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Development and evaluation of *Cannabis sativa* infused herbal Ointment for Psoriasis management

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Abstract: *Cannabis sativa L., a historically valued medicinal plant, has gained modern scientific interest due to its diverse pharmacological properties, particularly in managing inflammatory conditions. Psoriasis is a long-lasting autoimmune condition characterized by excessive growth of keratinocytes, chronic inflammation, and disturbances in immune function, often leading to a reduced quality of life for affected individuals. While traditional therapies such as corticosteroids and systemic immunosuppressive agents are commonly used, their effectiveness varies, and they may cause unwanted side effects. This has led to increased interest in identifying and developing alternative treatment approaches. This study aimed to develop and evaluate a herbal ointment incorporating ethanolic extracts of Cannabis sativa L. leaves for topical use in psoriasis management. Plant material was authenticated and extracted using Soxhlet apparatus with ethanol as the solvent. Three trial formulations were prepared using varying combinations of oils, waxes, and bioactive agents, followed by a finalized formulation incorporating Cannabis extract, turmeric oil, and cetyl alcohol, among others. Phytochemical analysis of the extract indicated the presence of flavonoid compounds. Terpenoids, alkaloids, tannins, and phenols, confirming its bioactive potential. The formulated ointment underwent comprehensive physicochemical evaluations including pH, homogeneity, spreadability, washability, irritancy, viscosity, and accelerated stability testing. Results demonstrated favorable topical properties with no signs of skin irritation and good stability over time. The therapeutic potential of cannabinoids, especially CBD, was emphasized due to their anti-inflammatory and antioxidant actions mediated via endocannabinoid pathways. The findings support the potential of Cannabis sativa-based topical formulations as a promising alternative for psoriasis treatment. Further clinical investigation is warranted to establish efficacy, safety, and patient tolerability.*

Keywords- *Cannabis sativa, Psoriasis, Herbal ointment, Phytochemical screening, Anti-inflammatory.*

INTRODUCTION

Psoriasis is a persistent, immune-mediated skin condition that significantly affects a person's quality of life [1]. It results from a complex interaction of genetic susceptibility, immune system dysregulation, and environmental triggers [2]. Characterized by itchy, inflamed, and scaly skin lesions, this autoimmune condition varies in severity and presentation. Though various treatment options exist including topical agents, phototherapy, and systemic drugs their success is inconsistent, and many may lead to adverse effects [1,2].
Genetic research has identified associations between psoriasis and specific markers such as the HLA-Cw6 allele and genes responsible for immune responses and skin barrier function [3]. External factors like stress, infections, and certain medications have been known to trigger or worsen the disease [4]. Psoriasis involves abnormal activation of immune cells, which speeds up skin cell turnover, leading to thickened, red, and scaly patches [5].Globally, psoriasis affects around 1% to 3% of the population, classifying it as a common chronic inflammatory disorder [1]. Although its exact cause remains unknown, the condition is widely considered a result of immune dysfunction influenced by genetics and lifestyle factors, including diet, stress, and comorbid illnesses.Currently, no definitive cure is available for psoriasis. It often coexists with systemic conditions like psoriatic arthritis, inflammatory bowel disease (IBD), obesity, and insulin resistance, all of which can further

reduce a patient's quality of life [1,6]. Emerging studies emphasize the importance of understanding metabolic comorbidities associated with psoriasis, as these insights can guide the development of more effective therapies [6].A particularly significant association is the two-way relationship between psoriasis and obesity. Obesity increases the risk of developing psoriasis, and in turn, having psoriasis may increase the likelihood of becoming obese due to chronic inflammation and reduced physical activity [7].

1.1 Pathophysiology

The exact underlying processes responsible for the onset of psoriatic disease skin remain complex and not yet fully elucidated. Researchers have adopted multiple strategies to investigate the pathophysiology of psoriasis. Among the most significant findings is the critical role of the adaptive immune system in the onset and progression of the disease [8]. A key feature of this is T-cell-mediated immune activation, where abnormal signaling pathways within immune cells contribute to chronic inflammation and skin changes. Several scientific studies examining psoriasis pathogenesis have consistently highlighted the involvement of this dysregulated T-cell response as a central factor [9].

1.2 Cannabinoids

The medicinal properties of *Cannabis sativa L.* are primarily linked to the presence of *cannabinoids (CNBs)* , a broad group of bioactive compounds that exert their effects by interacting with cannabinoid receptors (CBRs) in the body [10]. These compounds are structurally and biochemically related, particularly to *tetrahydrocannabinol (THC),* which is the principal psychoactive constituent of the plant. *Cannabis sativa L.* is known to produce an estimated 565 secondary metabolites [11], out of which approximately 120 have been identified as *cannabinoids* [12]. From a chemical standpoint, cannabinoids are part of the terpenophenolic compound class, with *THC* and *cannabidiol (CBD)* being the most widely recognized. Other important cannabinoids with notable pharmacological activity include *cannabinoid* acids, *cannabigerol (CBG),* and *cannabidivarin (CBDV)* [13].

Cannabis-based medicines (CBMs) have been explored for their effectiveness in managing a variety of medical conditions, particularly in the treatment of chronic pain and epileptic seizures [14]. Although the plant comprises over 400 bioactive compounds, recent scientific interest has primarily centered on two major constituents: *delta-9-tetrahydrocannabinol (THC)* and *cannabidiol (CBD)* [15,16]. THC is responsible for the psychoactive effects commonly associated with *cannabis* use, while *CBD* does not produce a high and is instead recognized for its anti-inflammatory and antioxidant properties [17].

In recent years, *cannabis* has attracted attention for its potential role in dermatology, especially in treating conditions such as pruritus (itching) and inflammatory skin disorders. Both *THC* and *CBD* Influence the endocannabinoid system, responsible for managing immune responses. Through this interaction, they may help reduce inflammation and immune-related skin flare-ups. Research suggests that topical *cannabis* applications might be beneficial in alleviating symptoms like itching and localized inflammation [18].

Historically, the therapeutic Cannabis has been utilized since Emperor Chen Nung, a figure often regarded as the Father of Chinese agriculture, who compiled one of the earliest Chinese pharmacopoeias. In this ancient text, *cannabis* was suggested for ailments such as fatigue, rheumatism, and malaria, while its seeds, rich in gamma-linoleic acid (GLA), were recommended for treating eczema, psoriasis, and inflammatory conditions [19,20,21]. Despite this long history of use, *cannabis* was banned in the 20th century, mainly due to concerns about its psychoactive effects.

However, by the end of the 20th century, *cannabis* began to re-emerge in scientific and medical discussions, becoming one of the most rapidly expanding agricultural products. In Western discourse, the terms Industrial-use hemp and therapeutic-use marijuana were introduced to differentiate between varieties of the plant based on their *THC* content and intended application. Hemp is defined as *cannabis* with 0.3% or less of *THC,* making it non-intoxicating and legal in many regions, whereas marijuana may contain up to 30% *THC* and is considered a controlled substance [22].

Because of hemp's minimal psychoactive content, the pharmaceutical industry has increasingly focused on it as a key source of *CBD*, which offers therapeutic effects without intoxication. In the realm of skincare, *CBD* oil, extracted mainly from the leaves, is notable for its non-psychoactive therapeutic potential. Additionally, hemp seed oil, derived from the seeds, is commonly used for its nutritional and emollient properties, though it contains little to no *cannabinoids* [23].

MATERIAL AND METHOD

2.1 Plant Material

Cannabis sativa L. leaves were collected from the local region of Peepal Chowk, located near D.B.U.U. The plant material was authenticated following collection to ensure proper identification. The harvested leaves were then air-dried under controlled conditions to reduce moisture content prior to extraction procedures.

2.2 Solvent

Ethanol was selected as the extraction solvent due to its high efficiency in extracting a broad range of bioactive compounds and its suitability for research applications. Its potency and effectiveness make it an ideal choice for obtaining phytoconstituents from *Cannabis sativa L.* leaves for further analysis.

2.3 Extraction Procedure

The extraction of *Cannabis sativa* leaves was carried out using a Soxhlet apparatus. A total of 25 g of coarsely powdered, An ethanol volume of 250 mL was used to extract the dried leaves. The Soxhlet assembly was run continuously for approximately 6 hours to ensure complete extraction. After completion, the extracted solvent was concentrated by placing it in a water bath until a semi-solid mass was obtained.The resulting extract was preserved at 4°C for subsequent analysis.

Formulation of Ointment (Procedure)

Table no. 1

Prepare the Oil Phase	Prepare the Water Phase	Combine Oil and Water Phases	Cooling the Mixture
Microcrystalline wax Beeswax Castor oil Turmeric oil Lanolin Camphor oil	In a separate beaker, heat the Purified Water (45.9%) to approximately 40–45°C and Add Emulsifying Wax	Slowly add the water phase (heated water and emulsifying wax) to the oil phase (heated waxes and oils) while stirring continuously. This should be done carefully to ensure proper emulsification.	Once the two phases are fully combined and emulsified, allow the ointment to cool down gradually while stirring occasionally to prevent air bubbles from forming and to maintain a smooth texture.

2.4 Preparation of Herbal ointment

Formulation A: (Trial)

S. No.	Ingredients	F1(10gm)	F2(15gm)	F3(20gm)	F4(25gm)	F5(30gm)
1	Microcrystalline wax	1gm	1.5	1gm	2.5	3
2	Beeswax	0.2gm	0.2	0.4gm	0.5	0.6
3	Castor oil	7.1gm	9.15	14gm	15.25	18.3
4	Lanolin	1gm	1.2	1.6gm	2	2.4

5	Cetyl alcohol	0.5gm	1.5	0.2gm	2.5	3
6	Emulsifying wax	0.5gm	0.75	1gm	1.25	1.5
7	Turmeric oil	0.2gm	0.3	0.4gm	0.5	0.6
8	*Cannabis extract*	0.5gm	0.75	1gm	1.25	3

Final Formulation

An herbal ointment was formulated with a total batch size of 50 g using a combination of waxes, oils, and active extracts. The formulation consisted of microcrystalline wax (2.5 g), beeswax (1 g), castor oil (30.5 g), lanolin (5 g), emulsifying wax (2.5 g), cetyl alcohol (5 g), turmeric oil (1 g), and Cannabis sativa L. extract (5 g). These components were carefully selected to provide a stable base with moisturizing, emollient, and therapeutic properties.

S. No.	Ingredients	Batch size (50gm)
1	Microcrystalline wax	2.5gm
2	Beeswax	1gm
3	Castor oil	30.5gm
4	Lanolin	5gm
5	Emulsifying wax	2.5gm
6	Cetyl alcohol	5gm
7	Turmeric oil	1gm
8	*Cannabis extract*	5gm

The oily phase was prepared by melting microcrystalline wax, beeswax, castor oil, lanolin, emulsifying wax, and cetyl alcohol together over a hot plate with constant stirring. Once a uniform molten mixture was obtained, the active components turmeric oil and *cannabis* extract were incorporated into the mixture at a suitable temperature to preserve their bioactivity.

4. Evaluation test

4.1 Determination of pH of the ointment:

4.2 Homogeneity: A small quantity of the cream formulation was evaluated for homogeneity through visual inspection and by assessing its texture via touch and gentle rubbing between the fingers.

4.3 Appearance: The cream formulations were evaluated based on their pearlescence, color, texture, and consistency.

4.4 Spreadability test: To ensure consistent thickness, an excess sample was positioned between two glass slides, and a 100 g weight was applied to the top slide for one minute. Subsequently, a 100 g weight was placed on the pan, and the time taken (in seconds) for the two glass slides to move apart was recorded as a measure of the formulation's spreadability. The spreadability was calculated using the formula:

S=M×LTS = \frac{M \times L}{T}S=TM×L

Where:

M (Weight applied to the upper slide) = 100 g

L (Distance between the slides) = 5 cm
T (Time required for the slides to separate) = 60 seconds
Based on the above values, the spreadability was determined to be 8.3 g·cm/s.
4.5 Washability test: To evaluate washability, a small quantity of the base was applied to the hand and gently rubbed to observe how easily it could be removed.
4.6 Irritancy Test: A small amount of ointment was applied to remain on the skin surface for approximately 10 minutes. After this period, the properties of the ointment on the skin were evaluated.
4.7 Stability testing: Accelerated stability testing (AST) for the ointment was conducted for 6 days at room temperature. The most stable ointment was then tested at 40°C ± 1°C for 20 days. Evaluations were done on days 0, 5, 10, 15, and 20 at both room and elevated temperatures.
4.8 Viscosity test: The Brookfield viscometer was used to measure the viscosity of the formulated ointment. An appropriate spindle Spindle No. 4 was chosen based on the consistency of the formulation. Once the correct spindle was attached, the instrument was adjusted to the required operating conditions. The measurement was conducted at a rotational speed of 6 RPM, ensuring that the torque remained constant throughout the process. The viscosity reading was taken directly from the dial, and the average of multiple readings was calculated to obtain a reliable result.
The viscosity was calculated afterward by applying the relevant formula:
Viscosity = Dial Reading × Factor
For Spindle IV-4 at 6 RPM, this factor is 1M, equivalent to 1000.

5. Selection of Bacteria:

The bacterium applied in this research was *Staphylococcus aureus*, a gram-positive microorganism belonging to the family Staphylococcaceae. *Staphylococcus aureus* is commonly associated with skin infections and is recognized for its involvement in initiating or worsening inflammatory skin disorders like psoriasis. It grows well on solid and liquid nutrient media and grows under aerobic conditions while also able to survive in low-oxygen environments. *S. aureus* is a resilient and adaptable bacterium, making it suitable for antibacterial screening and therapeutic evaluation. Standard microbiological techniques, including aseptic handling, were used to culture and maintain *S. aureus* for the experimental studies.

6. Qualitative Phytochemical analysis:

Table 2

S. NO.	Phytochemicals (Ethanolic Extract)	Method	Result
1	Alkaloids	Dragendroff's/ Kraut's test	+
2	Reducing sugar	Fehling's test & Benedict's test	+
3	Terpenoids	Salkowski test	+
4	Tannin	Ferric Chloride Test	+
5	Flavonoids	Conc. H2SO4 test & Lead acetate test	++
6	Phenols	Iodine test	+
7	Glycosides	Keller-Kiliani Test	-

7. For Anti-microbial activity:
Allow the sterile Mueller Hinton Agar (MHA) medium to cool to approximately 55°C, then introduce 10 µl of the bacterial culture into the MHA flask and mix gently. Label the Petri dishes appropriately and pour 25 ml of the prepared medium into each dish using a sterile measuring cylinder. Once the medium has solidified, create wells at equal distances using a sterile borer. Carefully dispense the test samples, standard, and blank into their designated wells according to the labels. After complete diffusion of the samples, place the MHA plates in a bacteriological incubator set at 25°C and incubate for 48 hours to observe the zones of inhibition.
S. aureus:

Figure 1: Zone of inhibition observed against *Staphylococcus aureus* on MHA plates.

7.1 Zone of Inhibition:- Zone of inhibition is the surrounding clear area on the agar plate where the *Cannabis sativa* extract was applied, indicating antimicrobial activity. It represents the diameter of the area where bacterial growth has been inhibited. To measure the zone of inhibition, the agar plate should be held upside down over a dark background, and the clear zone diameter is measured using a ruler in millimeters.

RESULT AND DISCUSSION
Ethanol was used to extract compounds from Cannabis sativa for the evaluation of its antipsoriatic activity.
8.1 Phytochemical analysis :-

S. NO.	Phytochemicals test	Method	Result
1	Alkaloids	Dragendorff's/Kraut's test: The addition of Dragendorff's reagent (1–2 mL) to a few milliliters of cannabis extract results in the formation of an orange hue or a reddish-brown precipitate.	Present
2	Reducing sugar	Fehling's Test: An equal mixture of Fehling's solutions A and B was prepared, and a small amount of the extract was added. Boiling the mixture led to the appearance of a brick-red cuprous oxide precipitate, confirming carbohydrate presence. Benedict's Test: An equal volume (2 mL) of the plant extract and Benedict's reagent was mixed in a test tube and then heated in a water bath for nearly five minutes.	Present
3	Terpenoids	Salkowski test: To test for terpenoids, the extract is mixed with 2 mL of chloroform, and then 3 mL of concentrated sulfuric acid is gently added along the side of the test tube to form a separate layer. The	Present

		appearance of a reddish-brown coloration at the boundary between the two layers confirms a positive result.	
4	Tannin	Ferric Chloride Test: 1 mL of the filtrate was diluted with distilled water, and two drops of ferric chloride were then added. A transient greenish to black coloration indicates the presence of tannins. Lead Subacetate Test: A creamy, gelatinous precipitate formed upon mixing 1 mL of filtrate with three drops of lead subacetate solution confirms the presence of tannins.	Present
5	Flavonoids	Concentrated Sulfuric Acid Test: A mixture of 5 mL plant extract, 2 mL glacial acetic acid, one drop of 5% $FeCl_3$, and concentrated H_2SO_4 was prepared. The development of a brown ring at the interface indicates a positive test for glycosides. Lead Acetate Test:A test tube containing 2 mL of the plant extract was treated with a few drops of 10% lead acetate solution, gently shaken, and examined for the appearance of a precipitate.	Present
6	Phenols	Iodine Test: When a few drops of diluted iodine were added to 1 mL of the extract, a short-lived red hue developed.	Present
7	Glycosides	Keller-Kiliani Test: Two milliliters of the plant extract were treated with glacial acetic acid, one drop of 5% ferric chloride solution, and concentrated sulfuric acid. The formation of a reddish-brown ring at the junction of the layers, along with a bluish-green tint in the upper layer, indicates the presence of glycosides.	Absent

Table no. 3 Showing the results of Phytochemical test

Fig. Phytochemical testing

8.2 Zone of Inhibition:-

Table no. 4

Sr. No.	Name of Sample	Zone of Inhibition
		S. aureus
1	Blank (MeOH: DMSO)	NZI

2	Ref. Std. – Clobetasol Cream	NZI
3	Ref. Std. – Mupirocin Cream	27 mm
4	Herbal Ointment	11 mm

8.3 UPLC Interpretation Report - *Cannabis sativa* Extract and Formulation

Purpose:- To confirm the presence of *CBD* in *Cannabis sativa* extract and ointment formulation samples by comparing their Retention Time (RT) with *CBD* standard.

Chromatographic Summary

Sample Name	Sample Type	CBD Retention Time (min)	Response Area
Std_CBD_10 ppm_01	Standard	2.882	691184
Std_CBD_10 ppm_02	Standard	2.881	653869
Std_CBD_10 ppm_03	Standard	2.883	649584
Spl_Hemp_Extract_01	Extract	2.882	71674
Spl_Hemp_Extract_02	Extract	2.892	67135
Spl_Ointment_01	Formulation	2.886	76256
Spl_Ointment_02	Formulation	2.891	71337
Blank_01	Blank	_	_

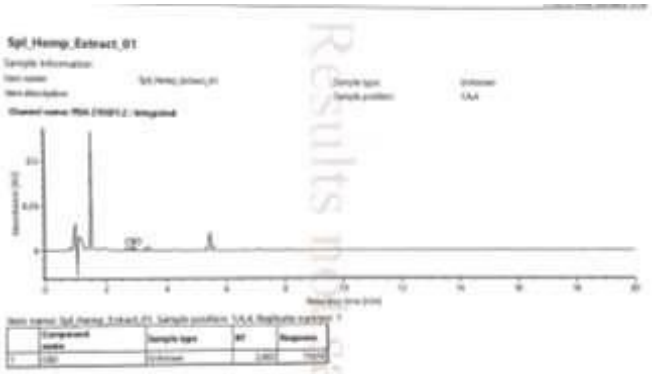
Observations

CBD standard samples showed consistent RT around 2.881–2.883 min.
All test samples (extract and ointment) displayed a distinct peak at similar RT, confirming presence of *CBD*.
Blank samples showed no peak at *CBD* RT, indicating no contamination or background interference.
Slight variation in RT (~±0.01 min) is acceptable and within instrument precision range.

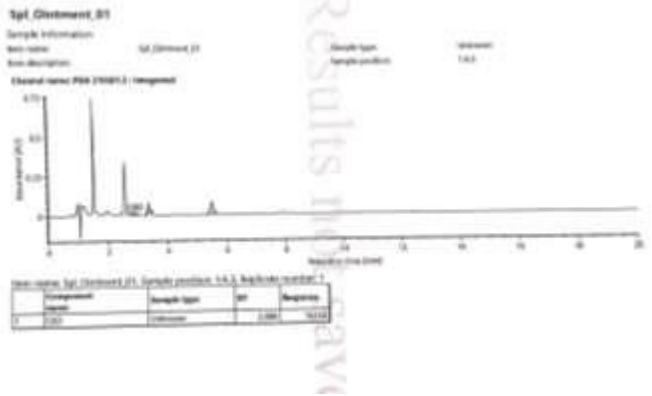
CONCLUSION

The UPLC analysis successfully identified *Cannabidiol (CBD)* in both the extract and formulation samples of *Cannabis sativa.*
The retention time in all unknowns closely matched that of the standard. No *CBD* was detected in the blank sample.
These results validate the presence of *CBD* in both raw extract and final ointment.

8.4 UPLC Chromatographic Analysis of *Cannabidiol (CBD)* in Extract of *Cannabis sativa*



8.5 UPLC Chromatographic Analysis of *Cannabidiol (CBD)* in Formulation of *Cannabis sativa*



CONCLUSION

The current study highlights the effectiveness of *Cannabis sativa L.* extract in developing a topical herbal ointment with promising antimicrobial activity. Phytochemical analysis of the ethanolic extract confirmed Enriched with bioactive molecules like flavonoids, such as terpenoids, tannins, phenols, and alkaloids, all of which contribute to anti-inflammatory and antibacterial effects. The formulated ointment showed a zone of inhibition measuring 11 mm against *Staphylococcus aureus*, a major bacterium implicated in skin infections and psoriasis. In comparison, standard Mupirocin cream showed a 27 mm inhibition zone, whereas Clobetasol cream and the blank base exhibited no inhibitory effect, indicating limited antibacterial action. Despite its relatively lower antimicrobial activity, the *Cannabis sativa*-based formulation offers advantages due to its herbal origin, reduced side effects, and therapeutic potential. These findings support the further investigation of this plant-based ointment as an alternative or complementary treatment for psoriasis and related inflammatory skin disorders.

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