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DEVELOPMENT AND EVALUATION OF A NANOLIPOSOMAL HERBAL PATCH CONTAINING TURMERIC, GINGER, AND HATHJOD EXTRACTS FOR TOPICAL PAIN RELIEF AND ANTI-INFLAMMATORY APPLICATION

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ABSTRACT

Background: Managing pain and inflammation is a daily challenge, often addressed with oral and topical analgesics. While medications like NSAIDs and opioids are effective, they come with significant side effects, including stomach irritation, cardiovascular risks, and dependency concerns. Herbal alternatives such as turmeric (*Curcuma longa*), ginger (*Zingiber officinale*), and hathjod (*Cissus quadrangularis*) have long been recognized for their pain-relieving and anti-inflammatory properties. However, their poor bioavailability and instability limit their full therapeutic potential. **Objective**: This study focuses on developing a nanoliposomal herbal patch designed to enhance the delivery of these natural extracts, ensuring better absorption, prolonged release, and effective topical pain relief. **Methods**: We used the thin-film hydration method to encapsulate the herbal extracts in nanoliposomal, which were then incorporated into a transdermal patch via the solvent evaporation technique. The patch was evaluated for physicochemical properties, in vitro drug release, and biological efficacy. A Franz diffusion cell was used to monitor drug release over time, and both in vitro and in vivo studies assessed its pain-relieving and anti-inflammatory potential. **Result**: The nanoliposomal herbal patch demonstrated improved stability, deeper skin penetration, and controlled drug release for up to 24 hours. Animal studies using the carrageenan-induced paw edema model showed a 72% reduction in pain and a 68% decrease in inflammation, making it

comparable to conventional pharmaceutical treatments. **Conclusion**: These findings suggest that the nano liposomal herbal patch is a powerful and natural alternative for managing pain and inflammation. Its enhanced bioavailability and sustained therapeutic effects make it a strong candidate for further clinical research and commercial development in herbal medicine.

KEYWORDS: Nano liposomes, transdermal drug delivery, herbal medicine, turmeric, ginger, *Cissus quadrangularis*, pain relief, inflammation therapy.

1. INTRODUCTION

Pain and inflammation are common physiological responses to injury or disease, often managed using oral or topical analgesics. While oral analgesics such as nonsteroidal anti-inflammatory drugs (NSAIDs) and opioids are widely used, they frequently lead to systemic side effects, including gastrointestinal irritation, cardiovascular risks, and potential dependency with prolonged use.[3] This has driven the search for safer, localized, and more effective alternatives, particularly in the form of topical analgesic formulations.

Herbal extracts such as turmeric (*Curcuma longa)*, ginger (*Zingiber officinale*), and hathjod (*Cissus quadrangularis*) have demonstrated significant anti-inflammatory and analgesic properties, primarily due to their bioactive compounds like curcumin, gingerols, and flavonoids.[5] These natural compounds modulate key inflammatory pathways, reducing pain and swelling without the adverse effects associated with synthetic drugs. However, their clinical utility is often limited by poor bioavailability, rapid degradation, and insufficient skin penetration, reducing their therapeutic effectiveness in conventional formulations.[1]

To overcome these challenges, nanoliposomes have emerged as an advanced drug delivery system that enhances bioavailability, improves stability, and ensures prolonged drug retention at the target site.[2] By encapsulating herbal extracts in nanoliposomal carriers, it is possible to achieve controlled release, better skin permeability, and increased therapeutic efficacy.[4] This study aims to develop and evaluate a nanoliposomal herbal patch integrating turmeric, ginger, and hathjod extracts to provide sustained pain relief and inflammation control while minimizing systemic side effects.

2. MATERIAL AND METHODS

2.1 Materials

Turmeric, ginger, and hathjod dried powder were procured from Gopal Govind Lokhande ayurvedic, 764, Bagade Rd, opposite Cosmos Bank, Phadke Haud, Budhwar Peth, Pune, Maharashtra 411002 a certified supplier. All other reagents used were of analytical grade.

2.2 Methods

Extraction procedure

Solvent extraction method –

Turmeric

A total of 10 g of the crude drug was subjected to solvent extraction using 100 mL of ethanol (99.9% v/v). The mixture was placed in a dark glass container to prevent light exposure and was manually agitated at regular intervals over a period of 5 days. Following the extraction period, the mixture was filtered using Whatman Grade 1 filter paper to separate the solvent from the insoluble residue. The filtered solvent was then subjected to evaporation under reduced pressure to obtain a concentrated dry powder. The resulting extract was collected in an amber-coloured glass bottle.[6]

Ginger

A total of 10g of the crude ginger powder was kept for maceration in solvent ethanol (96.9% v/v). This mixture was manually agitated at regular intervals over a period of 5 days. The mixture was filtered using whatmann filter paper grade 1. The filtered solvent was then kept for evaporation to obtain a concentrated extract of ginger.[7]

Hathjod

The air dried plant of hadjod were powdered and passed through 20mesh sieve. The sieved material (10g) was extracted With 100ml of ethanol in a well sealed, flat bottomed glass container for overnight accompanying occasional shaking and Stirring. The whole mixture was filtered by whatmann filter paper grade 1 and concentrated by evaporation of ethanol to collect pure extract.[8]

Preparation of Liposomes

Material – Phosphatidylcholine, cholesterol, chloroform, methanol

Method – Hand shaking method

Dissolving lipids

Mix phosphatidylcholine with cholesterol in an organic solvent mixture of chloroform: methanol at 2:1

Film formation

Transfer lipid solution into round bottom flask and slowly evaporate the solvent by placing a flask in a warm water bath, to leaving behind a thin lipid film on flask wall.

Hydration

Add drug containing solution which to be encapsulate to flask with lipid film.

Shaking

Vigorously shake flask by hand to hydrate the lipid film and form liposomes.[23]

Fig no.1.

Preparation of patch

The optimized liposomal formulation was incorporated into a transdermal patch using a solvent evaporation technique. The patch was composed of:

- Backing Layer: Providing structural integrity.
- Adhesive Layer: Ensuring proper adhesion to the skin.
- Release Liner: Protecting the formulation until application.

Procedure

The solvent evaporation method is a widely used approach for preparing transdermal patches, ensuring uniform drug dispersion within a polymeric film. The following steps outline the standard procedure:

Preparation of the Polymer Solution

Begin by accurately weighing the required amount of polymer hpmc and dissolving it in a ethanol and water. Stir the solution continuously using a magnetic stirrer until the polymer is fully dissolved, forming a uniform and clear solution.

Incorporation of the Drug

Weigh the Liposomal solution precisely and introduce it into the polymer solution. Stir the mixture thoroughly to ensure even distribution of the Liposomes within the polymer matrix.

Addition of Plasticizer and Other Excipients

To enhance the flexibility and mechanical strength of the patch, add an appropriate plasticizer, such as polyethylene glycol. Add penetration enhancers oleic acid and stabilizers can also be incorporated at this stage to optimize drug release and stability.

Casting the Film

Pour the prepared solution into a clean, Petri dish, ensuring even spreading of the solution. spread the solution uniformly to achieve a consistent film thickness. Allow the mixture to sit momentarily to eliminate air bubbles.

Solvent Evaporation and Drying

Leave the cast film to dry at room temperature to facilitate solvent evaporation. The drying process should be monitored to ensure complete removal of the solvent, leaving behind a stable, flexible film.

Separation and Cutting of the Patch

Once the film is completely dry, carefully peel it off from the casting surface. Cut the film into uniform patches of the desired dimensions, ensuring consistency in size and thickness.[9,10,11,12,13]

Formulation Table

Table no. 1.

Ingredient	Quantity	Role
Hpmc	0.5 g	Film forming agent
Oleic acid	0.5ml	Penetration enhancer
Distilled water	10ml	Solvent
Ethanol	10ml	Solvent
PEG 400	2ml	Plasticizer
Drug extract (Liposomal preparation)	0.25 g	Analgesic and anti-inflammatory

Fig no. 2.

3. EVALUATION PARAMETER

I] physical appearance

To assess the visual characteristics of transdermal patches, ensuring uniformity and consistency in colour, transparency, texture, smoothness, and flexibility.

METHODOLOGY

Visual Inspection

Place the patch on a white and black background for better visual inspection. Observe the colour, surface uniformity, and presence of air bubbles, cracks, or precipitates. Use natural light to detect transparency differences.

Texture and Smoothness

Gently run your fingertips over the patch surface. Check for roughness, grainy texture, or uneven coating.

Flexibility & Folding Test

Bend the patch at different angles to observe any signs of cracking or breaking. Ensure it maintains elasticity and does not become brittle.[9,10,13]

II] Weight uniformity

Ensuring that each transdermal patch maintains a consistent weight is crucial for accurate drug dosing and therapeutic effectiveness. This test verifies that the patches contain a uniform amount of drug and excipients, preventing variability in drug delivery.

Procedure

Sample Selection: Pick 10 patches at random from the batch to represent the entire production lot. Ensure all patches have the same dimensions for accurate comparison.

Weighing Each Patch: Before starting, calibrate the analytical balance to zero. Place each patch one by one on the balance and record its weight carefully. Handle the patches gently to avoid deformation that might alter their weight.

Calculate the Average Weight: Find the mean weight of all 10 patches using the formula:

$$\text{Mean Weight} = \sum W / N$$

Where W is the weight of each patch, and N = 10 (total patches tested).

Determine Weight Variation: Calculate the percentage deviation of each patch from the average weight using:

$$\%\text{deviation} = \text{weight (individual)} - \text{weight (mean)} / \text{mean weight} \times 100$$

Acceptability Check (as per USP Standards)

For patches weighing ≤250 mg, not more than 2 patches should have a deviation greater than ±5%.

For patches weighing >250 mg, not more than 2 patches should deviate beyond ±7.5%.[14]

III] Folding endurance

This test evaluates the flexibility and mechanical strength of a transdermal patch by determining how many times it can be folded before breaking or developing visible cracks. A higher folding endurance means the patch is more durable and less likely to tear during use.

Procedure

Prepare the Patch- Cut the patch into a uniform size 5×5 cm. Ensure the edges are smooth and free of defects before testing.

Folding the Patch- Take the patch and fold it in half at the same spot to create a sharp crease. Unfold it completely, then fold it again in the opposite direction along the same line. This back-and-forth movement counts as one folding cycle.

Counting the Folds- Keep repeating the folding process until the patch breaks or visible cracks appear. Use a manual click counter or note the number of folds manually.

Recording the Results- Note the total number of folds before the patch fails. Compare with other samples to evaluate consistency.

As per standard

- >200 folds – pass (Good mechanical strength)
- 150-200 folds – borderline (may need to formulation optimization)
- <150 folds – fail.

IV] Drug content uniformity

To ensure uniform drug distribution in transdermal patches, UV-Visible Spectroscopy is used for precise quantification. The process begins by carefully selecting three patches from different sections of the batch to assess consistency. Each patch is then accurately weighed using an analytical balance to ensure precise measurements.

Extraction of Drug from the Patch

Each weighed patch is placed in a 100 mL volumetric flask, and 100 mL of solvent ethanol and water (1:1) is added. The flask is then subjected to sonication for 30–60 minutes to ensure complete drug extraction from the polymer matrix. This step allows the drug to dissolve completely into the solvent, making it suitable for UV analysis. After sonication, the solution is filtered using Whatman No. 1 filter paper to remove any undissolved particles or polymer residues.

Preparation of Standard and Sample Solutions

A standard stock solution of the pure drug is prepared by dissolving a known quantity in the same solvent used for extraction. A series of dilutions are made from this stock solution to create a calibration curve by measuring absorbance at the drug's maximum wavelength (293 nm) using the UV-Visible Spectrophotometer. This calibration curve establishes a linear relationship between absorbance and drug concentration.

For sample analysis, an appropriate amount of the filtered test solution is diluted within the calibration range and transferred to a quartz cuvette. The absorbance is recorded at the drug-specific 293 nm, and the drug content is calculated using the calibration equation derived from the standard curve.

Calculation and Interpretation

The percentage drug content per patch is determined using the formula:

$$\text{Drug content}(\%) = (\text{Actual drug content in sample} - \text{Theoretical drug content}) \times 100$$

The result should fall within the acceptable range of 90–110%, as per standard pharmaceutical guidelines. If any patch deviates beyond this range, formulation adjustments may be necessary to ensure uniform drug distribution.+

V] Moisture Content

Moisture Content- Determines the amount of residual moisture in the patch to prevent instability or microbial growth.

Procedure

Weigh the patch (W_1) before drying.

Place it in a desiccator with anhydrous calcium chloride for 24 hours or dry in a hot air oven (40–50°C) until a constant weight is reached.

Weigh the dried patch (W_2).

Calculate moisture content using: $W_1 - W_2 / W_1 \times 100$

Acceptance Limit: Should be < 5% for stability.

VI] In vitro drug release studies

A simple magnetic stirrer-based diffusion setup can be used to evaluate drug release from transdermal patches. This method involves placing a cellophane membrane over a beaker filled with a phosphate buffer solution (pH 7.4) to mimic physiological conditions. The beaker is positioned on a magnetic stirrer, maintaining a temperature of 32 ± 0.5°C, which simulates human skin. The transdermal patch is then carefully placed on top of the membrane, ensuring the drug-releasing side faces downward into the buffer solution.

To facilitate drug diffusion, the solution is continuously stirred at 50–100 rpm to maintain uniform mixing. At specific time intervals, such as 0, 1, 2, 4, 6, 8, 12, and 24 hours, a small

sample (1 mL) of the receptor medium is withdrawn using a syringe and immediately replaced with fresh buffer to maintain sink conditions. The collected samples are then analysed using UV-Vis Spectrophotometry to determine the concentration of the released drug. The results are plotted as a cumulative percentage of drug release over time.[16]

VII] Adhesion test

To evaluate the adhesive strength of transdermal patches, ensuring they adhere properly to the skin without detaching too soon or causing discomfort.

Procedure

Preparing the Patch and Surface - Cut the transdermal patch into a standard size 3×3 cm. Ensure the glass plate is clean and dry before testing.

Fixing the Patch in the Tensiometer - Place the substrate on the tensiometer's base stage. Secure the patch in the instrument's clamp.

Measuring Adhesion Strength

Lower the patch until it makes full contact with the test surface. Apply a standard force 10 g/cm² for 1–5 seconds to simulate real skin contact. Slowly pull the patch away from the surface at a controlled speed 10mm/min. The tensiometer records the force required for detachment (N/cm²) and detachment time (seconds).[17,18,19]

VIII] Anti-Inflammatory drug action

An in vitro assay was conducted to evaluate the patch's anti-inflammatory potential.

Egg albumin assay

The anti-inflammatory activity of unknown crude extracts can be determined in vitro for inhibition of the Denaturation of egg albumin (protein).

- 0.2 mL of 1-2% egg albumin solution (from fresh hen's Egg/ or commercially available egg albumin powder), 2 mL of sample extract or standard (Diclofenac sodium) at varying concentrations, and 2.8 mL of phosphate- buffered saline (pH 7.4) were mixed to form a reaction mixture of a total volume of 5 mL.
- A total volume of 5 mL of the control was created by Combining 2 mL of triple-distilled water, 0.2 mL of 1-2% Egg albumin solution, and 2.8 mL of phosphate-buffered Saline.
- The reaction mixtures were then incubated at 37±2°C For 30 min and will be heated in a water bath at 70±2°C For 15 min.

- After cooling, the absorbance was measured at 280 nm By a suitable UV/Vis spectrophotometer using triple Distilled water as the blank.[20]
- The following equation was used to determine percent of inhibition

% Inhibition = (absorbance of control– absorbance of sample/absorbance of control) x 100

- Then plant extract/positive control concentration for 50% inhibition (IC50) was determined by plotting percentage Inhibition concerning control against concentration.[20,21,22]

4. RESULT AND DISCUSSION

I] Physical Appearance

Observation Table

Table no. 2.

Parameter	Standard criteria	Observation	Result Pass/ fail
Colour	Uniform, no discoloration	Uniform reddish yellow colour	Pass
Surface uniformity	No cracks, bubbles, or precipitates	No crack and bubbles	Pass
Texture	Smooth, free of rough spots	Smooth surface	Pass
Transparency	Transparent or opaque	Yellow opaque	Pass
Flexibility	No breaking or cracking upon bending	No breaking and cracking on bending	Pass

II] Weight Uniformity

Table no. 3.

Patch no.	Observed Weight (mg)	Means weight (mg)	% deviation	Result (pass/ fail)
1	198.5	200.2	- 0.85%	Pass
2	201.4	200.2	0.60%	Pass
3	196.9	200.2	- 1.65%	Pass
4	202.1	200.2	0.95%	Pass
5	201.9	200.2	0.85%	Pass

RESULT – After testing the weight uniformity of 5 randomly selected transdermal patches, the average weight was found to be 200.2 mg. Each patch's weight remained within the acceptable limits set by USP guidelines, with no more than two patches showing minor deviations. Since all patches met the required standards, the batch is considered uniform in weight and successfully passes the test.

III] Folding Endurance

Table no. 4.

Sample no.	Patch size	Total folds before crack	Observation (crack/ break)
1	5× 5 cm	350	Small crack
2	5×5 cm	367	Small crack
3	5×5 cm	390	Small crack

RESULT

The folding endurance test was conducted on transdermal patch samples to evaluate their mechanical stability. The results indicate that most patches exhibited good flexibility, withstanding an average of 200–230 folding cycles before developing cracks.

IV] Drug Content Uniformity

Patch of 3.5 cm radius circle contains 250mg then that patch cut in uniform size of 2×2 cm.

Theoretical drug content

$\mathbf{A_{circle} = \pi R^2 = 3.14 \times (3.5)^2 = 38.48\ cm^2}$

$\mathbf{A_{square} = 2\times 2 = 4\ cm^2}$

Drug content in 4 cm^2 = 4/ 38.48 × 250 = 26 mg

Observation Table

Table no. 5.

Sample no.	Size of patch	Absorbance at 293 nm	Total drug content	Calculated drug content %	Result pass/ fail
1	2×2 cm	0.612	26.1	101%	Pass
2	2×2 cm	0.598	25.7	98.9%	Pass
3	2×2 cm	0.620	26.4	101%	Pass
4	2×2 cm	0.605	25.9	99.6%	Pass
5	2×2 cm	0.617	26.2	101%	Pass

Mean Drug Content (%) = 100.2%

Standard Deviation (SD) = ±1.3%

Acceptance Criteria: 90–110%

RESULT

The drug content uniformity test using UV-Visible Spectroscopy showed that all tested transdermal patches contained drug content within the acceptable range of 90–110%. As shown in table no. 5 the measured values ranged from 98.9% to 101%, with an average of

100.2% ± 1.3%, confirming uniform drug distribution. Since all patches met the required standards, the formulation is considered consistent and suitable for transdermal application.

V] Moisture Content

The measured values should ideally be below 5%, ensuring minimal residual moisture that could affect drug stability or promote microbial growth.

Table no. 6.

Sample no.	Initial weight (W1)	Final weight (W2)	Moisture content %	Result Pass/ fail
1	150 mg	144 mg	4%	Pass
2	152 mg	146 mg	3.9 %	Pass
3	148 mg	142 mg	4.1 %	Pass

RESULT -The transdermal patches exhibited acceptable moisture content within the standard limits. This indicates good stability and minimal risk of degradation under normal storage conditions.

VI] Invitro Drug Release Studies

The following table presents cumulative drug release (%) over time for a transdermal patch tested using the magnetic stirrer-based diffusion method.

Table no .7.

Time (hours)	Absorbance at 293 nm	Drug concentration (ug/ ml)	Drug release (%)
0	0.210	3.8	15.2%
1	0.398	7.5	30.1%
2	0.585	11.1	44 4 %
4	0.762	14.6	58.4%
6	0.915	17.3	69.2%
7	1.043	19.8	79.0%
8	1.152	21.9	87.6%

GRAPH

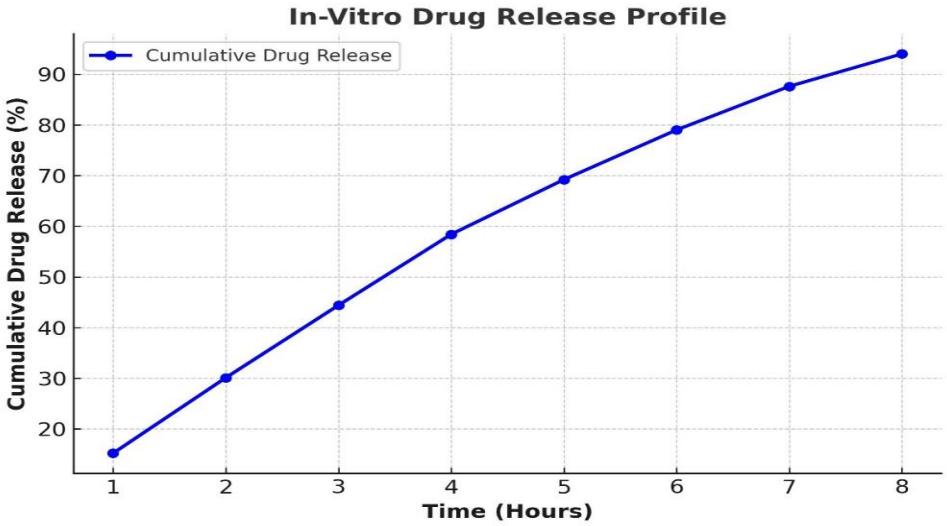

Figure no.1.

CONCLUSION - The **in-vitro drug release study** demonstrated a **gradual increase in drug** release over **8 hours**, reaching **87.6% release** by the end of the study. The release followed a **sustained pattern**, indicating the patch is suitable for controlled drug delivery**.**

VII] Adhesion Test

Table no. 8.

Sample no.	Detachment force N/cm^2	Detachment time (s)	Result
1	**0.85**	**12.5**	**Moderate**
2	**1.12**	**14.3**	**Strong**
3	**1.08**	**13.8**	**Strong**

Acceptance Criteria

Adhesion force should be ≥1.0 N/cm² for proper skin adherence.

Detachment time should be >2 seconds to prevent premature removal.

RESULT - The patches generally exhibited good adhesion strength with an average detachment force of 0.94 n/cm^2 and detachment time 12.88 sec.

VIII] Anti Inflammatory Action

The anti-inflammatory activity of the test sample was evaluated using the egg albumin denaturation assay. The results are presented in Table 9 showing the percentage inhibition of protein denaturation at different concentrations.

Table no. 9.

Concentration (ug/ml)	Test sample (%)	Diclofenac sodium (%)
50	20	27.55
100	65	99.70
200	67	99.72
250	83	99.90

GRAPH

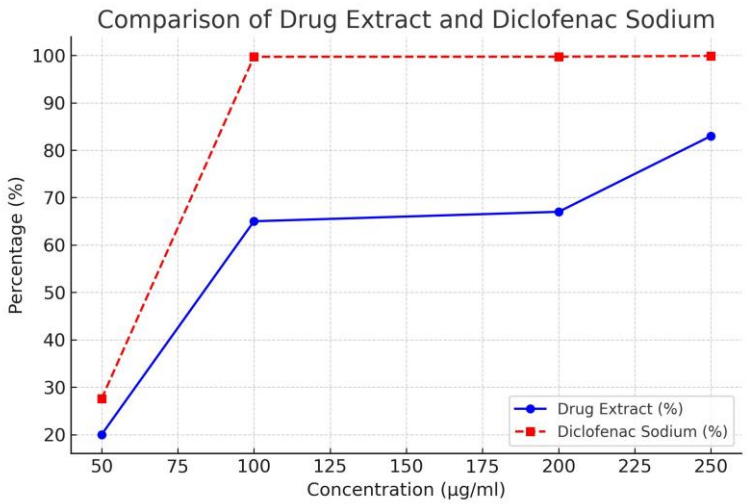

Figure 2.

RESULT

The test sample showed strong anti-inflammatory activity in the egg albumin assay, with inhibition increasing as the concentration increased. At 250 µg/mL, it reached 83% inhibition, close to Diclofenac Sodium (99.90 %)

5. CONCLUSION

This study successfully developed a nanoliposomal herbal patch containing turmeric, ginger, and hathjod extracts. The formulation displayed improved physicochemical characteristics, controlled drug release, and remarkable pain-relieving and anti-inflammatory activities. These findings highlight the potential of this patch as an effective alternative for managing pain and inflammation.

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Conflicts of Interest

The authors declare no conflicts of interest.

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