

A Research Project
On
“FORMULATION AND EVALUATION
OF HERBAL ANTIFUNGAL AND
ANTIBACTERIAL CREAM”

Submitted to
Dr.Babasaheb Ambedkar Technological University, Lonere,
Raigad.



BY
Miss. Pallavi Bhaskar Aher
B pharmacy (Final Year)

GUIDE
Ms. Priti B. Undre
M.Pharm (Pharmaceutical Assurance Techniques)



Shreeyash Pratishthan's
Shreeyash Institute of Pharmaceutical Education and
Research, Aurangabad
(2021-22)

(Gut No. 258 (P), Satara Parisar, Beed By Pass Road, Near SRPF Camp, Dist.
Aurangabad

A Project Research
on
“FORMULATION AND EVALUATION OF
HERBAL ANTIFUNGAL AND
ANTIBACTERIAL CREAM”

Project

Submitted in the partial fulfilment of
the requirement for the degree of

Bachelor of Pharmacy

in the

Pharmaceutical Sciences

Dr. Babasaheb Ambedkar Technological University, Lonere, Raigad.

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Certificate

This is to certify that Pallavi Bhaskar Aher (20257220181382310058) has completed his/her project work as per **Dr. Babasaheb Ambedkar Technological University, Lonere, Raigad** semester VIII syllabus. He/she was supervised by Ms. Priti B. Undre under the project entitled A Research Project on “**FORMULATION AND EVALUATION OF HERBAL ANTIFUNGAL AND ANTIBACTERIAL CREAM**”

Date:

Dr. Ganesh G. Tapadiya

Place: Aurangabad.

Principal



Shreeyash Institute of Pharmaceutical Education and Research,

Dr. Babasaheb Ambedkar Technological University, Lonere, Raigad.

2021-22

(Gut No. 258 (P), Satara Parisar, Beed By Pass Road, Near SRPF Camp, Dist. Aurangabad)



Certificate

This is to certify that investigations described in this project entitled, A Project *Research* on, "**FORMULATION AND EVALUATION OF HERBAL ANTIFUNGAL AND ANTIBACTERIAL CREAM**" were carried out by **Pallavi Bhaskar Aher** in the laboratories of the "Shreeyash Institute of Pharmaceutical Education and Research, Aurangabad under my guidance in partial fulfilment of the requirements for the degree of Bachelor of Pharmacy in Dr. Babasaheb Ambedkar Technological University, Lonere, Raigad.

This project is now ready for examination.

Date:

Ms. Priti B. Undre

Place: Aurangabad.

Guide

Affectionately dedicated to,

My Sister, Brother & Parents,

whose affection and love infinite.

My friends,

who are always with me in adversity and prosperity

My Teachers,

To whom, I shall remain indebted for giving new

shape and

path to my life....

Acknowledgement

“Success is simple. Do what’s right way, at the right time.”

A single project needs the heads and hands of many for its successful completion. Good number of well-wishers, some who are with me from the beginning some who joined me at some stage during the journey, to complete this project successfully with profound appreciation. I thank all the numerous acquaintances, which has extended support and contribution to my work.

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Pallavi Bhaskar Aher

Place : Aurangabad

B Pharmacy (Final Year)

Abstract :

The main aim of this research was to develop a novel cream formulation consisting of Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha for the treatment of secondary skin infections.

Topical route is most suitable route for skin infections. The development of topical drug delivery systems designed to have systemic effects appears to be beneficial for a number of drugs on account of the several advantages over conventional routes of drug administration.

A novel cream formulation consisting of Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha was prepared. Microbiological studies were performed to evaluate the fungal and bacterial activating.

The developed cream consist of Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha was found to be safe and effective for the treatment of skin infections.

Aim and Objectives :

The main aim of this research was to develop a novel cream formulation consisting of Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha for the treatment of secondary skin infections.

Keywords:

Antibacterial cream, Antifungal cream, Candida albicans, Onychomycosis, Ringworm.

List of Abbreviations

Serial No.	Abbreviations	Full form
1.	Mm	Millimetre
2.	Cm	Centimetre
3.	ml	Milliliter
4.	%	Percentage
5.	Mg	Milligram
6.	G	Gram
7.	L	Liter

Table of Contents

Chapter No.	Chapter Name	Page No.
1.	Literature Review	1
2.	Introduction	2
3.	Material and method	15
4.	Isolation of candida albicans	19
5.	Test for Chemical Constituent	20
6.	Methodology	21
7.	Evaluation of cream	22
8.	Discussion and conclusion	24
9.	Future scope	24
10.	Result	24
11.	References	25

List of Figures

Figures No.	Figures Name	Page No.
1	Physiology of Human Skin	3
2	Distal subungual onychomycosis	5
3	Superficial white onychomycosis	5
4	Proximal subungual onychomycosis	6
5	Candida onychomycosis	6
6	Total dystrophic onychomycosis	7
7	Fingernail Anatomy	8
8	Ringworm	11
9	Athlete's Foot	13
10	Jock Itch	13
11	Yeast Infections	14
12	Ginger Rhizomes	15
13	Withania somnifera	15
14	Curcuma longa rhizome	16
15	Neem Tree	17
16	Psidium guajava	17
17	Herbal Extraction	18
18	Chemical	18
19.1	Vegtable Sample Candiada albicans	19
19.2	Soil Sample Candida Albicans	19
20	antibacterial and antifungal herbal cream	21
21	pH measurement	22
22	Smear	22
23	formulated cream showing zone of inhibition	23

List of Table

Table No.	Table Name	Page No.
1	Scientific Classification of Ginger Rhizomes	15
2	Scientific Classification of Withania somnifera	15
3	Scientific Classification Curcuma longa	16
4	Scientific Classification of Neem Tree	17
5	Scientific Classification Psidium guajava	17
6	Chemical and Its Industry Name	18
7	Composition of Agar Media	19
8	Formula for cream	21
9	Antifungal and Antibacterial Studies	23
10	Result	24

1} Literature Review

1. **Neelam N. et al** Nails of mammals are specialized epidermal derivatives which protect the delicate tip of fingers and toes against trauma and act as tools or weapons. Fungal infections of the nails or onychomycosis accounts for about 50% of the nail diseases. Oral antifungal drugs and topical creams and lotions are used for treating fungal nail infections. The disadvantages of oral antifungal agents are toxicity and longer treatment period and topical preparation may get wiped off causing less absorption of drug into nails.
2. **Ashlesha Pravin Pandit et al** An attempt was made to prepare transparent nail lacquer containing natural antifungal agent obtained from extract of whole plant of *Cissus quadrangularis* for the treatment of onychomycosis. Methods: The extract of *C. quadrangularis* was evaluated for antifungal study against *Candida albicans*
3. **Amol Pimpale et al** : The main aim of our research was to develop a novel cream formulation consisting of Garlic oil for the treatment of secondary skin infections. Topical route is most suitable route for skin infections. The development of topical drug delivery systems designed to have systemic effects appears to be beneficial for a number of drugs on account of the several advantages over conventional routes of drug administration.
4. **Leodevico L Ilag et al** Ringworm is a frequently encountered infection commonly known as tinea. Most topical treatments are chemical-based and typically work against mild tinea infections, but are not effective in treating severe cases.
5. **Wysocki AB et al**: Human skin is the largest multifunctional organ of the body, and knowledge of its structure and function is essential to clinicians and researchers. The skin has two layers, the epidermis and dermis, separated by a basement membrane zone. It provides protection, sensation, thermoregulation, biochemical/metabolic, and immune functions.

2} Introduction:

2.1} Creams:

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. 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.

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).

- **Type of Skin Creams:** They are divided into two types

1} Oil-in-Water (O/W): Creams which 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.

2} Water-in-Oil (W/O): Creams which 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.

- **Classification of Creams:**

All the skin creams can be classified on different basis:

1. According to function, e.g., cleansing, foundation, massage, 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.

2.2} Physiology of Human Skin

Human skin is the largest multifunctional organ of the body. The skin has two layers, the epidermis and dermis, separated by a basement membrane zone. It provides protection, sensation, thermoregulation, biochemical/metabolic, and immune functions.

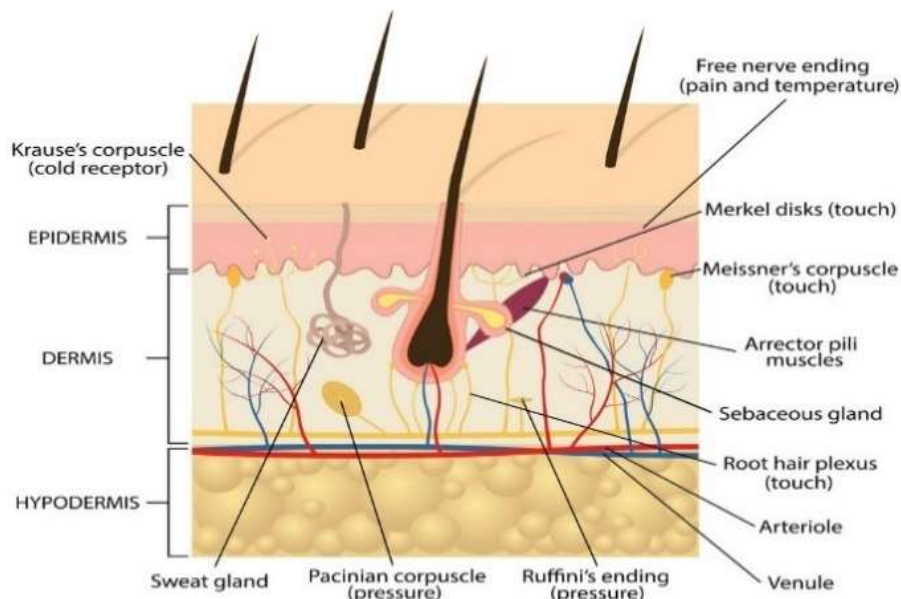


Fig. No.1 Physiology of Human Skin

- **Epidermis:** The epidermis is the most superficial layer of the skin and is composed of stratified keratinized squamous epithelium, which varies in thickness in different parts of the body. It is thickest on the palms of the hands and soles of the feet. There are no blood vessels or nerve endings in the epidermis, but its deeper layers are bathed in interstitial fluid from the dermis, which provides oxygen and nutrients, and drains away as lymph.
- **Dermis:** The dermis is tough and elastic. It is formed from connective tissue and the matrix contains collagen fibers interlaced with elastic fibers. Rupture of elastic fibers occurs when the skin is overstretched, resulting in permanent striae, or stretch marks, that may be found in pregnancy and obesity. Collagen fibers bind water and give the skin its tensile strength, but as this ability declines with age, wrinkles develop. Fibroblasts, macrophages and mast cells are the main cells found in the dermis. Underlying its deepest layer there is areolar tissue and varying amounts of adipose (fat) tissue. These consist of secretory epithelial cells derived from the same tissue as the hair follicles. They secrete an oily substance, sebum, into the hair follicles and are present in the skin of all parts of the body except the palms of the hands and the soles of the feet. They are most numerous in the skin of the scalp, face, axillae and groins. In regions of transition from one type of superficial epithelium to another, such as lips, eyelids, nipple, labia minora and glans penis, there are sebaceous glands that are independent of hair follicles, secreting sebum directly onto the surface.

- **Functions of Skin:** Skin performs the following functions:
 - a) Protection:** An anatomical barrier from pathogens and damage between the internal and external environment in bodily defense, Langerhans cells in the skin are part of the adaptive immune system.
 - b) Sensation:** Contains a variety of nerve endings that react to heat and cold, touch, pressure, vibration, and tissue injury, see soma to sensory system and haptics.
 - c) Heat regulation:** The skin contains a blood supply far greater than its requirements which allows precise control of energy loss by radiation, convection and conduction. Dilated blood vessels increase perfusion and heat loss, while constricted vessels greatly reduce cutaneous blood flow and conserve heat.
 - d) Control of evaporation:** The skin provides a relatively dry and semi-impermeable barrier to fluid loss. Loss of this function contributes to the massive fluid loss in burns.
 - e) Aesthetics and communication:** Others see our skin and can assess our mood, physical state and attractiveness.
 - f) Storage and synthesis:** Acts as a storage center for lipids and water, as well as a means of synthesis of vitamin D by action of UV on certain parts of the skin.
 - g) Water resistance:** The skin acts as a water-resistant barrier so essential nutrients aren't washed out of the body.

2.3} Onychomycosis:

Onychomycosis is a contagious contamination of fingernails and toenails which influences around 19% of world population. This infection occurs mainly in diabetic and elderly patients. The most common cause is dermatophytes, non-dermatophytes, moulds and yeast mainly *Candida albicans*. About 80% cases of onychomycosis, mostly toenails are affected. In diabetic patients, onychomycosis is prevalent than non-diabetic patients. Diseased nail of patient has thick sharp edges infect surrounding skin tissue due to pressure that leads to erosion of nail bed.

The infection of the nail folds is associated with proximal subungual onychomycosis, distal and lateral subungual onychomycosis and primary total dystrophic onychomycosis. Yellow-brown patches appear at lateral border of nail.

The masses of horny debris disperse and nail plate rapidly becomes thickened and broken. The irritation, pain and pressure also are the secondary effects.⁷ To treat various fungal infections of nail, variety of formulations are available either by oral or topical route like solution, cream, gel and nail patch.

There are four distinct types of clinical presentation of onychomycosis:

- i. Distal subungual onychomycosis
- ii. Superficial white onychomycosis
- iii. Proximal subungual onychomycosis
- iv. Candida onychomycosis

They are described as follows:

1) Distal subungual onychomycosis: It is the most common form of the fungal invasion of the nail. This occurs when fungus invades the nail bed and under surface of the nail plate via hyponychium and spreads proximally along the ridges of the nail bed.

Fig No. 2 Distal subungual onychomycosis



Distal and lateral subungual onychomycosis may be confined to one side of the nail or spread sideways to involve the whole of the nail bed, and progresses relentlessly until it reaches the posterior nail fold. Eventually the nail plate becomes friable and may break up, often due to trauma, although nail destruction may be related to invasion of the plate by dermatophytes that have keratolytic properties. A thickened nail and a large amount of debris under the nail may cause discomfort when wearing shoes. Figure 2 given below represents distal subungual onychomycosis.

Features of DLSO are as follows:

- Subungual hyperkeratosis and onycholysis is usually yellow-white in color
- Yellow streaks and/or yellow onycholytic areas in the central portion of the nail plate

2) Superficial white onychomycosis:



Fig. No. 3 Superficial white onychomycosis

The second most common type of fungal nail infection accounting about 10% of onychomycosis. It affects the top layer of the nail by forming white spots on the surface of the nail. Eventually the entire nail surface becomes covered with a crumbly and chalky powder. It occurs when fungus localizes superficially on the dorsal nail plate and form colonies seen as opaque scaly plaques with distinct edges that are scraped away easily.

The nail becomes rough, soft and crumbly. Figure 3 given below represents superficial white onychomycosis.

Features of WSO are as follows:

- Confined to the toenails
- Small, white, speckled or powdery patches on the surface of the nail plate
- The nail becomes roughened and crumbles easily
- Molds produce a deep variety of WSO characterized by a larger and deeper invasion of nail plate.

3) Proximal subungual onychomycosis:



Fig. No. 4 Proximal subungual onychomycosis

In proximal subungual onychomycosis the fungal element invades the deeper, ventral aspect of the nail plate from proximal portion of the nail and migrates distally, causing a band or a patch of leukonychia that moves distally with nail growth. It is relatively uncommon subtype that can suggest the possibility of HIV infection or other types of immunosuppression. It can also occur secondarily to paronychia i.e. infection and inflammation of nail folds. Figure 4 given below represents proximal subungual onychomycosis.

Features of PSO are as follows: It is an area of leukonychia in proximal nail plate that moves distally with nail infection. In PSO caused by molds, leukonychia is associated with marked periungual inflammation

4) Candida onychomycosis:



Fig. 5 Candida onychomycosis

Patients with chronic mucocutaneous candidiasis have more chances of developing candidal infection of the nails. Candida species invade the nails previously damaged by infection or trauma. Candidal paronychia commonly affects the hands and usually occurs in persons who frequently immerse their hands in water. Swelling of the posterior nail fold occurs secondary to chronic immersion in water or due to allergic reactions to some food items, and the cuticle becomes detached from the nail plate. Yeasts and bacteria, enters the subcuticular space causing swelling of the posterior nail fold. Figure 5 given up represents candida onychomycosis.

Features of candida onychomycosis are as follows:

- Affects several or all digits
- The digits often take on a bulbous or drumstick appearance
 - In more severe cases gross thickening of the nails occurs, which amounts to a Candida granuloma.
- Secondary candida onychomycosis occurs due to other diseases of the nail, mostly psoriasis
- The nails typically are discoloured white or yellow and deformed.

5) Total dystrophic onychomycosis:



Fig. No. 6 Total dystrophic onychomycosis

In total dystrophic onychomycosis, the nail is opaque, thickened, and yellow-brown and/or greenish-brown to black. The entire plate and matrix are affected. Total dystrophic onychomycosis may be end result of any of the four main forms of onychomycosis. This condition is characterized by the total destruction of the nail plate. It is the end stage of the disease. Figure 6 given below represents total dystrophic onychomycosis.

- **Causes of Fungal Infections:** Warm and moist environment helps fungi to grow and cause infection. Wearing occlusive footwear or using shower stalls, locker rooms or bathrooms can increase the risk of fungal infections.
- **Symptoms of Nail Infections:** At the start, there are usually no symptoms. The nails may become so thick that they hurt when pressed on the inside of a shoe. They then become hard to trim. The appearance of an infected nail, particularly a fingernail, may cause embarrassment. The skin nearby may also have a fungal infection; it may itch, crack, form a blister or appear white, especially between the

toes. When fungi infect a nail, they usually start at its free edge spreading down the side of the nail towards the base of the cuticle. Eventually the whole nail may be involved. The infected areas turn white or yellowish, and become thickened and crumbly. There may be white areas on the surface of the nail. The nails most commonly affected by fungal infections are on the big and little toes. Sometimes, especially in those who carry out regular wet work such as housewives or cleaners, the skin around the fingernail becomes red and swollen. This is called paronychia, and can allow infection.

Prevention: Fungal nail infections can be prevented by the following methods-

- Avoiding injury to nails, which may increase the risk of developing a nail infection.
- Wearing footwear such as flip-flops in public places, such as communal bathing/shower places, locker rooms, etc.
- Avoid towel sharing.
- Replacing old footwear, as it might be contaminated with fungal spores.

Treatment: Various oral and topical therapies have been used in the past to treat onychomycosis.

1) Oral: Antifungal tablets will often clear a fungal nail infection. The medication helps to clear any associated fungal skin infection, such as athlete's foot

2) Topical: Creams, lotions and gels are used for treating fungal infections of nails. The preparation is rubbed into infected nails after soaking. Rubbing helps the medication get through the hard nail surface to the underlying fungus. Topical preparations contain urea as antifungal agent.

3) Device based treatment: Device based treatment are rapidly expanding area for treatment of onychomycosis. Various devices can be used to enhance the delivery of drug, helps to activate the topical drugs applied o by killing the fungi photo thermally. It includes laser treatment and iontophoresis.

2.4} Nail:

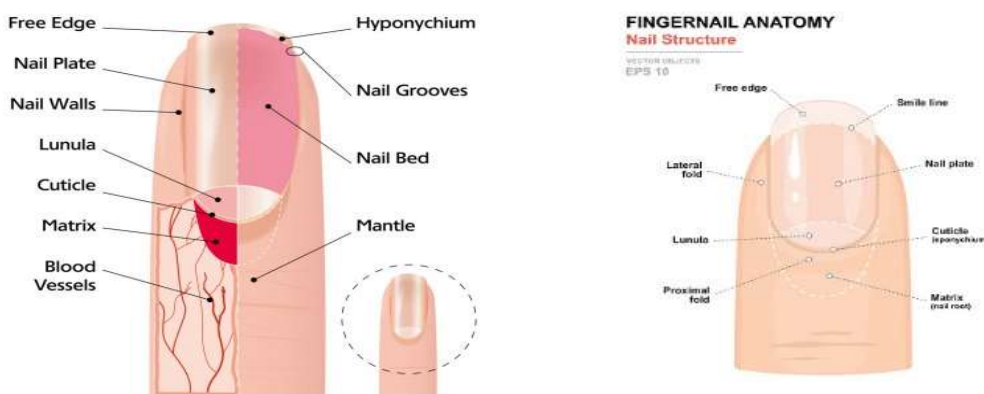


Fig. No. 7 Fingernail Anatomy

Nail is a hard covering of finger tips and toe tips. They are ectodermal appendages covering the dorsal aspects of the digits. It is a horn-like envelope used to protect soft ends of the phalanges or digits. It enhances the capacity for fine digital movements and tactile sensation. This allows humans to use them as precise tools for picking up objects. Human nails consist of many parts: the nail plate, the nail bed, matrix, hyponychium, etc. They are described below

1) The Nail Plate: The nail plate is a thin, hard, yet slightly elastic, translucent, convex structure and is made up of approximately 25 layers of dead keratinized, flattened cells. It is about 0.25 -0.6mm for fingernails and up to 1.3mm for toenails. The nail plate appears pink because of the underlying capillaries. The upper surface of the nail plate is smooth and shiny and may have variable number of longitudinal ridges that change with age resulting in the loss of nail plate lustre with age.

2) The Matrix: It is also called as nail root. It is the thickened part of the nail bed and is composed of 3-20 layers of actively dividing cells o stratum germinativum. New cells are added to the root and by this constant addition nail plate is pushed slowly forward over nail bed about 0.5mm/week.

3) Lunula: It is a whitish, crescent-shaped area. The opacity of lunula is probably caused by considerable thickness of nail matrix.

4) The Nail Bed: Nail bed is formed of several layers of epidermal cells, corresponding to stratum Basale and stratum spinosum of epidermis underlying the nail plat. Except the nail matrix, these cells do not participate in the formation of nail plate.

5) Cuticle: The cuticle is also known as eponychium. It is situated between the skin of the finger and the nail plate. It fuses these structures together, providing a waterproof barrier. It forms the seal between the nail and the digit to prevent foreign matter from entering.

6) Perionychium: It is the skin that overlies the nail plate on its sides also known as the paronychia edge. The perionychium is an indication of ingrown nails, hangnails and an infection of the skin called paronychia.

7) Hyponychium: It is the area between the nail plate and the fingertip. It is the junction between the free edge of the nail and the skin of the fingertip, providing a waterproof barrier and prevents the penetration of foreign bodies, dirt and invasion of pathogens that cannot digest keratin. It is composed of tough keratin layer that seals the subungual space. The rate of growth and length of nails is related to the length of the terminal phalanges i.e. outermost finger bones. The nail of index finger grows faster than that of the little finger. Fingernails grow faster than toenails up to four times. In humans, nails grow at a rate of 3 mm a month. Toenails require 12 to 18 months and fingernails require three to six months to regrow completely. Actual growth rate is dependent upon season, exercise level, sex, age, diet, etc. Nails grow faster in the summer.

- **Functions of Nails:**

- Fingernails help to protect fingers. If they weren't there the tips of your fingers would get all scratched up.

- It also helps us the grab hold of things.
- A healthy finger nail has the function of protecting the fingertip, the distal phalanx, and the surrounding soft tissues from injuries.
- It serves to enhance precise delicate movements of the distal digits through counter-pressure exerted on the pulp of the finger.
 - The nail then acts as a counterforce when the end of the finger touches an object, thereby enhancing the sensitivity of the fingertip
- The nail functions as a tool, enabling for instance, a so-called "extended precision grip" and helps in picking different objects.
- The most important function of the fingernails consists in enhancing tactile discrimination and fine movements.
- Fingernails are utilized for scratching and grooming and are an efficient natural weapon.
- Toenails protect the distal toes and contribute to pedal biomechanics. The nails also contribute to the aesthetic appearance of the hand and foot.
- **Histology, Ultrastructure and Composition:** The epidermis of the nail bed is similar to that of the skin, but has no stratum lucidum or stratum granulosum and no hair follicles or sweat glands. The nail plate is made up of impacted and adhering layers of flattened and cornified cells i.e. dead onychocytes that have lost their nuclei. The cells contain hard keratin, similar to that of hair, with high sulfur content mainly in the form of cysteine, which comprises about 9-12 % of the weight of the nail. Nails contain about 7-12% of moisture and 0.15-0.76% of fat, a little more about 1.38% in infants. Calcium constitutes about 0.02-0.04% of the weight and does not contribute to the hardness. The nail bed and matrix have a rich supply of blood from 2 arteries that run lateral along the digit and form an oxygen-rich capillary bed, which lies below the nail plate.
- **Nail Pathologies:** Nails are important to an individual's overall appearance, people spend time cutting, filing and decorating them in order to look presentable. Indeed, individuals with nail dystrophies often suffer from considerable pain and discomfort, can have difficulty in walking and are at risk for significant complications, including bacterial infection and cellulitis. Although, there are a variety of disorders that can affect the nails, more than half of all nail dystrophies are caused by fungal infections i.e. onychomycosis.

2.5} Ringworm:



Fig. No. 8 Ringworm

Ringworm of the body is a skin infection caused by a fungus. The medical term for ringworm is *tinea corporis*. “Tinea” means fungus, the cause of the rash, and “corporis” means the body. It’s a superficial fungal skin infection caused by dermatophytes, which are a type of fungus. It can occur on the:

- torso
- arms
- legs
- neck

The condition is common and highly contagious, but it’s not serious.

Symptom: Symptoms of ringworm on your body usually start about 4 to 14 days Trusted Source after contact with the fungus. Ringworm can affect any area of your skin, and it may also be found on fingernails and toenails. Symptoms usually include:

- a ring-shaped rash
- red skin that is scaly or cracked
- hair loss
- itchy skin

Symptoms may also vary based on where ringworm is located on your body:

- **Scalp ringworm (*tinea capitis*)** may produce a bald spot that is scaly, red, and itchy. Multiple bald spots can appear if the infection spreads across the scalp. This may be mistaken for severe dandruff.
- **Feet ringworm (*tinea pedis*)** may cause skin between your toes to peel, itch, or turn red and swollen. It can also cause a pink or scaly rash stretching across your feet. In some severe cases, *tinea pedis* may cause blisters on your feet.
- **Groin ringworm (*tinea cruris*)** can cause red spots that are scaly and itchy in the skin folds on your inner thigh.
- **Beard ringworm (*tinea barbae*)** can cause spots that are red, scaly, and itchy. This may occur on your chin, cheeks, and upper parts of your neck. The spots can be filled with pus, and the affected hair may fall out. The spots may also crust over.

- **Causes and risk factors:** Ringworm is caused by fungi. The types of fungi that cause ringworm tend to thrive in warm and humid areas like locker rooms and indoor pools. It can also be found in soil, gyms, and animals, or on objects like hats, towels, and combs.

It's still possible to get ringworm even in cool weather, due to how contagious it is factors that may increase your risk include:

- living in damp, hot, or humid areas
- excessive sweating
- participating in contact sports
- wearing tight clothing
- having a weak immune system
- sharing clothing, bedding, or towels with others
- diabetes

A ringworm infection can spread in many direct and indirect ways, including:

- **Person to person.** This happens when you have direct contact with the skin of a person who has ringworm.
- **Animal to person.** This occurs through direct contact with an animal that has ringworm. Both dogs and cats can spread the infection to people. Ferrets, horses, rabbits, goats, and pigs can also spread ringworm to people.
- **Inanimate item to person.** It's possible to get ringworm through indirect contact with objects, including the hair of a person with ringworm, bedding, clothing, shower stalls, and floors.
- **Soil to person.** Rarely, a ringworm infection can spread through contact with affected soil for an extended amount of time.

Prevention: We can prevent trusted source ringworm of the body by limiting contact with someone who has the infection. This includes both indirect and direct contact with that person.

Precautions we can follow include:

- not sharing towels, hats, hairbrushes, or clothing with someone who has ringworm
- taking your pet to a vet if you suspect a ringworm infection
- maintaining personal hygiene around other people if you have ringworm of the body, and not scratching the affected areas of your skin
- drying your skin well after a shower, especially between your toes and where skin touches skin, such as in your groin and armpits.

2.6} Athlete's foot:



Fig. No. 9 Athlete's Foot

Athlete's foot, also called tinea pedis, is a fungal infection of your foot. The fungi grow best in warm, moist places such as shoes, socks, swimming pools, locker rooms, and public showers.

Athlete's foot symptoms: Signs of athlete's foot vary from person to person. You might have:

- Peeling, cracking, and scaly feet
- Blisters
- Skin that's red, softened, or broken down
- Itching
- Burning

2.7} Jock Itch



Fig. No. 10 Jock Itch

A type of fungus called tinea causes jock itch. The infection is also known as tinea cruris. Tinea loves warm, moist areas like your genitals, inner thighs, and buttocks. Infections happen more often in the summer or in warm, wet climates. Jock itch is a red, itchy rash that's often ring-shaped.

- **Jock itch symptoms:** Symptoms of jock itch include:
 - Itching, chafing, or burning on your groin or thigh
 - A red, circular, rash with raised edges
 - Redness on your groin or thigh
 - Flaking, peeling, or cracking skin

2.8} Yeast Infections



Fig. No. 11 Yeast Infections

Yeast infections of your skin are called cutaneous candidiasis. A type of fungus called candida causes these infections when it grows too much. Yeast infections aren't contagious.

- **Yeast infection symptoms** : Signs of a yeast infection on your skin include:
 - Rash
 - Patches that ooze clear fluid
 - Pimple-like bumps
 - Itching
 - Burning

The infections are most common in warm, moist, creased areas of your body, including your armpits and groin. They often happen in people who are obese or who have diabetes. Candida can cause diaper rash in infants. It can also cause infections in your nails, vagina, or mouth (oral thrush). Signs of a yeast infection in your nail beds include:

- Swelling
- Pain
- Pus
- A white or yellow nail that separates from the nail bed
- This antifungal cream is used to treat fungal infection .

2.9} Bacterial skin Infections: Bacterial skin infections often begin as small, red bumps that slowly increase in size. Some bacterial infections are mild and easily treated with topical antibiotics, but other infections require an oral antibiotic. Different types of bacterial skin infections are:

- Cellulitis
- Impetigo
- Boils
- Hansen's disease (leprosy)

2.10} Antifungal cream: Antifungal cream is used for treat fungal infection fungal infection also called mycosis, is a skin disease caused by fungus.

- **Types of Fungal Infections:** Fungal skin infections can happen anywhere on your body. Some of the most common are athlete's foot, jock itch, ringworm, and yeast infections.

3} Material and Method

3.1} Materia Herbs with Antifungal Activity:

1} Zingiber Officinale:

Domain	Eukarya
Kingdom	Plantae
Phylum	Magnoliophyta
Class	Liliopsida
Order	Zingiberales
Family	Zingiberaceae
Genus	Zingiber Mill
Species	Zingiber officinale



Fig. No.12 Ginger Rhizomes

Table No.1 Scientific Classification of Ginger Rhizomes

- **Synonym:** Zingiber, Ginger, Sunthi
- **Parts Used:** rh'zomes
- **Chemical Constituents:** Rhizome of ginger contains essential oil upto 4%, phenolic compounds, flavonoids, carbohydrates, proteins, alkaloids, glycosides, saponins, steroids, terpenoids and tannin.
- **Method of Extraction:** Maceration with ethanol for 3 days.

2} Withania Somnifera:

Kingdom	Plantae
Subkingdom	Tracheobionta
Super division	Spermatophyta
Division	Angiosperma
Class	Dicotyledons
Order	Tubiflorae
Family	Solanaceae
Genus	Withania
Species	somnifera



Fig. No.13 Withania somnifera

Table No.2 Scientific Classification of Withania somnifera

- **Synonym:** Ashwagandha, Winter Cherry, asgandh
- **Parts Used:** Leaves, fruits, stem and roots.

- **Chemical Constituents:** The biologically active chemical constituents are alkaloids ashwagandhine, cuscohygrine, anahygrine, tropine. Steroidal compounds, withaferin A. Other constituents include saponins. The plant also contains constituents like withaniol, acylsteryl glucosides, starch, reducing sugar, hantreacotane, ducitol. A variety of amino acids including aspartic acid, proline, tyrosine, alanine, glycine, glutamic acid, cystine, tryptophan, and high amount of iron.
- **Method of Extraction:** Maceration with alcohol for 3 days.

3} *Curcuma Longa* :

Kingdom	Plantae
Subkingdom	Tracheobionts
Super division	Spermatophyta
Division	Mangoliophyta
Order	Zingiberales
Family	Zingiberaceae
Genus	Curcuma
Species	Longa
Scientific name	Curcuma longa



Fig. No. 14 *Curcuma longa* rhizome

Table No.3 Scientific Classification *Curcuma longa*

- **Synonym:** Curcum, Haridra, Haldi, Turmeric, Indian Saffron
- **Parts Used:** Rhizomes.
- **Chemical Constituents:** Turmeric comprises volatile as well as nonvolatile compounds. It contains fat, protein, carbohydrates, minerals and moisture. Essential oil of turmeric rhizomes possesses sabinene, borneol, α -phellandrene, cineol, sesquiterpines, zingiberene and curcumin. Volatile compounds are turmerone, zingiberene, curlone and arturnerone. The nonvolatile components include the curcuminoids.
- **Method of Extraction:** Maceration with Ethanol for 3 days.

4} Azadirachta Indica:

Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Order	Sapindales
Family	Meliaceae
Genus	Azadirachta
Species	A.indica
Scientific Name	Azadirachta indica



Fig. No. 15 Neem Tree

Table No.4 Scientific Classification of Neem Tree

- **Synonym:** Margosa, Nimba.
- **Parts Used:** Leaves.
- **Chemical Constituents:** Neem leaves contains several active ingredients such as desactylimbin, quercetin and sitosterol, triterpenes or the limonoids such as meliantriol, azadirachtin, desactylimpin, quercetin, sitosterol, nimbin, nimbinin, nimbidin, nimboesterol and margisine, different bitter substances such as alkaloids, phenols, resins, glycosides, terpenes and gums.
- **Method of Extraction:** Maceration with ethanol for 3 days.

5} Psidium Guajava:

Kingdom	Plantae
Clade	Tracheophytes
Clade	Angiosperms
Clade	Eudicots
Clade	Rosids
Order	Myrtales
Family	Myrtaceae
Genus	Psidium



Fig. No.16 Psidium guajava

Table No.5 Scientific Classification of Psidium Guajava

- **Synonym:** Guajava pyrifera
- **Common names.** Guava, lemon guava, mpera (Kiswahili), mubera (Kikuyu), mupeera (Luganda)
- **Parts Used:** leaves

- **Chemical Constituents:** triterpenoid guajanoic acid, beta-sitosterol, uvaol, oleanic acid, ursolic acid.
- **Method of Extraction:** Maceration with ethanol for 3 days.

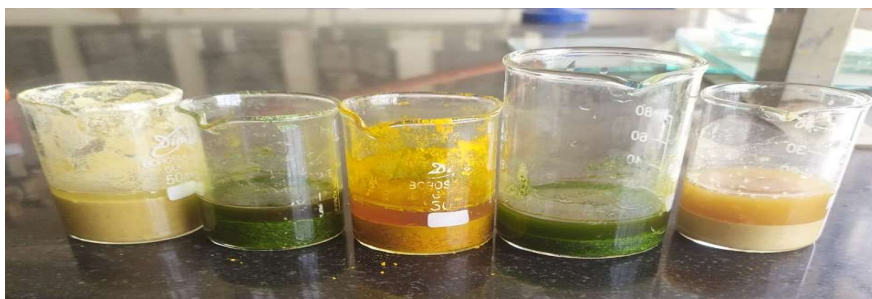


Fig. No. 17 Herbal Extraction

Sr. No.	Chemical Name	Industry
1	Propylene, glycol, propyl paraben, Mineral oil, cetyl alcohol	Dipa chemical Industry Aurangabad.
2	Bees wax, steric acid	Modern Industries, Nashik
3	Stearyl alcohol, Triethanolamine, methyl paraben	Merck specialities limited, Mumbai.
4	Extract of Neem, Guava, Turmeric, Ginger, Ashwagandhas	Prepared in laboratory.

Table No. 6 Chemical and Its Industry Name



Fig. No. 18 Chemical

3.2} Preparation of cream formulation

- **Preparation of oil phase:** White Bees Wax, stearic acid, stearyl alcohol, cetyl alcohol were melted in a stainless steel vessel. To this mixture Liquid paraffin were added and allowed to melt. The temperature of oil phase maintained between 65 – 70°C
- **Preparation of Aqueous phase:** Water was heated to 65 – 70°C. In this weighed propylene glycol, triethanolamine, methyl paraben and propyl paraben were added the temperature of the phase was maintained at 65 – 70°C.
- **Development of Cream formulation:** Oil portion was then slowly incorporated into the aqueous phase at 65-70°C and mixed for 10 to 15 Minutes. When the water and oil phase were at the same temperature, the aqueous phase was slowly added to the oil phase with moderate agitation and was kept stirred until the temperature dropped to 40°C. and Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha was added to it. The emulsion was cooled to room temperature to form a semisolid cream base. pH of cream kept between 4.5 – 6.

4} Isolation of candida albicans

1. Make a sabouraud dextrose Agar containing

Sr. No.	Name	Quantity
1	Dextrose	4 gm
2	Peptone	19 m
3	Agar	2.5 gm
4	Distilled water	100 ml

Table No. 7 Composition of Agar Media

2. After making agar, petri plate, test tube, measuring cylinder, agar place in Autoclave for sterilization at 154 rpm of 15 min.

3. After sterilization add agar on petri plate prepare dilution of soil sample and vegetable sample.

4. After solidifying agar by using nichrome wire loop by add soil sample by using four corner method and also add vegetable sample on another petri plate by using strike plate method.

5. After that place petri plate into incubator for 2 days at temp 35°C . After 2 days candida albicans grow on Petri plate.



Fig. No. 19.1 Vegetable Sample Candida albicans



Fig. No.19.2 Soil Sample Candida Albicans

5} Test for chemical constituents

5.1} Test for flavonoids: The stock solution (1 mL) was taken in a test tube and added few drop of dilute NaOH solution. An intense yellow colour was appeared in the test tube. It became colourless when on addition of a few drop of dilute acid that indicated the presence of flavonoids.

5.2} Litmus Test for phenol detection: Litmus paper changes colour if the solution is acidic or basic. Acidic solutions turn blue litmus paper into red and basic solutions turn red litmus paper into blue. Phenols are acidic, so they show the litmus test.

5.3} Liebermann-Burchard test for steroid detection: When chloform solution of steroid is treated with aseptically anhydride and concentrated sulphuric acid, green colour is formed.

5.4} Test for Terpenoids (Salkowski test) : Five ml of each extract was mixed in 2 ml of chloroform, and concentrated H₂SO₄ (3 ml) was carefully added to form a layer. A reddish brown colouration of the inter face was formed to show positive results for the presence of terpenoids

5.5} Molisch's test for Carbohydrates: Few drops of Molisch's reagent were added to the portion of sample dissolved in distilled water; this was then followed by addition of 1 ml of conc. H₂SO₄ by the side of the test tube. The mixture was then allowed to stand for two minutes and then diluted with 5 ml of distilled water. Formation of a red or dull violet colour at the interphase of the two layers was a positive test . Test for Quinones About 0.5 g of plant extract was taken and added 1 ml of extract and 1 ml of con. H₂SO₄ was added formation of red colour shows the presence of quinones.

6} Methodology:

Part A (Oily Phase)		Part B (Aqueous Phase)		F1	F2	F3
Ingredient	Quantity	Ingredient	Quantity			
Stearic Acid	2.5%	Propylene Glycol	5%			
White Bees Wax	1.5%	Triethanolamine	2%			
Stearyl Alcohol	5%	Methyl Paraben	0.01%			
Cetyl Alcohol	6.5%	Propyl Paraben	0.04%			
		Neem Extract	2%	1%	1.5%	1.7%
		Guava Extract	2%	1%	1.5%	1.7%
		Turmeric Extract	2%	1%	1.5%	1.7%
		Ginger Extract	2%	1%	1.5%	1.7%
		ashwagandha extract	2%	1%	1.5%	1.7%
Mineral Oil	5%	Water	Up to 100%			

Table No. 8: Formula for cream



cream

Fig. No.20 antibacterial and antifungal herbal cream

7} Evaluation parameters

7.1} Physical examination: The cream is yellow, appealing appearance and smooth texture, and they were all homogenous with no signs of phase separation.

7.2} Viscosity: The viscosity of formulated creams was measured by Brook field Viscometer .

7.3} Tube extrudability: The method adopted for evaluating cream formulation for extrudability was based upon the quantity in percentage cream extruded from tube on application of finger pressure . More quantity extruded better was extrudability.

7.4} pH measurement :



Fig. No. 21 pH measurement

The pH of the cream was found to 6.2. The pH should not be too acidic as it may cause skin irritation and should not be too alkaline as it may cause scaly skin.

7.5 Homogeneity: The formulation was tested for the homogeneity by visual appearance and by touch.

7.6} Removal: The ease of removal of the creams applied was examined by washing the applied part with tap water.

7.7} Type of smear: After application of cream, the type of film or smear formed on the skin were checked.



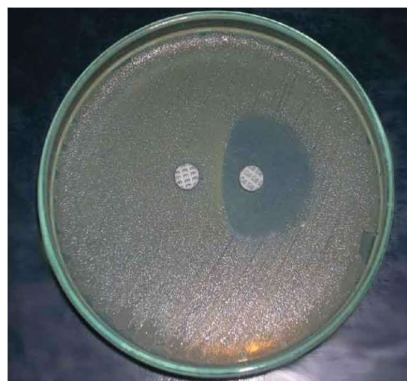
Fig. No. 22 Smear

7.8} Microbiological studies:

From the microbial study it was found that the cream showing good effects on microbial growth and the zone of inhibition was calculated by zone reader. The zone of inhibition of candida albicans was 42.32 mm and of E.coli it was 34.16 mm.



a) Against candida albicans



b) Against E.coli

Fig No.23: formulated cream showing zone of inhibition

bacteria	Candida albicans	E.coli
Zone of inhibition	42.32 mm	34.16 mm

Table No. 9 Antifungal and Antibacterial Studies

8} Discussion and Conclusion:

From the above compiled data the study clearly shows that the formulation is showing good anti-fungal activity against E.coli and candida albicans.

The formulation of antimicrobial agents along with Extract of Neem, Guava, Turmeric, Ginger, Ashwagandha exhibited enhanced rate of diffusion and antibacterial activity. The results of different chemical and physical tests of cream showed that it could use topically in order to protect against skin infections caused by fungus or bacteria.

9} Future Scope:

Advantage of herbal cream over cream by using chemical.

10} Result:

Sr. No.	Evaluation test	Result
1	Physical Appearance	Good
2	Viscosity	Good
3	Tube Extrudability	Good
4	PH	6.2
5	Homogeneity	Good
6	Removal Test	Good
7	Antimicrobial Studies	34.16 MM Zone of Inhibition
8	Antifungal Studies	42.32 MM Zone of Inhibition

Table No. 10 Result

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