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Development and Evaluation of Transdermal Patches Containing Ocimum Sanctum Linn.: Formulation, Physicochemical Properties, and Antimicrobial Activity Assessment

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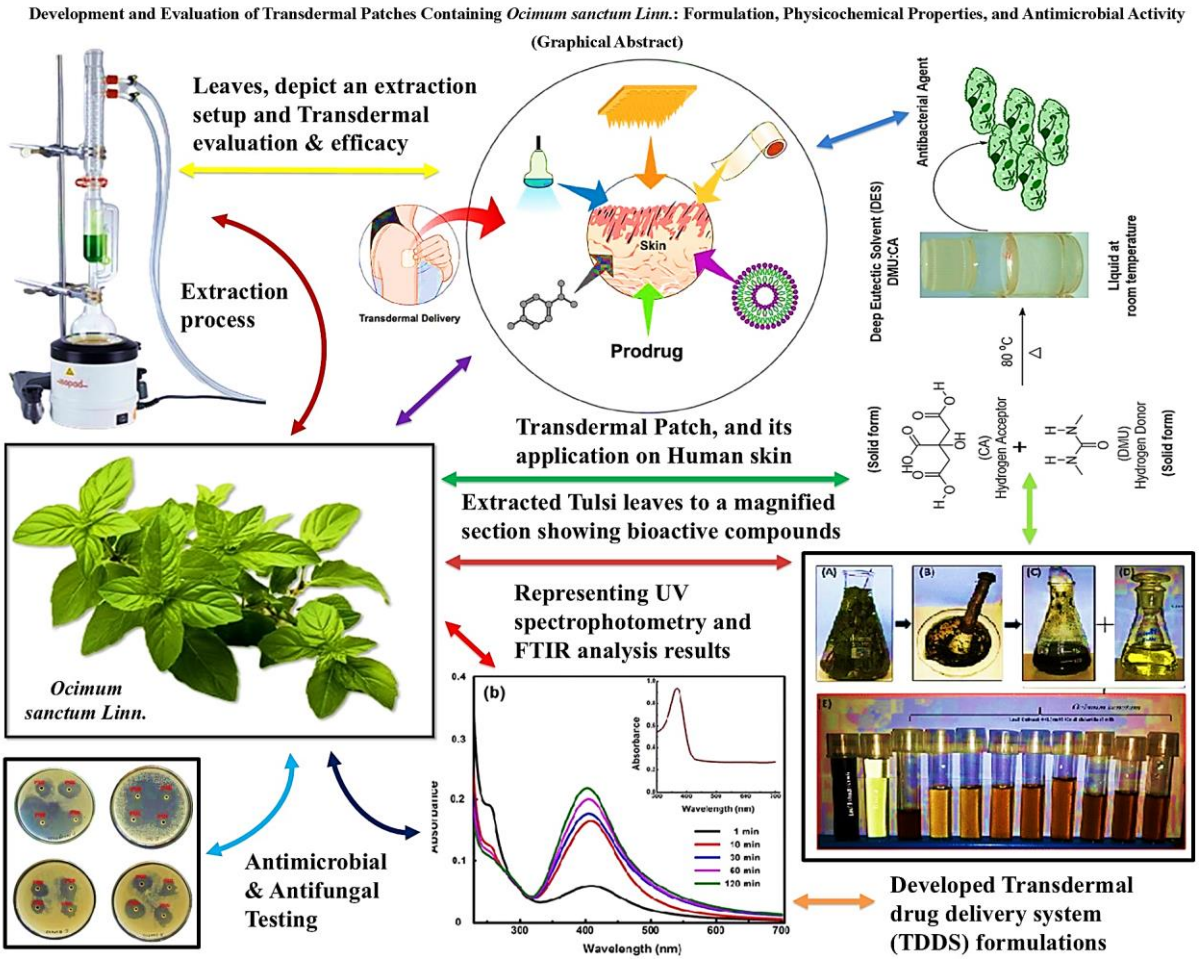
ABSTRACT:

Tulsi, often referred to as Tulasi, is a revered herb with both culinary and therapeutic attributes. It is a member of the Lamiaceae family and is indigenous to the Indian subcontinent. Ayurvedic medicine has relied on it for more than 3000 years as a crucial component. In the Ayurveda system, Tulsi is widely recognized as the "Elixir of Life" because of its therapeutic characteristics and its efficacy in treating a range of common health conditions. The objective of this research study is to develop and evaluate transdermal patches containing an aqueous extract of *Ocimum sanctum Linn.* leaves. The process involves extracting and purifying the leaves to obtain raw extracts, which are then used in the production of transdermal patches. The manuscript offers a thorough explanation of the chemicals and equipment used in patch manufacturing, followed by an examination of phytochemicals to determine their bioactive constituents. The formulation process entails the creation of the transdermal drug delivery system (TDDS) and the calibration of the extract using UV spectrophotometry. Physicochemical investigations are conducted to assess the quality and effectiveness of the patches. Furthermore, the research study includes stability testing in challenging environments and antibiotic susceptibility testing using the disc diffusion method. The patches are subjected to antimicrobial and antifungal testing, with a specific focus on microbial strains, growth medium, and testing standards. The research study also incorporates FTIR analysis to evaluate the compatibility of medications and excipients, as well as to examine the hygroscopic properties of the patches. The results demonstrate the effectiveness and long-lasting nature of the developed patches, highlighting their potential for use in transdermal medication delivery systems.

Keywords: Antimicrobial Testing, Aqueous Extract, Ocimum sanctum Linn., FTIR analysis, Phytochemical screening, Physicochemical evaluation, Transdermal patches



1. INTRODUCTION

Tulsi in Hindi or Tulasi in Sanskrit (holy basil in English) is a highly revered culinary and medicinal aromatic herb from the family Lamiaceae that is indigenous to the Indian subcontinent and been used within Ayurvedic medicine more than 3000 years. In the Ayurveda system Tulsi depicted in figure no. 1 is often referred to as an "Elixir of Life" for its healing powers and has been known to treat many different common health conditions. In the Indian Materia Medica *tulsi* leaf extracts are described for treatment of bronchitis, rheumatism, and pyrexia[i]. Other reported therapeutic uses include treatment of epilepsy, asthma or dyspnea, hiccups, cough, skin and haematological diseases, parasitic infections, neuralgia, headache, wounds, and inflammation[ii] and oral conditions[iii]. The juice of the leaves has been applied as a drop for earache[iv], while the tea infusion has been used for treatment of gastric and hepatic disorders. The roots and stems were also traditionally used to treat mosquito and snake bites and for malaria[v].Three types of tulsi are commonly described. *Ocimum tenuiflorum* (or *Ocimum sanctum* L.) includes 2 botanically and phytochemically distinct cultivars that include Rama or Sri tulsi (green leaves) and Krishna or Shyama tulsi (purplish leaves)[vi,vii], while *Ocimum gratissimum* is a third type of tulsi known as Vana or wild/forest tulsi (dark green leaves)[viii]. The different tulsi types exhibit vast diversity in morphology and phytochemical composition including secondary metabolites, yet they can be distinguished from other *Ocimum* species by the colour of their yellow pollen, high levels of eugenol[ix], and smaller chromosome number. Despite being distinct species with *Ocimum tenuiflorum* having six times less DNA than *Ocimum gratissimum* they are traditionally used in the same way to treat similar ailments[x]. Tulsi has been the subject of numerous scientific studies and its

pharmacological and wide range of therapeutic applications are the subject of more than one hundred publications during the last decade alone. Numerous in vitro and animal studies attest to tulsi leaf having potent pharmacological actions that include adaptogenic, metabolic, immunomodulatory, anticancer, anti-inflammatory, antioxidant, hepatoprotective, radioprotective, antimicrobial, and antidiabetic effects that have been extensively reviewed previously.[xi] The current research study focuses on the development and evaluation of transdermal patches containing ***Ocimum sanctum Linn.*** leaf aqueous extract. The process involves extraction and processing of the leaves to prepare crude extracts, which are then used in formulating transdermal patches. Various chemicals and equipment used in patch preparation are detailed, followed by phytochemical studies to screen for bioactive constituents. The formulation process includes preparing the transdermal drug delivery system (TDDS) and standardizing the extract using UV spectrophotometry. Physicochemical evaluations of the patches are conducted to assess their quality and performance. Additionally, the study includes stability testing under stress conditions and antibiotic susceptibility testing using the disc diffusion method. Antimicrobial and antifungal testing of the patches is performed, with a focus on microbial strains, media, and standards. The study also covers FTIR analysis for drug-excipient compatibility and the hygroscopic properties of the patches. The findings demonstrate the effectiveness and stability of the developed patches, highlighting their potential for use in transdermal drug delivery systems.

Figure No. 1: Picture of Ocimum sanctum Linn. Leaves

2. MATERIALS AND METHODS

2.1 Collection of Plant and Preparation of Extract

The leaves of *Ocimum sanctum Linn.* were gathered from the campus of CCS University of Pharmaceutical Sciences in Meerut, Uttar Pradesh. The collected leaves were verified by a botanist from the Department of Botany. The leaves were thoroughly cleansed and then dried in the shade at the ambient temperature of the room. The leaves, which had been dried in the shade, were reduced in size and then sifted through sieves numbered 20 and 40. A continuous extraction process was conducted using a Soxhlet apparatus with petroleum ether for 24 hours to eliminate the waxy substances from approximately 500 grams of dried powder. Subsequently, it was extracted using distilled water for a duration of 72 hours. After a period of 72 hours, the water underwent evaporation in order to get the crude extract, which had a concentration of 7.4% weight/volume. The extract was dehydrated using a vacuum oven. The Soxhlation of *Ocimum sanctum Linn.* leaves is depicted in Figure 2.

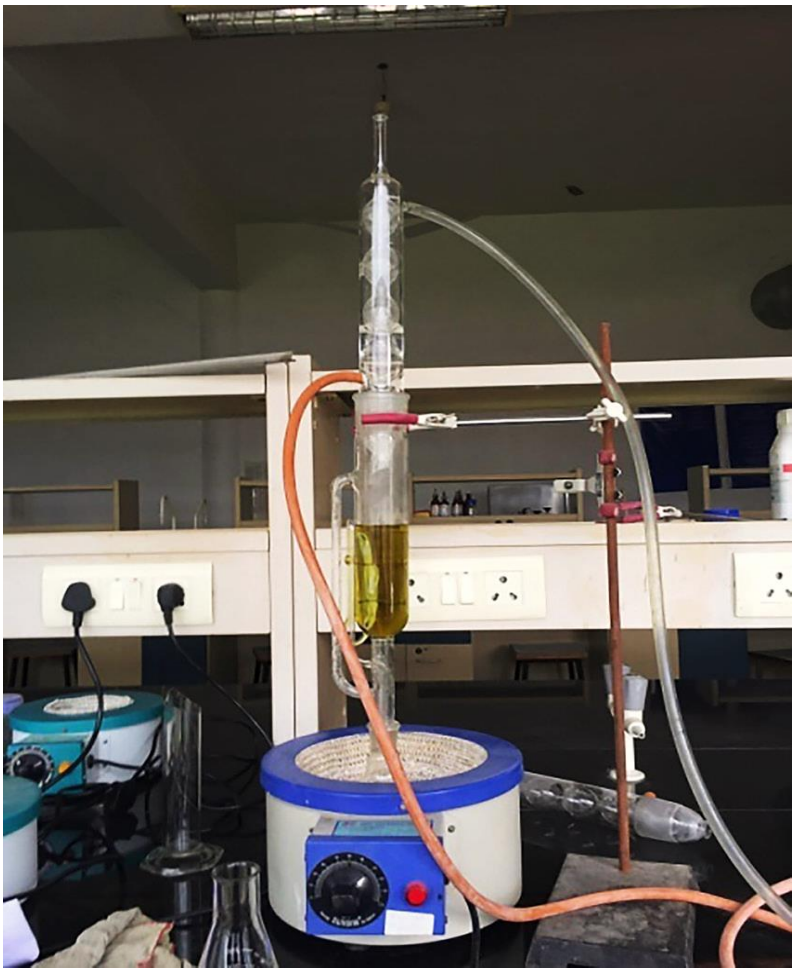


Fig No:2. Soxhlation Apparatus

2.2 Chemicals and equipment

Various chemicals and equipment were used for the preparation of transdermal patches. The list of chemicals and equipment is shown in Tables 1 and 2.

Table No.1. List of Chemicals

S. No.	Chemical Name	Company Name
1.	Pectin	Loba, Delhi
2.	Sodium Alginate	Kemphasol, Delhi
3.	DMSO	Merck Limited, Bhopal
4.	Glycerin	Merk Limited, Bhopal
5.	Potassium dihydrogen orthophosphate	Microfine Chemicals, New Delhi
6.	Sodium hydroxide	Microfine Chemicals, New Delhi

Table No.2. List of equipment's with their purpose

S.No.	Name of Equipment	Name of Manufacturer	Purpose
1.	Dessicator	-	Moisture content studies
2.	Digital Vernier caliper	-	Patch thickness Studies
3.	Electronic balance	Sortorius, Germany	Weighing purpose
4.	Digital pH meter	Elico Ltd, AP, India	Surface pH study

5.	Fourier transfor Infrared Spectrometer	Perkin-Elmer, USA	Compatibility studies
6.	Magnetic Stirrer	ROTEK, W. Vengola, Kerala, India	Diffusion Studies
7.	UV Spectrophotometer	Shimadzu 1700, Japan	Determination of Absorption maxima & concentration of active substances

2.3 Phytochemical Studies[xii]

The Aqueous Extract (AE) underwent phytochemical analysis to determine the presence or absence of its ingredients. A variety of tests were conducted to detect alkaloids, glycosides, saponin glycosides, tannins, phenolic compounds, reducing sugars, amino acids, flavonoids, terpenoids, and steroids.

2.4 Preparation of Transdermal Patch[xiii]

The aqueous extract of *Ocimum sanctum Linn.* leaf was divided into six batches. A. Juss transdermal patches were formulated using a medicine and two separate polymers in three varying ratios (1:4, 1:6, and 1:8). A certain quantity of water was employed to dissolve the polymer with a known mass, which was subsequently heated using a water bath. After incorporating the precise amount of calculated extract into the previous mixture, it was thoroughly blended to achieve a homogeneous mixture. Subsequently, glycerin and a permeation enhancer were added in the precise quantities as determined. The quantity of extract was consistent throughout all six batches. Once the mixture was thoroughly blended, it was transferred into a Petri dish and left to dry naturally for 24 hours at ambient temperature. Subsequently, the patches were extracted from the Petridis using a knife and placed in a desiccator for storage. The ingredients utilized for the formulation of transdermal patches are listed in Table 3.

Table No.3: Formula for TDDS[xiv]

Ingredients	**Formulation Code**					
	TP1	**TP2**	**TP3**	**TS4**	**TS5**	**TS6**
AE (mg)	50	50	50	50	50	50
Pectin (mg)	160	240	320	-	-	
S.Alginate (mg)	-	-	195	180	270	360
DMSO (ml)	0.2	0.2	0.2	0.2	0.2	0.2
Glycerin (ml)	0.3	0.3	0.3	0.3	0.3	0.3
Water	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.	Q.S.

2.5 Preparation of Calibration Curve of Ocimum Sanctum Linn. Extract

A 100 ml volumetric flask was filled with distilled water to dissolve an accurately weighed quantity (100 mg) of AE, resulting in a standard stock solution with a concentration of 1 mg/ml. The concentration ranges from 5 to 50 µg/ml. 1 ml of the stock solution was placed into a 10 ml volumetric flask and the remaining volume was filled with the buffer solution. From this resulting solution, 0.5 ml to 3 ml was transferred into another 10 ml volumetric flask and the remaining volume was filled with more distilled water. The absorbance of these solutions was determined at 382 nm using a UV spectrophotometer. The calibration curve was generated using the absorbance and concentration.

2.6 Physicochemical Evaluation of Ocimum Sanctum Linn. Transdermal Patch

The preliminary assessment tests were carried out on the created patches. Patches that exhibited any faults, trapped air, or inconsistencies in thickness, weight, or material consistency were not included in the subsequent investigation. The evaluation parameters include the uniformity of weight and the thickness of the patch.

- Drug content analysis
- Folding endurance
- The percentage of moisture absorbed
- Percentage moisture content
- The measurement of surface pH is being determined.
- Percent elongation refers to the increase in length of a material expressed as a percentage of its original length.
- The measure of a material's ability to withstand tension without breaking or deforming is referred to as tensile strength.
- Cellulose membrane treatment
- Drug permeation studies
- Diffusion kinetics refers to the study of the rate at which particles or molecules spread and mix in a medium.

2.7 Stability Studies[xv]

The period that a drug product maintains the same properties and features as when it was manufactured is the official definition of stability. This is an early stage of the development process. In current formulations, instability is frequently not noticeable until after a long period has passed under normal circumstances. To evaluate a product's stability, it is customary to subject it to extreme stress conditions, which accelerates deterioration and shortens the testing period. Humidity and temperature are frequent sources of excessive stress. This will get rid of the inadequate formulation. Stability studies parameter shown in Table No. 4.

Table No.4: Stability condition chart

Intended Storage Condition	Stability TestMethod	ICH Test Temperature and Humidity (period in months)	WHO Test Temperature and Humidity (period in Months)
Room Temperature	Long term	25±19C/65±5%RH	35±25C/65±5%RH or 20±10C/60±5%RH 50±30C/70±5%RH
	Intermediate	35±25C/60±5%RH	40±10C/60±5%RH
	Accelerated	45±25C/70±5%RH	60±20C/85±5%RH
Refrigerated	Long term Accelerated	50C/ambient 30±20C/65±5%RH	5±30C 15±10C/70±5%RH or 35±15C/65±5%RH
Freezer	Long term	-300C/ambient	-250C±50C

2.8 Anti-Bacterial Activity[xvi]

Discs loaded with antibiotics at a known concentration Discs are put on an agar plate that has been evenly seeded with a culture of the bacteria under investigation, or inoculated. The plate is incubated at 37°C for 18 to 24 hours. The antibacterial ingredient diffuses into the agar during this time and may stop the growth of the organism. The diameter of the inhibitory zone surrounding the disc determines how effective a susceptibility is. Organisms that extend to the disc's edge exhibit resistance.

2.8.1 Material required

Peptone, Sodium Chloride, Dil. Sodium Hydroxide, Dil. Sulphuric acid, Agar, Distilled water, pH paper, Conical flask, Culture tubes, Glass rod, non-absorbant cotton, Autoclave-Micropipette, Petri dishes and Incubator.

2.8.2 Experimental condition

1. **Organisms used:** *Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa*
2. **Media used** **:** Nutrient Agar.
3. **Test used** **:** AE patch
4. **Standard** **:** Ciprofloxacin, Nystatin

2.8.3 Preparation of Nutrient Agar

250 ml of water was used to dissolve 8.2 grams of agar powder. To precipitate any heat-coagulable substance, the medium was heated in a boiler. The media was filtered after that. Five millilitres of the filtrate were added to each culture tube. We used cotton that wasn't absorbent to clog the tubes. The medium within the tubes was autoclaved for a minimum of 15 minutes at 121 degrees Celsius and 15 pounds per square inch.

2.8.4 Preparation of Paper Disc

Standard paper with a diameter of 6.0 mm was created by cutting filter paper with a standard punching machine. The paper discs were sterilized for one hour at 160 degrees Celsius in a hot air oven. After that, the test solution was injected into the paper discs.

2.9 Anti-Fungal Activity[xvii]

Discs loaded with antibiotics at a known concentration Discs are put on a modified Sabouraud's glucose agar plate that has been evenly seeded with a culture of the fungus under investigation, or inoculated. The plate is incubated at 37°C for three days, 18–24 hours. The antifungal agent diffuses through the agar during this time and may stop the organism from growing. The diameter of the inhibitory zone surrounding the disc determines how effective a susceptibility is. Organisms that extend to the disc's edge exhibit resistance.

2.9.1 Materials Required

Glucose, Peptone, yeast extract, Glycerin monostearate, Olive oil, Tween 80, Chloramphenicol, Agar, Distilled water, pH paper, Conical flask, Culture tubes, Glass rod, Non-absorbant cotton, Autoclave, Micropipette Petri dishes and Incubator.

2.9.2 Experimental condition[xviii]

1. **Organisms used** **:** *Candida albicans, Aspergillus niger.*
2. **Media used** **:** Modified Sabouraud's Glucose Agar medium
3. **Test used** **:** AE patch,
4. **Standard** **:** Ciprofloxacin, Nystatin

3. RESULTS AND DISCUSSIONS

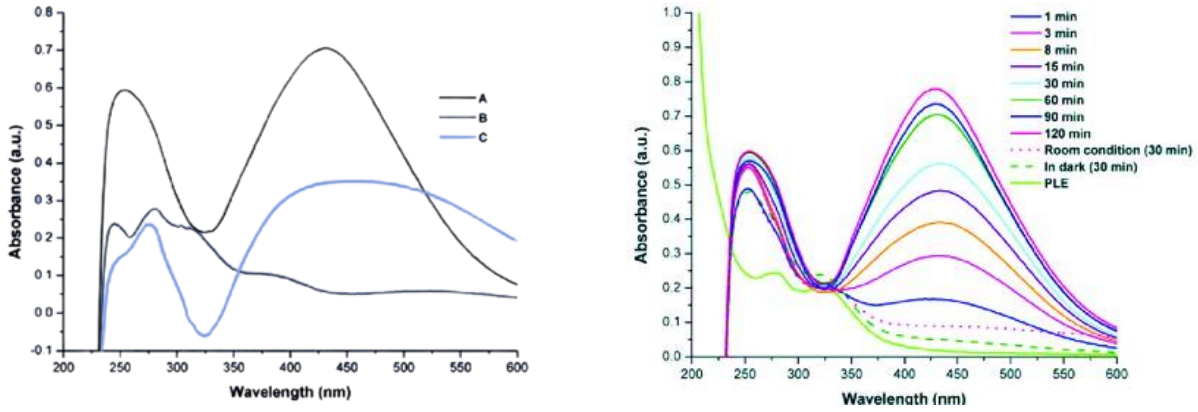
3.1 Absorption Maxima (Λ Max) Of Aqueous Extract of Ocimum Sanctum Linn.

The sharp peak was observed at 382 nm, further measurements were taken at 382nm. UV spectrum of *Azadiractha indica* A Juss aqueous extract[xix] given as Fig No. 3.A & B

3.2 Standard Curve Values of Aqueous Extract of Ocimum Sanctum Linn.[xx]

To draw the linearity of standard their concentration and absorbance are shown in Table No. 5 and calibration curve is shown in Figure No. 4

Fig No.3: Absorption maxima (λ max) of Aqueous Extract of *Ocimum sanctum Linn.*



A **B**

Table No.5: Standard values of Aqueous Extract of *Ocimum sanctum Linn.*

Concentration(g/ml)	**Absorbanceat 382 nm**
	Average - SD
0	0.000 - 0.000
10	0.091- 0.001
20	0.182 - 0.001
30	0.273 - 0.001
40	0.363 - 0.001
50	0.432 - 0.001
60	0.512 – 0.001

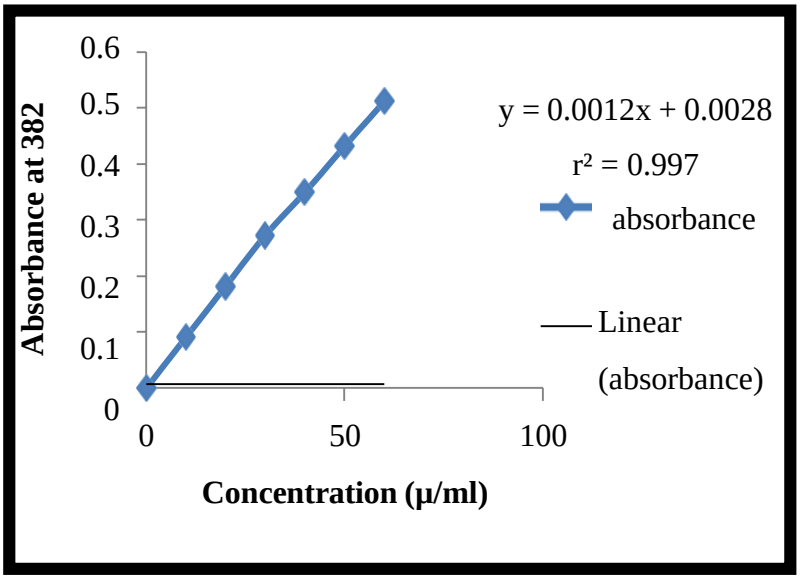


Fig No.4: Standard Curve of Aqueous Extract of *Ocimum sanctum Linn.*

3.3 Physical appearance

1.	**Color**	**:**	**Green**
2.	**Taste**	**:**	**Bitter taste**
3.	**Solubility**	**:**	Freely soluble in Distilled Water

3.4 Phytochemical Studies[xxi]

The phytochemical studies revealed the presence of alkaloids, saponins,tannins, phenolic compounds, flavonoids and sterols. Complete data is shown in Table No. 6.

Table No.6: Phytochemical constituents

S. No.	Chemical constituents	Aqueous Extract
1.	Alkaloids	+
2.	Saponins	+
3.	Tannins	+
4.	Phenolic	+
5.	Compounds	+
6.	Flavonoids	+
7.	Steroids	+
8.	Glycosides	+
9.	Amino acids	-
10.	Reducing sugar	-
11.	Terpenoids	

(+) **Presence of constituents** **(-) Absence of constituents**

3.5 Hygroscopic Nature

The results, it is observed that the prepared Transdermal patch is hygroscopic data represented in Table No. 7.

Table No. 7. Hygroscopic Nature determination

At Room Temperature	75%RH at 40^0C
Sample No-1	Sample No-1
Weight gain observed (0.40±2.0)	Weight gain observed (0.35±0.8)

3.6 Compatibility Study

The FTIR graphs of drugs, excipients and formulation results showed that there is no extra peak (or) broadening of the peak was observed and thus it indicates that there is no incompatibility between drug and excipients.

3.6.1 FTIR studies

FTIR spectrum of Aqueous Extract of ***Ocimum sanctum*** *Linn.*, Pectin, Pectin formulation, Sodium alginate and Sodium alginate formulation are shown in Figure No. 5 and FTIR Interpretation given in Table No. 8.

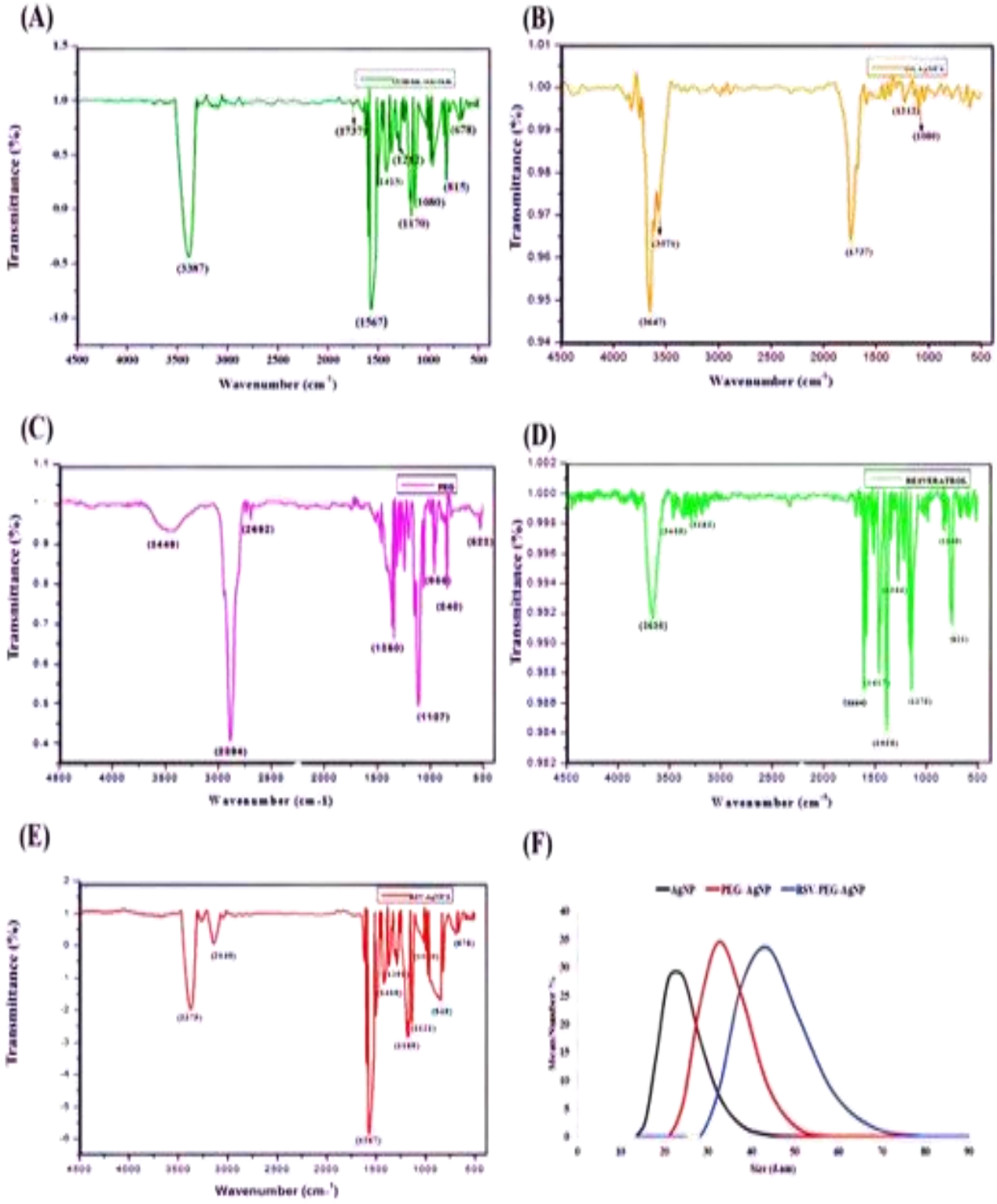


Fig No. 5: FTIR spectrum of Aqueous Extract of *Ocimum sanctum Linn.* Pectin formulation, Sodium alginate and Sodium alginate formulation[xxii]

Table No. 8. FTIR Interpretation of Aqueous Extract of *Ocimum sanctum Linn.,* Pectin

Wave number (cm^{-1})	**Functional Group**
Aqueous Extract of *Ocimum sanctum Linn.*	
3381.610	C=O stretching
2937.65	C-H Stretching
1678.90	C-C Stretching
1510.20	OH Bending
956.30	C-O Stretching
790.94, 783.02	C-H Rocking
Pectin	
3230.60	C=O stretching
2830.16	C-C stretching
1567.10	C-C stretching
1563.20	OH bending
945.30	C-O Stretching
780.70	CH Rocking
Pectin formulation	
3412.10	C=O stretching
1970.66	C-C stretching
1031.10	C-C stretching
111.01	OH bending
920.901	C-O Stretching
856.30	CH Rocking
Sodium alginate	
3720.50	C-H stretching
2876.81	C-H stretching
1980.01,1419.20	C-C multiple bonds stretching
1315.20	O-H stretching
Sodium alginate formulation	
3456.10	O-H stretching
1672.50	O-H bending
1078.81	C-O stretching
1254.20	C-O stretching
1009.08	C-O stretching
917.06	C-H (Out plane bending)
798.60	C-H (Out plane bending)

formulation, Sodium alginate and Sodium alginate formulation

3.7 Physicochemical evaluation of Aqueous Extract of *Ocimum sanctum Linn.* Transdermal patch

The six (P1, P2, P3, S4 S5, S6) batches of extract-loaded patches with different ratios of two different polymers weresubjected to various physicochemical evaluations.

Based on thickness, uniformity of weight, folding endurance, percentage moisture uptake

moisture content and tensilestrength, the formulations P2 and S5 were selected for further studies. Table 9 Shows data on physiochemical evaluation.

Table No.9: Physicochemical evaluation of Aqueous Extract of *Ocimum sanctum Linn.* Transdermal patches (Mean ± S.D: n = 3)

Formulation Code		Uniformity Of Weight (g)	Thickness (mm)	Drug content (%)	Folding Endurance (no's)	Moisture Uptake (%)	Moisture Content (%)	Surface pH	Percent Elongation (% mm)	Tensile Strength (Kg/mm^2)
Transdermal Patch of Pectin (P)	P1	0.37± 0.12	0.40± 0.70	75.20± 0.86	279± 0.70	2.10± 0.09	2.756± 0.06	7.0± 0.11	81± 0.30	5.308±0.87
	P2	**0.40± 0.80**	**0.47± 0.20**	**83.89± 0.50**	**247± 0.27**	**2.89± 1.00**	**3.421± 0.21**	**7.4± 0.70**	**94± 1.10**	**7.382±0.96**
	P3	0.47± 0.11	0.40± 1.11	80.90± 0.15	274± 0.26	3.70± 1.00	3.946± 0.30	7.1± 0.70	85± 1.04	6.700±0.60
Transdermal Patch of Sodium alginate (S)	S4	0.39± 0.43	0.36± 0.33	82.03± 0.22	241± 0.72	2.75± 0.65	1.736± 0.46	7.0± 0.33	82± 0.23	5.513±0.86
	S5	**0.40± 0.60**	**0.41± 0.70**	**82.30± 0.90**	**269± 0.43**	**1.90± 0.40**	**1.650± 0.02**	**7.0± 0.10**	**80± 0.90**	**6.250±0.56**
	S6	0.46± 0.51	0.49± 0.13	87.10± 0.40	247± 0.42	2.79± 0.35	2.457± 0.04	7.0± 0.10	86± 0.80	6.40±0.708

3.8 Optimized formula of *Ocimum sanctum Linn.* Transdermal patch

Formula for *Ocimum sanctum Linn.* Transdermal patch is given in Table No. 10

Table No.10: Optimized formula of *Ocimum sanctum Linn.* Transdermal patch

S.NO.	Ingredients	P (Pectin)	S (Sodium alginate)
1.	Extract (mg) *Ocimum sanctum Linn.*	40	39
2.	Polymer (mg)	242	241
3.	DMSO/SLS (ml)	0.4	0.3
4.	Glycerin (ml)	0.4	0.4
5.	Water (ml)	Q.S.	Q.S.

3.9 Prepared Patches

A figure of prepared patches is given in Figure 6.

3.10 Physicochemical Evaluation of Selected Formulations

Physiochemical evaluation of selected formulations on different parameters is shown in Table No. 11.

Table No.11: Physicochemical Evaluation of Selected Formulations

S. No.	Parameter	Formulation Code (Mean ± S.D: n= 3)	
		Pectin (P2)	**Sodium alginate (S5)**
1.	Uniformity of weight (g)	0.399±0.75	0.49±0.80
2.	Thickness of the patch (mm)	0.49±0.23	0.46±0.88
3.	% Drug content	84.70±0.80	89.21±0.80

4.	Folding Endurance	270±0.11	260±0.34
5.	% Moisture uptake	1.80±1.03	1.87±0.54
6.	% Moisture content	4.123±0.22	1.607±0.05
7.	Surface Ph	6.4±0.72	8.1±0.10
8.	% Elongation (%mm)	87±1.11	97±0.92
9.	Tensile strength (kg/mm^2)	5.401±0.65	6.209±0.20

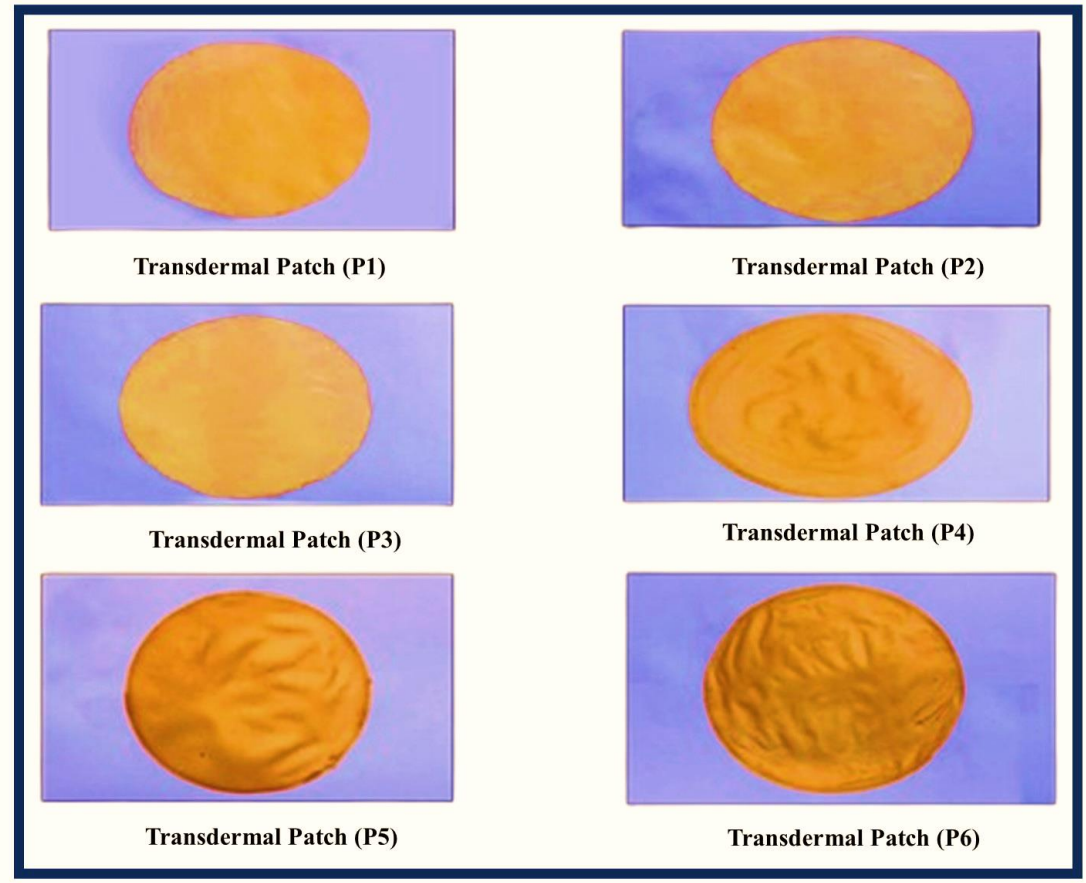


Fig No. 6: Pictures of Prepared Patches

3.11 Screening of Antimicrobial activity of *Ocimum sanctum Linn.*

Disc diffusion technique was used to determine the anti-microbial activity of the sample (Indian Pharmacopoeia 1996, vol II A-105). The National Chemical Laboratory (NCL) in Pune provided the test microorganisms of Staphylococcus aureus, Bacillus subtilis, Klebsiella pneumonia, Proteus vulgaries, and Aspergilus niger, as well as Candida albicans. These microorganisms were then routinely subcultured on nutrient agar and sabouraud dextrose agar medium for the bacteria and fungi, respectively. The sample's effect was contrasted with that of the positive control, the reference standard Ciprofloxacin (5µg/disc for bacteria and Nystatin 100µg/disc for fungi).

3.11.1 For Fungi

After 72h the plates were observed. The zone of inhibition was calculated by measuring the minimum dimension of the zone of no fungal growth around the patch The figures are shown in 7 & 8.

3.11.2 For Bacteria

After 24h the plates were observed. The zone of inhibition was calculated by measuring the minimum dimension of the zone of no bacterial growth around the patch. The Figures are shown in 9,10,11&12.

FUNGI

Fig No.7: *Bacillus subtilis*

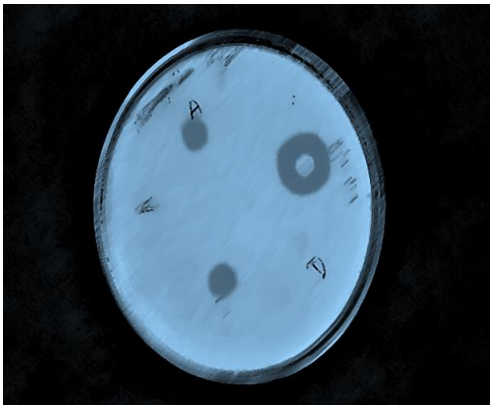
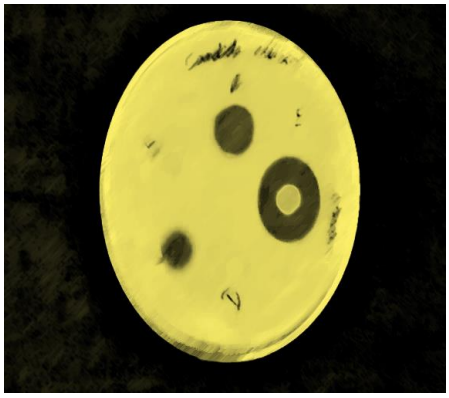


Fig No.8: *Staphylococcus aureus*



GRAM POSITIVE BACTERIA

Fig No.9: *Bacillus subtilis*

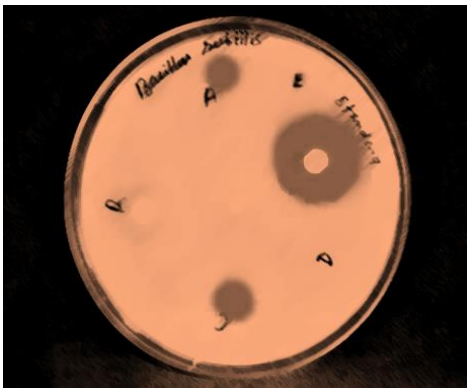
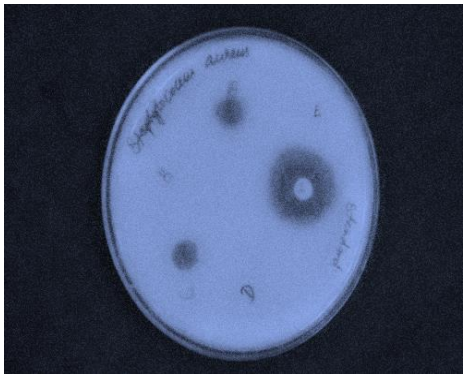


Fig No.10: *Staphylococcus aureus*



GRAM NEGATIVE BACTERIA

Fig No.11: *Klebsiella pneumonia*

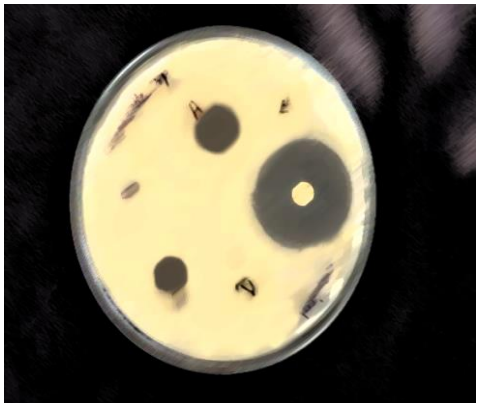
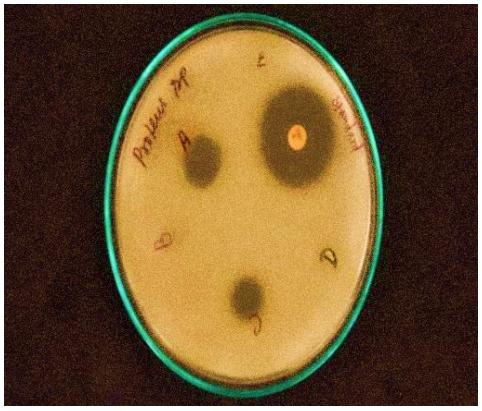


Fig No.12: *Proteus vulgaries*



3.12 Stability study of Aqueous Extract of *Ocimum sanctum Linn.* TransdermalPatch

The stability studies of formulation of aqueous extract of ***Ocimum sanctum Linn.*** transdermal patch were carried out for 3 months as per the procedure described in the Table No:12. During this period, the formulations were stable and showed no significant changes invisual appearance, colour, texture and drug content.

Table No: 12. Stability study of P2

Parameter	**Room temperature**	**40±3^{0}C& RH 70±5%**
Visual Appearance	Brown colour	Brown colour
Initial	No change	No change
At the end of 1st month	No change	No change
At the end of 2nd month	No change	No change
At the end of 3rd month	No change	No change
Color	Brown	Brown
Initial	No change	No change
At the end of 1st month	No change	No change
At the end of 2nd month	No change	No change
At the end of 3rd month	No change	No change
Texture	Smooth	Smooth
Initial	No change	No change
At the end of 1st month	No change	No change
At the end of 2nd month	No change	No change
At the end of 3rd month	No change	No change
Drug content	No change	No change
Initial	**80.60%**	**87.60%**
At the end of 1st month	83.20%	83.17%
At the end of 2nd month	80.90%	80.82%
At the end of 3rd month	83.20%	80.70%

4. SUMMARY AND CONCLUSION

Six batches/sets of transdermal patches containing aqueous extracts of *Ocimum sanctum Linn.* (P1, P2, P3, S4, S5, and S6) were made using the solvent casting technique. The formulation parameters, drug-polymer ratios, and permeation enhancers were tuned to achieve thin, clear, smooth, stable, and highly permeable transdermal patches. The FTIR spectra of the medicine, excipients, and formulations revealed no additional peaks or broadening of peaks, indicating the absence of any incompatibility between the drug and excipients.
The evaluation of all six batches included the assessment of Percentage Moisture uptake, Percentage Moisture content, Thickness, Folding Endurance, Percentage Drug content, percentage elongation, Tensile strength, and Adhesive strength. After conducting optimization, the two best formulations, P2 and S5, were chosen based on a physicochemical evaluation and an in vitro drug diffusion investigation. The formulations P2 & S5 exhibited the highest percentage of moisture absorption, moisture content, thickness, folding endurance, drug content, percent elongation, and tensile strength. There was no discernible variation in the

medication content among the six formulations of the patches. This demonstrates the uniform distribution of medication during the patch formulation. The drug diffusion from P2 & S5 exhibits zero-order and non-Fickian diffusion. These data suggest that the drug diffusion from the P2 & S5 patch was controlled by the process of diffusion.
Based on the findings of experiments involving the diffusion of substances in a controlled environment and the analysis of their physical and chemical properties, it was determined that the P2 & S5 formulation is the most effective. Subsequently, they underwent screening to determine their antibacterial activity. The antimicrobial screening result indicated that P2 strongly inhibited the growth of microorganisms around the patch. Therefore, P2 was chosen for additional assessments, including ex vivo and stability investigations. The findings of the ex vivo experiment demonstrated a drug diffusion rate of 61.01% after 8 hours. The study determined the controlled diffusion characteristic across the skin. The stability experiments revealed that there was no substantial alteration in its original characteristics for a duration of three months at a temperature of 40˚C ±2˚C and a relative humidity of 75±5%. The current study successfully accomplished the goal of developing a transdermal patch containing an aqueous extract of *Ocimum sanctum Linn.* This was achieved by utilizing several polymers. The diffusion kinetics indicate that the formulation adhered to a zero-order, non-fickian diffusion model.

Future Directions

Future study should give priority to several crucial areas in order to further improve and validate the transdermal patches of *Ocimum sanctum Linn.* Thorough and extended clinical trials are necessary to assess the effectiveness and safety of these patches in various groups of patients and real-life situations. To enhance the effectiveness of the treatment and ensure patient adherence, it is necessary to optimize the formulation by investigating further variants and fine-tuning the concentration of active substances. It is necessary to explore advanced methods of delivering drugs, such as nanoencapsulation or microneedle technology, in order to enhance the effectiveness and absorption of active components. Conducting extended stability studies will allow for the evaluation of the longevity of the patches under different environmental circumstances, hence ensuring their durability and efficacy throughout time. Moreover, by extending the scope of antimicrobial testing to encompass a wider array of microbial strains and resistance patterns, the patches' overall efficacy may be definitively established. Ultimately, it is essential to carry out patient-centric studies in order to assess the comfort, adhesion, and general happiness of patients with the transdermal patches, while also resolving any potential usability problems. These research initiatives will expedite the advancement of more efficient and dependable transdermal delivery methods, potentially providing novel therapeutic choices for the treatment of skin disorders and systemic diseases.

Compliance with ethical standards

Acknowledgments

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Disclosure of conflict of interest

The authors declare there is no conflict of interest in this Research study.

Statement of informed consent

Informed consent was obtained from all individual participants included in the Research study.

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