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FORMULATION AND EVALUATION OF POLYHERBAL CREAM FOR VITILIGO

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ABSTRACT

Vitiligo is a skin condition characterized by decreased pigmentation and the appearance of white patches due to destruction of melanocytes. The present study was undertaken to formulate and evaluate a polyherbal cream containing Psoralea corylifolia, Eclipta alba and turmeric for vitiligo. Psoralea corylifolia seeds and Eclipta alba were extracted using 95% alcohol after 24 h extraction, and three formulations (F1, F2 and F3) were prepared with suitable excipients. The creams were evaluated for physical properties, irritancy, washability, phase separation, spreadability, greasiness and pH. Phytochemical screening of powdered Psoralea corylifolia showed psoralen strongly present and phenol, alkaloids, anthraquinone glycosides, cardiac glycosides and coumarins present. In vitro hydrogen peroxide scavenging activity was assessed using a modified Ruch et al. method at 276 nm. The sample showed an absorbance of 0.222 compared with a control absorbance of 1.189, corresponding to 81.33% inhibition.

KEYWORDS: Psoralea corylifolia; Eclipta alba; Turmeric; Polyherbal cream; Vitiligo; Antioxidant activity.

1. INTRODUCTION

Traditional Medicinal System

Traditional medicine has a rich history of ethnic customs and cultural heritage. It is described as a practice and set of skills based on indigenous theories, beliefs and experiences in various cultures for maintenance of healthcare and for diagnosis, treatment and prevention of physical and mental illness. Traditional and ethno-medicine served as the foundation for human healthcare until 1899, when Bayer introduced aspirin. Each culture developed healing practices based on natural resources through gradual clinical trial and error. The dissertation states that an estimated 4.6 billion people worldwide rely on traditional medicinal systems for their drug supply.[1]

Herbal Medicine

Herbal medicine, also known as phytomedical or botanical medicine, refers to using a plant's seeds, berries, roots, leaves, bark or flower for medicinal purposes. The word phytomedicine is derived from the Greek word 'phyto', meaning plant. Herbal medicine uses plants to treat disease and enhance general health and wellbeing. Herbs can interact with pharmaceutical medications and should be taken with care.[2]

History of Traditional and Herbal Medicine

Traditional medicine has been used for thousands of years in Egypt, China, India, Greece and Rome. Ancient texts such as Ebers Papyrus, Shennong Herbal, Charaka Samhita and Sushruta Samhita documented medicinal plants. Many modern medicines are derived from natural products and their synthetic analogues. According to the WHO, traditional medicine includes knowledge and practices used for preventing and treating diseases. Natural products and plant-based medicines have been used for thousands of years, and until the early twentieth century most medicines came from plants and natural sources. In India, traditional medicine dates back about 5000 years BCE, with knowledge recorded in Rigveda, Yajurveda, Atharva Veda, Charaka Samhita, Sushruta Samhita and Dhanvantari Nighantu.[3]

Indian System of Medicine

India has the recognized traditional systems of Ayurveda, Siddha, Unani, Yoga and Naturopathy, and Homeopathy.

Ayurveda
Ayurveda, the science of life, considers physical, psychological, philosophical, ethical and spiritual wellbeing. The Ayurvedic concept emerged in India between 2500 and 500 BC. Nadi, Mootra, Mala, Jihva, Shabda, Sparsha, Druk and Aakruti are described as eight methods used for diagnosis. Ayurvedic medicines are available as tablets, fermented goods, liquids, powders, pastes and medicinal butters.[2]

Siddha
The Siddha medical system was established in India between 10,000 and 4000 BCE and remains an old medical practice of South India developed through practical knowledge of natural resources. The system describes the human body in terms of three humours, seven fundamental materials and waste products; equilibrium of humour is associated with healthy wellbeing, whereas imbalance results in illness.[3]

Unani
Unani is one of the oldest traditional medical systems and has been used for centuries to prevent and treat a wide range of illnesses. It is also known as Unani Tibb or Graeco-Arab medicine. Its principles include akhlat, aaza, mizaj, arkan, afaal, quwa and arwah, which work together to maintain balance.[4]

Yoga and Naturopathy
Yoga is a Sanskrit word generally understood as union and explores preventive and curative aptitudes as a training exercise for mindfulness. Naturopathy balances modern research and rational advancements with traditional healing methods, emphasizing the body's innate ability to heal, prevention of illness and individual accountability for wellbeing.[3]

Homeopathy
Homeopathy as practiced today evolved through the work of Dr. Samuel Hahnemann (1755–1843). The word is derived from the Greek words 'Homois' meaning similar and 'pathos' meaning suffering. In India, homeopathy has been practiced for at least 150 years and is recognized as one of the medical systems.[1]

Topical drug delivery system
Topical drug delivery is the application of a pharmaceutical dosage form to the skin to treat a cutaneous condition or a cutaneous manifestation of a general disease, with the intention of limiting pharmacological or other activity to the skin's surface. The skin is an accessible route for drug delivery. Solid powders, semisolids, liquids and sprays are used, while gels, creams and ointments are popular semisolid preparations.[5]

Advantages
Advantages stated in the dissertation include enhanced stability, less sonication, avoidance of first-pass metabolism, the possibility of self-medication and enhanced capacity for loading.[5]

Disadvantages
Disadvantages include excessive skin irritability, possibility of allergic reaction, poor permeability of certain medications through the skin, difficulty in absorption of drugs with large particles, contact dermatitis and formation of a bubble during the emulgel process.[5]

2. SKIN

The skin is the largest organ of the human body and forms a barrier between the external world and the internal environment. It comprises about 15% of total body weight and its thickness ranges from 0.1 mm at the thinnest part to 1.5 mm at the thickest part. It performs functions including regulation of body temperature, sensory perception, prevention of water loss, and protection against infection and mechanical harm. The three primary layers are epidermis, dermis and hypodermis.[6]

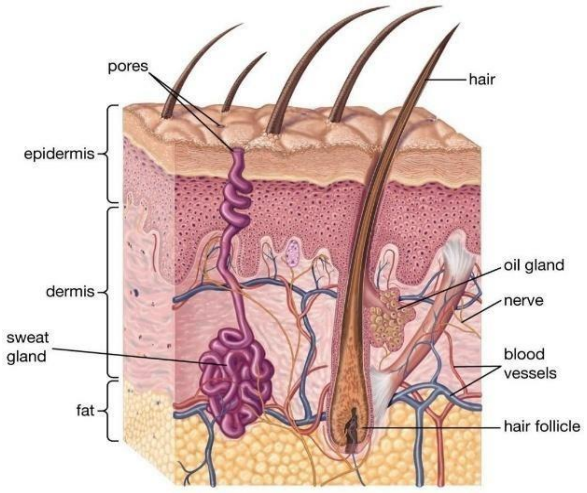

Figure No.1: Structure of skin.

Epidermis
The epidermis is the outermost layer of skin and is composed primarily of stratified squamous epithelial cells. It is non-vascular and serves as a major barrier. The stratum corneum contains dead keratinized cells, while living layers include the stratum basale where new cells are produced. Melanocytes generate melanin and Langerhans cells participate in immune defence.[6]

Dermis
The dermis lies beneath the epidermis and is a thick connective tissue layer providing elasticity and structural strength. It contains collagen and elastin fibres, lymphatic, blood and nerve vessels, sweat glands, sebaceous glands, erector pili muscles and sensory receptors for pressure, pain and temperature.[6]

Hypodermis
The hypodermis, or subcutaneous layer, lies beneath the dermis and consists mainly of loose and adipose

connective tissue. It acts as an energy store, thermal insulator and shock absorber, and helps anchor skin to underlying bones and muscles while allowing passage of larger nerves and blood vessels.[6]

Sebaceous glands
Sebaceous glands secrete sebum, which lubricates and protects the skin and hair. They are mainly found on the face, scalp, shoulders and upper chest, but not on the palms and soles. The dissertation states a density of 500–1000 glands per square centimetre.[6]

Sweat Glands
Sweat glands are coiled tubular glands present throughout the skin. They secrete sweat containing water, salts and small amounts of waste products. Their main function is regulation of body temperature through sweat production and evaporation, and they also help excrete salts and waste substances.[6]

Function of Skin
Protection is provided by the skin barrier and Langerhans cells against external damage and microbes. Sensation is mediated by nerve endings responding to heat, tissue damage, vibration, pressure, cold and touch. Thermoregulation involves skin blood supply, radiation-induced energy loss, conduction and convection; vascular dilation enhances heat loss, while restricted vessels help maintain body heat.[7]

Protection
Langerhans cells comprise of adaptive immune system and acts as barrier between the external and internal environments, safeguarding the body from damage and a variety of microbes.

Sensation
Haptics and the somatosensory system describe a collection of nerve endings that react to heat and tissue damage, vibration, pressure, cold, and touch.

Thermoregulation
The blood supply to the skin is much greater than it needs, management of radiation-induced energy loss, conduction as well as convection. Vascular dilation enhances heat loss and perfusion, while restricted blood vessels reduce the blood supply to the skin and aid to maintain body heat.

3. DISEASE PROFILE OF VITILIGO

Vitiligo is a skin condition in which pigmentation gradually decreases, producing white patches. The dissertation states that it affects about 0.5% of the global population, without preference for skin tone or gender, and often begins before 20 years of age. In classical Ayurvedic texts it was referred to as ‘Sweta Kustha’ or ‘white leprosy’. Vitiligo occurs due to destruction of melanocytes, resulting in depigmented or hypopigmented patches. The patches may also affect mucous membranes, retina and hair. Typical lesions include macules of less than 1 cm or patches greater than 1 cm surrounded by normal skin, and vitiligo is characterized as a multifactorial autoimmune disorder with loss of functioning melanocytes.[8]

Figure No. 2: Vitiligo.

Types Of Vitiligo[9]

Table No. 1: Types of vitiligo.

TYPES OF VITILIGO	SUBTYPES
Segmental	Bisegmental / Unisegmental / Pluriseg mental
Non segmental	Generalised / acrofacial / universal / mucosal
Unclassified /undetermined	Focal / mucosal (one site)

Vitiligo Symptoms
The primary symptom is skin depigmentation or loss of colour. Affected skin may become lighter than surrounding skin or turn white. Depigmentation commonly occurs on the face, hands and feet but may occur anywhere, including hair, genitals and inside the nose or mouth. Symptoms usually appear before age 20.[10]

4. MECHANISM OF ACTION

Psoralen administration (topical/oral) is followed by absorption into cells, intercalation between DNA base pairs, mainly pyrimidine bases such as thymine, and exposure to UVA light at 320–400 nm. Activated psoralen forms monoadducts with thymine followed by DNA cross-links, inhibiting DNA replication and transcription. This is followed by cell-cycle arrest at G1/S phase, induction of apoptosis and decreased proliferation of abnormal cells.[11]

SEMI-SOLID DOSAGE FORMS

Semi-solid dosage forms have a consistency intermediate between solid and liquid states and are designed for external application to the skin, mucous membranes or body cavities. Active ingredients are dispersed or dissolved in a suitable base. They provide prolonged

contact with the affected area, protect the skin and enhance drug absorption. Examples include ointments, creams, gels and pastes.[12]

5. CREAMS

Creams are viscous liquid or semi-solid emulsions of either oil-in-water or water-in-oil type, with consistency varying according to the amounts of oil and water. They are applied to the skin and consist of continuous and dispersed phases.[7]

Classification Of Cream

- Skin creams can be categorized based on function Ex: cleaning, foundation, massage.
- Based on the characteristic properties Ex: cold creams, vanishing creams.
- Based on their nature or type of emulsion:

Types Of Skin Cream

- oil-in-water (O/W) creams
- water-in-OIL (W/O) CREAMS

6. PLANT PROFILE PSORALEA CORYLIFOLIA

Introduction

The word Psoralea originates from the Greek term psoraleos, meaning 'affected with itch or leprosy'. Species of Psoralea are native to America and are widely distributed across India, particularly Uttar Pradesh, Bengal and Maharashtra. Psoralea corylifolia is an annual erect herb growing between 30–180 cm, thriving in warm sunny locations and tolerating clay, sand or loam soils under acidic, basic or neutral conditions. The best sowing season is March–April and seeds mature in November.[26]

SEEDS

Seeds are brownish black, oblong and flattened; kidney shaped, 2–4 mm long, 23 mm broad and 1–1.5 mm thick; hard, smooth, exalbuminous, with straw-coloured testa, agreeable aromatic odour and pungent bitter taste. The plant has been used for skin disorders including psoriasis, leukoderma and leprosy.[27]

Synonyms

Cullen corylifolium; Cullen corylifolia; Psoralea patersoniae; Trifolium unifolium; Purple Fleabane.

Scientific Classification

Kingdom: Plantae; Division: Angiospermae; Class: Dicotyledonae; Order: Rosales; Sub-order: Rutinae; Family: Leguminosae; Sub-family: Papilionaceae; Genus: Psoralea; Species: corylifolia Linn.[27]

Vernacular Names

Hindi: Babachi, Babchi, Bakuci; Gujarati: Babichi, Babchi, Bawchi; Tamil: Karpokarishi, karpuvanshi; Marathi: Babachi, Bavachi, Bavanchi; Sanskrit: Bakuchi, Vakuchi; Punjabi: Babchi; Urdu: Babechi; English: Psoralea seeds, Fountain bush; Telugu: Bavanchalu, Bavanchi-vittulu; Kannada: Somaraji, Karbekhiga.[27]

Organoleptic Characters

- Colour: dark brownish black;
- Odour: oily;
- Taste: characteristic;
- Touch: rough.[28]

Description

The genus Psoralea belongs to Fabaceae and comprises approximately 130 species, mainly found in South Africa, North and South America and Australia, with some species native to Asia and temperate Europe. The plant typically grows 25–170 cm in warm climates; sandy soil is suitable, sowing is March–April, maturation occurs in November, and the plant may survive 6–7 years. It produces perennial fruits and small reddish flowers.[29,30]

Phytoconstituents

Bakuchiol (meroterpene; seeds/fruit; anti-acne, anti-diabetic); Bavachinin (flavone; seeds; antibacterial); Bakuisoflavone (flavone; fruit; antibacterial); Bakuchicin (coumarin; seeds; topoisomerase inhibitor); Bavachinone A (flavonoid; fruit; antibacterial); Bavachinone B (flavonoid; fruit; antibacterial); Corylifolin (chalcone; seeds; carboxylesterase inhibitor); Isopsoralen (furanocoumarin; dried ripe fruits/seeds; antiprotozoal); Psoralen (furanocoumarin; dried ripe fruits/seeds; leukoderma, psoriasis); Hydroxy bakuchiol (meroterpene; seeds; lymphangiogenesis inhibition).[31]

Figure No. 3: ***Psoralea Corylifolia leaf.***

ECLIPTA ALBA

Introduction

Eclipta alba, synonym Eclipta prostrata, is a small perennial herbaceous plant grown in tropical areas worldwide and distributed in China, India, Brazil, Thailand, Sri Lanka, Malaysia and Nepal in moist soil, watercourses and hilly areas. It is commonly known as bhringraj or false daisy. Three varieties described are white flowering, black fruiting and yellow flowering. The plant occurs around rivers, lakes and mountain foothills in India.[32]

Synonyms

Verbesina prostrata L.; Eclipta undulata Willd.; Eclipta patula Schrad.; Micrelium tolak Forssk.[33]

Scientific Classification

- Subkingdom: Viridaeplantae
- Infrakingdom: Streptophyta
- Division: Tracheophyta
- Subdivision: Spermatophytina
- Infradivision: Angiospermae
- Class: Magnoliopsida
- Superorder: Asteranae
- Order: Asterales
- Family: Asteraceae
- Genus: Eclipta
- Species: alba.[34]

Figure No.4: Eclipta alba.

Phytochemicals Present In Eclipta Alba

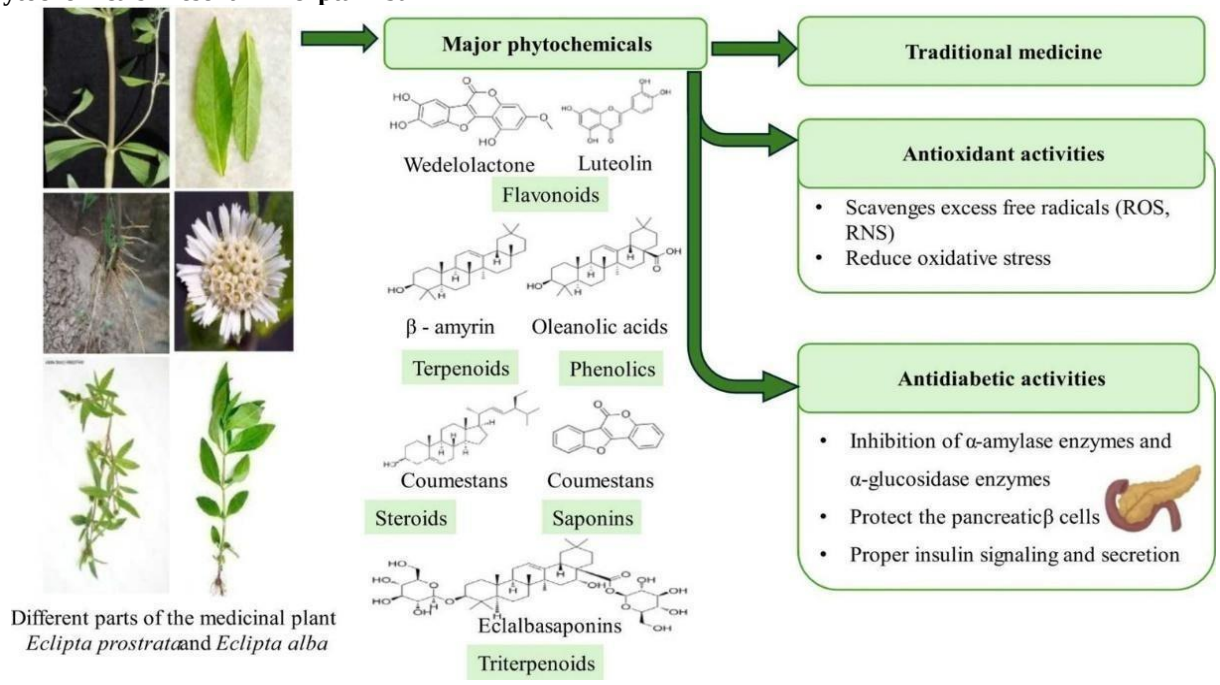

Figure No. 5: Phytochemicals in *Eclipta alba.*

TURMERIC

Introduction

Turmeric is a fragrant medicinal plant used since ancient times. Ayurveda, Siddha, Unani medicine and traditional Chinese medicine have made extensive use of Curcuma longa. About 40–45 Curcuma species are found in India. Chemical components described include polyphenols, sesquiterpenes, diterpenes, triterpenoids, sterols and alkaloids. Turmeric is grown in India, China, Taiwan, Indonesia, Sri Lanka, Africa and Australia.[37]

Synonyms

Indian Saffron, Turmeric, Haldi, Aridra.

Figure No. 6: Turmeric.

Scientific Classification[38]

- **Kingdom** : Plantae
- **Subkingdom:** Tracheobionta
- **Superdivision** : Spermatophyta
- **Division** : Magnoliophyta
- **Sub-class** : Zingiberidae
- **Order** : Zingiberales
- **Family** : Zingiberaceae
- **Genus** : Curcuma
- **Species** : longa

7. MATERIALS AND METHODS

Extraction Of Plant Extracts

Psoralea corylifolia: The required quantity of finely powdered Psoralea corylifolia seeds was suspended in 95% alcohol at a ratio of 1:10 w/v. The mixture was kept in a closed container at room temperature for 24 hours with occasional shaking. The extract was filtered using Whatman filter paper and the liquid ethanolic extract was collected.[46]

Figure No. 7: Extraction of Psoralea corylifolia.

Eclipta Alba: Leaves were collected from in and around Perambalur, authenticated by a Botanist, Department of Botany, St. Joseph's College, Trichy, cleaned and shade dried at room temperature. The material was dried in an oven at 40°C for 5 days and ground in an electric blender. The powder was suspended in 95% alcohol and left at room temperature for 24 h. The crude extract was filtered using Whatman filter paper under sterile condition and the alcoholic extract was collected.[47]

Figure No. 8: Extraction of Eclipta alba.

Ingredients And Formulation

Table No. 2: Formulation of cream.

S.No	Ingredient	F1	F2	F3
1	Psoralea corylifolia extract	3 ml	1.5 ml	1.5 ml
2	Eclipta alba extract	2.5 ml	1.4 ml	1 ml
3	Turmeric powder	1.5 g	0.28 g	0.28 g
4	Beeswax	6.78 g	4.97 g	4.97 g
5	Liquid paraffin	16 ml	22 ml	22 ml
6	Borax	0.20 g	0.056 g	0.056 g
7	Sodium benzoate	0.02 g	0.02 g	0.02 g
8	Rose oil	q.s	q.s	q.s
9	Distilled water	q.s	q.s	q.s

Formulation of Cream

The cream is a water-in-oil type of emulsion. Liquid paraffin and beeswax were heated in a beaker at 75°C while maintaining the temperature. Borax, sodium benzoate and psoralen were dissolved in another beaker and heated at 75°C to dissolve. The aqueous phase was slowly added to the heated oil phase with continuous stirring. Psoralea corylifolia extract, Eclipta alba extract

and rose oil were added individually drop by drop with vigorous stirring to obtain a uniform, smooth texture. The cream was kept at 4°C until use.[8]

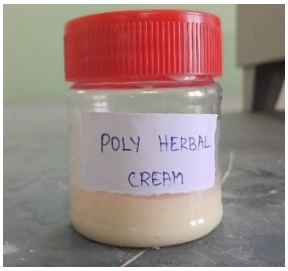

Figure No. 9: Polyherbal Cream.

PHYTOCHEMICAL SCREENING

Phytochemical screening was carried out on the powdered form of Psoralea corylifolia.[48]

Table no. 3: Phytochemical screening of *Psoralea corylifolia Linn*. Note: ++ = strongly present, + = present.

Test	Result
Psoralen	++
Phenol	+
Alkaloids	+
Anthraquinone glycosides	+
Cardiac glycosides	+
Coumarins	+

8. EVALUATION OF CREAM[8]

1. **Physical evaluation:** cream was observed for colour, texture, odour, etc.
2. **Irritancy:** mark 1cm2 area on the left-hand dorsal surface. Cream is applied to that area and note that time. After interval up to 24 hours it is checked for irritancy, erythema and edema if any reported
3. **Wash ability:** apply a small amount of cream on the hand and wash with the tap water.
4. **Ph:** 0.5 g cream was taken and dispersed in 50 ml distilled water and ph was measured by digital ph meter.
5. **Phase separation:** cream is kept in close container away from light at 25-100°c for one month and observed for phase separation.
6. **Spreadability:** spreadability is carried out for all formulations. The less time taken for separation of both slides better the spreadability**.**
7. **Greasiness:** the cream is applied in the form of smear on the surface of skin and observed if smear is oily or grease like.

EVALUATION OF CREAM[8]

1. PHYSICAL EVALUATION

Table No. 4: Physical Evaluation.

SR.NO	PARAMETERS	OBSERVATION
1	Colour	Pale yellow colour
2	Odour	Pleasant
3	Texture	Smooth
4	State	Semi-solid

Irritancy

Mark the area (1 cm^2) on the left-hand dorsal surface. After that, the cream was administered there, and the duration was recorded. Then it is checked for irritancy, erythema, and oedema if any for an interval up to 24 h and reported. According to the results, the formulation is showing no sign of irritancy, erythema, and oedema.

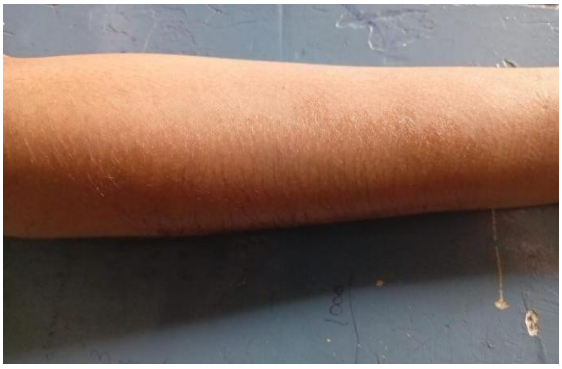

Figure No. 10: Irritancy.

Washability

Applying a small quantity of cream to the hand and then washing it with tap water served as the washability test.

Phase Seperation

The prepared cream was kept in a closed container at a temperature of 25-100 C away from light. The phase separation was then monitored for 24 hr 30 days. Any change in the phase separation was observed. According to the results, no phase separation was observed in the formulation.

Spreadability

The spreadability of the formulation was carried out and the time taken by the 2 slides to separate is less so as said in the description of the evaluation test lesser the time taken for separation of the two slides better the spreadability so according to this state better spreadability.

Figure No. 11: Spreadability.

Greasiness

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

three formulations were non-greasy.

pH

According to the results, the pH of the formulation was found to be 5.75 so it can be safely used on the skin.

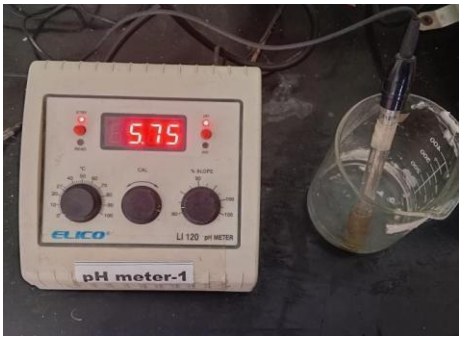

Figure No. 12: pH meter.

9. IN-VITRO ANTIOXIDANT ACTIVITY

The methodology described by Ruch et al. was slightly modified to evaluate hydrogen peroxide scavenging activity. A 43 mM hydrogen peroxide solution was prepared using phosphate buffer (1 M, pH 7.4). Various concentrations of the polyherbal formulation ranging from 10 to 500 µm/ml were mixed with 0.6 ml of hydrogen peroxide solution. After 10 min incubation, absorbance of hydrogen peroxide was measured at 276 nm. Phosphate buffer without hydrogen peroxide was used as blank and ascorbic acid was employed as the standard.[49]

% inhibition = [(control test) / control] × 100. Control absorbance = 1.189; sample absorbance = 0.222. Therefore, % inhibition = [(1.189 0.222) / 1.189] × 100 = 81.33%.

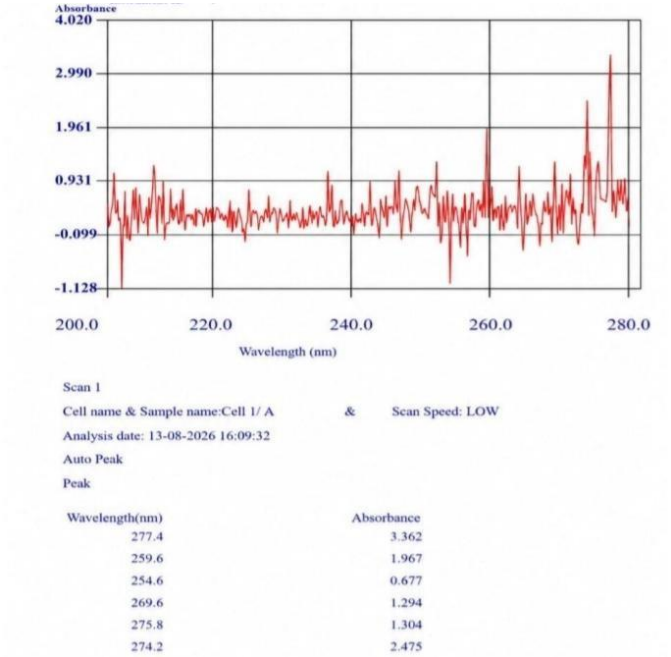

Figure no. 13: UV-Vis spectrophotometer spectrum scan from the supplied dissertation.

RESULT

The sample exhibited 81.33% inhibition according to the Ruch et al. method. The supplied spectrophotometer record shows the spectrum scan and peak readings reported in the dissertation.

10. DISCUSSION

The study formulated three polyherbal cream formulations containing Psoralea corylifolia extract, Eclipta alba extract and turmeric powder with beeswax, liquid paraffin, borax, sodium benzoate, rose oil and distilled water. The plant profiles and phytochemical information in the dissertation support the selection of the three herbal ingredients. The prepared cream was pale yellow, pleasant, smooth and semi-solid. It showed no irritancy, erythema or oedema, no phase separation, and the three formulations were reported as non-greasy. The pH was 5.75. The hydrogen peroxide scavenging study gave 81.33% inhibition, indicating antioxidant activity in the tested sample. These findings are limited to the evaluation parameters and results reported in the supplied dissertation.

11. ACKNOWLEDGEMENT

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