In-Vitro Study Of Formulation And Development Of A Transdermal Patches For Anti-Inflammation And Its Evaluation

Sachin Dimri[1*], Pallavi Sati[2], Mansi Nautiyal[3], Vijay Laxmi Trivedi[4] And Priyanka Bankoti[5], Jayanti Ballabh[6], Sunil Prakash[7], Gaurav Jain[8], And Neha Saini[9]

[1*]Sai College of Pharmacy, Dehradun-248001, Uttarakhand, India. Email: sachindimri04@gmail.com
[2] High Altitude Plant Physiology Research Centre, Hemvati Nandan Bahuguna Garhwal University (Central University), Srinagar-246174, Uttarakhand, India. Email: pallavisati45@gmail.com
[3]Department of Agriculture, Tula's Institute, Dehradun-248197, Uttarakhand, India. Email: mansinautiyal94@gmail.com
[4]High Altitude Plant Physiology Research Centre, Hemvati Nandan Bahuguna Garhwal University (Central University), Srinagar-246174, Uttarakhand, India. Email: vijaylaxmitrivedi@gmail.com
[5]School of Agricultural Sciences, Shri Guru Ram Rai University, Dehradun, 248001, Uttarakhand, India. Email: priyankabankoti28@gmail.com
[6]School of Agriculture, Uttaranchal University, Dehradun 248007, Uttarakhand, India. Email: jayantiballabh1987@gmail.com
[7]School of Agriculture, Uttaranchal University, Dehradun 248007, Uttarakhand; India Email: sonymt099@gmail.com
[8]School of Agriculture, Uttaranchal University, Dehradun 248007, Uttarakhand, India Email: gauravj888@gmail.com
[9]School of Agriculture, Uttaranchal University, Dehradun 248007, Uttarakhand, India. Email: neha.saini783@gmail.com

***Corresponding Author:** Sachin Dimri
Email: sachindimri04@gmail.com

Abstract
Conventional drug delivery system has many problems so bulk of research has now shifted from synthetic drugs to herbal drugs. This is possible because of the vast variety of bioactive molecules in the plants and their higher safety margin. This present study is focused on the use of *Adhatoda vasica* flower and *Ricinus Communis* leaves as an antioxidant agent. The main objective of present work was to develop the transdermal patch so that minimize the side effects and maximize the therapeutic efficacy. The formulations were also examined for organoleptic properties as well as other physicochemical features as thickness, weight homogeneity, folding durability, moisture content, and medication content.

Keywords: Transdermal patches, Herbal products, *Adhatoda vasica*, *Ricinus Communis*

Introduction

Drug delivery technologies are now receiving considerable attention from pharmaceutical companies. The main purpose of developing alternative drug delivery technologies is to increase efficiency and safety of drug and provide more convenience for the patient (Asbill et al., 2000). Substantial research conducted during the past several years has led to the development of technologies that meet the requisite criteria for delivering the drug through a non-invasive route. One of such technologies is transdermal drug delivery (TDD). It is a painless method of delivering drugs systemically by applying a drug formulation onto intact and healthy skin (Funke et al., 2002; Pastore et al., 2015; Al et al., 2019). The drug initially penetrates through the stratum corneum and then passes through the deeper epidermis and dermis without drug accumulation in the dermal layer. When drug reaches the dermal layer, it becomes available for systemic absorption via the dermal microcirculation. It can improve patient compliance due to the reduction of dosing frequencies and is also suitable for patients who are unconscious or vomiting, or those who rely on self-administration (Kunal et al., 2012; Ananda et al., 2021). The requirement for an inexpensive and non-invasive means of vaccination, especially in the developing world, has given rise to substantial research focused on the development of simple, needle-free systems such as TDD for vaccination purposes (Bird & Ravindra, 2020). Herbal transdermal patches are innovative products that deliver herbal or plant-based ingredients through the skin. They work by adhering to the skin's surface and gradually releasing these ingredients into the blood stream. They often contain natural ingredients like herbs, essential oils, and plant extracts known for their therapeutic properties. There are so many medicinal plants are reported for their pharmacological activities (Jamloki et al., 2022) such as *Hippophae salicifolia* (Trivedi et al., 2020), *Azadiracta indica*, *Dioscorea deltoidea*, *Curcuma longa* (Marrelli et al., 2021), *Aconitum heterophyllum* (Chandra et al., 2022), *Trillium govanianum* (Chandola et al., 2022). These plants are potential source of bioactive compounds and having various biological activities like, anti-carcinogenic, anti-diabetic, anti-helminthic, anti-oxidant and anti-inflammatory etc.

Adhatoda vasica and *Ricinus Communis* belongs to family Acanthaceae and Euphorbiaceae respectively, are known for their anti-inflammatory activities.
The goal of this study was to develop and test a transdermal patch that might be used to treat inflammation. Ingredients or an extract such as *Adhatoda vasica* flower and *Ricinus Communis* Leaves were included in the current formulation.

Materials and method
Plant Sample- Flower of *Adhatoda vasica* and leaves of *Ricinus Communis* were used in the study. The medicinal Plant species and identified and authentication from Botanical Survey of India, (BSI), Dehradun. Volatile oils from *Adhatoda vasica* flower and *Ricinus Communis* Leaves were extracted using Clevenger apparatus for the design and development of an Anti-Inflammation formulation as a novel dosage form. The equipment consisted of a 1000 mL round bottom flask attached to the separator, which automatically separated the oil from the distillate in a graduated tube, allowing a direct reading of the amount of oil collected. A condenser was connected to this separator. The temperature for the oil collection was kept at a moderate level (50–70 ^{0}C) and was modified throughout the experiment.

Preparation of Calibration Curve- Preparation of calibration curve prepared by using the method given by Vaddi et al. (2001). The dilution approach was used to create a standard curve. For the varied concentrations of 1, 2, 3, 4, and 5 μL/mL, a stock solution of 10 μL in 10 mL of methanol was utilised. At a wavelength of 304 nm, the absorbances of these solutions were measured in spectrophotometer.

Preparation and Optimization of Emulsion- Using tween 80 as a surfactant and ethanol as a cosurfactant, an oil-in-water (o/w) emulsion was formed. 75% *Adhatoda vasica* oil and 25% a blend of equal portions *Ricinus Communis* oil were used to make the oil phase. Sirka 5% solution was employed as the aqueous phase. Emulsions were made in various ratios of oil phase to surfactant to aqueous phase and stored at room temperature for a month to test their stability. The most stable emulsion was chosen for dosage form formulation after optimization.

Formulation of Anti-Inflammation Transdermal Patch- The transdermal patch was formulated by following the method of Roberts et al., 1993. Petri-dish having overall area of 50.24cm2 was utilized. The patch was made using the solvent evaporation method. Polymers were precisely pondered and a clear solution was formed by dissolving 10ml of water: ethanol (1:1) solution and 4% lactic acid solution. Later, 125 mg of chitosan was added gradually and dissolved with the help of a magnetic stirrer. After the chitosan had completely dissolved, 1 mL of PEG-400 (Polyethylene glycol) was added, followed by 1 mL of distilled water. The liquid was thoroughly mixed to produce a thick, homogenous solution. Further the obtained solution was allowed to cast on the interiors of petri-dish, which was lubricate with glycerine and dried at room temperature for 24hrs. The quick evaporation of the solvent was minimized using an inverted funnel which was placed over the Petri dish. On completion of mentioned time, dried out patches was isolated from petri-dish and kept in desiccator for further examinations (Figure 1).

Characterization of Developed Patches-
1. **Organoleptic Characteristics-**The prepared patch was physically examined for its facade, color, clearness, litheness, and smoothness.
2. **Thickness-** Vernier calipers was utilized to measure the thickness of the patches. Consistency in thickness was deliberated at various locations and mean values were computed.
3. **Weight Uniformity-** Uniformity in weight was determined by taking 3 patches weighed on digital balance and analyzed for distinctions in weight.
4. **Folding Endurance-** Developed patch was taken and subjected to repetitive folding at same point until it gets sever. Instances of time when patch folded without breaking was noted down.
5. **Moisture Content-** Developed patch after weighing was placed in a desiccator consisting compounded calcium chloride for 24hrs. Further after taking out from desiccators, patch was re-weighed. Under mentioned formula was used for compute % moisture content:

$$\text{Percentage of moisture content} = [\textit{Initial weight} - \textit{Final weight}]/\textit{Final weight} \times 100.$$

6. **Drug Content-** Drug content was analysed by dissolving patch in methanol and the residual volume was making up distilled water to 100mL. Further this solution was subjected to filtration and absorbance of the solution was notified at the wavelength of 304 nm, through which the concentration was measured (Kathe et al., 2017).

Results and Discussion
Organoleptic Characteristics- The organoleptic traits of herbal patch including appearance, colour, clarity, flexibility and smoothness id represented in table 1. It was jellified in appearance and pale Yellow in colour with slightly opaque clarity. However, the flexibility and smoothness of the preparation observed good and fair respectively. Herbal patches for inflammation are designed to deliver natural compounds through the skin to help alleviate inflammation and

associated discomfort (Vaddi et al., 2001; Pastore et al, 2015). These herbs can be combined or used individually in transdermal patches, allowing their active compounds to penetrate the skin and target the affected area directly, providing localized relief (Ananada et al., 2021). However, it's essential to consult a healthcare professional before using herbal patches, especially if you have underlying health conditions or are using other medications, to ensure safety and efficacy. In current formulation, we have observed it was jellified in appearance, pale yellow in colour, slightly opaque clarity, good flexibility and smoothness. These physical characteristics collectively contribute to the functionality, safety, and user satisfaction of transdermal herbal patches. The physical characteristics of transdermal herbal patches such as appearance, clarity and flexibility are crucial for their functionality, efficacy, and user experience (Kathe et al., 2017). Flexibility in the material ensures that the patch can conform to the contours of the skin, enhancing comfort for the user during wear. This characteristic is especially important for patches applied to areas that move frequently, like joints (Vaddi et al., 2001).

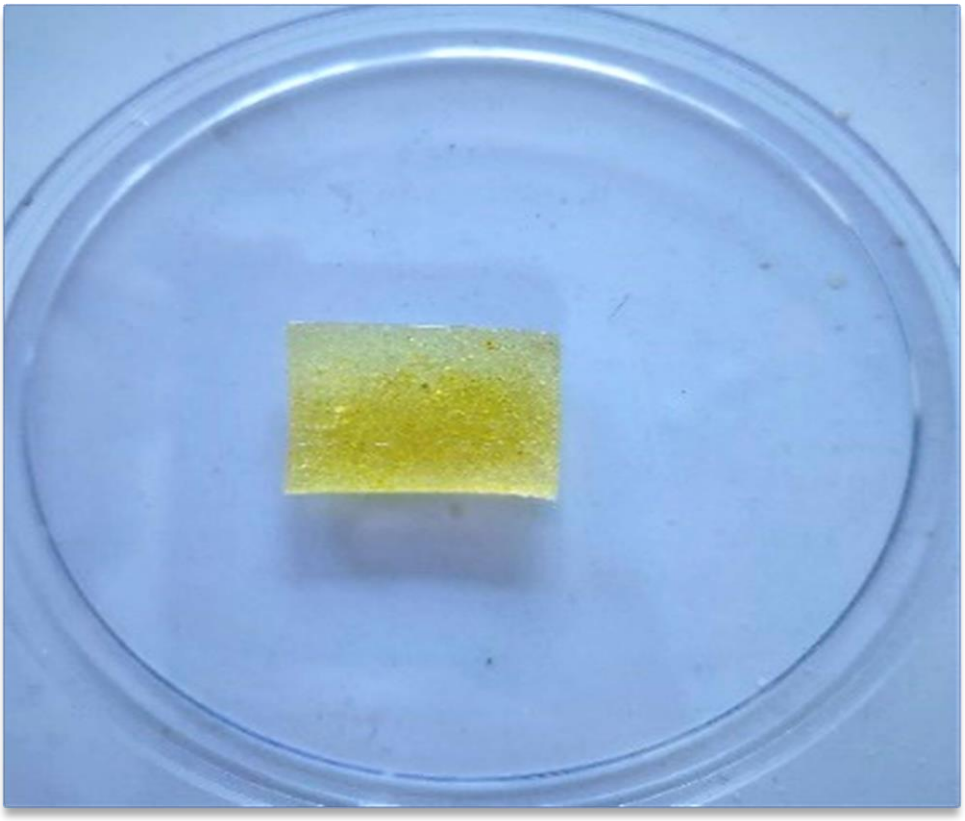

Figure 1: Calibration curve for formulated oil.

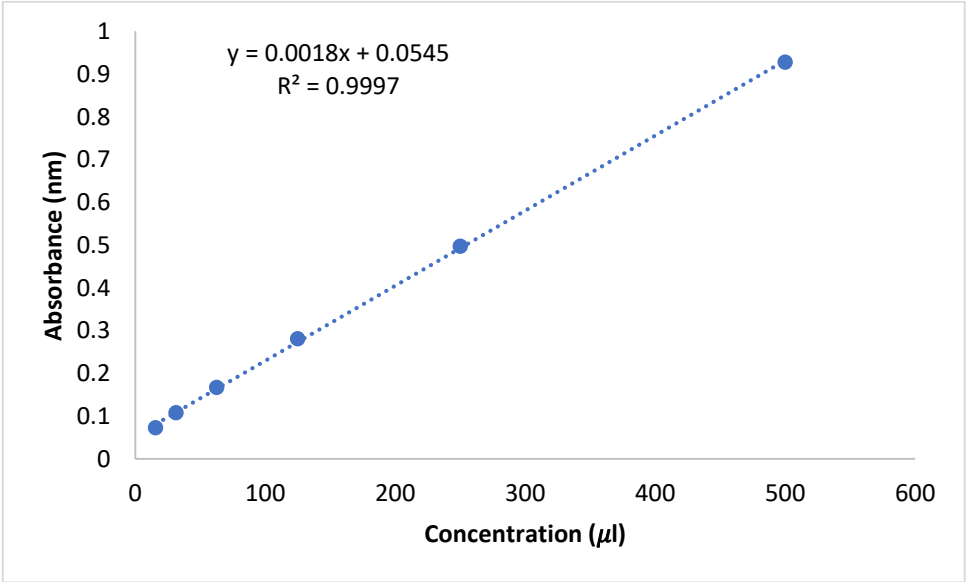

Figure 2: Prepared Transdermal Anti-Inflammation Patch Sample

S.No	*Physical Characteristic*	*Result*

1	Appearance	Jellified Preparation
2	Colour	Pale Yellow
3	Clarity	Slightly Opaque
4	Flexibility	Good
5	Smoothness	Good

Table 1. Results of Organoleptic property analysis of developed transdermal patches.

S.No	Thickness (mm)	Weight Uniformity (gm)	Folding endurance	Moisture content (%)	Drug Content (mg)
1	0.68	0.26	78	5.01	0.18
2	0.59	0.26	81	5.13	0.17
3	0.66	0.25	83	5.22	0.19
4	0.60	0.26	80.6	5.12	0.18
	0.63±04	0.26±0.10	80.65±2.05	5.12±0.08	0.18±01

Table 2- Results of thickness, weight uniformity, folding endurance, moisture content and drug content.

Other characteristics- Developed patches shown optimum thickness in the range of 0.60-0.68 mm. Results of weight uniformity depicted of mean weight of 0.26±0.10 gm whereas mean folding endurance of prepared patches was 80.65±2.05. The average moisture content of prepared patches was calculated to be 25.12±0.08 % and the content of drug was found to be 0.18±01 mg (Table 2). The materials used in the patch should be non-irritating to the skin. The patch should be smooth and appearance should be best for the skin. This helps prevent allergic reactions or adverse skin effects, ensuring the patch can be used safely by a wide range of individuals. The thickness and size of the patch can impact its comfort and ease of use. Thin patches are often preferred for discreetness, while the size might vary depending on the intended area of application (Vaddi et al., 2001; Pastore et al, 2015). Folding assurance, moisture content, drug content, weight uniformity, thickness and drug content of herbal patches also affect the quality of herbal patches. Folding assurance refers to the ability of the patch to withstand folding without damage. It's crucial for patches to maintain their structural integrity during handling, packaging, and application. Manufacturers perform tests to ensure that the patches can withstand folding as per specified standards without breaking or losing effectiveness (Ananda et al., 2021). Moisture content also important for the best quality of the herbal patches. Controlling moisture content is essential as excessive moisture can lead to microbial growth or degradation of active ingredients in the herbal patch. Maintaining a specific moisture level through proper manufacturing and packaging processes is crucial to preserve the patch's stability and efficacy. On the other hand, the drug content within the patch indicates the concentration of active herbal ingredients. It's crucial for the patch to contain the specified number of herbal compounds to ensure consistent therapeutic effects. Quality control measures verify the drug content to ensure it meets the intended dosage and efficacy (Vaddi et al., 2001; Ananda et al., 2021). Furthermore, uniformity in weight ensures consistent dosing across different patches. Variations in weight might lead to inconsistencies in the amount of active ingredients delivered, affecting the patch's efficacy. Quality checks assess weight uniformity to maintain consistent dosage in each patch (Anananda et al., 2021). These parameters are critical quality control checkpoints during the manufacturing process. Various testing methods, such as visual inspection, moisture analysis, content assays, and weight/thickness measurements, are employed to ensure herbal patches meet predefined standards for safety, efficacy, and consistency before they reach consumers.

Conclusion

The herbal transdermal patch of *Adhatoda vasica's* flowers and *Ricinus Communis's* leaves was prepared successfully by using different concentrations of extracted oil by solvent casting method. The research focused on the design and development of an anti-inflammatory transdermal formulation in a unique dosage form for a common clinical problem, such as abdominal discomfort, joint pain or stiffness, and redness. The patch was determined to be stable and free of skin irritation. Moreover, the transdermal patch has enormous potential to be used not only for the management of inflammatory reactions but also for other conditions requiring transdermal drug release. The present work can further be proceeding with in-vivo study on healthy animals to evaluate the pharmacokinetic profile.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

ACKNOWLEDGMENT:

This research is supported by Division of Research & Innovation, Uttaranchal University, Dehradun, India.

References

1. Al Hanbali, O. A., Khan, H. M. S., Sarfraz, M., Arafat, M., Ijaz, S., & Hameed, A. (2019). Transdermal patches: Design and current approaches to painless drug delivery. Acta Pharmaceutica, 69(2), 197-215.

2. Ananda, P. W. R., Elim, D., Zaman, H. S., Muslimin, W., Tunggeng, M. G. R., & Permana, A. D. (2021). Combination of transdermal patches and solid microneedles for improved transdermal delivery of primaquine. International Journal of Pharmaceutics, 609, 121204.
3. Asbill CS, Michniak BB. Percutaneous penetration enhancers: Local versus transdermal activity. Pharm Sci Technolo Today. 2000;3:36–41.
4. Bird, D., & Ravindra, N. M. (2020). Transdermal drug delivery and patches—An overview. Medical Devices & Sensors, 3(6), e10069.
5. Chandola, V., Chandra, S., Nautiyal, A. R., & Concenço, G. (2022). Antioxidant potential and impact of different extraction solvents on the free, esterified and insoluble-bound phenolics, flavonoid and tannin content of Trillium govanianum Wall ex D. Don, a rare Himalayan herb. Vegetos, 35(4), 953-960.
6. Chandra, S., Chandola, V., Sultan, Z., Singh, C. P., Purohit, V. K., Nautiyal, B. P., & Nautiyal, M. C. (2022). Climate change adversely affects the medicinal value of Aconitum species in Alpine region of Indian Himalaya. Industrial Crops and Products, 186, 115277.
7. Funke AP, Schiller R, Motzkus HW, Günther C, Müller RH, Lipp R. Transdermal delivery of highly lipophilic drugs: In vitro fluxes of antiestrogens, permeation enhancers, and solvents from liquid formulations. Pharm Res. 2002;19:661–8.
8. Jamloki, A., Trivedi, V. L., Nautiyal, M. C., Semwal, P., & Cruz-Martins, N. (2022). Poisonous plants of the Indian Himalaya: an overview. Metabolites, 12(6), 540.
9. Kathe K, Kathpalia H. Film forming systems for topical and transdermal drug delivery. AJPS. 2017; 6: 487-497.
10. Kunal M, Hetalk KP, Vishnu AP. Formulation and characterization of drug in adhesive transdermal patches of Diclofenac acid. Int J Pharm Pharm Sci. 2012;4:296–9.
11. Marrelli, M. (2021). Medicinal plants. Plants, 10(7), 1355.
12. Pastore, M. N., Kalia, Y. N., Horstmann, M., & Roberts, M. S. (2015). Transdermal patches: history, development and pharmacology. British journal of pharmacology, 172(9), 2179-2209.
13. Roberts, M.S. Walker, M., In Pharmaceutical Skin Penetration Enhancement, Walters, K.A. Hadgraft, J., Eds. Marcel Dekker: New York. 1993; pp. 1-30
14. Trivedi, V. L., Nautiyal, M. C., Sati, J., & Attri, D. C. (2020). Antioxidant enzyme activities in male and female plants of Hippophae salicifolia D. Don in different pheno-phases. Acta physiologiae plantarum, 42, 1-11.
15. Vaddi, H.K., Wang, L.Z., Ho, P.C., Chan, S.Y., 2001. Effect of some enhancers on the permeation of haloperidol through rat skin in vitro. Int. J. Pharm. 212, 247–255.