



Formulation and Evaluation of Herbal Antiseptic Cream

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I. Introduction-

Creams are the topical preparations which can be applied on the skin. Creams are defined as “viscous liquid or semi-solid emulsions of either the oil-in-water or water-in-oil type” dosage forms which consistency varies by oil and water.[1] Creams are used for cosmetic purposes such as cleansing, beautifying, improving appearances, protective or for therapeutic function. These topical formulations are used for the localized effect for the delivery of the drug into the underlying layer of the skin or the mucous membrane. These products are designed to be used topically for the better site-specific delivery of the drug into the skin for skin disorders.[2]

Creams are considered as a pharmaceutical product as they are prepared based on techniques developed in the pharmaceutical industry; unmedicated and medicated creams are highly used for the treatment of various skin conditions or dermatoses. Creams can be ayurvedic, herbal or allopathic which are used by people according to their needs for their skin conditions. They contain one or more drugs substances dissolved or dispersed in a suitable base. Creams may be classified as o/w or w/o type of emulsion on the basis of phases. The term ‘cream’ has been traditionally applied to semisolid formulated as either water-in-oil (e.g.: cold cream) or oil-in-water (e.g.: vanishing cream).[3]

TYPES OF SKIN CREAMS-

They are divided into two types:

Oil-in-Water (O/W) creams-

These are composed of small droplets of oil dispersed in a continuous phase, and an emulsion in which the oil is dispersed as droplets throughout the aqueous phase is termed an oil-in-water (O/W) emulsion.

Water-in-Oil (W/O) creams-

These are composed of small droplets of water dispersed in a continuous oily phase. When water is the dispersed phase and an oil the dispersion

medium, the emulsion is of the water-in-oil (W/O) type. [4-6]

CLASSIFICATION OF CREAMS -

All the skin creams can be classified on different basis:

1. According to function, e.g., cleansing, foundation, massage, Medicinal Creams (Antiseptic, Antimicrobial, Wound Healing) etc.
2. According to characteristics properties, e.g., cold creams, vanishing creams, etc.
3. According to the nature or type of emulsion.

Types of creams according to function, characteristic properties and type of emulsion:

1. Make-up cream (o/w emulsion): a) Vanishing creams. b) Foundation creams.
2. Cleansing cream, cleansing milk, cleansing lotion (w/o emulsion)
3. Winter cream (w/o emulsion): a) Cold cream or moisturizing creams.
4. All-purpose cream and general creams.
5. Night cream and massage creams.
6. Skin protective cream.
7. Hand and body creams. [7-10]

Antiseptic Activity-

These are chemical substance which inhibit the growth or kill micro-organisms on living surface such as skin and mucous membrane or these are anti-microbial substances that are applied to living tissue or skin to reduce possibility of infection, sepsis, putrefaction.

Properties of good Antiseptic Cream-

- 1) Cidal
- 2) Non-staining and good odour.
- 3) Active against all pathogens.
- 4) Active in presence of pus, blood and exudates.
- 5) Rapid Acting
- 6) Non-Irritating to Tissues or non-corrosive
- 7) Non-absorbable
- 8) Non-Sensitizing



Mechanism of Action of Antiseptic Cream-

- Oxidation of bacterial protoplasm.
- Coagulation (Denaturation) of bacterial proteins and disrupt cell membrane.
- Detergent like Action increases the permeability of bacterial cell membrane.[11]

II. Aim and Objective

Aim: –

- Formulation and Evaluation of Herbal Antiseptic Cream using Giloy stem extract.

Objective: –

- To extract the stems of *Tinospora Cordifolia* by Soxhlet apparatus.
- To evaluate the prepared extract of stems of *Tinospora Cordifolia*.
- To formulate herbal Antiseptic cream of *Tinospora Cordifolia*.
- To evaluate the herbal Antiseptic cream of *Tinospora Cordifolia*.

III. Literature Review

1.N. N. Navindgikar et.al. (2020) - Formulation and Evaluation of Multipurpose Herbal Cream.

The extract of Neem leaves, Tulsi leaves and Aloe vera gel is prepared and cream was prepared by using Slab Technique.

2. B. Venkatappa et.al. (2021) - A Review on Medicinal Properties of *Tinospora Cordifolia* (Giloy).

All parts of the plant constitute immunomodulatory properties. It is one of the important medicinal plants used for the treatment of cold, fever, diabetes, and even rheumatoid arthritis. This paper presents a review on importance of *tinospora cordifolia*.

3. Shubhangi E. Savant et.al. (2016) - Formulation and Evaluation of Herbal Ointment Containing Neem and Turmeric Extract

The present work is to formulate and evaluate the Ointment of Neem and Turmeric extract. The ethanolic extract were prepared by using maceration method. The ointment base was prepared and formulation of ointment was done by incorporating the extract in the base by levigation method.

4. Abinash Kumar Sahu et.al. (2021) - *Tinospora Cordifolia*: phytochemical screening & anthelmintic study of it's different extracts

Study is to investigate phytochemical screening and anthelmintic study of *tinospora cordifolia* by petroleum ether and aqueous extract. The data revealed that the aqueous extract has a better wormicidal effect than petroleum ether extract with compared with the standard drug.

5. Abinash Kumar Sahu et.al. (2021) - *Tinospora Cordifolia*: phytochemical screening & anthelmintic study of it's different extracts

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Drug Profile-

Kingdom : Plantae

Family : Menispermaceae

Genus : *Tinospora*

Species : *T. cordifolia*

Common Names :

Latin : *Tinospora cordifolia*

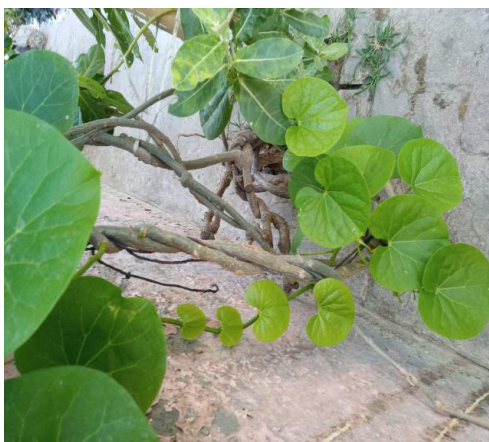


Fig.01: Giloy Stems

Marathi : Shindilakodi, Gudvel

English : *Tinospora Gulancha*

India is a traditional medicinal practicing country; from the centuries this traditional method is followed for cure disease. Medicines obtained from herbal plant show multiple therapeutic effects in human beings. Herbal aqueous extract of medicinal plant prepared and examine for their activity against various disease is checked. The present herbal medicine shows the antiseptic activity on strains of micro-organism. The antiseptic activity were checked by using the herbal medicinal plant extract on the strain of microorganism which will affect the human beings.[12]



**Fig.02: Tinospora cordifolia (Giloy) plant
(Leaves, stems and root)**

Biological Source: -

It is consisting of dried, matured pieces of stems of **Tinospora Cordifolia Miers**. It belongs to the family Menispermaceae. *Tinospora cordifolia* is a deciduous woody climbing shrub distributed throughout India, China, Africa. The pharmaceutical significance of this plant is mainly because of root, stem, leaf. It constitutes various phytoactive compounds such as alkaloids, steroids, glycosides.[13] It is one of the important medicinal plants used in Ayurvedic Medicine for the treatment of Cold, Fever, Diabetes, Rheumatoid Arthritis and also Antiseptic and Antibacterial activity. *Tinospora cordifolia* is a deciduous woody climbing shrub distributed throughout India, China, Africa. It belongs to the family Menispermaceae. The pharmaceutical significance of this plant is mainly because of root, stem, leaf.[14] It constitute various phytoactive compounds such as alkaloids, steroids, glycosides. It is one of the important medicinal plant used in Ayurvedic Medicine for the treatment of Cold, Fever, Diabetes, Rheumatoid Arthritis and also Antiseptic and Antibacterial activity.

Botanical description and Distribution: -

Tinospora cordifolia, a semi-evergreen deciduous climbing shrub which is often found on the trunks of large trees like mango and neem. It can survive in different kinds of soils from acidic to basic with average moisture. Stems of the plant are moist and thick, having large aerial roots, emerging from the branches with varying thickness; younger stem are green coloured with smooth surfaces, while older stems being light brown colored The shrub is succulent, twinning, rapid growing vine with green branches turning brown with age and aerial roots, ovate, juicy, acute membranous young

leaves with round petiole, 5 to 14 cm in diameter. [15] Wood is porous, soft grey-brown or creamy white in colour which peels off easily. The plant blossom during May-June to small unisexual yellow flowers. Male flowers are small yellow-green axillary and female flowers are solitary. Fruits are ovoid, fleshy, reddish having single seed. Fruiting is seen in September-October. The plant is endemic to tropical and subtropical Indian subcontinent, China also in Srilanka and Myanmar. In India, it is a very common plant found from the kumaun to Kanyakumari. It can grow in any temperature and almost all soil types, also has a long lifespan. It is also found in countries such as China, Srilanka and Myanmar ascending upto an altitude of 310 m. [16]

Biological and phytochemical properties: -

The ayurvedic medicinal significance of *Tinospora* species is ascribed to the presence of various phytochemicals viz, alkaloids, glycosides, diterpenoids, steroids, flavanoids, lignins and various other chemical compounds. Alkaloids like jatrorrhizine in *T. cordifolia* contains antimicrobial properties. The aqueous extract from stem of giloy has revealed the existence of arabinogalactan that possess immunological properties.[17] The alkaloids like Tinocordifolin, Tinosporide and Tinocordifolioside have been identified in stems and leaves which are anti-hypertensive and rich in proteins. *T. cordifolia* species acts as diuretic and are effective against urinary disorders. Giloy also acts as a memory booster and contain rejuvenating and healing properties. It also helps in liver related problems and prevent hepatic tissue and fibroids. Glycosides like Pregnane and Palmatoside helps in heart related problems. Alkaloids present in giloy reduce glucose level in blood and controls diabetes. It also prevents eye disorders. The stem is rich in Sesquiterpenoids, Alkaloids, Steroids are used as anti-pyretic and antiviral which prevents all kinds of fever and urinary disorders.[18] It is one of the best natural remedies to boost immune system. It helps us fight against various infections and disorders. This plant is useful for treatment of skin related problems, like acne vulgaris, hives, psoriasis and wound healing as it contains anti-bacterial characteristics. Recent research showed that this plant contains alpha-D-glucan which plays a major role in humoral immunity. Its extract also possesses anti-tumor properties which can prevent cancer. This plant extract is diuretic which stimulates bile secretion, prevents digestive disorders, relief burning sensation and jaundice. Its stem contains anti-inflammatory property which is helpful in arthritis. It is said that giloy which grows on neem



tree is considered to be double effective as it incorporates remarkable neem healing characteristics. *Tinospora cordifolia* therefore is the immortal elixir of life, it acts as a natural rejuvenator, a potent tonic and can be grown anywhere in any atmosphere.[19]

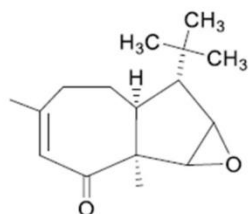


Fig.02: Structure of Tinocordifolin

Medicinal usage and therapeutic properties: -

Ayurveda without this plant may not be possible as it is the main component of various ayurvedic medicines. *Tinospora cordifolia* is beneficial in viral and bacterial fever, diarrhea, jaundice, low immunity, bone weakness, recurrent infections, common flu cold, fatigue, asthma, diabetes, arthritis, cancer, etc. Various parts of the plant are used for medicinal benefits since ancient time by folks and tribals.[20]

Tinospora cordifolia have different constituents and used to treat various diseases. It is a multipurpose plant and its different dosage forms are used for various purpose.

□ Immunity Enhancer: -

Giloy is used to improve or boost immunity". It contains number of antioxidants which fight free-radicals, keep your cells healthy and get rid of diseases. Giloy helps to remove toxins and purifies blood, fights against bacteria.[21]

□ Antiseptic and Anti-microbial:-

These are chemical substance which inhibit the growth or kill micro-organisms on living surface such as skin and mucous membrane or these are anti-microbial substances that are applied to living tissue or skin to reduce possibility of infection, sepsis, putrefaction.

□ In Chronic Fever: -

Giloy helps to recover fevers. Giloy is anti-pyretic drug, it can reduce signs and symptoms of several life-threatening conditions like Dengue, Swine Flu and Malaria as well". It helps to improve blood platelets in fever.[22]

□ In Digestion: -

Giloy is very beneficial in improving digestion and treating bowel related issues. Giloy powder with

some amla can use regularly to maximum results, or with jaggery for treating constipation. [23,24]

□ Treats Diabetes: -

Giloy is also a hypoglycaemic agent which help to treat diabetes particularly Type 2 diabetes. It also helps to lower blood sugar. It has been reported to mediate its anti-diabetic potential through mitigating oxidative stress (OS), promoting insulin secretion and also by inhibiting gluconeogenesis and glycogenolysis, thereby regulating blood glucose. The major phytoconstituents of *Tinospora cordifolia* are alkaloids, tannins, cardiac glycosides, flavonoids, saponins, and steroids as have been reported to play an anti-diabetic role.[25]

□ Treats Arthritis: -

Giloy contains anti-inflammatory and anti-arthritic properties that help treat arthritis and its several symptoms. For joint pain, the powder from giloy stem. It can be used along with ginger to treat rheumatoid arthritis. *Tinospora cordifolia* have been reported to affect the proliferation, differentiation and mineralization of bone like matrix on osteoblast model systems in vitro and hence finds potential application as an anti-osteoporotic agent. [26,27]

□ Reduces Asthmatic Symptoms: -

Asthma causes chest tightness, shortness of breath, coughing, wheezing, etc. Giloy have an anti-inflammatory action and helps to reduce respiratory problems like frequent cough, cold, tonsils.[28]

□ Improves Vision and reduces Signs of Ageing: -

In several parts of India, Giloy plant is helps to boost up vision clarity. For this, boil giloy powder in water, let it cool down and apply over the eyelids. This plant contains anti-aging properties that help reduce dark spots, pimples, fine lines and wrinkles. It provides flawless, glowing skin of an individual.[29]

□ Anti-HIV effects: -

TCE has been shown to demonstrate a decrease in the recurrent resistance of HIV virus thus improving the therapeutic outcome. Anti -HIV effects of TCE was revealed by reduction in eosinophil count, stimulation of B lymphocytes, macrophages and polymorphonuclear leucocytes and haemoglobin percentage thus, revealing its promising role of application in management of the disease.[30]

Excipient Profile-

Giloy Extract (Antiseptic)-

These are chemical substance which inhibit the growth or kill micro-organisms on living surface such as skin and mucous membrane or these are anti-microbial substances that are applied to living



tissue or skin to reduce possibility of infection, sepsis, putrefaction. Due to the presence of Tinocordifolin, it shows Antiseptic activity.

Beeswax (emulsifier)-

Emulsifying agent, stabilizer and gives thickness to the cream It is an ingredient in surgical bone wax, which is used during surgery to control bleeding from bone surfaces; shoe polish and furniture polish can both use beeswax as a component, dissolved in turpentine or sometimes blended with linseed oil or tung oil; modeling waxes can also use beeswax as a component; pure beeswax can also be used as an organic surfboard wax. Beeswax blended with pine rosin is used for waxing, and can serve as an adhesive to attach reed plates to the structure inside a squeezebox. It can also be used to make Cutler's resin, an adhesive used to glue handles onto cutlery knives.

Liquid paraffin (Lubricating Agent)-

It is used as Lubricating agent. In the topical form of this medicine is used in a combination with other medicines to treat certain skin conditions.

Borax (Alkaline Agent)-

It is an alkaline agent which reacts with emulsifying agent in formulation of cream. Borax is used in the cosmetic industry to prevent or slow bacterial growth in moisturising products like creams, shampoos, gels, lotions, bath bombs, scrubs and bath salts. Manufacturers of cosmetics use borax as

a buffering agent or an emulsifier to keep product ingredients.[31]

Methyl Paraben (Preservatives)-

The use of preservatives in cosmetics is essential to prevent alteration caused by microorganism and contamination during formulation, shipment, storage and consumer use. Antioxidants can also be used to protect alteration caused by exposure to oxygen. Synthetic preservatives when used in low concentration effectively preserve the products.[32]

Rose Oil (Fragrance)-

Rose oil has antifungal and antibacterial properties and can help deal with a variety of infection-inducing microbes. In fact, it has also been proven to be beneficial in getting rid of the bacteria that cause staph and strep throat. Additionally, rose oil benefits by working towards various gut, mouth, and vaginal infections as it soothes and gets rid of the infection-causing microbes

Distilled Water (Vehicle)-

This is the most important and widely used raw material in any cream formulation. These are the cheapest and easily available. In skin creams, water is used as solvent to dissolve other ingredients of creams. Water, which is free of any toxins, pollutants, microbes, etc. is used in preparation of creams. Water can also form emulsions, it depends upon how much quantity of water is used in the formulation and sometimes referred to as oil-in-water emulsions and sometimes water-in-oil emulsions depending upon the quantities of oil phase and water phase used.[33]

Plan of work -

Sr. No	Title	Days
1	Selection Of Drug	7 Days
2	Authentication	2 Days
3	Collection of plant	2 Days
4	Preparation of extract	15 Days
5	Evaluation of extract	20 Days
6	Formulation of Herbal Antiseptic Cream	25 Days
7	Result and Discussion	5 Days
8	Conclusion	1 Day

Table No.01: Plan of Work

Experimental Work –

Introduction -Excipient and Formula for Herbal Cream –

Sr. No.	Ingredient	Role	Quantity
1	Giloy Extract	Antiseptic	0.5 ml
2	Bees Wax	Emulsifier	3 gm
3	Liquid Paraffin	Lubricant	10 ml



4	Borax	Alkaline Agent	0.2 gm
5	Methyl Paraben	Preservative	0.02 gm
6	Rose Oil	Fragrance	q.s.
7	Distilled Water	Vehicle	q.s.

Table No.02: Formula for Herbal Cream

IV. Material and Methods

Collection –

It is collected from native places. *Tinospora cordifolia*, a semi-evergreen deciduous climbing shrub which is often found on the trunks of large trees like mango and neem. It can survive in different kinds of soils from acidic to basic with average moisture. Stems of the plant are moist and thick, having large aerial roots, emerging from the branches with varying thickness. [34]

Authentication – The collected plant was identified and authenticated by R.K. Choudhary, Agarkar Research Institute, Pune-411004.

महाराष्ट्र असोसिएशन फॉर द कल्चिवेशन ऑफ सायन्स
आधारकर अनुसंधान संस्थान
(विज्ञान और प्रौद्योगिकी विभाग, भारत सरकार के अधिन स्वायत्त संस्थान)
गो. ग. आगरकर पथ, पुणे - ४११ ००४.
Maharashtra Association for the Cultivation of Science
AGHARKAR RESEARCH INSTITUTE
(An Autonomous Body under the Department of Science and Technology, Govt. of India)
G. G. Agarkar Road, Pune - 411 004.

दिनांक / Date: 01.03.2022

प्रमाणीकरण पत्र / AUTHENTICATION CERTIFICATE

नाम Name : Ms. Vrushali S. Malusare, Mr. Atharva S. Joshi,
Ms. Revati V. Mahanwar and Mr. Onkar P. Kolapkar

पता Address : Navsahyadri Institute of Pharmacy,
Sr. No. 69, 70, 71, Naigaon (Nasarapur),
Pune-412 213

संदर्भ Reference : Your letter No: NESNIP/21-22/, dated 17/02/2022

नमूने का नाम / Name of the sample : *Tinospora cordifolia* Stem

नमूने की मात्रा / Amount of sample : Approx. 100 gm.

प्राप्ति की तिथि / Date of the receipt : 17/02/2022

Report (AUTH 22-20):

प्राप्त नमूने का मैक्रोस्कोपिक, माइक्रोस्कोपिक और ऑर्गेनोलेप्टिक चरित्रों की मदद से सावधानीपूर्वक अध्ययन किया गया है। हम एवं द्वारा प्रमाणित करते हैं कि दिया गया नमूना *Tinospora cordifolia* (Willd.) Hook.f. & Thomson, [Family: Menispermaceae] के तने का है।

The sample has been critically studied with the help of macroscopic, microscopic and organoleptic characters. We hereby authenticate that the given sample belongs to stem of *Tinospora cordifolia* (Willd.) Hook.f. & Thomson, [Family: Menispermaceae].

रिश्तेज कुल / Dr. R. K. Choudhary
(वैज्ञानिक / Scientist)

वैज्ञानिक / Scientist
औषधीय वनस्पति प्रमाणीकरण / Plant Drug Authentication
जैवविविधता एवं पुरावैद्यिकी (वनस्पति एवं झगटम) समूह
Biodiversity and Palaeobiology (Plants & Diatoms) Group

CONDITIONS AND REMARKS:
The user has collected the sample at ARI. We assume that user has obtained necessary permission from territorial state/foreign country. The user has been advised to carry out analysis and examination of the sample with care and to the condition of the sample. It should not be used as an evidence of authenticity in any official, governmental, legal, statutory correspondence or certificate. The contents of this report are confidential and being disclosed only to the party responsible for sample. The user needs to acknowledge the services provided by ARI in their reports, publications, books, monographs etc.

Fig.03: Authentication Letter



EXTRACTION OF GILOY – The powdered giloy stems is extracted for 16 hrs. by using soxhlet apparatus with methanol and acetone in the ratio 70:30 at 40⁰ C. [35]



Fig.04: Extraction of Giloy by Soxhlet Apparatus

Formulation of Cream –



Fig.05- Antiseptic Cream of Giloy

Procedure – Heat liquid paraffin and beeswax in a borosilicated glass beaker at 75⁰ C and maintain that heating temperature (Oil phase). In another beaker dissolve borax, methyl paraben in distilled water and heat this beaker to 75⁰ C to dissolved borax and methyl paraben and to get a clear solution (aqueous phase). Then slowly add this

aqueous phase to heated oily phase, then add measure amount of giloy extract and stir vigorously until it forms a smooth cream then add few drops of rose oil as a fragrance.

Mix the cream in geometric manner in the mortar and pestle to give a smooth texture to the cream and mix all the ingredients.[36]



Evaluation-

• Evaluation of Extract-

Test Performed	Chemical Test	Observation	Result
Test For Alkaloids	A saturated solution of picric acid+Filtrate	Yellow Coloured ppt	Positive
Test for Glycoside	Dil.H2SO4+Filtrate+5% sol of NaOH+Add Fehling solution A&B	Red colour Produce	Positive
Test for Steroids	Conc.H2SO4+Filtrate	Yellow Colour on Top	Positive
Test for Flavonoids	NaOH sol+Filtrate	Yellow Colour Obtained	Positive
Test for Diterpenoids	Conc.H2SO4+Filtrate	The Green Colour on Bottom	Positive

• Evaluation of Cream –

Physical evaluation –

In this test, the cream was observed for colour, odour, texture, state.[37]

Sr. No	Test	A1	A2	A3	A4
1	Colour	Dark Brownish	Yellowish Brown	Brown	Creamy Yellowish
2	Odour	Pleasant	Pleasant	Pleasant	Pleasant
3	Texture	Smooth	Smooth	Smooth	Smooth
4	State	Semi Solid	Semi Solid	Semi Solid	Semi Solid

Table No.03: Physical Evaluation

pH - 0.5 gm cream was taken and dispersed in 50 ml distilled water and then pH was measured by using digital pH meter. [38]



Fig.06: pH Test



Batch No.	Formulation	pH
1	A1	6.9
2	A2	6.5
3	A3	6.6
4	A4	6.8

Table No.04: pH Test Results

Irritancy Test –

Mark the area (1cm) on the left-hand dorsal surface. Then the cream was applied to that area and the time was noted. Then it is checked for irritancy if any for an interval up to 24 hr and reported.

Batch No.	Formulation	Irritant Effect
1	A1	Not Irritant
2	A2	Not Irritant
3	A3	Not Irritant
4	A4	Not Irritant

Table No.05: Irritancy Test Observation

Washability Test –

A small amount of cream was applied on the hand and it is then washed with tap water.

Batch No.	Formulation	Washability
1	A1	Easily Washable
2	A2	Easily Washable
3	A3	Easily Washable
4	A4	Easily Washable

Table No.06: Washability Test Observation

Spreadability –

The spreadability was expressed in terms of time in seconds taken by two slides to slip off from the cream, placed in between the slides, under certain load. Lesser the time taken for separation of the two slides better the spreadability. Two sets of glass slides of standard dimension were taken. Then one slide of suitable dimension was taken and the cream formulation was placed on that slide. Then other slide was placed on the top of the formulation. Then a weight or certain load was placed on the upper slide so that the cream between the two slides was pressed uniformly to form a thin layer. Then

the weight was removed and excess of formulation adhering to the slides was scrapped off. The upper slide was allowed to slip off freely by the force of weight tied to it. The time taken by the upper slide to slip off was noted.

$$\text{Spreadability} = m \times l/t$$

Where,

m = Standard weight which is tied to or placed over the upper slide

l = length of a glass slide

t = time taken in seconds.[39]

Batch No.	Formulation	Time in sec	Spreadability (gm.cm/sec)
1	A1	10	22.5
2	A2	7	32.14
3	A3	8	28.12
4	A4	7	32.14

Table No.07: Spreadability Test Observation



Greasiness Test –

Here the cream was applied on the skin surface in the form of smear and checked if the smear was oily or grease-like.[40]

Batch No.	Formulation	Greasiness
1	A1	Non-Greasy
2	A2	Non-Greasy
3	A3	Non-Greasy
4	A4	Non-Greasy

Table No.08: Greasiness Test Observation

Stability Test –

Physical stability test of Herbal Antiseptic Cream was carried out for four weeks at various temperature conditions like 2°C, 25°C, 37°C. The Herbal ointment was found to be physically stable at different temperature i.e., 2°C, 25°C, 37°C within four Weeks. [41]

Anti-Microbial Test –

Nutrient Agar was prepared using peptone, Yeast extract, Beef extract, Sodium Chloride and agar. For evaluation 2 micro-organism was used S.A and B.S using the cup plate method. The Invitro Antibacterial activity was evaluated using the agar well Diffusion technique.[42]

Sr. No	Ingredients	Gram/Litre
1	Peptone	5.0
2	Yeast extract	1.5
3	Beef extract	1.5
4	Sodium Chloride	5.0
5	Agar	15.0

Table No.09: Nutrient Agar Formulation

$$\text{Zone of Inhibition} = \pi r^2$$



Fig.07: Zone of Inhibition of S.A



Fig.08: Zone of Inhibition of B.S

Viscosity –

The cream was taken in small beaker, P7 spindle was used to check viscosity at 100 RPM using Viscolead Instrument.[43]

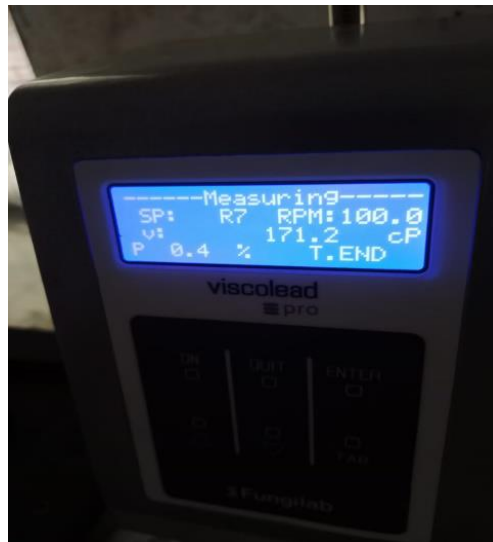


Fig.09: Viscolead Instrument

Batch No.	Formulation	Viscosity	Viscosity
1	A1	2142.3	
2	A2	2182.4	
3	A3	2123.1	
4	A4	2183.7	

Table No 10: Viscosity Result



V. Results and Discussion -

Physical evaluation –

In this test, the cream was observed for colour, odour, texture, state of the four formulations were checked.

pH -

According to the results, the pH of all the four formulations that is A1, A2, A3 and A4 were found to be nearer to skin pH, so it can be safely used on the skin.

Irritancy Test –

Mark the area (1cm) on the left-hand dorsal surface. Then the cream was applied to that area and the time was noted. Then it is checked for irritancy if any for an interval up to 24 h and reported. According to the results all the four formulations that is A1, A2, A3 and A4 showed no sign of irritancy.

Washability Test -

Washability test was carried out by applying a small amount of cream on the hand and then washing it with tap water. All four formulations were easily washable.

Spreadability -

The Spreadability of the four formulations that is A1, A2, A3 and A4 was carried out and out of that for A2 and A4 the time taken by the 2 slides to prepare is less so as said in the description of evaluation test lesser the time taken for separation of the two slides better the spreadability so according to this statement A2 and A4 showed better spreadability.

Greasiness -

Here the cream was applied on the skin surface in the form of smear and checked if the smear was oily or grease-like. According to the results, we can say that all four formulations were non-greasy.

Stability Test -

The Herbal cream was found to be physically stable at different temperature i.e., 2°C, 25°C, 37°C within four Weeks.

Anti-Microbial Test -

The Anti-Microbial Test was performed on two strains of bacteria *S. Aureus* and *B. Subtilis* using cup plate method

Viscosity Test -

The Viscosity Test was performed using Viscolead Instrument.

VI. Conclusion

The extract of *Tinospora Cordifolia* stem was prepared by Soxhlet extraction method and evaluated for phytochemical tests. Antimicrobial

test has been performed and Zone of inhibition of *Tinospora Cordifolia* extract using *S. Aureus* was found to be 8 mm ±, and for *B. Subtilis* was found to be 6 mm ±. By using different concentrations of *Tinospora Cordifolia* extract four antiseptic cream formulation A1, A2, A3 & A4 were prepared. All formulations have been stable at room temperature, formulation A4 showed good spreadability, Viscosity, pH as compare to A1, A2 & A3. So Formulation A4 has been selected as final formulation. As stability studies of herbal formulations is challenging because of complexity of constituents present in it, so further stability studies should be performed to know the shelf life of formulation.

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