of waxes and oils, the formulation achieves water resistance, durability, moisturization, sun protection, and versatile finishes. This unique formulation enables seamless blending of multiple shades, providing users with a customizable lip makeup experience. The transformative qualities of Layered Lipstick empower individuals to confidently express their unique identities, underscoring the cosmetics industry's ongoing commitment to innovation and consumer satisfaction.

B) Evaluation of Layered Lipstick

Results showed that all evaluation parameters of Layered Lipstick are resemble with standard values. (Table 2.)

Table 1: Evaluation of Layered Lipstick

Sr. No.	Parameter	Layered Lipstick Formulation	Standard Values
1.	Color	Multi colour	-
2.	Texture	Smooth	Smooth
3.	pH	6.2	6.4
4.	Melting point	60-64°C	60-66°C
5.	Breaking point	190 gm	-
6.	Softening point	60°C	50-60°C
7.	Surface anomalies	No defects	No defects
8.	Aging stability	Smooth	Smooth
9.	Perfume stability	+++	+++

Conclusion

In conclusion, the formulation and development of Layered Lipstick, exemplified by the innovative Layering Lipstick series, mark a significant advancement in cosmetic design. Through meticulous blending of waxes and oils, this lipstick achieves water resistance, durability, and versatile finishes. Evaluation parameters confirm its adherence to standards, underscoring its transformative potential in the cosmetics industry. Layered Lipstick offers users a customizable experience, empowering them to confidently express their unique identities.

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3

The Persimmon fruit extract: Its Dematological and Cosmetics Benefits

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Abstract
Persimmon is fleshy fibrous tropical, deciduous fruit belonging to *Ebenaceae* family. It is commonly cultivated in warm regions of the world including China, Korea, Japan, Brazil, Turkey, and Italy. In 2007, the global production of persimmon reached over 3.3 million tons, with 70.0 % from China, 10.0 % from Korea and 7.0 % from Japan. The persimmon is not so popular in European communities but its demand is increasing owing to consumer's awareness regarding its hidden health promoting potential. Mediterranean region is also suitable for persimmon production that has reached up to 110,000 tons.(1)
Generally, over 400 species of persimmon are planted globally. Among these, *Diospyros kaki*, *Diospyros virginiana, Diospyros oleifera,* and *Diospyros lotus* are of significant importance. It is interesting that *D. kaki* (Japanese persimmon) is the most promising specie. The popular varieties grown in Japan.(1)
Diospyros kaki belonging to family Ebenaceae, commonly known as persimmon is used as a medicinal plant in Chinese traditional medicine since many years for different ailments including cosmetics and dermatologic applications. Traditionally this plant is used to treat different skin conditions including pimples, skin eruptions and eczema. Present interest has been focused toward use of natural bioactive compounds in various curative and beautifying applications in dermatological and cosmeceutical disciplines.(2)

1. Introduction

A potent antioxidant and loaded with vitamins, Persimmon Extract has traditionally been used in Japan for its naturally purifying and deodorizing benefits. This prized, natural ingredient is particularly effective in eliminating Nonenal, the source of hormonal imbalance or aging body odor, leaving skin squeaky clean and freshly hydrated.(3)

Persimmon is enriched with many nutritious and bioactive components including proteins, sugar, lipids, vitamin A, vitamin B6, vitamin B12, vitamin D, ascorbic acid(AA), Vitamin E polyphenols, flavonoids and carotenoids. Elemental micronutrients present in persimmon fruit include potassium, sodium, iron, calcium and many others. The fruit have been used as a key ingredients in some marketed cosmetic products including soaps, deodorizing and purifying body lotion, body wash, skin toner and body serum (Mirai Clinical, 2017). Different reviews have been published about reported pharmacological activities and phytoconstituents profile of various parts of this plant, with very limited or no emphasis on its potential use in dermatology and cosmetics. This review describes available data about potential utilization of different parts of *D. kaki* and its bioactive phytoconstituents in different dermatological and cosmeceutical applications.(2)

Scientific data has revealed an excellent position of *D. kaki* in both dermatology and cosmetic discipline making it a valuable choice in respective field. Active principles from different plant parts have shown to possess anti-inflammatory, antiallergic, photo-protective, deodorant and anti-wrinkle effects with appreciable activities against tyrosinase, elastase, and collagenase enzymes. Promising antioxidant activity and skin whitening potential, augmented by reduction in sebum contents, and reduction in size and number of skin pores make it a suitable choice as cosmetic ingredient. Data has been summarized and presented on

available molecular mechanism that can contribute toward phytoconstituents usage in cosmetics and dermatology mediated by different cellular pathways. Crude extracts and some of phytochemical obtained from this plant such as isoquercitrin and hyperin have better reported activities than well-known cosmetic ingredients viz., arbutin, kojic acid and hydroquinone with possibility of having no side effects. Photo protection against degenerative effects of UVA, UVB and gamma radiation can help skin to fight well against oxidative stress and reactive oxygen species.[4] Further investigation need to be directed toward human subjects for evaluation of these reported activities for obtaining optimum commercial and industrial benefits from this valuable plant.[2]

2. Phytochemicals of and cosmetics interest obtained from *Diospyros kaki*

2.1 Phenolic acids

Phenolics (or phenolic acids) are widely distributed aromatic secondary metabolites in plant kingdom. They contain an aromatic hydrocarbon and one or more than one functional hydroxyl (or carboxylic acid) group attached to it.[16-17] They can be categorized into simple phenols bearing one phenol unit or polyphenols having multiple phenol units in chemical structure. They perform a range of different functions in plants and human being including structural maintenance and protection against oxidative stress disorders such as coronary heart disease, stroke and cancer. [23] [28][31][34-35]

2.2 Flavonoids

Flavonoids, also called bioflavonoids are naturally occurring secondary metabolites of botanical origin having a general structure of 15-carbon skeleton comprised of two phenyl rings and one heterocyclic ring. More than 8000 phytoconstituents have been identified with this characteristic flavonoid structure. Basic benzo-γ-pyrone ring is subjected to different combinations of hydroxyl, methoxyl, and *O*-glycosyl group substituents resulting in numerous individual flavonoids.[22][29-30]

Flavonoids are further classified into twelve different subgroups, however six of them have gained a significant dietary importance, including anthocyanidins, flavan-3-ols, flavonols, flavones, flavanones, and isoflavones[33-34]. In *D. kaki* following examples are found in different parts of the tree including (I) anthocyanidins *e.g.* cyanidin, (II) flavan-3-ols *e.g.* (+)-catechin, (−)-epicatechin and (−)-epigallocatechin, (III) flavonols *e.g.* kaempferol (H), quercetin and their glycosides. [12-14]

2.3 Carotenoids

Carotenoids are colored, fat soluble pigments generated as secondary metabolites in fruits, vegetables, algae, fungi, and some microbes. Most important carotenoids include beta-carotene, lycopene, lutein, and zeaxanthin. Carotenoids can be categorized into two groups *i.e.*, "xanthophylls" which are oxygenated carotenoids and "carotenes" being non-oxygenated. Approximately 700 carotenoids have been identified with around 100 being considered for their dietary benefits. They have wider applications in food, cosmetics and nutrition because of their color producing tendency and free radical scavenging activity. Peroxyl radicals, singlet molecular oxygen and superoxide anions are the major ROS formed in human skin exposed to UV irradiation, which may result in degradation of lipids, proteins and nucleic acids. Such degradation outcomes in various skin pathological conditions such as erythema, pre-mature skin aging and even dermatological carcinomas. β-Carotene also known as "provitamin A" which resides in the skin imparting a golden yellow color, have no doubt a selective cosmetic value. Lutein and zeaxanthin provide protection to retina against oxidative damage to UV light. Lycopene can reduce erythema induced by UV light. [19][24]

2.4 Hydrolysable tannins

Another group of bioactive phytoconstituents present in persimmon are tannins. Tannins are comprised of either gallic acid subunits (e.g. hydrolysable tannins), flavone subunit (non-

hydrolysable or condensed tannins) or phloroglucinol subunits (phloro-tannins)[5]. Tannins from different sources have been studied for their antiviral, antioxidant, pediatirc dermatoses, anti-inflammatory and radioprotective effects. Tannins have been used medically for many years and their importance in dermatological application have gained significant importance because of their astringent effects, management of superficial skin conditions, weeping, inflammation and itching with acceptable tolerability. [10][32]

Astringent feeling upon eating persimmon fruit is due to soluble tannins which are released from tannin vacuoles making complex with protein in oral cavity. When these tannins are transformed into insoluble form, the fruit loses its astringent nature considerably. In persimmon major tannin present include flavanoellagitannin (molecule of flavan-3-ol attached with hydrolysable tannin through C-C linkage), procyanidinoellagitannin (proanthocyanidins and ellagitannins) and their degraded products such as gallo-catechin, catechin, catechin-gallate and gallocatechin-gallate.[25-27]

2.5 Terpenoids

Different triterpenoids have been separated from leaves of *D. kaki* including ursolic acid, 19-hydroxy ursolic acid and 19,24-dihydroxy ursolic acid, which demonstrated suppressive activity against stimulus induced super oxide generation and tyrosyl phosphorylation. Other terpenoids reported from leaves of *D. kaki* include lupeol, betulinic acid, betulic acid (Yoshihira et al., 1971) and pomolic acid (Thuong et al., 2008). Coussaric acid and betulinic acid have been separated from leaves of persimmon plant. [18]

2.6 Ascorbic acid, vitamins A, D and E

Ascorbic acid (AA) is hexuronic acid lactone micronutrient being lipophobic in its nature. It cannot be synthesized by human being and hence should be supplied externally from food. AA performs different biochemical functions inside the body including synthesis and maintenance of collagen, immunostimulant, anti-aging, and skin rejuvenating agent, skin whitening effects, neuromodulator, anti-oxidant, free radical scavenger and antiviral. In the skin AA plays a vital role as a substrate for oxidative stressors and hence prevents damage to skin caused by ROS and other reactive oxidants produced as a result of UV exposure.[15][36-37]

3. Dermatological and Cosmetics Benefits

3.1 Anti-inflammatory effects

Inflammation is a vital immune mechanism of innate immunity that protects body against various harmful factors. Inflammation is usually mediated by different exogenous and endogenous stimuli that may activate cellular immune system, which intern can produce some pro-inflammatory cytokines. Cyclooxygenase-2 (COX-2) in human skin, is a main key player in UV-induced inflammation, wrinkle formation, edema, epidermal hyperplasia and carcinogenesis. Antiallergic properties and potential use in prevention of dermatitis Skin is the largest protective organ at the interface between host and environment. It protects from pathogens as a physical barrier and defends our body against different allergens by activating immune system. Mast cells are widely distributed in mammalian tissues and play an important role in regulation of allergic inflammation in different immune mediated disorders. Mast cells upon activation can release histamine and other inflammatory mediators. Dermatitis is a common skin condition characterized by inflamed, red, itchy skin that may become blistered and weepy.[11]

3.2 Antiallergic properties and potential use in prevention of dermatitis

Skin is the largest protective organ at the interface between host and environment. It protects from pathogens as a physical barrier and defends our body against different allergens by activating immune system. Mast cells are widely distributed in mammalian tissues and play an important role in regulation of allergic inflammation in different immune mediated disorders. Dermatitis is a common skin condition characterized by inflamed, red, itchy skin that may

become blistered and weepy. There are different types of dermatitis and all of them are precipitated onto the skin by reacting with allergens or irritants. When allergens or irritants become in contact with skin, they may lead to a skin reaction, this condition is termed as contact dermatitis. A skin damage is usually seen with an irritant while an allergen initiates immune response advancing to allergic reaction. Atopic dermatitis or eczema occurs due to hypersensitivity to certain types of food (*e.g.* cow's milk) and/or allergens. Neurodermatitis is because of irritation to nerve endings down the skin, leading to sever itchy sensation and an irresistible desire to scratch the skin repeatedly resulting in thickening and redness of the skin.[38]

3.3 Anti-radiation activity (protection against photo damage)

Electromagnetic radiation emitted from sun, is comprised of ultraviolet radiation (UVR; 200–400 nm), visible light (400–780 nm), and infrared (IR; 780 nm to 1 mm). International commission on illumination (CIE) divides UVR into three categories: UVA (315–400 nm), UVB (280–315 nm) and UVC (100–280 nm). [6] UVC portion being most dangerous for skin, is entirely absorbed by the upper atmospheric layers. Human body needs a very limited UVA and UVB photons for vitamin D synthesis, longer exposure to UVR may lead to various skin abnormalities including photoaging and photocarcinogenesis through production of ROS, DNA damage, immunosuppression, photo-inflammation.

Polyphenol enriched extracts have been evaluated for their efficacy toward skin cancer with greatly promising outcomes indicating their potential role in preventing or curing different skin cancer condition.[7-9]

3.4 Effects on sebum contents, oil contents, number and size of skin pores

Excessive sebum production and accumulation on the skin may increase the skin pore size. An effective skin cleanser is capable to reduce skin pore size by reducing production rate of sebum and promoting its removal from skin, hence reducing chances of comedones development. Careful face washing helps improve skin lesions and prevents acne development by washing away excessive sebum and avoiding hairfollicular obstruction. Many cosmetic ingredients used in skin cleansers have some unwanted effects, such as sodium lauryl sulphate may irritate the skin. Similarly, retinoid and its derivatives are known to be sever local skin irritants. Natural products usually have lesser side effects, that is why cosmetics industry is going through a shift from synthetic to natural cosmetic ingredients. [20-21]

Extract from *D. kaki* folium, Polygonum cuspidatum, and *Castanea crenata* (DPC) loaded to cosmetic cleanser formulation was evaluated for its effects on skin parameters including number and size of skin pores and removal of sebum from the skin in 23 healthy volunteers. On application of test formulation containing DPC extract, oil contents decreased by 77.3%, number of skin pores were reduced by 24.83% and skin pore size was reduced by 71.43% as compared to the control formulation. The preparation was also capable to remove solidified sebum from skin and can facilitate removal of Demodex mites (causative microbe for rosacea and seborrheic dermatitis) from the skin. Further studies can be directed for evaluation of different formulation containing persimmon extract for their effects on other skin parameters using non-invasive *in-vivo* evaluation techniques. [39]

3.5 Inhibition of melanogenesis (skin whitening effects)

Skin color is usually determined by four chromophoric substances known as carotenoids, hemoglobin, oxyhaemoglobin and melanin, the last being most abundant relatively. Melanin is produced by melanosomes which are present in the skin, eyes, inner ear, and hairs. In human being pigmentation may increase as a result of UV or solar light exposure to the skin, which intern, stimulates melanin production by melanosomes. Melanin provides protection against UV radiations, skin burn and cancer. Melanogenesis is the production of melanin from melanocytes in basal epidermal layer. Every individual usually have a particular

number of melanocyte, however the skin color in not determined by the number of melanocytes, rather its being determined by melanin producing genes.(2)

3.6 Collagenase and elastase inhibition (prevention of wrinkle formation)

Collagen represents 30% of total protein in man with almost same weightage in other animals. Collagen can exist in 27 different types however, type I, II, and III are most prominent in man, comprising approximately 80–90% of total collagen in the body. Some body organs are relatively richer in collagen type-I including dermis, bones, tendon, and ligament while skin, blood vessels and intestine are enriched with type-III. In the skin collagen may be degraded by aging or by activity of collagenase, producing wrinkles. Collagen is produced by mature cells called fibroblasts. Firstly, procollagen is produced by fibroblasts, which is subjected to different modifications including proline and lysine hydroxylation. Cross linkage occurs as a result of proline hydroxylation producing strong collagen fibers. [40]

Skin aging is usually estimated by wrinkles on the face. So extract of *D. kaki* can be used in cosmetic preparation as a natural whitening and anti-wrinkle agent. Persimmon leaves have a long been used in Chinese medicines to treat different skin conditions traditionally including pimples, skin eruption and eczema. These traditional uses can be appreciated momentously by cosmetic and dermatological beneficial profile of *D. kaki*.[41]

3.7 Potent antioxidant activity

Skin aging being a dynamic process depends on both intrinsic and extrinsic factors, resulting in various skin changes at both esthetic and functional levels. Two distinct mechanism of skin aging are chronological aging (determined genetically) and photoaging due to repeated exposure to UV light resulting in microscopic changes in stratum corneum. UV radiations results in generation of ROS leading to oxidative damage and oxidative products which are indicators of oxidative stress. Skin damage caused by ROS is the major factor driving toward photoaging. Skin, acting as a physical barrier between internal body and environment, is also a major target for oxidative stress. It contains numerous biochemical molecules which are prone to oxidative damage induced by ROS, including lipids, proteins, carbohydrates, and DNA. UV radiation exposure is a major contributory factor in photoaging, so preventive strategies may include avoiding sun light exposure or by maintaining cellular redox balance caused by UV radiations. In both cases, i.e. chronological aging or photoaging, utilization of different antioxidants in various skin care products has produced promising result.[25]

Food, especially fruits are a major source of antioxidants for the body. Persimmon fruits is enriched with many antioxidants including polyphenols, phenolic acids, flavonoids, carotenoids, tannins, proanthocyanidins, catechin, vitamins and others. Many reports have been published indicating potent radical scavenging activity of crude extracts and their purified fraction from different parts of *D. kaki*, and their effects on different biological functions have well been established. The antioxidants obtained from *D. kaki* have capability of scavenging ROS, hydroxyl ion radicals, superoxide radicals, peroxyl radicals, singlet molecular oxygen species and shows metal chelating activity. Flavonoids from leaves can increase levels of catalase, super oxide dismutase, and glutathione peroxidase in a manner better than rutin. Total antioxidant activity and total phenolic contents in persimmon were significantly higher than that of apple, grapes and tomato.[42]

4. Conclusion

- From the above study it could be concluded that Persimmon fruit extract have shown to possess anti-inflammatory, antiallergic, photo-protective, and anti-wrinkle effects with appreciable activities against tyrosinase, elastase, and collagenase enzymes.
- Crude extracts and some of phytochemical obtained from this plant such as isoquercitrin and hyperin have better reported activities than well-known cosmetic ingredients viz., arbutin, kojic acid and hydroquinine with possibility of having

no side effects. Photo protection against degenerative effects of UVA, UVB and gamma radiation can help skin to fight well against oxidative stress and reactive oxygen species.

- It could also be concluded that this extract has many cosmetics properties and could be used in various types of products such as:
- Antiallergic products
- Sunscreens
- Skin whitening creams
- Anti-wrinkle preparation
- Deodorant preparations

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4

3D Printing Technology for development of Transdermal Drug Delivery Systems

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Abstract:
The innovative approach of three-dimensional printing enables the on-demand production of transdermal drug delivery systems. This technology, already applied in dentistry, orthopedics, and pharmaceuticals, particularly stands out in the latter field. It facilitates the printing of medical devices and diverse formulations of active pharmaceutical ingredients, featuring controlled-release characteristics and varied geometries. This study provides an overview of these pharmaceutical applications, focusing on the 3D printing of transdermal patches. The discussion encompasses different printing technologies and material systems known for their customization capabilities, generating intricate geometries with precise characteristics crucial for transdermal systems, thereby enhancing bioavailability. The study includes case studies, explores advantages and limitations of the technology, and forecasts industry growth, projecting a value exceeding USD 8 Billion by 2025. Despite this potential, the conservative nature of the pharmaceutical industry leans toward cost-effective methods for large-scale production. Nevertheless, 3D printing has the potential to revolutionize the current 'one size fits all' manufacturing approach, becoming an integral part of the drug development timeline.
Keywords: 3D Printing, Inkjet printing, Microneedles, Patches, Transdermal delivery, Pharmaceutics

1. Introduction

The historical application of therapeutic substances, including herbal ointments and various drugs (such as scopolamine, estradiol, fentanyl, rivastigmine), on the human skin serves both medical and cosmetic purposes. Over the past decades, the skin has proven to be an accessible surface for drug administration, making systematic therapy through percutaneous drug absorption feasible (Prausnitz and Langer, 2008; Alkilani et al., 2015; Pastore et al., 2015). The transdermal route emerges as an attractive alternative to traditional methods like oral administration or hypodermic injections. Oral administration may face issues of partial drug absorption, complications related to gastrointestinal metabolism, and slow onset, making it impractical for emergency cases. Hypodermic injections, while effective, are invasive, pose infection risks, require skilled administration, and generate medical waste (Awodele et al., 2016).

Contrarily, transdermal systems offer advantages such as bypassing metabolic systems, ensuring higher bioavailability, and promoting sustained and controlled drug release. Additionally, transdermal drug delivery (TDD) holds promise for vaccinations due to the abundance of dendritic cells in the skin. This patient-friendly approach is noninvasive, contributing to psychological well-being, and provides independence as it doesn't require professional care for repositioning, removal, or replacement.

However, TDD faces limitations, primarily stemming from the skin barrier's nature. The Stratum Corneum, the outermost skin layer, acts as a significant barrier due to its density and low hydration (15–20%). Overcoming this impermeable barrier has been the focus of TDD research, presenting both challenges and opportunities for future progress.

In the current global trend towards personalized patient care, traditional mass-production methods of drug delivery systems are being questioned for their ability to tailor dosages cost-

effectively. New technologies, particularly Additive Manufacturing (AM), such as 3D printing, are investigated for their potential in pharmaceutical technology. Initially introduced in the 1980s, 3D printing has gained attention across various industries, contributing to the production of complex structures beyond the capabilities of conventional techniques.

The application of 3D printing in pharmaceuticals is relatively recent, aiming to produce targeted-release and customized drug delivery systems (Goole and Amighi, 2016). In the field of TDD, although studies are limited, they demonstrate the transformative potential of 3D printing. This review explores the existing research on 2D and 3D printing as direct or indirect fabrication methods for TDD systems. The materials and drugs associated with 3D printing in TDD systems are also examined in this context.

2. 3D printing techniques for optimized Transdermal Drug Delivery Systems

Additive Manufacturing (AM), commonly referred to as 3D printing or Solid Freeform Fabrication (SFF), encompasses various techniques that utilize a virtual Computer Aided Design (CAD) model to construct a physical object by depositing consecutive layers. Introduced in the 1980s, 3D printing has revolutionized industrial and scientific sectors, offering fast and precise production of intricate structures beyond the capabilities of traditional methods. The medical field quickly recognized the transformative potential of 3D printing, leading to the creation of customized implants, prosthetics, and ongoing investigations into live tissue printing (Chia and Wu, 2015).

The application of 3D printing in drug delivery has recently gained attention, with the FDA approval of Spritam, the first 3D printed oral administration tablet. This has given rise to the term 'pharmacoprinting' (Prasad and Smyth, 2015; Jacob et al., 2014; Goyanes et al., 2015; Di Prima et al., 2016). While its impact on oral drug delivery is well-established, 3D printing's potential in transdermal drug delivery (TDD) is currently under exploration, with a growing body of relevant studies.

2.1. Inkjet Printing

Inkjet printing, involving the controlled deposition of small droplets, has seen successful applications in medicine but is yet to be extensively explored for TDD. Studies have used inkjet printing for coating microneedles with various agents, demonstrating its potential for controlled and selective deposition on suitable substrates (Boehm et al., 2011, 2013, 2014; Ross et al., 2015; Uddin et al., 2015). While its application for building complex three-dimensional TDD structures remains unexplored, inkjet printing's high resolution and selective deposition make it promising for microneedle coating, enabling personalized dosages with high reproducibility.

2.2. Photopolymerization-based Technologies

A significant group of 3D printing technologies relies on selective polymerization of photo-sensitive polymers through laser emissions or light projections. Techniques like Stereolithography (SLA) and Digital Light Processing (DLP) enable layer-wise polymerization of UV-sensitive polymers. These technologies offer versatility in geometric complexity and resolution, making them suitable for TDD applications. Studies have utilized micro-stereolithography (DLP) to create microneedle arrays indirectly, contributing to the customization of therapeutic approaches (Boehm et al., 2014, 2011, 2012). Photopolymerization-based 3D printing has proven applications in fabricating TDD systems, offering high resolution and flexibility.

2.3. Fused Deposition Modelling (FDM)

Fused Deposition Modelling (FDM), based on the melt-extrusion process, is a versatile 3D printing technique with potential applications in TDD. While FDM's limitations in resolution and sensitivity to process parameters are acknowledged, its ability to produce structures through extrusion without solvents makes it a compelling choice for certain pharmaceutical applications. The combination of FDM with hot-melt extrusion (HME) processes holds promise for producing drug/polymer blends for cost-effective TDD system fabrication.

3. Materials:

In the contemporary landscape, 3D printing technologies possess the capability to manipulate a diverse array of materials, ranging from ceramics and metals to polymers. The categorization of these techniques implies that the choice of a specific technology inherently limits the materials compatible with the corresponding printing apparatus. For instance, SLA or DLP printers exclusively handle photo-cured polymers, while FDM printers utilize thermoplastic filaments. This limitation poses challenges for the widespread adoption of 3D printing as a direct manufacturing technique for Transdermal Drug Delivery (TDD) systems, as the material must meet specific criteria for integration into such systems. (Sharma et al.2011) Essential parameters include stability, biodegradability without toxic by-products, mechanical strength, and non-reactivity with the drug. Material biocompatibility is a critical consideration, as evidenced by studies involving Gantrez, a biocompatible copolymer used in TDD applications. While there's evidence of manufacturing Gantrez biocompatible microneedles using 3D printed molds, the multi-step nature of this approach may hinder mass production scalability. (Boehm et al., 2014, 2011, 2012, Donnelly et al., 2012).

Numerous polymers with biomedical and pharmaceutical applications show promise for integration into 3D printed TDD systems. Polyvinyl alcohol (PVA) and poly lactic acid (PLA) are examples. However, challenges exist, such as PVA's limited biodegradability and PLA's slow degradation rates and poor mechanical properties when employed in FDM technology. Biopolymers, like chitosan and collagen, exhibit favorable attributes for TDD. Bioprinting advancements further expand the possibilities. Yet, ongoing research on materials remains vital for the evolving field of 3D printed TDD. (Economidou et al. 2018)

4. 3D Printed Transdermal Drug Delivery Systems- Future Challenges and Expected Impact:

Transdermal Drug Delivery (TDD) systems, facilitated by 3D printing, hold potential as a user-friendly, personalized pharmaceutical therapy. The layer-by-layer fabrication inherent in 3D printing aligns well with TDD requirements. This technology enables the creation of systems with varying drug concentrations across layers, catering to individual needs. Customization possibilities enhance TDD efficiency, addressing factors like skin hydration and thickness variations among patients. In vaccination, microneedles offer promise, particularly in regions facing challenges with traditional administration methods. 3D printing's role in reducing costs and providing needle-free solutions is crucial for global health initiatives. However, challenges such as limited biomaterial options, dosing constraints, and drug degradation characteristics need resolution. The development of 3D printable materials and improvements in existing technologies could drive the evolution of TDD systems. (Economidou et al. 2018)

5. Regulatory Considerations:

For the commercialization of 3D printed TDD systems, adherence to regulatory requirements is essential. Despite the FDA's approval of the first 3D printed oral tablet, TDD systems face unique regulatory challenges. Microneedle patches, viewed as medical devices, must adhere to Good Manufacturing Practice (cGMP) guidelines. The FDA emphasizes technical considerations, including the impact of printing parameters, in-situ quality control, design validation, sterilization, and post-process cleaning. A 2017 guidance document provides recommendations for 3D printed medical devices, addressing issues like patient-matched devices and data protection. Sterilization, a regulatory requirement, presents challenges for microneedles. Future success depends on addressing material limitations, improving technology, and establishing specific regulatory frameworks for 3D printed TDD systems. (Economidou et al. 2018)

6. Conclusion:

Since its inception, 3D printing has revolutionized fabrication methods, with potential applications in medicine and pharmaceutics. Despite being in the early stages, advancements in 3D printed Transdermal Drug Delivery (TDD) systems show promise. Inkjet printing, photopolymerization-based technologies, and FDM have been explored, with inkjet printing successfully depositing films on microneedle surfaces and commercializing 3D inkjet printed microneedles. The integration of elaborate microneedle array systems with precise 3D printing techniques has the potential to reshape modern drug administration. Overcoming engineering, chemistry, and material challenges through interdisciplinary research is crucial. Regulatory considerations, addressed by the FDA, are imperative to ensure the safety and effectiveness of 3D printed TDD systems. Success hinges on resolving material limitations, advancing technology, and establishing specific regulatory frameworks.

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5

Development and Characterization of Metallic Nanoparticles for Antibacterial activity

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Abstract:
The emergence of antibiotic-resistant bacteria has become a major global health concern, leading to the search for alternative antibacterial agents. Metallic nanoparticles (NPs) have gained significant attention due to their unique properties, such as high surface area-to-volume ratio, biocompatibility, and tunable size and shape. This research paper aims to explore the development of title and characterization of metallic NPs for enhanced antibacterial activity. The paper will give detail studies on metallic NPs' synthesis and their antibacterial activity. Additionally, the paper will discuss the challenges and limitations of using metallic NPs as antibacterial agents and suggest future research directions.
Key words: *AgNPs, Antibacterial activity, Multidrug Resistance, Antibiotics.*

Introduction:
Antibiotic resistance has become a major global health challenge, with over two million infections and 23,000 deaths annually in the United States alone (CDC, 2021). The overuse and misuse of antibiotics have led to the emergence of antibiotic-resistant bacteria, making it increasingly difficult to treat bacterial infections. Therefore, there is an urgent need to develop alternative antibacterial agents with novel mechanisms of action. Metallic nanoparticles (NPs) have gained significant attention due to their unique properties, such as high surface area-to-volume ratio, biocompatibility, and tunable size and shape (Kumar et al., 2021). This paper aims to explore the development of title and characterization of metallic NPs for enhanced antibacterial activity [1].
Nanoparticles are viable alternative to antibiotics and appear to possess a high potential for bacterial multidrug resistance. In particular, AgNPs have attracted much attention towards field of nanotechnology. In the past years, it was found that silver was very useful as an antiseptic and antimicrobial agent against Gram-positive and Gram-negative bacteria due to its low cytotoxicity [2]. From a structural point of view, AgNPs have at least one dimension in the range of 1 to 100 nm and more importantly, as particle size decreases, the Surface area-to-volume ratio greatly increases [3].

Antibacterial Mechanisms of Metallic NPs:
The antibacterial mechanisms of metallic NPs are complex and multifaceted, involving various interactions with bacterial membranes, enzymes, and DNA (Kumar et al., 2021). The small size and high surface area-to-volume ratio of metallic NPs enable them to interact with bacterial membranes, leading to membrane disruption, leakage of intracellular contents, and cell death. Additionally, metallic NPs can interact with bacterial enzymes, such as proteases and DNA polymerases, leading to enzyme inhibition and DNA damage. The antibacterial mechanisms of metallic NPs are dependent on various factors, such as size, shape, surface charge, and functional groups [4].

Mechanism of AgNPs AgNPs show antibacterial effect by attaching to the cell membrane of bacteria and also penetrating inside the bacteria. The Nanoparticles preferably attack the respiratory chain and inhibit cell division. The release of silver ions in the bacterial cells enhances their bactericidal activity. Silver is used in different forms such as metals, nitrates,

and sulfadiazine. Nanoparticles are effectively a bridge between bulk materials and atomic or molecular structures [5].

In ancient days Romans treated their water with silver coins, a tradition still being continued in many societies and even in space programs for purifying water. The use of silver as an antimicrobial agent a German obstetrician used 1% silver nitrate solution to eliminate blindness caused by postpartum infections in newborns. US Food and Drug Administration approved colloidal silver for wound treatment. The 0.5% silver nitrate solution use in the burn area. 1% silver sulfadiazine (SSD) cream, which has become one of the leading topical antimicrobial agents used to treat burn wound infections over the last four decades [6-7].

Material and Methods

Material

Silver nitrate, Sodium borohydrate and ascorbic acid all chemical are made by Hi-media (AR Grade)

Methods

Formulation of AgNPs

In formulation of AgNPs, Silver nitrate (AgNO3) was used as a precursor material while Sodium borohydrate and Ascorbic acid (C6H8O6) were used as the reducing agent and surfactant, respectively [8].

The concentrations of Sodium borohydrate and ascorbic acid were varied in order to observe the effect of these parameters especially on the size and morphology of the AgNPs. In detail, 80 ml of AgNO3 was first heated to 60°C and was then added (with vigorous stirring) to 20 ml of a sodium borohydrate and ascorbic acid solution that was pre-heated to 60°C. The mixture was stirred for 20 minutes. Then heating was stopped and the solution cooled to room temperature with continuous stirring. The as synthesized AgNPs was characterized by XRD (using model D5000 Siemens Diffractometer). In plan of AgNPs silver nitrate (AgNO3) was utilized as an antecedent material while sodium borohydrate and ascorbic acid were utilized as the reducing agent and surfactant, individually [9-11].

Table 1: Different Methods of Nanoparticles formulation

Physical methods	Chemical methods	Biological method
Ion beam technique	Sol gel method	Using plant extracts
ric arc deposition	Sol gel method	Using plant extracts
hanical methods	Co precipitation	Using microorganisms

X-ray diffraction Analysis

The X-ray diffraction (XRD) measurement of AgNPs was carried out using Cu-Kα radiation source in scattering range (2θ) of 20– 70 on the instrument operating at a voltage of 45 kV and a current of 40 mA. The presence, crystalline nature, phase variety, and grain size of synthesized AgNPs were determined by X-ray diffraction spectroscopy [13]. The particle size of the prepared samples was determined by using Scherer's equation as follows:

$$D = K\lambda\ \beta 1/2 \cos\theta$$ [14].

Particle size Determination by Laser particle size analyzer

Particle size determination was carried out by means of laser Diffractometer, using an Omec instrument Co ltd. Model Omec LS (POP)9. Measurements were taken in the range between 0.1 and 1000 µm. The instrument was set on the following parameters, particle refractive index 0.54, particle absorption coefficient 4, water refractive index 1.33, and general calculation

model for irregular particles. Three measurement cycles of each were taken, and the data obtained were averaged by software LS (POP) [15].

Particle size Determination by XRD

The particle size 'D' was calculated for the samples using Scherer's equation

$$\frac{dy}{dx} = \frac{0.9\lambda}{(W\text{Cos}\,\theta)}$$

Where 'λ is the wavelength of x-ray, 'W' is FWHM (Full width at half maximum), 'ɵ' is the diffraction angle and 'D' is particle diameter (Size).

When the crystalline domain size calculated by the Scherer equation matches the average diameter of particles determined Particle size analyzer. This observation suggests that the particles are single crystals rather than polycrystalline X-ray diffraction [16, 17].

In-Vitro Antibacterial Activity

This examination was pointed toward deciding the MIC and MBC of AgNPs against Staphylococcus Aureus and Escherichia coli. The antibacterial effects of silver are mostly attributed to silver ions [18]. AgNPs continuously release silver ions in an aqueous microenvironment. Because of the bigger surface area of AgNPs, they show a stronger and better bactericidal effect [19]. The main reasons for bactericidal properties of AgNPs interfere with the integrity of the bacterial cell by binding to essential cellular structural, particularly to their SH-groups. AgNPs also generate reactive oxygen species (ROS) and free radicals which damage the bacterial cell wall and inhibit the respiratory enzymes [20]. AgNPs disturb the DNA replication and terminate the bacteria [21].

Minimum Inhibitory Concentration (MIC) determination

Antibacterial activity of the synthesized AgNPs was studied by the standard disc diffusion method. The overnight grown bacterial culture of Staphylococcus Aureus and Escherichia, coli. Were taken for study [24]. The dilutions of synthesized AgNPs varying from 0.030 mg/ml, 0.070 mg/ml, 0.150mg/ml, 0.310 mg/ml, and 0.620 mg/ml were prepared. The preparation of nutrient media was done by taking 20 g of solidified nutrient media (Soyabean casein digest media) with 2 % of Agar added in 500 ml of distilled water and sterilized in autoclave at 15 lb of pressure and 1210c temperature [25]. This mixture was poured equally into Petri-plates. Keep this plate to solidify, after solidification bacterial cultures were spread on surface of solidified agar with help of spreader. The bore of 8 mm was made up to the lower surface of solidified media. [26, 27] Then organisms to be tested were inoculated in four bores (8 mm diameter) in different dilutions of AgNPs (0.030 mg/ml, 0.070 mg/ml, 0.150mg/ml, 0.310 mg/ml, and 0.620mg/ml) solutions [28, 29]. The plates containing different concentration of AgNPs was incubated at 370C and then examined for confirmation, the appearance of a clear area around the bore was observed. The diameter of such zones of inhibition was measured using a meter ruler, and the mean value for each organism was recorded and expressed in millimeters [30, 31].

Result & discussion

The AgNPs were successfully prepared by chemical reduction method with low cost. The process utilizes, in the aqueous solution, the mixing of Silver Nitrate as organic precursor. The sodium borohydrate and ascorbic acid were use as reducing agents for preparation of AgNPs. The concentrations of Sodium borohydrate and ascorbic acid were varied in order to observe the effect of these parameters especially on the size and morphology of the AgNPs.

Fig 1: Prepared AgNPs

After formulation of AgNPs the confirmation of synthesized AgNPs was characterized by using UV spectrophotometer (Shimadzu 1800). The formation of these can be confirmed by means of spectrum for the colloids that Plasmon band is observed near 416 nm, which confirms that the silver ions were reduced to Ag° in watery phase. The prepared AgNPs can change the absorption spectra at different wavelength according the synthesized range of Nanoparticles in the solution

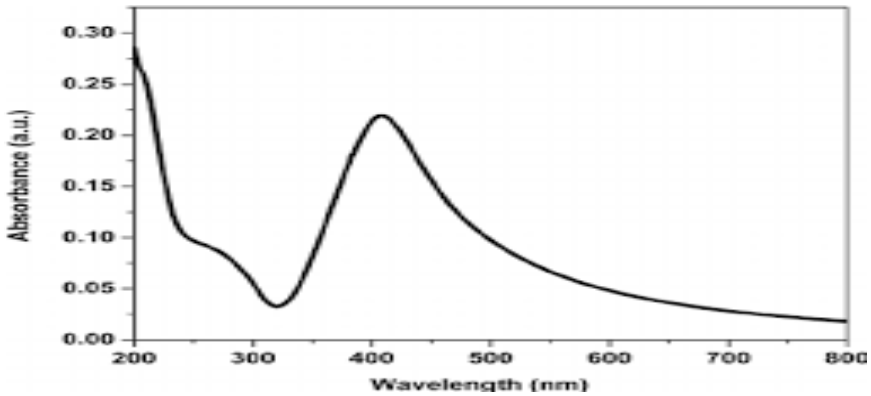


Fig 2: UV spectra of prepared Silver Nanoparticles

As per the absorption spectrum mention above it is clear that there is formation of AgNPs in the range of 1-200 nm. UV spectrum is primary characterization to confirm the synthesis of AgNPs.

Particle size

Particle size determination was carried out by means of laser Diffractometer, using an Omec instrument Co ltd. Model Omec LS (POP) 9. In which parameters of the instruments has been set. Material Refractive Index was 1.52, Dispersant RI 1.333 , Obscuration 0.45% was set then result was taken the average particle size of AgNPs was found in the range of 1- 200 nm.

From the above data particle size distribution was observed which shown in Fig: 3

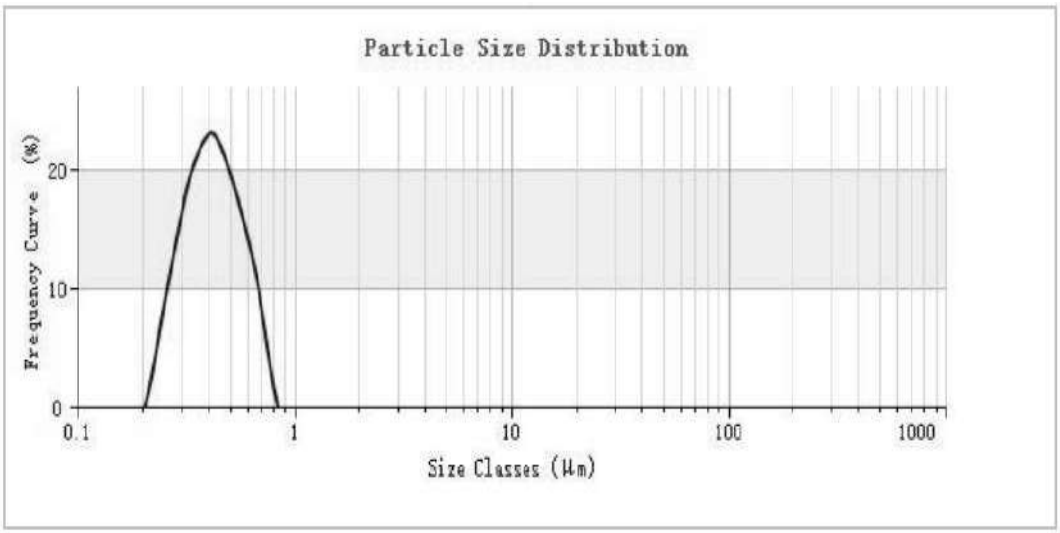


Fig 3: Particle size distribution

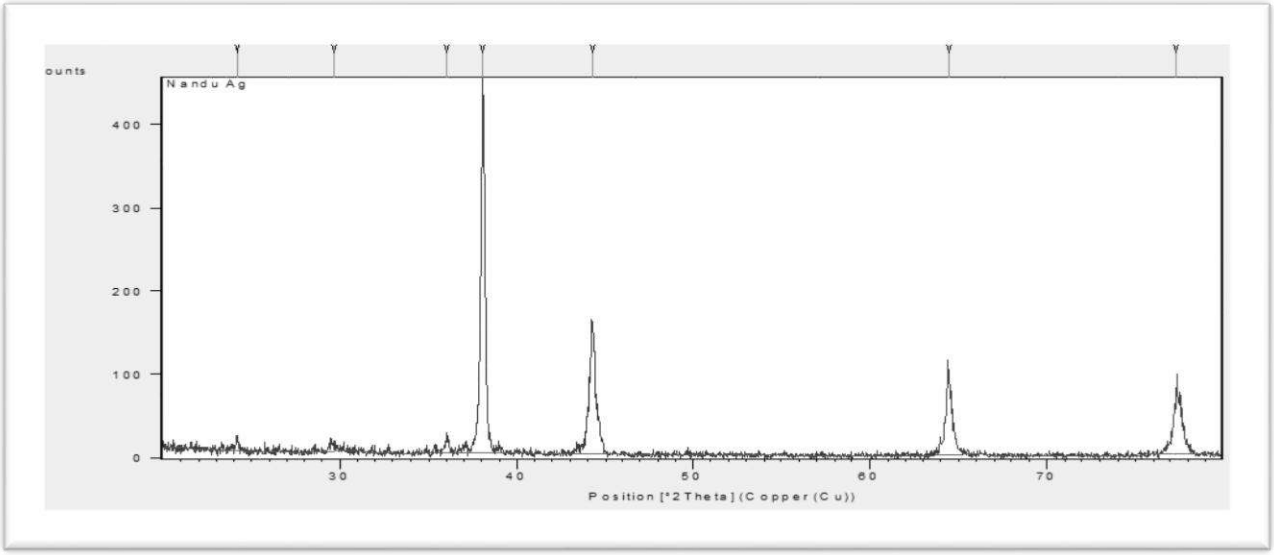


Fig 4: XRD spectrum of AgNPs

The Nanoparticles synthesized in this method were characterized using powder form. The evaluations are shown in Table 3. Diffracting angle in degree, FWHM(radians) , d spacing(nm), Rel. Int. [%] .By considering the values given in table the particle size 'D' was calculated for the samples using Scherer's equation (Cullity & Stock, 2001). From the calculation the particle size of synthesized Nanoparticles was observed in the range of 1-200 nm. Which was again compared with the particle size obtain by the Particle size analyzer. The result obtain by both the methods was matched with each other.

Table 3: XRD Data of Prepared AgNPs

)iffracting angle in degree (expt.)	**Diffracting angle in degree(JCPDS)**	**WHM adians)**	**acing(nm)**	**d spacing(nm) JCPDS data.**	**Rel. Int. [%]**
.2131	24.2125	5904	.67585	.6729	2.39
).6854	29.6875	5904	.00952	.0068	1.70
.0417	36.0375	2952	.49201	.49023	3.60
.0910	38.0875	2460	.36253	.36078	.00.00

.2875	44.2875	2460	.04529	.0436	34.23
.4234	64.4125	2460	.44628	.4453	22.76
.3330	77.3375	3000	.23290	.23284	16.10

Minimum inhibitory concentration (MIC) and Minimum bactericidal concentration (MBC)

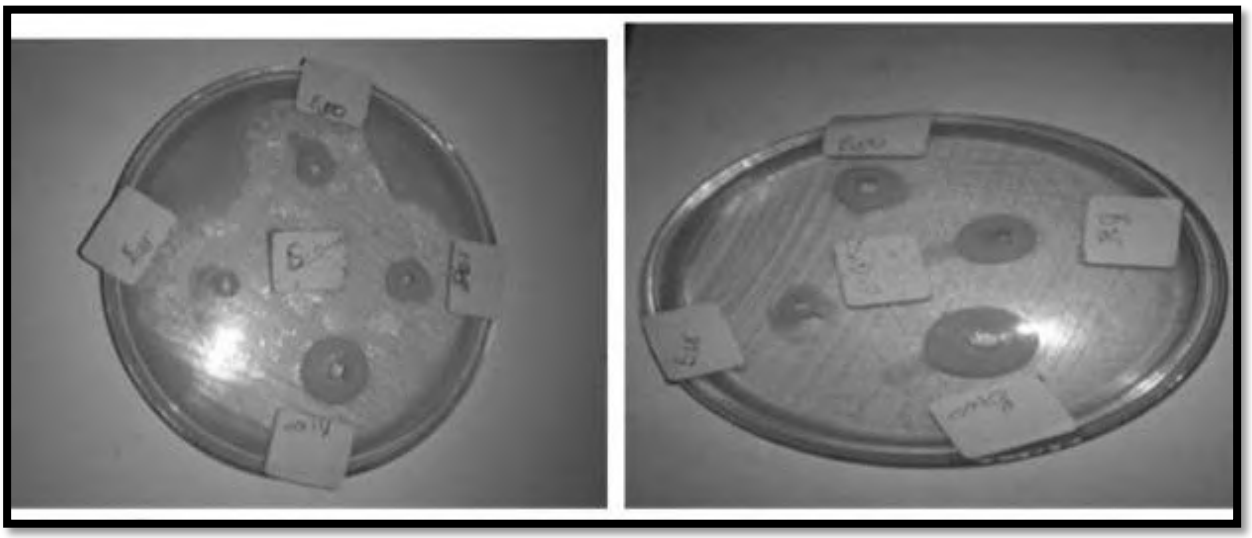
Determination of MIC was done by taking 0.030 mg/ml, 0.070 mg/ml, 0.150 mg/ml, 0.310 mg/ml, 0.620 mg/ml of conc. of AgNPs and inoculated to well and incubated for 24 hrs. zone of inhibition was measured against the same bacterial culture result shows that 0.030 mg/ml concentration of silver does not show zone of inhibition whereas 0.070 mg/ml concentration of AgNPs showed zone of inhibition against Staphylococcus Aureus and Escherichia Coli. From this it confirms that 0.070 mg/ml was the minimum inhibitory concentration of AgNPs.

Then MBC of AgNPs was determined by taking 0.070 mg/ml 0.150mg/ml, 0.310 mg/ml, 0.620 mg/ml and inoculated to well and incubate for 24 hours. 0.070 mg/ml conc. shows the zone of inhibition after that it was kept under observation no visible bacterial growth reappear on area of zone of inhibition that menace 0.070 mg/ml conc. consider as Minimum bactericidal concentration. This was done by observing pre and post-incubated agar plates. In this study, The MIC and MBC of AgNPs against gram positive and gram negative bacteria were determined and were found to be effective at 0.070 mg/ml. (Tables 4). The AgNPs showed MIC at concentration of 0.070 mg/ml against the culture of both gram positive and gram negative bacteria and same concentration act as Minimum Bactericidal Concentration. (Shown in Table: 4 & 5)

Table 4: MIC and MBC Determination of AgNPs against S, Aureus

	MIC observations					**MBC observations**			
	Staphylococcus Aureus					**Staphylococcus Aureus**			
Conc. of silver NP	**.030 mg/ml**	**.070 mg/ml**	**50mg/ml**	**.310 mg/ml**	**20mg/ml**	**.070 mg/ml**	**50mg/ml**	**.310 mg/ml**	**520 /ml**
Zone of Inhibition	-	+	+	+	+	+	+	+	+

Positive (+): Indicating Zone of Inhibition; Negative (–): Indicating No Zone of Inhibition



Staphylococcus Aureus (5a) Escherichia Coli (5b)

Fig: 5 Determination of MIC by AgNPs

CONCLUSION

The AgNPs were synthesized by chemical reduction method. In this method silver nitrate ($AgNO_3$) work as precursor which further interact with reducing agent and stabilizing agent. UV -VIS absorption spectrum and XRD result give the confirmation about synthesis of AgNPs. The size of prepared Nanoparticles was confirmed by Scherer equation and particles size analyzer. The resulted sizes compared with each other which showed the average particles size in the range of 1-200 nm. In further study the MIC and MBC of AgNPs against S. Aureus was determined and found to be effective at 0.070 mg/ml. The AgNPs show MIC and MBC at concentration of 0.070 mg/ml against the culture of Staphylococcus Aureus and Escherichia Coli.

Conflict of Interest: None.

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6

Formulation and Development of Acne Repair Cream Using Matcha Actives

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Abstract:
This research paper investigates the formulation and efficacy of an advanced acne control cream fortified with bioactive compounds derived from Matcha, a finely ground green tea. A comprehensive literature review is conducted, elucidating the multifaceted etiology of acne vulgaris and the potential therapeutic benefits associated with Matcha's rich polyphenolic profile, with a focus on the key compound, epigallocatechin gallate (EGCG). The formulation process is meticulously detailed, encompassing the incorporation of Matcha actives within a sophisticated matrix of emollients, humectants, and stabilizers to optimize not only the cream's bioavailability but also its sensorial attributes and stability over time.

Clinical trials involving human subjects are conducted to comprehensively evaluate the safety, tolerability, and efficacy of the Matcha-infused acne control cream. Parameters such as lesion count reduction, improvement in skin texture, and participant-reported outcomes are systematically documented and statistically analyzed. The findings not only underscore the potential of Matcha as a valuable botanical resource in dermatological formulations but also contribute to the growing body of evidence supporting natural-based skincare solutions.

KEYWORDS:
Acne vulgaris, Matcha bioactive compounds, green tea polyphenols, Epigallocatechin gallate (EGCG).

INTRODUCTION:

The acne repair cream with matcha actives combines the antioxidant and anti-inflammatory properties of matcha powder and green tea extract to provide a potent dose of active ingredients for the skin. The cream also contains allantoin, which has moisturizing and soothing properties and can help to reduce irritation associated with acne.

The matcha actives in the cream can help to reduce inflammation and redness associated with acne, while also providing antioxidant protection to prevent further damage to the skin. The caffeine in matcha powder can help to improve circulation and reduce puffiness, while the polyphenols in green tea extract can help to stimulate collagen production and improve skin elasticity

MATCHA

Matcha powder is known for its high concentration of antioxidants, particularly EGCG. Antioxidants play a vital role in protecting the skin from damage caused by free radicals, which are unstable molecules that can damage cells and contribute to aging. EGCG has been shown to have potent antioxidant activity and can help to protect the skin from UV radiation-induced damage. In addition to its antioxidant properties, EGCG also has anti-inflammatory and antimicrobial properties. These properties make it an effective ingredient for reducing inflammation and redness associated with acne and other skin conditions. Matcha powder also contains caffeine, which can help to improve circulation and reduce puffiness in the skin. Caffeine has been shown to constrict blood vessels, which can help to reduce the appearance

of dark circles and bags under the eyes. Additionally, caffeine has been shown to have anti-inflammatory properties, which can help to reduce redness and inflammation associated with acne.

GREEN TEA EXTRACT:

Green tea extract is a rich source of polyphenols, including catechins, flavonoids, and EGCG. Like matcha powder, green tea extract has antioxidant, anti-inflammatory, and antimicrobial properties. It can help to protect the skin from damage caused by UV radiation, reduce inflammation and redness, and improve the overall appearance and texture of the skin.

Green tea extract has also been shown to have anti-aging benefits. The polyphenols in green tea extract can help to reduce the appearance of fine lines and wrinkles by stimulating collagen production and improving skin elasticity. Additionally, green tea extract can help to brighten the skin by reducing the appearance of dark spots and hyperpigmentation.

ACNE REPAIR CREAM WITH MATCHA ACTIVES:

The acne repair cream with matcha actives combines the antioxidant and anti-inflammatory properties of matcha powder and green tea extract to provide a potent dose of active ingredients for the skin. The cream also contains allantoin, which has moisturizing and soothing properties and can help to reduce irritation associated with acne.

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MATCHA FOR SKINCARE

Matcha is a popular ingredient in skincare due to its high concentration of antioxidants and anti-inflammatory properties. The antioxidants in matcha, such as epigallocatechin gallate (EGCG), can help protect the skin against damage from UV rays and other environmental stressors that can contribute to premature aging.

Matcha also contains chlorophyll, which has detoxifying properties and can help to remove impurities from the skin. Additionally, the caffeine in matcha can help to reduce puffiness and dark circles around the eyes, making it a common ingredient in eye creams and serums. Matcha is also a natural source of L-theanine, an amino acid that promotes relaxation and can help to reduce stress-related inflammation in the skin.

Matcha can be used in a variety of skincare products, including cleansers, toners, masks, and moisturizers. Some skincare experts also recommend using matcha as a natural exfoliant, either by mixing it with a gentle exfoliating agent like sugar or by using a matcha-infused scrub.

Overall, incorporating matcha into your skincare routine can provide a range of benefits for your skin, including improved hydration, increased brightness and clarity, and protection against environmental stressors.

MATCHA FOR ACNE TREATMENT

Matcha can potentially be helpful in treating acne due to its anti-inflammatory and antibacterial properties. Acne is an inflammatory condition caused by a build-up of bacteria and excess oil in the pores, which can lead to the formation of pimples, blackheads, and other blemishes. The anti-inflammatory compounds in matcha, such as EGCG, can help to reduce the redness and inflammation associated with acne, while the antibacterial properties can help to kill acne-causing bacteria.

Matcha can be used in a variety of ways to treat acne, including as a topical application or as a dietary supplement. As a topical treatment, matcha can be mixed with other ingredients like

honey or aloe vera gel to create a soothing face mask. This can help to reduce inflammation and promote healing of existing acne blemishes. As a dietary supplement, matcha can be consumed as a tea or added to smoothies or other beverages. Some studies have suggested that consuming matcha may help to regulate hormonal imbalances that can contribute to acne.
It Is Important to note, however, that more research Is needed to fully understand the effects of matcha on acne. While some individuals may find it helpful, others may not see any improvement in their acne symptoms. It is also important to speak with a healthcare professional before using matcha as a treatment for acne, particularly if you are currently taking any medications or have underlying health conditions.

MATERIALS:
DISTILLED WATER, GLYCERINE, PROPELENE GLYCOL, XANTHUN GUM, ALLANTOIN, CAPRYLIC TRIGLYCERIDE, CETEARYL ALCOHOL, STEARIC ACID, GLYCERYL STEARATE, MATCHA POWDER, GREEN TEA EXTRACT, TEA TREE OIL, PHENOXY ETHANOL, ETHYLHEXYLGLYCERIN

FORMULATION OF PRODUCT: - (TRIALS)

B1

SERIAL NO.	INGREDIENTS	QUANTITY(IN%)
1	DISTILLED WATER	60
2	GLYCERINE	3
3	PROPELENE GLYCOL	3
4	XANTHUN GUM	0.1
5	ALLANTOIN	0.5
6	CAPRYLIC TRIGLYCERIDE	8
7	CETEARYL ALCOHOL	3
8	STEARIC ACID	3
9	GLYCERYL STEARATE	2
10	MATCHA POWDER	2
11	GREEN TEA EXTRACT	1
12	TEA TREE OIL	1
13	PHENOXY ETHANOL	1
14	ETHYLHEXYLGLYCERIN	0.5

B2

SERIAL NO.	INGREDIENTS	QUANTITY(IN%)
1	DISTILLED WATER	60
2	GLYCERINE	3
3	PROPELENE GLYCOL	3
4	XANTHUN GUM	0.1
5	ALLANTOIN	0.5
6	CAPRYLIC TRIGLYCERIDE	8
7	CETEARYL ALCOHOL	3
8	STEARIC ACID	3
9	GLYCERYL STEARATE	2
10	MATCHA POWDER	1.5
11	GREEN TEA EXTRACT	1
12	TEA TREE OIL	1
13	PHENOXY ETHANOL	1
14	ETHYLHEXYLGLYCERIN	0.5

B3

SERIAL NO.	INGREDIENTS	QUANTITY(IN%)
1	DISTILLED WATER	60
2	GLYCERINE	3
3	PROPELENE GLYCOL	3
4	XANTHUN GUM	0.1
5	ALLANTOIN	0.5
6	CAPRYLIC TRIGLYCERIDE	8
7	CETEARYL ALCOHOL	3
8	STEARIC ACID	3
9	GLYCERYL STEARATE	2
10	MATCHA POWDER	3
11	GREEN TEA EXTRACT	1
12	TEA TREE OIL	1
13	PHENOXY ETHANOL	1
14	ETHYLHEXYLGLYCERIN	0.5

In the above given formulations, the B1 is seen to have the good spread ability, texture and feel. Also, it is seen that it passes all the quality parameters. Thus, the formulation B1 is selected as the final formulation.

EVALUATION OF ACNE REPAIR CREAM

Stability testing: This involves subjecting the product to various conditions such as high temperature, low temperature, and exposure to light to assess its stability over time.The product was kept at room temperature, 40^0C, and in 5^oC to check the colour appearance and the texture of the acne repair cream

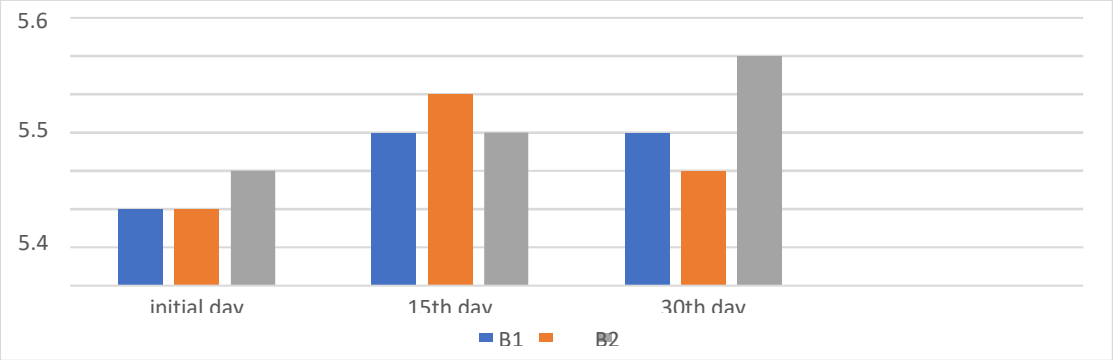
Stability testing:-

SR NO	***TIME INTERVAL***	***B1***	***B2***	***B3***
1	***INITIAL DAY***	***5.5***	***5.1***	***5.2***
2	***15^{th}. DAY***	***5.3***	***5.4***	***5.3***
3	***30^{th}. DAY***	***5.5***	***5.2***	***5.5***

Physical appearance and texture: -

SR NO	PARAMETERS	B1	B2	B3
1	**COLOUR**	++	++	+++
2	**ODOUR**	++	+++	+++
3	**TEXTURE**	+++	++	+++
4	**FEEL**	++	++	+++

Graphical Representation of Ph test:-



SUMMARY

Acne repair cream using Matcha actives is a topical skincare product designed to help treat and prevent acne breakouts. It contains Matcha tea powder as the active ingredient, which is known for its anti-inflammatory and antioxidant properties. These properties can help to reduce inflammation, prevent breakouts, and promote skin healing.

Matcha tea powder is made from ground green tea leaves and has been used for centuries in traditional Japanese medicine. It contains high levels of polyphenols, which are antioxidants that can help to protect the skin from free radical damage. It also contains catechins, which have been shown to have anti-inflammatory effects that can help to reduce redness and swelling associated with acne.

When using an acne repair cream with Matcha actives, it's important to follow the instructions carefully to avoid over-drying or irritating the skin. Some creams may cause redness, peeling, or sensitivity, especially when first starting use. It's also important to use a sunscreen during the day, as some acne repair creams can increase skin sensitivity to the sun.

In summary, acne repair cream using Matcha actives is a topical skincare product that can help to treat and prevent acne breakouts. It contains Matcha tea powder as the active ingredient, which is known for its anti-inflammatory and antioxidant properties. When using this type of acne repair cream, it's important to follow the instructions carefully to avoid over-drying or irritating the skin.

CONCLUSION

Acne is a common skin condition that affects many people worldwide and can have a significant impact on a person's self-esteem and quality of life. Acne repair creams can be an effective tool in managing and preventing acne breakouts, including those using Matcha actives. These creams contain active ingredients that work to unclog pores, reduce inflammation, and promote skin healing.

However, it's important to choose the right product for your skin type and to use it correctly to avoid over-drying or irritating the skin. Additionally, it's important to take other steps to promote skin health, such as keeping your skin clean, using non-comedogenic products, and managing stress. If you are experiencing persistent or severe acne, it's important to consult with a dermatologist, who can recommend a personalized treatment plan.

Overall, with the right skincare routine and treatment plan, it is possible to manage and prevent acne breakouts, leading to clearer, healthier-looking skin.

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7

Use on snail secretion as multifunctional cosmetic active

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Abstract-
Snail mucus is material of a natural origin that is a source of valuable active ingredients. Due to the content of vitamins, allantoin, acids and proteins fulfilling specific roles, snail mucus has many applications used in skin care. Snail mucus accelerates the healing of wounds and sunburns. It nourishes the skin, reduces imperfections, and protects against free radicals. The varied composition allows the use of snail mucus according to the needs of the skin.
Products that contain snail mucus make it possible to rejuvenate and beautify the skin. They can be used to treat skin diseases, such as melanoma, acne, and inflammation, as well as burn wound infections. The presence of a very large amount of nutrients makes snail mucus widely used in cosmetics and medicine, but at the same time, it makes it impossible to produce it artificially in the laboratory.

Index Terms- Mucus, Mucin,

I. INTRODUCTION

Intrigue in the mucus slime trails left by snails and slugs date back to ancient Greece, where they utilized the mucus for its ability to reduce inflammation and the signs of aging. Today snail mucus is still used in skin care products by various companies and is a growing market whose value is expected to approach $770 million by2025. Despite its commercial applications, the field of mucus research remains surprisingly underdeveloped. The primary constituent that is responsible for the properties of mucus are secreted mucins, a family of heavily glycosylated proteins produced in epithelial cells in most animals. Mucins are either bound to the plasma membrane or secreted out of the cell, and each type has major differences in their functions and capabilities (Dhanisha et al., 2018). Membrane bound mucins are glycolipids that act as markers for cell signaling and also protect the cell from extracellular affronts that might lead to damage, such as infections and physical strain (Van Putten and Strijbis, 2017). Secreted mucins can be either gel forming or non-gel forming biopolymers. Secreted biopolymers form mucous membranes macroscopic scale. These mucosal membranes account for a large portion of the surface area of multicellular organisms exposed to the environment. In humans, mucosal membranes account for 99% of the bodies surface area (Sompayrac, 2012; Maet al., 2018; Cerullo, 2020). Each snail species secretes multiple distinct functional mucuses. The mucus produced by a snail's foot is used for adhesion and for lubrication, allowing the snail to stick onto or walk across any surface, even while inverted.
Snail slime is a clear, slightly amber liquid with a pH value of 4.80 and a density of 1.02 g/ml. It contains many active ingredients, including: allantoin, elastin, collagen, proteins, antioxidants, enzymes, metal ions, proteoglycans, glycosaminoglycans, vitamins,minerals as well as mucin, mitamycin AF and achacin.The mucus obtained from snails is a cosmetic raw material,rich in many ingredients that exhibit beneficial effects on human skin. It can be found mainly in facial care products as it demonstrates regenerative properties of the skin after

mechanical damage or sunburn and reduces imperfections and discoloration . Moreover, snail mucus promotes the longevity of fibroblasts and the structure of the extracellular matrix, thus delaying the symptoms of skin ageing . Such valuable properties of snail mucus for human skin were the reason to describe in this work the production, the qualitative and quantitative composition, features and potential cosmetic and medicinal utilization based on literature data. Additionally, the mucus produced on the back of the snail is used for microbial defense and tissue hydration. Certain snail species have specialized uses for mucus.

For example, Falsilunatia eltanini (Moon Snail) uses mucus to protect their eggs, and Tikoconus costarricanus (Costa Rican Land Snail), uses mucus for load-bearing activities, such as to hide from the Sun on the bottom of leaves during droughts (Gould et al., 2019; Barrientos,2020). Recent advances in omics (genomic, transcriptomic, proteomic, glycomics) technologies have expanded the exploration of gastropod mucins as a scientific resource with wide ranging applications across chemistry, biology, biotechnology, and medicine. For example, the antimicrobial properties of snail mucus are being used to combat disorders seen in humans ranging from gastric ulcers, to post-surgical- related infections (Amah et al., 2019; Gentili et al., 2020). Mucins are also being coupled with approved therapeutics in order to potentiate the drug's abilities to cure diseases, such as diabetes and ulcerative colitis (Gugu et al., 2020). Additionally, snail mucins are being investigated in a vast array of other biotechnical applications that exploit their surfactant-like properties (Petrou and Crouzier, 2018). Despite their potential, little is known about how the hierarchical mucin structures account for their diverse functional properties damage, such as infections and physical strain (Van Putten and Strijbis, 2017). Secreted mucins can be either gel forming or non-gel forming biopolymers. Secreted biopolymers form mucous membranes macroscopic scale. These mucosal membranes account for a large portion of the surface area of multicellular organisms exposed to the environment.

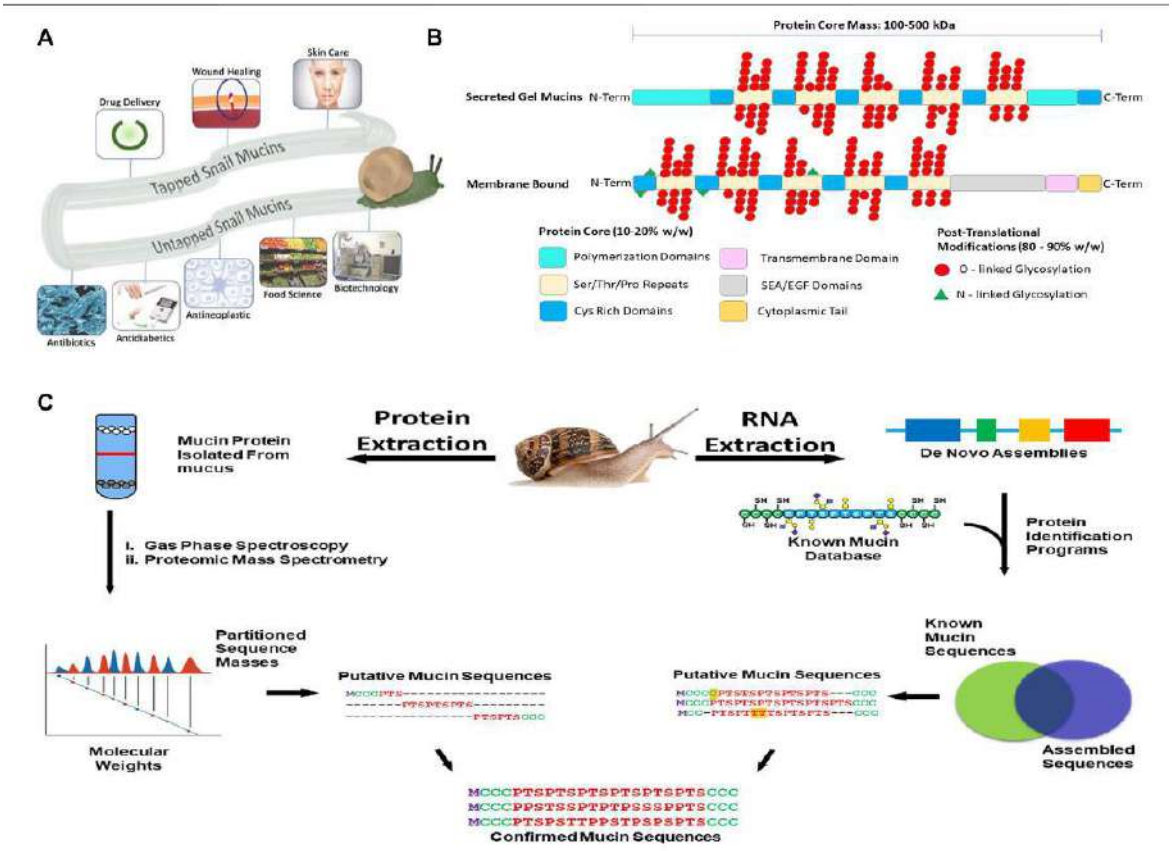

FIGURE 1 | (A) APPLICATIONS OF SNAIL MUCUS. SNAIL MUCUS HAS BEEN USED FOR SKIN CARE, WOUND HEALING AND REJUVENATION, AND DRUG DELIVERY. SNAIL MUCUS IS BEING EXPLORED IN FOOD SCIENCE, IMPLANT COATINGS, AND OTHER BIOTECHNICAL SECTORS ARE CURRENTLY RESEARCHING MUCINS TO BE EXPLORED FOR POTENTIAL USE. (B) A 2-DIMENSIONAL REPRESENTATION OF THE MUCIN STRUCTURES. MUCINS ARE CHARACTERIZED BY TWO PARTS OF THEIR STRUCTURE, THEIR PROTEIN CORE, AND THEIR GLYCAN BRANCHING. THE PROTEIN CORE IS A PROTEIN SEQUENCE OF VARIABLE LENGTH DEPENDING ON THE MUCIN GENE, WHICH HAS BEEN FURTHER MODIFIED WITH GLYCOSYLATION BRANCHES. THE PROTEIN STRUCTURE, HOWEVER HAS MULTIPLE DOMAINS, AND THESE DOMAINS VARY DEPENDING ON THE FUNCTION AND THE CELLULAR LOCATION OF THE MUCIN. THE GLYCAN BRANCHES ARE SUGAR BRANCHES RANGING FROM 3 TO 18 SUGARS, AND MAKE UP THE MAJORITY OF THE MUCIN MASS. SHOWN ARE 2 DIMENSIONAL REPRESENTATIONS OF THE DIFFERENT TYPES OF MUCINS, AND THEIR STEREOTYPICAL FEATURES. (C) APPLYING AN INTEGRATED OMICS APPROACH TO IDENTIFY SNAIL MUCIN SEQUENCE, STRUCTURE, AND FUNCTION. PATH 1(LEFT) EXTRACT CRUDE MUCIN PROTEINS AND SEPARATE FROM THE CELLULAR DEBRIS TO OBTAIN SEQUENCE MASSES FROM SPECTROSCOPIC AND MASS SPECTROMETRIC ANALYSES. PATH 2(RIGHT) RNA EXTRACTION FROM MUCUS GLANDS OR WHOLE ANIMAL FOLLOWED BY DE NOVO ASSEMBLY OF MUCIN GENE SEQUENCES TO GENERATE A DATABASE TO BLAST AGAINST BY A COMPARISON OF ASSEMBLED SEQUENCES TO A KNOWN MUCIN DATABASE, WE OBTAIN PUTATIVE MUCIN SEQUENCES. COMBINING THE PROTEOMIC AND RNA PIPELINES WE CONFIRM THE NATIVE TYPE MUCIN SEQUENCE FOR FURTHER ANALYSIS.

I. CONTENTS OF SNAIL MUCILAGE :

Snail mucus contains allantoin, collagen, elastin, glycolic acid, natural peptides and proteins, vitamins A, C and E, as well as antioxidants (e.g. polyphenols) and enzymes (superoxide dismutase - SOD and glutathione S-transferase - GST). Among the metal ions, copper (Cu), iron (Fe) and zinc (Zn) were found. The other ingredients are proteoglycans, glycosaminoglycans - including hyaluronic acid, copper peptides, and antimicrobial peptides, as well as lactic acid, matrix metalloproteinases and their inhibitors.Snail mucus also contains mucin, mitamycin AF and achacin .Mucin is the main macromolecular component of mucus, which is responsible for its regenerative properties. Mucin contains active antimicrobial proteins against gram- positive and gram-negative bacteria .Their activity was found against Pseudomonas aeruginosa AP9 and Bacillus. Snail mucus is composed of ingredients such as Proteins (Collagen and Elastin), Hyaluronic acid, Copper peptides, Antimicrobial peptides, Antioxidants, Glycolic acid, Allantoin, and more. All of these components are beneficial for your skin in different aspects – that's what gives snail mucin its many benefits.

Collagen and elastin: Natural proteins that form the connective tissues in the body. Collagen promotes skin strength, while elastin provides skin elasticity.

Glycolic acid: An exfoliant often used to remove layers of dead skin and curb hyperpigmentation.

Allantoin: An organic compound that moisturizes the skin, which may have anti-inflammatory effects and promote wound healing.

II. SNAIL MUCINS AS ANTIMICROBIAL AGENT

Antibiotic-resistant bacteria are becoming an increasingly prevalent issue without many viable solutions. Because mollusks lack adaptive immunity, they depend on physical barriers and innate immunity for protection against pathogenic agents (Gerdol,2017). For most snails, the foot has the most contact with

surfaces that are contaminated with pathogens and parasites, and secretion of mucus along the feet protects against such microbes.

BIOSLAB (Rio de Janeiro) was used for microbiological testing. The antimicrobial activity was performed by diffusion wells according to the method described by Shriyan et al. (1995). BHI broth was inoculated with the microorganisms and incubated at 37 ° C/24horas. After this period, the inoculum was adjusted to McFarland scale tube 0.5 (1.5 x 108 CFU) in saline. Was inoculated by spreading on plates containing Mueller-Hinton Agar m and Sabouraud agar inocula adjusted to the McFarland scale. We used 5 mL, 10 mL and 20mL respectively, of mucus containing the feeds, which were deposited into wells and incubated for 24 hours at 37 °C. The inhibitory activity was determined by measuring the zone of inhibition.

III. Snail mucous as multifunctional cosmetic active

Snail mucus has moisturizing, nourishing, soothing, exfoliating, cleansing, anti-wrinkle and ultraviolet radiation-absorbing properties. It reduces acne, wrinkles and stretch marks as well as the signs of skin photoaging and also damage caused by free radicals. Trapella et al. proved that mucus obtained from Helix Complex can promote cell migration and support the wound-healing process. H. aspersa mucus has an antibacterial effect and accelerates the reconstruction of damaged skin. In turn, Gentili et al. proved that the mucus obtained from this snail species was protective against damage caused by ozone, thus highlighting the possibility of using a given raw material as a new method of protection againstcontamination. Lim et al.showed in in vitro studies that the active snail mucus extract had a positive effect on skin ageing (including transepidermal water loss (TEWL), number of wrinkles, skin roughness and elasticity). Whereas Mencuccet al. showed that the solution extracted from snail mucus (GlicoPro ®) reduces the biomarkers of inflammation and eye damage. The anti-inflammatory.

One of the earliest mucuses evaluated for antimicrobial activity was that of Achatina fulica (Giant African Land Snail) Mucus from A. fulica demonstrated promising antibacterial activity against the Gram-positive bacteria, Bacillus subtilis and Staphylococcus aureus, and the Gram-negative bacteria, Escherichia coli and Pseudomonas aeruginosa . The mucus secretions of A. fulica inhibited the bacterial growth of both S. aureus and S. epidermidis when applied via wound dressing films on a mouse model.

The wound dressings improved the maturation of granulation tissue and the rate of collagen deposition, which are known to expedite the healing process . In a similar study, the mucus of Helix aspersa demonstrated antimicrobial activity against several strains of Pseudomonas aeruginosa .Further, the mucus of both A. marginata and A. fulica, were utilized as wound dressinsg on 28 clinical wound samples collected with known common infections. The mucus showed anti-bacterial potency against Staphylococcus, Streptococcus, and Pseudomonas isolated from wounds. In the same study, when compared to seven common antibiotics, including amoxicillin, streptomycin, and chloramphenicol, some of the mucus secretions were more inhibitory to infections than commercial antibiotics. Understanding the antimicrobial properties of snail mucus is an active and growing area of research. analgesic and moisturizing properties of the cornea were proven. Due to the above, the mucus can be used in the treatment of atopic dermatitis,psoriasis, burns, ulcers and acne.

IV. Antioxidant activity of snail mucus

Brieva et al. discovered that the mucus of H. aspersa contains antioxidant activities of superoxide dismutase and glutathione-s-transferase. Therefore, the richness of our sample in biomolecules is the basis of their antioxidant activity. The identified compounds have been found to possess anti-inflammatory properties and inhibit angiogenesis, a process crucial for

tumor growth. As a result, they help in restoring the immune system. In addition to their antioxidant properties, these compounds exhibit a range of biological activities, including anticoagulant, antiallergic, anti-inflammatory, and vasodilatory activities . These activities could also be explained by the presence of hydroxyl groups in phenolic compounds that can trap free radicals. Furthermore, these antioxidant properties have been reported and confirmed in the mucus of H. aspersa Muller due to the presence of allantoin, whose antioxidant properties have been demonstrated . The main function of antioxidants depends on their ability to reduce oxidative damage. Hatuikulipi et al. confirmed that the mucus of H. aspersa was found to have anti-inflammatory and antioxidant properties, which helped in reducing colon inflammation. Various bioactive compounds and antioxidant properties were identified in extracts from different parts of several snail species . Due to their antioxidant properties specifically, their ability to donate a hydrogen atom-reductones are crucial to iron reduction capability. Similarly, the beneficial substances in H. aspersa Muller mucus can combine with radicals, giving electrons to transform them to more stable molecules and stopping the free radical chain reaction.

V. It Contain Anti-Aging Properties

"Snail mucin is known for its anti-aging properties due to its collagen and elastin content," researchers investigated the effects of a skin care regimen containing snail secretion filtrate in women between the ages of 45 and 65 years. At the end of the three-month trial period, researchers found that the women who followed the regimen experienced "significant improvements in skin roughness, firmness and elasticity"

VI. It Aid in Wound Healing

Snail mucin may also be used as a wound-healing agent in skin care. One 2016 in vitro study found that snail mucin had antibacterial effects on bacteria isolated from wounds. The study involved African snails, which secrete a substance called achacin that kills bacteria by generating hydrogen peroxide . The allantoin in snail mucin may prove useful for minor cuts, acne and scarring, according to reseacherThe anti-inflammatory properties of snail mucin have also been used to treat burns and radiation dermatitis. The mechanism of action is thought to involve the antioxidant properties of snail mucin and the resulting control of the free radicals that contribute to inflammation and skin injury

VII. It Moisturize the Skin

Snail mucin contains substances that trigger the production of hyaluronic acid, a natural substance that retains moisture, and is found in the eyes, skin and joints. This increased hyaluronic acid could reduce dryness and help the skin stay hydrated.

VIII. Sun Damage Recovery

One 2021 study in mice found that oral intake of snail mucin could reduce symptoms of ultraviolet B (UVB) radiation, a type of radiation emitted by the sun that could lead to sunburn, aging and skin cancer. However, more research is needed to confirm similar effects in humans.

IX. It Can Help Exfoliate Skin

The glycolic acid in snail mucin can act as a gentle exfoliant, revealing brighter and smoother skin, Additionally, glycolic acid can break down keratin (a protein that helps form the hair, skin and nails), which helps eliminate rough or dry patches of skin. It's also used as an exfoliant in skin care products targeted to address acne and post-inflammatory hyperpigmentation.

CONCLUSION

Snail mucus having multiple actions such as inflammatory process during wound healing, sooths sun exposure action, it also acts as hydrating, moisturizing, anti-aging, it can also helps to exfoliate the skin, Snail mucus is a potential promising material to be developed into a drug

to accelerate multifunctional . However, it is necessary to test the toxicity, biocompatibility, and stability of the 96% snail slime gel to produce a useful cosmetic active.

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8

Formulation and Evaluation of Anti-aging Cream by using Mushroom Extract

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ABSTRACT:

The main objective of work is to formulate Anti-Aging cream by using Mushroom Extract.Today, there is a growing consumer demand for cosmetics containing natural and/ororganic ingredients as paving their way into cosmetics, such as ceramides, lentinan, schizophyllan, omega 3, 6,and 9 fatty acids, carotenoids, resveratrol, and others
Many mushroom ingredients possess potent antioxidant, as well as anti-inflammatory, properties, which are used frequently These compounds show excellent antioxidant, anti-aging, anti-wrinkle, skin whitening, and moisturizing effects, which make them ideal candidates for cosmetics products.Formulation FW2 shows anti-aging properties which also showed good Reheologicalchatracterstics ,pH, greater active content. Hence this study showed that F2 was the best formulation for anti-aging cream. According to In- Vivo study, the product has no skin irritation and redness form after applying on the skin.

Keywords : Anti-aging, Mushroom, Antioxidant, Moisturizing and Sajor-caju

INTRODUCTION:

Now-a-days herbal extracts are used in the cosmetic preparations for augmenting beauty and attractiveness. Herbal cosmetics are classified on the basic of dosage from like- cream, powder, soaps, solutions etc. and according to part or organ of the body to be applied for like: cosmetics for skin, hair , nail , teeth and mouth etc. The use of cosmetics requires both their efficacy as well as minimal risk of skin irritation/skin sensitization. This is influenced by their formulation, nature of their use and quantity and quality of ingredients. Mushrooms are rich in protein, vitamins, minerals, and excellent sources of _-glucan, selenium,thiamine, riboflavin, niacin, panthothenic acid, and folic acid, etc. It has reported that **mushrooms** provide beneficial effects as invigorating vital energy, maintaining one's optimal weight, favoring longevity, and avoiding unnecessary aging Recently mushrooms have drawn worldwide attention as the most interesting natural sources with diverse and unique bioactivities, including immunomodulatory, antioxidant, anti-inflammatory, antidiabetic, antibacterial, antifungal, antiviral, antitumor, hepatoprotective, reducing glucose and lipidic levels. Sajor-caju gives best anti-aging properties. Many mushroom ingredients possess potent antioxidant, as well as anti-inflammatory, properties, which are frequently used in an effort to address cosmetic concerns, such as fine lines, wrinkles, uneven tone, and texture. **[33],[34]**

Sajor-caju Mushroom

MATERIALS AND METHODS

Glycerol mono stearate, Sodium laureth sulfate, Distilled water, Lanolin, Glycerin, Stearic acid, Bees wax, Mushroom extract, Phenoxy ethanol and Perfume are used for anti-aging cream.

Table No. 1 Base Formulation

Sr. No.	Ingredients	FW1	FW2	FW3
1	Glycerol mono stearate	3.1gm	3gm	3.5gm
2	Sodium laureth sulfate	0.4 gm	0.3 gm	0.4 gm
3	Distilled water	76 ml	77 ml	76 ml
4	Lanolin	3gm	3gm	3.5gm
5	Glycerin	4ml	4ml	4.5ml
6	Stearic acid	3gm	3gm	3.2gm
7	Bees wax	5.5 gm	5.5 gm	5.5 gm
8	Perfume	q.s	q.s	q.s
9	Phenoxy ethanol	q.s	q.s	q.s

We selected FW2 Because its shows best results

Table 2: Formulation by using mushroom active

Sr. No.	Ingredients	FW1	FW2	FW3
1	Glycerol mono stearate	3.1gm	3gm	3.5gm
2	Sodium laureth sulfate	0.4 gm	0.3 gm	0.4 gm
3	Distilled water	76 ml	77 ml	77 ml
4	Lanolin	3gm	3gm	3.5gm
5	Glycerin	4ml	4ml	4.5ml
6	Stearic acid	3gm	3gm	3.2gm
7	Bees wax	5.5 gm	5.5 gm	5.5 gm
8	Mushroom extract *(sojar-caju)*	1ml	2.2 ml	2.5ml
9	Perfume	q.s	q.s	q.s
10	Phenoxy ethanol	q.s	q.s	q.s

We selected FW2 Because its shows best results

Procedure

Firstly use clean and dry apparatus.

Part A – The water- soluble components like sodium laureth sulfate, glycerine, Distilled water (70%), are dissolved in an aqueous phase and mix it well. These are heated to 70°C on Hot plate.

Part B – The oil-soluble components like Bess wax, Glycerol mono stearate(GMS), Stearic acid, lanolin are dissolved in oil phase and mix it well. These are heated to 70°C on hot plate.

After that we incorporate oil in water and at 40°C add perfume preservative and mushroom extract.

Procedure is same in base formula but extract is added after evaluation

EVALUATION

In- Vitro Studies

Determination of Physical Parameters

1) **Color :** The color of the cream was observed by visual examination. The result was shown in table no.3
2) **Odour:** The odour of cream was found to be characteristics.
3) **State :** The state of cream was examined visually. The cream was semi-solid state was shown in table no.3
4) **Consistency :** The formulation was examined by rubbing cream on hand manually. The cream having smooth consistency.
5) **pH :** pH of prepared herbal cream is measured by using pH paper. The average pH value of cream is 6.5 formulation FW2
6) **Non-irritancy test :** Herbal cream formulation was evaluated for the non-irritancy test. Preparation shown no redness and irritancy. Observation of the state was done for 24 to 28 hours.
7) **Viscosity :** The viscosity of cream was done by using viscometer at the room temperature. Viscosity of formulated cream was determined by brook field viscometer at 30 rpm using spindle no.s05. The viscosity of cream was in the range of, 472,000 to 25,438 cp which indicates that the cream is easily spreadable by small amount of shear. The formulated cream shows the viscosity within range i.e.39,077cp.at temperature 37°C.

In-Vivo Studies

Determination of moisture content of skin by Corneometer

Principle : Corneometer is device which is equipped with a moisture sensitive probe which is used to determine the accurate moisture content of stratum cornerum. Hence it plyas important role in determining the moisturizing activity of product on stratum corneum after its application on skin.

Apparatus : Corneometer equipped with a probe.

Procedure : The volunteers were selected and the probe of Corneometer was applied onto the selected part of skin before application o product and the reading was recorded. The the seclected part of skin was rinse with product allowed to dry properly and again the probe was applied onto the skin and reading was recorded. The volunteer were allowed to wash the selected area of skin with the product twice a Day and then same procdure was followed 14 days. Within these intervals the recorded after 7^{th} Days and then 14^{th} Days and the graps were plotted.

Results and Discussion

Determination of Physical Parameters

Table 3. Results of anti-aging cream

Sr. no	Parameters	Results
1	Color	White
2	Odour	Characteristics
3	State	Semi-solid
4	Consistency	Smooth
5	pH	6.5
6	Spreadability	Good
7	Non-irritancy	Non-irritant
8	Viscosity	39,077
9	Phase separation	No phase separation
10	After feel	Emollient

Table no 4: Ph of Anti-aging cream

Sr.No.	Time interval	FW1	FW2	FW3
1	Initial Day	6.5	5.6	5.6
2	15thDay	5.6	6.7	6.6
3	30th Day	6.7	6.5	6.9

Non-irritancy test : Anti-aging cream formulation was evaluated for the non-irritancy test. Preparation shown no redness and irritancy. Observation of the state was done for 24 to 28 hours

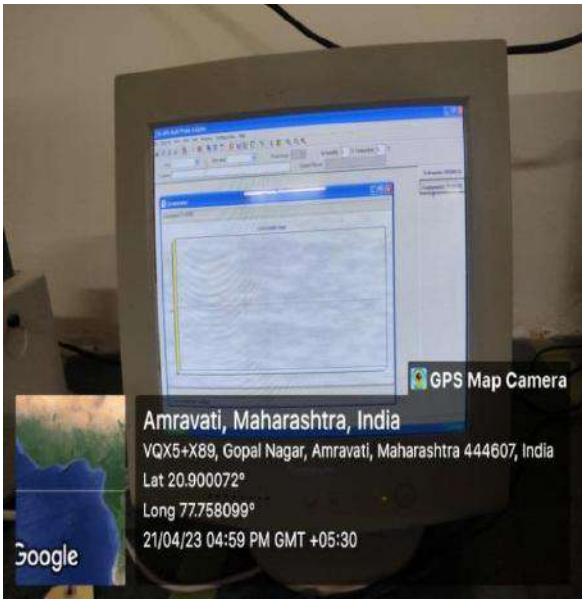
Determination of viscosity

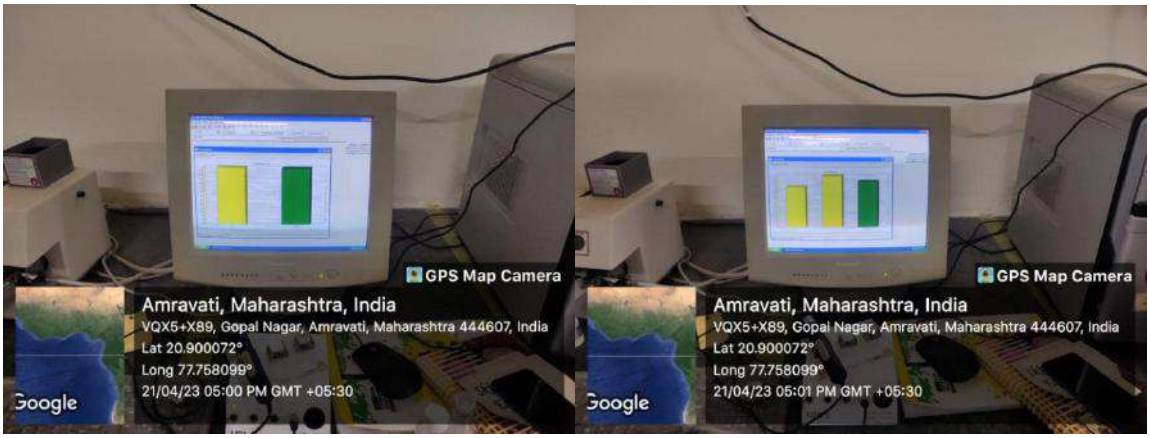
Table no 4: Determination of viscosity

The viscosity of face wash determine by using Brookfield Viscometer. The values obtained from the sample noted.

Sr. No.	No. of days	FW1	FW2	FW3
1	Initial Day	72500cp	6200cp	6950cp
2	15th Day	71530cp	71000cp	68080cp
3	30th Day	71820cp	39,077cp	67500cp

Determination of moisture content of skin by Corneometer



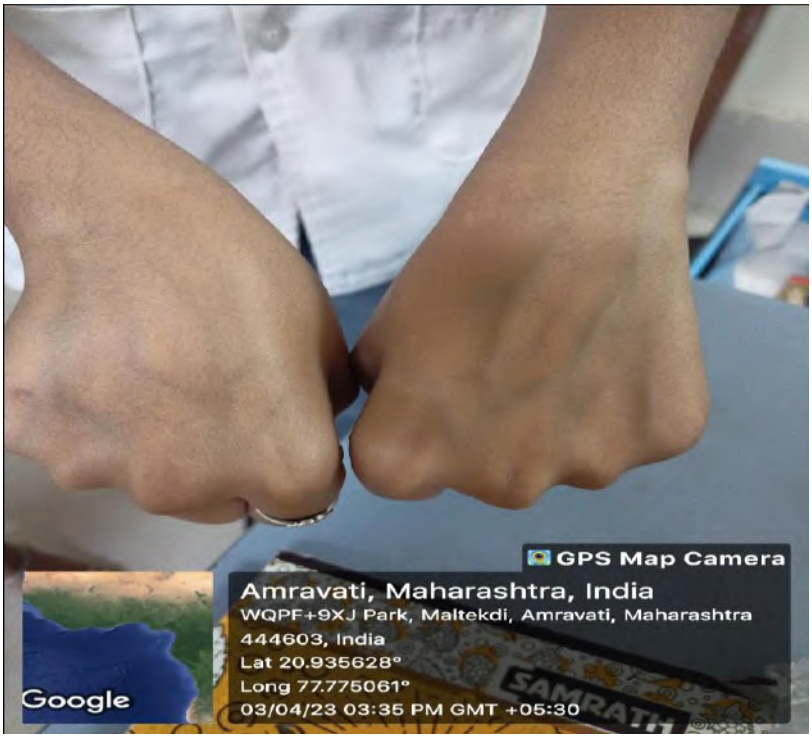


Corneometer analysis before application **Corneometer analysis after application**

Result : The moisturizing activity was carried out by using Corneometer. It was observed that before application of cream, the mositure content of skin was less and after application of Cream moisture content was increased.

Photographic Evaluation The study of effectiveness of product was done by the help of the volunteer study. This was carried out human volunteers. Face wash were applied on skin. The photograph were taken before and after application of product.

Before and After application of anti-aging cream



CONCLUSION

Mushroom *(sojar-caju)* in different ratio to get multipurpose effect such as whitening, antiwrinkle, antiaging and sunscreen effect on skin. As we know that it is not possible to increase the extent of efficiency of medicinal and cosmetic property of single plant extract. The study indicated that the formulation was found to be more stable with constant pH, homogenous, emollient, non-greasy and easily removed after the application. The stable formulations were safe in respect to skin irritation and allergic sensitization. The present study was aimed to develop and formulate the herbal cream containing Mushroom (sojar-caju) extract which may maintain aging is a gradual process that results in a dysfunction and reduced

reserve capacity to all body organs. The benefits of an effective anti-aging skincare regime include retaining the skin's firmness, refining the skin tone, reducing the appearance of fine lines and wrinkles, and boosting brightness and radiance. Several types of mushrooms are used in topical cream, serums, and facial preparation as anti-aging ingredients

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9

Development of shampoo with superior hair conditioning properties of Lipoaminoacid technology

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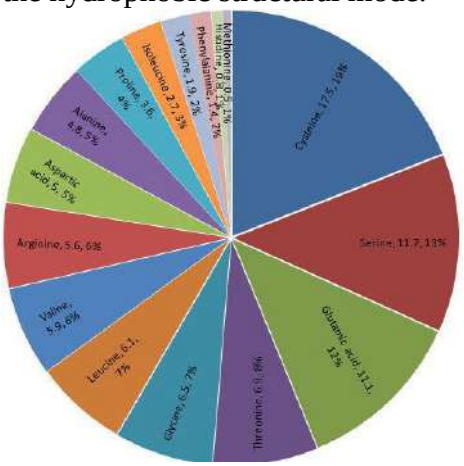
Abstract-
Scalp hair forms an important part of overall appearance of human body and contributes heavily to the impression it may create about one's personality. Thus making 'cleansing and beautifying' the hair an area of importance. Hair care is ever-present human habit which is often driven by conscious or sub conscious social and evolutionary pressures. Hence this report is devoted to design the shampoo formula and lipoaminoacid technology catering to specific needs of the category, also a superior functional delivery is obtained against a benchmark through careful consideration and selection of active ingredients in experiment approach.
Apart from basic cleansing that a shampoo can provide, study of beautification and hair health improvement that a shampoo can deliver still remains a huge platform of further research and exploration with evolving consumer lifestyle. Living tissues are made of amino acids, building blocks of life, creating proteins- collagen, elastin. "Essential" amino acids cannot be synthesized on their own. The use of cosmetics & supplements are the only way to supply them for a healthy strong substrate. Non-essential- amino acids can be produced by the body. But with age, the production slows down, resulting in unbalanced skin & hair. **A** lipoaminoacid is the association of an amino acid & a fatty chain. The fatty chain being the vector of the amino acid to its target. By masterizing the lipid profile of substrate and by combining specific amino acids with the right vector for perfectly targeted activity on skin & hair.

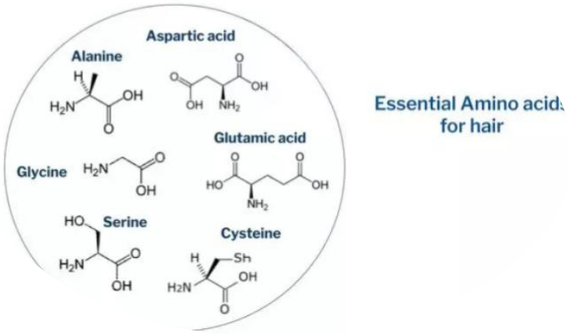
Key Words- Cleansing, Conditioning, lipoamino acid, proteins, surfactant, hair morphology.

I. INTRODUCTION

Hair a composition of living organism often idealizes for beautification purpose and have protective mechanism and health implications are made of amino acid which is a fibrous and helical protein with 90- 95% Keratin in the form of helical structure. As the insolubility of keratin in water it represents the hydrophobic structural mode.



Amino acid given the diversity of their structure and omnipresence in nature, constitute an essential resource for the creation of bio-inspired ingredients. As essential components of proteins, amino acids are encoded by the DNA of the cells of all living beings and play a

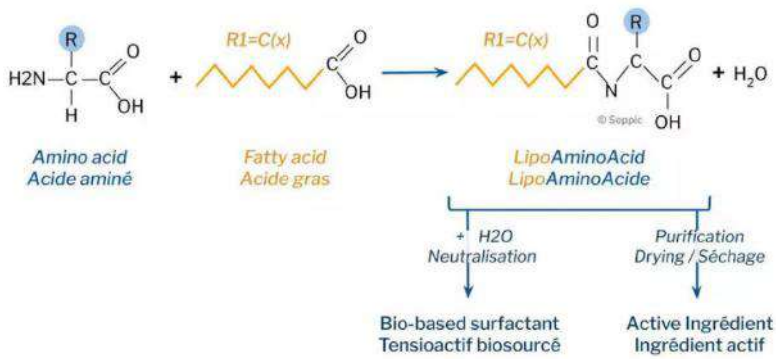


determining role both in the structure of organisms, and their biological metabolisms. Originating from plant sources or biotechnologies, they are carboxylic acids with an amine functional group. While their general structure is

NH2 – HCR – COOH, they differ in terms of the R group side chain and as the well-known fact of main constituent of hair is protein which is altogether amino acid.

Amino acid transformation

Amino acids are naturally hydrophilic molecules, potentially anionic or cationic depending on the pH. Grafting one or more hydrocarbon chains gives them an amphiphilic character, which amplifies their biological effects or surface properties. This is the essence of lipoaminoacid technology. On the one hand, grafting hydrocarbon chains (acylation) improves their solubilization in hydrophobic media (lipophilization). This facilitates their vectorization in living media and increases their bioavailability to create active ingredients with multiple biological targets. On the other hand, depending on the hydrocarbon chain selected, the amphiphilic character gives the possibility of creating biosourced surfactants. One advantage is that lipophilization processes are carried out in a single step at room temperature without sophisticated catalysts. The sources of lipophilic chains essentially come from fatty acid derivatives, which themselves originate from the oleochemical sector. Following this transformation, the lipophilized amino acids, also called lipoaminoacids, are subjected to two types of finishing operations. These come in two forms, either as aqueous solutions with a lipoaminoacid concentration of between 20 and 40%, or else in powder form with a high concentration of lipoaminoacids (>90%). In both cases the compositions, whether liquid or solid, are perfectly defined, and each constituent analytically quantified.



Lipophilization / acylation reaction
Lipoaminoacids, concentrated active ingredients for multiple purposes Acylation technology combines various amino acids and fatty acids with a chain length of 8 to 16 carbons that exist in nature. Adding a purification and drying step creates an inexhaustible source of concentrated active ingredients that can be used for cosmetics with a wide variety of biological targets

Selection of active ingredients:

1. Glycosphingolipids – Glycolipids (Highly purified Wheat exreact)
2. Hydrolyzed Wheat protein extract

Final Formula:

Sr no	Ingredients	% Quantity Placebo	% Quantity with Active (with Hydrolysed wheat protein)	% Quantity with Active (Glycosphingolipids – Glycolipids)
1	Deminarlised Water	25.24	23.24	23.24
2	Di Sodium EDTA	0.1	0.1	0.1
3	Sodium lauryl ether sulphate	39	39	39
4	Cocomonoethanolamide	1.5	1.5	1.5
5	Ethyleneglycolmonostearate	1.5	1.5	1.5
6	cocoamidopropyl betaine	5	5	5
7	xiameter 1785 Emulsion	3	3	3
8	Deminarlised Water	5	5	5
9	Gaur Hydroxy Propyltrimonium Chloride	0.06	0.06	0.06
10	MCIT MIT (Euxyl K120)	0.1	0.1	0.1
11	NaCl	0	0	0
12	Deminarlised Water	7.5	7.5	7.5
13	TRIquat(TRI K 10L)	0.5	0.5	0.5
14	water	4	4	4
15	Perfume(pompodour imp4)	0.5	0.5	0.5
16	Proteol APL	5	5	5
17	Glycosphingolipids – Glycolipids	0	0	2
18	Hydrolysed Wheat protein	0	2	0
	Total	100	100	100

Analysis Data:-

Details	% Quantity Placebo	% Quantity with Active (with Hydrolysed wheat protein)	% Quantity with Active (Glycosphingolipids – Glycolipids)
pH	**6.5**	**6.2**	**6.13**
Viscosity	**11230**	**12140**	**12420**
Moisture content (% w/w)	**76.8%**	**77.1%**	**77.32%**

Stability report: 1Month:-

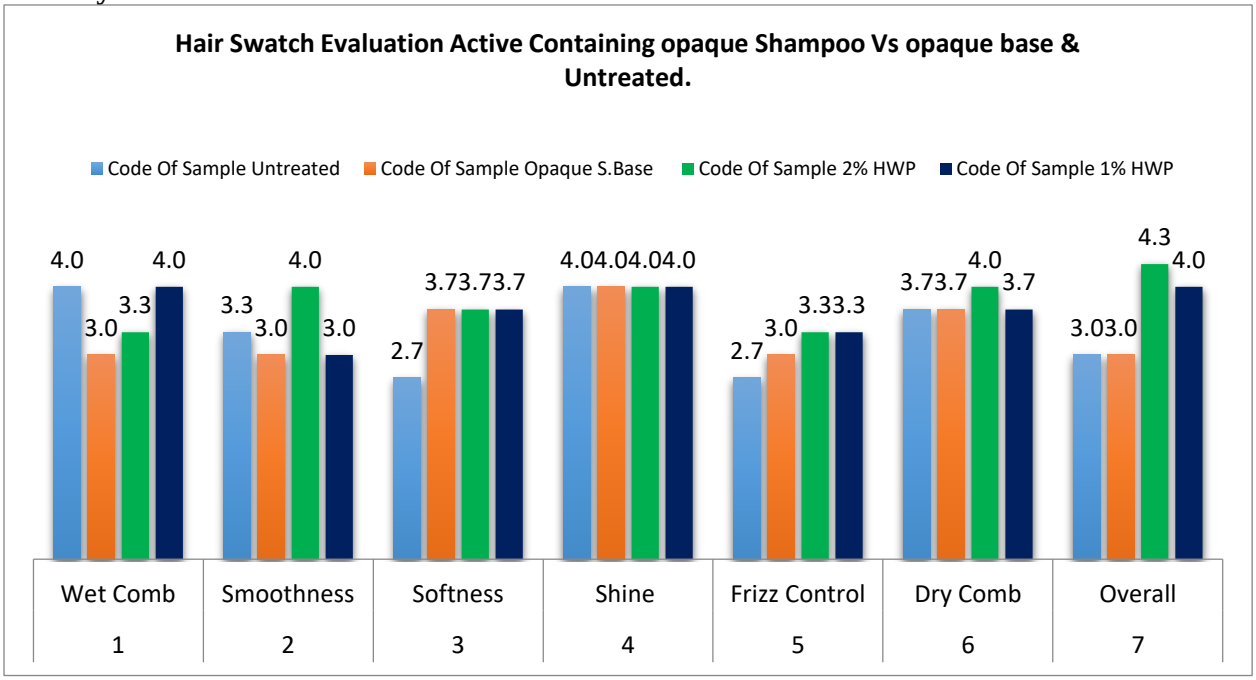
Batch Number-12	Condition			
	RT	REF	50°C	45°C/75 RH
PH	5.59	NA	NA	5.52
Viscosity(S.6 at 10 rpm)	11000cp	NA	NA	12800cp
M.C in %	77.58%	NA	NA	78.18%

2 Month

Batch Number-12	Condition			
	RT	REF	50°C	45°C/75 RH
PH	6.29	NA	NA	6.2
Viscosity(S.5 at 10 rpm)	13500cp	NA	NA	13200cp
M.C In %	76.37%	NA	NA	76.55%

3 Months

Sensory evaluation of hair swatch:



IN VITRO INSTRUMENT EVALUATION OF ACTIVES

The impact of actives on hair swatches through Diastron instrument for wet combing and dry combing force measurement and hair strength through tensile strength tester.

Study Objective:

To evaluate conditioning efficacy of actives through wet combing and dry combing force data.

Batch Number-12	Condition			
	RT	REF	50°C	45°C/75 RH
PH	6.27	NA	NA	6.14
Viscosity(S.05 at 10 rpm)	14000cp	NA	NA	14480cp
M.C in %	75.93%	NA	NA	75.88%

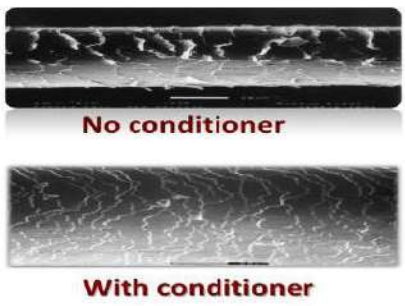
To evaluate strength imparted to hair shafts by the actives through tensile hair strength tester.

Instrument details:

A) Wet combing and dry combing force calculated with help of Diastron instrument:

- Diastron calculates combing force required to comb through hair tress.
- When conditioning shampoo is applied on hair, less force is required, that means conditioning effect of shampoo on that particular hair tress is good
- It is helpful technical measurement of product performance with respect to conditioning that provide guidance to formulation chemist, while also potentially being useful in claim derivation and product marketing.

Diastron Combing



Hair Fiber

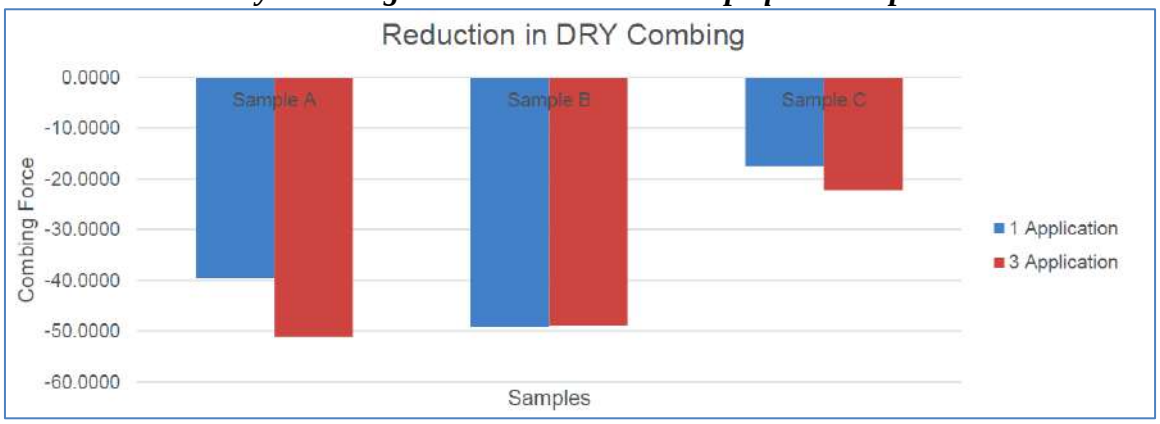
B) Tensile strength testing using Di-Stron instrument:

- Tensile testing is one of the most widely used methods in hair care industry, where strength of hair shaft can be determined by measuring the force used to break the shaft.
- More the force required to break the hair shaft post product application, better is the efficacy of product for against hair breakage.
- This method helps obtain claims in hair care industry related to hair strength, hair damage repair, hair hydration claims, etc.

Study Protocol: Wet and Dry Combing through Diastron Instrument:

Category	Details using Shampoo
Test Model	•9 Indian Semi Bleached Hair Tresses divided in to 3 groups (0.25 g each)
Materials	•Shampoo sample A, B and C as received
Treatments	•Group 1: Sample A •Group 2: Sample B •Group 3: Sample C
Instrument	Dia-Stron MTT175 Miniature Combing Tester
Treatment Methods	•Hair tresses were Washed with 10% SLES solution, and tested as Untreated Wet and Dry •Hair was then treated and washed with 0.5 g of shampoo A, B and C for 1 min •Hair dried under ambient conditions between cycles
Conditions	Water Temperature: 30°C ± 2, Relative Humidity: 50% ± 5, Room Temperature: 20°C ± 2
* *Protocol remains same for Transparent and Opaque shampoo evaluation*	

Dry Combing Force Reduction with Opaque Shampoo



Observations-

1. Shampoo A (with Glycosphingolipids – Glycolipids (Highly purified Wheat exreact) has shown reduction in DRY combing force as 39% after 1 application as compared to untreated
2. Shampoo A (with Glycosphingolipids – Glycolipids (Highly purified Wheat exreact) has shown reduction in DRY combing force as 51 % after 3 applications as compared to untreated
3. Shampoo B (Hydrolyzed Wheat protein extract) has shown reduction in DRY combing force as 49 % after 1 application as compared to untreated
4. Shampoo B (Hydrolyzed Wheat protein extract) has shown reduction in DRY combing force as 49% after 3 applications compare to untreated

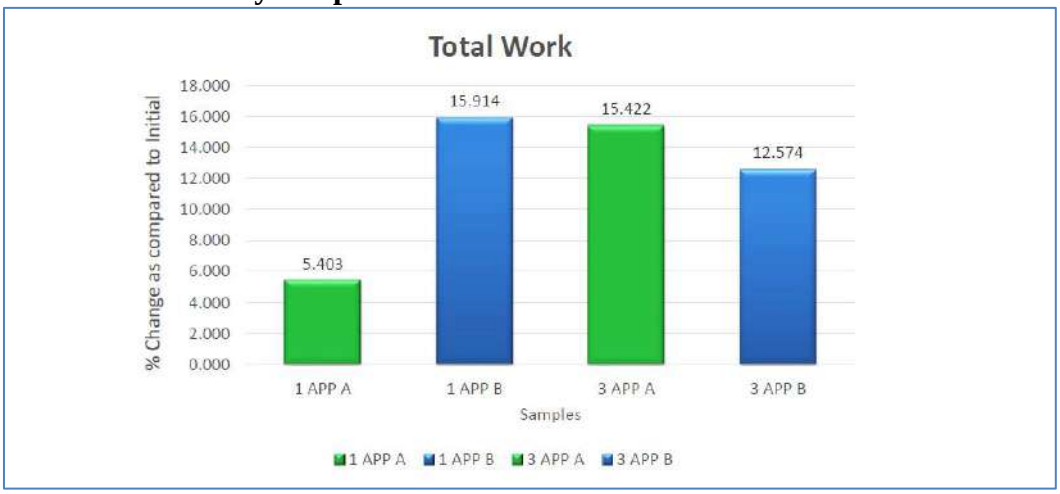
5. Shampoo C (Placebo) has shown increase in DRY combing force as 18% after 1 application compare to untreated
6. Shampoo C (Placebo) has shown reduction in DRY combing force as 22% after 3 applications compare to untreated

Study Protocol: Improvement in Indian Hair Tensile Properties:

Category	Tensile
Test Model	•3 swatches(5 g each) Partially bleached Indian Hair
Materials	•Control –Untreated Opaque Shampoo from JK Helen Curtis •Sample A •Sample B
Treatments	•Group 1: Untreated •Group 2: Sample A with 2% Quinoa Pro NPNF •Group 3: Sample B with 2% Barla Tein NPNF
Instrument (s)	Dia-StronMTT175 Miniature Tensile Tester & Dia-Stron Crimp Assembly System
Treatment Methods	•Hair swatches were clarified with 10% SLES Solution and dried at ambient condition overnight •Hair was treated with 1 and 3 cycles of shampooing (1 min) for both set of sample A and B separately. Hair were dried at ambient conditions in between the cycles •Brass crimps were added to the ends of the fibres following Dia-Stron method. •Measurements were taken after 1 and 3 cycles (average of 50 fibres)
Conditions	Water Temperature: 30°C ± 2, Relative Humidity: 50% ± 2, Room Temperature: 20°C ± 2

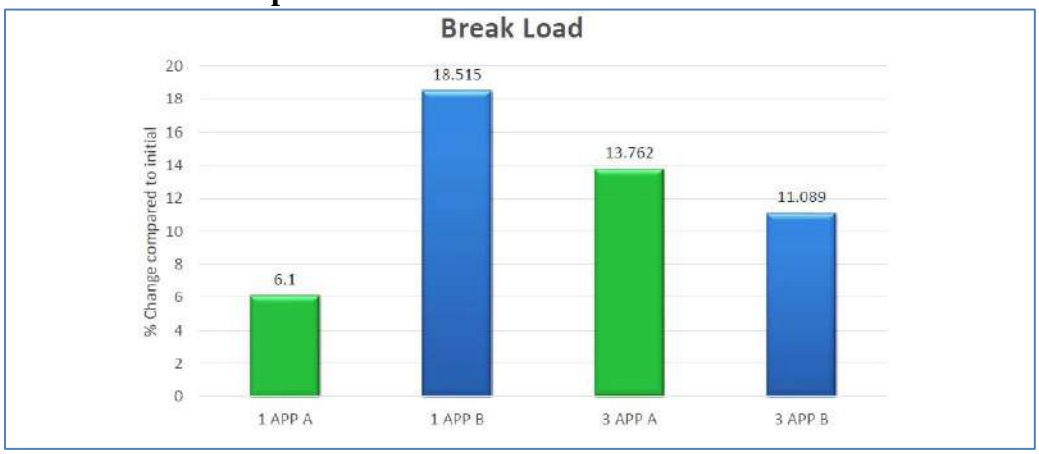
Study Findings:

Study –Improvements in Indian Hair Total Work



Study –Improvements in Indian Hair Break Extension

Improvements in Indian Hair Break Load



Study Inference:

1. Both Barley Tein Pro and Quinoa pro NPNF have shown positive results in conditioning efficacy and hair tensile test against bases without actives.
2. Within the 2 actives Quinoa Pro NPNF has outperformed other active and shows a substantial improvement in both the tests in a wash off format.
3. Considering the baseline as untreated value, bases developed for both formats transparent and opaque have shown positive readings to a substantial extent. Hence efficacy of both bases – transparent and opaque is successfully validated through the above In Vitro Study.

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10

Design and evaluation of skin brightening serum with potent antioxidant

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Abstract-
A fresh complexion is a sign of vitality and well-being. A luminous and homogeneous skin is associated with youth and good health. It is a sign of a social status and attraction asset. A radiant skin requires a good reflection of light on its surface. A glow can be influenced by uniformity, smoothness, skin color. The efficacy of product active ingredient with antioxidant action, uniformity of skin color and skin hydration are the key area to study and evaluate the impact of product and active on luminosity of substrate.

Key words- Brightening, TEWL, Melanin, Suger complex, Vitamin C derivative, Tyrosinase inhibition.

Introduction

Traditionally, moisturization was believed to inhibit trans-epidermal water loss by occlusion. Research suggests that the stratum corneum acts an active membrane consisting of intercellular lipids (i.e. ceramides, cholesterol and fatty acids), thereby forming a water-barrier function. In addition, the stratum corneum contains a natural mixture of amino acids, lactates, urea and electrolytes, which also help retain water. Dry skin is noted when the moisture content is less than 10%, and there is loss of continuity of the stratum corneum.

The basic function of moisturizers is to help treat your skin when it's dry and prevent it from drying out again. Moisturizers do this by holding water in the stratum corneum, the outermost layer of the skin. But they have other functions as well. They can help protect your skin from the environment applying moisturizer, creates a barrier on your skin that keeps oils from escaping and harmful outside elements from causing dryness or irritation escaping and harmful outside elements from causing dryness or irritation escaping and harmful outside elements from causing dryness or irritation.

Scientifically, the moisturizing treatment involves a four-step process:

- Repairing the skin barrier
- Increasing water content
- Reducing trans-epidermal water loss
- Restoring the lipid's water barrier function

Class	Compounds	Mode of action
Occlusive	Petrolatum, lanolin, mineral oil, silicones, zinc oxide	Physically block TEWL
Humectants	Glycerin, propylene glycol, sorbitol, hexylene glycol, butylene glycol, urea, α-hydroxy acids (AHA's)	Attract water to stratum corneum
Emollient	Plant oils, polyisobutenesqualene, fatty acids, ceramide (e.g. lacto ceramide)	Smooth skin by filling spaces between skin flakes with droplets of oil
Protein	Collagen, keratin, elastin protein mixtures (e.g. wheat protein)	Replenish proteins in stratum corneum

Table 1- Moisturizing compounds and their mode of action

Skin Pigmentation:

Human skin colour ranges in variety from the darkest brown to the lightest hues. An individual's skin pigmentation is the result of genetics. The actual skin colour of different humans is affected by many substances, skin pigmentation in humans evolved to primarily regulate the amount of ultraviolet radiation (UVR) penetrating the skin, controlling its biochemical effects. The chromospheres are the part of a molecule responsible for its colour.

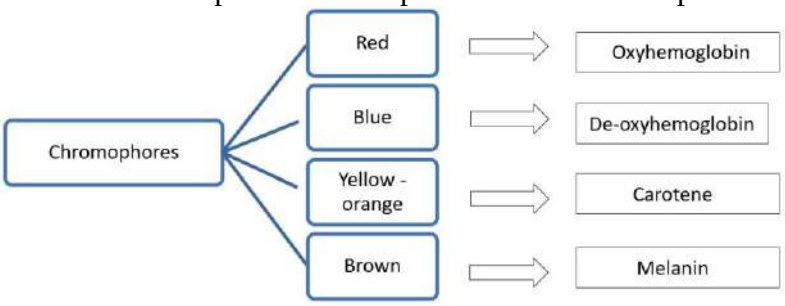
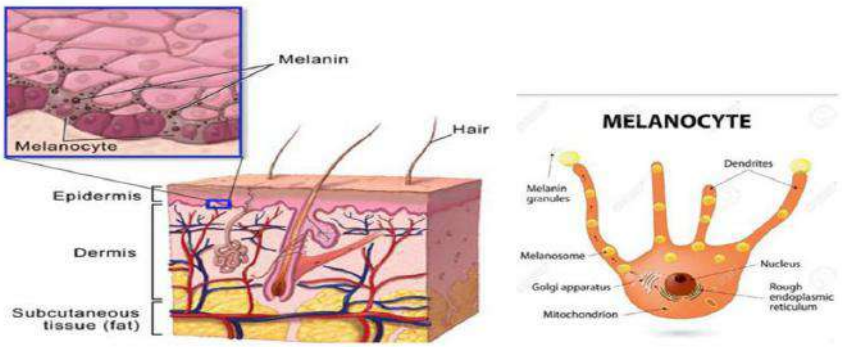


Fig: Chromospheres responsible for skin colour

It concerns about the excessive release of particular pigment in specific area or unprotected area. Melanocytes synthesize melanin. From precursors in the neural crest, melanocytes migrate normally to the epidermis. Normally in human skin scattered along the plan of the epidermal/dermal junction and in tube, hair bulb. In the latter area responsible for the colour of hair while in the former they are response for colour of the skin. And are basically located in basal layer of epidermis



Mechanism of skin brightening ingredients

Before going for skin brightening it is necessary to understand pigmentation or darkening process, though tyrosinase is an enzyme behind the colouration of skin which stimulates the release of melanin from melanocytes. As Melanin is of 2 types

1. Eumelanin (Black-brown colour)
2. Pheomelanin (Red to yellow colour)

In this the activation of cycle starts with tyrosinase act as catalyst for tyrosine to release melanin so Brightening or lightening ingredients start working here by suppressing the tyrosine inhibition or reducing the overproduction of melanin which is responsible for skin darkening or can say as pigmentation.

Ingredient selection

Emulsion a system consisting of two immiscible phases stabilised by using emulsifier. Among these two immiscible phases the dominant one is called as continuous phase or dispersion phase or external phase and the other one is called as dispersed phase or internal phase.

Emulsifiers:

The inclusions of emulsifying agents are necessary for the emulsification process during manufacture, and also to ensure emulsion stability during the shelf-life of the product. A useful

method has been suggested for calculating the quantities of these emulsifying agents necessary to produce physically stable emulsion. This is called the hydrophilic-lipophilic balance (HLB) method.

I. Synthetic emulsifiers

1. Anionic surfactants:
2. Cationic
3. Amphoteric surfactants:

II. Natural emulsifiers

III. Semi synthetic emulsifiers

Other formulation additives

In addition to the emulsifier, other additives such as buffers, preservatives, thickening agents, sensory modifiers, colours and sweeteners are also used.

Other Serum Additives:

Polymers and Viscosity modifier: -

Emollients: -

Humectants

Sensory modifiers:

Preservatives:

Miscellaneous:

Along with above key additives, emulsion systems may contain UV / photo stabilisers, for avoiding colour fading of product, Colours, pigments, fragrances for aesthetic appeal of product , Sequestering agents, pH regulators like buffer solutions or neutralising materials.

Formulation Development:

2 types of actives are considered for incorporation in selected bases.

1. Moisturising Active
2. Brightening Active

Moisturising Active

Sr. No.	INCI	Major benefits	Purpose
1	Xylitylglucoside (and) Anhydros xylitol (and) Xylitol	Optimized circulation of water and reserves	To Give the plus plusmoisturisation to the skin
		oost the production of Hyluronic acid in keratinocytes and fibroblast	As it imboost the hyaluronic production the skin will feel more moisturised and hydratedas because it reduces TEWL and keep balance in loss and gain of moisture by restoration to keep fresh skin
		It maintain the epidermal water content i.e. reduces transepidermal water loss	

Brightening Actives

Sr. No.	INCI	Major benefits	Purpose
1	L-Ascorbyl 2-phosphate magnesium salt	Skin Brightening, depigmentation,	It claims that as it is magnesium salt it is stable compared to sodium salt which is also commonly used in benchmarks.
		UV Protection	Existing active ingredient used in many skin lightening preparations so in order to understand how it perform this one selected
		Anti-ageing, Acne treatment	
2	3-O-Ethyl ascorbyl ether Ethyl ascorbic acid	Skin Lightening	It consisting of a stabilized ethyl L-ascorbic acid by etherification, and claim to be providing comparatively better skin brightening effect as compared to Magnesium ascorbyl phosphate
		Antioxidant and ant ageing	
		High index of stability	
3	Water, Hydrolyzed Oat Protein, Ethyl Ascorbic Acid, Alpha-Arbutin	Inhibits tyrosine better than individual ingredients & benchmarks (Comparative study provided by vendor)	Claims to be containing multiple ingredients with optimized ratios for superior performance
		Contains calming oat oligopeptides which will provide reduction in dullness of skin with bright appearance	Alpha arbutin present with oat protein which is going to boost the skin lightning with calming

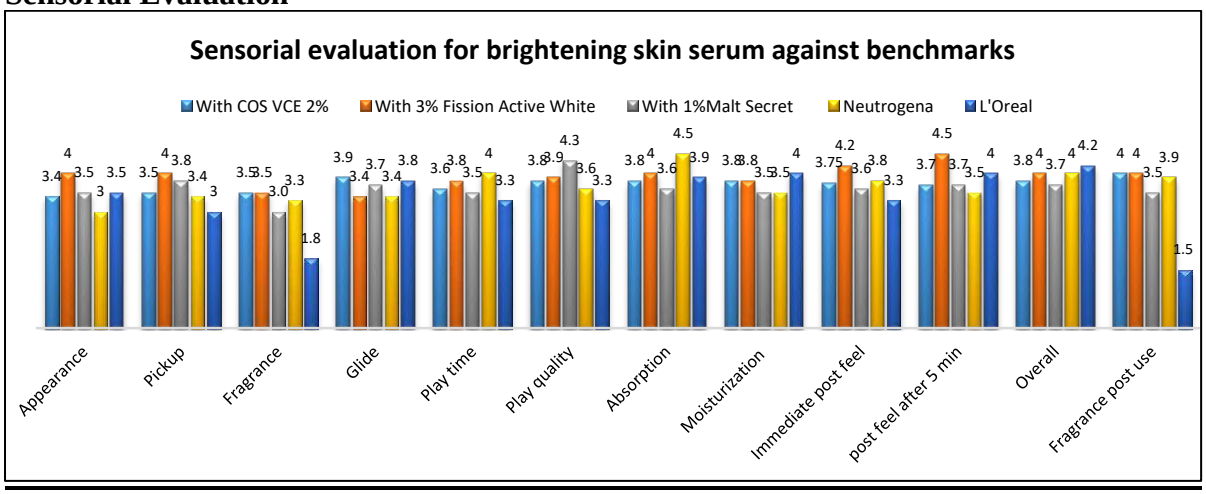
Parameters used for evaluating serum formulation:

Pre-use parameters	Appearance
	Pickup
	Fragrance
In-use parameters	Strokes to absorb product in skin
	Glide
	Play time
	Play quality
	Absorption
Post-use parameters	Immediate post feel
	post feel after 5 min
	Fragrance post use

Formulation:

Sr. No	Ingredient	INCI	Quantity (%) (Placebo)	Quantity (%) (Placebo)
1	Di Water	Aqua	Up to 100	Up to 100
2	Xanthan Gum	Xanthan Gum	0.50	0.50
3	Aristoflex AVC	Ammonium acryloyldimethyl taurate/ VP Copolymer	0.75 (0.75 – 1.2%)	0.75 (0.75 – 1.2%)
4	Di EDTA	Ethylene diamine tetra-acetic acid	0.10	0.10
5	Brij L 23	Laureth 23	1.00	1.00
6	CCTG	Capric Caprylic Tri Glyceride	3.00	3.00
7	Glycerin	Glycerol	2.00	2.00
8	Propylene glycol	Propane-1,2-diol	1.00	1.00
9	Simulsol 165	PEG-100 Stearate (and) Glyceryl Stearate	1.00	1.00
10	Span 120	Sorbitan Isostearate	1.00	1.00
11	SF 1202	Cyclopentasiloxane	2.00	2.00
12	Element 14 PDMS 350	Dimethicon	2.00	2.00
13	Velvesil 125	Cyclopentasiloxane (and) C30-45 Alkyl Cetearyl Dimethicone Crosspolymer	2.00	2.00
14	Dryflo PC	Aluminum Starch Octenylsuccinate	2.00	2.00
15	Euxyl PE 9010	Phenoxyethanol (and) Ethylhexylglycerin	0.50	0.50
16	Xylityl glucoside (and) Anhydros xylitol (and) Xylitol	Xylityl glucoside (and) Anhydros xylitol (and) Xylitol	0.0	3
17	Water, Hydrolyzed Oat Protein, Ethyl Ascorbic Acid, Alpha-Arbutin	Water, Hydrolyzed Oat Protein, Ethyl Ascorbic Acid, Alpha-Arbutin	0.0	3

Sensorial Evaluation



The resultant products are comparable with benchmarks and though the product skin brightening serum with Fision Active White 3% is selected for external consumer study and claim substantiation

Stability studies: Stability conducted as per ICH guidelines:

Parameters / Conditions and duration	pH @ 27°C	Viscosity (Cps) @ spindle no. 5, 10 rpm	Moisture Content (%)	Appearance	Fragrance
Initial	**5.02**	**18960**	**80.96**	**Pale white**	
1 Month					
RT	5.08	18220	83.2	No Change	No Change
Ref	4.71	18700	83.5	No Change	No Change
45°C/75°RH	4.72	18320	83.07	No Change	No Change
50°C (Dry Heat)	5.13	17280	84.12	No Change	No Change
2 Months					
RT	4.88	18900	83.6	No Change	No Change
Ref	4.98	18720	83.3	No Change	No Change
45°C/75°RH	4.71	19210	82.9	No Change	No Change
3 Months					
RT	5.04	19400	83.37	No Change	No Change
Ref	5.14	18920	83.54	No Change	No Change
45°C/75°RH	4.79	19600	82.2	No Change	No Change

IN VITRO INSTRUMENT EVALUATION OF ACTIVES

Brief Overview:

Out of four one of the active (Fision Active white) was tested in in-vitro instrument evaluation by the supplier of active. This chapter will cover the impact of active ingredient through Mexameter® MX18 (Courage Khazaka) instrument for studying skin lightening property of product with Diffuse Reflectance Spectroscopy (DRS)

Active Details:

Fision Active White:

A complex blend of Water, Oat Oligopeptides, Ethyl Ascorbic Acid (Vitamin C), α-Arbutin with multifunctional ingredient which serves as a skin lightening agent, anti-ageing ingredient, photo-protective ingredient and calming agent (Oat oligopeptide).

Sample details:

1. 2 samples of skin serum were given to the principal lab for evaluation
2. These contained finalised trials based on internal sensorial evaluation. The sample was tested in comparative evaluation against the formulation base without active.

Study objective:

To evaluate skin lightening efficacy of product with reduction in melanin index, the Mexameter® MX 18 is used to check the efficacy of product containing active (Fision Active White).

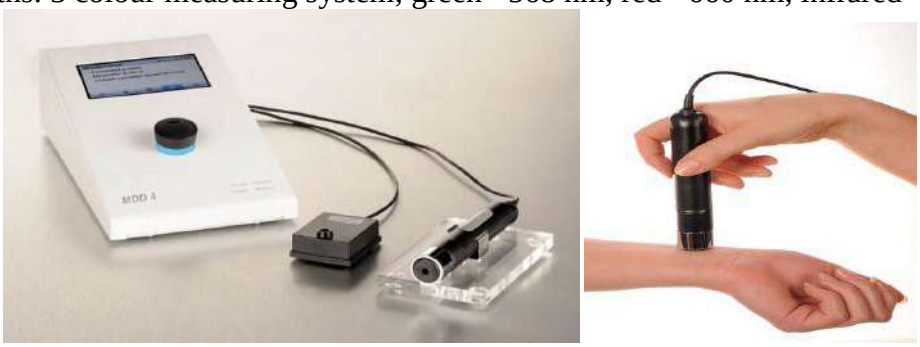
Instrument working principal:

The Mexameter is a spectrometer measurement technique, based on light reflection and absorption. The probe emits three wavelengths of light, chosen to correspond to the different absorption rates of melanin and haemoglobin.

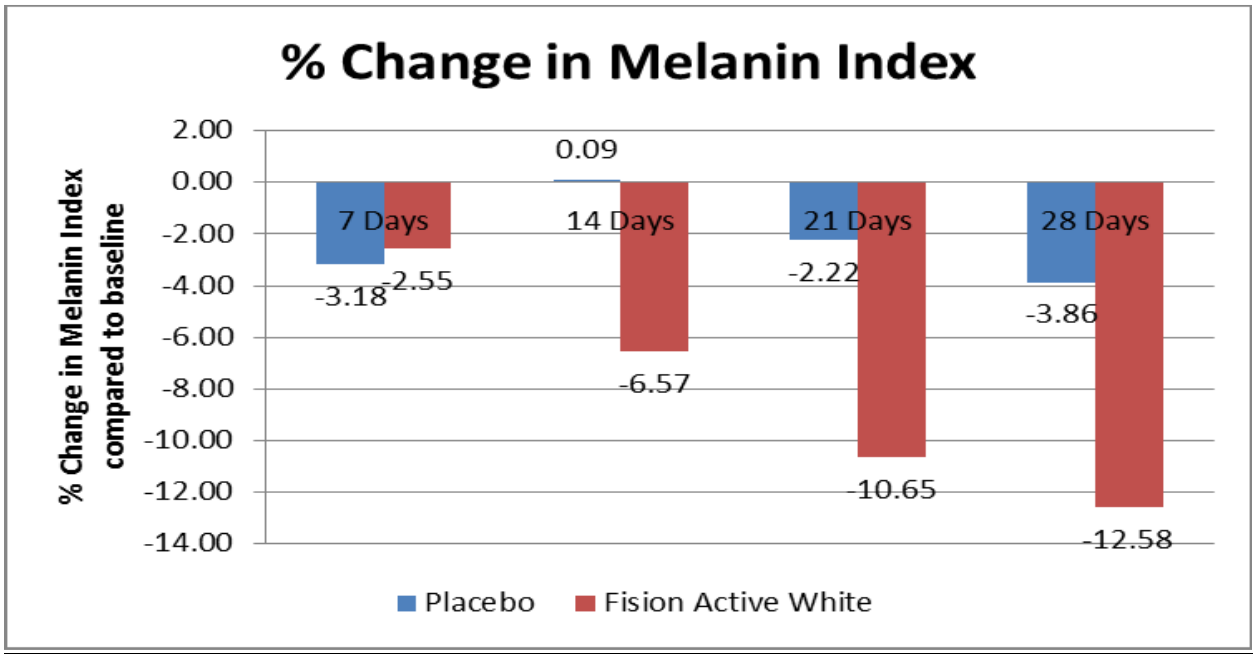
The light emitted by the probe is reflected by the skin and the receiver in the probe measures this reflected light. It is only the diffuse and scattered light that is measured. The results are shown in 1 second as index numbers between 0 and 999.

The probe allows the measurement to be made quickly (1 second). The probe head is spring loaded so that a constant pressure is provided.

(Wavelengths: 3 colour measuring system; green =568 nm, red =660 nm, infrared =870 nm)



Mexameter with probe



Reading: Negative (-ve) quadrant indicates the reduction in melanin index of skin (Improved skin lightening).

Observations:

- 7 days post application of product on regular basis the melanin index reduces to 2.25% which when compared to placebo (Product sample without Fision Active White) it was performing better as it reduces 3.18% melanin index.
- After 14 days on regular use of product the product with fission active white reduces the melanin index up to6.57% which proves to work better than placebo.
- After 21 days of regular product use the melanin index drops to 10.65% and when compared with placebo (melanin index 2.2%) is better.
- On continuous use of product with Fision Active White the melanin index drops to 12.58% which shows the better results than placebo (melanin index 3.86%).

For Moisturization efficacy-

A finalised moisturising active ingredient Aquaxyl 1% was tested in In-vitro evaluation for hydration/ moisturization property against placebo.

Active Details:

Xylityl glucoside (and) Anhydros xylitol (and) Xylitol

A moisturising active ingredient which helps to optimize circulation of water and reserve it, with boosting the Hyaluronic acid in Keratinocytes and fibroblast

Sample details:

1. 2 Samples were used for this test to check the hydration property of serum.
2. 1 placebo (Base) and 1 with 1% Aquaxyl

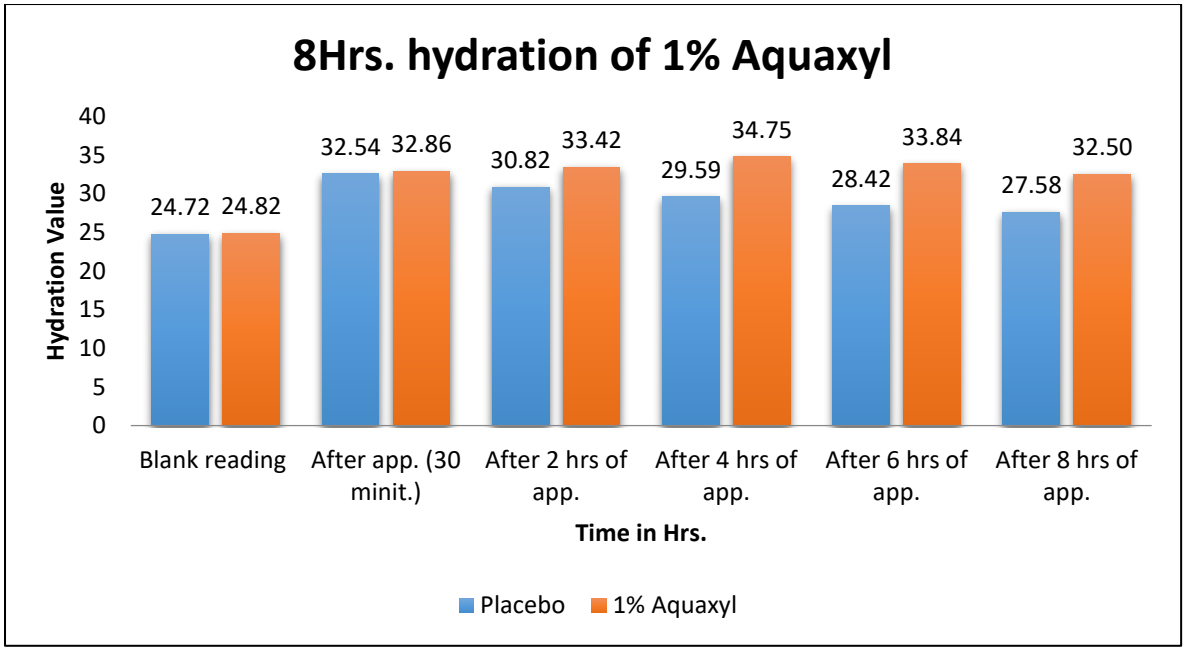
Study objective:

To Study the hydration property of skin serum with a moisturising active ingredient using Corneometer®

Instrument working principal

The Corneometer® is the mostly used instrument worldwide to determine the hydration level of the skin surface, mainly the Stratum corneum. The measurement is based on capacitance measurement of a dielectric medium. The Corneometer® measures the change in the dielectric constant due to skin surface hydration changing the capacitance of a precision capacitor.

<u>Skin hydration effect with 1% Aquaxyl</u>



Observations:

- 30 min. post application the hydration of skin is almost the same which is 32.54 and 32.86 hydration number.
- After 2 hrs. the hydration level of placebo drops to 30.82 which is comparatively less than product with 1% aquaxyl (33.42).
- Subsequently after 4 hrs. and 6 hrs. the product with 1% aquaxyl is maintaining the hydration value.
- 8 hrs. post product application the hydration value of skin is better than placebo and blank which 32.50 hydration number.

Conclusion:

The skin brightening serum with active ingredient Fision Active White proves to substantiate the claim skin brightening (Lightening) by reduction in melanin index upto 12.58% in 28 days.

The skin serum with 1% aquaxyl is providing good hydration property for 8 hrs. & better in comparison to the placebo.

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11

Formulation and Evaluation of Anti-Acne Face Wash Gel by using Extracts of Curry Leaves and Bael Leaves

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ABSTRACT:
The objective of this work is to formulate and evaluate gel base face wash by using herbal extracts. From ancient times, there has been awareness among the people regarding the use of plants for the essential needs of healthy and beautiful skin. Cosmetic designed with incorporating natural sources, such as herbs. Face Wash are used to get rid from dirt, oil, pollution etc. Many plants has been shown to be effective for anti-acne treatment. Curry leaves are used in traditional medicine as a source of many therapeutic agents in the Indian culture and grows well in the tropical countries. Curry leaves, known as Murraya Koeniggi, belonging to Rutaceae family are widely used as a medicinal herb and has characteristic aroma. It is rich source of carbazole alkoids, Carbohydrates, steroids and flavonoids are also present in the root extracts of the plant. It showed some antimicrobial activity as well as antifungal activity. The gel face wash Leaves, fruits, stem and roots of Aegle marmelos have been used in ethno medicine to exploit it's medicinal properties including astringent, antidiarrheal, demulcent, and anti-inflammatory activities. Formulation FW2 shows anti acne properties which also showed good Reheological chatracterstics ,pH , spredability , stickiness, greater active content. Hence this study showed that FW2 was the best formulation for anti-acne face wash. According to In-Vivo study, the product has no skin irritation and redness form after applying on the skin.
KEYWORDS: Anti-Acne Face Wash, Murraya Koeniggi, Aegle marmelos , antimicrobial activity, astringent.

INTRODUCTION:

Face wash

Face wash is the products which are used to cleanse face without drying it out. Face wash is very helpful in removing dirt, oil and provide moisture to the skin. Face Wash are used to get rid from dirt, oil, pollution etc. A cleanser dissolves away excess oil makeup and grime from your face. These are oil soluble impurities. Facial skin is the delicate and ordinary soaps can cause it to lose moisture. The purpose of face wash may be to impart cleansing, anti-acne property and moisturizing effect to the skin. And it is commonly called as cleansers.

Forms of face wash

1. Cream base face wash
2. Gel base face wash
3. Liquid base face wash
4. Face wash in powder form

Gel based face wash

A gel is a solid jelly like material that can have properties ranging from soft and weak to hard and tough. Gels are defined as a substantially dilute cross-linked system, which exhibits no flow when in the steady-state. By weight, gels are mostly liquid, yet they behave like solids due to a three-dimensional cross-linked network within the liquid.

Types of face wash

- Oily skin face wash
- Dry skin face wash
- Normal skin face wash

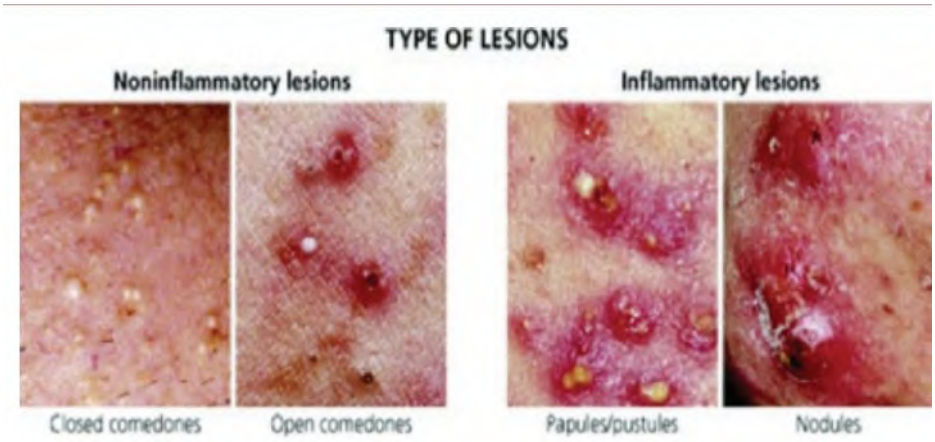
Ideal properties of face wash

- Removing the dead cell from the skin
- Removing oil and dirt from the skin
- Reduces microbial flora of skin.
- Leave skin fresh and breathing. [52]

ACNE

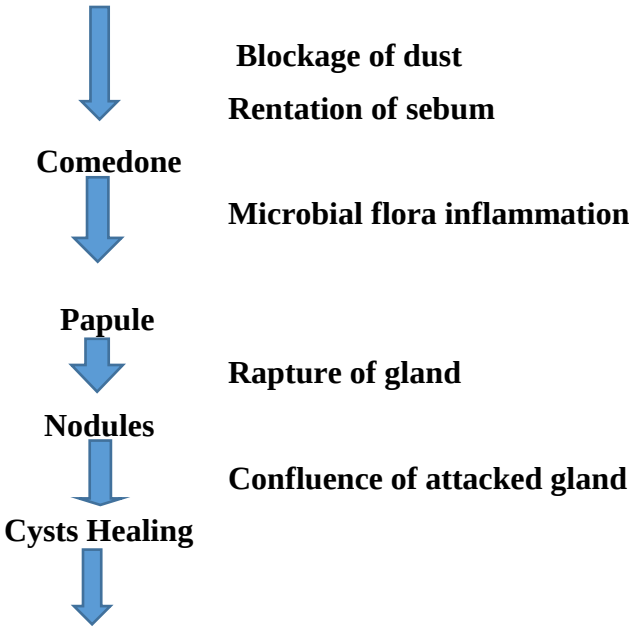
Acne is an infection of the skin, caused by changes in the sebaceous glands. The most common form of acne is called acne vulgaris, which means "common acne". The redness comes from the inflammation of the skin in response to the infection. Oils from the glands combine with dead skin cells to block hair follicles. Under the blocked pore, oil builds up. Skin bacteria can then grow very quickly. This infection makes the skin become swollen and red, which becomes visible. The face, chest, back, and upper arms are most common places for acne to happen. Acne is common during puberty, when a person is turning from a child into an adult, because of high levels of hormones. Acne becomes less common as people reach adulthood. Acne vulgaris is an extremely common disorder of skin (pilocebaceous unit) that affects virtually all individuals at least once during life. The incidence of acne peaks at teenage, but substantial numbers of men & women between 20-30 years of age are also affected by the disorder. Adolescent patients have reported low self-esteem and symptoms of depression leading to a lower quality of life. Acne vulgaris is an extremely common disorder of skin [pilocebaceous unit] that affects virtually all individuals at least once during life. Acne vulgaris is one of the most common dermatological disorders that afflict people in their adolescence. Acne vulgaris or simply known as acne is a human skin disease characterized by skin with scaly red skin (seborrhea), blackheads and whiteheads (comedones), pinheads (papules), large papules (nodules), pimples and scarring. Acne vulgaris is a disease of pilosebaceous unit characterized by the formation of open and closed comedones, papules, pustules, nodules and cysts. Acne affects skin having dense sebaceous follicles in areas including face, chest and back. Acne is not life threatening but severe acne can affect psychological status and social activities. The present review focuses on an epidemiology, etiology, pathogenesis, diagnosis, differential diagnosis and management of acne with the pharmaceutical dosage forms of oral and topical administrations. Currently laser and light devices and minor subcision surgery have been also performed for acne treatment.[3],[4],[5],[6]

Stages of development and formulation of Acne

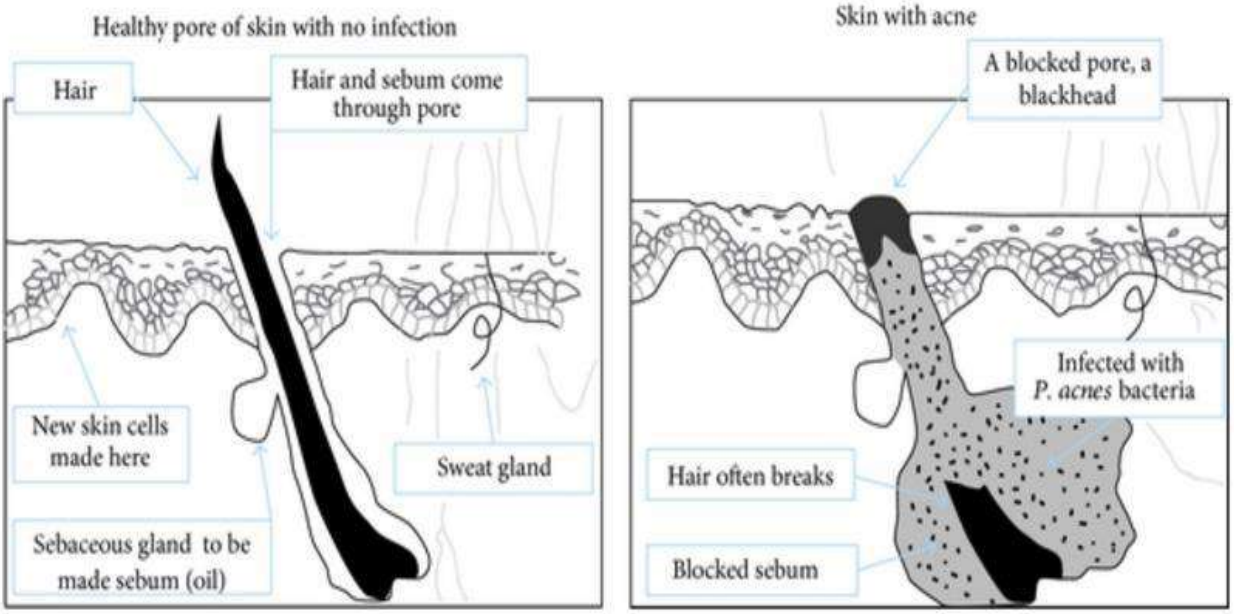


Stages of development and formation of Acne

Sebaceous Glands (Sebum)



Flow Charts of stages of acne



Anti- Acne Agents:
MURRAYA KOENIGII

Plant products nowadays play an important role in the world population. People use herbal product because they are considered as safe, inexpensive and less side effects. Murraya koenigii

contains phytochemicals such as saponins, proteins, Murraya koenigii contains phytochemicals such as saponins, proteins, steroids, tannin, carbohydrates, alkaloids, flavonoids and glycoside. It has antimicrobial, antifungal, antidiarrheal, anticancer, antidiabetics and anti-inflammation. It has the skin improving effect. The Murraya koenigii extract from leaves provide a higher amount of polyphenols and antioxidant activity. Murraya koenigii showed significant antibacterial activity against Staphylococcus aureus and Staphylococcus Epidermidis. The curry tree Murraya koenigii is a tropical to sub-tropical tree in the family Rutaceae. Curry tree is also called curry leaf tree or curry bush, among numerous local names, depending on country. This plant is known to be the richest source of carbazole alkaloids, Mahanine, Mahanimbicine, Mahanimbine, Vitamin A and Isomahanimbine.[47]

AEGLE MARMELOS

Aegle marmelos, commonly known as bael, also stone apple or wood apple, is a species of tree native to the Indian subcontinent and Southeast Asia. It is present in India, Sri Lanka, Nepal, Thailand, and Malesia as a naturalized species. The tree is considered to be sacred by Hindus. Bael (Aegle marmelos (L.) Corr.) is an important medicinal plant of India. Leaves, fruits, stem and roots of Aegle marmelos have been used in ethno medicine to exploit it's medicinal properties including astringent, antidiarrheal, demulcent, and anti-inflammatory activities. Compounds purified from bael have been proven to be biologically active against several major diseases including cancer, diabetes and cardiovascular diseases. Preclinical studies indicate the therapeutic potential of crude extracts of Aegle marmelos in the treatment of many microbial diseases, diabetes and gastric ulcer. This review covers the biological activities of some isolated chemical constituents of Aegle marmelos and preclinical studies on some crude extracts and pure compounds to explore novel bioactive compounds for therapeutic application.[16],[17]

MATERIALS AND METHODS:

List of ingredients required:

1. **Carbopol Ultrez 20** - Lubrizol
2. **Sodium lauryl sarcosinate**- Galaxy Surfactant Ltd.
3. **Cocoamidopropyl Betain**- Galaxy Surfactant Ltd.
4. **Propylene Glycol** -VBMV
5. **Triethanol amine**-VBMV
6. **Distilled Water**- VBMV
7. **Disodium EDTA**-VBMV
8. **Phenoxyethenol**-VBMV
9. **Bael Leaves Extract**-Konark Herbal Pvt. Ltd.
10. **Curry Leaves Extract**- Konark Herbal Pvt. Ltd.

List of Equipments

1. Precision balance: CA series contech

2. Mechanical stirrer: Shettal Scientific industry Pvt ltd.,Mumbai

3. pH meter: Digital Model 111E-E-1 Electronic india

4. Brook field Viscometer: S.M.S Scientific Industry Pvt.Ltd. Mumbai (DV-E-version1, E-34/03)

Method of prepration of Face wash:

The prepration of face wash is very important and before incorporation of active ingredients. The ingredients used in preparation of Face wash are mentioned in table no.1

Formulation of gel face wash :

Table No 1: Formulation of Base face wash

Sr. No.	Ingreadients	FW1 For 100%	FW2 For 100%	FW3 For 100%
1	Carbopol Ultrez 20	0.5	0.6	0.6
2	Sodium Lauryl Sarcosinate	18.3	18.6	17.6
3	Cocoamidopropyl Betain	5	3	3
4	Propylene Glycol	4	4	4
5	Triethanol Amine	0.4	0.4	0.4
6	Distilled Water	71	72	72
7	Disodium EDTA	0.1	0.1	0.1
8	Phenoxyethanol	0.3	0.3	0.3

Optimization of Face wash Procedure

Procedure: Sprinkle some Ultrez 20 in water and then add triethanol amine to adjust the pH then add Sodium Lauryl Sarcosinate ,Cocoamidopropyl Betain , Propylene Glycol Disodium EDTA and Phenoxyethanol in the end.

Formulation of Face wash containing curry leaves and bael leaves extract

Table no 2: Formulation of face wash containing bael leaves and curry leaves extract

Sr. No.	Ingredients	FW1	FW2	FW3
1	Carbopol Ultrez 20	0.5	0.6	0.6
2	Sodium lauryl sarcosinate	18.3	18.6	17.6
3	Cocoamidopropyl Betain	5	3	3
4	Propylene Glycol	4	4	4
5	Triethanol amine	0.4	0.4	0.4
6	Distilled Water	71	72	72
7	Disodium EDTA	0.1	0.1	0.1

8	Phenoxyethanol	0.3	0.3	0.3
9	Bael leaves extract	0.2	0.5	1
10	Curry leaves extract	0.2	0.5	1

Procedure of face wash:

- Clean all the ingredients as per the formulation of face wash
- 0.1 gm of disodium EDTA was dissolve in water
- The ultrez 20 was dispersed in the water
- pH were adjust with the help of TEA
- Surfactant were added into the formulation
- At the end propylene glycol
- And Phenoxyethanol which is first mixed with propylene glycol
- In the end both actives are incorporated.

FW2 was the best formulation for anti-acne face wash

Final formulation of Curry leaves and Bael leaves extracts

Table no. 3 Final formulation of face wash FW2 containing Curry leaves and Bael leaves

Sr. No.	Ingredient	FW2
1	Carbopol Ultrez 20	0.6
2	Sodium lauryl sarcosinate	18.6
3	Cocoamidopropyl Betain	3
4	Propylene Glycol	4
5	Triethanol amine	0.4
6	Distilled Water	72
7	Disodium EDTA	0.1
8	Phenoxyethanol	0.3
9	Bael leaves extract	0.5
10	Curry leaves extract	0.5

EVALUATION:

In- Vitro Studies:

1) Determination of physical parameters:

Apperance: Visually appearance of the formulation observed

Colour: Colour of the formulation check visually.

Consistency: Consistency was check weather its satisfactory or poor or good.

Tacky feel: Tackiness were check after application on palm.

2) Determination of pH

Principle: Face wash are used for topical application so there ph should be similar to that of with the skin. The skin these acidic mantle and the pH of the face wash as per standard should be in the range of 5-9

Appratus : pH meter

Procedure: Take 1gm of sample and dissolve in 100ml of water in beaker that is 1% solution is prepared. Then with the help of pH meter reading were taken.

3) Determination of viscosity:[27]

Appratus: Brookfield viscometer

Principle: The viscosity most important parameter in the evaluation of cosmetic product. Viscosity governs the many properties such as spreadability, pourability of the product from the container. As viscosity is affected by many factors such as change in temperature, change in manufacturing condition, quality of the raw material. Hence it is very important to measure viscosity of product.

Proceduce: The viscosity of Face wash was determined by using spindle no. 6 at 10 to 100 rpm

4) Determination of foaming power:[26]

Appratus: Beaker and Measuring cylinder.

Procedure: Firstly 5 ml of face wash was taken in a beaker and then add 45ml of water in it. Stir it well before solubilizing the face wash in water, then this solution taken in 500 ml measuring cylinder give 12 shakes to it stand a cylinder for a 2 min then take the reading by using ruler in centimeter.

5) Determination of microbial Testing [50,51,52]

Principle: The disc-diffusion test is based on the fact that for a given antibiotic, the size of the zone of inhibition is inversely related to the MIC (determination by dilution method of the strain being tested when the test conditions are the dilution method) of the strain being tested when the test conditions are held constant. Antimicrobial susceptibility testing with discs is a simple and rapid method and provides a reproducible means of testing bacterial sensitivity to various antibiotics and chemotherapeutic agents.

In- Vivo Studies:

1) Skin irritation:

The skin irritation was carried out on human volunteers. For formulated Face wash, five volunteers were selected and 1.0 gm of formulated cream was applied on an area two square inch to the back of the hand. The voluntrees were observed for lesions of irriation.

Parameter	Skin irritation test
F1	No irritation
F2	No irritation
F3	No irritation

Table no. 4 skin irritation

2] Photographic evaluation:

Photographic evaluation is carried to see the effect of the product visually. In case of determination of cleansing activity photographic evaluation was adopted. In this method the photograph of skin before and after rinsing of skin were taken out and effect of product was determined.

RESULTS AND DISSCUSSION

In-Vitro studies:

1) Determination of physical parameter of face wash

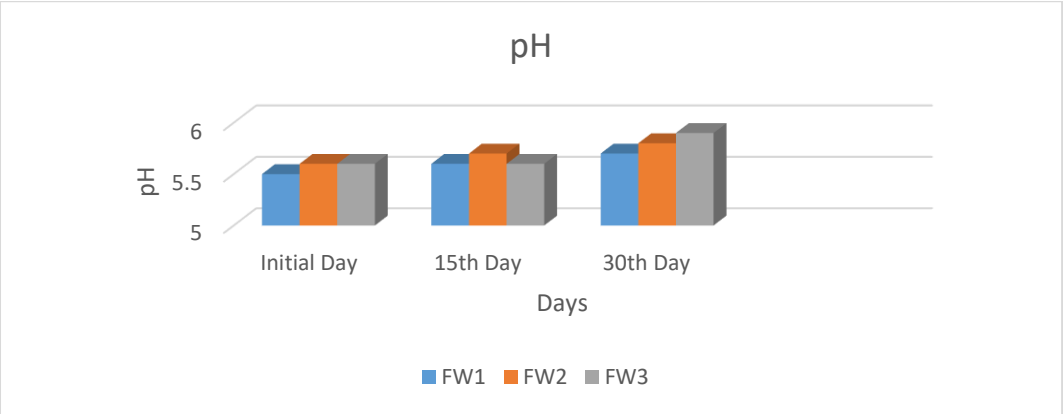
Table no 5: Parameters of face wash

Sr. No.	Parameters	FW1	FW2	FW3
1	Appearance	Clear	Clear	Clear
2	Colour	Colourless	Colourless	Colourless
3	Consistency	Not Good	Good	Good
4	Tacky Feel	No	No	No

2) Determination of pH of Face wash

Table no 6: pH of Face wash

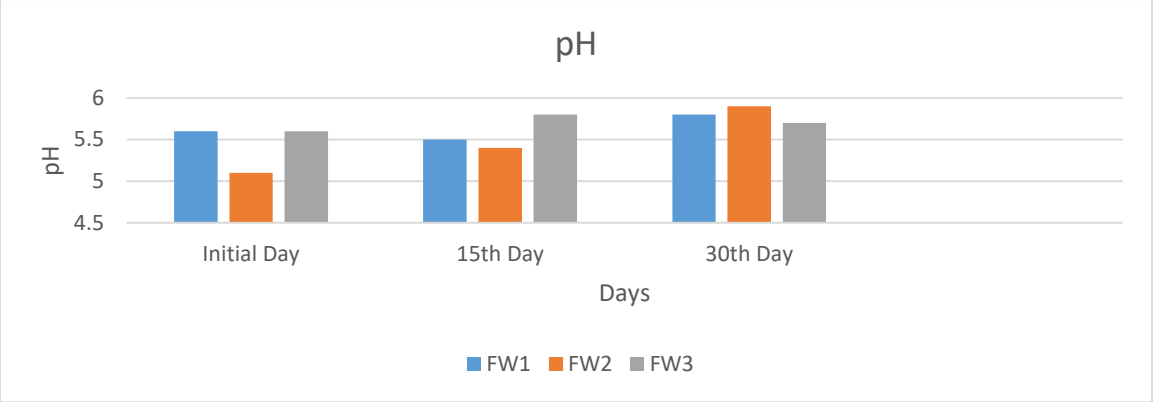
Sr.No.	Time interval	FW1	FW2	FW3
1	Initial Day	5.5	5.6	5.6
2	15thDay	5.6	5.7	5.6
3	30th Day	5.7	5.8	5.9



1) Determination of pH by using Curry leaves and Bael leaves

Table no 7: Determination of pH

Sr. No.	Time interval	FW1	FW2	FW3
1	Initial Day	5.6	5.1	5.6
2	15th Day	5.5	5.4	5.8
3	30th Day	5.8	5.9	5.7



3) Determination of viscosity

Table no 8: Determination of viscosity

The viscosity of face wash determine by using Brookfield Viscometer. The values obtained from the sample noted.

Sr. No.	No. of days	FW1	FW2	FW3
1	Initial Day	7250cp	7200cp	6950cp
2	15th Day	7150cp	7100cp	6800cp
3	30th Day	7120cp	6840cp	6750cp

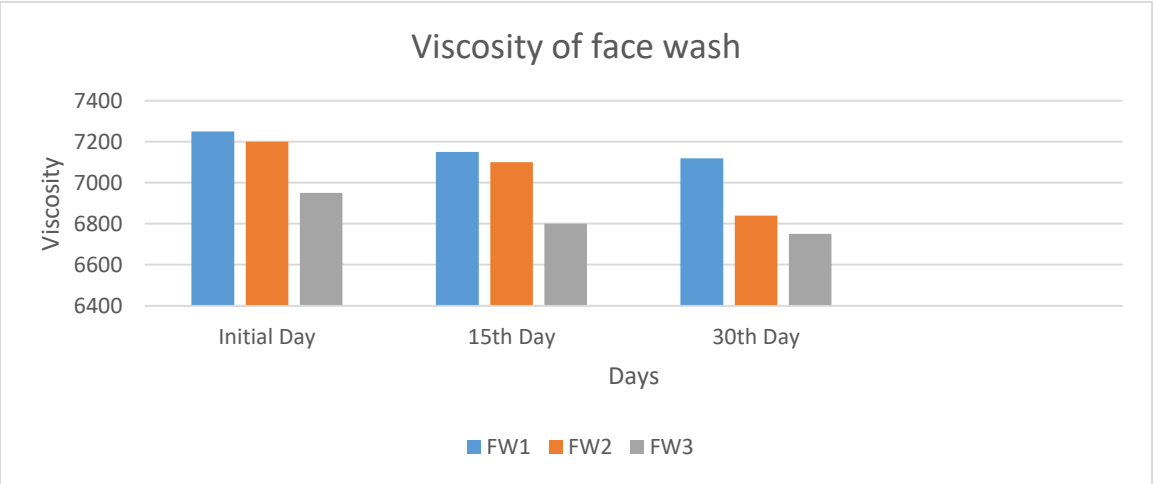
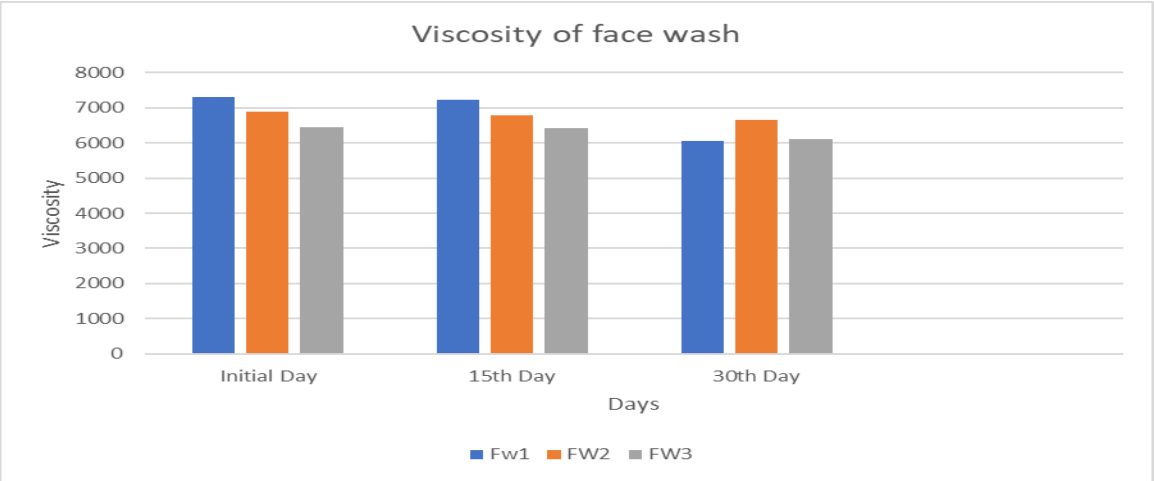


Table no 9: Determination of viscosity of face wash with Bael leaves and Curry leaves

Sr. No.	Time interval	FW1	FW2	FW3
1	**Intial Day**	7312cp	6900cp	6450cp
2	**15th Day**	7220cp	6780cp	6423cp
3	**30th Day**	6059cp	6660cp	6100cp

It was observed that viscosity of formulation was found to be which was good. Therefore formulation passes the test.



4) Determination of Foaming Power

Table no. 10 Determination of Foaming Power

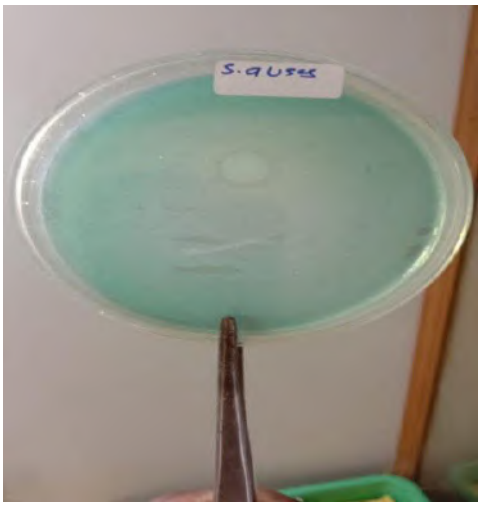
Sr. No	Determination of foaming	Result
1	Foaming	64

Determination of Microbial Testing:[50,51,52]

5) Determination of Microbial Testing

Interpretation of result:

Although there is some correlation between the size of the zone of inhibition and the susceptibility of the organism to the antibiotic, the former is a function of many variables e.g density of the inoculum, depth of the medium, diffusibility of antibiotic etc. The size of the inhibition zone at which the organisum is considered Resistant, Intermediates or sensitive is given in the zone size interpretative chart as a part of this literature.



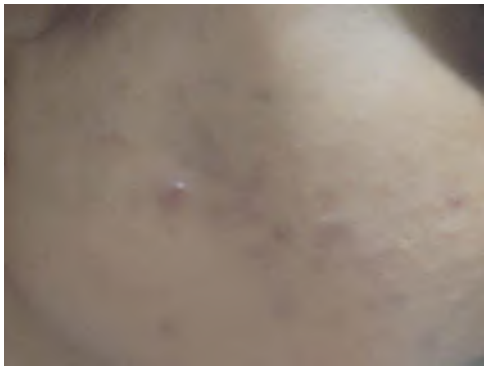
In-Vivo Studies

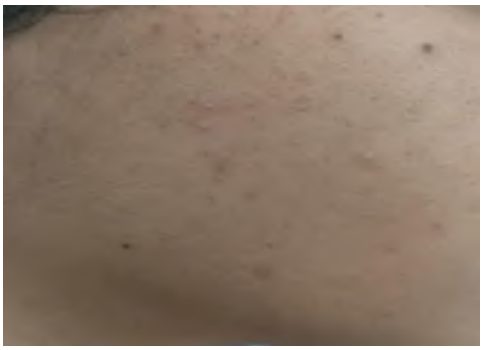
1) Table no. 11 Skin irritation study:

Parameters	Skin irritation
FW1	No irritation
FW2	No irritation
FW3	No irritation

2) Photographic Evaluation

The study of effectiveness of product was done by the help of the volunteer study. This was carried out human volunteers. Face wash were applied on skin. The photograph were taken before and after application of product.





CONCLUSION

At Present because of availability of cosmetic products in market, consumers are giving special attention Towards the selection of cosmetic product to develop a well standard formula; the new product viz. herbal face wash was formulated by incorporating active extract singly and also in combination for good effect.

Anti-acne face wash was selected for sebum regulation activity because anti-acne face wash Contain good quality of extracts which helps to reduce sebum secreation and helps to remove oil and reduce pimple. Face wash prepared on synthetic base containing polymer, surfactant, humectant and preservatives etc. One formulation was selected from prepared base formulation on the basis of physical parameter for futher incorporation. Incorporation of active and sebum regulation property. Different formulation were prepared with varying concentraction of actives i.e anti-acne face wash with curry laves and bael leaves. Evaluation studies like physical parameter, pH, viscosity, stability was done for selecting the final batch. In-Vivo study of final batch was taken. Cleansing activity were determine photographically.

Over the post few year, several methods are developed for an efficient cleansers with profound effect for various applications. There are various types of cleansers available depending on purpose and need. Curry leaves and bael leaves are used to remove acne and clear scars from the skin. Anti-acne face wash is used to remove all the acne from the skin and reduce the scars. The single formulation Shows all the activities like sebum regulation and moisturization. Curry leaves and Bael leaves is the key active ingredient of face wash helps to remove dirt from the skin and acne. So it is concluded that, the formulation of anti-acne face wash give the satisfactory result to the skin.

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Formulation and Development Of Hair Colour Spray

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ABSTRACT
The beauty and personal care industry have witnessed a surge in demand for innovative and convenient hair coloring products. In response to this trend, our research focuses on the formulation and development of a novel hair color spray, aiming to offer consumers a user-friendly and effective solution for temporary hair color enhancement. This research encompasses a thorough exploration of key aspects, including formulation optimization, stability studies, and performance evaluation. The formulation process involves selecting safe and effective colorants, solvents, and additives to achieve a balanced and vibrant hair color spray. We delve into the chemical compatibility of ingredients to ensure stability over time and under various environmental conditions. Additionally, our study emphasizes the incorporation of ingredients that contribute to hair health, such as conditioning agents and UV protectants.
Stability studies play a pivotal role in determining the product's shelf life and maintaining its efficacy. We assess the impact of factors like temperature, light, and air exposure on the formulation, employing analytical techniques to monitor changes in color, texture, and chemical composition. Our findings contribute to the development of a hair color spray that remains reliable and appealing throughout its intended lifespan. Performance evaluation encompasses both laboratory assessments and consumer trials. Objective measurements, such as color intensity, coverage, and ease of application, are conducted to quantify the product's performance. Furthermore, consumer feedback regarding factors like odor, residue, and overall satisfaction aids in refining the formulation to meet diverse preferences and expectations. In conclusion, our research on the formulation and development of a hair color spray provides valuable insights into creating a product that not only delivers vibrant and temporary hair color but also prioritizes stability and user experience. This study contributes to advancing the field of cosmetic science, offering a promising solution for individuals seeking a convenient and customizable hair coloring experience.
KEYWORDS: COFFEE, BLACK TEA, HAIR COLOUR, HAIR CARE.

INTRODUCTION TO HAIR COLOUR SPRAY:

A hair color spray is a form of temporary hair dye, also known as wash-out hair color, that offers a non-damaging, short-term way to color your hair.
Colouring your hair is both Experimental and painstaking task
Experimental, because you don’t know how the hair dye would look on your hair until you get it done on your hair And painstaking coz it is effort and time-consuming. Though, coloring your hair can change your appearance and can transform the way you look. But if you are one of those experimental types, who does not want to stay with one hair color for long, temporary hair color sprays are for you
These hair color sprays are fun, lively and will not make you feel monotonous wearing them for longIt was already mentioned that adolescents and young people are the main target of these hair spray painting, but the use is not exclusive. In fact**,** modern adults, seniors and anyone willing to hide white hair (which do not only appear in elderly people) may also use this product**.**These aerosols present many advantages and the application is exactly the same as in the other dye sprays. The only difference is that you should focus the application on the specific

areas where white hair appears.Furthermore, you should be careful when choosing the proper colour scheme, based on your hair colour, so it is not noticeable. Once applied, you will feel how the colour is distributed uniformly, providing a natural appearance.For the best outcomes, use a comb to separate the hair creating lines, apply the product and distribute this with a comb in order to remove the excess**.** Do not worry if by mistake your front or ears are spotted**.** You just simply use a cotton moistened with soap and clean it up Hair color spray is a temporary hair dye that is used to change the color of hair temporarily. It is a type of spray-on hair color that is typically available in aerosol cans. The color spray is usually applied to dry hair and it can be used to create highlights, cover up roots, or even create an ombre effect.

Handling and correct usage

- As certain gases are integrated in the aerosol product, handling should be taken seriously. Speaking about seriousness, it should be understood that a wrong usage may result from fire to death.
- When an aerosol filler, which contains a gas at environment temperature, experiences a sharp change in temperature, this product will seek the way to expand or to release abruptly, which will consequently cause an explosion.
- Such explosion will cause damage in people, animals or facilities, depending on the magnitude of the incident.For this reason, our company shares some recommendations about the correct usage that should be given to aerosol products in order to avoid any inconveniences.

COFFEE:

Cosmatological importance of Coffee in Hair colour:

Coffee contains compounds like antioxidants, which may have potential benefits for the hair and scalp. Some people believe that coffee can enhance hair color, add shine, and potentially darken hair. However, these effects are generally subtle, and the efficacy of coffee as a hair colorant is not comparable to synthetic hair dyes.If there have been developments in the beauty industry since my last update, it's advisable to check for more recent sources or consult with cosmetologists, dermatologists, or other beauty professionals for the latest information on the cosmetological importance of coffee in hair color sprays.

Coffee has several characteristics that make it an effective ingredient in hair care products:

Caffeine: Coffee contains caffeine, which is a stimulant that can help to promote hair growth. Caffeine also improves blood circulation to the scalp, which helps to deliver essential nutrients to the hair follicles.

Antioxidants: Coffee is rich in antioxidants that protect hair from damage caused by free radicals. Antioxidants also help to reduce hair fall by preventing damage to hair follicles.

Natural Dye: Coffee can be used as a natural dye to enhance the natural color of your hair. It can darken hair and make it more vibrant.

Moisturizing Properties: Coffee also has moisturizing properties that can help to hydrate and soften hair. This can make hair more manageable and reduce frizz.

Material And Methods:

Coffee decoction can be used in a hair spray to provide a range of benefits for the hair. Coffee contains caffeine, which can help to stimulate the scalp and promote healthy hair growth. Additionally, coffee can help to darken the hair and add a subtle reddish tint, making it a useful natural ingredient in hair color sprays.

Ethyl acrylate is a chemical compound commonly used in the manufacturing of adhesives, coatings, and plastics. It is also sometimes used in hair care products, including hair color sprays, as a binding agent to help the product adhere to the hair.

Ethyl alcohol is a common ingredient in many hair care products, including hair color sprays. It is often added to these products as a solvent and a carrier for the other ingredients. Ethyl

alcohol can help to distribute the hair color spray evenly over the hair, and Ethyl alcohol can help to distribute the hair color spray evenly over the hair

EDTA (ethylenediaminetetraacetic acid) can be used in hair color spray formulations as a chelating agent to improve the stability and shelf life of the product

Phosphoric acid can be used in hair color sprays as a pH adjuster to maintain the proper acidity of the product.

Propylene Propylene glycol is commonly used as a solvent and a humectant in hair color sprays, particularly in semi-permanent and temporary hair color formulations.

Phenoxyethanol sometimes used as a preservative in hair color sprays to prevent the growth of harmful bacteria and other microorganisms that could spoil the product or cause infections

DM water, or Deionized water, is commonly used as a solvent and diluent in hair color sprays. It is a highly purified form of water that has had its mineral ions removed, making it ideal for use in cosmetic applications where purity is important.

SR NO	INGREDIENTS	%
1	ETHYL ACRELATE	5
2	ALCOHOL	20
3	EDTA	0.25
4	PHOSPHORIC ACID	1
5	PROPYLENE GLYCOL	0.5
6	PHENOXY ETHANOL	0.2
7	COLOUR BLEND	10
8	DM WATER	15
9	LIQUIFIED PETROLEUM GAS	40

TRAIL A

HERE FROM TRAIL A IT IS OBSERVED THAT THS FILM FORMATION IS NOT PROPER

SR NO	INGREDIENTS	%
1	ETHYL ACRELATE	10
2	ALCOHOL	20
3	EDTA	0.25
4	PHOSPHORIC ACID	0.5
5	PROPYLENE GLYCOL	0.5
6	PHENOXY ETHANOL	0.2
7	COLOUR BLEND	10
8	DM WATER	7
9	LIQUIFIED PETROLEUM GAS	40

TRIAL B

HERE FROM TRIAL B WE OBSERVE THAT THE HAIR COLOR SPRAY HAVE MORE DRYING TIME

TRIAL C

SR.NO	INGREDIENTS	%	USE
1	ETHYL ACRYLATE	10	FIM FORMING POLYMER
2	ALCOHOL	40	DRYING AGENT
3	EDTA	0.25	CHELATING AGENT
4	PHOSPHORIC ACID	0.5	COLOUR DEVELOPER
5	PROPYLENE GYCOL	0.5	VEHICAL/HUMACTANT
6	PHENOXY ETHANOL	0.2	PRESERVATIVE
7	COLOUR BLEND	10	COOURING AGENT
8	D.M WATER	7	SOLUBLIZER
9	LEQUIFIED PETROLIUM GAS	30.00	PROPPELENT /SRAYER
10	ACTIVE	2%	ACTIVE INGREDIENT

From trial C we observe that the feel effect and coverage of the product is satisfactory.

Process :

Step I : Dissolve E.D.T.A 0.25 G In 7 Ml Of D.M Water Folowed By Addition Of Phosphoric Acid 500 Mg Mix Well (Keep Aside)

Step II : Diperse Ethyl Acrylate 10 Kg In Alcohol 20 Ml In Clean And Dry Vessel No 1 Mix Under Stirring

Step III : Mix Propylene Gycol 500 Mg And Phenoxyethanol 200 Mg And Add In Vesel 1 Ii Under Stirring.

Step IV: Now Diperse Colur Blend 10 Gm In 20 Ml Alcohol Mix Well Filter Through 100# Nylone Cloth And Add In Vessel 1 Under Stirring

After Blending Add Active Element.

Step V : Add Filtered Batch 68.45 Ml In Filling Can Of 100ml Capacity Add 3.-4 Small Ball Bearings For Shacking . Seal It With Cap

Step VI : Fill Can With 31.55 Ml Of Lpg Carefully.

Step VII: Cheack Whether Lpg From The Can Is Leaking Or Not .Then Put Proper Lable On It And Use It.

FINAL FORMULATION FOR HAIR COLOUR SPRAY

SR.NO	INGREDIENTS	%	USE
1	ETHYL ACRYLATE	10	FIM FORMING POLYMER
2	ALCOHOL	40	DRYING AGENT
3	EDTA	0.25	CHELATING AGENT
4	PHOSPHORIC ACID	0.5	COLOUR DEVELOPER
5	PROPYLENE GYCOL	0.5	VEHICAL/HUMACTANT
6	PHENOXY ETHANOL	0.2	PRESERVATIVE
7	COLOUR BLEND	10	COOURING AGENT
8	D.M WATER	7	SOLUBLIZER
9	COFFEE DECOCTION	2	ACTIVE INGREDIENT
	LIQUIFIED PETROLIUM GAS	30	PROPELLENT/SPRAYER

QC PARAMETERS

SR.NO	TEST	RESULT
1	PHYSICAL APPEARANCE	SPRAYBLE COLOURED LIQUID
2	PH (10% SOLUTION IN WATER)	5.5-6.5
3	COLOUR	SPECIFIED
4	ADDATION OF POLYMER	STCKING NTO HAIR DOSENOT REMOVED IN NORMAL RINCE (SHOULD BE REMOVED WITH SOAP /SHAMPOO)
5	PARTICALSIZE OF DISPERSION	SHOULD BE PASS THROUGH 100 # FILTER.
6	LEAK TEST AFTER LPG FILLING	PASSES (NO LEAKADGE) (CONTAINER KEPT IN HOTWATER BATCH)
7	SPRAYING AFFICIENCY	IQUID SPRAYED EVENLY WITHOUGHT BLOCKING NOZEL

EVALUATION OF HAIR COLOUR SPRAY:

Evaluations of hair color spray can be done through various methods to determine its effectiveness and safety. Here are some common evaluations for hair color spray:

- Color matching: This evaluation involves comparing the color of the hair color spray to a standardized color chart to ensure that the product produces the desired color.
- Coverage: This evaluation determines the ability of the hair color spray to evenly cover the hair, including any gray or white hair.
- Safety: This evaluation involves testing the hair color spray for any potential harmful chemicals or irritants that may cause skin or scalp reactions.
- pH testing: This test determines the acidity or alkalinity of the product. Hair color sprays usually have a pH between 8-10, which is alkaline to open up the hair cuticle and allow the color to penetrate.

RESULT AND DISSCUSSION

Results and discussion on hair color spray can vary depending on the specific research or analysis being conducted. Here are some potential results and discussions related to hair color spray:

Effectiveness: One potential area of research could be on the effectiveness of hair color spray. Results could show how well the spray covers gray hairs, how long the color lasts, and how easy it is to apply and remove.

Safety: Another potential area of research could focus on the safety of hair color spray. Results could include any potential risks or side effects associated with the product, such as allergic reactions or damage to hair.

Color options: Hair color spray is available in a wide range of colors. Results and discussions could focus on the popularity of different color options and trends in hair color spray.

Application techniques: Results and discussions could also focus on the different application techniques for hair color spray. This could include the use of brushes or combs, how to avoid overspray, and best practices for achieving an even and natural-looking color.

PATCH TEST;

A) Patch Test Result Patch test was performed on sensitive part of skin, e.g. blend of elbow, popliteal space of skin behind ears. The cosmetic was tested by applying to an area of 1 sq.cm of the skin. Central patches were also applied. The site of the patch wasinspected after 24 hours. There were no reactions and then test was repeated once moreon the same side. Since there was no reaction as the person was considered as not hypersensitive and product pass the test.

Sr.no.	Parameter	M1	M2	M3
1	Immidiately after removalof product	N.R.	N.R.	N.R
2	After 24 hrs.	N.R.	N.R.	N.R
3	After 48 hrs.	N.R.	N.R	N.R

Disscussion :

1. In the patch test there is no reaction from the product which is determined that theproduct is safe and compatible for all age group between 18 to 45.

2. The product is transparent and non comedogenic because of that these product is also compatible with acne prone skin also etc.

3. After the discussion of product is also stable in all conditions. That is also provenfrom stability

Cyclic Stability Study

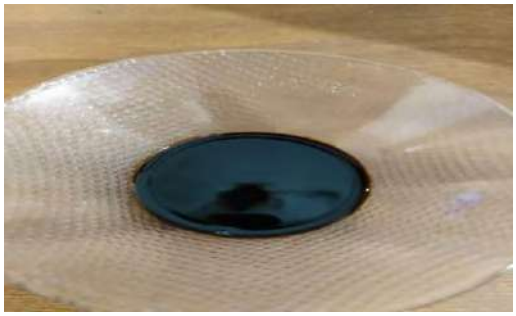
These tests are not carried out at fixed temperature and humidity. In this test,temperature was changed cyclically every day e.g. low-high-low-high to stimulate the changes in temperature daily

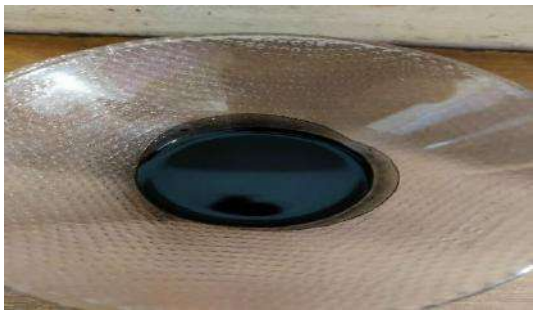
Sr.no	Parameter	F1	F2	F3
1	Freeze temperature	Stable	Stable	Stable
2	Room temperature	Unstable	Unstable	Stable
3	High temperature	Unstable	Unstable	Stable

Ph Test:

Sr.no.	Days	FA1	FA2	FA3	FINAL
1	Initial Day	6.5	6.1	6.6	6.6
2	8 Days	6.4	5.8	6.5	6.5
4	15 Days	6.3	5.6	6.4	6.4
5	30 Days	6.2	5.6	6.3	6.3

COLOUR MATCH:BLACK,BROWN





CONCLUSION:

In conclusion, the formulation of a hair color spray for black and brown hair requires careful consideration of various ingredients and factors. Some important ingredients that may be used in such a formulation include colorants, such as dyes or pigments, as well as solvents, emulsifiers, preservatives, and other functional ingredients like EDTA and phosphoric acid.

It is essential to ensure that the ingredients used in the formulation are safe and effective, and that they work together to create a product that is easy to use, delivers consistent results, and is suitable for the targeted hair types and colors. It is also important to follow good manufacturing practices, such as using sterile equipment and ensuring proper storage conditions, to ensure the safety and stability of the product. Overall, a successful hair color spray formulation for black and brown hair will depend on careful selection of ingredients, expert formulation, and rigorous quality control processes

In conclusion, hair color spray can be a fun and temporary way to experiment with new hair colors without committing to a permanent change. They work best on light or bleached hair and can be messy, so it's important to use them in a well-ventilated area and protect clothing and surfaces. Additionally, hair color sprays can dry out hair and may cause irritation or allergic reactions for those with sensitive skin. As with any hair product, it's important to use hair color sprays safely and cautiously.

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Formulation and Development of Cleansing Spray using Marine Extract

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ABSTRACT:-
This study investigates the efficacy of Chondrus crispus marine extract in formulating a skin cleansing face spray. The research explores the natural cleansing properties of the red seaweed extract, assessing its ability to remove impurities, balance skin pH, and enhance overall skin health. Additionally, the paper discusses the development process, formulation optimization, and sensory attributes of the Chondrus crispus-based face spray. The findings suggest that this marine extract holds promise as a sustainable and effective ingredient for skincare products, providing a refreshing and nourishing cleansing experience.
Keywords: Chondrus Crispus, Cleansing, Skin health, Nourishing, Optimization.

I. INTRODUCTION:-

Chondrus crispus, colloquially known as Irish moss, boasts a rich tradition of use, extending beyond culinary applications to encompass diverse biological activities. With a family classification within the Rhodophyta phylum, this seaweed is recognized for its anti-inflammatory, antioxidant, and immunomodulatory properties. As skincare formulations increasingly gravitate towards sustainable practices, the exploration of marine-derived ingredients gains significance.

Chondrus crispus belongs to the family Rhodophyceae within the phylum Rhodophyta. The Rhodophyta, commonly known as red algae, are characterized by their red pigmentation due to the presence of chlorophyll a and phycobiliproteins. The family Rhodophyceae encompasses a diverse group of red algae, and Chondrus crispus, or Irish moss, is specifically classified within this family. Dry matter (DM) on wet weight (average %) 22%

It belongs to the category-Class: Rhodophyceae

Order: Gigartinales

Hydra I Rich is rich in Vitamins C, B2, B3 and A

a)Hydra I Rich is a blend of Active Molecules from Seaweeds – predominantly containing Chondrus crispus.

b)It acts as Anti-Pollution shield for the skin with added benefits of high hydration

c)It has high concentration of “mannose” – an oligosaccharide studied and established for its effectiveness - protection of cells from cigarette smoke, pesticides and heavy metals along with anti- inflammation action.

Vitamin C : Pollution Shield + Moisture Retention

a) protects the skin from free radicals, from excess exposure to the sun, environmental pollution and regular smoking.

b)Wrinkles will become less prominent, as the Vitamin C increases elastin formation &

c) helps to retain moisture

Vitamin B3 : Natural Niacinamide

a)According to the Journal of Cosmetic Dermatology, an article published in 2004 showed niacinamide helps improve the moisture content in the toplayer of skin.

b)Vitamin B3 also reducestopical inflammation and can help with sun damage.

c)It helps reduce wrinkles, reduce uneven skin tone, help heal acne and reduce hyperpigmentation.
Vitamin A : Skin Maintenance
a)Vitamin A is necessary for the maintenance and repair of skin tissue
b)Medical studies show a reduction in lines and wrinkles, good acne control, and some psoriasis relief, all from using creams containing Vit. A
Vitamin B2 : Protects Healthy Skin and Hair
a)Riboflavin plays a key role in maintaining levels of collagen.
b)Low levels of this vitamin can result in premature aging.
c)Some studies suggest that riboflavin can provide relief from skin inflammation and chapped lips.

Cosmetological Importance of Marine Extract:-

There's a reason why red algae is being incorporated into so many skincare products nowadays. Specifically, it can offer many of the same benefits as traditional cleansers and creams without the side effects associated with chemical-based ingredients. It's a natural ingredient that comes straight from our oceans, making it super effective for a variety of benefits.

Hydrating Properties Anything that exists in the ocean naturally does a pretty good job of retaining moisture, and algae is no different. Some forms of algae have been found to have humectant properties, which is a common ingredient in moisturizers to help your skin lock onto moisture and remain hydrated for longer.This makes red algae great for dry skin, as it can help to rejuvenate your complexion. Additionally, it might be able to help reduce the appearance of fine lines or wrinkles by preserving moisture in your skin to make it look more vibrant.

Antioxidant Effects You've probably heard about how important it is to eat foods that are high in antioxidants. But using skincare products with antioxidant effects is just as necessary. Antioxidants are chemicals that help stop or limit the damage caused by free radicals, contributing to oxidative stress. Oxidative stress can trigger cell damage, and it's one of the primary causes of the signs of skin aging. It's been found that algae is high in antioxidant activity because they are high in nonenzymatic 32 components such as ascorbic acid. This makes it a great natural alternative to chemical preservatives in most other skincare products, as it can nourish your skin and keep it looking healthy.

Protection From the Sun When you think about it, there's no shade in the ocean. So marine organisms such as red algae need a way to protect themselves from the harmful ultraviolet rays of the sun. Luckily, algae have been found to have a UV absorbing capacity that can help shield your skin from harmful rays. Using a cleanser or cream with red algae can help to reduce the harmful effects of photoaging from blue light or UV light exposure. Not to mention, much of the reason for algae's ability to protect from the sun is associated with its polyphenol constituents. Polyphenol is a micronutrient in plants that have been found to have a large number of benefits for other parts of your body.

Shields from Pollution Algae has loads of unique properties that make it stand out from most ingredients in other skincare products. For example, it can repair your skin's natural barrier to mimic its native biome. This gives it the ability to protect your skin from harmful outside pollutants, helping to keep it healthy and younglooking.

II. MATERIAL AND METHODS

Preparation of extract-

The extraction of Chondrus crispus (Irish moss) typically involves several steps to isolate the desired compounds, especially carrageenan. Here is a simplified overview of the extraction process:

1. Harvesting:- Chondrus crispus is usually harvested from marine environments. The seaweed is gathered from rocks or cultivation areas, ensuring sustainable harvesting practices.

2. Cleaning:- The harvested seaweed undergoes thorough cleaning to remove any impurities such as sand, shells, or other debris. This step is crucial to obtain a high-quality extract.
3. Drying:- After cleaning, the seaweed is often dried to reduce its water content. This can be done through sun-drying or using specialized drying equipment. Drying helps in preserving the seaweed for storage and further processing.
4. Milling or Grinding:- The dried seaweed is then milled or ground into a coarse powder. This increases the surface area, facilitating the extraction of carrageenan during subsequent steps.
5. Extraction:- The milled seaweed undergoes an extraction process to isolate carrageenan. Typically, this involves soaking the seaweed in an alkaline solution to remove cell wall components, followed by precipitation and separation steps to isolate carrageenan from the liquid.
6. Purification:-The extracted carrageenan may undergo further purification steps to remove any remaining impurities, such as colorants, proteins, or minerals. This purification process ensures a more refined and standardized extract.
7. Drying the Extract:- The purified carrageenan extract is then dried to obtain a powder or gel-like substance. This final form makes it easier to incorporate into various cosmetic and skincare formulations.
It's important to note that the specific extraction methods may vary among manufacturers, and some may use additional steps or variations in the process. The goal is to obtain a carrageenan-rich extract with the desired properties for use in cosmetic and skincare applications. Additionally, adherence to sustainable harvesting practices is crucial to preserve marine ecosystems and ensure a long-term supply of Chondrus crispus.

III.Face cleansing Spray Formulation

Weigh Water, Disodium EDTA, Allantoin, Glycerine, D-Panthenol, Coco Apple Amino Acid, Coco glucoside in one glass beaker and heat it till 75-80°C. Stir it slowly when it cools down to room temperature add Preservative, Marine extract (hydraI Rich), Fragrance and tween 20 need to be premixed for clear solution lastly citric acid is used for buffering.

Sr.No.	Ingredients	Quantity
A	Water	88.3
	Disodium EDTA	0.1
	Allantoin	0.2
	Glycerine	3
	D-Panthenol	0.2
	Coco Apple Amino Acid	2
	Coco Glucoside	1
B	Phenoxyethanol and ethylhexylglycerine	0.5
	Fragrance	0.5
	Tween 20	1
C	Hydra I Rich (Marine Extract)	3

Table No.1- Formula of Face Cleansing Spray using Marine Extract

Evaluation of face cleansing spray:-
Evaluation of face cleansing spray was following.
Physical Evaluation Formulated face cleansing spray was further Evaluated by using the following physical parameter physical parameter clarity, odour, appearance,feel and state of the formulation.
Clarity: The clarity of the face cleansing spray was observed by visual examination. The result was shows in table 2.

Odour: The odour of cleansing spray was found to be pleasant
Feel: The feel of the cleansing spray was observed after using it as a patch test manually after feel was very smooth and hydrated.
Ph: Take 1 ml of sample and dissolve in 100ml of water in beaker that is 1%
solution is prepared. Then with the help of pH meter reading were taken.Results are shown in table 2
Non- irritancy test: Face cleansing spray formulation was evaluated for the non-irritancy test. Preparation shown no redness and irritancy. Observation of the state was done for 24 h 28. Results are shown in table 2

Table no.2- Physical Parameters

Sr.No.	Parameter	Results
1	Clarity	Clear Transparent
2	Odour	Pleasant
3	After feel	Clean and hydrated
4	pH	5.6
5	Non Irritancy	Not irritant
6	Viscosity	1.04 cp

IV.Analysis of Moisture Content using Corneometer:

The moisturizing activity was carried out by using coreometer. It was observed that before application of Cleansing Spray, the moisture content of skin was less and after application of Cleansing spray moisture content was increased.

Sr.No.	Time Interval	% **of moisture content**
1	Before Application	68.4
2	After Application	87.4

V.Result

The present research was the formulation and development of Face cleansing spray using marine extract. The evaluation parameters were coming under results, like the physical evaluation of cleansing spray , PH ,Clarity,Odour, non-irritancy test, viscosity and of the Cleansing Spray was shown in table.

VI.DISCUSSION

The product is a very handy and anywhere use product. A couple of sprays and simply wipe off with a hankey/tissue. The cleanser removes all the dirt and oil from the face and makes the face look absolutely clean. And all of this without a drop of water. Hydra I Rich (Marine Extract) is rich in Vitamins C, B2, B3 and A Hydra I Rich is a blend of Active Molecules from Seaweeds – predominantly containing Chondrus crispus. It acts as Anti-Pollution shield for the skin with added benefits of high hydration.

VII. Conclusion

The formulation of Cleansing spray using marine extract was so formulated for instant cleansing and provide hydration.The tests Carried out simply manually and by

observation Ph,Viscosity, Clarity,Odour, After feel were all noted down and mentioned in table 2.

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Formulation and Evaluation of Antiacne Cream by using Cinnamon oil

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Abstract
Annoying skin problem 'acne' is often related with the microbial infection and requires antimicrobial agents for the treatment. A growing trend in Cosmetic market is towards natural products containing essential oils as antimicrobial agents. It is reported that Cinnamon oil shows antimicrobial activity against microorganisms responsible for acne such as *Propionibacterium acnes*, *Staphylococcus epidermidis*, *Staphylococcus aureus*, and *Candida albicans*. Hence the present work was undertaken with the aim to formulate and develop Antiacne cream by using Cinnamon oil. The Cinnamon essential oil was extracted by steam distillation method and cream formulations were developed with three different concentrations of Cinnamon oil; all the formulations were evaluated as per Bureau of Indian Standards (BIS) guidelines and for antimicrobial activity against the microorganisms responsible for acne by agar well diffusion technique. Also, all Antiacne cream formulations were subjected to stability studies and subjective evaluation on panel of human volunteers. The results showed that, Antiacne cream (C-12) containing Cinnamon oil is effective against microorganisms responsible for acne. Study results indicated that Cinnamon oil is effective Antiacne agent hence can prove to be beneficial for adding in Antiacne products.

Keywords: - Cinnamon oil, Antiacne cream, Formulations, Evaluation, Antimicrobial, Acne

1. Introduction

Acne is a common skin problem, characterized by the formation of comedones, papules, pustules, inflamed nodules, superficial pus, filed cysts and in extreme cases canalizing and deep scaring [1]. The bacteria *Propionibacterium acnes*, *Staphylococcus epidermidis* [2], *Staphylococcus aureus* [3], the fungus *Candida albcans* are almost commonly present in the pustular contents of the acne [4]. Acne skin problem is often related with the microbial infection and requires antimicrobial agents for the treatment. Normally synthetic materials are used because of low cost and strong antimicrobial property but these may cause adverse effect [5]; also, faith of consumer on herbal products is growing fast. In recent years, a large number of essential oils and their constituents have been investigated for their antimicrobial properties [6,7]. It is reported that essential oils provide a gentle and inexpensive way of treating acne infections and healing acne scarring [8]. Cinnamon consists of the dried inner bark (Fig. 1) of the shoots of coppiced trees of *Cinnamomum zeylanicum* Nees, (Synonym- *Cinnamomum verum* J.S.Presl) belonging to family Lauraceae [9] (Fig. 2).

Fig. 1 – Cinnamon Bark

Fig. 2 Cinnamon Tree

Cinnamon bark contains about 0.5 to 1.0 % of volatile oil, 1.2% of tannins, and sweet substance mannitol. The volatile (essential) oil is the active constituent of the drug. Cinnamon bark oil contains 60-70% of cinnamaldeyhde, 5-10% eugenol, other major constituents include sesquiterpenoids (4-5%) such as α-humulene, β-caryophyllene and limonene. Oil also contains eugenol acetate, methyl eugenol, cinnamyl acetate, cinnamyl alcohol, benzaldehyde, cuminaldehyde and benzyl benzoate. Cinnamon bark oil is also reported to contain monoterpenes (for example, linalool, pinene, phellandrene, and cymene), carophyllene and safrole [10]. It is widely used as spice and has been used as flavours, carminative, stomachic, tonic, counter irritants in pharmaceutical and cosmetic preparations, including liniments, suntan lotions, mouthwashes and in toothpastes. In European phytomedicine, Cinnamon bark (2.0-4.0 g daily) or essential oil (0.05-0.2g daily) are used in tea and other galenicals as antibacterial and carminative remedies [11]. It is reported that Cinnamon bark oil can inhibit the growth of human pathogenic fungi and bacteria [12]. It is also reported to have antiseptic, aromatic, astringent, deodorant, anti-inflammatory properties and helps in restoring normal skin colour on the face [13]. Cinnamon oil is reported to have antimicrobial activity against acne causing microorganisms such as *Propionibacterium acnes*, *Staphylococcus epidermidis*, *Staphylococcus aureus*, and *Candida albicans* [14]. Despite these properties, Cinnamon is not popular in cosmetics, may be because of insufficient scientific proof to support the claims made about the various properties of Cinnamon oil in cosmetic preparations. Hence, the present work is aimed at scientific evaluation of the Cinnamon oil as Antiacne agent to treat acne by incorporating it in cream formulations and evaluating the Antiacne cream formulations.

2. Materials and Methods

Collection and authentication of Cinnamon: -Dried inner bark of Cinnamon, were collected from the local market of Nagpur, M.S., India and authenticated (with number 9864) (Fig. 3) from the P.G. Dept. of Botany, RTMNU, Nagpur.

Fig. 3 Authenticated Herbarium Sheet of Cinnamon

Extraction of Cinnamon oil [15]:- Dried inner bark of Cinnamon, was subjected to steam distillation for obtaining Cinnamon oil. It was done by the distillation of 100g of Cinnamon with water (300 ml) by using Clevenger's apparatus [16]. Distillation was continued for 5 hours and oil thus obtained was stored in a refrigerator at 4^0C until use. Extractive value is shown in (Table 4) in Result section.

Validation of Cinnamon oil:- To validate the purity of extracted Cinnamon oil, qualitative analysis of the extracted Cinnamon oil was carried out for determination of organoleptic properties (i.e. colour, odour, and taste) [17], specific gravity[18], refractive index[19], optical rotation[20] and results were compared with the standard values. The results are shown in (Table 5) in Result section.

Formulation of Antiacne Cream Using Cinnamon Oil [21]

Antiacne cream base was formulated by using formulation given in (Table 1).

S. N.	Ingredients	Use	Trial I (Quantity in %)	Trial II (Quantity in %)	Trial III (C-2) (Quantity in %)
	Phase A				
1	Stearic Acid	Emulsifier	12	9	9
2	Cetyl Alcohol	Emulsion stabilizer	2	1	1
3	Bees wax	Emollient	3	-	-
4	Glyceryl Mono Stearate	Self-emulsifier and emollient	-	2	1
5	Mineral oil	Occlusive film former	3	1	1
6	Propyl Paraben	Preservative for oil phase	0.15	0.15	0.15
	Phase B				
7	Tri-ethanolamine	Emulsifier	1.5	0.5	0.5
8	Methyl Paraben	Preservative for water phase	0.15	0.15	0.15
9	Water	vehicle	Upto 100 (78.2)	Upto 100 (86.2)	Upto 100 (87.2)

Table No. 1: Formulation of Antiacne cream base

Since, formulation Trial III (C-2) gave a satisfactory cream base; it was selected as a suitable Antiacne cream base for incorporation of Cinnamon oil. Three different concentrations of Cinnamon oil (i.e. 1%, 0.75% and 0.5%) were incorporated in Cream base (C-2) to formulate three Antiacne creams (C-12, C-13, C-14) respectively. (Table No. 2)

Ingredients	Cream Formulation (Quantity in %)		
	C-12	C-13	C-14
Cream Base C-2	99	99.25	99.50
Cinnamon oil	1	0.75	0.5
Observations: Colour - Bright White – BW	BW	BW	BW
pH	6.47	6.03	6.41
Consistency - Satisfactory - (S)	S	S	S

Table No. 2: Formulation of Antiacne cream with Cinnamon oil

Evaluation of Antiacne creams as per Bureau of Indian Standards (BIS) Guideline [22]:

Antiacne cream base (C-2) and all the Antiacne cream formulations (C-12, C-13, C-14) were

evaluated as per BIS Guidelines for the various parameters like determination of thermal stability, pH, Total Fatty Substance content-% by mass, Total residue-% by mass, Microbial content limit and results were compared with standard and are summarized Result section in (Table 6).

Stability study of Antiacne creams [23]: -The cream base C-2 and Antiacne creams (C-12, C-13, C-14) were subjected to stability studies. Changes in parameters like Colour, Odour, pH, Viscosity, Particle size at three temperatures i.e. in oven at (45 ± 2^0C), in refrigerator at (4 ± 2^0C) and at room temperature were recorded for 45 days, at the interval of 4 days for colour, odour, pH and at the interval of 6 days for viscosity and particle size. Results indicated stable products. (Result section -Graph No. 1 to 12).

Evaluation of antimicrobial activity of Antiacne creams: - The method based on zones of inhibition – agar well diffusion technique is used in present study [24]. Antiacne creams C-12, C-13, C-14 and Antiacne cream base C-2 were evaluated for their antimicrobial activity against pure cultures of *Pseudomonas aeruginosa* (MTCC 1688), *Escherichia coli* (MTCC 1687), *Staphylococcus aureus* (MTCC737), *Candida albicans* (MTCC 227), *Staphylococcus epidermidis* (MTCC 6810) and *Propionibacterium acnes (MTCC 1951)* which were procured from Institute of Microbial Technology, Chandigarh, India.

Cultivation and Maintenance of Microorganisms [25]: -All the microorganisms selected for study were cultivated-maintained on suitable growth medium and conditions as recommended by Institute of Microbial Technology, Chandigarh. The details of conditions for cultivation and maintenance of microorganisms are summarized in Table no. 3. Cultures of these microorganisms were used to evaluate antimicrobial activity of Antiacne creams C-12, C-13, C-14 and Antiacne cream base C-2. Evaluation of antimicrobial activity was based on the measurement of diameter of zone of inhibition in millimetre. The results are given in Table No. 7, Fig. 4 in Result section.

S. N.	Micro organisms	Growth medium used for cultivation and maintenance of stock culture (as recommended by Institute of Microbial Technology, Chandigarh, India)	Medium used for Screening antimicrobial activity	Incubation Period	Incubation Temperature	Growth condition
1.	*P. aeruginosa* (MTCC 1688)	Medium No. 3 (Nutrient Agar Medium)	Muller Hinton Agar Medium	24 Hours	37^0C	Aerobic
2.	*E. coli* (MTCC 1687)	Medium No. 3 (Nutrient Agar Medium)	Muller Hinton Agar Medium	24 Hours	37^0C	Aerobic
3.	*S. aureus* (MTCC 737)	Medium No. 3 (Nutrient Agar Medium)	Muller Hinton Agar Medium	24 Hours	37^0C	Aerobic
4.	*C. albicans* (MTCC 227)	Medium No. 6 (Malt Yeast Agar)	Medium No. 6 (Malt Yeast Agar)	48 Hours	25^0C	Aerobic

5.	*S. epidermidis* (MTCC 6810)	Medium No. 3 (Nutrient Agar Medium)	Muller Hinton Agar Medium	24 Hours	37^0C	Aerobic
6.	*P. acnes* (MTCC 1951)	Medium No. 41 (Blood Agar Medium)	Medium No. 41 (Blood Agar Medium)	48 Hours	37^oC	Anaerobic

Table No. 3: Conditions for cultivation and maintenance of microorganisms under study

Subjective evaluation [26] of Antiacne creams containing Cinnamon oil: - Antiacne cream (C-12) showed maximum antimicrobial activity and hence was selected for subjective evaluation on panels of 20 human volunteers (age group: 18-35 years) having acne skin condition for the time period of 28 days. Subjective evaluation study was carried out by instructing volunteers to use the C-12 cream on face twice daily so that the cream should remain on face for 3-4 hours daily. The volunteers were instructed not to take any other treatment for acne and also to wash the face before applying Cream C-12. The observations were made before starting of the study and after using the Antiacne cream i.e. at the end of 28 days. Assessment of the Antiacne cream was determined based on the functional parameters like Appearance of cream, ease of spreadability, Antiacne efficacy, Improvement in texture of skin, irritancy on application. The self-assessment questionnaires were filled by the volunteers and results were analysed.

3. Results and Discussion

Acne skin disease requires antimicrobial agents for its treatment. Generally, microorganisms associated with acne skin conditions are found to be *Propionibacterium acnes*, *Staphylococcus aureus*, *Staphylococcus epidermidis, and Candida albicans*. It is reported that Cinnamon oil possesses antibacterial, antiseptic, astringent, anti-inflammatory properties and helps in restoring normal skin colour on the face [13]. Hence the present study was undertaken with the aim to formulate and develop Antiacne cream by using Cinnamon oil and evaluation of Antiacne property of cream formulations against microorganisms responsible for acne i.e. against *Propionibacterium acnes, Staphylococcus aureus, Staphylococcus epidermidis, Candida albicans* and also antibacterial activity of the Antiacne creams was evaluated against two common bacteria i.e. *Pseudomonas aeruginosa* and *Escherichia coli*. In the present study, the dried inner bark of Cinnamon was subjected to extraction by steam distillation with Clevenger's apparatus and the percent extractive value of oil (Table 4) was within the range of standard extractive value [27].

S. N.	Name of Oil	Wt. of Crude Drug	Wt. of Oil Obtained	% Extractive value	Standard Extractive Value
1.	Cinnamon oil	100 gm	0.5159 gm	0.5159%	0.5-1% [27]

Table No.4: Percent extractive value of Cinnamon oil

To validate the purity of extracted Cinnamon oil, it was studied for organoleptic properties (i.e. colour, odour, and taste), specific gravity, refractive index and optical rotation. The results were compared with the Standard values [27] which showed that the extracted oil is of standard quality. The results are recorded in the Table No. 5

S. N.	Parameter for validation of Cinnamon oil	Standards [27]	Laboratory Extracted Cinnamon Oil
1.	Color	Light yellow	Light yellow
2.	Odor	Characteristic aromatic, spicy	Characteristic aromatic, spicy
3.	Taste	Characteristic, aromatic spicy	Characteristic, aromatic spicy

4.	Specific Gravity at 25^0 C	1.00 to 1.030 wt./ml	1.01 wt./ml
5.	Refractive Index at 20^0 C	1.562 – 1.582	1.570
6.	Optical Rotation at 20^0C	0^0 to -2^0	$-2^011'$

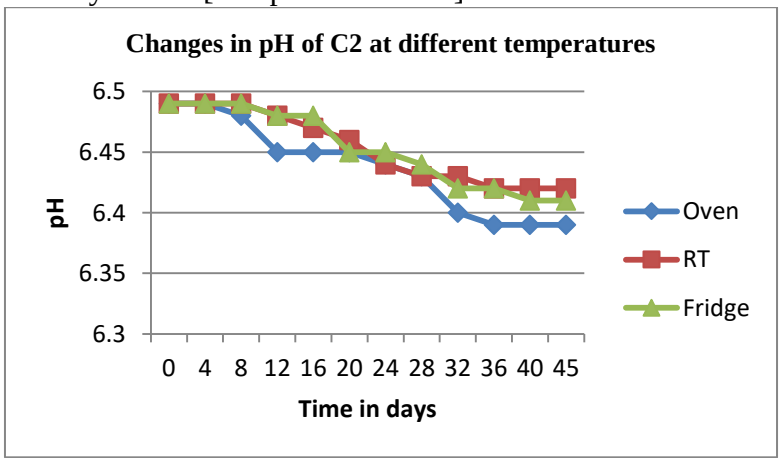
Table No. 5 Validation of Cinnamon oil

Evaluation of Antiacne creams C-12, C-13, C-14 and Antiacne cream base C-2 as per BIS Guideline showed that all creams passes the tests and standards as per BIS Guidelines. (Table No. 6)

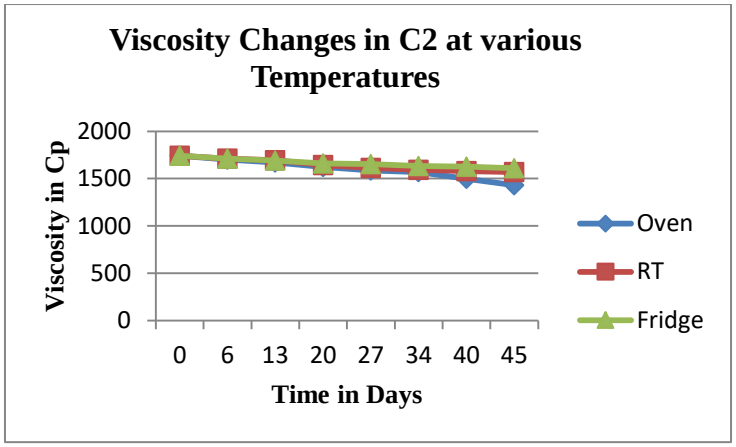
S. N.	Test	BIS Requirement [22]	Observations for C-2	Observations for C-12	Observations for C-13	Observations for C-14	Inference
1	Thermal Stability	No oil separation	No oil separation	No oil separation	No oil separation	No oil separation	Passes test.
2	pH	4.0 to 9.0	6.49	6.47	6.03	6.41	Passes test.
3	Total Fatty Substance content, % by mass	Min. 5%	10.86	11.15	10.64	10.74	Passes test.
4	Total residue, % by mass	Min. 10%	13.21	12.98	13.29	13.17	Passes test.
5	Microbial content limit	Not more than 1000 cfu/g	130 cfu/g	50 cfu/g	60 cfu/g	80 cfu/g	Passes test.

Table No. 6: Evaluation of Antiacne cream bases C-2 and Antiacne Formulations (C-12, C-13, C-14) as per BIS Guidelines

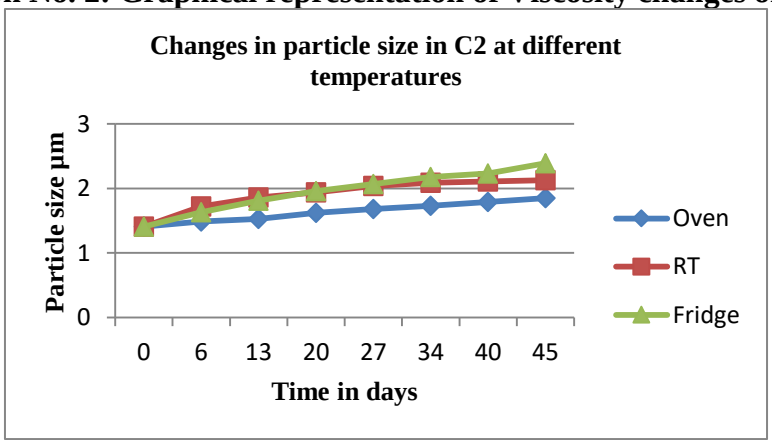
Stability of Antiacne creams was determined by studying changes in parameters like colour, odour, pH, viscosity and particle size under extreme conditions and all Antiacne creams were found to be substantially stable. [Graph No. 1 to 12].



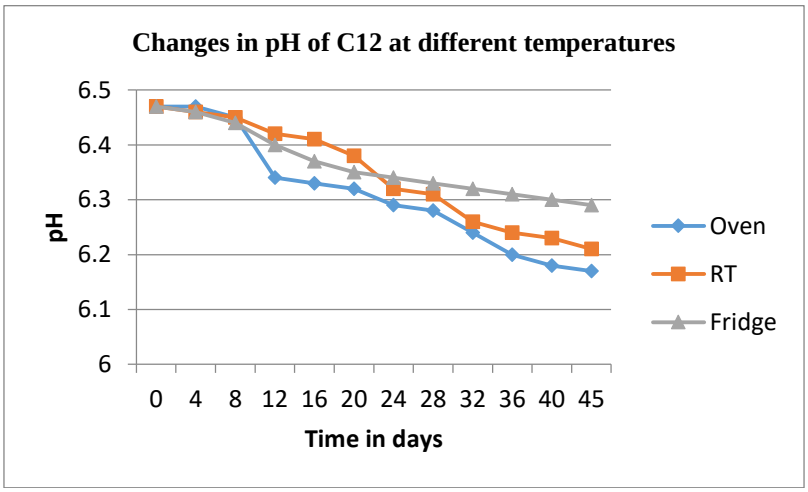
Graph No. 1: Graphical representation of changes in pH of C-2



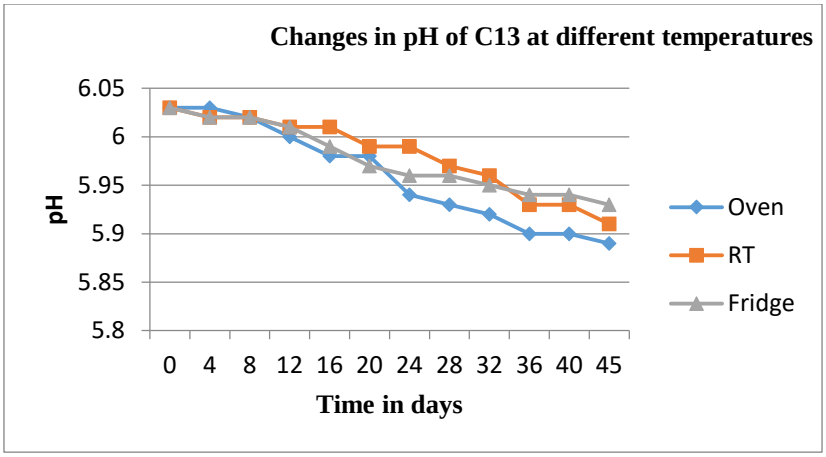
Graph No. 2: Graphical representation of Viscosity changes of C-2



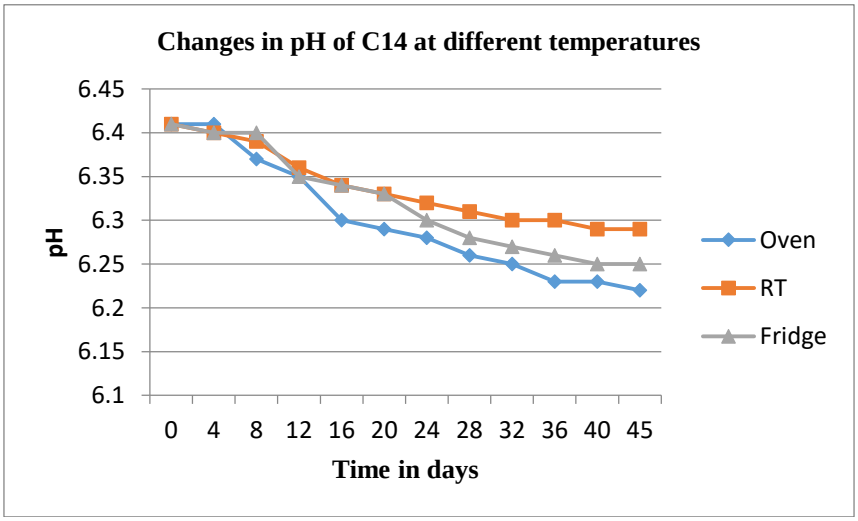
Graph No. 3: Graphical representation of changes in particle size of C-2



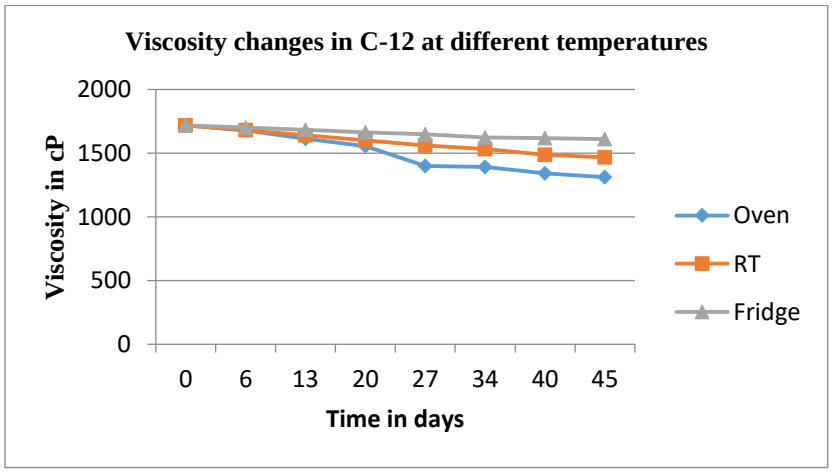
Graph No. 4: Graphical representation of changes in pH of C-12



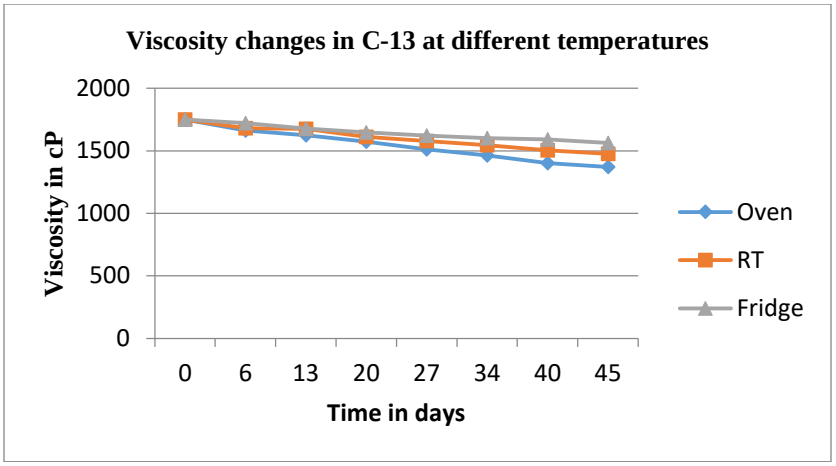
Graph No. 5: Graphical representation of changes in pH of C-13



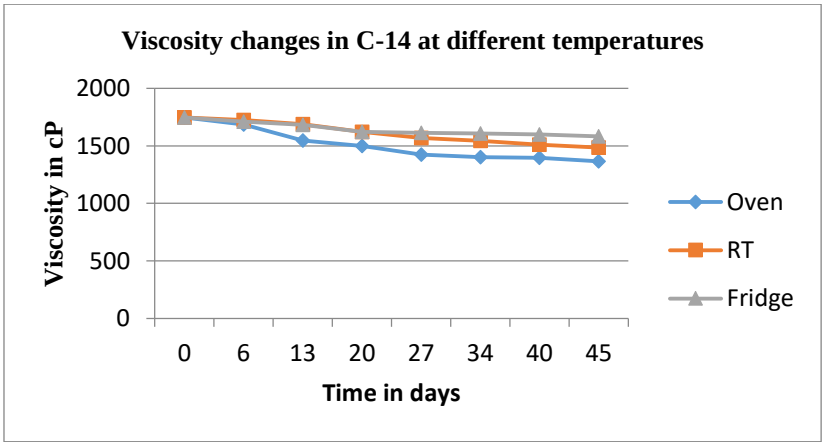
Graph No. 6: Graphical representation of changes in pH of C-14



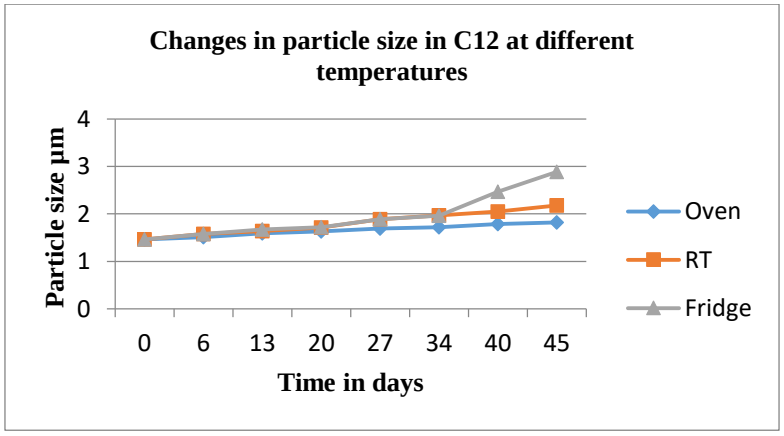
Graph No. 7: Graphical representation of Viscosity changes in C-12



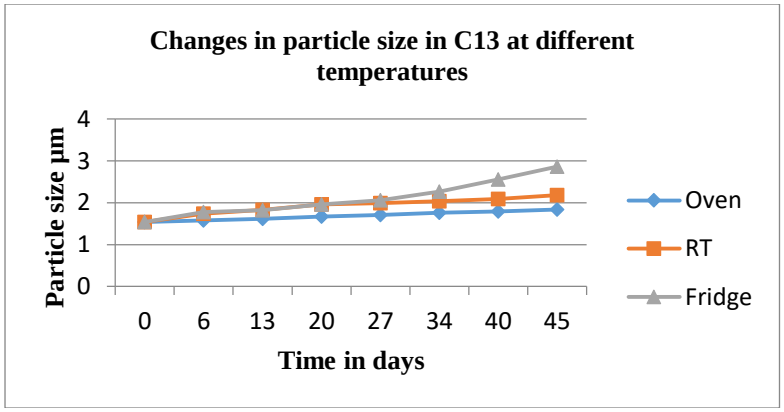
Graph No. 8: Graphical representation of Viscosity changes in C-13



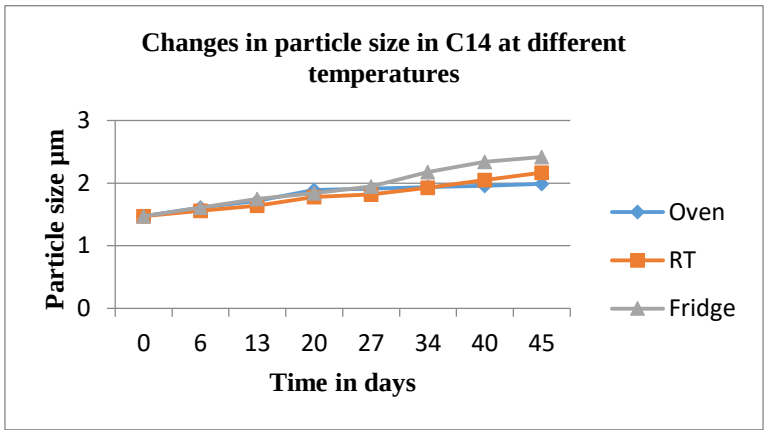
Graph No. 9: Graphical representation of Viscosity changes in C-14



Graph No. 10: Graphical representation of particle size changes in C-12



Graph No. 11: Graphical representation of particle size changes in C-13



Graph No. 12: Graphical representation of particle size changes in C-14

Evaluation of antimicrobial activity of Antiacne creams

The Antiacne cream base C-2 and Antiacne creams C-12, C-13, C-14 were subjected to agar-well diffusion technique to evaluate their antimicrobial activity; the zones of inhibition were measured and recorded [Table No. 3, Fig. 4].

Microorganisms	**Antiacne cream base**	**Antiacne creams containing Cinnamon oil**		
	C-2	**C-12**	**C-13**	**C-14**
P. aeruginosa	-	16	15	14
S. aureus	-	15	13	11
S. epidermidis	-	14	12	11
E. coli	-	14	13	11
C. albicans	-	17	15	14
P. acnes	-	15	14	13

Table No. 7: Zone of Inhibition of Antiacne cream base C-2 and Antiacne creams C-12, C-13, C-14 against acne causing microorganisms

Diameter of Cork borer used – 8 mm, '-' indicates no zone of inhibition, All zones of inhibition in mm

Fig. No. 4: Zone of inhibition of Antiacne creams (C12, C13, C14) containing Cinnamon oil against *S. aureus, S. epidermidis, C. albicans, P. acnes*

Antiacne cream base C2 was non inhibitory to the growth of experimental microorganisms. All the Antiacne creams containing Cinnamon oil (C-12, C-13, C-14) showed inhibitory activity against *P. aeruginosa, S. aureus, S. epidermidis, E. coli, C. albicans, P. acnes.*

From the evaluation of antimicrobial activity of Antiacne creams, it was observed that Antiacne cream [C-12(containing Cinnamon oil 1%)] showed maximum antimicrobial activity against microorganisms under study hence was selected for subjective evaluation on panels of human volunteers. This study was designed to find out the Antiacne efficacy of C-12 cream in terms of reduction in acne condition after application of the C-12 cream. The Antiacne cream was evaluated for functional parameters like, appearance, ease of spreadability, Antiacne efficacy, improvement in texture of skin and irritancy by panel of human volunteers. The results of subjective evaluation indicated that, Antiacne cream C-12 was well appreciated by volunteers for its appearance, ease of spreadability and non-irritancy. Also, C-12 showed remarkable reduction in acne condition and significant improvement in texture of skin. Interestingly 15% of volunteers specifically mentioned that cream C-12 helped them to restore skin colour.

4. Conclusion

India has a rich heritage of many medicinal plants which are used from ancient times for skin and hair care. Acne skin problem is often related with the microbial infections and many other causes also. Herbal remedies are more in demand. Hence, this study was undertaken with the aim to formulate, develop and evaluate Antiacne cream by using three different concentrations of Cinnamon oil. From results of the present study it can be concluded that Antiacne cream containing 1% Cinnamon oil (C-12) was acceptable in view of reduction in acne problem and contains all good characters of skin cream. It can be concluded that Cinnamon oil is effective Antiacne agent. The optimum concentration of Cinnamon oil to be used as Antiacne agent can be 1% for Antiacne cream formulations.

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15

Advance Formulation & Development of Scrub Face Wash With Active Kojic Acid

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ABSTRACT :
The objective of this work is to formulate and evaluate a cosmetic Instant Whitening Face Wash by using natural ingredient. Since the ancient times, there has been awareness among people regarding the use of plants for the essential needs of a healthy and beautiful skin. Scrub is a cosmetic product that contains slightly rough material that can remove dead skin cells. Natural ingredients such as Halimeda macroloba is potential to be used as scrub.

Keywords :- Subject Study, Facial scrub, Face Wash, Kojic Acid , Ph , Visvcosity

INTRODUCTION

Face wash is the products which are used to cleanse face without drying it out. Face wash is very helpful in removing dirt, oil and provide moisture to the skin. Face Wash are used to get rid from dirt, oil, pollution etc. A cleanser dissolves away excess oil makeup and grime from your face. These are oil soluble impurities. Facial skin is the delicate and ordinary soaps can cause it to lose moisture. The purpose of face wash may be to impart cleansing, anti-acne property and moisturizing effect to the skin. And it is commonly called as cleansers.

ACTIVE :

KOJIC ACID

Kojic Acid is a chelation agent produced by several species of fungi, especially *Aspergillus oryzae*, which has the Japanese common name *koji*.[2][3][4] Kojic acid is a by-product in the fermentation process of malting rice, for use in the manufacturing of sake, the Japanese rice wine.[2] It is a mild inhibitor of the formation of pigment in plant and animal tissues, and is used in food and cosmetics to preserve or change colors of substances.[5] It forms a bright red complex with ferric ions.

Classification

INCI Name : Kojic Dipalmitate
Chemical Name / Synonyms : 2-palmitoyloxymethyl-5-palmitoyloxy- γ- pyrone; Hexadecanoic acid, 4-oxo-6-[[(1-oxohexadecyl)oxy]methyl]-4H-pyran-3-yl ester; KAD
Trade Name : MC-KAD
CAS No. : 79725-98-7
Molecular Formula : C38H66O6
Molecular Weight : 618.9

Cosmetic Uses:

Kojic acid's properties allow it to be a bleaching agent when used in creams, gels, and other cosmetics. Kojic acid is similar to a chemical called hydroquinone. They are both effective treatments for hyperpigmentation. Treatment with kojic acid isn't immediate.

The science behind how kojic acid works as a lightening agent involves its effect on melanin production.

Skin Benefits:

Unclogs skin pores. One of the most obvious benefits of using a face scrub is that it cleanses and detoxifies the skin pores , Tackles uneven skin tone , Combats signs of aging, Improves texture, Allows better penetration, Fights ingrown hair, Makes skin soft and supple.

Scrub :

A face scrub is a skincare product used to exfoliate your skin. It helps in the removal of dead skin cells from the surface of your skin, reducing the chances for clogged pores and acne breakouts. With the particle of scrubing and exfoliation dating back to ancient times, history indicates that people used something abrasive to exfoliate their skin.

For example, the American Indians used dried corn cobs for skin exfoliation. Crushed sea shells were also a popular option. Nowdays, scrubs are made with ingredients like poppy seeds , sugar, finely ground sea salt, coffee grounds, cinnamon, honey oats, etc.

Benefits Of Using A Face Scrub

1. Removes Dead Skin Cells
2. Unclogs Skin Pore
3. Removes Flakes
4. Reduces Acne Scars
5. Prevents Ingrown Hair
6. Provides Smoother Skin
7. Improves The Texture Of Skin
8. Better Absorption Of Skincare Products

MATERIALS AND METHODS

Materials (Ingredients) :

1. Sodium lauryl ether sulphate (SLES)
2. Cocoamidopropyl Betain (CAPB)
3. Sodium Chloride
4. EDTA Diasodium
5. Glycerine
6. Distilled Water
7. Preservative
8. Tulsi Extract
9. Kojic Acid
10. Poppy Seeds
11. Aleo Vera Gel
12. Lemon Extract

List Of Equipment :

1. pH meter
2. Brook field Viscometer
3. Beaker
4. Weighing Balance
5. Stirrer

Method:

Preparation of base formulation:

In any cosmetic preparation it is necessary to have stable formulation before Incorporation of active. Preparation of base formulation is important before incorporation of active ingredient, to prepare a stable cosmetic formulation. The Effectiveness and stability of product was depending upon the compatibility of active ingredients

Sr. No.	Ingredients	F1 For 100%	F2 For 100%	F3 For 100%
1	Sodium lauryl ether sulphate (SLES)	25	26	25
2	Cocoamidopropyl Betain	10	12	15
3	Sodium Chloride	Q.S.	Q.S.	Q.S.
4	EDTA Diasodium	Q.S.	Q.S.	Q.S.
5	Glycerine	4	5	4
6	Distilled Water	55	70	65
7	Preservative	Q.S.	Q.S.	Q.S.
8	Tulsi essence	2	4	2
9	Aleo vera gel	Q.S.	Q.S.	Q.S
10	Lemon essence	Q.S.	Q.S.	Q.S.

Table No. 1

Procedure:

1. Firstly add some EDTA in Water and
2. add SLES to adjust the Foaming agent
3. add Cocoamidopropyl Betain , Glycerine, Preservative
4. NaCl in the end to viscosity build up.

Parameter of Base Formulation Of Scrub Face wash:

Sr. No.	**Parameter**	**F1**	**F2**	**F3**
1	**Appearance**	+++	+	+
2	**Colour**	++	+	+
3	**Consistency**	+++	+	
4	**Spreadability**	+++	+	++
5	**Feel**	+++	+	+
6	**Odour**	++	++	++

Here, += Good, ++= Better, +++= Best

From the above observation formula F1 was Stable and it shows consistency, spreadability, and feel therefore it was selected and extract was added with different concentration and forward for in vitro study as per IS and in vivo study with human volunteer.

Final Formulation of base Herbal Scrub Face wash using Activated Kojic Acid with Scrubing Agent (Poppy seeds)

Sr. No.	Ingredients	Formulation
1	Sodium lauryl ether sulphate (SLES)	25
2	Cocoamidopropyl Betain	10
3	Sodium Chloride	0.2
4	EDTA Diasodium	0.1
5	Glycerine	4
6	Distilled Water	55
7	Preservative	Q.S.

8	Tulsi essence	2
9	Kojic Acid	0.7
10	Poppy Seeds	3
11	Aloe vera gel	Q.S.
12	Lemon extract	Q.S.

Table No. 3

parameter of herbal scrub face wash

Sr. No.	Parameter	Formulation
1	Apperance	++
2	Colour	++
3	Consistency	+++
4	Foaming power	++
5	Feel	++
6	pH	**5.5**

Table No. 4

Abbrevation :

‘’+’’= poor, “++”=good, “+++= Satisfactory

Procedure :

1. Firstly add some EDTA in Water and
2. add SLES to adjust the Foaming agent
3. add Cocoamidopropyl Betain , Glycerine, Preservative
4. NaCl in the end to viscosity build up.
5. Add Poppy seeds at Temp 45℃
6. Continuesly Stirr of product after add scrubing agent.
7. Then reapet heat the product at 60-70℃
8. And at the last add Activated Kojic acid at Temp. 45℃.
9. At the end Stirr the product properly .

RESULT AND DISCUSSION:

a) In-Vitro Studies of face wash with different actives

a) Determination of physical parameters of Herbal Scrub face wash

Sr. No.	Parameters	FW1	FW2	FW3
1	Appearance	Clear	Clear	Clear
2	Colour	Colourless	Colourless	Colourless
3	Consistency	Not Good	Good	Good
4	Tacky Feel	No	No	No

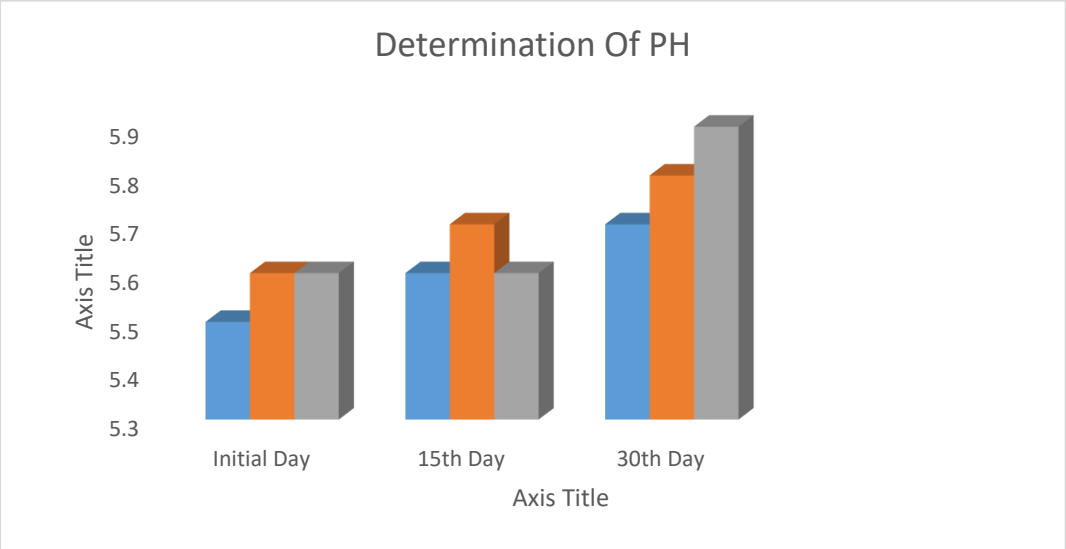
Table No. 5

Determination of pH of Face wash

Sr.No.	Time interval	FW1	FW2	FW3
1	Initial Day	5.5	5.6	5.6
2	15thDay	5.6	5.7	5.6
3	30th Day	5.7	5.8	5.9

Table No.6

Graphical Representation of Determination of pH



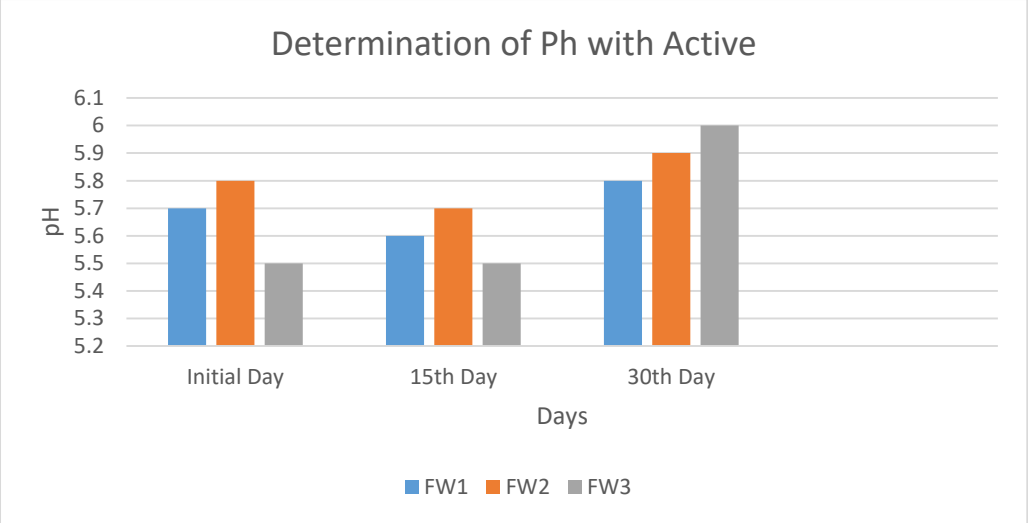
Determination of pH by using Active

Determination of pH Using Scrubing agent poppy seeds and Kojic acid extract :

Sr. No.	Time interval	FW1	FW2	FW3
1	Initial	5.7	5.8	5.5
2	15th Day	5.6	5.7	5.5
3	30th Day	5.8	5.9	6

Table No.7

Graphical Representation of Determination of pH

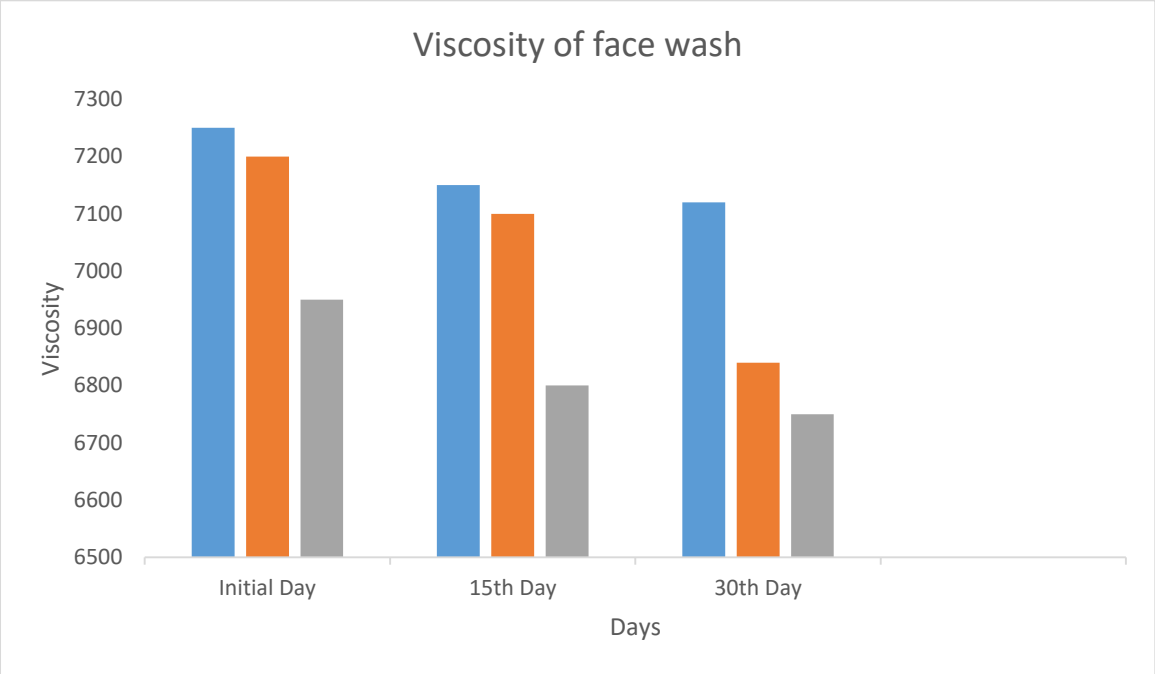


Determination of viscosity

The viscosity of face wash determine by using Brookfield Viscometer. The values obtained from the sample noted.

Sr. No.	No. of days	FW1	FW2	FW3
1	Initial Day	7250cp	7200cp	6950cp
2	15th Day	7150cp	7100cp	6800cp
3	30th Day	7120cp	6840cp	6750cp

Table No.8



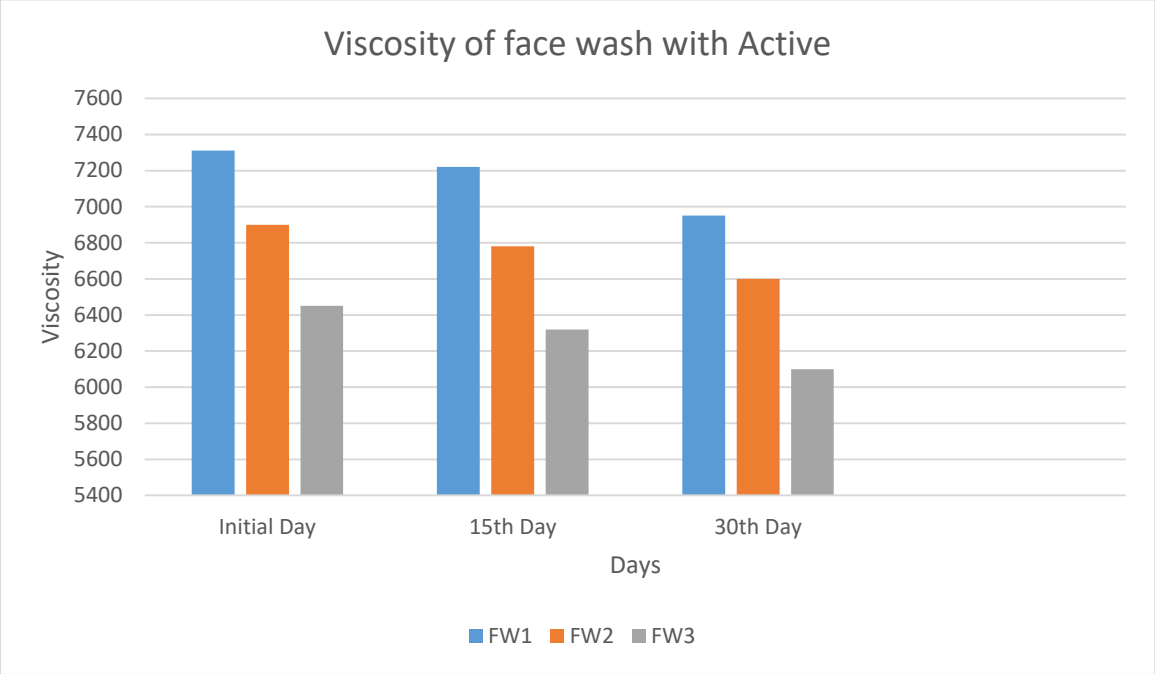
Determination of Viscosity by using scrubing agent Poppy seeds and Kojic acid extract

Sr.no	Time in Interval Time of interval	FW1	FW2	FW3
1	Initial Day	7310cp	6900cp	6450cp

2	**15th Day**	7220cp	6780cp	6320cp
3	**30th Day**	6950cp	6600cp	6100cp

Table No. 10

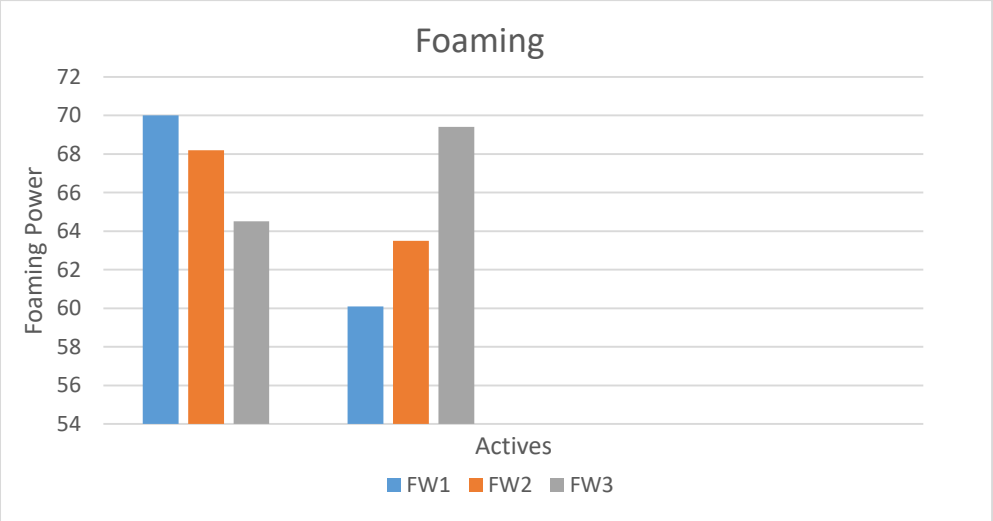
Graphical Representation of Determination of Viscosity



Determination of foaming Power

Sr. No.	Actives	FW1	FW2	FW2
1.	Kojic acid	70	68.2	64.3
2.	Scrubing poppy seeds	60.1	63.5	69.4

Table No. 11



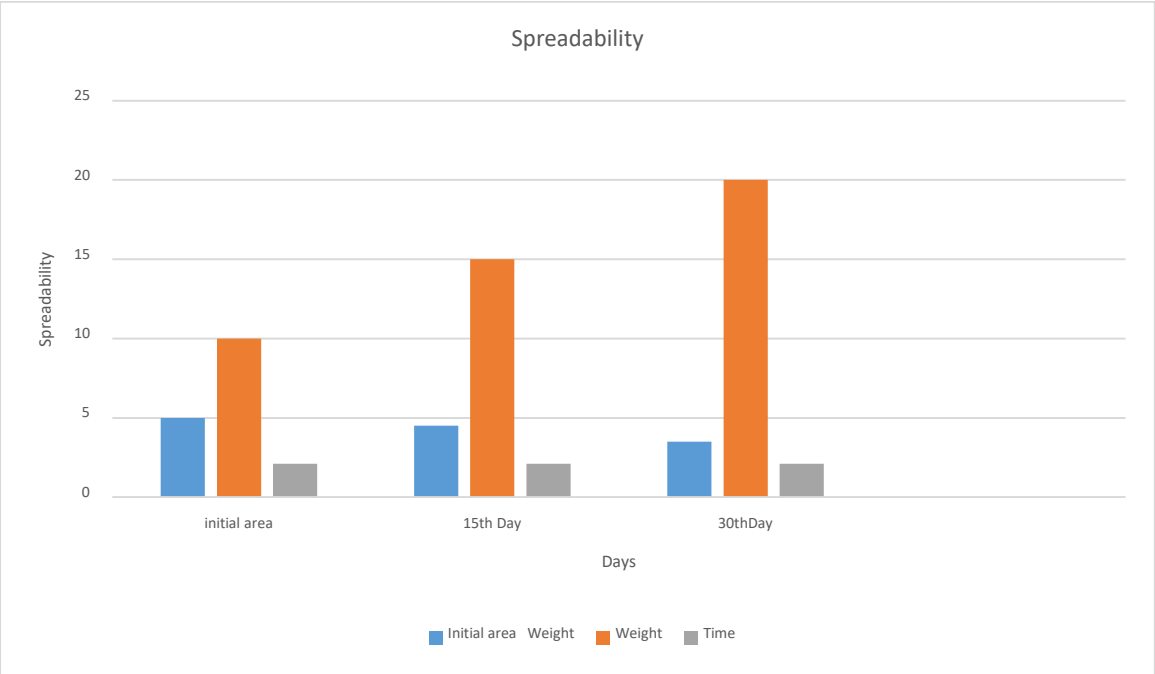
Determination of Spreadability Time

As per the formulation from scrubing poppy seeds shows results of foaming power FW2 It was observed that viscosity of formulation was found to be which was good. Therefore formulation passes test.

Determination of Spreadability

Sr. No.	Days of interval	Initial area	Weight	Time
1	Initial day	5cm	10gm	2.1 Sec
2	15th Day	1.5 cm	15 gm	2.1 Sec
3	30th Day	3.5 cm	20gm	2.1 Sec

Table No. 12



Cyclic Temprature test:

These tests are not carried out at fixed temprature and humidity. In this test, temprature was changed cyclically every day e.g. low -high-low-high to stimulate the changes in temprature daily.

Cyclic Temprature test

Sr. No.	**Parameter**	**F1**	**F2**	**F3**
1	Freeze Temprature	Stable	Stable	Stable
2	Room Temprature	Unstable	Unstable	Stable
3	High Temprature	Unstable	Unstable	Stable

Table No. 13

Determination of pH

Sr. No.	Name of Test	Result
1.	pH Determination	5.9

Table No. 14

Determination of Viscosity of Scrub Face Wash

Determination of Viscosity

Sr. No.	Viscosity	Result
1.	Viscosity	6660cp

Table No. 15

G) Determination of Foaming Power

Determination of Foaming Power

Sr. No.	Determination of Foaming	Result
1.	Foaming	64

Table No. 16

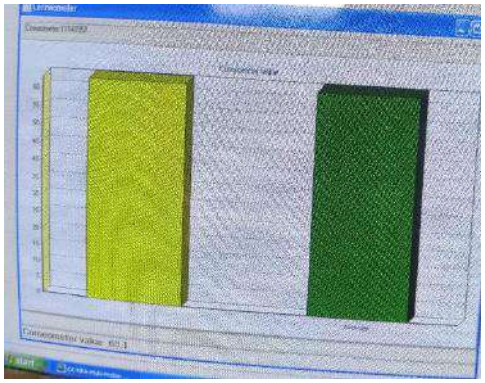
H) Determination of Spreadability

Determination of Spreadability

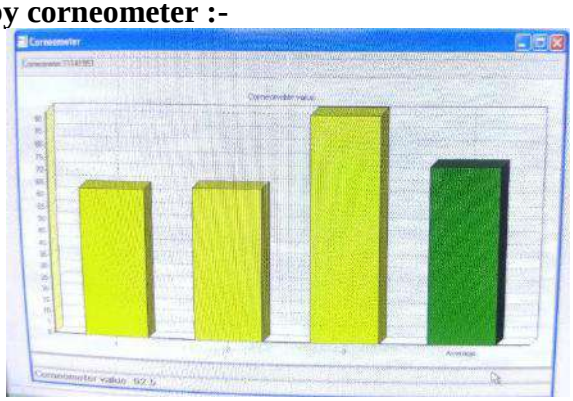
Sr. No.	Spreadability	Result
1	Spreadability	3.5

Table No. 17

Determination of Moisture content of skin by corneometer :-



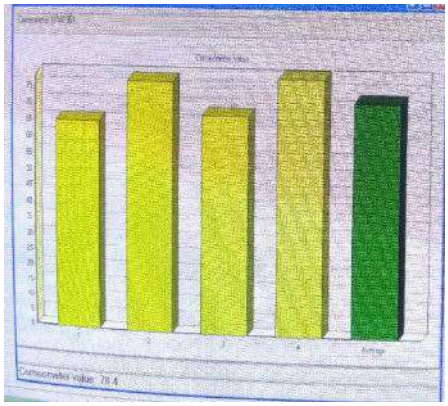
Corneometer analysis before application



Corneometer analysis after application

Result after 7 days :

Coroneometer analysis before application

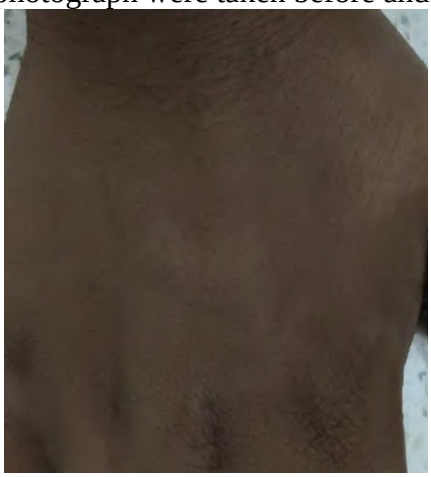


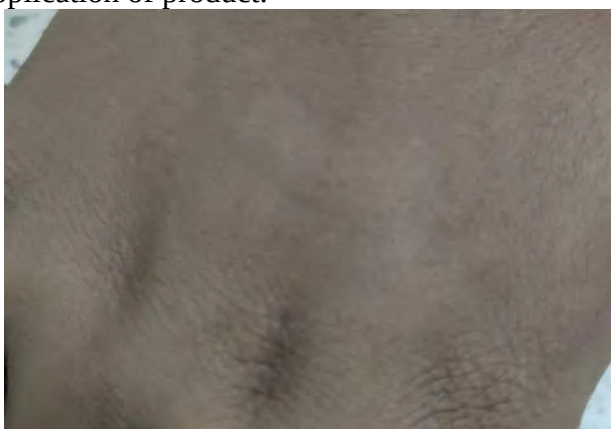
Corneometer analysis after application

Result :

The moisturizing activity was carried outby using corneometer. It was oberseved that before application of face wash , the moisture content of skin was less and after application of face wash moisture content was increased.

Photographic Evaluation : The study of effectiveness of product was done by the help of the volunteer study. This was carried out human volunteers. Face wash were applied on skin. The photograph were taken before and after application of product.



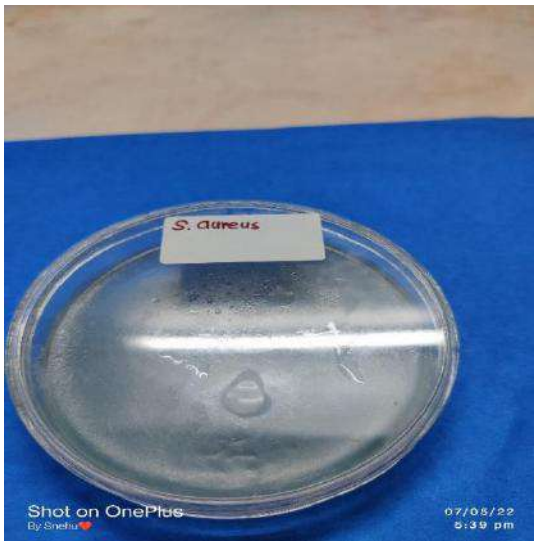


Before After

Determination of Microbial Testing

Interpretation of result:

Although there is some correlation between the size of the zone of inhibition and the susceptibility of the organism to the antibiotic, the former is a function of many variables e.g density of the inoculum, depth of the medium, diffusibility of antibiotic etc. The size of inhibition zone at which the organisum is considered Resistant, Intermediates or sensitive is given in the zone size interpretative chart as a part of this literature.



Conclusion :

At Present because of availability of cosmetic products in market, consumers are giving special attention Towards the selection of cosmetic product to develop a well standard formula; the new product viz. herbal face wash was formulated by incorporating active extract singly and also in combination for good effect.

Herbal scrub face wash was selected for sebum regulation activity because anti-acne face wash Contain good quality of extracts which helps to reduce sebum secreation and helps to remove oil and reduce pimple. Face wash prepared on synthetic base containing polymer, surfactant, humectant and preservatives etc. One formulation was selected from prepared base formulation on the basis of physical parameter for futher incorporation. Incorporation of active and sebum regulation property. Different formulation were prepared with varying concentraction of actives i.e anti-acne face wash with kojic acid. Evaluation studies like physical parameter, pH, viscosity, stability was done for selecting the final batch. In-Vivo study of final batch was taken. Cleansing activity were determine photographically.

Over the post few year, several methods are developed for an efficient cleansers with profound effect for various applications. There are various types of cleansers available depending on purpose and need. Kojic acid is used to remove acne and clear scars from the skin. Herbal scrub face wash is used to remove all the acne from the skin and reduce the scars. The single formulation Shows all the activities like sebum regulation and moisturization. Kojic acid is the key active ingredient of face wash helps to remove dirt from the skin and acne and lighteing the skin. So it is concluded that, the formulation of anti-acne face wash give the satisfactory result to the skin.

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16

Formulation and Development of Hair Conditioner With Glycolic Acid and Sesame Seed Oil

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ABSTRACT:
The hair care market is one of the largest personal care markets all over the world. Hair care is important to a person's overall appearance. Washing the hair and scalp has a become a near-universal practice. Good hair care is very important shampooing, conditioning and moisturizing the hair to restore healthy luster and sheen. In the past, the main aim of using hair care products was to clean the hair by removing spoilage and dirt. Today, hair care products are desired to provide additional benefits, such as beautifying the hair, making it easy to handle, or repairing damages.Hair care can be described as " preparations intended for placing in contact with the hair and scalp, with the purpose of cleansing, promoting attractiveness, altering appearance, and/or protecting them in order to maintain them in good condition".
The main objectives of the present study is to design and development of Hair care product for the damage and dry hair and maintained healthy looking hair. Studies have shown that glycolic acid penetrates throughout the hair shaft, and effect that persists even after the hair dries. Glycolic acid when used in hair care products it gives following application.
KEYWORDS : Hair care, Hair Conditioner, Glycolic Acid, Sesame Seed Oil, Stability testing, Physiochemical parameters.

INTRODUCTION –
Hair care can be described as " preparations intended for placing in contact with the hair and scalp, with the purpose of cleansing, promoting attractiveness, altering appearance, and/or protecting them in order to maintain them in good condition". Conditioners are one of the most popular hair care products. A conditioner consists of moisturising ingredients such as oils, humectants, silicones, butters, and emollients that nourish the hair and replenish it with moisture. Some conditioners also contain special proteins that help bind split-ends. Conditioners work by forming a protective coating around the cuticles. This coating helps cut the frizz, makes the hair soft, and also helps prevent damage from environmental aggressors. Conditioners are used to decrease friction, detangle the hair, minimize frizz and improve combability. Conditioners act by neutralizing the electrical negative charge of the hair fiber by adding positive charges and by lubricating the cuticle that reduces fiber hydrophilicity.
Hair is an imperative part of the human body which protects the scalp. Hair Conditioner is a hair care product, which is applied to the hair and hair tips after shampoo in order to condition the hair and then it is rinsed out. Hair Conditioner is used to improve the managebility and to enhance lustrous look of hair. Its main purpose is to reduce friction between the hair strands to allow easier brushing and combing. Hair conditioners are skin care product that are applied to the ends of the hair and later used for cleansing, conditioning the hair, and rinsing. It is used to make the hair shiny and smooth. Increases the luster of hair. Mainly prevents hair breakage, reduces split ends and improves manageability. Its main purpose is to reduce friction between hairs, making brushing and combing easier.
Conditioners are available as liquids, creams, or gels. The main ingredients in hair conditioners are the conditioning ingredients. There are various types of conditioning agents available, including lipids, silicones, quats, protein derivatives, silicones, and glycols, among others. The

product is beneficial to all types of hair. It works by restoring moisture, and smoothing the cuticles of the hair follicles. Hair conditioner comprising of powerful antioxidants can reduce UV damage to the hair including hair colour changes and protein damage.

CHARECTERISTICS OF GOOD CONDITIONER: -

1. Premium Moisture - The inherent quality that must exist is the conditioner must provide maximum moisture. It should be super hydrating and restore natural oils removed from hair from daily styling and shampooing. Emollients and humectants will provide moisture and shine.

2.Slip baby - In addition to moisture, a good conditioner will provide slip and thus have detangling ability. Slip is imperative to length retention and effective detangling.

3. Consistency - A thick and creamy conditioners. The conditioner must be able to absorb and protect at a high level, which means it needs to penetrate the hair shaft. This can be achieved by using a water based conditioner.

4. Leaves hair feeling uber soft

5. Good Ingredients- If you are partial to all natural ingredients, reading the ingredients is essential. Once you identify what your hair responds positively to, this will aid in your selection process.

BENEFITS OF CONDITIONER: -

1. It provides shine and smoothness
2. It tames split ends and flyaway hair
3. It improves manageability
4. It reduces the fiber hydrophilicity
5. It hydrates the hair

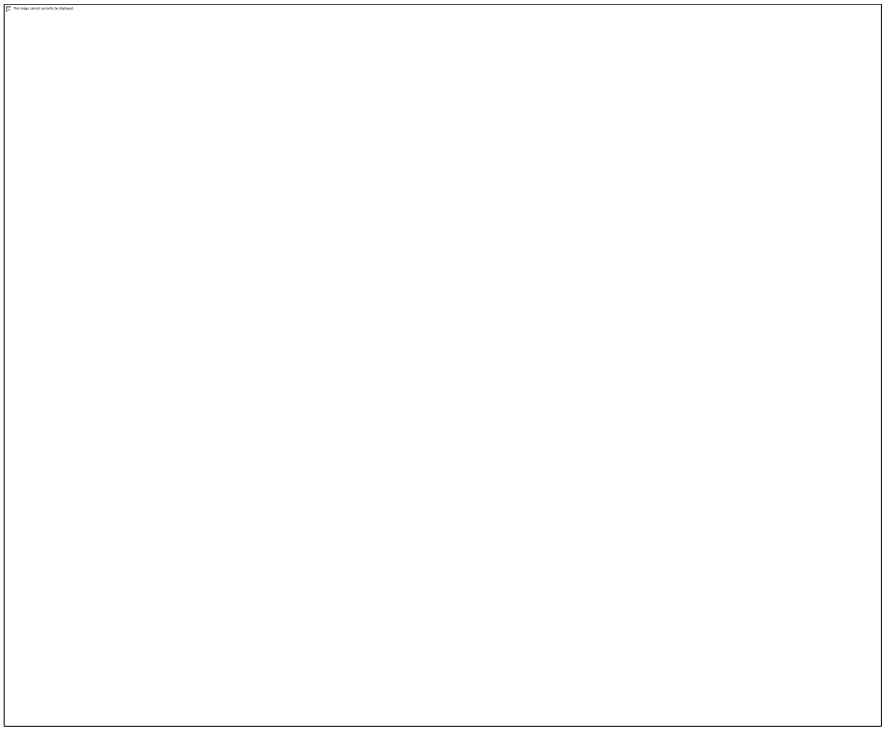
TYPES OF CONDITIONERS: -

1. Rinsed-out conditioners - Rinse-Out conditioners work by closing the cuticle scales of the hair. This is because they can be rinsed quickly and instantly. Their balanced composition and instant results often complement shampoos as a pre-wash, co-wash, or after-wash accessory. They contain ingredients that deliver instant moisturization and softness to hair. They are often made of mild yet hydrating ingredients that make them better suited for natural or straight hair.

2. Leave-in Conditioners - These conditioners are made with nourishing ingredients that penetrate the shaft to nourish strands. They are generally lighter, and the composition contains a fine blend of penetrative agents to maintain hair’s softness. This product comes in different forms, such as liquids, creams, or sprays. Leave in conditioners also help protect your hair from chlorine and salt.

3. Deep Conditioners - Deep Conditioners or hair masks are treatments that will help hydrate, repair or nourish you hair. They are designed to provide an even more potent effect than everyday conditioners. These usually have a thicker consistency.

4. Cleansing Conditioners - These are the ones that you can also wash your scalp and hair with (these are also known as co washes). Their cleansing properties will help remove residue and buildup and they are best for thick, curly hair as it helps to maintain moisture.



GLYCOLIC ACID HAIR BENEFITS

3. As the simplest alpha hydroxy acid (AHA) and smallest fruit acid, glycolic acid easily penetrates the cuticle layer of hair shafts. This characteristic allows glycolic acid to attachitself to keratin—a fibrous protein that protects hair from damage and promotes hair growth and length—to further strengthen hair and decrease breakage.

Glycolic acid is an alpha-hydroxy acid (or AHA, as it is commonly seen on product bottles) that can be naturally derived and acts as a gentle exfoliant. AHAs are typically made up of things like sugar cane, cirtus fruits, grapes, and even sour milk. Additionally, the size of the molecule itself is so small, it actually allows the acid to travel further, and deeper, within your skin and hair, making it more effective.

3. Glycolic acid's effectiveness in hair care applications that include

a. Revitalizing shampoos
b. Conditioners
c. Detanglers
d. Hair and scalp masks

Specifically, adding cosmetic-grade glycolic acid to a conditioner or a hair treatment product aid moisturizes both hair and scalp by improving the penetration and delivering the active ingredients into the hair shaft and skin. Also, glycolic acid in conditioning formulations provides moisturizing-like effects and softness that helps prevent hair breakage, giving hair overall better manageability.

4. **Glycolic Acid: Protecting Hair from the Inside Out**

Traditional hair conditioning agents repair and protect the exterior of the hair shaft. Less commonly known is that the hair shaft's interior can also suffer damage and weaken the hair shaft. The cortex—made of keratin—gives hair its pigment and provides hair's structural integrity, strength, and swelling/stretching ability. Chemours has demonstrated that glycolic acid penetrates the hair shaft, stabilizes the keratin within, and provides stronger, more manageable hair.

The biggest takeaway from glycolic acid and hair is its ability to help with dandruff. By moisturizing and exfoliating the scalp, glycolic acid will actually help treat and keep dandruff at bay. What's key here is the gentle exfoliation using the small AHA molecules along with the moisturization. Most other dandruff treatments focus on stripping the scalp of its woes, rather than removing and replacing what it lacks.On the flip side, glycolic acid can help balance an oily scalp as well. This again harkens back to its exfoliation properties, which in turn increase cell turnover. Regular cell turnover on the scalp, just like on our face, helps balance our skin and keep our follicles happy.While glycolic acid may become your scalp health holy grail, it is important to remember thatall good things come in moderation. Overusing glycolic acid on the scalp or leaving it on too long too frequently can actually cause irritation and further flakiness issues, as well as weaken your hair. Limit your use of this product to once, maybe twice a week if that, and no more than 20 to 30 minutes at a time.

Material and Methods:-

List of ingredients required:

1. Ceto stearyl alcohol - VBMV, Amravati
2. Shea butter- VBMV, Amravati
3. Glyceryl monostearate - VBMV, Amravati
4. Sesame seed oil - VBMV, Amravati
5. Iso-propyl-myristate- VBMV, Amravati
6. Behentrimonium chloride - Solvay Novecare
7. Cetrimonium chloride Solvay Novecare
8. Argon oil- VBMV, Amravati
9. Guar hydroxy trimonium chloride- Solvay Novecare
10. Dimethicone - VBMV, Amravati
11. Phenoxy ethanol - VBMV, Amravati
12. Water - Ganesh scientific, Amravati
13. Glycolic acid - Chemico health and beauty India Pvt.Ltd. Mumbai
14. Glycerin- VBMV, Amravati

List of equipment: -

1) pH meter – Labline, Mumbai
2) Brook field Viscometer – Brookfield engineering labs
3) Round electronic Hot plate – Bio Technics India, model BTI-22
4) Remi Motor– Remi Elektrotechnik Ltd. Vasai, Type – RQ- 122
5) Stability chamber – Labline
6) Humidity chamber – Bio techno lab
7) Weighing balance – VBMV, Amravati

Method of preparation of Hair Conditioner -

Sr.no.	**Ingredients**	**Quantity for 100 gm**			
		F1	F2	F3	F4
1.	Ceto-stearyl alcohol	6	5	5.5	5
2.	Glyceryl monostearate	1.5	1.2	1	2
3.	Emulsifying wax	0.8	1	0.5	0.4
4.	Light liquid paraffin	1.2	2.2	3	2.5
5.	Behentrimonium chloride	3	1	2.5	2
6.	Cetrimonium chloride	1	2	1	1.2
7.	Argan oil	0.8	1	0.4	0.6
8.	Iso-propyl myristate	1	3	1.5	2
9.	Jaguar (GHTC)	1	0.5	0.4	0.3

10.	Glycerine	1.2	0.8	1.5	1
11.	Phenoxy ethanol	0.2	0.2	0.2	0.2
12.	Citric acid	0.42	0.12	0.12	0.12
13.	Water	80	81	81	82
14.	Dimethicone	1,5	0.8	1	0.7
15.	Perfume	0.1	0.2	0.3	0.4

Base Formulation of Hair ConditionerTable no. 1: -

PROCEDURE –

1) Clean all apparatus and weigh all the ingredients properly.
2) Phase A i.e., oil phase was weighed accurately in a beaker and Phase B i.e. water phase inanother beaker.
3) Both phases are heated on hot plate until it melts completely and reaches up to temperature70 -80°C
4) Pour Oil phase in Water phase with slow stirring as it is oil in water type of emulsion.
5) Mix both the phases properly with continuous slow stirring.
6) Add Cetrimonium chloride and dimethicone (silicone oil) in it when it reaches to 50 -45°C,
7) When it reaches to proper texture and consistency check pH and pH was adjusted by citricacid.
8) At last step add perfume with slow stirring check if it affects viscosity.
9) Pour the product in proper clean container.

Optimization of Hair Conditioner Base-

Optimization of formulation means selection of a stable formulation which is carried out on the basis of various parameters such as appearance, color, feel, odour, Spreadability, etc. The optimization of hair conditioner base was carried out after a particular time period and at different temperature. The optimization of hair conditioner base was done on the basis of above-mentioned parameters and the results are as follows,

Sr. No.	**Parameters**	Formulation			
		F1	F2	F3	F4
1.	Appearance	-	++	+++	+++
2.	Colour	++	++	++	+++
3.	Spreadability	+	++	++	+++
4.	Flowability	-	++	++	++
5.	Odour	++	+++	+++	+++
6.	Feel	-	+	++	+++

Table – Showing Characteristic of Hair Conditioner Base

Abbreviation: - (+) = good, (++) = better, (+++) = best, (-) = average

Based on the results obtained from the stability study of hair conditioner base of formulation F1, F2, F3, and F4**, "base F4"** was selected for the final formulation as it shows all desired properties.

Incorporation of glycolic acid and sesame seed oil in final baseformulation of hair conditioner

After the optimization of hair conditioner base, as per optimization results F4 was selected and now active was added into the formulation into different concentration.

Table – Formulation of hair conditioner with glycolic acid sesame seed oil

Sr.no.	Ingredients	Quantity for 100 gm		
		F1	F2	F3
1.	Ceto-stearyl alcohol	5	5	5
2	Glyceryl monostearate	2	2	2
3.	Emulsifying wax	0.4	0.4	0.4
4.	Light liquid paraffin	2.5	2.5	2.5
5	Behentrimonium chloride	2	2	2
6.	Cetrimonium chloride	1	1	1
7.	Sesame oil	0.4	0.2	0.5
8.	Argan oil	0.6	0.6	0.6
9.	Iso-propyl myristate	1.5	1.5	1.5
10.	Jaguar (GHTC)	0.3	0.3	0.3
11.	Glycerin	1	1	1
12.	Phenoxy ethanol	0.2	0.2	0.2
13.	Water	82	82	82
14.	Dimethicone	0.7	0.7	0.7
15.	Glycolic acid	1	1.5	2
16.	Citric acid	0.2	0.2	0.2

PROCEDURE –

1) Clean all apparatus and weigh all the ingredients properly.
2) Phase A i.e., oil phase was weighed accurately in a beaker and Phase B i.e. water phase inanother beaker.
3) Both phases are heated on hot plate until it melts completely and reaches up to temperature70 -80°C
4) Pour Oil phase in Water phase with slow stirring as it is oil in water type of emulsion.
5) Mix both the phases properly with continuous slow stirring.
6) Add Cetrimonium chloride and dimethicone (silicone oil) in it when it reaches to 50 - 45°C,
7) When it reaches to proper texture and consistency add Glycolic acid and check pH and pHwas adjusted by citric acid if necessary.
8) At last step add perfume with slow stirring check if it affects viscosity.
9) Pour the product in proper clean container.

Above formulation was optimized to check the stability of product after incorporation of active.

OPTIMIZATION

Table – Examination of hair conditioner base with active

Sr.no.	Parameters	Day 1			Day 5			Day 8		
		F1	F2	F3	F1	F2	F3	F1	F2	F3
1.	Appearance	+++	+++	++	+++	+++	++	+++	+++	++
2.	Colour	+++	+++	++	++	+++	++	-	++	++
3.	Spreadability	++	+++	++	++	+++	++	++	+++	-
4.	Flowability	++	++	+++	++	++	+++	+	++	+++
5.	Odour	+++	+++	+++	+++	+++	++	+++	+++	++
6.	Stability	+++	+++	++	+++	+++	++	+	++	+

Abbreviations: - (+) = good, (++) = better, (+++) = best, (-) = average

Based om the above examination **formulation F2** was selected for the final formulation as well as evaluation.

Final Formulation of Hair Conditioner –

Table – Formulation of hair conditioner with glycolic acid sesame seedoil

Sr.no.	Ingredients	Quantity for 100 gm
1.	Ceto-stearyl alcohol	5
2.	Glyceryl monostearate	2
3.	Emulsifying wax	0.4
4.	Light liquid paraffin	2.5
5.	Behentrimonium chloride	2
6.	Cetrimonium chloride	1
7.	Sesame oil	0.2
8.	Argan oil	0.6
9.	Iso-propyl myristate	1.5
10.	Jaguar (GHTC)	0.3
11.	Glycerin	1
12.	Phenoxy ethanol	0.2
13.	Water	82
14.	Dimethicone	0.7
15.	Glycolic acid	1.5
16.	Citric acid	0.
17.	Perfume	0.3

The final formulation of hair conditioner was formulated using same procedure as mentioned for final base formulation. On the final formulation mentioned above further quality tests were performed as per the BIS (Bureau of Indian Standard).

EVALUATION -

In- vitro evaluation of parameters:

A. Determination of physical parameter of Hair Conditioner

Appearance: Visually appearance of the formulation observed.
Colour: Colour of the formulation check visually.
Consistency: Consistency was check weather its satisfactory or poor or good.
Tacky feel: Tackiness were check after application on palm.
Odour : odour was check by smelling.

B. Determination of thermal stability –

Test for thermal stability –

13. **Apparatus – humidity chamber / stability chamber controlled at 60-70 % RH and 37 +1°C.**

Procedure -

1. Spread a 20 mm broad and 5 mm thick stripe from the material to be tested on the internal wall of beaker of 100 ml capacity in its total height. Keep the beaker for 8 hours in the humidity chamber at 45% – 50% relative humidity. Physical stability test of the formulation was carried out for two weeks at various temperature conditions like at 45^{o}C temperature, room temperature, 4^{o}C and 45% relative humidity in a close container.

C. Determination of pH –

Apparatus – a pH meter, beaker

Procedure: -

For oil-in water emulsions creams – Weigh accurately 5 + 0.01 g of the cream in a 100 ml beaker. Add 45 ml of water and disperse the cream in it for some time. The reading is recorded to Determine the pH of the suspension at 27°C using the pH meter.

D. **Determination of Viscosity**

Apparatus: Brookfield viscometer, beaker

The selection of spindles is based on the viscosity of the sample being tested, as thespindle needs to be able to rotate freely in the sample without getting stuck or jammed.

- Rheology experiment: Rotational spindle Brookfield viscometer (Model DV-I plus, LV, USA) instrument was used for rheology experiment.100 mL of the conditioner is taken in a beaker and the spindle is dipped in it for about 5 minand then the reading is taken

The viscosity of hair conditioner was determined by using **spindle no. 5** at 10 to 100 rpm.

Stability study of conditioner

The sample of hair conditioner was kept at 5^{o}C, room temperature 40^{o}C. The changes in physical
appearance, color, feel etc were studied.

F. **Accelerated stability study**

Accelerated stability studies at various temperature for hair conditionerProcedure:

The stability of the final formulation was checked at room temperature, 45^{0}C ±2^{0}C, and freeze thaw cycles. To ensure that a cosmetic remain stable till the consumers has used the entire cosmetic or has stopped using it, a number of special accelerated test procedures have been developed. The evaluation employs a combination of tests. This method of evaluation not only indicates stability of Base formulation but also indicates the stability of functional ingredient.

Cyclic Temperature Test:
These tests are not carried out at fixed temperature and humidity. In this test, temperature was changed cyclically every day e.g. low-high-low-high to stimulate the changes in temperature daily.

G. **Determination of Spreadability Time:**
Apparatus: Petri dish
Procedure:
Clean and dry Petri dish well to perform the test. Place 1gm of Conditioner on outer side of the Petri dish on it, press a little. Kept a side for a min then note the diameter of sample in centimeter (cm) using scale.

In- vivo evaluation of parameters:
A. Determination of Patch test –

If there is no redness, itching, or irritation, then the conditioner is safe to use. However, if there is any redness, itching, or irritation, then the conditioner is not suitable for use and should be tested on another patch of skin.
B. Skin irritation:
The skin irritation was carried out on human volunteers. For formulated conditioner, five volunteers were selected and 1.0 gm of formulated product was applied on an area two square inch to the back of the hand and on small part of hair. The volunteers were observed for lesions of irritation.
C. Photographic Evaluation
The study of effectiveness of product was done by the help of the volunteer study. This was carried out human volunteers. Hair conditioner were applied on skin. The photograph were taken before and after application of produc.

Results-
A. Determination of physical parameter of Hair Conditioner –

Sr. No.	**Parameters**	**Quantity of glycolic acid**		
		1 %	1.5 %	2%
1.	Appearance	Opaque	Opaque	Opaque
2.	Colour	Good	Very Good	Very good
3.	Spreadability	Good	Very Good	Good
4.	Flowability	Flowable	Flowable	Flowable
5.	Odour	Good	Very good	Good
6.	Feel	Very good	Good	Very good

Table - Determination of physical parameter of Hair Conditioner

It was observed that, Formulation FW2 parameters are satisfactory as compared to F1 andF3 hence **F2 selected.**

B. Determination of thermal stability –

Sr. No.	Thermal Stability	At 4^{o} Temp.	At Room Temp.	At 45^{o} Temp.
1.	Initial Day	Stable	Stable	Stable

2.	After 7 Days	Stable	Stable	Stable
3.	After 15 Days	Stable	Stable	unstable
4.	After 30 Days	Unstable	Stable	Stable

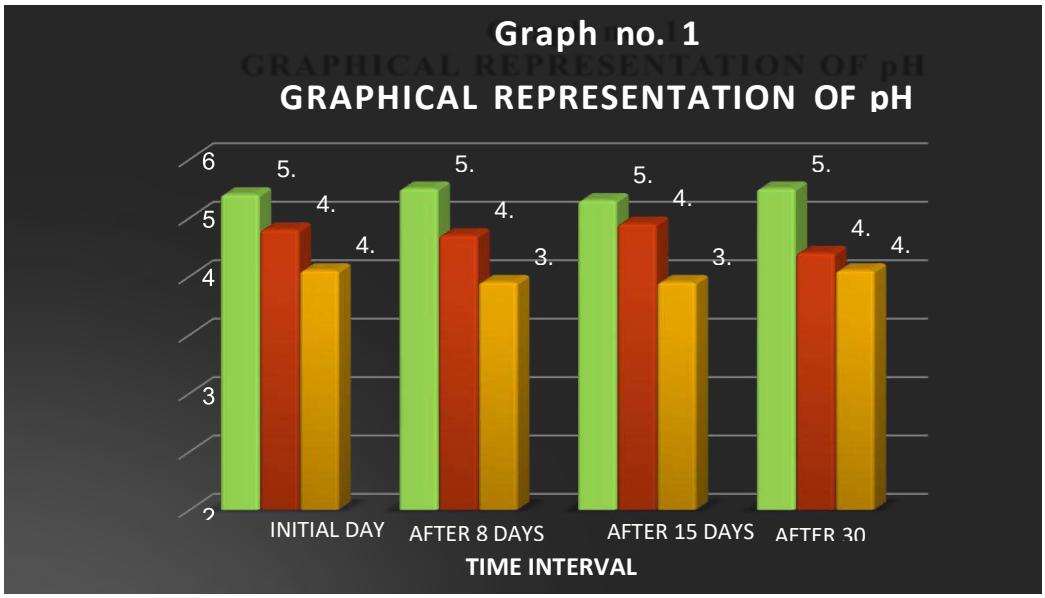
Table - Showing Thermal Stability of Conditioner

The product shall be taken to have passed the test if, on removal from the thermostat, no oil separation is observable.

C. Determination of pH –

Parameter	**Time interval**	**Result**		
		1	2	3
pH	Initial day	5.4	4.8	4.1
pH	After 8 days	5.5	4.7	3.9
pH	After 15 days	5.3	4.9	3.5
pH	After 30days	5.5	4.4	3.6

Table - Determination of Ph



D. Determination of Viscosity -

Parameter		Result		
		1.	**2.**	**3.**
Viscosity	Initial day	68500 cp	66800 cp	59500 cp
	After 15 days	68250cp	65870 cp	58380 cp
	After 30 days	68300 cp	66710 cp	56000 cp

it shows that the product is viscous paste like and can easily be removable from its packing

E. Test For Stability Study –

Sr. No.	Parameter	F1	F2	F3
1	Appearance	++	++	++
2	Colour	++	++	++
3	Spreadability	+++	+++	++
4	Oily/tacky feel	++	++	++

Table - Stability study of conditioner:

F. Accelerated stability study

Accelerated stability studies at various temperature

Sr. no.	Parameter	Room Temperature			45±2ºC			Freeze Thaw Cycle		
		F1	F2	F3	F1	F2	F3	F1	F2	F3
1	Apearance	NC	NC	NC	NC	NC	NC	SC	NC	NC
2	Colour	NC	NC	NC	NC	NC	NC	NC	NC	NC
3	Consistency	NC	NC	NC	SC	NC	SC	SC	SC	SC
4	Flowability	NC	NC	NC	NC	NC	NC	NC	NC	NC
5	Spreadability	NC	NC	NC	NC	NC	NC	SC	NC	NC
6	Feel on application	E	E	E	E	E	E	E	E	E

Table - Accelerated stability studies

NC= No Change, **SC**= slightchange, **E** = Excellent

From the above table, it is observed that F-2 had not changed its physical properties exceptfor consistency at $45^0C \pm 2^0C$. Hence, it was stable. Therefore, there formulation**, F-2** was selected for subjective study.

- **Cyclic Temperature test**

Sr.No.	Parameter	F1	F2	F3
1	Freeze Temperature	Stable	Stable	Stable
2	Room Temperature	Unstable	Stable	Stable
3	High Temperature	Unstable	Stable	Unstable

Table - Cyclic Temperature test

G. Determination of Spreadability Time:

Sr. No.	Spreadability	Result
1		3.5

2	Spreadability	3.4
3		3

Table - Determination of Spreadability

In- vivo evaluation of parameters:

A. Determination of Patch test –Table

Sr.no	Parameter	Results
1.	Immediate after application	No reaction
2.	After 1 hour	No reaction
3.	After 24 hours	No reaction

Result – The product passes the test as no reaction was occur.

C. Skin irritation:

Parameter	Skin irritation test
F1	No irritation
F2	No irritation
F3	No irritation

C. **Photographic Evaluation -**

CONCLUSION

From the above studies it is concluded that the hair conditioners show an excellent property of conditioning. hair conditioner has a cationic surfactant which gives good cleansing action Hair conditioner is one of the cosmetics which is widely used in daily life. They will act basically on shaft of the hair. The conditioner functions to impart manageability, gloss and antistatic properties to hair. Conditioners also attempt to recondition hair that has been damaged by chemical/mechanical trauma common sources of trauma. Include excessive brushing, hot

blowing, drying, permanent hair waves, bleaching etc. This article gives an ideaabout hair parts, types , benefits, purpose, functions of conditioner and some commonly used ingredients in formulation of hair conditioner.

The main causes of hair damaged and dry are due to cheap hair product, styling hair with heat without protecting it and not replenishing moisture that is last during styling, blow drying, harmful environment. So considering this present study was carried out formulate hair care product with active ingredient alpha hydroxy acid such as glycolic acid, in different concentration, and they were tested according to Indian standard.

Thus conclusion can be made that the conditioner containing active ingredient glycolic acid have been able to deliver positive benefits to hair by penetrating throughout the hair shaft, so enhancing softness, shine, elasticity and manageability of hair by conditioning, moisturizing allowing hair to better withstand heat and preventing breakage. The data from these experiments clearly shows that both healthy hair and bleached hair can benefit from the multiple positive effects of active.

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Nanotechnology In Cosmetics And Cosmeceuticals

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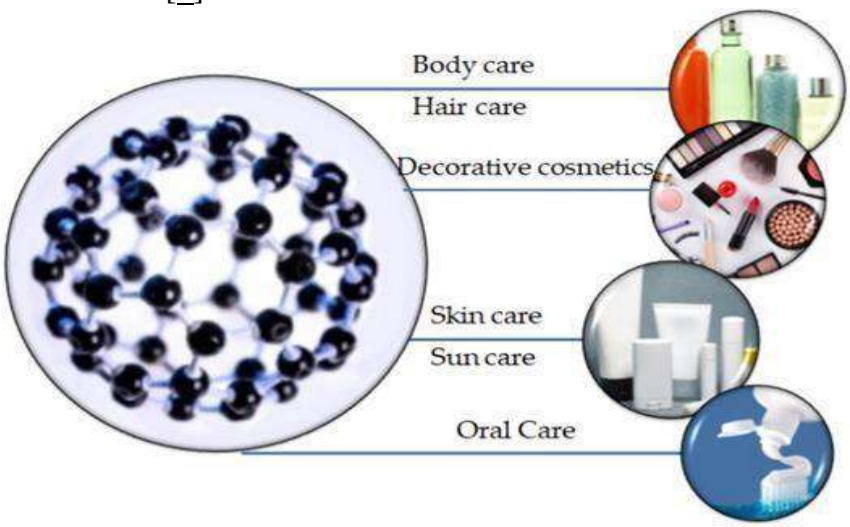
ABSTRACT:
Nanotechnology is now known as the 'hottest technology' that recently available in cosmetic field, being used by most of the beauty-concerned consumers. Nanotechnology and nano delivery systems are innovative areas of science that comprise the design, characterization, manufacturing, and application of materials, devices, and systems at the nanoscale level (1–100 nm). Nanotechnology is the science of manipulating atoms and molecules in the nanoscale – 80000b times smaller than the width of a human hair. Incorporation of nanotechnology in cosmeceuticals is aimed at making incense of perfumes last longer, sunscreens to protect the skin, antiaging creams to fight back the years, and moisturizers to maintain the hydration of skin. The different types of nanomaterials employed in cosmetics include nanosomes, liposomes, fullerenes, solid lipid nanoparticles etc. The aim of the review is, thus, to provide an update on the current status and trends of research and industrial development related to the use of nanotechnology in cosmetics and to give an indication of where the field could be heading in the future.
It outlines their benefits, as well as potential health and environmental risks. Further, it highlights the regulatory status of cosmeceuticals. Finally, this article seeks to provide an overview of nanocosmetics and nanocosmeceuticals and their applications in cosmetic industries, which may help consumers and regulators to gain awareness about the benefits as well as the toxicity related to the continuous and long-term uses of these products, thus encouraging their judicious use.

KEYWORDS: nanotechnology; nanomaterial; cosmetics; cosmeceuticals; nanocosmetics; nanocosmeceuticals; patent; regulation; health hazards; toxicity

INTRODUCTION –

Nanotechnology incorporated in cosmetics has now been highly regarded by companies and manufacturers due to its low production cost and enriched characteristics. One of the most common cosmetic products utilized in nanotechnology include moisturizers, sunscreens, lip care, and hair care products.[1] Nanotechnology is regarded as the most imminent technology of 21st century and is contemplated as a big boon in the cosmetic industry. The term nanotechnology is the combination of two words: namely, technology and the Greek numerical **"nano"** which means **dwarf**. Nanotechnology and nanodelivery systems are innovative areas of science that comprise the design, characterization, manufacturing, and application of materials, devices, and systems at the nanoscale level (1–100 nm). Nanotechnology, being recognized as one of the revolutionizing technologies, is extensively studied in the area of cosmetics and cosmeceuticals [1,2]. Nanotechnology can increase the surface area of a material. This allows more atoms to interact with other materials. An increased surface area is one of the chief reasons nanometer-scale materials can be stronger, more durable, and more conductive than their larger-scale counterparts. The Drugs and Cosmetics Act 1940 and Rules 1945 defines a cosmetic as "any article intended to be rubbed, poured, sprinkled or sprayed on, or introduced into, or otherwise applied to the human body or any part thereof for cleansing, beautifying, promoting attractiveness, or altering the appearance, and includes any article

intended for use as a component of cosmetic" [4]. In the cosmetic area it is believed that the smaller particles are readily absorbed into the skin and repair damage easily and more efficiently. Incorporation of nanotechnology in cosmeceuticals is aimed at making incense of perfumes last longer, sunscreens to protect the skin, antiaging creams to fight back the years, and moisturizers to maintain the hydration of skin. Some of the nanotechnology-based innovations are nanoemulsions (which are transparent and have unique tactile and texture properties), nano capsules (which are used in skin care products), nanopigments (that are transparent and increase the efficiency of sunscreen products), liposome formulations (which contain small vesicles consisting of conventional cosmetic materials that protect oxygen or light sensitive cosmetic ingredients), niosome, nanocrystals, solid lipid nanoparticles, carbon nanotubes, fullerenes, and dendrimers. Despite these definitions, the legal meaning of cosmetics in many nations is more extensive. In some Western nations, cosmetics are normally interpreted as just beautifying products, such as lipstick, mascara, eyeliners, highlighter, and a few other items of this kind [5].



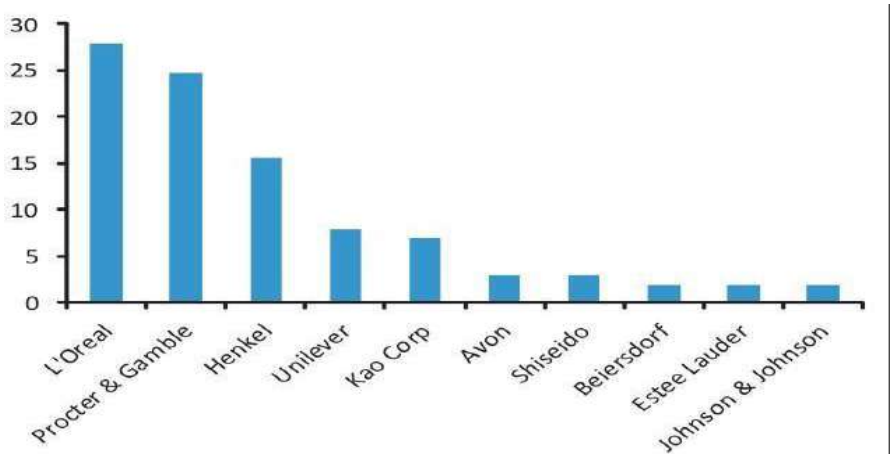
NANOCOSMETICS: -

Nanocosmetics are personal care products containing nanocarriers or nanoparticles. The Nanocosmetics aims products intended for application to the skin of the face and body, with antiaging action and photo protection, capable of penetrating into the deep layers of the skin, potentiating the effects of the active. Fronza and collaborators in 2007 defined nanocosmetic as "a cosmetic formulation that carries actives or other nanostructured ingredients, which has superior properties regarding its performance if compared with conventional products".[9]

FRONT-RUNNING BRANDS OF NANOCOSMETICS -

It has been found out from different surveys that almost all the major cosmetic manufacturers use nanotechnology in their various products. Cosmetics giant Estee Lauder entered the Nano Market in 2006 with a range of products containing "NanoParticles" L'Oréal, the world's largest cosmetics company, is devoting about $600 million dollars, of its $17 billion dollar revenues, to Nano patents, and has patented the use of dozens of "nanosome particles". It ranks number 6 in nanotech patent holders in the U.S. Other examples include Freeze 24/7, DDF (Doctor's Dermatologic Formula), and Colorescience. An estimation of how the top 10 cosmetic companies of the world rank in terms of nano-related patents, based on Espacenet database, is depicted in Graph 1.[11]

Graph 1: Ranking of top 10 beauty companies in terms of number of nano-related patents.



The first ever nanocosmetic product to be launched was anti-aging liposomes, "Capture Totale" in 1986 by Dior, this was followed by L'Oreal Paris' "Plentitude Revitalift", again an anti-aging cream consisting of polymeric nanocapsules of active agent retinol [5], [6]. The global leads of the cosmetic industry who extensively employ nanotechnology in their products are companies like Estee Lauder, Purology, Dior, L'Oreal, Proctor & Gamble, Colorescience, and Revlon to name a few [7]. L'Oréal: The brand's Age Perfect Glow Renewal Facial Oil contains **nanopeptides, which are nanomaterials that help strengthen and repair the skin's barrier.** Estée Lauder: Estée Lauder's Double Wear Stay-in-Place Makeup Foundation has a complex of nanoparticles that help to keep the product in place.Maybelline: Maybelline's Fit Me Matte + Pore less Foundation contains **silica, which is a type of nanomaterial that helps to create a matte finish.**

Clinique: Clinique's Superprimer Face Primers feature various nanomaterials, such as **zinc oxide, titanium dioxide, and iron oxides, that help to give the skin a smoother, more even appearance.**

Neutrogena: Neutrogena's Hydro Boost Gel-Cream contains **hyaluronic acid, which is a nanomaterial that helps to hydrate the skin.** According to the Nanotechnology Products Database, there are currently 903 nanocosmetic products of 104 types, made by 276 companies in 31 countries [8].

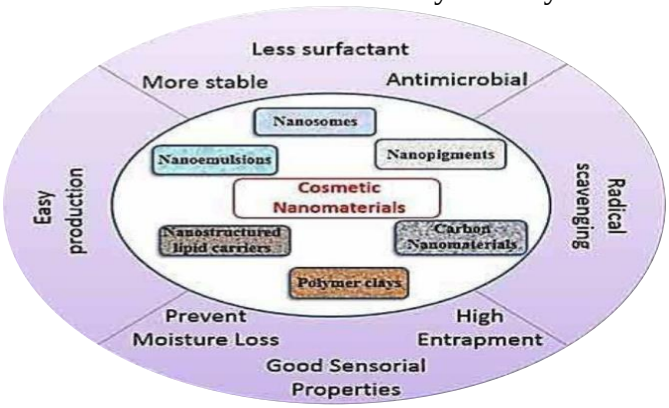
NEED OF NANOMETERIALS IN COSMETICS –

A number of nanomaterial types are already in use, including nanoemulsions, and nanoparticles of minerals present in our natural environment, such as titanium dioxide (TiO_2), zinc oxide (ZnO), alumina, silver, silicon dioxide, calcium fluoride and copper. The rationale for the use of nanomaterials in cosmetic products is, of course, that they offer added value in terms of product performance.

The unique properties and behavior of nanomaterials mean that nanotechnologies could profoundly transform industry and everyday life. In formulation of cosmetics, Titanium dioxide (TiO_2) and Zinc Oxide (ZnO) nanopigments are the main compounds used as highly efficient UV-filters, able to reflect and scatter the visible part of solar radiation while absorbing UV light. Given these properties, they are extensively used in sunscreens. Other examples of nanocosmetic products on the market include body firming lotion, bronzer, exfoliant scrub, eye liner, and styling gel, to name but a few. Nanocosmeceuticals have also been highly exploited for formulating various anti-aging formulations. They are successfully marketed as skincare, hair care, and nail care products, among others, claiming to stimulate their growth, protect their structure, and increase hydration power, thus improving their effectiveness as cosmetic products [12,13].

- ❑ Nanomaterials used in cosmetics differ from nanomaterials used by other industries.

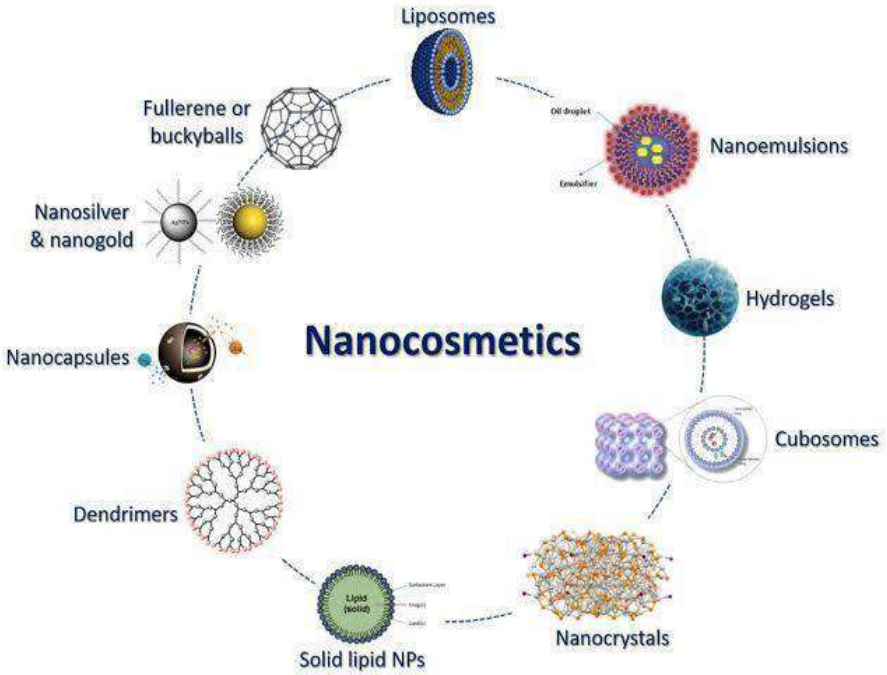
- They differ by their shape, their molecular structure, their mode of use and their specific interactions with the living world and the environment.
- Recently, Nanoparticles can be used to create containers that are more resistant to air and light, keeping cosmetics fresher for longer. Nanoparticles can also be used to create packaging materials that are more environmentally friendly.



The primary advantages of using nanoparticles in cosmeceuticals include improvement in the stability of cosmetic ingredients (e.g., vitamins, unsaturated fatty acids, and antioxidants) by encapsulating within the nanoparticles; efficient protection of the skin from harmful ultraviolet (UV) rays; aesthetically pleasing products (e.g., in mineral sunscreens, using smaller particles of active mineral allows them to be applied without leaving a noticeable white cast); targeting of active ingredient to the desired site and controlled release of active ingredients for prolonged effect.

TYPES OF NANOMATERIALS USED IN COSMETICS -

Most popular nanomaterials applied in cosmetics is provided, grouped into two broad classes— organic and inorganic NPs



Liposomes - Liposomes are concentric bilayered vesicles in which the aqueous volume is entirely enclosed by a lipid bilayer composed of natural or synthetic phospholipids which are GRAS (generally regarded as safe) products. The lipid bilayer of liposomes can fuse with other bilayers such as the cell membrane, which promotes release of its contents, making them useful

for cosmetic delivery applications. Their ease of preparation, enhanced absorption of active ingredients by skin and continuous supply of agents into the cells over a sustained period of time make them suitable for cosmetic applications.[14,15]

Nanoemulsions - They are dispersions of nanoscale droplets of one liquid within another.[18] They are metastable systems whose structure can be manipulated based on the method of preparation. The components used for their preparation are GRAS products and are safe to use. Their smaller particle size provide higher stability and better suitability to carry active ingredients; they also increase the shelf life of the product.[19,20]

Nanocapsules - Nanocapsules are sub microscopic particles that are made of a polymeric capsule surrounding an aqueous or oily core. It has been found that the use of nanocapsules decreases the penetration of UV filter octyl methoxycinnamate in pig skin when compared with conventional emulsions.[21]

Solid lipid nanoparticles - They are oily droplets of lipids which are solid at body temperature and stabilized by surfactants. They can protect the encapsulated ingredients from degradation, used for the controlled delivery of cosmetic agents over a prolonged period of time and have been found to improve the penetration of active compounds into the stratum corneum.[22] In vivo studies have shown that an SLN-containing formulation is more efficient in skin hydration than a placebo.They have also been found to show UV-resistant properties, which were enhanced when a molecular sunscreen was incorporated and tested. Enhanced UV blocking by 3,4,5-trimethoxybenzoylchitin (a good UV absorber) was seen when incorporated into SLNs.[23]

Nanocrystals - They are aggregates comprising several hundred to tens of thousands of atoms that combine into a “cluster”. Typical sizes of these aggregates are between 10 and 400 nm and they exhibit physical and chemical properties somewhere between that of bulk solids and molecules. They allow safe and effective passage through skin.[24]

Nanosilver and Nanogold - Cosmetic manufacturers are harnessing the enhanced antibacterial properties of nanosilver in a range of applications. Some manufacturers are already producing underarm deodorants with claims that the silver in the product will provide up to 24-hour antibacterial protection. Nano-sized gold, like nanosilver, is claimed to be highly effective in disinfecting the bacteria in the mouth and has also been added to toothpaste.[25]

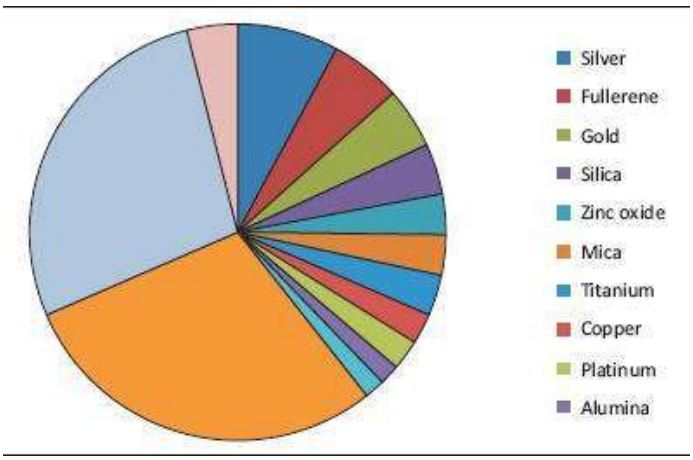
Dendrimers - Dendrimers are unimolecular, monodisperse, micellar nanostructures, around 20 nm in size, with a well-defined, regularly branched symmetrical structure and a high density of functional end groups at their periphery. They contain large number of external groups suitable for multi functionalization. [26,27]

Cubosomes - Cubosomes are discrete, sub-micron, nanostructured particles of bi-continuous cubic liquid crystalline phase.[28] It is formed by the self-assembly of liquid crystalline particles of certain surfactants when mixed with water and a microstructure at a certain ratio. Cubosomes offer a large surface area, low viscosity and can exist at almost any dilution level. They have high heat stability and are capable of carrying hydrophilic and hydrophobic molecules.[29] Combined with the low cost of the raw materials and the potential for controlled release through functionalization, they are an attractive choice for cosmetic applications as well as for drug delivery.

Hydrogels - They are 3D hydrophilic polymer networks that swell in water or biological fluids without dissolving as a result of chemical or physical cross-links. They can predict future changes and change their property accordingly to prevent the damage. [30]

Buckyballs - Buckminster polymer networks that swell in water or biological fluids without dissolving as a result of chemical or physical cross-links. They can predict future changes and fullerene, C60, is perhaps the most iconic nanomaterial and is approximately 1 nm in diameter.

It has found its way into some very expensive face creams. The motivation is to capitalize on its capacity to behave as a potent scavenger of free radicals.[31]



Top Nanotechnology Cosmetic Products in the World

1) Sunscreens

No other cosmetic product has seen widespread use of nanotechnology as sunscreens. They are widely used on skin as a cream or a lotion to protect the skin from harmful effects of sun rays especially in the ultraviolet (UV) range. Severe UV exposure can lead to skin darkening, sun burns and in worse situations skin cancer. The most popular nanomaterials used in sunscreens are nanoparticulate Zinc oxide (ZnO) and Titanium dioxide (TiO2). These nanoparticles blocks both UVA and UVB rays from penetrating down to the deeper layers of skin providing broad spectrum sunscreen effect. Traditional sunscreens can be bulky, usually leaves a chalky layer on the skin and less stable to provide long term protection. However, due to the small size of ZnO and TiO2 nanoparticles, sunscreen remains transparent and less greasy. They are much more stable and have good aesthetic appeal. Lancôme and Dior have leading sunscreen products that contain nanotechnology. Find out more information about nano sunscreens. [7,8]

2) Anti-wrinkle products

Many factors can lead to wrinkling of the skin, including lack of nutrients, age, excessive use of chemicals on the skin, pollution, stress and over exposure to sun. The main reason for wrinkling of the skin is weakening of collagen structure in our skin triggered by any of the reasons noted above. Antiwrinkle products that are being made using nanotechnology is a big hit in the market today. The most popular brand in nanotechnology antiwrinkle products is L'Oreal, which markets an antiwrinkle cream named Ravtalift which contains nanosomes with Pro-retinol A; a modified version of vitamin A specially designed to deliver the nutrient to cells. Nanosomes can deliver the drug to the heart of the cell, leading to efficient absorption of the nutrient. These cosmeceutical compounds can improve the moisturizing effect while slowing down the collagen breakdown. Lancôme also markets a moisturizing antiwrinkle product by the name of Hydra zen cream which contains nutrients encapsulated in a nanosized particle. [7,8,10]

Nano ZnO based sunblock cream

RevitaLift : nanotechnology based antiwrinkle product

3) Skin moisturizers-

Human skin is the largest organ in the body. It's design to keep inside in and outside out. Dry skin can lead to many problems, including dry patches, loss of stretch, wrinkles and in worse cases premature aging. Nanotechnology based cosmetics use special nanoscale materials to form a humectant layer on top of the skin. This primarily limits the excessive water loss through the skin while keeping the skin moist due to constant moisturization by the humectant layer. The moisturization efficiency and longevity of nanotechnology-based skin moisturizers are much higher compared to conventional products. The most widely used nanomaterials are Liposomes, nano-emulsions and solid lipid nanoparticles. Lancôme and Nano-Infinity Nanotech are two cosmetic companies who have nanotechnology skin moisturizers in their product line.[8,10]

4) Hair care

Glowing beautiful skin is a major part of anyone's beauty and nanotechnology applications in hair care products have emerged as a promising field in future products. Cosmetic companies and institutes, have engaged in ongoing research to discovery new nanotechnology applications in major hair issues like, preventing hair loss and maintaining shine and health of the hairs. Especially nanoemulsions is emerging as a great method of delivering hair cosmetics to the inner structure of the hair fiber without damaging outer shell called cuticle due to their small size. Another very active area of nanotechnology hair care is sericin nanoparticles, primarily as a sealing agent for damaged hair to treat damaged cuticles.[7]

Nanoparticles used in hair care products -

1. Clay nanotubes, 2. Graphene-based nanosheets,
3. Clay halloysite nanotubes, 4. Cationic nanoemulsions.

5) Skin cleansers

Our skins are covered with a lipid film that is designed to protect us from pathogenic organisms like bacteria and fungi. However, due to the hydrophobic nature this film can also attract dirt and pollutants from the environment. It also can collect cellular debris and body oils secreted out from our skin. Sometimes, bacteria soap. naturally present on the skin can act on this nutrient media to produce body odor. Nanoemulsions and antibacterial nanoparticles have been used to prepare nanotechnology based cosmeceuticals that can provide better cleansing effects. Due to the small size of the nanoemulsions, they can reach deep layers and pores of the skin to provide a better cleaning action which is not possible with the conventional products. This can help eliminate skin problems like acne. Both organic and inorganic nanoparticles have been used in antibacterial skin cleaning products. They act on harmful bacteria from the skin to give further functional benefit than just cleaning the skin with [7]

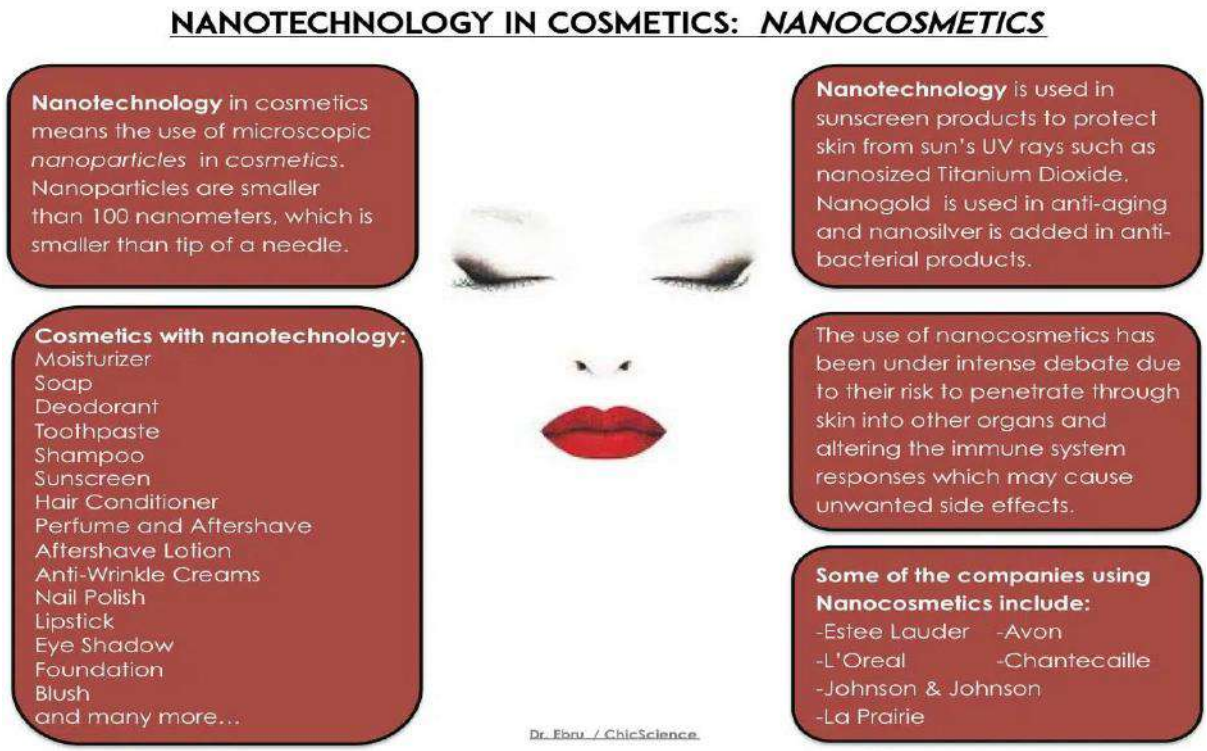
6) Nail care products: Nanoparticle nail polishes (from left): none, platinum, silver, gold-silver alloy, and gold. Nanoparticles repair the damaged, opaque, whitish and brittle nails normally diagnosed as leukonychia.

7) Lip care product: Lipstick, lip balm, lip gloss, and lip volumizer are examples of nano cosmeceutical lip care products. Lip volumizer with liposomes boosts lip volume, moisturizes and defines the lip, and smooths out wrinkles in the lip contour. Nanoparticles used in lip care products -

1. Dendrimer, 2. Liposome, 3. Nanocrystals, 4. Nano gold and nanosilver, 5. Niosomes, 6. Silica

8) Oral care - Nanosilver is one of the active ingredients used in the making of toothpaste. Silver contains antimicrobial properties. There are three main toothpaste ingredients that may be made of nano-sized particles:

Hydroxyapatite as cavity filler, Silver as bacteria killer, Titanium dioxide as whitener



BENEFITS OF NANOMATERIALS -

1. Protect the encapsulated active ingredients from degradation.
2. Provide better UV protection.
3. Nanomaterials are useful in the prevention and treatment of hyperpigmentation.

4. Long lasting effect.
5. Give texture and transparency to the cosmetic formulation.
6. Maintain hydration of skin by preventing moisture loss
7. Penetrate into deeper layer of skin
8. Avoid skin irritation and many more..

APPLICATION OF NANOPARTICLES IN COSMETICS

1) NPs as active ingredients in cosmetics :-

a) UV filters, b) Antibacterial and antifungal agents, c) Moisturizing and anti-aging nanomaterials, d) Cleansing Agents, e) Rheology modifiers,etc.

2) Nanoparticles as delivery vehicles :-

The use of nanomaterials as delivery systems is intended to improve the performance of the active ingredients.

d) a) Retinoids, b) Antioxidants, c) Enzymes, d) Peptides, e) Ceramides, f) Hyaluronic acid etc.

NANOTECHNOLOGY BENEFITS AND IMPACT ON SKINCARE -

Because of its effective approach to skincare, manufacturers are using nanotechnology in a wide range of cosmetics. According to studies, the average adult uses nine cosmetic products per day. These are among the most widely used products in the world and are available in a variety of forms these days, but not all of them are effective and provide long-term results. Nanotechnology's minute size makes it ideal for any skincare routine or requirement. As a result, it is preferred for almost any product that must specifically remove the problem from its roots. Issues such as dark circles and puffy eyes necessitate the most extensive use of nanotechnology to restore the freshness of the targeted area like never before.

PENETRATION OF NANOPARTICLES VIA SKIN –

Scientific studies have shown that nanoparticles can penetrate skin, especially if skin is flexed.[16] Broken skin is a direct route for the penetration of particles even up to a size of 7000 nm. The presence of acne, eczema and wounds may enhance the absorption of nanoparticles into the blood stream and may lead to further complications. A preliminary study found that nanoparticle penetration was deeper in skin affected by psoriasis than in unaffected skin.[17] Recently, the base carriers are being modified in order to enhance the skin penetration by incorporating certain penetration enhancers, both physical and chemical, and also by preparing newer vesicular systems with increased skin penetrability like ethosomes and transferosomes. Even flexing and massage can increase the skin penetration of nanoparticles. One study found that even particles up to 1000 nm in size can be taken up through intact skin to reach living cells, when skin is flexed.[18]

Food and Drug Administration (FDA): Guidance for Industry Safety of Nanomaterials in Cosmetic Products

This document provides guidance to industry and other stakeholders on the FDA's current thinking on the safety assessment of nanomaterials in cosmetic products. or altered properties, data needs and testing methods should be evaluated to address any unique properties and functions of the nanomaterials used in the cosmetic products. The FDA recommends that the safety assessment of cosmetic products using nanomaterials address several important factors, including:

- The physicochemical characteristics,
- Agglomeration and size distribution of nanomaterials under the conditions of toxicity testing and as expected in the final product,
- Impurities,

- Potential routes of exposure to the nanomaterials,
- Potential for aggregation and agglomeration of nanoparticles in the final product,
- Dosimetry for in vitro and in vivo toxicology studies, and
- In vitro and in vivo toxicological data on nanomaterial ingredients and their impurities, dermal penetration, potential inhalation, irritation (skin and eye), sensitization studies, and mutagenicity/genotoxicity studies.

- Conclusions and Future Direction

Currently, nanotechnology is regarded as a promising and revolutionizing field and is being utilized and appreciated in the areas of cosmetics, cosmeceuticals, dermatology,biomedical applications, etc. The introduction of newer advancements and novel drug delivery systems make cosmetics and cosmeceuticals more popular with increased market share. Today, these cosmetics are an indispensable part of the daily routine; further, the introduction of nanotechnology to cosmetics has enhanced its acceptance among users all around the world. However, its associated toxicity owing to its penetrability is a major concern that is often overlooked, leading to adverse health issues. Presently, novel nanocarriers such as liposomes, ethosomes, cubosomes, NLC, SLNs, nanoemulsions, niosomes, etc.,are exploited to formulate various cosmetics and cosmeceuticals with enhanced outcomes. Nanosystems carry and deliver these formulations across the skin by diverse mechanisms and impart several functions, such as sun protection, moisturization, wrinkle reduction, etc. Even though these nanomaterial products are gaining impressive market value, there is tremendous debate concerning their safety and toxicity in humans, demanding more careful investigations. Hence, the cosmetic legislation should provide a specific list of references as well as the ingredients that produce unintended environmental effects for all users of cosmetic products, such as consumers and professional users, thus ensuring the safety of the usage of cosmetic products. With so much claimed possibilities of nanoparticles due to their improved properties, the rush to applying them in cosmetic preparation is on the increase and the market is already flooded with so much “nano-enhanced” skin formulations.

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Cutaneous Benefits of Snow Mushroom in Cosmetics

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Abstract:
Tremella fuciformis, also known as snow mushroom, is an edible mushroom that has historically been popular in herbal and Asian medicine and cuisine. Also have excellent mosisturizing and antioxident properties. The main polysaccharide ingredients have been extracted and used as treatment in a variety of conditions, demonstrating positive effects in a range of biological functions including those involved in antioxidation, antitumor, antidiabetic and immunomodulatory, Studies have demonstrated the role this extract may play in skin antiaging, photoprotection, wound healing, and barrier protection. Most studies have been limited to in vitro and in vivo animal models. Future clinical research is needed to further understand the role of T. fuciformis in dermatology. This review will discuss the existing research findings and potential future applications for T. fuciformis as a treatment in skin conditions.
Keywords: Snow Mushroom, antioxidents, Anti-Aging, Photoprotection, wound healing.

Introduction:
Mushroom have been valued as a tradition source of natural bioactive compounds and recently been exploited for potential component in cosmetics industry. The Tremella fuciformis typically called as 'yin er' silver ear fungus, snow mushroom and white jelly mushroom and was discovered in china. Termella polysaccharides are major components in Termella fuciformis. The name mushroom refers to a fruiting body, formed by several hyphae that grow upwards and produce spores basidiospores.

Snow Mushroom in Fig no. 1

History
The article titled 'Research on tonic Termella fuciformis' in journal of Natural History, 1914 . Cultivation techniques began in 1968. Snow mushrooms are native to subtropical and tropical regions around the world including China, Japan and in other Asian countries.

Technical aspects:
INCI: Tremella Fuciformis Sporocarp Extract
Appearance: White or light yellow
Ph: 5.5-7.5
Odour: Characteristic

Solubility: Water soluble
COSMOS-approved: yes
Ecocert: yes
Bioactives:

- Polysaccharides
- Phenolic Compounds
- Flavonoids
- Vitamine D
- Fatty acids
- B-glucan

Dry Skin Sings ,Causes and problems sings:

- Itching
- Scaling
- Redness

Causes

Bath, swimming and showers. Freaquent showering with hot water breakdown the lipid layer. Same change with heavy chlorinated.
Harsh Scrubbing of Skin.
Tight clothing or compression. Tight clothing and compression can incresses the risk of dry skin. And worsen the condition of dry skin.

Problems of Dry Skin:

Xerodermatic

Xeroderma, is a skin condition characterized by excessively dry skin.
Detergents such as washing powder and dishwashing liquid can cause xeroderma.

Desquamation

Desquamation is the natural process in which skin cells are created, sloughed away, and replaced.
Nonpathologic visible desquamation can be observed after immersion of the skin in warm or hot water.
In pathologic desquamation, such as that seen in the Startum Corneum become thicker, imparting a “dry” or scaly appearance to the skin
Wrinkles A wrinkle, also known as a is a fold, or crease in otherwise smooth surface, such as on skin sun damage, smoking and poor hydration . Age wrinkling in the skin is promoted by habitual facial expressions, aging.

NMF:

In addition to keratin, which can bind a substantial amount of water, the stratum corneum contains a number of other hydrophilic agents. These materials are called natural moisturizing factors (NMF).
The NMF concentration varies as a function of age and skin depth.
The conversion of filaggrin to NMF occurs as the corneocytes are moving to the stratum corneum it is possible for the outermost skin layers to maintain an adequate water supply when exposed to dry environments
Althought the corneocytes are biologically dead, but biochemically they are active there are number of enzymes in corneocytes and they need nmf for example when desqumation occur these enzyme weaken the forces hold coreneocytes and they need water that NMF provide for desqumation process.

Cosmetics Uses:

Moisturizing effect: The water content in stratum corneum is important factor in appearance and function of skin. T. fuciformis polysaccharides have been developed for use in cosmetics on account of their excellent skin moisture retention. The polysaccharide isolated from a hot

water extract of a Tremella mushroom without adding a chemical reagent was found to have a novel effect of inhibiting melanin formation effects and lightening the spots on the skin when applied to the skin.

It is used in Cream, Lotions, Serums, and facial preparations as Cosmetics ingredients.It shows Anti-inflammatory and wound healing properties used in cosmetics. Termella fuciformis are loaded with Vitamine D which also helps makes it great for healing acne lesions.

Snow Mushroom VS. Hyaluronic Acids:

Benefits of Hyaluronic acid

Hyaluronic acid is already present in our body that fills out, plumps up and firms our skin thereby keeping it free of wrinkles and fine lines.

Hyaluronic acid can hold up to 1,000 times its weight in water, which helps to hydrate the skin.

Benefits of Snow mushroom

Tremella generates a natural flexible hydration film on the skin that reinstates dry skin to its op timally hydrated and mobile state, enabling it to develop elasticity and a fit appearance.

The polysaccharide from Tremella Mushroom was applied to skin topically.

The water holding capacity of the skin and horny layer was greatly improved and had better results than the Hyaluronic Acid control.

Extraction Processes:

In the past most studies have focused on the extraction of polysaccharides from the fruiting body and mycelium.

The generally adopted polysaccharide extraction method is to stir the pulverized fruiting bodies in hot water for several hours.

After the extraction method is selected, the supernatant is collected by centrifugation or filtration, and the residue is generally extracted three-times.

The precipitate is collected by centrifugation and then freeze-dried or low temperature dried to obtain crude polysaccharide.

Conclusion:

Antioxidant will protect against free radical damage for the prevention of various diseases and aging. A significant advantage in antioxidant compounds extraction from mushrooms is that fruit bodies or mycelium can be manipulated to produce active compounds in a relatively short period of time. Snow mushroom extract is a moisturizing ingredients which is capable of moisture retention on the skin and the small particles penetrate more easily to the skin than hyaluronic acid. There are various products in the market that uses the power of Snow mushroom extract to get better result to the consumer.

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Formulation And Development of Skin Lightening Cream using Daisy flower Extract

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ABSTRACT
Skin Lightening cream using daisy flower extract is usefull in lightening skin tone or simply to obtain a lighter skin tone as whiter skin may be synonymous of wealth, health,youth and beauty in different culture. Also this cream gives the properties of Moisturising, Nourishing and the treatment of hyperpigmentation to the skin.
The cream is naturally safe as it contains natural flower extract, easy to formulate making it attractive in the cosmetic field. The cream is non-irritating, non allergic and non-toxic.
KEYWORDS
Skin Lightening preparation, Bellis perennis, melanin inhibition, melanosome uptake.

1.INTRODUCTION

INCI Name: - Bellisperennis (Daisy) Flower
Kingdom: Plantae – Plants
Botnical name: - Bellisperennis
Family: – Asteraceae or Compositae
Genus – Bellis L Aster
A brighting agent it is the perfect Natural solution for sunspot. The Daisy Flower is rich in antioxidants, along with malic acid & tartaric Acid this acid assists the skin firming and effective fighting the wrinkle. Daisy flower use on its own is the wonder fully scented and healing moisturizer also excellent for both the skin & hair.
It is effective in skin whitening and improving pigmentation. Other beneficial substances in daisy flower are antioxidants, malic, and tartaric acids (skin-firming natural acids).

2) MATERIALS AND METHOD

Sr.No	Materials	Properties
1	Bellisperennis (Daisy) Flower	Lightening agent
2	EDTA-disodiumsalt	Chealating agent
3	Propylene glycol	Humactant
4	Sodium benzoate	Preservative and anti corrosive
5	Liquid paraffin (light)	Hydrating and cleansing agent
6	Stearic acid	Thickening Agent
7	Cetostearyl alcohol	Opacifyingagent,surfactant-foam booster,viscosity increaser
8	Glyccerylmonosterate	Thickening, Emulsifying, Anti-Sticking, Dispersing.
9	Potassium hydroxide	pH adjuster
10	Iso propyl myristate	Emollient, Thickening Agent
11	Perfume	Lavender

Table no. 1s

Method of preparation of cream base

Ingredients	Base Formulation code				
	F1	F2	F3	F4	F5
EDTA-disodiumsalt	0.01	0.02	0.03	0.04	0.05
Propylene Glycol	1	2	3	4	5
Sodium Benzoate	0.40	0.40	0.40	0.40	0.40
Liquid paraffin	0.3	0.5	1	1.5	2
Stearic acid	9	10	11	12	13
Cetostearyl alcohol	0.3	0.5	0.7	0.8	1
Glyccerylmonosterate	0.3	0.5	0.7	0.8	1
Potassium hydroxide	0.3	0.4	0.5	0.6	0.7
Iso propyl myristate	0.3	0.5	1	0.5	2
Water	Up 100	Up 100	Up 100	Up 100	Up 100

F- Formulation Table no. 2

preparation of cream base:

Step I: water phase was prepared by collecting distilled water and the water was remove aside from this for final volume makeup. Take EDTA- disodiumsalt, water soluble components methyl paraben, propyl, Glycerine, Pot hydroxide were dissolved in distilled water; heated up to 70-75^{0}c.

Step II: Oil phase was prepared by heated propyl paraben, stearic acid, sunflower oil, cetostearyl alcohol, Glycerylmonostarate, Isopropyl Myristate heated up 70-80^{0}c.

Step III: Oil phase was added in water phase at 80^{0}c with continuous stirring for 20-25min and then it was homogenized till uniform emulsion is formed.

The finished products have cream like consistency.

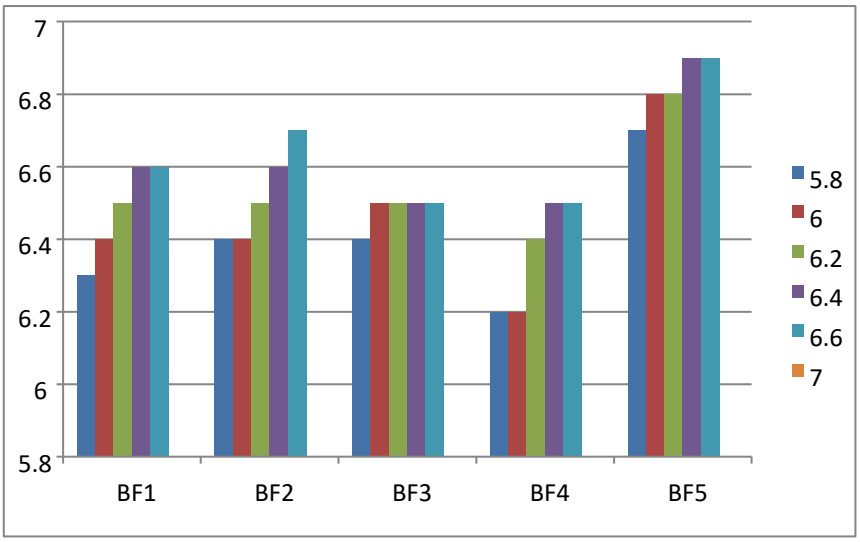
Base formulation i.e. F5 were selected for further studies.

3)EVALUATION OF BASE FORMULATION

a)Result of pH base formulation

Sr no.	Days	Base Formula code				
		BF1	BF2	BF3	BF4	BF5
1	0	6.3	6.4	6.4	6.2	6.7
2	7days	6.4	6.4	6.5	6.2	6.8
3	15days	6.5	6.5	6.5	6.4	6.8
4	21days	6.6	6.6	6.5	6.5	6.9
5	30days	6.6	6.7	6.5	6.5	6.9

BF- Base Formulation Table no.3



Discussion: From the above table the result was found that pH formulation BF4 and BF4 is desirable range. But BF1, BF2, BF5 have higher range.

b) Determination of Viscosity

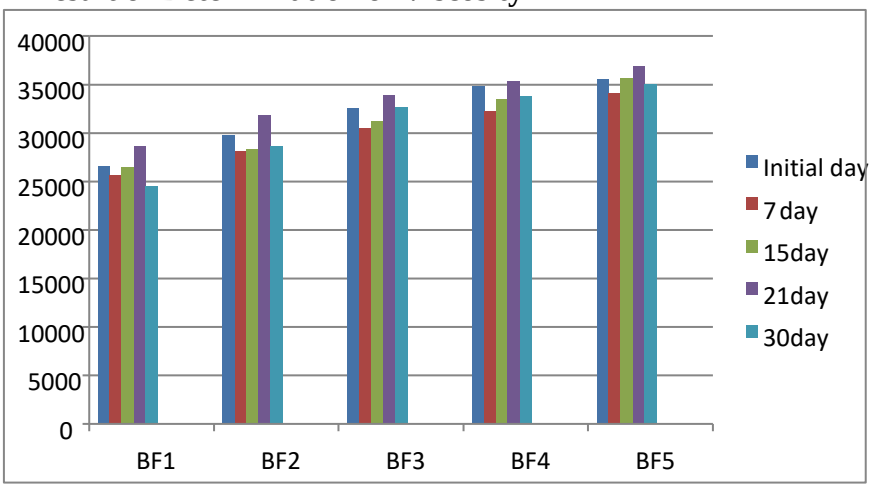
Viscosity test was performed for base formulation

Result of Determination of Viscosity

Sr. no	parameters	Base formulation code				
		BF1	BF2	BF3	BF4	BF5
1	Initial	26580cps	25670cps	26500cps	28560cps	24540cps
2	7 Days	29730cps	28130cps	28340cps	31760cps	28590cps
3	15 Days	32540cps	30450cps	31200cps	33890cps	32640cps
4	21 Days	34820cps	32240cps	33450cps	35340cps	33810cps
5	30 Days	35550cps	34100cps	35660cps	36900cps	34980cps

Table no.4

Result of Determination of Viscosity



Discussion: According toobservation the formulation BF5 was good viscosity like lightening cream, formulation BF1, BF2, BF3, BF4 low viscosity as compare to BF5.

c) Stability study of base formulation

Formulation code	Physical Characteristics		
	Colour	Consistency	Feel
Initial day			
BF1	White	No Change	Smooth
BF2	White	No Change	Smooth
BF3	White	No Change	Smooth
BF4	White	No Change	Smooth
BF5	White	No Change	Smooth
After 7 days			
BF1	Pale White	Change	Smooth
BF2	White	Change	Smooth
BF3	White	No Change	Smooth
BF4	White	Change	Tacky
BF5	White	No Change	Smooth
After 14 days			
BF1	Pale White	Change	Smooth
BF2	White	Change	Tacky
BF3	Pale White	Change	Smooth
BF4	White	Change	Tacky
BF5	White	No Change	Smooth

Table no.5

Discussion: From the above observation, the formulation BF5 was stable into room temperature, other formulation i.e.BF1, BF2, BF3, BF4 change its viscosity.

d) Skin Irritation Test

parameter	Formulation				
	BF1	BF2	BF3	BF4	BF5
Skin Irritation	NI	NI	NI	NI	NI

NI- No Irritation

After applying cream base there was no irritation or redness even after 48 hours. Hence, the all formulation was non-irritant and safe for human being.

Discussion: For the all above observations the formulation BF5 was selected as final formulation of active incorporation because it showed good result for pH, thermal stability, stability study, viscosity, skin irritation test, etc.

Final Selection of Base Formulation

Ingredients	Base Formulation (BF5)
EDTA-disodiumsalt	0.05
Propylene Glycol	5
Sodium Benzoate	0.40
Liquid paraffin	2
Stearic acid	13
Cetostearyl alcohol	1
Glyccerylmonosterate	1
Potassium hydroxide	0.7
Iso propyl myristate	2
Water	Up 100

Table no.6

4)Evaluation of Active cream base

Component%	Ingredients	Active Formulation code				
Active Ingredients	Daisy flower oil	AF1	AF2	AF3	AF4	AF5
		1	2	3	4	5
Oil Phase	Steric acid	13	13	13	13	13
	Cetosteryl Alcohol	1	1	1	1	1
	Glycerylmonostarate	1	1	1	1	1
	Iso Propyl Myristate	2	2	2	2	2
	Liquid paraffin	2	2	2	2	2
Water Phase	Disodium EDTA	0.05	0.05	0.05	0.05	0.05
	Potassium hydroxide	0.7	0.7	0.7	0.7	0.7
	Propylene glycol	5	5	5	5	5
	Sodium Benzoate	0.40	0.40	0.40	0.40	0.40
	Perfume	0.1%	0.1%	0.1%	0.2%	0.2%

Table no.7

Evaluation of Active base cream formulation

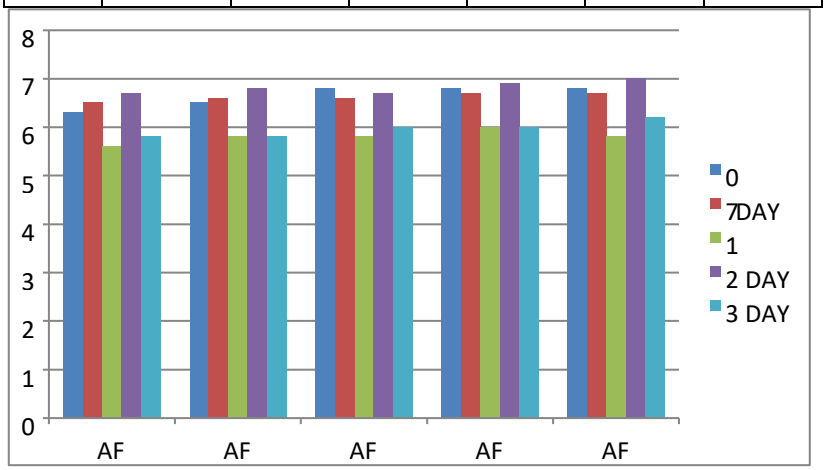
Different concentration of active ingredients was added selected base formulation i.e. Active base formulation and optimized. Based on the results the best formulation BF5 was selected for further dissertation work.

a) Determination of pH:

Result of pH base formulation

Table no.8

Sr no.	Days	Active Formulation code				
		AF1	AF2	AF3	AF4	AF5
1	0	6.3	6.5	5.6	6.7	5.8
2	7days	6.5	6.6	5.8	6.8	5.8
3	15days	6.8	6.6	5.8	6.7	6
4	21days	6.8	6.7	6	6.9	6
5	30days	6.8	6.7	5.8	7	6.2



Result of pH base formulation

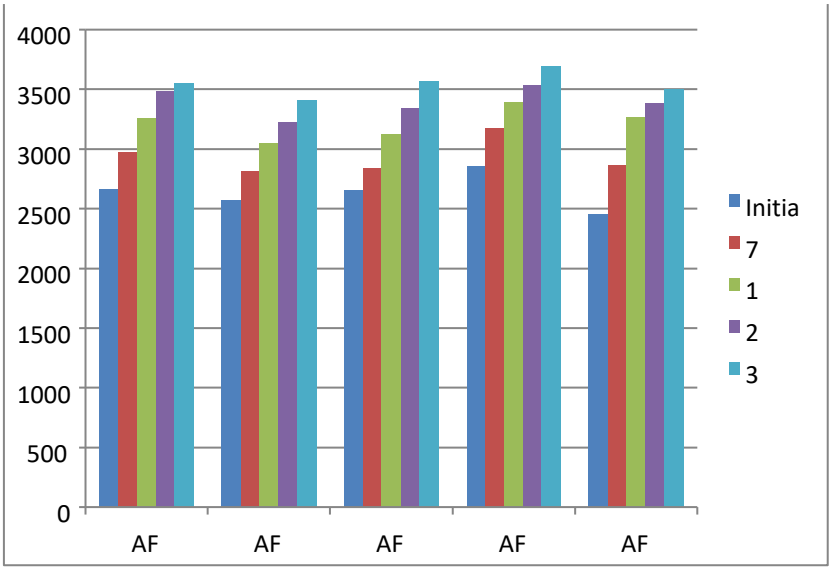
Discussion: From tableAfter 8 days pH of AF1,AF5 decreases, AF2,AF4 increases, no change observed in AF3 observed after 15,21 and 30days the pH of AF2,AF4,have more pH which can irritation to skin, AF1 also more pH than normal skin pH, AF3and AF5moreto normal pH range. Hence AF5 was selected.

b) Determination of Viscosity

Result of Determination of Viscosity

Sr. no	parameters	Base formulation code				
		AF1	AF2	AF3	AF4	AF5
1	Initial	26580cps	25670cps	26500cps	28560cps	24540cps
2	7 Days	29730cps	28130cps	28340cps	31760cps	28590cps
3	15 Days	32540cps	30450cps	31200cps	33890cps	32640cps
4	21 Days	34820cps	32240cps	33450cps	35340cps	33810cps
5	30 Days	35550cps	34100cps	35660cps	36900cps	34980cps

Table no.9



Result of Determination of Viscosity

Discussion: According to observation the formulation AF5 was good viscosity like lightening cream, formulation AF1, AF2, AF3, AF4 low viscosity as compare to AF5.Hence, AF5 was selected.

c) Stability study

Table no .3.1 physical characteristics of skin lightening cream

Formulation code	Physical Characteristics		
	Colour	Consistency	Feel
Initial day			
AF1	White	No Change	Smooth
AF2	White	No Change	Smooth
AF3	White	No Change	Smooth
AF4	White	No Change	Smooth
AF5	White	No Change	Smooth
After 1 week			
AF1	Pale White	Change	Smooth
AF2	Pale White	Change	Smooth
AF3	Pale White	No Change	Smooth
AF4	White	Change	Tacky
AF5	White	No Change	Smooth
After 2 week			
AF1	Pale White	Change	Smooth
AF2	Pale White	Change	Tacky
AF3	Pale White	Change	Smooth
AF4	White	Change	Tacky

AF5	White	No Change	Smooth

Table no.10

d) Skin Irritation Test

Skin Irritation of Active base formulation

parameter	Formulation				
	AF1	AF2	AF3	AF4	AF5
Skin Irritation	NI	NI	NI	NI	NI

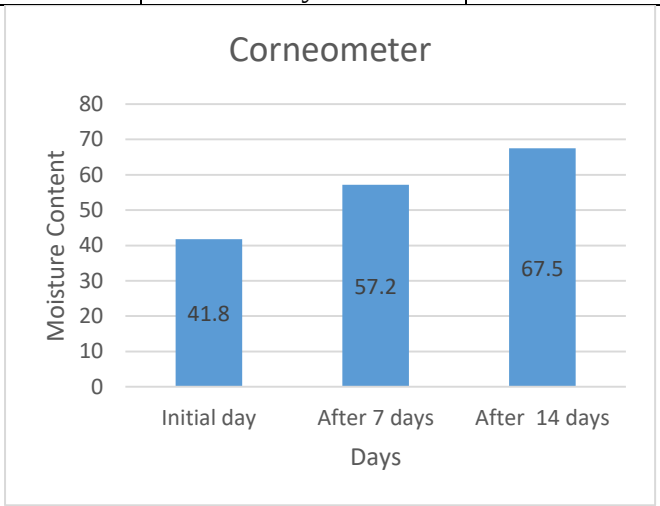
NI- No Irritation

Discussion: After applying cream base there was no irritation or redness even after 48 hours. Hence, the all formulation was non-irritant and safe for human being.

e) Determination of moisturising property by using Corneometer

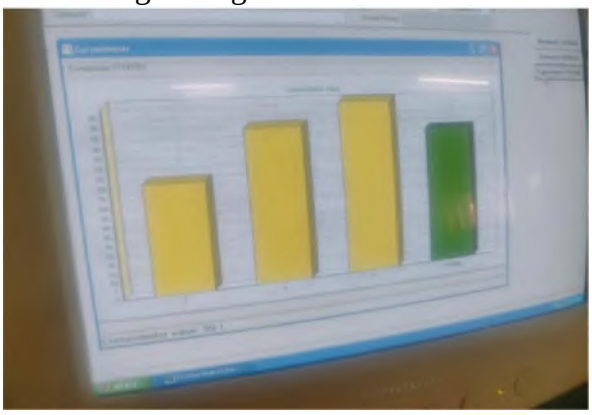
Table no. Analysis of moisturising activity by using Corneometer

Sr. No.	Time Interval	% of moisture Content
1	Initial day	41.8
2	After 7 days	57.2
3	After 17 days	67.5



Result of Moisture Content Using Corneometer

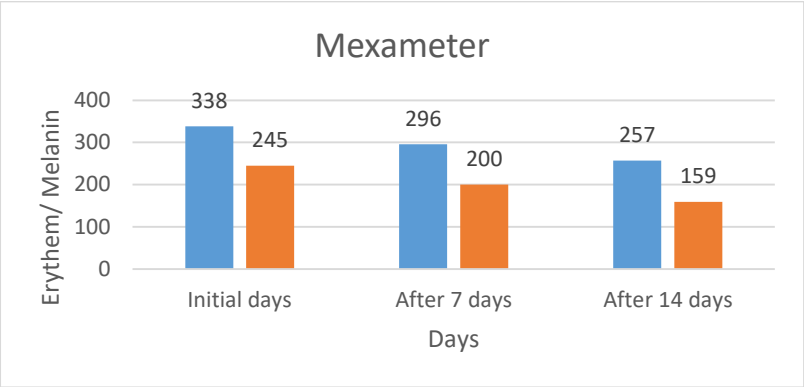
Discussion: The moisturising activity was carried out by using corneometer. It was observed that befor application of skin lightening cream , the moisture content of skin was less and after application of skin lightening cream moisture content was increased.



f) Determination of melanin by using Mexameter

Analysis of melanin by using mexameter

Sr.no	Time Interval	Melanin value	Erythem value
1	Initial day	245	338
2	After 7 days	200	296
3	After 14 days	159	257



Discussion: The melanin content was carried out by using mexameter. It was observed that befor application of skin lightening cream , the melanin content of skin was more and after application of skin lightening cream melanin content was decreased.

Base Formulation **Active Base formulation**

g) Photographic Evaluation

Photographic Evaluation was carried out for the formulation AF5.The study was done on volunteers for 21 days and photographic were taken after initial day, 7 days, 15 days and 21 days.

Initial day **After 7 days**

After 15 days **After 21 days**

Determination of photographic evaluation.

Discussion : For the picture shown in figure it was observed that, and Daisy Flower Oil proves itself as skin lightening agent.

Discussion: For the all above observations the formulation AF5 was selected as final formulation because it showed good result for pH, Viscosity, stability study, skin irritation test, decreased amount of melanin, microbial activity, etc.

Final selection of Active Formulation

Ingredients	Active Formulation (AF5)
EDTA-disodiumsalt	0.05
Propylene Glycol	5
Sodium Benzoate	0.40
Liquid paraffin	2
Stearic acid	13
Cetostearyl alcohol	1
Glyccerylmonosterate	1
Potassium hydroxide	0.7
Iso propyl myristate	2
Water	74.85

5) CONCLUSION

Skin lightening cream AF5 which was formulated showed a good physical characteristics, pH, spreadability, stability parameters like visual appearance, nature, viscosity, Absorption study, Hence, this study showed that AF5 was the best formulation for skin lightening cream.

The external application of skin lightening protective cream was studied at different time interval for 15 days on volunteers and is was observed that as compared to marketed cream it give better result. Photographic evaluation shows the improvements in skin tone, color and good moisturizing and softness of skin compared with standard skin lightening cream and protective cream. From the present work study it can be concluded that it is possible to develop creams containing herbal extract having good lightening property and to protect skin. Form the result obtained in the study we can positively conclude that Daisy Flower Oil have significant lightening property.

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Formulation and Development of Gel Shampoo Using Grape Seed Extract

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Abstract :
The goal of the present study was to formulate and evaluate the hair gel shampoo which would help to reduce the dandruff and dermatitis, to remove dirt and dust, to nourish the hair scalp etc. The present study was conduct with a view to formulate and evaluate on removing dust, dirt, sebum, greasiness, oil from scalp n hairs of given formulation by using grape seed extract. The gel shampoo was prepared with natural active agents. The concentration of active agent were kept in range of 0.5 %, 1%, 2%, each were incorporated and three combination of each were prepared and evaluated on different stability parameters like Ph ,viscosity, appearance, feel, and efficacy was evaluated by subjective evaluation and formulation F3 with 1% Grape seed extract was passed all the stability parameters and also approved by the volunteers and it was really helps to reduce hair loss and restart the hair growth again. Grape seed extract was selected as reducing hair loss, dandruff and dermatitis and incorporated them in the cosmetic product i.e. shampoo
Grape seed extract has a characteristic tocopherol and tocotrienol content. In addition to exhibiting vitamin e activity, tocopherols occur in seed extract, such as α-, β-, γ-, and δ-tocopherol, with γ-tocopherol as one of the most potent antioxidants Grape seed oils are richer in tocotrienols (unsaturated forms of vitamin E) than tocopherols though, among which γ-tocotrienol is the most abundant, followed by α-tocotrienol.α-tocopherol and γ-tocotrienol have been reported to show the highest variability between grape varieties. They also contain phenolic compound, flavonoid, and vitamin A.
In other words, it was always kept in mind to developed value-added cosmetic products containing blood circulation, increase and promoters or product having multi-functional benefits to the client. Thus, choice of actives was very crucial and even choice of the product category to effectively deal the problem. Grape seed extract was selected as reducing hair loss, dandruff and dermatitis and incorporated them in the cosmetic product i.e. shampoo.
Key words: Grape seed extract, Tocopherilic acid,Hair growth cycle, Tocopherol, Hair matrix keratinocytes, Gel shampoo.

1.Introduction

Cosmetics are substances or products used to enhance or alter the appearance of the face or fragrance and texture of the body. Many cosmetics are designed for use of applying to the face, hair, and body. They are generally mixtures of chemical compounds; some being derived from natural sources (such as coconut oil), and some being synthetics or artificial.[1] Cosmetics applied to the face to enhance its appearance are often called make-up or makeup. Common make-up items include: lipstick, mascara, eye shadow, foundation, blush, and contour. Whereas other common cosmetics can include skin cleansers, body lotions, shampoo and conditioner, hairstyling products (gel, hair spray, etc.), perfume and cologne.

In the U.S., the Food and Drug Administration (FDA), which regulates cosmetics,[2] defines cosmetics as "intended to be applied to the human body for cleansing, beautifying, promoting attractiveness, or altering the appearance without affecting the body's structure or functions". This broad definition includes any material intended for use as a component of a cosmetic product. The FDA specifically excludes pure soap from this category

Shampoo is a hair care product, typically in the form of a viscous liquid, that is used for cleaning hair. Less commonly, shampoo is available in bar form, like a bar of soap. Shampoo is used by applying it to wet hair, massaging the product into the hair, and then rinsing it out. Some users may follow a shampooing with the use of hair conditioner.

The typical reason of using shampoo is to remove the unwanted build-up of sebum in the hair without stripping out so much as to make hair unmanageable. Shampoo is generally made by combining a surfactant, most often sodium lauryl sulfate or sodium laureth sulfate, with a co-surfactant, most of cocoamidopropylbetaine in water.

Specialty shampoos are available for people with dandruff, color-treated hair, gluten or wheat allergies, an interest in using an organic product, and infants and young children ("baby shampoo" is less irritating). There are also shampoos intended for animals that may contain insecticides or other medications to treat skin conditions or parasite infestations such as fleas.

For individuals suffering from Androgenetic Alopecia (also known as male-pattern baldness), the sex hormone DHT leads to the miniaturization. As the follicles become smaller, the hair cycle shortens.

Grape seed extract, however, has actually been proven to jumpstart the hair cycle and push the follicle from the telogen phase (the phase in which the most hair is lost) to the anagen phase (the phase in which active hair growth occurs).Grape seed Extract Is Full of Antioxidant. Grape seed Extract is Antibacterial. Scientists have been researching treatment options for years, and in 2010, researchers found that grape seed extract is actually an effective treatment for this debilitating, and sometimes fatal, bacterium. If grape seed extract is effective at treating MRSA, then surely it's an effective treatment for a num. Individuals suffering from Androgenetic Alopecia (also known as male-pattern baldness), the sex hormone DHT leads to the miniaturization. As the follicles become smaller, the hair cycle shortens. To give cleansing effect to hairs. Removes dirt and dust from hairs. Also removes greasiness and sweat from scalp.

2.Material and Methods :

In this gel shampoo formulation Carbopol-940 was the main ingredient used as a gelling agent with Triethanolamine as a neautralizer,both gave body to the product,then glycerine as a humectants and SLS,CAPB were used for cleansing property which was essential property in the shampoo. Along with these basic raw materials Grape Seed Extract was used as active agent which was reported to have antioxidant property and reduces hair fall was procured for the present study from RP chemicals Kalyan, Mumbai, India, along with Certificate of Analysis. The procured sample was validated for parameters such as Color, assay, pH, specific gravity, water content, tocopheryl content.

2.1 Formulation and optimization of base formulation

In any cosmetic preparation it is important to have stable formulation before incorporation of active. The effectiveness and stability depends upon the compatibility of active ingredients

2.1.1 Formulation of gel shampoo

Procedure-

Firstly weighed all the wet and dry ingredients accurately. After that heat the demineralized water up to 80 to 85 ^{0}C. Clean all the equipments properly. After that heat demineralized water and add citric acid and EDTA one by one. Dissolve carbomer properly in main batch and add Sodium Lauryl Ether Sulphate,Coca Amido Propyl Betaine,glycerin and cocodiethanolamine in the beaker n start low stirring with the help of stirrer.

2.1.2 Formulation of gel shampoo base:

Table No.1: Formulation of shampoo base

Sr. No	Ingredient	F1 For100%	F2 For 100%	F3 For100%
1.	Water	68.1	66.8	**65.3**
2.	Carbopol 940	2	1.50	**2**
3.	Triethanol amine	1	0.30	**0.30**
4.	Glycerin	3	3	**3**
5.	Disodium EDTA	0.10	0.10	**0.10**
6.	Sodium Lauryl Ether Sulphate	20.00	21	**22**
7.	CAPB(Coca Amido Propyl Betaine)	5	5	**5**
8.	Citric Acid	0.10	0.10	**0.10**
9.	CDEA (cocodiethanolamine)	0.5	2	**2**
10.	Methyl Paraben	0.20	0.20	**0.20**

Optimization of gel shampoo base:

Sr. No.	Parameters	Qty for 100 gm		
		F1	F2	**F3**
1.	Appearance	++	+++	**+++**
2.	Color	+++	++	**+++**
3.	Odour	+++	++	**+++**
4.	Spreadability	++	+++	**+++**
5.	Feel	++	+++	**++**
6.	Consistency	+	++	**++**

Abbreviations:- + - Good, ++ - Better, +++ - Best.

From the above observation F1, F2, F3, the F3 have the most desirable properties and was stable. So the F3 formulation is selected for the further experimentation. Therefore extracts with different concentration will be added to this formulation.

2.2 Incorporation of Grape Seed Extract at different concentration in base formulation:

Table no.2 : Incorporation of Grape Seed Extract at different concentration in base formulation.

Sr No.	Ingredients	F1 For 100%	F2 For 100%	F3 **For 100%**
1	SLES(Sodium Lauryl Ether Sulphate)	20.00	21.00	**22.00**
2	CAPB(Coca Amido Propyl Betaine)	5.00	5.00	**5.00**
3	Citric Acid	0.10	0.10	**0.10**
4	Disodium EDTA(Ethylene DiamineTetraacetic Acid)	0.10	0.10	**0.10**
5	CDEA (cocodiethanolamine)	5.00	2.00	**2.00**
6	Glycerin	3.00	3.00	**3.00**
7	Carbopol-940	2.00	1.50	**2.00**
8	Methyl paraben	0.20	0.20	**0.20**
9	Perfume	0.30	0.30	**0.30**
10	TEA(Triethanolamine)	0.30	0.30	**0.30**
11	Grape seed Extract	0.50	1.50	**1.00**
12	Water	68.1	66.8	**65.3**

2.3 Analysis of Gel Shampoo with Grape Seed Extract:

Gel shampoo formulation (i.e F3 with 1% Grape seed extract) was subjected to study the parameters like Appearance, Color, Odor, Consistency, Thermal stability, pH, Foam height removefficacy, moisture retention by corneometer and Skin irritation.

2.4 Stability Study:

The sample of gel shampoo was kept at 5ºC, room temperature and at 40ºC. The changes in physical appearance, colour etc were studied.

Table no. 3 : Stability studies of gel shampoo

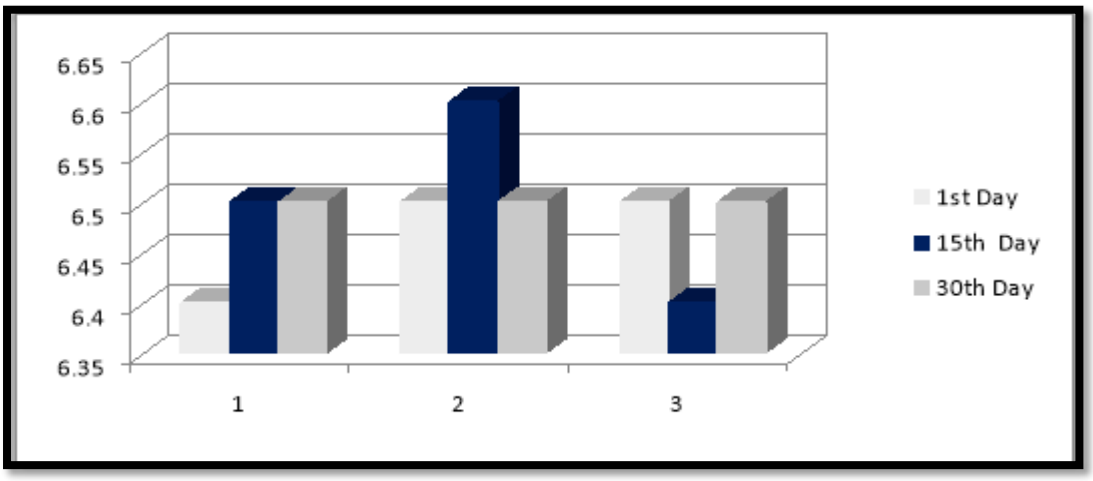
Sr. No	Parameters	F1	F2	F3
1	Appearance	Opaque	Opaque	Opaque
2	Colour	Clear	Clear	Clear
3	Spredability	Good	Good	Very Good

Determination of pH:

Determination of pH of gel shampoo incorporated with grape seed extract:

Table no.4 : Determination of pH of gel shampoo incorporated with grape seed extract

Time interval	F1	F2	F3
1st Day	6.4	6.5	6.5
15th Day	6.5	6.6	6.4
30th Day	6.5	6.5	6.5



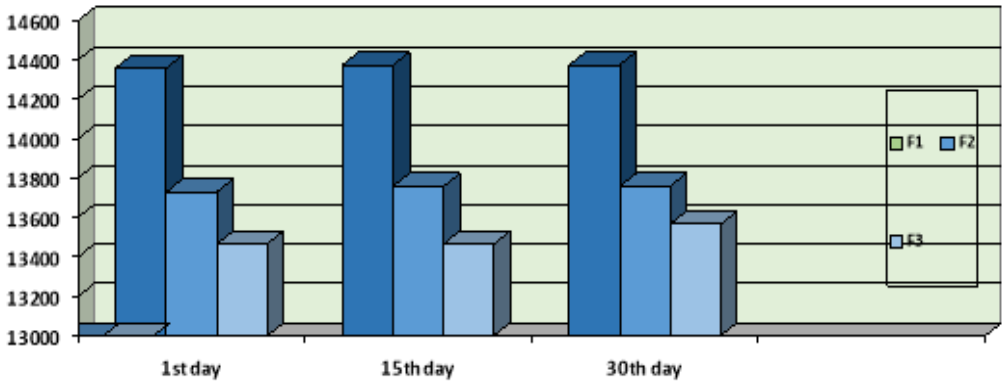
Graph no 1: Graphical representation of pH of gel shampoo with grape seed extract.

Determination of Viscosity: Determination of viscosity for gel shampoo incorporated with grape seed extract:

Table no. 5 : Determination of viscosity for gel shampoo incorporated with grape seed extract

Sr.no.	No. of days	F1	F2	F3
1	**1st day**	13360	13730	13466
2	**15th day**	14375	13759	13466
3	**30th day**	14375	13759	13567

Graph no. 2 : Graphical representation of viscosity of gel shampoo with grape seed extract

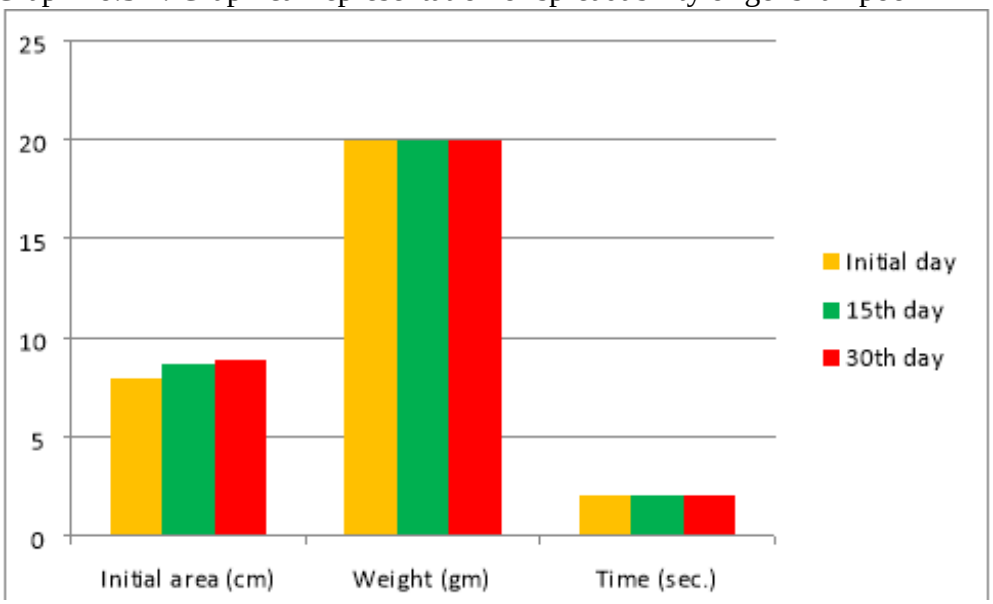


Determination of spreadability

Table no. 6 : Determination of spreadability

Sr.no.	Days of interval	Initial area	Weight	Time
1	Initial day	8cm	20gm	2.1 sec
2	15th day	8.7cm	20gm	2.1 sec
3	30th day	8.9cm	20gm	2.1 sec

Graph no.3 : Graphical representation of spreadability of gel shampoo

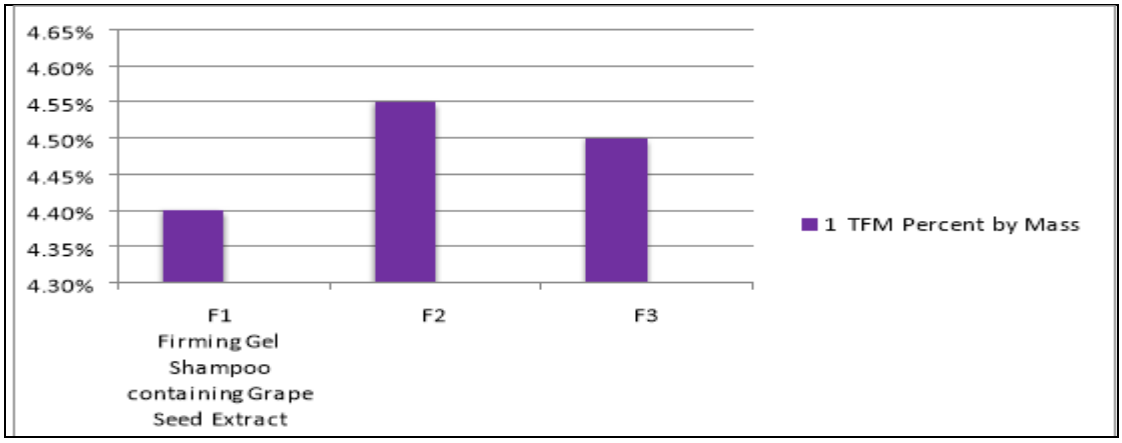


Determination of Total Fatty Matter:

Table no. 7 : Determination of Total Fatty Matter:

Sr. No.	Characteristics	Firming Gel Shampoo containing Grape Seed Extract		
		F1	F2	F3
1	TFM Percent by Mass	4.4%	4.55%	4.5%

Graph no.4 : Graphical representation of TFM of gel shampoo



Microbial examination of Gel Shampoo

Sr.no.	Test	Result	Specification	Unit
1.	Total bacterial count	20 CFU/gm	NMT100 CFU/gm	CFU/gm
2.	Total fungal count	NIL	NMT10 CFU/gm	CFU/gm

Table no. 8 : Microbial examination of Shampoo

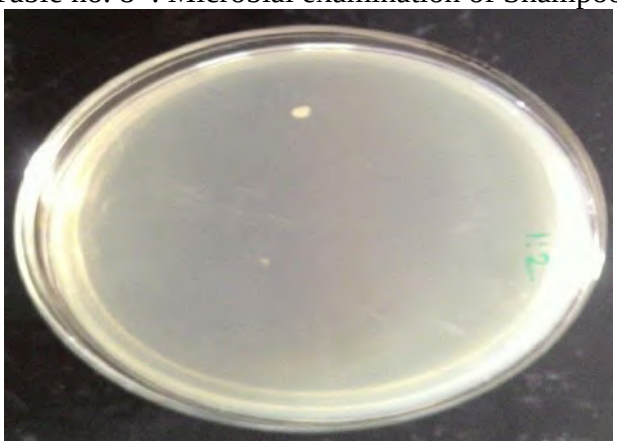


Fig: The total bacterial count

Result: The total bacterial count of Shampoo containing grape seed extract was found to be 10CFU/gm that is <100 CFU/gm. Therefore, the shampoo passes the test.

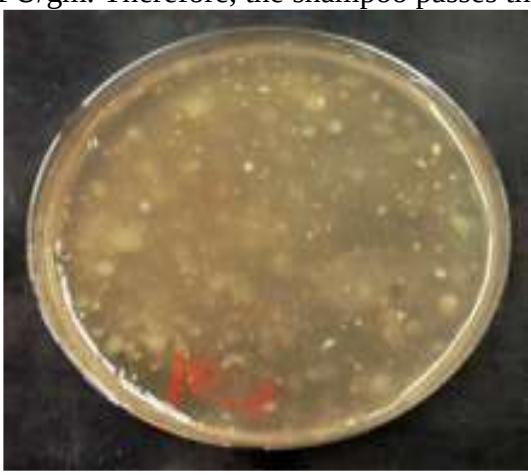
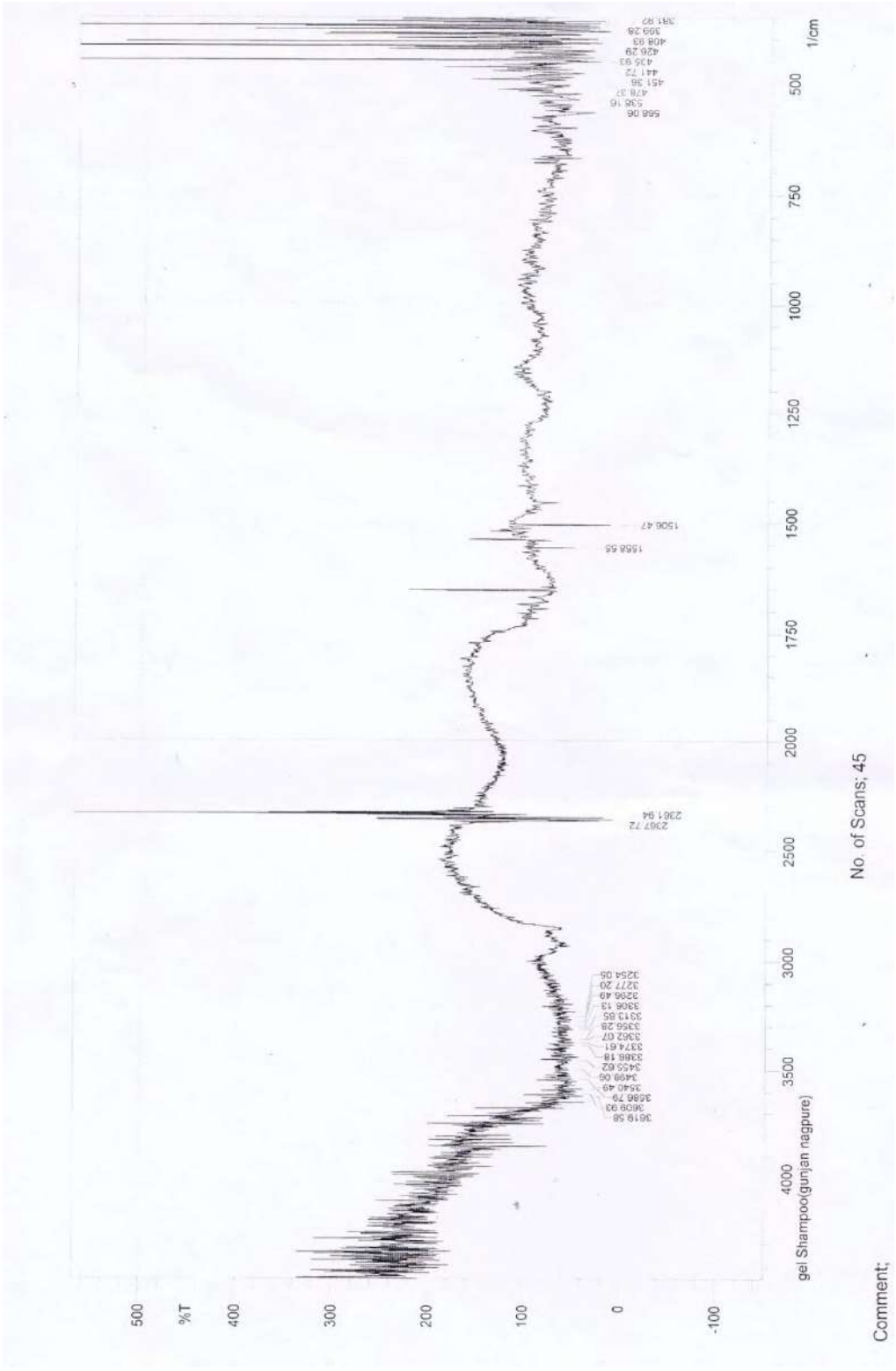


Fig : The total Fungal count

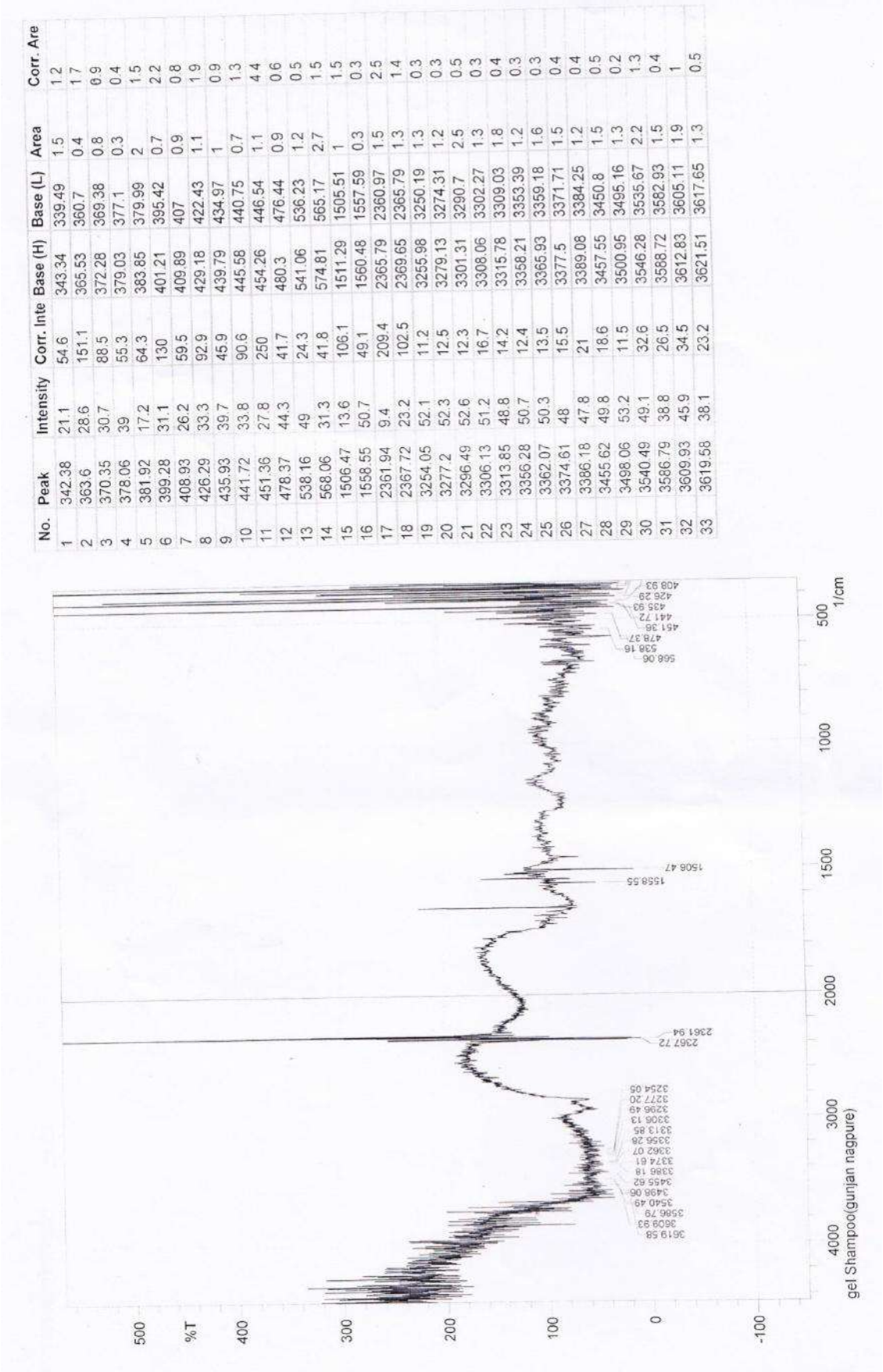
Result: The Total bacterial count Of a Shampoo containing grape seed Extract was found to be 10 CFU/gm that is <100 CFU/gm therefore, the Shampoo passes the test. The fungal count

of a Shampoo containing grape seed extract was found to be NIL. Therefore the Shampoo passes the test.

FTIR ANALYSIS:

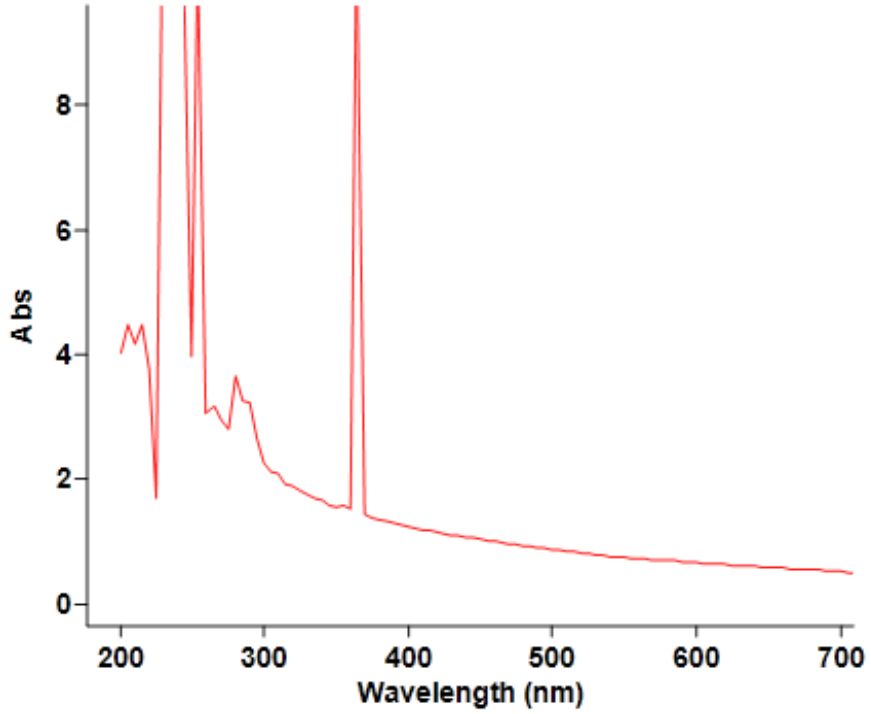


No.	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	342.38	21.1	54.6	343.34	339.49	1.5	1.2
2	363.6	28.6	151.1	365.53	360.7	0.4	1.7
3	370.35	30.7	88.5	372.28	369.38	0.8	0.9
4	378.06	39	55.3	379.03	377.1	0.3	0.4
5	381.92	17.2	64.3	383.85	379.99	2	1.5
6	399.28	31.1	130	401.21	395.42	0.7	2.2
7	408.93	26.2	59.5	409.89	407	0.9	0.8
8	426.29	33.3	92.9	429.18	422.43	1.1	1.9
9	435.93	39.7	45.9	439.79	434.97	1	0.9
10	441.72	33.8	90.6	445.58	440.75	0.7	1.3
11	451.36	27.8	250	454.26	446.54	1.1	4.4
12	478.37	44.3	41.7	480.3	476.44	0.9	0.6
13	538.16	49	24.3	541.06	536.23	1.2	0.5
14	568.06	31.3	41.8	574.81	565.17	2.7	1.5
15	1506.47	13.6	106.1	1511.29	1505.51	1	1.5
16	1558.55	50.7	49.1	1560.48	1557.59	0.3	0.3
17	2361.94	9.4	209.4	2365.79	2360.97	1.5	2.5
18	2367.72	23.2	102.5	2369.65	2365.79	1.3	1.4
19	3254.05	52.1	11.2	3255.98	3250.19	1.3	0.3
20	3277.2	52.3	12.5	3279.13	3274.31	1.2	0.3
21	3296.49	52.6	12.3	3301.31	3290.7	2.5	0.5
22	3306.13	51.2	16.7	3308.06	3302.27	1.3	0.3
23	3313.85	48.8	14.2	3315.78	3309.03	1.8	0.4
24	3356.28	50.7	12.4	3358.21	3353.39	1.2	0.3
25	3362.07	50.3	13.5	3365.93	3359.18	1.6	0.3
26	3374.61	48	15.5	3377.5	3371.71	1.5	0.4
27	3386.18	47.8	21	3389.08	3384.25	1.2	0.4
28	3455.62	49.8	18.6	3457.55	3450.8	1.5	0.5
29	3498.06	53.2	11.5	3500.95	3495.16	1.3	0.2
30	3540.49	49.1	32.6	3546.28	3535.67	2.2	1.3
31	3586.79	38.8	26.5	3588.72	3582.93	1.5	0.4
32	3609.93	45.9	34.5	3612.83	3605.11	1.9	1
33	3619.58	38.1	23.2	3621.51	3617.65	1.3	0.5



Graph No. 5 – FTIR of Gel Shampoo

UV SpectrophotometerAnalysis:



Scan Analysis Report

Report Time :	Sat 16 Mar 01:10:58 PM 2019
Method	
Batch:	C:\Users\ABCD\Desktop\Gunjan Nagpurkar VBMV\Gel Shampoo.DSW
Software version:	5.0.0.999
Operator:	

Instrument Parameters

Instrument	Cary 60
Instrument Version	2.00
Start (nm)	800.0
Stop (nm)	200.0
X Mode	Nanometers
Y Mode	Abs
UV-Vis Scan Rate (nm/min)	24000.000
UV-Vis Data Interval (nm)	5.00
UV-Vis Ave. Time (sec)	0.0125
Beam Mode	Dual Beam
Baseline Correction	Off
Cycle Mode	Off
Comments	

Sample Name: **Gel Shampoo**

Graph No. 6 – UV of Gel Shampoo

Collection Time	3/16/2019 1:11:17 PM
Peak Table	
Peak Style	Peaks
Peak Threshold	0.0100
Range	800.0nm to 200.0nm
Wavelength (nm)	Abs
365.0	10.000
355.0	1.572
280.0	3.671
265.0	3.179
255.0	10.000
245.0	10.000
215.0	4.476
205.0	4.478

Cyclic temperature test :

These tests are not carried out at fixed temperature and humidity. In this test, temperature was changed cyclically every day e.g. low-high-low-high to stimulate the changes in temperature daily.

Table no. 9 : Cyclic temperature test

Sr. no	Parameter	F1	F2	F3
1	Freeze temperature	Stable	Stable	Stable
2	Room temperature	Unstable	Unstable	Stable
3	High temperature	Unstable	Unstable	Stable

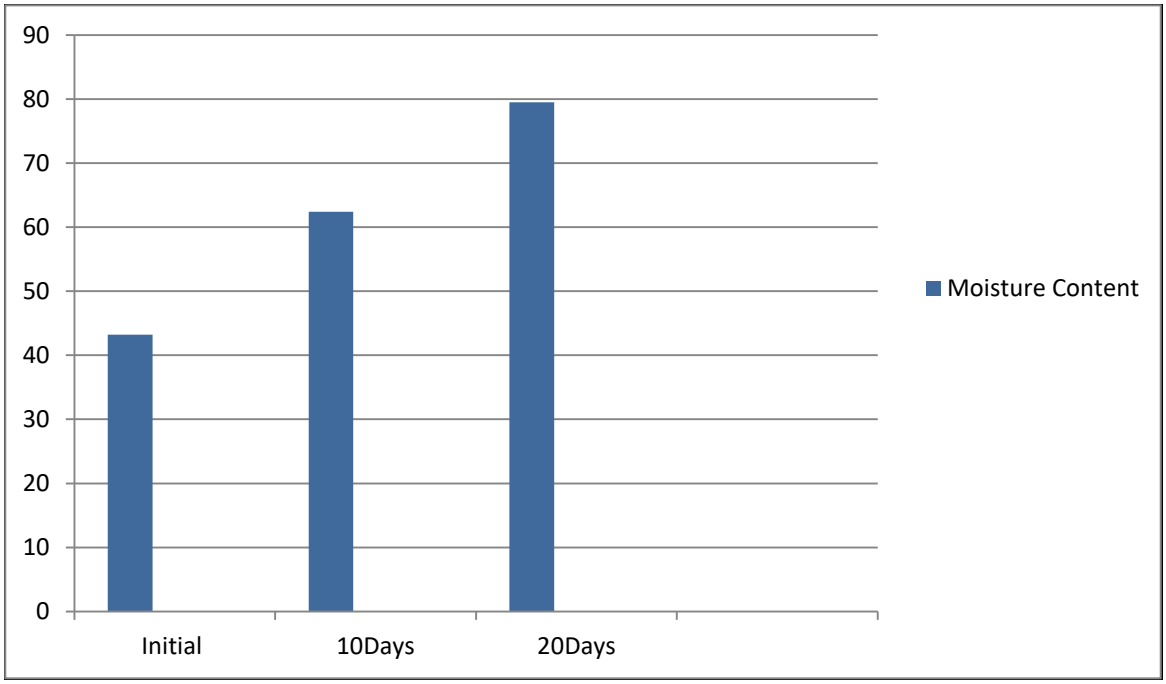
In Vivo Studies

Determination of moisturizing activity by corneometer

Table No.10: Determination of moisturizing activity by corneometer

Sr.No	Days	Moisture Content
1.	**Initial**	**43.2**
2.	**10 Days**	**62.4**
3.	**20 Days**	**79.5**

Graph No 7 : Graphical representation of Determination of moisturizing activity of Shampoo



Sensory Evaluation:

Objective: Comparison between two products based on following parameters

1. Appearance
2. Spreadiability
3. Moisturization
4. Over All

Total Voluntaries -6

Product details: Product A- Gel Shampoo
Product B- Ayur Gel Shampoo

Sensory evaluation of Gel Shampoo Vs. Benchmark product

S.No	Voluntaries	Appearance		Spreadiability		Moisturization		Overall	
		A	B	A	B	A	B	A	B
1.	Anjali	4	4	4.5	4	4.5	4	4	4
2.	Roshani	4	4	4	4.2	4	4	4	4.3
3.	Shrutika	4.2	4	4	4.1	4	4	4.5	4
4.	Meghana	4.5	4	4	4	4	4.2	4	4.2
5.	Pallavi	4	4.4	4	4.2	4	4	4.3	4
6.	Antara	3.7	4	3.8	4	4.3	4	4	4.1

S.No	Parameters	Product A	Product B
1.	Appearance	4.75	4.1
2.	Spreadiability	4.1	4
3.	Moisturization	4.18	4
4.	Overall	4.1	4

Status: Sensory evaluation and performance evaluation against benchmark is done and result found more than satisfactory.

Certificate of Analysis:

Grape Seed Extract

Description:

Proanthocyanidin grape seed extract. Dry in powder form, for dietary supplement applications.

Applications:

Free radical scavenging and antioxidant action

Batch nr. : GSE500306

Chemical Classification	**Organic, Nutritive**
Physical appearance	**Fine Reddish Brown Powder**
Colour	**Reddish Brown**
Odor	**Characteristic Tannin**
Taste	**Astringent**

Analysis:

	Specification:	Result:
%Moisture	**< 5%**	**4.0%**
Ash content	**< 8%**	**5.5%**
Total polyphenols (gallic acid equivalent) Folin method	**> 50%**	**54.1%**
Heavy metals:		
Arsenic (As)	**< 3 ppm**	**Complies**
Cadmium (Cd)	**< 1 ppm**	**Complies**
Lead (Pb)	**< 10 ppm**	**Complies**
Mercury (Hg)	**< 1 ppm**	**Complies**
Microbiology:		
Total Plate count (CFU/g)	**< 1000**	**Complies**
Coliforms (CFU/g)	**< 5**	**Complies**
Yeast and Mould (CFU/g)	**< 100**	**Complies**
Escherichia coli	**0**	**No growth**
Salmonella	**0**	**Absent**
Staphylococcus aureus	**0**	**No growth**

2.5 Subjective Evaluation:

To study the efficacy of the Gel shampoo formulation (i.e. F3 with 1% Grape seed extract) was selected for subjective evaluation and given to the volunteers for satisfactory results. Hence subjective Evaluation was carried out on the panel of human volunteers. Formulation F3 with 1% Grape seed extract was given to 30 volunteers and evaluation was carried out on the basis of their feedback after 30 days for parameters like Appearance, Ease of spreadability, Hair wash efficacy, Moisturisation and overall hair fall control experience. Hence,from the feedback by volunteers and by the photographic evaluation given below ,it was clear that formulation F3

with 1% Grape Seed Extract was proved to be excellent product in Gel shampoo and helps to regrowth of hairs as seen in the photographs.

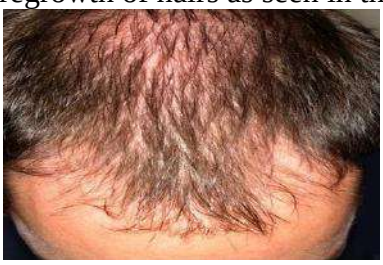


Fig: Hair fall before use

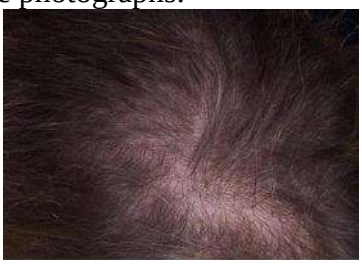


Fig: Hair growth after use

3. Result and Discussion:

3.1 Result

From the analysis of Grape seed extract,it was observed that procured sample passes the test as per Certificate of Analysis and hence was used for incorporation in formulation.

3.2 Result of Formulation and Development of Gel Shampoo using Grape seed extract

In the present study after analyzing all the three formulations, (i.e.F1,F2 and F3), on the basis of functional parameters, it was observed that,in the formulation-F1, base was not stable, hence emulsifying agents were increased. In F2, hair were not removed properly, hence the concentration of Grape seed extract was increased, and F3 containing1% Grape seed extract was giving satisfactory results. Hence, Gel shampoo (i.e. F3 with 1% Grape seed extract) was selected for further study

3.3 Result of stability study

The objective of stability study is to ensure that product will remain stable till the consumer has used the entire product. The stability not only indicates stability of formulation but also the stability of other ingredients present in the formulation of Gel shampoo. After analyzing all the three formulations, on the basis of functional parameters, it was observed that the Gel shampoo formulation (i.e. F3 with 1% Grape seed extract) was giving satisfactory results. Hence the formulation (i.e. F3 with 1% Grape seed extract) was subjected to accelerated stability studies. Changes in parameters like Color,Odor, pH at three different temperatures (i.e. in oven at (45^0C), in refrigerator at (4^0C) and at room temperature) was recorded for 45 days at interval of two days.

4.Conclusion: The present study was conduct with a view to formulate and evaluate on removing dust, dirt, sebum, greasiness, oil from scalp and hairs of given formulation by using grape seed extract. The gel shampoo was prepared with natural active agents. The concentration of active agent were kept in range of 0.5 %, 1%, 2%, each were incorporated and three combination of each were prepared.

Antioxidant testing of extract was done by Reducing power method. The Antioxidant activity was determined by power reducing method which showed that high absorbance which indicates it has good antioxidant property. The moisture content of hairs increases with the continuous use of product.

In the present work firming gel shampoo formulation gave satisfactory good cleansing and hair growth promoting property and this is achieved by the use of natural actives like grape seed extract.

The gel shampoo was prepared by the conventional procedure and all the factors, parameters such as pH, viscosity, stability, microbial analysis were determined. It was also kept accelerated by stability testing for 30 days. Then the product i.e. gel shampoo was applied on human volunteers and progressive effective result were found Thus the formulation F3 of gel shampoo containing 1% grape seed extract were found stable and gave most effective result respectively. At present because of availability of wide range of cosmetic products in market, consumers are giving special attention towards the selection of cosmetic product to develop a well standard formula, the new product viz. herbal firming gel shampoo was formulated by incorporating active extract singly for good effect.

Thus, F3 formulation of gel shampoo with grape seed extract were found to be most effective and stable. Thus, conclusion can be made that the gel shampoo containing grape seed extract have been able to remove dust, dirt, oil, stickyness and other signs also promotes hair growth without any side effect making hairs clean n healthy.

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Multi Functional Ingredients Used In Cosmetics

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Abstract
The minimalist movement has transcended various aspects of life, including personal care routines. The cosmetic industry has an undeniable need to design and develop new products to respond to the demands of consumers and international regulations. This requires substituting some traditional ingredients with new ones with more effective profiles. However, this transition towards the use of multi-functional ingredients in the cosmetic industry cannot compromise the effectiveness of the obtained products. This article reviews the emerging use of multi-functional ingredients in this new direction of the cosmetic industry. They often include benefits such as emolliency, film forming, fragrance, antimicrobial/antimicrobial boosting and viscosity building, among the most popular. In addition, the use of multi-tasking ingredients opening a promising future path to the design of cosmetic formulations as the formulator's work of creating innovative formulations being boosted by new multifunctional ingredients is also reviewed. Consumers are becoming more conscious of their purchasing decisions, seeking products that align with their values and have a minimal environmental footprint. Finally, with a focus on quality over quantity, minimalist personal care emphasizing use of essential multi-functional ingredients that are effective, sustainable, and personalized is also discussed.
Key Words: Multi-functional ingredients, cosmetics, minimal cosmetics, innovation.

Introduction

Long gone are the days when cosmetic products escaped scrutiny by the consumers and sold solely on the appeal of look and feel, glorified by savvy marketing. Today's consumers are more educated than ever. This is not surprising considering how social media revolutionised the way information is shared between individuals. Earlier, cosmetics were formulated with many ingredients, each serving different purposes. However, with the advancing innovation individuals are seeking more simpler and mindful approach to personal care.

Multifunctional ingredients are having a measurable impact, per a recent Morning Consult and EWG survey [1]. The report, which compared 2004 survey results to data collected in 2023, revealed that the number of ingredients/chemicals per personal care/beauty product seems to have dropped in the last 19 years. The takeaway of the survey is clear: formulations are becoming simpler, perhaps driven by a desire for minimalism, as well as the introduction of many multifunctional materials.

Multi-Functional Ingredients In Cosmetics

Whether it's a skincare product offering hydration, anti-aging, and blemish treatment properties, or an anti-frizz, strand-repair, in-shower conditioning hair mask, consumers are demanding more of their cosmetic products than ever before. This demand, however, puts pressure on the cosmetics industry to keep formulating products that tackle multiple concerns at once.

Adapting the "less is more" approach, formulators can continue to deliver innovative products to this fast-moving and high-demand market. Multi-Functional ingredients offer multiple

benefits simultaneously, enabling formulations with only a few ingredients to tackle multiple concerns.

Following are some examples of multi-functional ingredients used widely:

1. Niacinamide: Niacinamide is a type of vitamin B3. This ingredient promotes more youthful-looking skin and may also minimize the appearance of fine lines and wrinkles with its powerful hydrating and skin-plumping abilities. Niacinamide has been shown to ease inflammation, which can help calm redness. It can also soothe irritation caused by strong exfoliants like retinol or glycolic acid that remove dead cells from the surface of your skin. It is also dermatologist-approved for brightening skin tone.

2. Caffeine: Caffeine being an anti-aging powerhouse acts as an antioxidant and protects premature aging and wrinkle formation on skin. It draws excess fluids out of the cells creating a lightening and tonic effect, thereby improving the texture of the skin. It reduces redness and inflammation of skin by constricting blood vessels and thus provides an even skin tone. It even treats puffiness and dark circle under the eye by boosting blood circulation. Caffeine penetrates deep into the hair roots and stimulates them. Besides preventing male baldness and hair loss, it stimulates the hair roots in the scalps of women and triggers stronger hair growth. It is a stimulant for hair follicle and helps to restore hair growth. Besides strengthening the hair shaft, caffeine adds natural shine to your hair and makes them more manageable. It also enhances hair color thereby making your hairs more black!

3. Caprylyl Glycol: Caprylyl glycol works as an humectants. It also works as a skin softener, or emollient, and helps support a healthy skin barrier. It helps the product to last longer while improving the product texture. It also exhibits wetting properties as well as builds viscosity of the product.

4. Shea Butter: From repairing dry, cracked hands to reversing moisture loss and even reducing the appearance of stretch marks and scars, this versatile ingredient can be applied from head to toe! It effortlessly blends into the skin, leaving you with a smooth, moisturized feeling without clogging pores. The healing properties of shea butter make it the perfect choice for a natural after-sun moisturizer to soothe skin and reduce inflammation. Shea butter is also great for promoting optimum scalp health. The soothing properties of shea butter work wonders for itchy, dry scalps. In it also promotes collagen production, smoothes wrinkles, and restores moisture in areas prone to dryness, such as under your eyes. Shea butter can also aid in reducing the appearance of fine lines, blemishes, and other skin imperfections.

5. Polymers: Polymers present a broad range of applications in cosmetics, e.g., rheological modifiers, emulsifiers, stimuli-responsive reagents, conditioners, film formers, fixations, foam stabilizers, skin-feel beneficial agents, or antimicrobial agents, which allows considering these types of molecules as the most used ingredients in different families of cosmetic products.

6. Fragrance + Preservation: This multifunctional cosmetic ingredient exhibits its primary function of a fragrance as well as its secondary function of broad spectrum efficacy to protect the integrity of the finished formulation.

7. Fermented Skin Care: Fermented actives addresses the shift in consumer focus from fighting visible signs of aging to proactively maintaining healthy skin. It aids in barrier repair and promotes skin regeneration while contributing to a more youthful appearance.

Consideration of societal, economic and environmental issues is of primary interest in the design of cosmetic products. In the case of skincare products, nowadays, consumers are looking for highly effective, custom cosmetics that are adapted to their skin problems. Several active

cosmetic molecules are already in use and provide different benefits to the skin (UV-protection, anti-aging, anti-oxidant, anti-acne, skin lightening, skin hydration, etc.).
Multi-functional ingredients provide an edge to the formulators as it helps in controlling the inventory for manufacturing, cutting down the selection of multiple ingredients for the formulation process and easing the procedure to be followed.
Additionally, minimalism extends to ingredient selection with a focus on shorter INCI lists. By prioritizing products with concise ingredient lists, individuals can simplify formulations, avoid unnecessary additives, and gain a better understanding https://www.webmd.com/vitamins/ai/ingredientmono-1512/shea-butterof what they apply to their skin, potentially reducing the risk of irritation or sensitivity. This combination of naturality, sustainability and streamlined formulations exemplifies the minimalist philosophy in personal care.

Conclusion:
Whatever the choice of concept for the multifunctional cosmetic is developing, the range of available ingredients and textures is on the rise. The alignment with safety, stability testing, regulatory, scale-up functions during the development is of utmost importance for a winning formula.
Minimalism extends to multi-functional ingredient selection with a focus on shorter INCI lists. By prioritizing products with concise ingredient lists, individuals can simplify formulations, avoid unnecessary additives, and gain a better understanding of what they apply to their skin, potentially reducing the risk of irritation or sensitivity.
Staying up to date with the latest research in multifunctional cosmetics will continue to support innovative development and break the boundaries of what consumers can expect from their cosmetic products.

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Formulation & Development of Night Cream Using Butterfly Pea Extract

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Abstract
The present study is done to develop a cosmeceutical night cream using Butterfly Pea Extract, evaluate it and to carry out comparative study of prepared formulation with the marketed cream.The Butterfly Pea is a climbing plant[1] whose blue flowers are commonly used as a food dye, particularly among the Peranakans (Straits Chinese).This plant was widely used in traditional medicine because it is rich in bioactive compounds. In treating diabetics, blood pressure, retinal damage, edema, and indigestion both the aerial and underground parts of this plant are being used. Researchers proved this plant's medicinal activities such as nootropic activity, antioxidant activity, analgesic activity, anti-inflammatory and antibacterial activity. Currently, this plant's uses are widely spread in the nanotechnology field as well. The present formulation is evaluated by modern scientific parameters. Most of the night cream used as a moisturizing cream use different herbal extracts as base.Cream formulation was evaluated by checking pH, viscosity, spreadability and moisturizing test. From the result it is found that present cream containing Butterfly Pea Extract shows the better moisturizing activity than the marketed formulation. The formulated product does not show any irritation signs on volunteers and has more moisturising activity than marketed formulation.
Keywords : ClitoriaTernatea, Night Cream, Formulation, Skincare, Antioxidant.

Introduction:-
Creams are semi-solid mixtures of oil and water, categorized into oil-in-water (O/W) and water-in-oil (W/O) types. Water-in-oil creams create a moisturizing oily barrier, reducing water loss from the outermost skin layer. The main cream ingredients include water, oil, emulsifier, and thickening agent. Night creams, containing skincare ingredients and anti-aging agents, target concerns like lines, wrinkles, and dark spots. Nighttime application is optimal for addressing issues such as water loss, skin aging, fatigue, and dark spots through natural reparative processes, leading to a more hydrated and youthful complexion over time.

Active Ingredient:

1. **Butterfly Pea Extract (Clitoriaternatea):**

- A plant species with blue flowers, it has been traditionally revered in India and is known for its potential skincare benefits.
- Rich in polyphenols, flavonoids, and proanthocyanidin, it acts as an antioxidant, collagen, and elastin booster.
- Exhibits anti-glycation properties, slowing down skin aging, and has been found to have strong free-radical scavenging and anti-inflammatory abilities.

Potential health benefits of butterfly pea flower
Butterfly pea flowers may be associated with several health benefits.

Supports skin and hair health
Cosmetic manufacturers boast about butterfly pea flowers' effectiveness in everything from skin care serums to hair mists and shampoos.
According to a 2021 study, butterfly pea extract may increase your skin hydration by 70% one hour after topical application (8Trusted Source).

A 2012 animal study found that butterfly pea extract may be more effective at promoting hair growth than monoxide, which is a common product used to treat hair loss (9Trusted Source).
Butterfly pea flower contains a rich array of antioxidants, which may also be beneficial for promoting hair and skin health (10Trusted Source, 11Trusted Source, 12 Trusted Source).
Still, more research is needed to fully understand how butterfly pea flower may affect your hair and skin.

May promote weight loss

Some studies even suggest that butterfly pea flower may aid in weight loss efforts.
One test-tube study suggests that butterfly pea flower extract may slow the formation of fat cells by regulating certain pathways involved in cell progression (13Trusted Source).
Some older test-tube and animal studies have found that ternatins, which are found in butterfly pea flower, may also block the synthesis of fat cells in your body (14Trusted Source, 15Trusted Source, 16 Trusted Source).
Further research is necessary to evaluate how butterfly pea flower may impact your weight, especially when worked into your diet.

Stabilizes blood sugar levels

Studies indicate that butterfly pea flower may reduce your risk of diabetes and related symptoms.
For instance, one study in 15 men showed that drinking a beverage containing butterfly pea flower extract increased antioxidant levels and reduced blood sugar and insulin levels, despite the sugar levels in the drink (17Trusted Source).
Moreover, an animal study found that administering butterfly pea flower extract to rats with diabetes significantly reduced their blood sugar levels compared with a control group (18).
One study even reported that the antioxidant properties of butterfly pea flower may protect against cell damage and complications related to diabetes (4Trusted Source).
However, additional studies are needed to determine how butterfly pea flower may impact your long-term blood sugar control.

Night Cream Formulation with Butterfly Pea Extract

Materials (Ingrdients) :

1)Stearic Acid
2)Cetyl Alcohol
3)Light liquid paraffin
4) Glyceryl Monostearate
5) Shea Butter
6) Distilled Water
7) Disodium EDTA
8) TEA
9) Phenoxyethanol

List Of Equipment :

1) Beaker
2) Brook Field Viscometer
3) Mechanical Stirrer
4) Weighing Balance
5) Spatula

EXPERIMENTAL WORK

Table No. 1 Method of Preparation of Night Cream:

The night cream formulation involves a careful selection of active ingredients. Three formulations (F1, F2, F3) were created with varying quantities of Stearic Acid, Cetyl Alcohol, Light Liquid Paraffin (LLP), Glyceryl Monostearate (GMS), Shea Butter, Distilled Water, Disodium EDTA, TEA, and Phenoxyethanol. The optimization procedure includes

heating oil and water phases, mixing, adjusting pH with triethanolamine, and adding Phenoxyethanol.

Sr. No.	Ingredients	F1 For 100%	F2 For 100%	F3 For 100%
1	Stearic Acid	2	3	4
2	Cetyl Alcohol	1	2	3
3	Light liquid paraffin	3	5	6
4	Glyceryl Monostearate	1	1.5	2
5	Shea Butter	0.5	1	1.5
6	Distilled Water	71	72	72
7	Disodium EDTA	0.1	0.1	0.1
8	TEA	0.2	0.3	0.3
9	Phenoxyethanol	0.3	0.3	0.3

Optimization of Night cream Procedure :

Procedure

Heat oil phase and water phase differently up to 70 – 75C Mix oil phase into water phase with continuous stirring then add triethanol amine to adjust the pH then and Phenoxyethanol in the end.

EVALUATION:

Parameter of Base Formulation of Night Cream

Table No. 1Parameter of Base Night Cream

Sr. No.	Parameter	F1	F2	F3
1	**Appearance**	++	+++	++
2	**Color**	+	++	++
3	**Consistency**	+	++	+++
4	**Spreadability**	++	+++	++
5	**Feel**	+	+++	++
6	**Odour**	++	++	++

Here, += Good, ++= Better, +++= Best

From the above observation formula F3 was Stable and it shows consistency, Spreadability, and feel therefore it was selected and extract was added with different concentration and forward for in vitro study as per IS and in vivo study with human volunteer.

Final Formulation of Butterfly Pea Extract Night Cream

The final selected base formulation includes Stearic Acid, Cetyl Alcohol, LLP, GMS, Shea Butter, Distilled Water, Disodium EDTA, TEA, and Phenoxyethanol.

Sr. No.	Ingredient	Formulation
1	Stearic Acid	4
2	Cetyl Alcohol	3
3	Light liquid paraffin	6
4	Glyceryl Monostearate	2
5	Triethanol amine	0.3
6	Distilled Water	72
7	Disodium EDTA	0.2
8	Glycerine	3
9	Phenoxyethanol	0.3
10	Butterfly pea extract	5

Parameters of Night Cream with Butterfly Pea Extract:
The final formulation with butterfly pea extract was evaluated for appearance, colour, consistency, feel, and pH, showing positive attributes and stability.

Parameter of night cream of butterfly pea extract

Sr. No.	Parameter	Formulation
1	Appearance	++
2	Color	++
3	Consistency	+++
4	Feel	++
5	pH	**5.5**

Here, += Good, ++= Better, +++= Best

RESULT AND DISCUSSION:
A) In Vitro Evaluation Study:
a) Physical parameters:

Appearance	Viscous cream
Colour	Light blue
Consistency	Good
Spreadability	Good
Odour	Pleasant

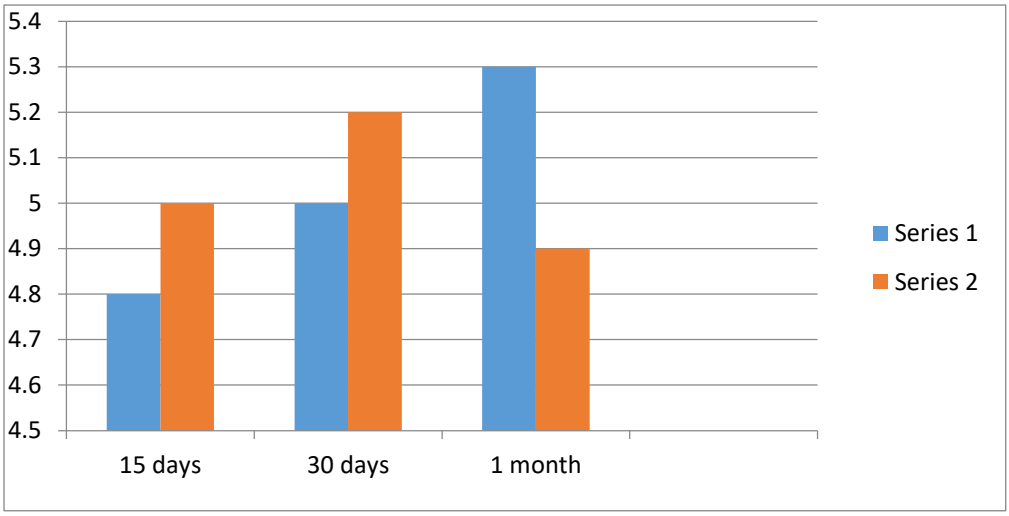
b)Determination of Ph

Sr. No	Time interval	Ph
1	15 days	5.2
2	30 days	5.4
3	1 Month	5.6

b) Determination of pH of Night Cream With Active:
The pH of the selected formulation (F3) was measured over a 30-day period, showing consistency between 5.4 and 5.6.

Sr. No.	Time interval	A	B	C
1	**Initial day**	5.5	5.6	5.6
2	**15thday**	5.6	5.4	5.5
3	**30thday**	5.5	5.6	5.6

Graphical representation of Determination of pH:



Sr. No.	Parameter	Formulation
1	Appearance	++
2	Color	++
3	Consistency	+++
4	Feel	++
5	Ph	**5.5**

Result:

From the above graph we can conclude that pH of the product at RT, 5℃ and 45℃ are comparatively equal (the pH range is acceptable) in three months as compare to initial value. The product passed in pH value

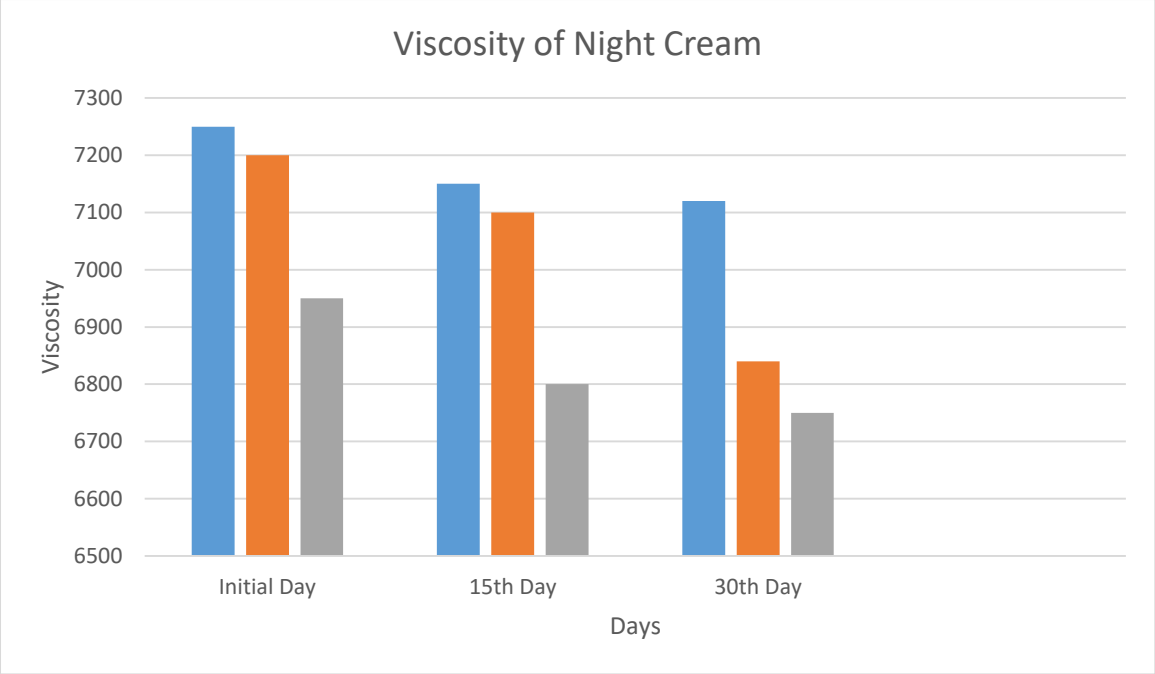
c) Determination of Viscosity Principle :

The viscosity of night cream determined by using Brookfield Viscometer. The values obtained from the sample note

Sr. no	Time interval	Viscosity
1	15 days	6950 cp
2	30 days	6800 cp
3	1 Month	6750 cp

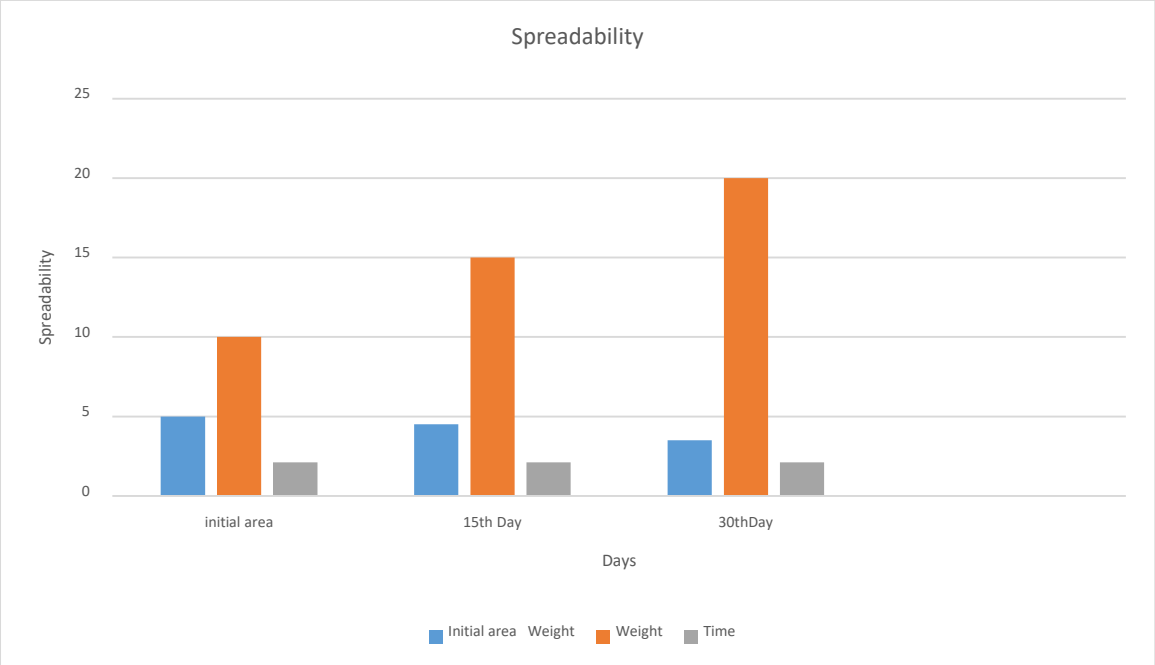
The viscosity of night cream determine by using Brookfield Viscometer. The values obtained from the sample noted.

Sr. No.	No. of days	1	2	3
1	Initial Day	7250cp	7200cp	6950cp
2	15th Day	7150cp	7100cp	6800cp
3	30th Day	7120cp	6840cp	6750cp



d)Determination of Spreadability

Sr. No.	Days of interval	Initial area	Weight	Time
1	Initial day	5cm	10gm	2.1 Sec
2	15th Day	1.6 cm	15 gm	2.1 Sec
3	30th Day	3.5 cm	20gm	2.1 Sec



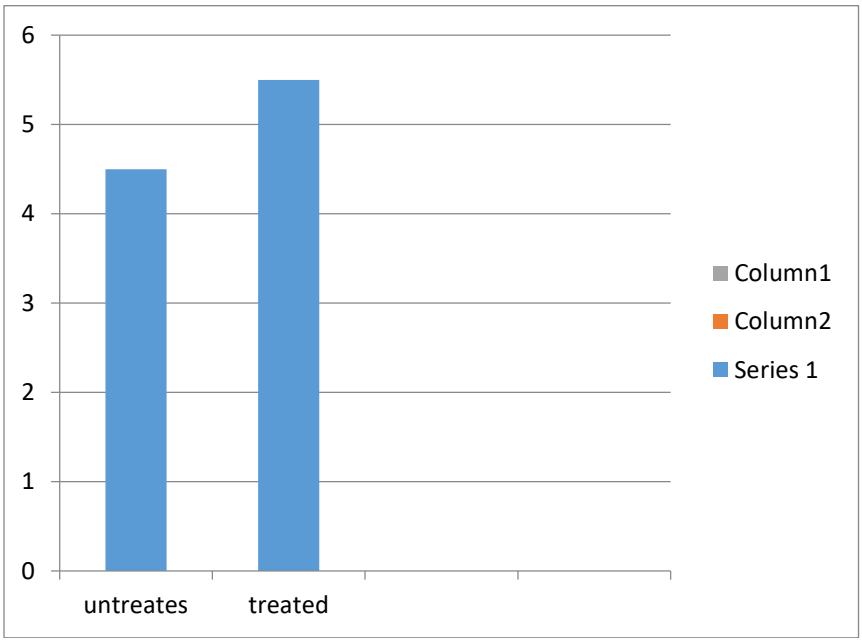
e)Temperature variation test (Thermal stability test) Result:

From the above three graphs, we can conclude that the product has passed thermal stability test. Because at all the temperatures the product remains constant in pH, viscosity & moisture content parameters, as well as its physical parameters such as appearance, color and odor**.**

B) In Vivo Evaluation Study:

a) Effect of anti-ageing agent on skin:

Graphical representation of anti-ageing effect of night cream:



Result:

From the above graph we can con conclude that the night cream has good anti-ageing effect on skin.

Determination of Microbial Testing:

Table: Determination of microbial testing

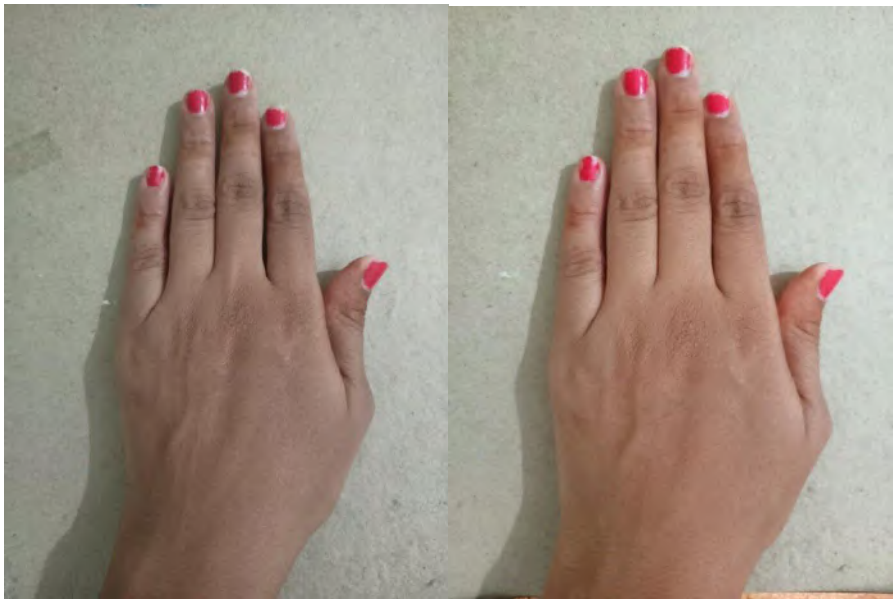
Interpretation of result:

Although there is some correlation between the size of the zone of inhibition and the susceptibility of the organism to the antibiotic, the former is a function of many variables e.g. density of the inoculum, depth of the medium, infusibility of antibiotic etc. The size of the inhibition zone at which the organism is considered Resistant, Intermediates or sensitive is given in the zone size interpretative chart as a part of this literature.

Photographic evaluation:

Photographic evaluation is carried to see the effect of the product visually. In case of determination of activity photographic evaluation was adopted. In this method the

photograph of skin before and after application on skin were taken out and effect of product was determined.



Before After

Conclusion:

The formulation of the night cream, enriched with an anti-ageing active, successfully passes rigorous stability tests and sensory evaluations. The product demonstrates resilience in both physical and chemical assessments, including aesthetic parameters. In vivo tests substantiate the positive effects of the night cream. Given the discerning consumer focus on selecting high-quality cosmetic products, the developed night cream stands out as a well-formulated solution. The synthetic base, containing polymers, humectants, and preservatives, underwent meticulous selection, and the final formulation exhibited superior performance in dirt removal and overall skin enhancement.

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23

Formulation and Development of Body Mask With Active Dead Sea Mud

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ABSTRACT:
A body mask with active Dead Sea mud offers a mineral-rich skincare experience. The mud, sourced from the Dead Sea, is renowned for its high concentration of salts and minerals. When applied as a mask, it helps exfoliate, detoxify, and nourish the skin, promoting a smoother and more revitalized appearance. The mask's unique properties make it effective for deep cleansing and may contribute to improved skin texture and hydration.
An active dead sea mud-based body mask combines the purifying properties of dead sea mud with the benefits of a body mask.This unique formulation is rich in minerals like magnesium, calcium, and potassium,renowned for their skin-nourishing qualities. When applied, the mask helps in drawing out impurities, unclogging pores, and revitalizing the skin. Additionally, the dead sea mud's exfoliating effect can leave the skin feeling smoother and more radiant, making it a popular choice for spa-like treatments and skincare routines.

KEYWORDS:
Dead Sea Mud Mask, Dead Sea Mud, Body Mask, Clay Mask.

INTRODUCTION:

Body Mask

Masks are cosmetics product which have been used since long ago. Packs & masks are simple mix of chemicals & natural ingredients. colloidal & adsorptive clay & earth which are present in some packs will absorb grease and dirt from the facial skin. The main purpose of packs & masks is to achieve tightening and cleansing effect. Masks are just not for the face but are used all over the body.
You are likely well acquainted with face masks, those intended to unclog pores or for some other benefits, but what about masks for skin below the neck?
The concept of the body masks is simple. The same skincare active ingredients found in your favorite serums and moisturizers (like collagen, charcoal, and niacinamide, or some others) are formulated into larger tubs of beauty goop that's meant to be spread across your arms, legs, decolletage, butt, and elsewhere. Why body masks, you might you ask? Well, whether you're dealing with stubborn body acne, stretch marks, hyperpigmentation, wound healing, ageing or simply want to indulge in a nourishing beauty treatment, the benefits of body masking are actually kind of endless.

Dead sea mud powder

INCI NAME:- Maris limus (Sea Silt Extract)
The main chemical constituents of Dead Sea Mineral Mud Powder are: Dead Sea mud, Organic Matter, and Minerals (expressed in Oxides: Silicon Dioxide, Calcium Oxide, Magnesium Oxide, Sodium Oxide, Potassium Oxide, Iron Oxide, Aluminum Oxide, Phosphorous Pentoxide, Titanium Oxide, Sulfur Trioxide, Manganese Oxide, Zirconium Dioxide, Chromium Oxide, Zinc Oxide, Nickel Oxide, Copper Oxide, Indium Oxide, Chloride, and Bromide).
Dead Sea Mud is the grainy silt obtained from the shores of the Dead Sea. The mud collected is further processed to eliminate sand particles, dirt, etc., till a smooth paste-like solution and after further process the powder is obtained. It contains abundant sources of minerals like calcium, sodium, potassium, magnesium, etc. Color of this mud changes as per season as it is

available in light grey color in summer whereas its colors might vary from dark grey to black in summer and rainy seasons.

The presence of natural medicinal properties in this mud powder makes it an essential ingredient to cure numerous skin diseases. Several cosmetic products also use Dead Sea Mud Powder as it does not contain any synthetic ingredients. The different forms of sea mud are added in mud baths, cleansing body and body masks as it has rejuvenating, exfoliating, and cleansing properties. This Mineral-rich salt maintains its reputation for having Soothing, Strengthening, Regenerative properties.

Cosmetic Uses:-

Cosmetically or topically in general, Dead Sea Mineral Mud is beneficial for dry, oily and normal skin types. It removes dead cells from the skin's surface, purges the pores of impurities, and balances the skin's oil production and ph level. While it remains warm and moist, usually for approximately an hour, Dead Sea Mud produces internal heat and stimulates circulation, which reduces the appearance of cellulite. Additionally, it accelerates the skin's detoxification process on the surface and in the tissues by gently drawing out visible and invisible impurities such as air pollution, allergens, dust, and dirt. As the mud dries, it exhibits gentle pulling action that draws out excess oil, tightens, and exfoliates to remove dead skin, which reveals a healthier layer of skin.

Furthermore, it enhances skin elasticity, reduces the appearance of pores, and smooths the appearance of fine lines and wrinkles.

Hair Benefits:

Makes hair thick and shiny, Eliminates excess fat content of hair, Relieves Dandruff, Strengthens the hair follicles and relieves of split ends, Increase scalp blood circulation.

MATERIALS AND METHODS:

Body Mask is prepared by using following ingredients and equipment.

Materials

Stearic acid, Glycerol monostearate, Shea butter, Cocoa butter, Glycerine, Coconut oil, Olive oil, Jojoba oil, Avacado oil, Propylene glycol, EDTA, Niacinnamide, Water, Kaoline, Fuller's earth, Salicylic acid, Sepicalm vg, Vitamin E, Aloe Extract, Phenoxyethanol, Dead Sea Mud

List Of Equipment

Mechanical Stirrer, Spatula, Borosilicate Glassware, Weighing Balance, PH Meter, Hot Plate, Brookfield Viscometer, Corneometer.

Method:

Preparation of base formulation:

In any cosmetic preparation it is necessary to have stable formulation before Incorporation of active. Preparation of base formulation is important before incorporation of active ingredient, to prepare a stable cosmetic formulation. The Effectiveness and stability of product was depending upon the compatibility of active ingredients.

Sr.no	Ingredients	F1 For 100%	F2 For 100%	F3 For 100%
1	Stearic acid	1.5%	2%	2%
2	Glycerol monostearate	2%	2%	1.5%
3	Shea butter	1.5%	2%	1.5%
4	Cocoa butter	1.5%	2%	2%
5	Glycerine	2.5%	2.5%	2%
6	Coconut oil	2%	2.5%	3%
7	Olive oil	2%	1.5%	1.5%
8	Jojoba oil	1.5%	2%	2%

9	Avacado oil	1.5%	2%	2%
10	Propylene glycol	4%	3%	2.5%
11	EDTA	0.1%	0.1%	0.1%
12	Niacinnamide	0.5%	0.8%	0.8%
13	Water	55.4%	56.3%	58.8%
14	Kaoline	9%	7%	8%
15	Fuller's earth	9%	7%	8%
16	Salicylic acid	0.5%	0.5%	0.5%
17	Sepicalm vg	0.3%	0.5%	0.5%
18	Vitamin E	1%	1%	1%
19	Aloe Extract	2%	2%	2%
20	Phenoxyethanol	0.2%	0.3%	0.3%

Table no.1

Procedure:

1.All the apparatus were cleaned and take all the ingredients as per the formulation of body mask.

2. Weigh all The Ingredients as per the formulation.

3. Heat oil phase upto 60^0C and water phase upto 75^0C seperately.

4. Then added oil phase into water phase by slow stirring.

5. Add clays by sprinkling on slow stirring into emulsion.

6. Then the mixture was continuously stirred until Uniform.

7. Half quantity of propylene glycol was added to water phase and remaining quantity of propylene glycol was mixed with salicylic acid for proper mixing and this slurry then added into emulsion.

8. At 45^0 C added vitamine E oil, Sepicalm vg and Aloe extract.

9. Then added phenoxy ethanol drop by drop.

10. Clay base was stored in suitable container.

11. From the above formulation F2 was found to be most stable hence it was selected.

Parameter of base formulations of Body mask:

Sr. No.	**Parameter**	**F1**	**F2**	**F3**
1	**Appearance**	++	+++	+++
2	**Colour**	+	++	++
3	**Consistency**	+	+++	+++
4	**Spreadability**	++	+++	++
5	**Feel**	+	+++	+++
6	**Odour**	++	++	++
7	**PH**	6.4	6.6	6.3

Table no 2

Here, += Good, ++= Better, +++= Best

From the above observation formula F2 was Stable and it shows consistency, spreadability, and feel therefore it was selected and extract was added with different concentration and forward for in vitro study as per IS and in vivo study with human volunteer.

Final selection of base formulation:

Sr.no	Ingredients	Quantity For 100%
1	Stearic acid	2%
2	Glycerol monostearate	2%
3	Shea butter	2%
4	Cocoa butter	2%
5	Glycerine	2.5%
6	Coconut oil	2.5%
7	Olive oil	1.5%
8	Jojoba oil	2%
9	Avacado oil	2%
10	Propylene glycol	3%
11	EDTA	0.1%
12	Niacinnamide	0.8%
13	Water	56.3%
14	Kaoline	7%
15	Fuller's earth	7%
16	Salicylic acid	0.5%
17	Sepicalm vg	0.5%
18	Vitamin E	1%
19	Aloe Extract	2%
20	Phenoxyethanol	0.3%

Table no 3

Parameters of final base Body mask:

Sr. no.	**Parameter**	**Formulation**
1	Apperance	**+++**
2	Colour	**++**
3	Consitency	**+++**
4	Spreadability	+++
5	Feel	**+++**
6	Odour	++
7	pH	6.6

Table no 4

Abbrevation

‘’+’’= poor, “++”=good, “+++= Satisfactory

Form the above table of parameter has required property and has selected as a base formulation.

Formulation with Addition of Active Ingredient

Sr.no	Ingredients	F1 For 100%	F2 For 100%	F3 For 100%
1	Stearic acid	2%	2%	2%
2	Glycerol monostearate	2%	2%	2%
3	Shea butter	2%	2%	2%
4	Cocoa butter	2%	2%	2%
5	Glycerine	2.5%	2.5%	2.5%
6	Coconut oil	2.5%	2.5%	2.5%
7	Olive oil	1.5%	1.5%	1.5%
8	Jojoba oil	2%	2%	2%
9	Avacado oil	2%	2%	2%
10	Propylene glycol	3%	3%	3%
11	EDTA	0.1%	0.1%	0.1%
12	Niacinnamide	0.8%	0.8%	0.8%
13	Water	56.3%	56.3%	56.3%
14	Kaoline	7%	7%	7%
15	Fuller's earth	7%	7%	7%
16	**Dead sea mud powder**	**2%**	**3%**	**4%**
17	Salicylic acid	0.5%	0.5%	0.5%
18	Sepicalm vg	0.5%	0.5%	0.5%
19	Vitamin E	1%	1%	1%
20	Aloe Extract	2%	2%	2%
21	Phenoxyethanol	0.3%	0.3%	0.3%

Table no. 5

Procedure:

1.All the apparatus were cleaned and take all the ingredients as per the formulation of body mask.
2. Weigh all The Ingredients as per the formulation.
3. Heat oil phase upto 60^0C and water phase upto 75^0C seperately.
4. Then added oil phase into water phase by slow stirring.
5. Add clays and Dead sea mud powder by sprinkling on slow stirring into emulsion.
6. Then the mixture was continuously stirred until Uniform.
7. Half quantity of propylene glycol was added to water phase and remaining quantity of propylene glycol was mixed with salicylic acid for proper mixing and this slurry then added into emulsion.
8. Then sepicalm vg was added.
9. At 45^0 C added vitamine E oil and Aloe extract.
10. Then added phenoxy ethanol drop by drop.
11. Perfume Was Added In Required Quantity.
12. Clay Mask with active was stored in suitable container for further study.

Parameter of formulation of Dead Sea Mud Body Mask

Sr. No.	**Parameter**	**F1**	**F2**	**F3**
1	**Appearance**	++	+++	+++
2	**Colour**	+	+++	++

3	**Consistency**	+	+++	+++
4	**Spreadability**	++	+++	++
5	**Feel**	+	+++	+++
6	**Odour**	++	+++	++
7	**PH**	6.4	6.6	6.3

Table no 6

Here, += Good, ++= Better, +++= Best

From the above observation formula F2 was Stable and it shows consistency, spreadability, and feel therefore it was selected and extract was added with different concentration and forward for in vitro study as per IS and in vivo study with human volunteer.

Final formulation of Dead Sea Mud Body Mask

Sr.no	Ingredients	Quantity For 100%
1	Stearic acid	2%
2	Glycerol monostearate	2%
3	Shea butter	2%
4	Cocoa butter	2%
5	Glycerine	2.5%
6	Coconut oil	2.5%
7	Olive oil	1.5%
8	Jojoba oil	2%
9	Avacado oil	2%
10	Propylene glycol	3%
11	EDTA	0.1%
12	Niacinnamide	0.8%
13	Water	53.3%
14	Kaoline	7%
15	Fuller's earth	7%
16	**Dead sea mud powder**	**3%**
17	Salicylic acid	0.5%
18	Sepicalm vg	0.5%
19	Vitamin E	1%
20	Aloe Extract	2%
21	Phenoxyethanol	0.3%

Table no 7

Parameter of final formulation of Dead Sea Mud Body Mask

Sr. no.	Parameter	Formulation
1	Apperance	+++
2	Colour	+++
3	Consitency	+++
4	Spreadability	+++
5	Feel	+++
6	Odour	+++
7	pH	6.6

Table no 8

Abbrevation

‘’+’’= poor, “++”=good, “+++= Satisfactory

Form the above table of parameter has required property and has selected as a base formulation.

RESULT AND DISCUSSION:

To assure the consistency and quality of the product the analytical parameters play an important role. Analytical Process also gives brief idea about formulation. Mostly Analytical parameters for clay based body masks are pH, Spreadbility.

A.In-vitro Evaluation

a)Determination of Physical Parameter

Appearance - Visual appearance of the formulation was observed.

Colour - Colour of the formulation also checked visually.

Consistency - Consistency was also checked whether it feels tacky or not.

Spreadability - If its Spreadable or not.

Result:

Sr. No	Physical parameters	F1	F2	F3
1	Appearance	Smooth Paste	Smooth Paste	Smooth Paste
2	Colour	Light Cream	Cream	Cream
3	Spreadability	Good	Good	Good
4	Consistency	Good	Better	Good
5	Odour	Pleasant	Pleasant	Pleasant

Table no 9

b) Determination Of pH

Body Masks are used for topical application, So their pH should be similar to that of the skin. To Ensure the required shelf life of clay mask, chemical inertness is essential i.e it should neither be too acidic nor too alkaline. Based on above point it was through the standard ph of skin should be in the range of 5.5-7.0. The Skin has acidic mantle and the pH of the body mask as per the standards should be in the range of 5.5 - 7.0

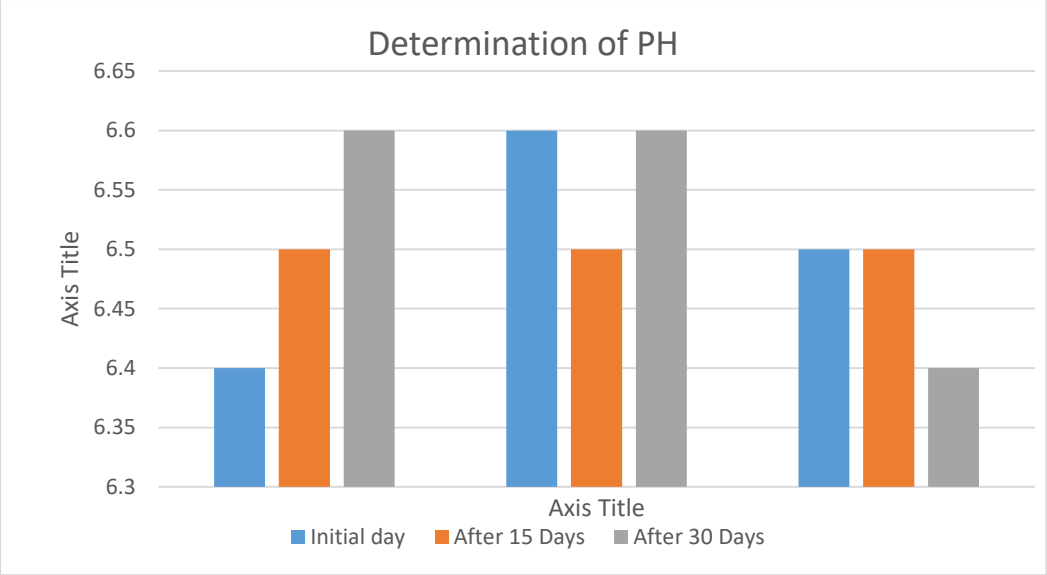
Procedure:- 2gm sample was taken in a 50 ml beaker and 8 ml distilled water was added and stirred. Then pH rod was dipped in the sample and the readings were noted pH was measured at 25°C.

Result:

Sr.No.	Time interval	F1	F2	F3
1	**Initial Day**	**6.4**	**6.6**	**6.5**
2	**15th Day**	**6.5**	**6.5**	**6.5**
3	**30th Day**	**6.6**	**6.6**	**6.4**

Table no 10

Determination of pH with graphical representation



From the above graph we can conclude that pH of the product at RT, 5 °C, 45 °C are comparatively equal (the pH Range is acceptable) In three Months as compares to initial value. The Product passed in PH value.

c) Determination of Spreadability Time:

Principle:- It is very important for any cosmetic product that after application the product must be easily spread over the skin. Spreadability is affected by many factor such as temperature, viscosity etc. The spreading time must be very less. The apparatus consists of a wooden block, with a movable glass slide with one end tied to weighted pan rolled on pulley.

Procedure:- 2 Gm Of sample was placed on a surface. A slide was placed on a surface. A slide was attached to a pan to which 20 gm of weight was added. The time seconds required to separate the upper slide from surface was taken as a measure of spreadability.

Result:-The product has good feel with excellent spreadability.

d) Accelerated Stability Studies

The purpose of stability testing of cosmetic product is to ensure that a new or modified products meets the intended physical, chemical quality standards as well as functionality and aesthetics when stored under appropriate conditions.

Because the development cycle of cosmetic manufacturer should design their products is relatively short each own stability testing program economically reasonable and efficiently address the testing required.

Because of the wide variety of cosmetic products 'standard' stability test cannot be prescribed. Manufacturers require the flexibility to modify testing protocols and to build a sound scientific basis for assessing stability of their own products. Thus. specific tests may be developed in order to be adapted to products having extended shelf lives. Stability tests can be conducted in real time or under accelerated conditions and should address the stability of a product under appropriate condition of storage, Transport and use.

Basically, there are three forms of stability tests:

Physical and chemical integrity tests which evaluate color, odour, pH value, Texture, flow and stability, Microbiological stability tests which evaluate the degree of contamination with bacteria, mold and yeast and packaging stability tests which evaluate the impact of packaging on the contained product.

Physical and Chemical Stability tests

This describe to approaches to predicting how well cosmetics will resist common stresses such as temperature extremes and light. Typically, manufacturers determine whether to perform

such specialized testing based on the vulnerabilities of the particular cosmetics products and its anticipated shipping, storage, display and use condition.

Common test Procedures include :

1. Temperature Variations:

High temperature testing is now commonly used as a predictor of long term stability. Most Companies Conduct their high temperatures testing at 37° C. and 45°C. If a product is stored at 45 °C. for three months (and exhibits acceptable stability) then it should be stable at room temperature for three months and for excellent stability product must be stored at 25° C for a period of two years. The product should also be subjected to - 5 °C for three months.

2. Freeze Thaw Stability testing :

During Transportation of cosmetic products, it is uncommon for them to encounter extreme temperature conditions, such as freezing or over -Heating, thus it is necessary for cosmetic products to be able to withstand a certain degree of temperature changes in transport. Freeze thaw cycle testing is a part of stability testing that allow you to determine if your formula will remain stable under various conditions. This type of test puts your sample through a series of extreme, rapid temperature changes that it may encounter during normal shipping and for liquid based cosmetics.

These products may experience phase separation that can negatively may experience phase separation that can negatively affect the intended function.

Procedure:-Freeze thaw testing is conducted by exposing the product to freezing temperatures (approximately sssss. 10° C) for 24 hrs, and then allowing it to thaw at room temperature for 24 hrs. The Sample is then Placed in a higher temperature (approximately 45 ° C) for 24 hrs. The Sample is then analysed for significant changes are observed, you can be confident that the stability of product is sufficient for transport.

Report of Accelerated Stability Studies:

❖ Freeze thaw Cycle

Representation of freeze thaw cycle

Temperature	Cycle 1	Cycle 2	Cycle 3
RT	NC	NC	NC
5^0C	NC	NC	NC
45^0C	NC	NC	NC

NC= No Change Table no 11

Result: From the above chart we can conclude that the product has passed the Freeze - Thaw cycle. It includes the appearance, Colour and odour of the product at RT, 5° C, 45° C.

❖ Temperature Variation test (Thermal stability test)

Result: From the above three graphs and freeze- thaw cycle, we can conclude that the product has passed thermal stability test. Because at all the temperatures the product remains constant in pH, Appearnce and spreadability as well as in colour and odour.

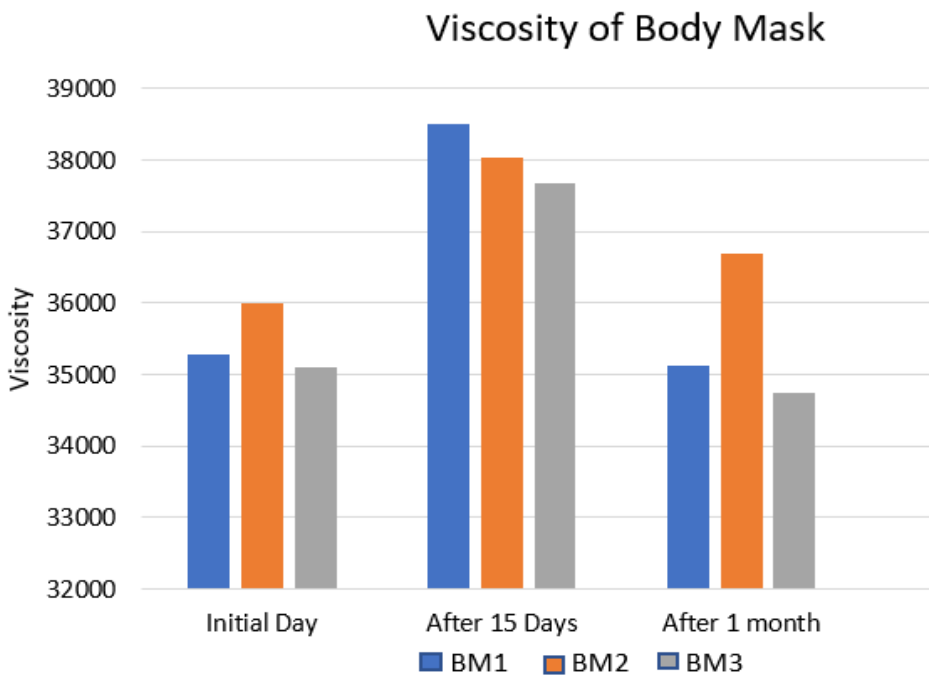
e) Determination of viscosity

The viscosity of Body mask determine by using Brookfield Viscometer.

The values obtained from the sample noted.

Sr. No	**Intervals**	**BM1**	**BM2**	**BM3**
1.	Initial Day	35280cp	36010cp	35100cp
2.	15^{th} Day	38500cp	38040cp	37680cp
3.	30^{th} Day	35120cp	36700cp	34750cp

Table no 12



B. In-vivo Evaluation

a) Determination of Patch Test

Patch test was performed on sensitive part of skin , eg. Blend of elbow. Popliteal space of skin behind ears. The Cosmetic was tested by applying to an area of 1sq. cm of skin. Central patches were also applied. The site of the patch was inspected after 24 hrs. There were no reactions and then test was repeated once more on the same side Since there was no reaction as the person was considered as not hoper sensitive and product pass the test.

Patch testing is usually used to detect allergic reactions to substances such as poisonous. ie. household chemicals, metals and their substances. It May be also be used to diagnose food allergies. Using this technique, we can test for allergies to as many as 60 different substances at the same time.

The test is very simple and accurate. First we will examine and clean the area to be tested . which is usually the upper back. Small samples of potential allergens are placed on our skin, and secured with a waterproof, medical tape. The patches well sealed, so they should not interfere with normal daily activities.

We will examine the tested skin and record data after you have worn the patches for 48 hrs and again 72 hrs. The information collected will help us determine if allergies to an of the tested substances are likely to be responsible for your symptoms.

Patch testing is painless,and causes minimal discomfort. However. You may experience itching under the patch. If you are allergic to any of the tested substances you may develop redness, small welts,or ocassionally small blisters. However, these are easily treated because a very small amount of the allergen is applied, and the area of contact is carefully confined by the small patch. A Reaction to the patch test is a very good indication that we have identified the source of the problem, which is the most important step in solving.

Patch test for Clay mask:

N.R= No Reaction

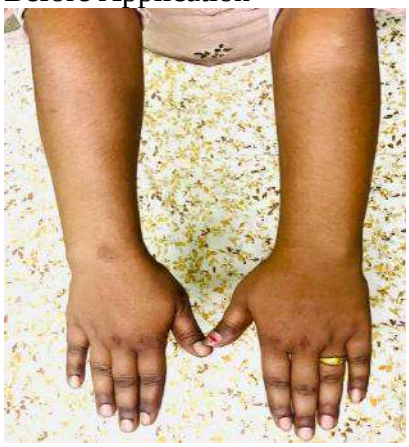
Sr. No	Parameter	F1	F2	F3

1	Immediate after removal of product	NR	NR	NR
2	After 24 hours	NR	NR	NR
3	After 48 hours	NR	NR	NR

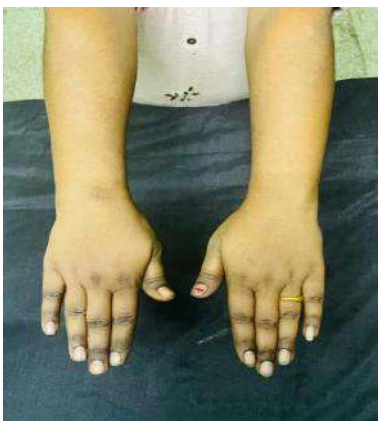
Table no 13

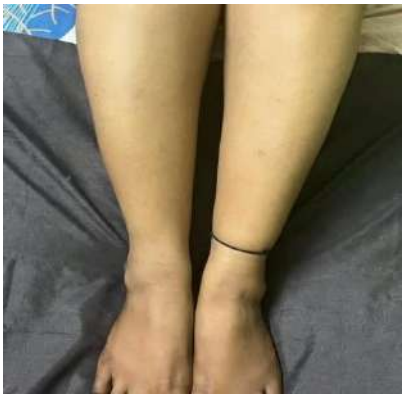
b) Photographic Evaluation:

Before Application



After application





c) Determination of moisture content of skin by corneometer

NOTE:- Only the best batch out of the three batches is tested by corneometer.

Priciple: Corneometer is device which is equipped with a moisture sensitive probe which is used to determine the accurate moisture content of stratum corneum. Hence it plays important role in determining the moisturizing activity of product on stratum corneum after its application on skin.

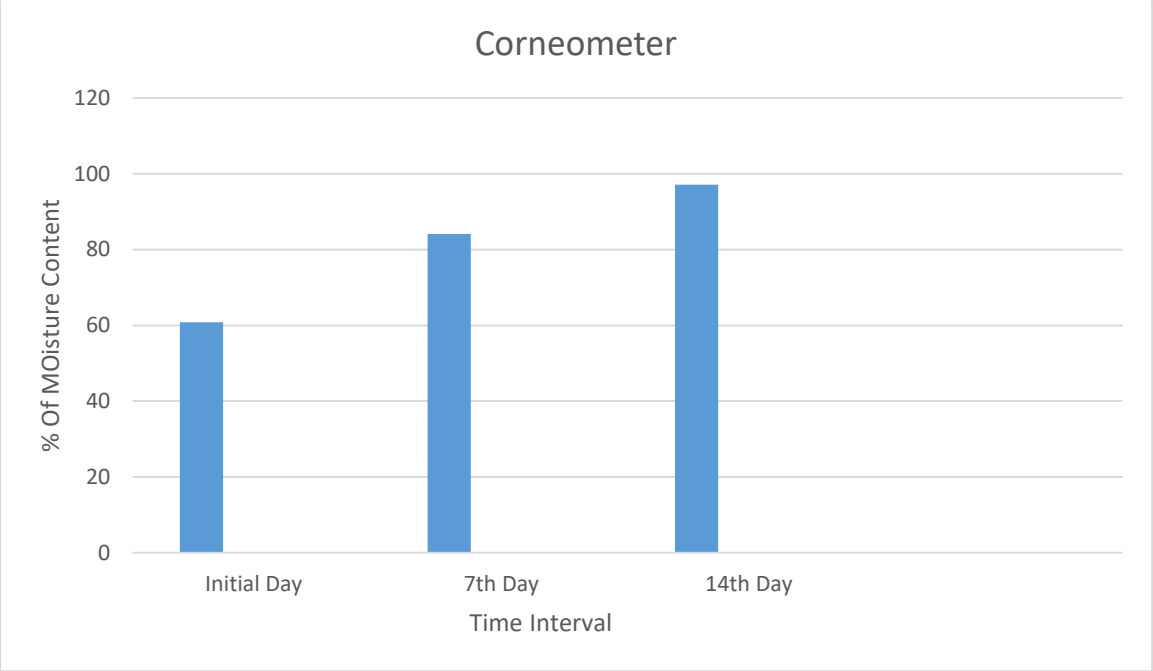
Apparatus: Corneometer equipped with a probe.

Procedure: The volunteers were selected and the probe of corneometer was applied onto the selected part of skin before application of product and the reading was recorded. Then the selected part of skin was rinsed with product allowed to dry properly and again the probe was applied onto the skin and reading was recorded. The voluntree were allowed to wash the selected area of skin after application of the product twice a Day and then same procedure was followed 14 days. Within these intervals the recorded after 7^{th} Days and then finally on 14^{th} Days and the graphs were plotted.

Result:

Sr. No.	Time Interval	% of Moisture Content
1	Intial	60.8
2	After 7 Days	84.1
3	After 14 Days	97.1

Table no 14



Analysis of Moisture Content using Coreometer:

The moisturising activity was carried out by using coreometer. It was observed that before application of body mask, the moisture content of skin was less and after application of body mask moisture content was increased.

CONCLUSION:

A body mask enriched with active Dead Sea mud presents a holistic skincare solution. With its potent blend of minerals, the mask offers deep cleansing, exfoliation, and nourishment for the skin. The application of this mud mask can contribute to a revitalized and smoother complexion, showcasing the benefits of harnessing the natural elements found in the Dead Sea for a luxurious and effective skincare experience. Furthermore, the mask's ability to detoxify and hydrate the skin adds to its appeal, making it suitable for various skin types. Regular use may assist in promoting overall skin health, addressing issues such as impurities and uneven texture. The incorporation of active Dead Sea mud in a body mask aligns with a natural, mineral-driven approach to skincare, providing users with a rejuvenating and spa-like experience from the comfort of their own routine.

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Formulation, Development and Evaluation of Manicure - Pedicure Soaking Liquid

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Abstract
The aim of present work was to formulate, develop and evaluate Manicure-Pedicure soaking liquid with aqueous dispersion of silver particles as active, since it is reported to give anti-bacterial and antifungal effect. Manicure and pedicure are one of the most important skin care treatments which are gaining significant popularity in recent years.The idea of formulation of manicure-pedicure soaking liquid was conceptualized by observing that there are more than one component which are required in 1st step of manicure and pedicure for soaking hands and feet in warm water. Hence, the present study was undertaken with the aim to formulate and develop one single product instead of using various products for soaking purpose. Suitable manicure-pedicure soaking liquid was developed with 2% aqueous dispersion of silver particles and final formulation(Trial II+active) was evaluated for parameters such as color, odor, pH, foaming power, cleansing ability, antimicrobial activity, moisturizing ability and accelerated stability study. Subjective evaluation was carried out to study the functional parameters of final formulation like, cleansing ability, moisturizing effect, softening and cooling property, and irritancy on human volunteers. The study showed that the manicure-pedicure soaking liquid with 2% aqueous dispersion of silver particles, when added to soaking water gives satisfactory cleansing ability, moisturization, softness and cooling effect on skin of hands and feet without any irritation.

Keywords: Aqueous dispersion of silver particles, cleansing ability, soaking ability, antimicrobial activity, Manicure- Pedicure Soaking liquid

1. Introduction

Well-maintained hands and feet are a true reflection of one's personality but they are most neglected parts of the body. Athlete's foot, corns, in growing toe nails, cracked heels, abrasion, dryness, eczema, itching etc. are some common examples of their disorders.Hands and feet are continuously exposed to a lot of friction, temperature differences, pollution, air, light and pressure. Hence,it is important to take good care of them to keep them healthy and beautiful[1].Generally, in manicure and pedicure procedure there are multiple components required in 1st step for soaking hands and feet in warm water such as hydrogen peroxide, antiseptic liquid, shampoo, tea tree oil, epsom salt, baking soda, sugar, white vinegar etc.[2].Aqueous dispersion of silver particles reported to have antibacterial and anti fungicidal properties[3]. For manicure pedicure procedures one single soaking liquid is not very popular, may be due to lack of awareness. Hence, the present study was undertaken with the aim to formulate and develop Manicure-Pedicure Soaking liquid with aqueous dispersion of silver particles as a skin care cosmetic product to be added in the soaking water which is the requirement of manicure-pedicure treatments. which can be used to clean the dirt, oil, germs, dead cells and also to kill microbes. This soaking liquid can prove to be all in one product, which will be more convenient over multiple components with similar results. It can stimulate blood circulation, repair cracked heels, prevents epidermal thickening and gives relaxing and cooling effect to hands and feet[4].

2.Materials and Methods

2.1.Analysis of active ingredient[Aqueous dispersion of silver particles

Aqueous dispersion of silver particles was procured for present study from Nano Tech Chemical Brothers Private Limited,Ludhiana, India, along with Certificate of Analysis. The procured sample was validated for parameters such as color, odor, pH, solubility, particle size and silver content.

2.2. Formulation and development of Manicure- Pedicure Soaking Liquid

Two different base formulations of Manicure-Pedicure soaking liquid (i.e., Trial I and Trial II) were formulated.Trial II was selected for incorporation of 2% Aqueous dispersion of silver particlesas active as it was found to be satisfactory and this trial II with active was selected for further study. (Table No.1)

S.N.	Ingredients	Trial I [Base]100%	Trial II [Base]100%	Trial II +Active [100%]	Use of Ingredients
1	SodiumMethyl Cocoyltaurate	38	38	38	Surfactant
2	Cocamidopropyl betaine	1	1	1	Foam Booster Stabilizer
3	Water	10	11.1	9.1	Solvent
4	Glycolic Acid	20	20	20	Exfoliant and Antioxidant
5	Germall Plus Liquid	0.5	0.5	0.5	Preservative
6	Hydroxyethyl Urea	20	20	20	Humectant
7	L-Glutathione	0.6	0.6	0.6	Skin Lighting agent
8	Aqueous dispersion of silver particles	--	--	2	Antimicrobial agent
9	Menthol	1	0.5	0.5	Cooling agent
10	Perfume[Japanese Cherry Blossom]	8.9	8.3	8.3	Fragrance
11	Color [Golden Yellow] q.s. – Quantity sufficient	q.s.	q.s.	q.s.	Coloring agent

Table No 1: Formulation of Manicure- Pedicure soaking liquid with Aqueous dispersion of silver particles

2.3 Analysis of Manicure-Pedicure soaking liquid[5,6]

Manicure -Pedicure soaking liquid (Trial II+2%Aqueous dispersion of silver particles) was subjected to analysis for parameters likecolor, odor, pH, foaming power, cleansing ability, antimicrobial activity and moisturizing ability.The results are summarized in Table no.2**.**

2.4 Evaluation of Antimicrobial Activity of Aqueous dispersion of silver particles and Manicure-Pedicure Soaking Liquid [7,8]

The evaluation of anti-microbial activity of active, final product (i.e. Trial II+2%Active), its dilutionsi.e.,1.5% dilution(with water) of final product and 3% dilution(with water)of final product was done by Agar Well Diffusion Method. The study was carried out against microorganisms namely *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*,

Proteus, Aspergillus niger, Bacillus subtilis and Candida albicans .The results are summarized in Table No.3.

2.5 Accelerated stability study [9,10]

The objective of stability study is to ensures the maintenance of product safety, quality and efficacy throughout the shelf life.The data generated during the stability testing is an important necessity for regulatory approval of any formulation.Such information can be helpful in terms of creating the successful product and product development.TheManicure-Pedicure soaking liquid(i.e.Trial II +2% Active) was subjected to accelerated stability study.Changes in parameters like colour, odour, pH at three temperatures i.e., in oven [±45°C],in refrigerator [±4°C] and at room temperature was recorded for 30 daysat the interval of 5 days.

2.6 Subjective Evaluation [11]

To study the effectiveness of Manicure- Pedicure soaking liquid with aqueous dispersion of silver particles subjective evaluation was carried out on the panel of human volunteers.Final product i.e.Trial II with 2% aqueous dispersion of silver particleswas given to 30 volunteers and evaluation was carried out on the basis of their feedback.Parameters such as, cleansing, softening, cooling effect, moisturization by using corneometer and irritancy were evaluated.

3.Results and Discussion

3.1.Analysis of Aqueous dispersion of silver particles

Analysis of aqueous dispersion of silver particlesshowed that procured sample passes the test as per the certificate of analysis and hence it was used for incorporation informulation.

3.2.Formulation and development of Manicure-Pedicure Soaking Liquid

Analysis of base formulations (i.e. Trial I and Trial II), on the basis of functional parameters, indicated that, Trial -II gave the satisfactory results, hence was selected for incorporation of aqueous dispersion of silver particles as an active,for further study.

3.3Analysis of Manicure-Pedicure soaking liquid From the results of analysis of Manicure-Pedicure soaking liquid (Trial II with 2% aqueous dispersion of silver particles), it was observed that product was satisfactory with respect to all parameters.(Table 2)

S.N.	Parameters	Results
1	Color	Golden Yellow
2	Odor	Pleasant
3	Foaming Power	110mm
4	Cleansing ability	Satisfactory
3	pH of Product	4.60

Table No.2 Analysis of Manicure-Pedicure soaking liquid

3.4Evaluation of antimicrobial activity of active (aqueous dispersion of silver particles) and Manicure-Pedicure Soaking Liquid

From the analysis of antimicrobial activity of manicure pedicure soaking liquid with aqueous dispersion of silver particles it was observed that the product shows its potency as an antimicrobial agent and exerted an inhibitory effect against bacteria and fungi. The results are shown in Table No. 3.

S.N.	Name of Microorganisms	Zone of Inhibition			
		Active	Product 100%	1.5% dilution (with water) of final product	3% dilution (with water) of final product
	Bacteria				
1.	*Escherichia coli*	15mm	26mm	10mm	11mm
2.	*Staphylococcus aureus*	19mm	39mm	10mm	11mm

3.	*Pseudomonas aeruginosa*	10mm	36mm	10mm	18mm
4.	*Proteus*	10mm	23mm	10mm	13mm
5.	*Bacillus Substilis*	10mm	17mm	10mm	10mm
	Fungi				
6.	*Aspergillus niger*	17mm	25mm	10mm	10mm
7.	*Candida albicans*	10mm	19mm	12mm	10mm

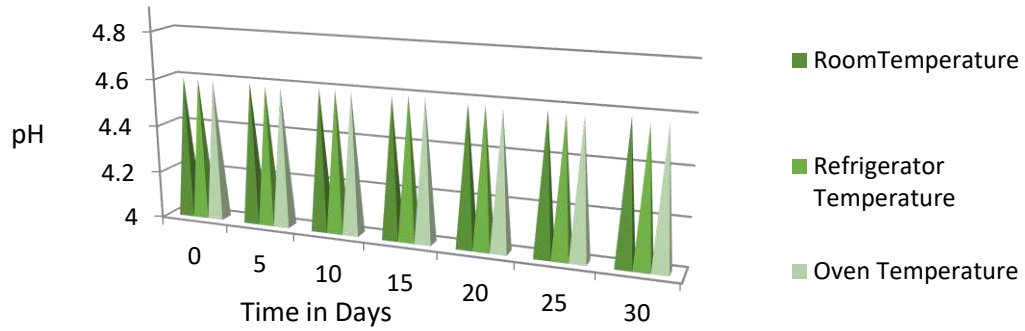
TableNo.3: Evaluationof antimicrobial activity of active, Manicure-Pedicure Soaking Liquid and its dilutions(Cork Borer Size 8 mm)

3.5Accelerated stability study

From the results of accelerated stability study, it was observed that the Manicure-Pedicure Soaking Liquid(i.e.Trial-II+2% active) was stable with respect to physical parameters such as color, odor and pH at all the three different temperatures i.e. at 4^0C,at room temperature and at 45^0C. Table No. 4 ,Graph No. 1.

Sr.No.	Parameters	Oven[±45 °C]	Refrigerator[±4°C]	Room Temperature
1.	Odour	No Change	No Change	No Change
2.	Colour[Golden Yellow]	No Change	No Change	No Change

Table No.4: Result of Accelerated stability study



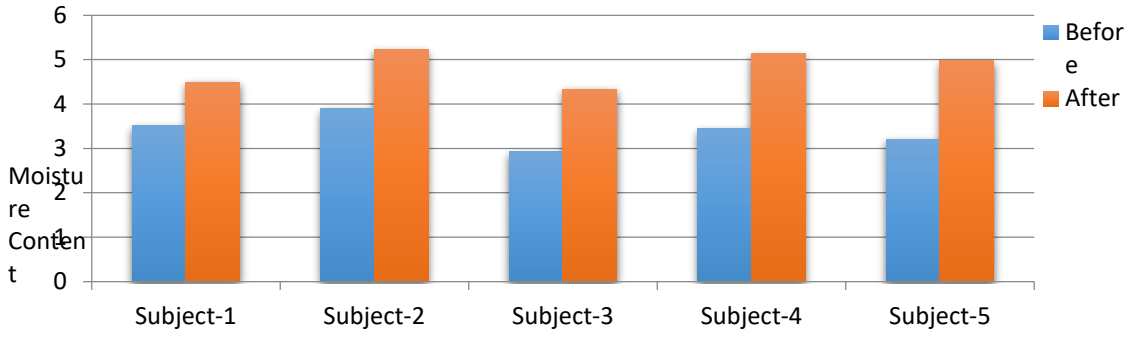
Graph No. 1. Graphical representation of change in pH of final product.

3.6 Subjective Evaluation

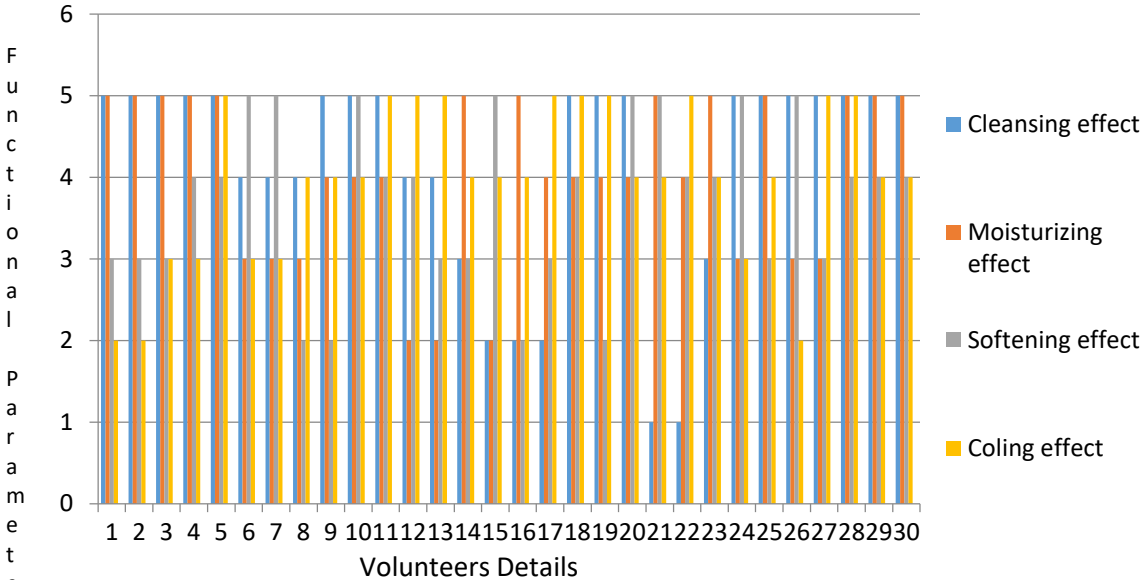
From the results of subjective evaluation, it was observed that the Manicure-Pedicure Soaking Liquid– final product (i.e. Trial II+ 2% active) was well appreciated, it showed satisfactory cleansing, softening, cooling effect,without any irritancy on skin of hands and feet. Also, from the analysis of moisturizing effect on skin by using Corneometer it was observed that the moisture content of the skin was improved satisfactorily after using the product. Table No.5, Graph No. 2, Graph No. 3

S.N.	Subjects	Before	After
1.	Subject-1	3.51%	4.49%
2.	Subject-2	3.89%	5.23%
3.	Subject-3	2.92%	4.33%
4.	Subject-4	3.45%	5.14%
5.	Subject-5	3.2%	4.98%

Table No.5: Analysis of Moisture Measurement by using Corneometer.



Graph No. 2. Graphical representation of Moisturizing effect of final product



Graph No. 3. Graphical representation of subjective evaluation

4.Conclusion From the above study, it can be concluded that Manicure-Pedicure soaking liquid with aqueous dispersion of silver particles is a beneficial product for addition into soaking water of manicure pedicure procedures, as it removes dirt, dead skin from hands and feet and moisturizes them with cooling effect. It has no harsh effects on skin and can remove bad odor from feet and hands. The Manicure-Pedicure soaking liquid is all in one product instead of using multiple products. Manicure Pedicure soaking liquid with aqueous dispersion of silver particles will be more convenient product and it will be a useful addition in the portfolio of manicure- pedicure kit.

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Formulation and Development of Hair Removal Spray

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Abstract :
The aim of the present work was to study the formulation and development of Hair removal spray. Teenage girls and women are often concerned about the unwanted hair development. The idea of formulation was conceptualized by observing that hair removers are the products designed to eliminate or reduced unwanted body hair. They come in various forms including creams, waxes and electronic devices which are very messy solution for hair removal. Hence, the current study highlights the importance of choosing easy and convenient method for hair removal by using spray with Potassium Thioglycolate. It is an organic substance which is main active component in hair depilatories. Because it may have ability to weakening the hair protein structure and allowing for effortless hair removal. Suitable hair removal spray was developed with three different concentration of potassium thioglycolate and final formulation with 4% was evaluated for parameters such as color, odor, consistency, thermal stability, pH and accelerated stability study. Subjective evaluation was carried out to study the functional parameters of final formulation like, hair removal efficacy, time required for hair removal and skin irritation on human volunteers. The study showed that the hair removal spray with 4% Potassium Thioglycolate gives satisfactory hair removal activity, minimum time required for hair removal and no irritation on skin.
Keywords: Potassium Thioglycolate, Efficient hair removal, Removal time, irritation, Hair Removal spray.

1.Introduction :

The cosmetics, hygiene industry and the hair removal market are growing continuously. For many people, hair is a natural part of their look and an expression of their personality. Human hair grows at a rate of 0.35 mm/day, and around 100 hair are shed daily. Hair is a filamentous, usually pigmented out growth from the skin, found only on mammals [1]. Hair can be found on all areas of the skin except the lips, finger tips, palms and soles. Hair removal is an increasingly important sector of the cosmetic and personal care industry. Both men and women are becoming more concerned about the aesthetic aspect of their appearance [2]. Human being choose to remove unwanted body hair for cosmetic, social, cultural or medical reasons [3]. The number of hair removal techniques has been developed over the years, including methods for temporary and permanent hair removal, which are very messy and painful [4]. Hair removal spray is a modern and mess free solution for effectively removing unwanted body hair. It is designed to provide convenient way and ease of use. This product has an alternative to traditional hair removal methods such as shaving, waxing and plucking [5]. Hair removal spray are chemical depilatory creams in form of spray which contain chemicals like Potassium Thioglycolate that help in removing unwanted body hairs by dissolving the disulfide bonds between the keratin proteins in the hair [6]. Hair removal Spray is a more convenient way to get rid of hair because it's quick, painless, and efficient.

2. Material and Methods

2.1 Analysis of Potassium Thioglycolate

Potassium thioglycolate was procured for the present study from RP chemicals Kalyan, Mumbai, India, along with Certificate of Analysis. The procured sample was validated for

parameters such as Color, assay, pH, specific gravity, water, Iron content.The result are summarized in table no. 2

2.2 Formulation and Development of Hair Removal spray

Three different formulations of Hair removal Spray (i.e. Trial I, II and III) were formulated with three different concentration of Potassium Thioglycolate i.e. 2%, 3%, and 4% respectively. Since formulation Trial III with 4% potassium Thioglycolate gave a satisfactory hair removal with no skin irritation and required minimum time for hair removal. Hence, it was selected for further study (Table No.1). The result are summarized in table no. 3

S. N.	Ingredients	Use of Ingredient	Trial I (Quantity in %)	Trial II (Quantity in %)	Trial III (Quantity in %)
Phase A					
1.	Gaur Gum	Conditioner	0.5 g	0.8gm	0.8gm
2.	Urea	Emulsifier	7 g	10gm	10gm
3.	Methyl cellulose	Thickener	1 g	1gm	1gm
4.	Sodium Benzoate	Preservative	0.1 g	0.1gm	0.1gm
5.	Glycerine	Humectant	5 ml	7ml	7ml
6.	DM water	Vehicle	Upto 100 ml	Upto 100 ml	Upto 100 ml
Phase B					
7.	Emulsifying Wax	Emulsifier	-	0.4gm	0.4gm
8.	Cetostearyl Alcohol	Emulsifier	3.5 g	3.5gm	3.5gm
9.	Liquid paraffin	Emollient	1 ml	2ml	2ml
10.	Jojoba Oil	Emollient	-	2ml	2ml
11.	Potassium hydroxide	Increases hair loss	4 g	3gm	3gm
12.	Potassium Thioglycolate	Increases hair loss	2ml	3ml	4ml
13.	Perfume (Lavender)	Odor	0.5 ml	0.5ml	0.5ml
14.	Propellant (Butane)	Propellant	8%	10%	12%

Table No.1: Formulation and Development of Hair Removal spray

2.3 Analysis of Hair Removal Spray

Hair removal spray formulation (Trial III with 4% Potassium Thioglycolate) was subjected to study parameters like Appearance , Odor, Consistency, Thermal stability, pH, Hair removal efficacy, Time required for hair removal and Skin irritation. The result are summarized in table no.4.

2.4 Stability Study

The objective of stability study is to ensure that product will remain stable till the consumer has used the entire product. The stability not only indicates stability of formulation but also the stability of other ingredients present in the formulation of hair removal spray. After analyzing all the three formulations, on the basis of functional parameters, it was observed that the hair removal spray formulation (i.e. Trial III with 4% Potassium Thioglycolate) was giving satisfactory results. Hence the Trial III with 4% Potassium Thioglycolate was subjected to accelerated stability studies. Changes in parameters like Color, Odor, pH at three different temperatures (i.e. in oven at (45^0C), in refrigerator at (4^0C) and at room temperature) was recorded for 45 days at interval of two days [7].

2.5 Subjective Evaluation

To study the efficacy of hair removal spray with Potassium Thioglycolate and its effects, subjective evaluation was carried out on the panel of human volunteers. Trial III containing 4% Potassium Thioglycolate was given to 30 volunteers and evaluation was carried out on the basis of their feedback for parameters like Appearance, Ease of spreadbility, Hair removal efficacy, Time required for hair removal and Skin irritation.

3. Result and Discussion

3.1 Result of Analysis of Potassium thioglycolate

From the analysis of Potassium thioglycolate it was observed that procured sample passes the test as per Certificate of Analysis and hence was used for incorporation in formulation. The result are summarized in table no. 2

S.N.	Parameters	Requirment as per C.O.A of Potassium thioglycolate	Result	Inference
1.	Appearance	Colourless to light pink colour liquid	Colourless to light pink colour liquid	Passes the test
2.	Colour	Colourless to light pink colour liquid	Colourless to light pink colour liquid	Passes the test
3.	Assay	30-31	Complies	Passes the test
4.	pH	6.5 -8	Complies	Passes the test
5.	Specific Gravity @ 25º C	1.250-1.255	Complies	Passes the test
6.	Water	0.3% MAX.	0.09%	Passes the test
7.	Iron	5 ppm MAX.	Pass	Passes the test

Table No. 2: **Result of Analysis of Potassium thioglycolate**

3.2 Result of Formulation and Development of Hair Removal Spray

In the present study after analyzing all the three formulation (i.e. Trial I, II and III)on the basis of functional parameters it was observed that Trial III with 4% Potassium thioglycolate was giving satisfactory results. Hence, hair removal spray (Trial III with 4% Potassium thioglycolate) was selected for further study.

S.N.	Formulations	Observation	Changes to be made
1	Trial-I	Base was not stable	Emulsifying agents were increased
2	Trial-II	Hairs was not remove properly	Concentration of active increased
3	Trial-3	Hairs was removed	Formulation was selected for further study

Table No. 3 Result of Formulation and Development of Hair Removal Spray

3.3 Result for Analysis of hair removal spray

From the result of analysis of hair removal spray (Trial III with 4% Potassium Thioglycolate) it was observed that the spray was giving satisfactory results with respect to all the parameters.

S.N.	Characteristic	Requirements	Results
1.	Appearance	Creamy White	Creamy White

2.	Odor	Good	Good
3.	Consistency	Semisolid	Semisolid
4.	Thermal Stability	Stable	Pass the test
5.	pH	11.0 to 12.7	12.5
6.	Hair Removal Efficacy	Satisfactory	Satisfactory
7.	Time required for hair removal	10 minute	7 Minute
8.	Skin Irritation	No Irritation	No Irritation

Table No.4: Result for Analysis of hair removal spray

3.4 Result of stability study

From the result of stability study, it was observed that the hair removal spray (Trial III with 4% Potassium thioglycolate) was stable with respect to physical parameters such as colour, odour and pH at three different temperatures i.e at 4^0C, at room temperature and at 45^0 C.

3.5 Subjective Evaluation

From the above results of evaluation, it was observed that the hair removal spray (i.e. Trial III with 4% Potassium Thioglycolate) was well appreciated. It showed satisfactory hair removal efficacy, with minimum time required for removing hair without any irritancy on skin.

4. Conclusion

From the above study it can be concluded that the hair removal spray Trial III with 4% potassium thioglycolate is giving satisfactory results in terms of appearance, odor, consistency, and it removes hair in gentle way with no skin irritation.

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A Comparative Study of Consumer Awareness towards Composition of Different Mehandi Types Available In The Market: A Survey

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ABSTRACT:
In India Mehandi has been used cosmetically and medicinally for over the years. The relation between Mehandi and cosmetics lies in their shared purpose of beautification. Concerning Mehandi, at least seven different types are reported, based on where they were grown, its parts used and products with additional ingredients. While natural Mehandi is generally safe, some commercial Mehandi products may contain harmful additives such as para-phenylenediamine (PPD) to produce darker colours quickly. PPD may cause severe allergic reactions and skin damage. The problem lies in educating users about the potential risks involved while using such products, promoting the use of natural and safe Mehandi and ensuring proper labelling and regulation of Mehandi products. The survey was conducted with the help of Google form and analysing the collected data, with the help of correlation analysis and Cronbach's alpha test. The survey results rejected null hypothesis and supported the alternative hypotheses. The findings revealed a significant knowledge gap among consumers regarding Mehandi composition and potential hazards associated with it. Thus, the present study indicates the need for improved labelling of readymade Mehandi, education initiatives regarding Mehandi composition and awareness campaigns to enhance consumer knowledge and promotion of safe Mehandi product usage.
Keywords: Mehandi, Cosmetics, Paraphenylenediamine, Labelling, Mehandi Composition

1. INTRODUCTION [1, 2]:

The benefits of Mehandi are mentioned in abundance in the history of India. As reported in studies, it is described that a fertile soil and moist conditions produce Mehandi plants with low dyeing power which means a low active constituent content, whereas dry and hot condition and an iron rich soil give the opposite results. Nowadays, Mehandi is commonly sold in packages available in grocery stores as a cosmetic to dye hair and body. In most of the market, sale and use of Mehandi are completely unregulated. Usually, Mehandi as a raw material comes from distinct countries, without any investigation of the constituents. Currently control over the selling of Mehandi is complex and difficult to be performed by the authorities. Origin, purity, preparation and utilization, are important aspects for safety and quality of Mehandi. The problem lies in educating users about the potential risks, promoting the use of natural and safe Mehandi and ensuring proper labeling and regulation of Mehandi products. This survey was undertaken with the aim to study the consumer behavior towards awareness of composition of Mehandi and their buying behavior. The study aims to provide useful advice that guarantees consumer safety and well-informed choices while choosing ready-made Mehandi.

2. RESEARCH METHODOLOGY [3,4, 5]:

2.1 Sources of Data: The study is based on both primary data and secondary data.
2.2 Sampling: Convenience sampling method has been used for collecting the response from the respondents.
2.3Data Collection: The data has been collected by using Google form.

2.4Tools for analysis: The statistical tool used for the purpose of the analysis of this study is simple percentage technique and correlation analysis.

Hypothesis:

Null hypothesis H_0: Consumer is aware about the composition of different Mehandi types available in market.

Alternative hypothesis H_1: Consumer is not aware about the composition of different Mehandi types available in market.

Alternative hypothesis H_2: Consumer is not aware about the hazards effect of Mehandi composition.

2.5 Analytical Method [6, 7, 8]:

The aim of this research is to determine the correlation between single dependent variables and different independent variables. A multiple regression model is utilized to test the impact of the independent variable and dependent variable. The demographic factors are included in the analysis to get a good profile of the respondents. In this part a full statistical analysis executed upon the data where first, a correlation analysis between variables is shown to study the relation and the statistical difference between variables. Nevertheless, a hypothesis testing will follow to prove if the relationship between variables is valid.

2.5.1 Reliability and Validity:

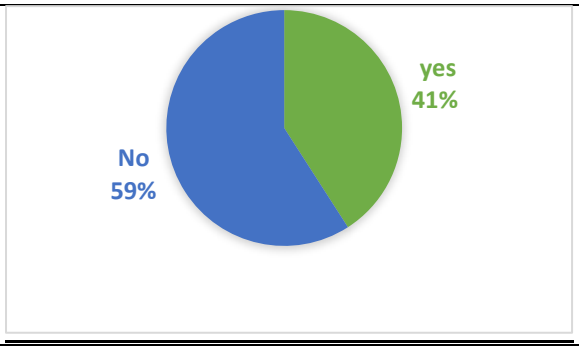
Reliability and validity are used to estimate the quality of the quantitative research. Cronbach's alpha test is used to measure the consistency of responses to a set of questions (scale items) that are combined as a scale to measure a particular concept. It consists of an alpha coefficient with a value between 0 and 1. Validity refers to the examination that satisfies its motivation of the investigation and measures what it planned to gauge or the honesty of the exploration results.

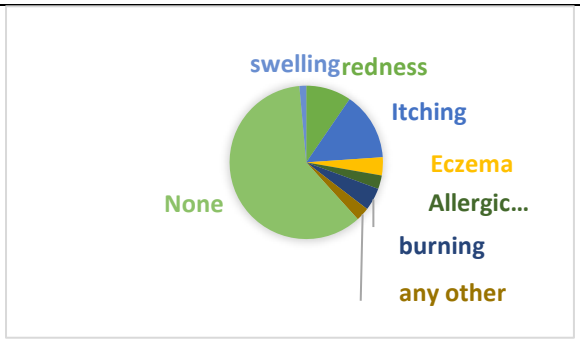
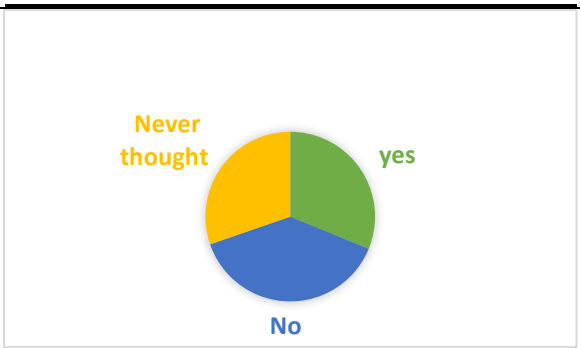
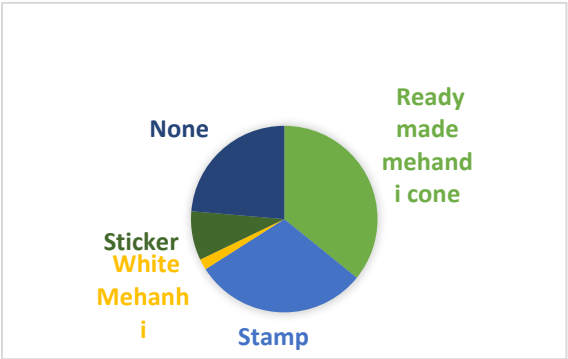
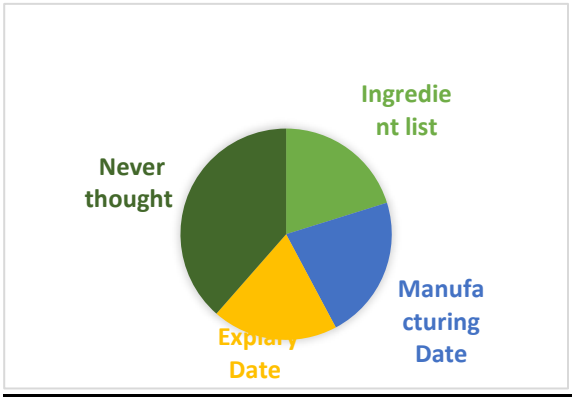
3. RESULT

3.1 Sources of Data: The primary data has been collected by using a questionnaire and the secondary data has been collected from books, magazines and the internet.

3.2 Sampling: A total of 109 respondents were selected for the study.

3.3Data collection: A survey was conducted with the help of Google form link - https://forms.gle/QHRkJmGUeFS8vjUx6 , to capture as many responses as possible. (Table no. 1)

Variables	Theoretical purpose	Survey questions	Graphs of survey findings
Demographic	Demographical information allows to better understand certain background characteristics of an audience	Name Age Gender Occupation Mobile No. City State	
Awareness towards hazard	Mehandi awareness mixed awareness connection with people's behavioral.	Have you ever experienced a skin problem during or after applying mehandi?	

		Which of the following adverse effect did you face after applying mehandi?	
Awareness towards composition	Awareness towards composition is main criteria for skin health	Have you ever thought about the composition of mehandi types? Did you get an irritant smell from which of these types mehandi?	 
Awareness towards parameters	Awareness towards parameter is consumer first right	What labeling parameters do you look for while buying mehandi? Did you judge the quality of mehandi by its rate ?	

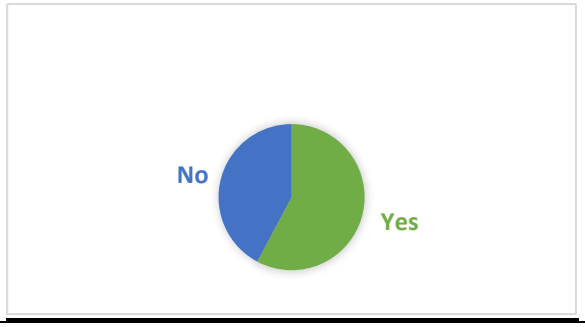
			

Table 1: Data Collection

3.4 Tools for analysis: The data collected was analyzed and interpreted with the help of tables and figures.

Hypothesis: The findings of survey strongly support the validity of alternative hypothesis. (H1- Consumer is not aware about the composition of different Mehandi types available in market).

3.5 Analytical Method:

Hypotheses testing is the technique to recognize whether there is a particular connection between the dependent variable with independent factors.

Correlation analysis and statistical significance

Hypothesis 1

The significant value (P-value = 0.13089) shows the data is not existence of a strong statistically significant correlation between consumer awareness towards composition of Mehandi as the P-value 0.13089 is greater than 0.05 (95% confidence level).

Hypothesis 2

The significant value (P-value = 0.9281) shows the existence of no statistically significant correlation between consumer parameter and their purchase intention towards Mehandi products since the P-value of 0.9281 is greater than 0.05 (95% confidence level).

In the hypothesis both significance values are greater and Positive than the critical value, which is the sufficient evidence to reject null hypothesis and accept the alternative hypothesis.

3.5.1 Reliability and Validity:

a. Standard Value

Alpha Value	Variables
0.90 and above	Excellent
0.80 – 0.89	Good
0.70 – 0.79	Acceptable
0.60 – 0.69	Questionable
0.50 – 0.59	Poor
Below 0.50	Unacceptable

Table no.2: Cronbach's alpha test results of Standard Value

b. Observed Value

Variables	Alpha values
Awareness towards hazard	0.51045
Awareness towards composition	0.84553
Parameter Awareness	0.75702

Table no.3: Cronbach's alpha test results of Observed Value

4. CONCULSION

Mehandi is a decorative product used throughout the world for the purpose of beautification. Most of the time manufacturer may add harmful chemicals to the raw Mehandi to enhance its shade. As per literature study, the health risks associated with such Mehandi products are evident and known. Currently, control measures for this purpose are not properly regulated in India as far as consumer safety is concerned. Hence there is need to create awareness towards Mehandi composition in consumers. The main purpose of this study was to examine consumer behaviour as far as Mehandi products are concerned. It has been done by examining and analyzing the attitudes of different consumers who choose to buy different types of Mehandi. The study further revealed that the different factors affected one's purchasing behaviour towards Mehandi products. Based on the results of the survey analysis, it can be concluded that the null hypothesis is rejected and alternative hypothesis is accepted, which states that consumers are not adequately informed about the composition of different Mehandi types. Hence, from the survey results it can be concluded that the consumers lack the awareness about the composition of different Mehandi types available in the market. It is needed to address this knowledge gap and provide consumers with accurate information to ensure their safety and help decision-making when choosing safe Mehandi products. Thus, the present study indicates the need for improved labeling of ready made Mehandi, education initiatives regarding Mehandi composition and awareness campaigns to enhance consumer knowledge and promotion of safe Mehandi product usage.

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Development of skin moisturizer with proven moisturizing properties of plant stem cell active

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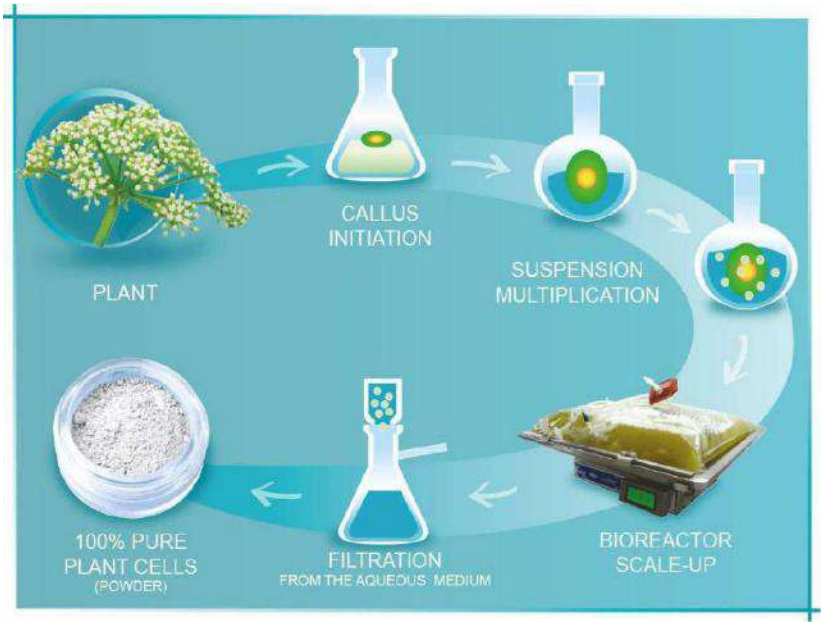
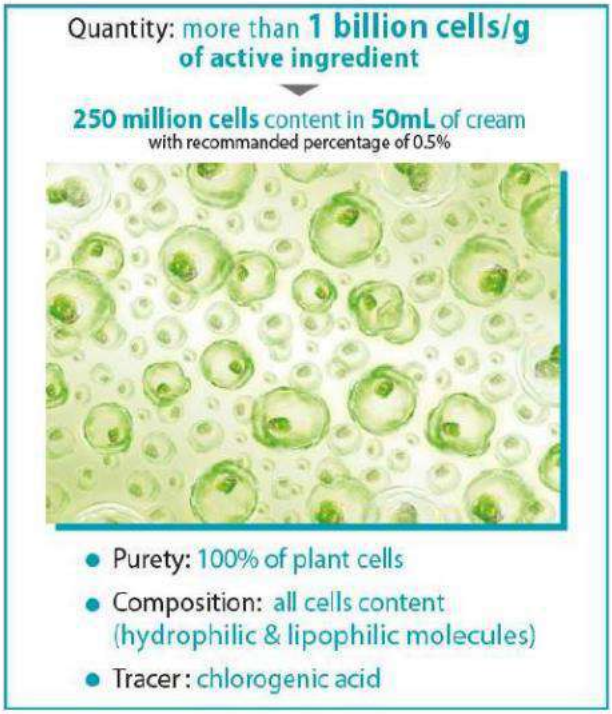
Abstract-
Plant cells have the amazing ability to either differentiate into all cell types, or to self-renew at an undifferentiated stage from which they can regenerate the whole plant. Thorough knowledge of the raw materials & technological expertise allows to cultivate cells at an industrial level in their "native" stage, thus potentially producing any type of compounds of interest.
Plant stem cells located in the meristematic tissue of the plant & serves as the origine of vitality with potency provides consistent supplement of precursor cells to form differentiated tissues and organs in the plant. Plant cell culture technology is a technique for growing of plant cells under strictly controlled environmental conditions & so control on its potent active constituent.

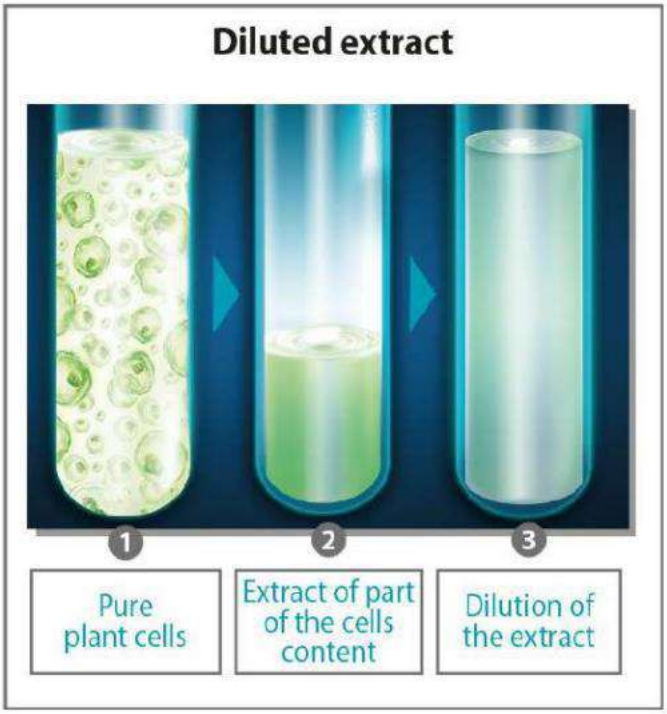
Key words- Plant stem cell, tissues, moisturization, hydration, dryness, emulsion

INTRODUCTION:

Plant cells are undifferentiated "native cells" located in meristematic areas, which can be found at the tip of the roots, at the apex of the stem & in the primordial leaf. These zones are responsible for the growth of the plant.
Plant cells have two key abilities: the ability to differentiate into all cell types found in the plant & the ability to self-renew at an undifferentiated stage. Thus, plant cells are responsible for every organ formation & every differentiation resulting in the growth of the plant.







Benefits of plant cells:

- Plant cells simultaneously manage two metabolisms:

✓primary metabolism, necessary for essential functions (growth, development & reproduction),

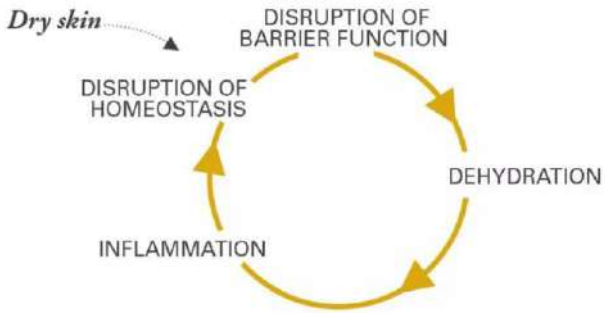
✓secondary metabolism, to adapt to external stress.

- Each metabolism generates specific molecules: primary & secondary metabolites.
- Endangered & Protected Raw Material: Plant Cells Culture: no need to harvest/ sustainable development
- Culture: Laboratory Conditions- Sterile environment: no potential pollution or contamination
- Undifferentiated Cells: Each undifferentiated cell potentially contains all of the plant's components.

Dry skin is characterized by tightness, itching, lack of elasticity and an overall discomfort. The altered barrier function leads to dehydration, impaired lipid production and an inflammation signalling cascade. Recent scientific progress has linked skin dryness to inflammation: this is the **Inflamm'dryness™ phenomenon**.

Breaks the Inflamm'dryness™ vicious circle:

- Manage the resolution of inflammation (decrease of pro-inflammatory and an increase of pro-resolutive mediators)
- Restores the barrier function & homeostasis



The vicious circle of Inflamm'dryness™ phenomenon.

Ingredient selection:

Emulsion: Is a system comprising 2 immiscible phases with the stabilization of emulsifier. 2 phases also called as dispersed phase (internal) & dispersion medium (external/continuous).

Emulsifiers: important constituent for emulsification process also for its stability.

I. Synthetic emulsifiers

1. Anionic surfactants:
2. Cationic
3. Amphoteric surfactants:

II. Natural emulsifiers

III. Semi synthetic emulsifiers

Other formulation additives: Polymers and Viscosity modifier, Emollients, Humectants, Sensory modifiers, Preservatives & Miscellaneous (i.e. UV / photo stabilisers, for avoiding colour fading of product, Colors, pigments, fragrances for aesthetic appeal of product, Sequestering agents, pH regulators like buffer solutions or neutralizing materials)

Active: Aqua (and) Glycerin (and) Helichrysum Stoechas Callus Culture Lysate

Hydrachrysum (Seppic active)- new natural ally of dry skin. Bio-inspired by Helichrysum stoechas, the Everlasting maritime plant which adapts to its arid ecosystem. Hydrachrysum™ has been developed using our stem cell technology. It offers a unique molecular richness made up of hydrophilic & lipophilic molecules derived from the dedifferentiated plant cells and specific ones secreted in the medium.

Hydrachrysum is a patented moisturizing active ingredient that breaks the Inflamm'dryness vicious circle. It induces a **decrease of pro-inflammatory and an increase of pro-resolutive mediators** to allow a return to homeostasis and improved barrier function. Hydrachrysum **increases the number of lacunae**, these markers of hydration acting as extracellular water tanks and representing up to 40% of the volume of the stratum corneum. It boosts skin moisturization after only 5 days by increasing significantly the number of lacunae **+82%* vs placebo (Proven study mentioned by Seppic- Wesource)**.

Hydrachrysum is Cosmos and NaTrue approved, Halal certified, and scientifically proven with in-vitro, ex-vivo and in-vivo data at 1%.

Active:

Sr. No.	INCI	Major benefits	Purpose/useful for
1	Hydrachrysum: Aqua (and)	It offers a unique molecular richness made	Long term moisturization

	Glycerin (and) Helichrysum Stoechas Callus Culture Lysate	up of hydrophilic & lipophilic molecules: • Sugars: polysaccharides of various size, • Phenolic compounds in particular caffeoylquinic derivatives, • Lipids: polyhydroxylated unsaturated fatty acids, for a powerful hydration.	

Formulation Development: with & without active

INCI NAME	**QUANTITY IN % (Placebo)**	**QUANTITY IN % With active**
Water	83.80	82.80
Ethylenediaminetetraacetic acid	0.10	0.10
Glycerin	3.00	3.00
Polyacrylamide (and) C13-14 Isoparaffin (and) Laureth-7.	0.50	0.50
Hydroxyethyl Acrylate/Sodium Acryloyldimethyl Taurate Copolymer	0.80	0.80
Acrylates/C10-30 Alkyl Acrylate Crosspolymer	0.20	0.20
Capric Caprylic Triglyceride	10.00	10.00
Hydrachrysum: Aqua (and) Glycerin (and) Helichrysum Stoechas Callus Culture Lysate	-	1.00
Phenoxyethanol (and) Ethylhexylglycerin (and) Octenidine HCl	1.00	1.00
Perfume	0.10	0.10
Sodium Hydroxide (20% solution)	0.50	0.50
	100.00	100.00

Analysis Data: -

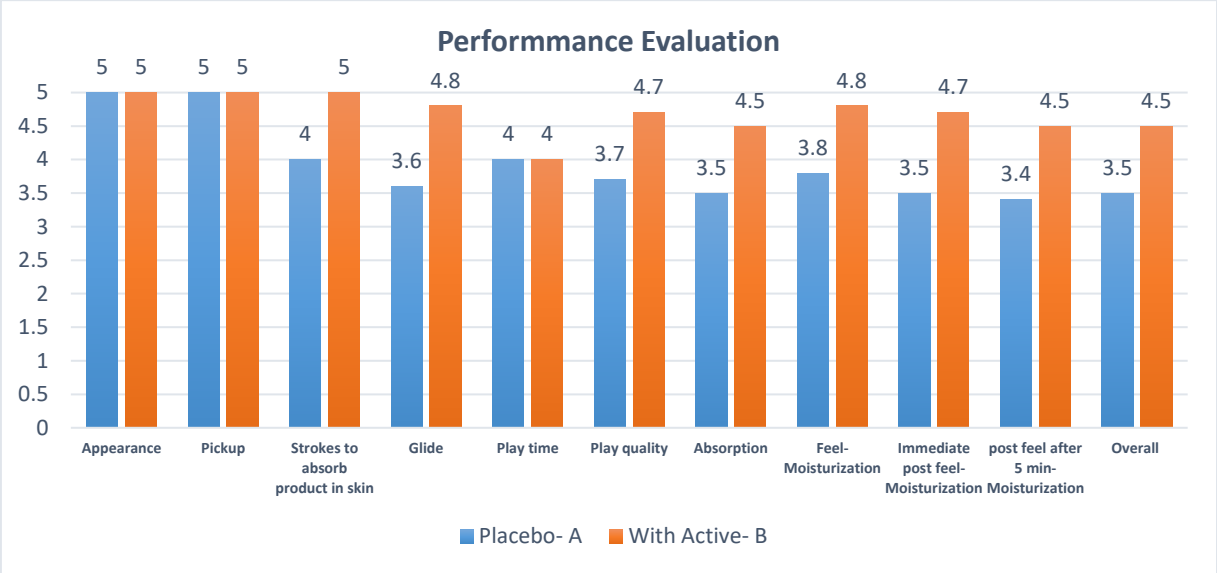
Name	**Viscosity**	**pH**	**Appearance**	**Moisture**
BODY LOTION	6520 CPS (RV 04 RPM 20)	5.75	White translucent lotion	80.00- 82.00 %w/w

Sensory evaluation- Parameters:

Pre-use parameters	Appearance
	Pickup
	Fragrance

In-use parameters	Strokes to absorb product in skin
	Glide
	Play time
	Play quality
	Absorption
	Feel- Moisturization
Post-use parameters	Immediate post feel- Moisturization
	post feel after 5 min- Moisturization
	Overall

Sensorial Evaluation:



The resultant products are comparable & better than the placebo in terms of overall performance. Although the perception of product B (with active) for skin Moisturization (while & post) found to be better than the product A- placebo in the consumer study.

Stability studies: Stability conducted as per ICH guidelines:

Parameters / Conditions and duration	pH @ 27°C	Viscosity (Cps) @ spindle no. 4, 20 rpm	Moisture Content (% w/w)	Appearance	Fragrance
Initial	5.75	6520 cps	80.21	White translucent lotion	
1 Month					
RT	5.08	6500 cps	80.00	No Change	No Change
Ref	4.71	6430 cps	81.00	No Change	No Change
45°C/75°RH	4.72	6240 cps	80.10	No Change	No Change
50°C (Dry Heat)	5.13	6150 cps	79.90	No Change	No Change
2 Months					
RT	4.88	6490 cps	79.80	No Change	No Change

Ref	4.98	6450 cps	80.20	No Change	No Change
45°C/75°RH	4.71	6200 cps	79.90	No Change	No Change
3 Months					
RT	5.04	6480 cps	80.10	No Change	No Change
Ref	5.14	6400 cps	80.00	No Change	No Change
45°C/75°RH	4.79	6240 cps	79.80	No Change	No Change

Sample passes the 3 months stability studies at all the stability temperature.

Way Forward:

Next step to carry out the in- vitro efficacy evaluation for moisturization, through external CRO for the claim validation.

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Effect of salt and polymers and their combination on rheology of rinse-off cosmetic products composed of combination of surfactants

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Abstract-
Surface-active agents are the organic molecules that when dissolved in a solvent at low concentration, have the ability to adsorb at interfaces, thereby altering significantly the physical properties of those interfaces. Thickening of surfactant can be achieved with with different hydrophobic thickeners, the hydrophilic thickener and the combination of both

Key words- Surfactant, Electrolyte, Rehology, Ionic behaviour, Miscell formation.

Introduction

Surface-active agents are the organic molecules that when dissolved in a solvent at low concentration, have the ability to adsorb at interfaces, thereby altering significantly the physical properties of those interfaces.

A surfactant base has been thickened with different hydrophobic thickeners, the hydrophilic thickener and the combination of both. The main task of thickener for surfactant formulation is of course to increase the viscosity. Good stabilizing effect can be obtained by choosing right rheological profile and low temperature dependence of the final viscosity. Rheology plays an important role in the functionality and usage profile of the system. The rheology of the system is maintained by using various rheology modifiers. Rheology modifiers like thickeners are used like salt, polymers, co-surfactants, etc.

The viscosity is determined by using the technically designed instrument known as viscometer. To make the system viscous one need to add viscosity modifier in the formulation i.e. thickeners. The most common way to thicken the anionic surfactant-based formula is to use Sodium Chloride (NaCl). In standard surfactant systems based on Sodium Laureth Sulphate (SLES) and cocoamidopropyl betaine (CAPB) this works quite well. This thickening effect depends on the presence of an anionic surfactant (mostly SLES) and it works up to a conc. maximum. Gelling agents like Xantum gum, Cellulose types or Carbomer types thicken or gel the water.

Surfactants are generally classified on the basis of their ionic behaviour or by hydrogen bonding. Four basic classes therefore emerges as:

1. Anionic surfactants (-ve charge on dissociation)
2. Cationic surfactants (+ve charge on dissociation)
3. Non-ionic surfactants(NO charge on dissociation)
4. Amphoteric or zwitterionics (+-ve charge on dissociation)

Rinse off products mostly cover a category which is Shampoo used for hair cleansing. Hair is soiled by sebum, shade, scales of stratum corneum. Atmospheric pollutants and residues from hair care products. 3 type of dirt in hair to be dealt with shampoo

1. Oily soil or sebum
2. The soluble soil- it is the proteineous matter from stratum corneum and protein contents from sweat.

3. Insoluble soil – It is the atmospheric pollutants and residue from hair care product.

Oliy soil or sebum is removed by process called 'myelenesis' where detergent particle make contact with lipid surface (oiliness to make lipid detergent) compound which loosen and floats away into bulk aqueous solution.

The molecule and detergent and static electricity remove insoluble soil. Surfactants are also termed as detergents as they remove dirt. The presence of surfactant in product is determined by method active detergency. Active detergency is mostly calculated for semisolid or liquid type ingredients because powdered ingredients are 100% active but the semisolid or liquid ingredients contains diluents so the total active matter get reduced. So this method is used to determine the presence of concentrated detergent in ingredient or products. Active detergency in cleansing system is higher in shampoo i.e. shampoo contains more surfactant concentration as compard to body wash and face wash. Face wash contains very low concentration of surfactants.

The active detergency is calculated on the basis of supplied product or ingredient active or dilution.

Example: If we want to add 15% active matter on the formulation and the surfactant material is 70% active as supplied then we need to calculate as follows

$$\text{A.D.} = \frac{15\times100}{70} = 21.42(\%)$$

Micelles structure

Micelles are the spherical aggregates whose alkyl groups form a hydrocarbon liquid like core, and whose polar group forms a charged surface. Later with the development of zwitterionic and non-ionic surfactants, micelles of very different shape are encountered. The different geometries were found to depend mainly on structure of surfactants, their concentration, electrolyte and co-surfactant curve.

The Concentration at which the surfactant molecules in solution start forming aggrigates are micelles and the concentration is called as critical micelle concentration (CMC)

Defining miscell structure

The main structures associated with two-componants surfactant-water system are: hexagonal, lamellar, and several cubic phases

- The hexagonal phase is composed of a close- packed array of long cylindrical micelles, arranged in a hexagonal pattern. The micelle may be 'normal' in that the hydrophilic head groups are located on the outer surface of cylinder, or 'inverted', with the hydrophilic group located internally
- The lamellar phase(L_α) is built of alternating water surfactant bilayers. The hydrophobic chain possess a significant degree of randomness and mobility, and the surface bilayer can range from being stiff and planar to being very flexible undulating.

Critical packing parameter (P_c) as the ratio of volume to surface area:

$$P_c = v/(a_0 l_c)$$

The parameter v varies with the number of hydrophobic groups, chain unsaturation, chain branching and chain penetration by other compatible hydrophobic group, while a_0 is mainly governed by electrostatic interaction and head group hydration. P_c is usually quantity since it allows the prediction of aggregate shape and size.

Figure-
Critical packing parameter

P_c	**General surfactant type**	**Expected Aggregate structure**
<0.33	Single chain surfactant with large head groups	Spherical or ellipsoidal micelles
0.33-0.5	Single chain surfactant with small head groups, or ionics in the presence of large amount of electrolyte	Large cylindrical or rod-shaped micelles vesicles and flexible bilayers structure
0.5-1.0	Double chain surfactant with large head groups and flexible chains	Planar extended bilayers
1.0	Double chain surfactant with small head groups or rigid immobile chains	Reversed or inverted micelles
>1.0	Double chain surfactants with small head groups, very large and bulky and hydrophobic groups	

- The cubic phase may have a wide variety of structural variations and occurred in many part of the phase diagram . These are optically isotropic system and so cannot be characterised by polarising light microscopy.

There are two main groups of cubic phase such as-

- Themicellar cubic phase- Built up of regular packing
- The bicontinuous cubic phase

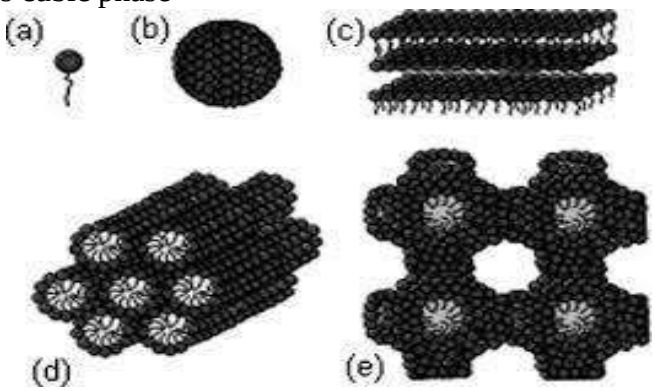


Figure: Structures of micelles

Mechanism of rheology modification:

Typical thickening agents for surfactant system can be generally divided into 2 groups:

1. The hydrophobic, monomeric or oligomeric type with a low molecular weight. These types are mostly non-ionic surfactant
2. The hydrophilic, polymeric type with high molecular weight. These types are based on highly ethoxylatedoleochemical derivatives.

These two type of thickeners provides two important difference in performance: **the flow behaviour** and **the temperature dependence of the viscosity.**

- **Flow behavior**

The hydrophobic thickener provides a shear thinning flow behavior,that means the viscosity decreases with increasing shear rate. This can be easily be observed by measuring the viscosity with a rotational viscometer at different speeds. The hydrophilic thickener provides a Newtonian flow behavior, which means the viscosity is independent of shear rate.

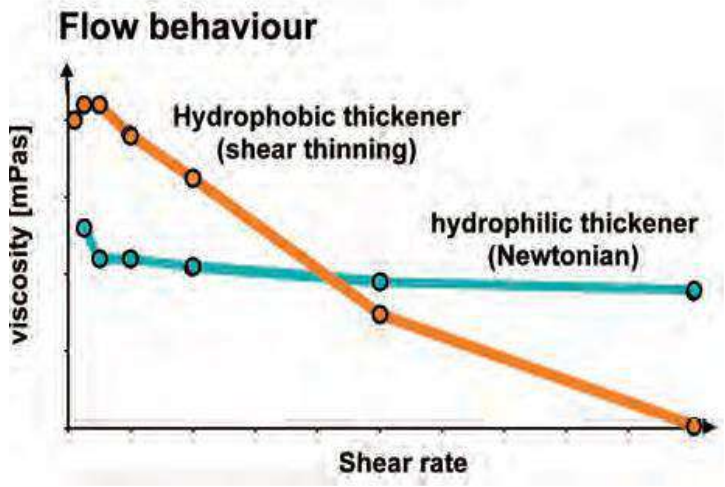


Figure- Differences of flow behavior

- **Temperature dependence of the viscosity**

The hydrophobic thickener provides a decrease in viscosity at low temperature, but a mostly stable viscosity at higher temperature .The hydrophilic thickener provides the strong temperature dependence of the viscosity

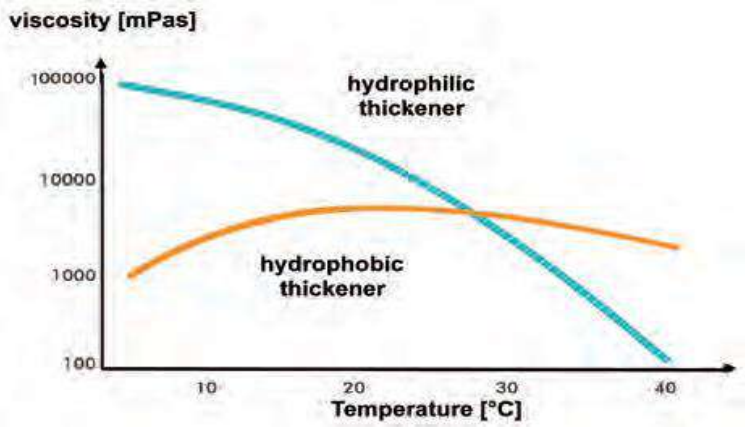


Figure- Differences of temperature dependence of the viscosity

Thickening mechanism

To explain the different flow behavior it is necessary to understand the General mechanism of surfactant thickening. Basically the thickening agents modify the micellar structure. In case of polymeric hydrophilic thickeners, the hydrophobic groups of the molecules are incorporated in the surfactant micelles. This leads to bridging of the spherical micelles and an increase of the micelle size occurred. The micells have the more limited space to move which lead in the increase of viscosity and a Newtonian flow behavior.

The hydrophobic thickeners are also incorporated into the surfactant micelles, but since their hydrophilic head is rather small, they change the shape of micelles. The shape changes from spherical to rod like. At rest the micelles are arrange randomly, which leads to a high viscosity.

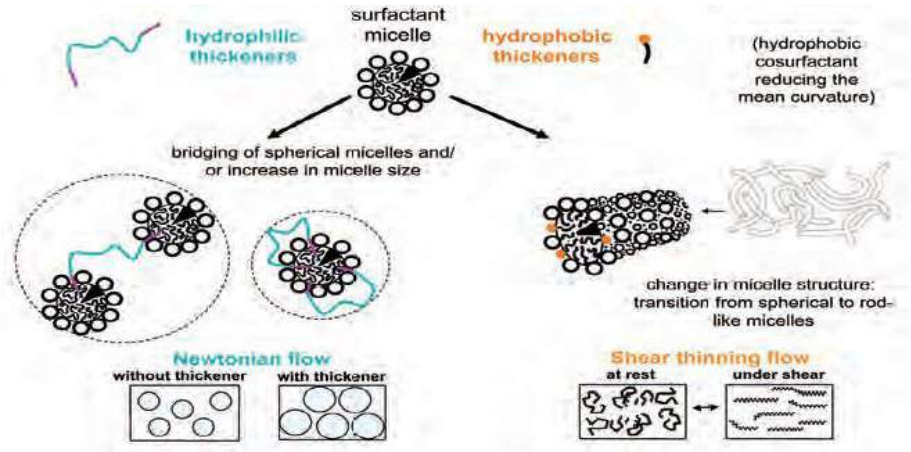


Figure: Thickeners for surfactant systems modify the micellar structure

Good stabilizing effect can be obtained by choosing right rheological profile and low temperature dependence of the final viscosity. Figure indicating differences of temperature dependence of the viscosity shows that hydrophilic thickeners tend to show a strong decrease in viscosity at higher temperatures. On other hand this lead to a higher sedimentation sped of disperse particles. At low temperature a hydrophilic thickener tend to provide an increase of viscosity. Hydrophobic thickeners are weak at low temperatures, so tend to drop the viscosity

Rheology plays an important role in the functionality and usage profile of the system. The rheology of the system is maintained by using various rheology modifiers. Rheology modifiers like thickeners are used like salt, polymers, co-surfactants, etc. Salt mainly Sodium chloride is used as thickener.

Polymers are used to thicken the system and to increase the viscosity of system.

Formulation:

- Shampoo formulation with SLES and salt
  - With 5 % CAPB (as supplied)

As the supplied surfactant (SLES) is 28% active and here need to add 15% so for 100gm. Formulation its concentration is calculated as

$$\text{A.D. (SLES)} = \frac{15\times100}{28} = 53.57$$

Sr. No.	Ingredients	Shampoo formulation with SLES +salt				
		Conc. Of salt				
		0.1	0.5	1	1.5	2
1	DI water	38.23	37.83	37.33	36.83	36.33
2	Sodium gluconate	0.05	0.05	0.05	0.05	0.05
3	SLES(28% active)	53.57	53.57	53.57	53.57	53.57

4	CAPB(30% active)	5	5	5	5	5
5	Polyquaternium-39	1	1	1	1	1
6	Glycerin	2	2	2	2	2
7	Aqua Methylchloroisothiazolinone (and) Methylisothiazolinone	0.05	0.05	0.05	0.05	0.05
8	NaCl	0.1	0.5	1	1.5	2
	Total	100	100	100	100	100

As the supplied surfactant (Sarcosinare) is 30% active and here need to add 15% so for 100gm. Formulation its concentration is calculated as

$$\text{A.D. (Sarcosinate)} = \frac{15\times100}{30} = 50(\%)$$

Sr. No.	Ingredients	**Shampoo formulation with Sodium Lauryl Sarcosinate + salt**				
		Conc. of salt				
		0.1	0.5	1	1.5	2
1	DI water	41.8	41.4	40.9	40.4	39.9
2	Sodium gluconate	0.05	0.05	0.05	0.05	0.05
3	Sodium Lauryl Sarcosinate(30% active)	50	50	50	50	50
4	CAPB(30% active)	5	5	5	5	5
5	Polyquaternium-39	1	1	1	1	1
6	Glycerin	2	2	2	2	2
7	Aqua Methylchloroisothiazolinone (and) Methylisothiazolinone	0.05	0.05	0.05	0.05	0.05
8	NaCl	0.1	0.5	1	1.5	2
	Total	100	100	100	100	100

Observation and Evaluation:

A typical salt curve

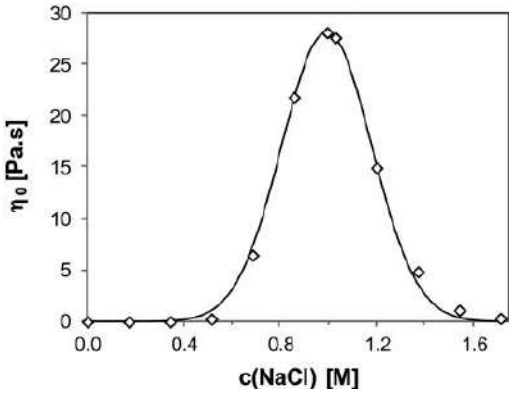
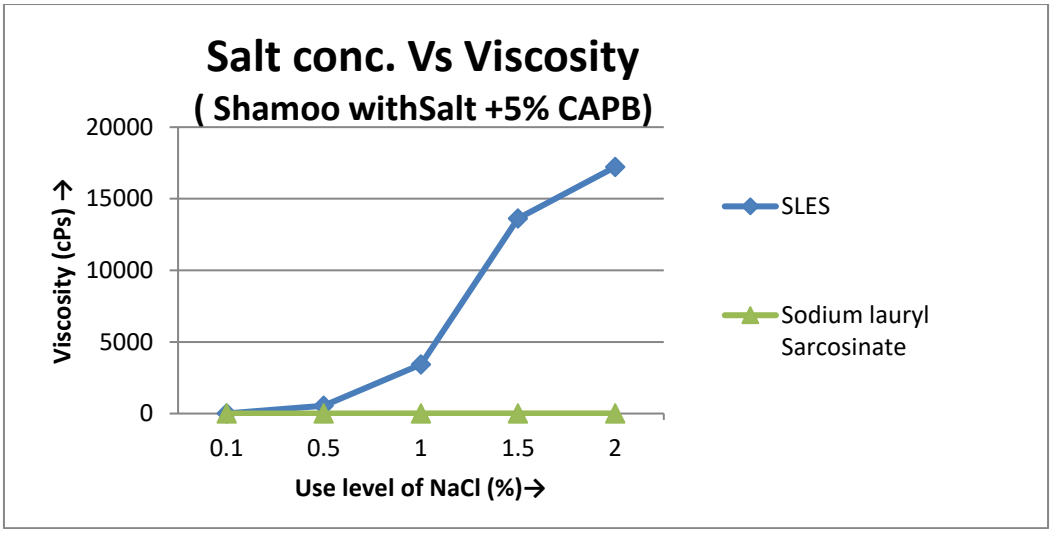


Figure: The impact of NaCl and small hydrophobic molecules, used in perfumery, on the viscoelastic properties of aqueous solutions of sodium lauryl ether sulphate is studied. As the salt concentration increases, the viscosity passes through a maximum. Empirically, this behaviour is well known and is referred to as the 'salt curve'.



OBSERVATIONS AND RESULT:

- In case of SLES the viscosity increases with increasing salt concentration
- Sarcosinate shows minor change in viscosity and it fluctuates.

It shows that SLES shows synergistic effect with increasing salt concentration as compared to Sodium lauryl sarcosinate.

WAY FORWARD-

The rinse off products is generally prepared by using combination of ingredients. Ingredients include surfactants, foam stabilizers, thickeners, and other additives like conditioners for shampoo, colors, humectants, and preservatives. By making change in the concentrations of the formulation one can innovates a different product of same ingredient with differing effects. This project is based on effect on rheology of surfactant system with addition of polymers, thickeners as well as foam stabilizer in the rinse off formulation. Polymers are also available in the market and we can go with them with the combinations of ingredients.

We can use more different polymers, thickeners available in market and work with them with differing ingredient concentrations. So combination of the other polymers the effect will be different and we can find different result and we will get more knowledge and idea about the chemistry of surfactants compatibility with different polymers.

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Microbiomics for cosmetics

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Abstract:

Over time, the idea of beauty goods has changed from being primarily about appearance to including a broader sense of well-being. It is hypothesised that the usage of artificial chemical compounds in contemporary cosmetics damages skin. For the first time, a method to test for healthy skin was made possible by the finding that the only currently valid indication of skin health on humans is biodiversity on the skin. Microbiomes are a collection of microorganisms that are naturally present in the skin and actively contribute to its health. Every type of skin has a unique microbiome. The skin is home to a rich and amazingly diverse community of microorganisms called the skin microbiota, which includes viruses, bacteria, fungus, archaea, and protozoa. Approximately one billion bacteria per square centimetre can be found on human skin, and these microorganisms are in charge of preserving the skin's immunity and halting the development of harmful growths. The foundation of healthy skin is a balanced microbiota; healthy skin is more resilient and resistant to external stresses. A disturbance of this dermal ecology can lead to a number of issues, including dermatitis, greasy skin, dandruff, and foul odours. The goal of biome-based skincare is to protect against the overabundance of harmful bacteria and encourage the growth of beneficial bacteria. Microbiome skincare includes prebiotics, probiotics, and postbiotics, which are similar to health supplements and strengthen and enhance the skin's immune system.

Keywords- Microbiomics, microbiomes, biome-based skin care, microbiome skin care

Introduction:

Recent research in dermatology and cosmetics has concentrated on the cutaneous microbiota and how it interacts with the skin and its surroundings. Microbiomes are a collection of microorganisms that are naturally present in the skin and actively contribute to its health. Every type of skin has a unique microbiome. The skin is home to a rich and amazingly diverse community of microorganisms called the skin microbiota, which includes viruses, bacteria, fungus, archaea, and protozoa. Approximately one billion bacteria per square centimetre can be found on human skin, and these microorganisms are in charge of preserving the skin's immunity and halting the development of harmful growths. The foundation of healthy skin is a balanced microbiota; healthy skin is more resilient and resistant to external stresses. This dermal environment becomes disturbed as a result.The goal of biome-based skincare is to protect against the overabundance of harmful bacteria and encourage the growth of beneficial bacteria.[1] One of the most alarming environmental elements impacting skin tone is UV radiation. Few research have looked into the cutaneous microbiota; most have concentrated on the impact of UV radiation on the skin. The generation of vitamin D is one good consequence that might exist, but the majority seems to be detrimental.Thus, photoprotection is essential. The cosmetics industry has recently asserted that physical or chemical sunscreens can be made better by adding molecules that reduce the damaging effects of UV light or compounds with inherent UV-absorbing potential. This review's goal is to examine and talk about cutaneous

microorganisms' potential benefits for skin health as well as their capacity to defend against UV radiation harm. Because of their anti-oxidant and/or anti-inflammatory properties, probiotic and postbiotic substances are used in cosmetic product formulations to protect or restore the balance of the cutaneous microbiota and block the effects of UV radiation. [2].Live microorganisms are referred to as probiotics when they are given to a host in sufficient quantities and offer health benefits. Postbiotic, on the other hand, describes metabolic byproducts produced by a probiotic organism over its lifetime, including peptides, enzymes, teichoic acid, muropeptides derived from peptidoglycan, exopolysaccharides, secreted and cell surface proteins, bacteriocins, and organic helpers [3]. We want to uncover protective methods associated with the cutaneous microbiota in this study, including the use of probiotics and postbiotics. Like probiotics, postbiotics, and prebiotics in health supplements, these microbiome skincare ingredients boost and enhance the skin's immune system.Probiotics are "friendly bacteria" that are fed prebiotics, or nutrients, while postbiotics are the materials that remain after microorganisms die, like whole cells or the walls of dead microbes.[4]

TheSkinAllergyEpidemic:

Western behaviors have been linked to a catastrophic loss of microbial diversity in the human gut microbiome in developed nations. As a result, within the past 75 years, there has been a sharp rise in food allergies. The same is true for skin allergies, whose severity has increased over the last five to ten years to the point where some have dubbed it a "skin allergy epidemic." Although a variety of environmental factors may be involved, artificial compounds found in cosmetics are becoming more and more associated with this. [5]This modification has frequently been connected to an increased risk of infection and illness. Studies on the microbiome of the skin are far behind those on the gut, where it is widely accepted that a non-diverse, out-of-balance gut flora is directly associated with a host of health issues. It is now understood that maintaining and enhancing the gut microbiota—rather than eliminating it—is crucial for general health.[6] Skin that is injured or diseased has a lower diversity of microbial species than skin that is healthy or normal, which is consistent with other studies that show a positive correlation between high bacterial diversity and enhanced host protection provided by an adaptive and creative immune system. [7]

Ingredients:

The following ingredients are recognized to support the preservation of the skin's ecosystems: *lactobacillus ferment lysate, saccharomyces lysate, Bifida ferment filtrate*, and bacillus ferment. Each component works through a unique method of action. Understanding the functions of each ingredient is crucial for the formulator to create a skin care product that is successful.[8]

TheRoleof SkinMicrobiota:

The market for microbiome skin care products offers four primary approaches to preserving the skin's microbiome: eliminating harmful bacteria, introducing prebiotics to nourish beneficial bacteria, probiotics to increase the number of good bacteria, and postbiotics to provide bacterial byproducts to the skin. The most widely used strategies involve developing prebiotic and probiotic skin care products, which are more readily adapted to the skin care industry and are the most familiar to consumers due to dietary trends**.** [9] Most of the microbiota that live on the skin are either helpful or safe and are essential to the host's defense. The benefits to both the host and the microbe indicate that the relationship is "mutualistically symbiotic." Despite the widespread misconception that certain microorganisms are always dangerous, pathogenicity only arises when the ecosystem's delicate balance is upset and variety is reduced. Many skin disorders are caused by this "perturbation" or "dysbiosis" of the skin microbiome. Not the intrinsic characteristics of the microbe, but the skin's overall resistance to illness and infection, makes the distinction

between benign bacteria and a pathogenic agent. Many diseases, such as acne, dermatitis, rosacea, psoriasis, general allergies, sunburn, athlete's foot, ringworm, wound healing, diabetic skin, leishmaniasis, blepharitis and conjunctivitis, and skin cancer, have been related to dysbiosis and decreased microbial biodiversity of the skin microbiome. Cosmetics and the diversity of the human skin microbiome.[10]Better skin is associated with higher biodiversity; yet, we have seen changes in skin moisture, trans epidermal water loss, and diversity that seem to be related to product use. [11-13]

Today's consumers are gravitating towards simpler, greener, and natural cosmetics. Natural skincare solutions that balance the microbiome can address the growing issues of environmental pollution, sensitive skin, and early ageing. Researchers studying cosmetic formulations have started to look into the connection between a healthy human microbiome and skin. By strengthening the skin's natural defenses and immune system, these products can help the skin heal itself. The goal of these natural products is to eradicate pathogens while preserving the mutualistic and commensal organism composition. Recently, the cosmetics industry has adopted this unique strategy of focusing on the skin microbiome on several fronts.[14]

Challenges:

The formulation of probiotic/prebiotic skin care products differs from that of fresh foods used to treat digestive system disorders because beauty goods have a natural shelf life. Since it is challenging to include live bacteria into cosmetics, the majority of them really contain bacterial cell wall fragments that can compromise immunological response. Another difficulty with using probiotics and prebiotics in the production of cosmetics is that each person has a unique set of needs for their skin microflora, and even the microflora that is naturally present on different body parts varies. [15-17]

Conclusion:

A product seemed to have less of a favorable impact on average biodiversity the more synthetic chemicals it included, especially in the first two weeks. In the future, a single, truly global norm for a range of skin health conditions will be developed, together with benchmark diversity levels, to be utilized as an international standard for evaluating product efficacy in lab settings. It will be carried out over a longer time frame in order to provide insight into the impacts of utilizing probiotics or other compounds, as well as how long ecosystems like the skin microbiome require to re-adjust to a healthier, more natural form. For the beauty business, the skin microbiome offers intriguing opportunities for new product development and marketing strategies. Amidst the COVID-19 epidemic, consumer interest in microbiome beauty has increased because to worries about the skin's critical function in protecting and reestablishing the equilibrium of the skin microbiota.

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