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RESEARCH ARTICLE

Development and Characterization of Synergistic Transdermal Patches Containing *Zingiber officinale* Extract and Diclofenac Sodium for Improved Anti-Inflammatory Action

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Abstract: A synergistic matrix-type transdermal drug delivery system containing a combination of *Zingiber officinale* (ginger) rhizome extract and diclofenac sodium was developed to improve therapeutic outcomes in inflammatory disorders while reducing the systemic adverse effects associated with conventional oral non-steroidal anti-inflammatory drugs. Matrix patches were prepared via the solvent casting technique using variable ratios of hydrophilic hydroxypropyl methylcellulose E15 and hydrophobic ethyl cellulose polymers, plasticized with propylene glycol. Physical evaluation of the developed batches showed excellent film uniformity, with thickness values ranging between 0.21±0.01 mm and 0.31±0.01 mm , and weight variations within acceptable pharmacopoeial limits. The folding endurance exceeded 240 folds across all formulations, indicating substantial mechanical flexibility, which was further substantiated by tensile strength assessments. Biophysical evaluations showed that formulations containing higher hydrophilic polymer proportions exhibited enhanced swelling indices and accelerated in vitro drug release dynamics. *In vitro* release studies using Franz diffusion cells showed a sustained and controlled drug permeation profile over twenty-four hours, governed predominantly by non-Fickian anomalous diffusion kinetics. *In vivo* pharmacological tests using carrageenan-induced paw edema and hot plate thermal nociception models in rats showed that the co-delivery of ginger extract and diclofenac sodium yielded a significantly higher inhibition of inflammatory edema and pain sensitivity at lower drug concentrations compared to patches containing the synthetic drug alone. These results confirm the synergistic potential of combining natural phytoconstituents with synthetic analgesics within a transdermal platform to establish an effective, stable, and biocompatible therapeutic system for chronic inflammatory diseases.

Keywords: Transdermal patches; Diclofenac sodium; Ginger extract; Synergistic anti-inflammatory; Matrix-type films.

1. Introduction

Chronic inflammatory diseases, such as rheumatoid arthritis, osteoarthritis, and various musculoskeletal disorders, are a major global health burden associated with debilitating pain, joint degradation, and progressive functional impairment [1]. The physiological cascades of inflammation involve the up-regulation of cyclooxygenase enzymes, particularly cyclooxygenase-2, leading to the excessive biosyntheses of pro-inflammatory prostaglandins that mediate tissue damage, swelling, and nociceptor sensitization [2]. Although non-steroidal anti-inflammatory drugs represent the primary therapeutic class employed to mitigate these symptoms, their prolonged systemic administration is severely limited by a high incidence of gastrointestinal erosion, peptic ulcerations, nephrotoxicity, hepatotoxicity, and adverse cardiovascular events [3]. These secondary pathological complications arise primarily from the systemic inhibition of cytoprotective prostaglandins in the gastric mucosa and renal parenchyma, as well as the sharp peak-and-trough plasma concentrations characteristic of conventional oral dosage forms [4]. There is an urgent clinical requirement for alternative drug delivery systems capable of maintaining steady-state therapeutic levels at the pathological site while minimizing systemic drug exposure and avoiding gastrointestinal contact.

Transdermal drug delivery systems have gained substantial interest as superior therapeutic systems capable of delivering active compounds directly through the skin barrier into the systemic circulation at a controlled rate [5]. Transdermal patches bypass the hepatic first-pass metabolism by using the skin as an entry portal, eliminate gastric mucosal irritation, reduce dosing frequency, and enhance patient compliance through non-invasive and painless application [6]. However, the outer layer of the epidermis, the stratum corneum, functions as a highly formidable physical barrier consisting of keratin-rich corneocytes embedded within a highly ordered intercellular lipid matrix [7]. To overcome this structural barrier and ensure sufficient therapeutic flux, formulations must be

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optimized utilizing appropriate polymeric matrices, plasticizers, and chemical penetration enhancers that temporarily modify the lipid packing of the stratum corneum [8].

The combination of bioactive herbal extracts with synthetic pharmacotherapy has emerged as a promising strategy to achieve synergistic therapeutic responses [9]. *Zingiber officinale*, commonly referred to as ginger, contains a rich array of bioactive non-volatile pungent phytoconstituents, including gingerols, shogaols, and zingerone, which possess potent anti-inflammatory, antioxidant, and analgesic properties [10]. These phytoconstituents exert their anti-inflammatory action by suppressing the activation of nuclear factor kappa β, thereby down-regulating the transcription of pro-inflammatory cytokines such as tumor necrosis factor-alpha, interleukin-1 β, and interleukin-6, while simultaneously inhibiting both cyclooxygenase and lipoxygenase pathways [11]. When combined with a potent synthetic agent like diclofenac sodium, the dual-action mechanism targets multiple nodes of the inflammatory cascade, facilitating a reduction in the required therapeutic dose of the synthetic drug and consequently lowering the risk of dose-dependent toxicities [12]. This research work includes the design, development, and evaluation of matrix-type transdermal patches containing a synergistic combination of diclofenac sodium and standardized *Zingiber officinale* extract.

2. Materials and Methods

2.1. Materials

Diclofenac sodium was obtained as a gift sample from modern pharmaceutical suppliers. Dried rhizomes of *Zingiber officinale* were procured from local specialized botanical repositories and authenticated. Hydroxypropyl methylcellulose E15 (HPMC E15, viscosity 15 cP for a 2% w/v aqueous solution at 20 °C) and ethyl cellulose (EC, ethoxyl content 48- 49%) were purchased from standard chemical suppliers. Propylene glycol (purity ≤ 99.5%) and analytical grade ethanol (99.9%) were obtained for use as plasticizers and solvents, respectively. Distilled water was prepared using an in-house Milli-Q purification system. All other reagents, chemicals, and solvents utilized throughout the analytical and biological experiments were of laboratory or analytical grade.

2.2. Extraction of Phytoconstituents from *Zingiber officinale* Rhizomes

The authenticated rhizomes of *Zingiber officinale* were subjected to thorough washing under running distilled water to remove adhering soil and extraneous particulate matter. The outer epidermal peel was carefully removed using sterilized stainless-steel instruments, and the remaining parenchymal tissue was sliced into uniform, thin segments. These segments were subjected to controlled shade drying at a temperature of 35±2 °C for a period of seven days until a constant dry weight was achieved. The dried rhizome slices were then pulverized into a coarse powder using an electric laboratory grinder and passed through a number forty mesh sieve to ensure particle size uniformity.

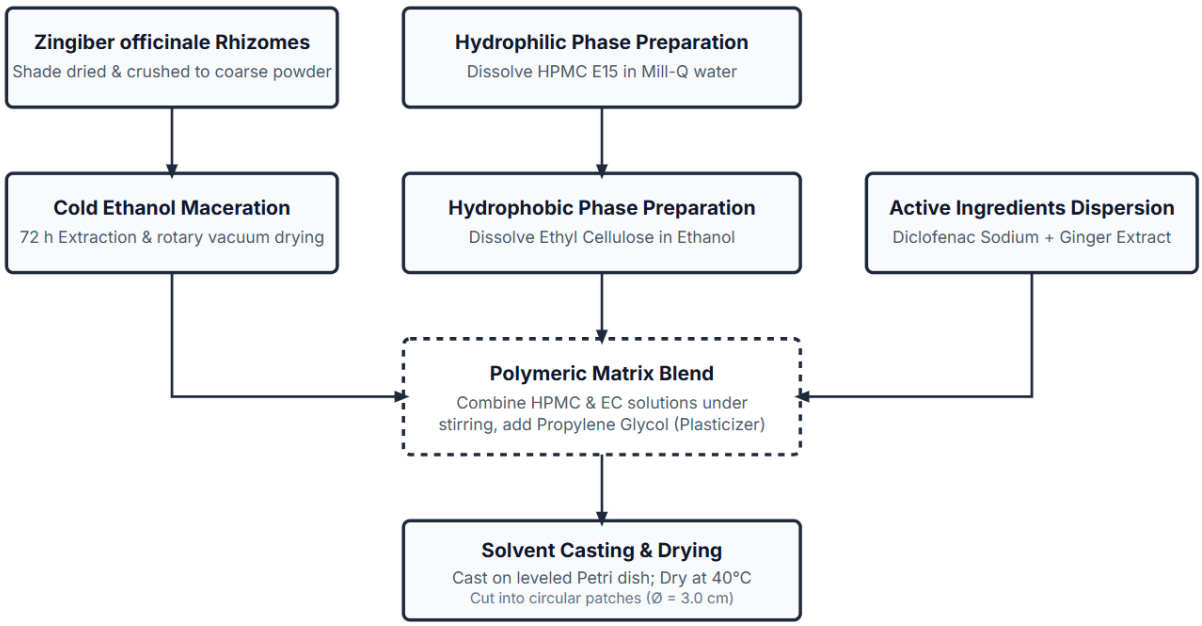

Figure 1. Process flow diagram for the synthesis of synergistic transdermal patches

The extraction was carried out using a cold maceration process to preserve thermolabile active phytoconstituents. An accurately weighed quantity of 100 g of the coarse ginger powder was placed into an airtight glass maceration vessel, to which 500 mL of analytical grade ethanol was added. The mixture was kept at room temperature for seventy-two hours with periodic manual agitation at regular intervals of six hours to facilitate maximum cellular lysis and extraction of active constituents. After the maceration period, the mixture was filtered through a triple-layered muslin cloth, followed by filtration through a Whatman number one filter paper under vacuum. The filtrate was collected and concentrated under reduced pressure using a rotary vacuum evaporator maintained at a controlled temperature of 40±1 °C. The concentrated syrupy mass was dried completely in a vacuum desiccator over anhydrous silica gel to yield a dark brown, semi-solid viscous extract, which was stored in airtight, amber-colored glass vials at 4 °C until further usage.

2.3. Preformulation and Physicochemical Studies

2.3.1. Melting Point and Thermal Transition Analysis

The melting point of pure crystalline diclofenac sodium was determined using the conventional capillary tube method in a calibrated digital melting point apparatus. A small quantity of the dry drug powder was tightly packed into a glass capillary tube sealed at one end and heated at a constant rate of 1 °C/min. For the complex crude extract of *Zingiber officinale*, which contains a heterogeneous mixture of volatile oils, resins, and polyphenols, a sharp crystalline melting point does not exist. Instead, the thermal behavior and physical state changes of the extract were characterized by examining the range of thermal softening, phase transition, and volatilization in a digital melting point apparatus, recording the temperature range spanning from initial softening to complete liquefaction.

2.3.2. Thermodynamic Solubility Studies

Thermodynamic solubility profiles of both diclofenac sodium and the dried *Zingiber officinale* extract were determined by adding an excess quantity of each material into glass vials containing 10 mL of different solvent media, specifically distilled water, methanol, ethanol, and phosphate buffer (pH 7.4). The vials were placed in a water bath shaker maintained at a constant temperature of 37±0.5 °C and agitated continuously for forty-eight hours to achieve thermodynamic equilibrium. After equilibrium, the samples were centrifuged at 5000 rpm for fifteen minutes. The supernatant was carefully decanted, filtered through 0.22 µm membrane filters, diluted appropriately with respective solvents, and analyzed spectrophotometrically. Diclofenac sodium concentration was measured at its characteristic absorption maximum (λ max) of 276 nm, while the total dissolved phytoconstituents of ginger extract were indexed qualitatively based on standard reference curves.

2.3.3. Qualitative Phytochemical Screening

To confirm the presence of the main anti-inflammatory classes within the extracted *Zingiber officinale* mass, qualitative phytochemical screening tests were performed. These screening tests were conducted on the reconstituted extract solutions using standard colorimetric and precipitation protocols.

The Mayer's test was conducted by treating 2 mL of the extract solution with a few drops of Mayer's reagent (potassium mercuric iodide), where the formation of a cream-colored precipitate indicated the presence of alkaloids. The Wagner's test was performed by reacting 2 mL of the extract with Wagner's reagent (iodine in potassium iodide), where the appearance of a reddish-brown precipitate confirmed the presence of alkaloidal bases. Phenolic compounds and tannins were evaluated using the ferric chloride test, in which the addition of three drops of freshly prepared 5% w/v ferric chloride solution to the extract produced a blue-black or green coloration. Flavonoids were identified using both the lead acetate test, where the addition of lead acetate solution yielded a yellow precipitate, and the alkaline reagent test, which involved treating the extract with dilute sodium hydroxide solution to produce an intense yellow color that became colorless upon the addition of dilute hydrochloric acid.

2.4. Formulation of Matrix-Type Transdermal Patches

Matrix-type transdermal patches containing diclofenac sodium and *Zingiber officinale* extract were fabricated using the solvent casting technique. The polymeric matrix was designed using HPMC E15 as the hydrophilic polymer and ethyl cellulose as the hydrophobic polymer in varying weight ratios to control drug release kinetics.

To prepare the polymeric casting solutions, the designated quantity of HPMC E15 was weighed and dissolved in 10 mL of distilled water under continuous stirring at 500 rpm using a magnetic stirrer, allowing the polymer to hydrate and swell completely to form a clear, bubble-free viscous solution. Concurrently, the specified quantity of ethyl cellulose was dissolved in 10 mL of analytical grade ethanol with constant stirring to obtain a uniform polymeric solution. The HPMC E15 aqueous solution and the ethyl cellulose organic solution were then blended together slowly under constant stirring to produce a homogeneous hydroalcoholic polymeric mixture.

Following the blending of the polymers, propylene glycol was added to the mixture as a plasticizer and permeation enhancer at a constant concentration of 20% w/w of the total polymer weight, and stirring was maintained for thirty minutes to ensure uniform distribution of the plasticizer within the polymeric network. In a separate step, 25 mg of diclofenac sodium and 20 mg of the dry ginger extract were dissolved in 5 mL of a hydroalcoholic solvent mixture (consisting of ethanol and water in a 1:1 volume ratio) and subjected to mild sonication for five minutes. This active drug-extract solution was then added dropwise to the plasticized polymer mixture under continuous mechanical stirring at 800 rpm for one hour, yielding a completely uniform casting fluid.

The casting fluid was kept stationary for two hours to allow for the complete escape of entrapped air bubbles. The bubble-free mixture was then poured carefully into a leveled glass Petri dish with an inner diameter of 9 cm (giving a total casting surface area of approximately 63.6 cm^2). The cast films were dried in a hot air oven maintained at a controlled temperature of 40±2°C for twenty-four hours to facilitate slow and uniform evaporation of the solvents. After drying, the transparent, flexible polymeric films were carefully peeled from the glass surface. The films were cut into circular patches of 3 cm diameter (equivalent to a surface area of 7.07 cm^2) using a sharp circular die cutter. Each cut patch was designed to contain a nominal dose equivalent to 25 mg of diclofenac sodium. The prepared patches were wrapped in aluminum foil and stored in a desiccator containing anhydrous silica gel until evaluation. The precise compositions of the formulation batches (F1 to F5) are shown in Table 1.

Table 1. Quantitative formulation batches of synergistic transdermal patches containing ginger extract and diclofenac sodium

Formulation	Diclofenac Sodium (mg)	Ginger Extract (mg)	HPMC E15 (mg)	Ethyl Cellulose (mg)	Propylene Glycol (mL)	Solvent Mixture Ethanol:Water (mL)
F1	25	20	100	50	0.03	25
F2	25	20	125	50	0.035	25
F3	25	20	150	50	0.04	25
F4	25	20	100	75	0.035	25
F5	25	20	125	75	0.04	25

2.5. Evaluation of Patches

2.5.1. Thickness and Weight Variation

The thickness of three randomly selected patches from each formulation batch was measured at five different anatomical points (one central and four peripheral locations) using a digital micrometer with an accuracy of ±0.001 mm. The average thickness and standard deviation were calculated to ensure batch uniformity. For weight uniformity, ten patches from each batch were weighed individually using a calibrated high-precision analytical balance. The mean weight and standard deviation were determined, ensuring that deviations remained well within the limits defined by pharmacopoeial specifications.

2.5.2. Folding Endurance and Surface pH

The mechanical flexibility of the transdermal patches was evaluated by determining their folding endurance. This was performed by repeatedly folding a single strip of the patch (2 x 2 cm^2) manually at the same central crease line until it either developed visible cracks or broke completely. The maximum number of folds achieved before structural failure was recorded as the folding endurance value.

The surface pH of the patches was assessed to evaluate their biocompatibility with the skin surface. Individual patches were allowed to swell in contact with 1 mL of double-distilled water in sealed glass vials for one hour at room temperature. The electrode of a micro-electrode digital pH meter was placed in direct contact with the swollen patch surface, and the pH value was recorded after stabilization.

2.5.3. Moisture Content and Moisture Uptake Dynamics

To determine the percentage of residual moisture present in the formulations, three patches from each batch were weighed individually to establish their initial wet weight (Wi). These patches were then placed in a laboratory desiccator containing active, dry fused calcium chloride at room temperature. After twenty-four hours, the patches were removed and weighed again to obtain their dry weight (Wd). The percentage moisture content was calculated using the following equation:

$$\text{Moisture Content}(\%) = \left[\frac{W_i - W_d}{W_i}\right] \times 100$$

For moisture uptake studies, three pre-weighed patches (Wd) were exposed to a high-humidity environment to assess their hygroscopic nature. The patches were placed inside a sealed desiccator containing a saturated solution of potassium chloride, which maintained a constant relative humidity of approximately 84% at 25±1 °C. After twenty-four hours, the patches were taken out, wiped gently with filter paper to remove surface moisture, and weighed to determine their final wet weight (Ww). The percentage moisture uptake was calculated as:

$$\text{Moisture Uptake}(\%) = \left[\frac{W_w - W_d}{W_d}\right] \times 100$$

2.5.4 Tensile Strength and Percentage Elongation

The mechanical strength and elasticity of the transdermal films were characterized by measuring their tensile strength and percentage elongation using a custom-designed pulley-based tensile testing apparatus. Rectangular strips of the cast films measuring 4 x 1 cm^2 were prepared. One end of the strip was held flat in a fixed upper clamp, while the opposite end was secured in a movable lower clamp connected to a weight hanger via a frictionless pulley system. Weights were added gradually to the hanger at a constant rate of 10 g/sec until the film broke. The force required to cause film rupture was recorded, and the tensile strength was calculated using the following formula:

$$\text{Tensile Strength}\left(\text{kg/cm}^2\right) = \frac{\text{Break Force(kg)}}{\text{Cross-sectional Area of Strip}(\text{cm}^2)}$$

The percentage elongation at the point of break was evaluated by measuring the distance between the two reference marks placed on the strip before applying the force. The initial length (L0) was compared with the final length at rupture (Lt), and the percentage elongation was calculated using the equation:

$$\text{Percentage Elongation}(\%) = \left[\frac{L_t - L_0}{L_0}\right] \times 100$$

2.5.4. Swelling Index and Matrix Erosion

The swelling kinetics of the transdermal matrix patches were evaluated to understand the hydration and gel-forming behavior of the polymers. Pre-weighed dry patches (Wd) were placed on a fine stainless-steel mesh basket and immersed in 20 mL of phosphate buffer (pH 7.4) at 37±0.5 °C. At predetermined intervals of 5, 10, 15, 30, 45, and 60 minutes , the mesh basket containing the swollen patch was removed, blotted gently with lint-free filter paper to remove excess surface water, and weighed immediately (Ws). The swelling index was calculated as:

$$\text{Swelling Index}(\%) = \left[\frac{W_s - W_d}{W_d}\right] \times 100$$

2.5.5. Drug Content Uniformity

To determine the concentration and spatial homogeneity of diclofenac sodium in each batch, single patches of 3 cm diameter were placed in 100 mL volumetric flasks containing 50 mL of methanol. The flasks were placed in an ultrasonic bath and sonicated continuously for forty-five minutes to facilitate complete dissolution of the polymeric matrix and extraction of the incorporated drug. The volume was then adjusted to 100 mL using phosphate buffer (pH 7.4), and the solution was filtered through a 0.45 µm syringe filter. Aliquots of the filtrate were diluted appropriately with phosphate buffer (pH 7.4), and the absorbance was measured spectrophotometrically at 276 nm against a blank patch extract. The percentage drug content was calculated from the standard calibration curve of diclofenac sodium.

2.5.6. In Vitro Drug Release and Permeation Dynamics

The *in vitro* skin permeation and release kinetics of diclofenac sodium from the developed transdermal formulations were investigated using a vertical Franz diffusion cell system with an effective diffusion area of 3.14 cm^2 and a receptor compartment capacity of 15 mL. A high-grade cellophane dialyzing membrane (molecular weight cut-off 12,000 Da), which had been pre-soaked in phosphate buffer (pH 7.4) for twelve hours, was mounted securely between the donor and receptor compartments of the diffusion cell.

The receptor compartment was filled with freshly prepared phosphate buffer (pH 7.4), which was maintained at a physiological temperature of 37±0.5 °C and stirred continuously at 300 rpm using a magnetic bar to prevent concentration polarization. A single transdermal patch of each batch was placed flat on the upper surface of the dialyzing membrane in the donor compartment, ensuring intimate contact without any air bubbles. The top of the donor cell was sealed with parafilm to prevent moisture loss.

At specific time intervals of 0.5, 1, 2, 4, 6, 8, 12, and 24 hours, aliquots of 1 mL were withdrawn from the sampling port of the receptor compartment. Each sample was immediately replaced with an equal volume of temperature-equilibrated, fresh phosphate buffer (pH 7.4) to maintain sink conditions. The collected samples were filtered, and the concentration of diclofenac sodium was determined spectrophotometrically at 276 nm. The cumulative percentage of drug released was calculated and plotted against time.

To analyze the physical mechanisms governing drug release from the swelling matrix, the *in vitro* release data were fitted to various mathematical models, including Zero-order, First-order, Higuchi square-root, and Korsmeyer-Peppas models. The mathematical expressions used were:

$$Q_t = K_0 \cdot t$$

$$\ln(100 - Q_t) = \ln(100) - K_1 \cdot t$$

$$Q_t = K_H \cdot t^{0.5}$$

$$\frac{Q_t}{Q_\infty} = K_P \cdot t^n$$

Where Qt represents the cumulative percentage of drug released at time t, K0, K1, KH, and KP are the respective release rate constants, and n is the diffusional exponent that indicates the underlying transport mechanism.

2.5.7. In Vivo Biocompatibility and Skin Irritation Study

To evaluate the safety and skin tolerability of the optimized transdermal patches, an *in vivo* skin irritation study was conducted on healthy Wistar rats weighing 180 - 220 g. The animals were housed in standard laboratory conditions with free access to food and water. The experimental protocols were conducted in strict accordance with institutional animal ethics guidelines.

The dorsal hair of the rats was carefully shaved over an area of approximately 4 cm^2 using electric clippers, taking care not to damage the epidermal barrier. The animals were divided into three groups of six rats each: Group I served as the untreated control; Group II received a topical application of a standard chemical irritant solution containing 0.8% w/v formalin; and Group III was treated with the optimized synergistic transdermal patch (F3) applied directly to the shaved dorsal skin.

The patch was held in place using non-sensitizing surgical adhesive tape for forty-eight hours. The application sites were examined visually at regular intervals of 24, 48, and 72 hours to detect signs of dermal toxicity, specifically erythema (redness) and edema (swelling). Dermal changes were scored on a scale ranging from zero to four based on severity, where a score of zero represented complete absence of skin irritation and a score of four indicated severe erythema and edema.

2.5.8. In Vivo Pharmacological Evaluation of Analgesic and Anti-Inflammatory Activity

The synergistic *in vivo* pharmacological performance of the co-delivered ginger extract and diclofenac sodium from the optimized transdermal patch was evaluated using animal models.

The anti-inflammatory efficacy was assessed using the carrageenan-induced hind paw edema model in rats. The animals were divided into four experimental groups of six rats each: Group I was the inflammatory control (untreated); Group II was treated with a plain transdermal patch containing only 25 mg of diclofenac sodium; Group III was treated with a plain transdermal patch containing only 20 mg of standardized ginger extract; and Group IV was treated with the optimized synergistic combination patch F3 (containing both 25 mg diclofenac sodium and 20 mg ginger extract).

Thirty minutes prior to inducing inflammation, the respective patches were applied to the shaved dorsal skin of the rats. Acute inflammation was then induced by injecting 0.1 mL of a sterile 1% w/v carrageenan solution in normal saline into the sub-plantar aponeurosis of the right hind paw of each rat. The volume of the inflamed paw was measured at 0, 1, 3, and 5 hours after carrageenan injection using a digital plethysmometer. The percentage inhibition of paw edema was calculated using the following formula:

$$\text{Edema Inhibition}(\%) = \left[1 - \frac{V_t}{V_c}\right] \times 100$$

Where Vt is the mean paw edema volume in the treated group and Vc is the mean paw edema volume in the control group.

The systemic analgesic activity was evaluated using the hot plate thermal nociception method in mice weighing 25 -- 30 g. The animals were placed on an electronically controlled laboratory hot plate maintained at a constant temperature of 55±0.5 °C. The reaction time, defined as the latency period before the animal showed nociceptive behaviors such as paw licking or jumping, was recorded. A maximum cut-off time of fifteen seconds was maintained to prevent thermal tissue damage.

Baseline latency was determined for all animals prior to patch application. The mice were then divided into four groups corresponding to those used in the paw edema study, and the respective patches were secured on their shaved skin. The reaction time was measured again at 30, 60, 120, and 180 minutes after patch application to evaluate the onset and duration of analgesic efficacy.

2.5.9. Accelerated Stability Protocols

Accelerated stability studies were conducted on the optimized transdermal formulation batch according to International Council for Harmonisation (ICH) Q1A(R2) guidelines. The patches were wrapped in aluminum foil and packed in heat-sealed laminated pouches. The packages were placed inside an environmental stability chamber maintained at a temperature of 40±2 °C and a relative humidity of 75±5% for a duration of three months.

Samples were withdrawn at monthly intervals and evaluated for physical appearance, folding endurance, surface pH, drug content uniformity, and *in vitro* drug release. Visual inspections were performed to identify any signs of film degradation, color changes, drug crystallization, or loss of adhesion.

3. Results and Discussion

3.1. Preformulation

The preformulation melting point analysis of pure crystalline diclofenac sodium yielded a sharp melt transition at 283.5±0.5 °C , confirming the purity of the drug candidate. In contrast, the thermal analysis of the crude *Zingiber officinale* extract did not show a sharp melting point. Instead, it showed a broad thermal softening and phase transition range spanning from 82.4 °C to 88.1 °C. This behavior is attributed to the complex amorphous nature of the extract, which contains volatile essential oils, non-volatile gingerols, shogaols, and resinous substances that undergo gradual thermal softening and continuous mass loss due to the volatilization of essential oils at elevated temperatures.

The thermodynamic solubility studies, summarized in Table 2, showed that diclofenac sodium has high solubility in methanol (142.15±3.24 mg/mL) and phosphate buffer pH 7.4 (84.62±1.95 mg/mL), but only low solubility in distilled water (4.12±0.32 mg/mL). Conversely, the standardized *Zingiber officinale* extract was freely soluble in absolute ethanol (215.40±5.60 mg/mL) and methanol, but only moderately soluble in phosphate buffer (12.45±0.84 mg/mL). These solubility results guided the selection of a hydroalcoholic solvent system (ethanol-water 1:1 ratio) to ensure complete molecular dispersion of both active components within the polymeric blend during patch preparation.

Table 2. Thermodynamic equilibrium solubility of active components in different solvent systems at 37±0.5 °C

Solvent Medium	Diclofenac Sodium Solubility (mg/mL)	Ginger Extract Solubility (mg/mL)
Distilled Water	4.12 ± 0.32	1.85 ± 0.14
Methanol	142.15 ± 3.24	168.20 ± 4.12
Ethanol	62.45 ± 1.80	215.40 ± 5.60
Phosphate Buffer pH 7.4	84.62 ± 1.95	12.45 ± 0.84

(mean ± SD, n = 3)

The qualitative phytochemical screening of the *Zingiber officinale* extract confirmed the presence of key bioactive anti-inflammatory compound classes. The results of these screening tests are summarized in Table 3.

Table 3. Qualitative phytochemical screening profile of the standardized *Zingiber officinale* extract

Phytochemical Test	Procedure	Visual Observation	Inference
Mayer's Test	Reconstitution + Mayer's Reagent	Formation of cream-colored precipitate	Alkaloids Present
Wagner's Test	Reconstitution + Wagner's Reagent	Formation of reddish-brown precipitate	Alkaloids Present
Ferric Chloride Test	Reconstitution + 5% w/v $FeCl_3$	Development of deep blue-green coloration	Phenols and Tannins Present
Lead Acetate Test	Reconstitution + 10% w/v Lead Acetate	Formation of yellow precipitate	Flavonoids Present
Alkaline Reagent Test	Reconstitution + NaOH solution	Intense yellow color, which becomes colorless with acid	Flavonoids Present

3.2. Physicochemical and Mechanical Optimization

The formulated matrix patches (F1 to F5) were obtained as smooth, flexible, and cohesive films with no visible cracks, phase separation, or drug crystallization. This indicates excellent physical compatibility among the drug, extract, HPMC E15, and ethyl cellulose. The biophysical and mechanical parameters of all formulations are summarized in Table 4.

Table 4. Comprehensive biophysical and mechanical evaluation data of the transdermal patches

Formulation	Weight (mg)*	Thickness (mm) *	Folding Endurance*	Surface pH*	Moisture Content (%)*	Moisture Uptake (%)*	Tensile Strength (kg/cm ^{2})*	Elongation (%)*	Drug Content (%)*
F1	118 ± 2	0.21 ± 0.01	245 ± 5	6.4 ± 0.1	2.12 ± 0.12	3.45 ± 0.18	0.48 ± 0.03	18.2 ± 0.5	94.21 ± 0.8
F2	122 ± 3	0.24 ± 0.02	268 ± 4	6.6 ± 0.1	2.35 ± 0.15	3.89 ± 0.22	0.54 ± 0.04	21.4 ± 0.6	96.45 ± 0.6
F3	126 ± 2	0.26 ± 0.01	290 ± 3	6.7 ± 0.2	2.48 ± 0.14	4.12 ± 0.20	0.62 ± 0.03	24.8 ± 0.4	98.12 ± 0.5
F4	130 ± 3	0.29 ± 0.02	312 ± 5	6.8 ± 0.1	2.71 ± 0.11	4.56 ± 0.25	0.78 ± 0.05	27.1 ± 0.5	99.04 ± 0.4
F5	134 ± 2	0.31 ± 0.01	328 ± 4	6.9 ± 0.1	2.84 ± 0.10	4.98 ± 0.28	0.85 ± 0.04	29.8 ± 0.4	99.36 ± 0.3

(mean ± SD, n = 3)

The mean weight of the patches ranged from 118±2 mg (F1) to 134±2 mg (F5), reflecting the increase in total polymer weight in the formulations. The patch thickness values were highly uniform, ranging between 0.21±0.01 mm and 0.31±0.01 mm. This level of thickness uniformity is essential to ensure a consistent drug concentration per unit surface area and to achieve reproducible drug release across the skin.

The folding endurance values ranged from 245±5 to 328±4 folds. These high values indicate that the films possess excellent mechanical durability and resistance to tearing during patient movement. The high folding endurance is attributed to the uniform distribution of the propylene glycol plasticizer, which inserts between the polymer chains, reduces intermolecular forces, and increases the free volume and molecular mobility of the polymeric network.

The surface pH of all formulations was close to neutral, ranging from 6.4±0.1 to 6.9±0.1. This range is highly compatible with the physiological pH of human skin (4.5 - 6.5), indicating that the patches are unlikely to cause skin irritation or discomfort when applied.

The percentage of residual moisture content remained low, between 2.12±0.12% and 2.84±0.10%. This low moisture content is desirable because it helps prevent microbial growth and maintains the physical integrity and stability of the patches during storage. The percentage moisture uptake was also controlled, ranging from 3.45±0.18% to 4.98±0.28%. This low moisture uptake behavior prevents the patches from becoming excessively sticky or liquefying when exposed to high-humidity environments. Both moisture parameters showed a slight increase as the concentration of hydrophilic HPMC E15 was increased. This trend is due to the highly hydrophilic nature of HPMC, which contains numerous free hydroxyl groups that can readily form hydrogen bonds with water molecules.

Tensile strength and percentage elongation are key indicators of a patch's mechanical durability and elasticity. The tensile strength of the formulations increased from 0.48±0.03 kg/cm ^{2} (F1) to 0.85±0.04 kg/cm ^{2} (F5). This increase is directly correlated with the higher proportion of the hydrophobic polymer ethyl cellulose, which forms a dense, rigid, and cohesive polymeric network. The

percentage elongation also increased from 18.2±0.5% to 29.8±0.4%, indicating that the patches have sufficient elasticity to withstand the mechanical stresses of skin stretching and body movement without rupture.

The drug content analysis showed high uniformity across all formulation batches, with values ranging from 94.21±0.8% to 99.36±0.3%. These results confirm that the solvent casting technique and the chosen hydroalcoholic solvent system achieved a highly uniform dispersion of diclofenac sodium and ginger extract throughout the polymer matrix.

3.3. Swelling Behavior

The swelling behavior of transdermal patches is the factor governing drug release, as the hydration and subsequent swelling of the polymer matrix create channels for drug diffusion. The swelling kinetics of batches F1 to F5 are shown in Table 5.

Table 5. Cumulative swelling kinetics profile of the formulated transdermal patches

Time (minutes)	F1 Swelling (%)	F2 Swelling (%)	F3 Swelling (%)	F4 Swelling (%)	F5 Swelling (%)
0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
5	12.4 ± 0.8	15.6 ± 0.9	18.9 ± 1.1	10.2 ± 0.6	13.4 ± 0.8
10	22.5 ± 1.1	28.4 ± 1.2	34.2 ± 1.5	18.6 ± 0.9	24.1 ± 1.2
15	31.8 ± 1.4	39.5 ± 1.6	48.6 ± 2.1	26.4 ± 1.3	33.5 ± 1.5
30	45.2 ± 1.9	54.1 ± 2.0	68.4 ± 2.4	38.9 ± 1.7	47.2 ± 1.9
45	52.4 ± 2.1	62.8 ± 2.3	79.5 ± 2.6	44.1 ± 1.8	54.8 ± 2.1
60	54.1 ± 2.0	64.5 ± 2.1	82.1 ± 2.8	45.2 ± 1.6	56.2 ± 2.0

**(mean ± SD, n = 3).*

Formulation F3, which contains the highest ratio of HPMC E15 to ethyl cellulose, exhibited the highest swelling capacity, reaching a swelling index of 82.1±2.8% at sixty minutes. This high swelling rate is due to the rapid hydration of HPMC's hydrophilic cellulose chains, which quickly swell to form a highly porous hydrogel matrix.

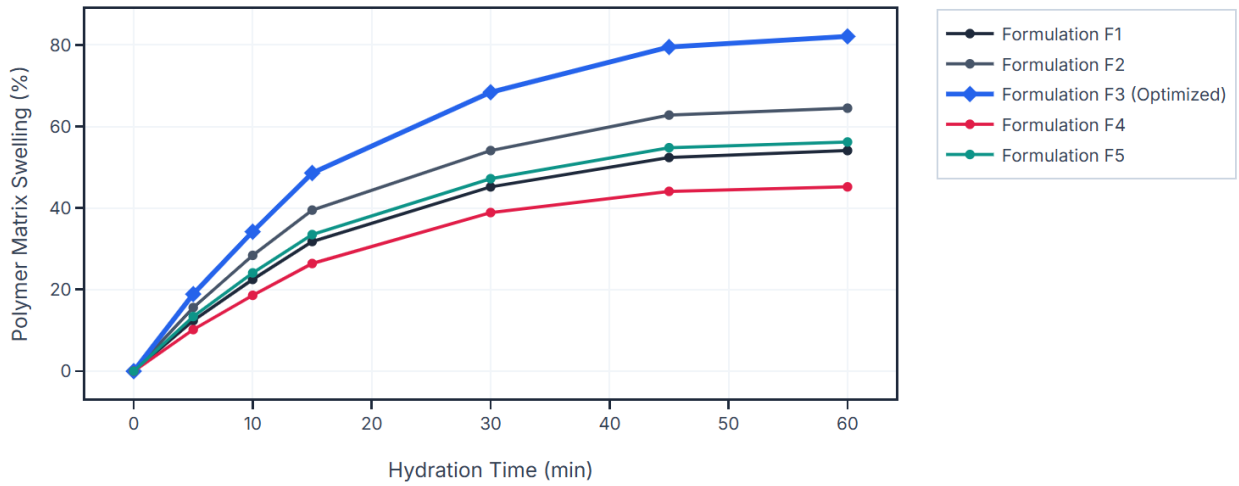

Figure 2. Swelling Behavior of formulations F1–F5 in phosphate buffer (pH 7.4) over 60 min

In contrast, formulations F4 and F5 showed lower swelling capacities, with swelling indices of 45.2±1.6% and 56.2±2.0%, respectively, at sixty minutes. This reduced swelling is due to the higher concentration of hydrophobic ethyl cellulose in these formulations. Ethyl cellulose acts as a physical barrier to water penetration, reinforcing the matrix and preventing rapid hydration and expansion of the HPMC polymer chains.

3.4. *In Vitro* Drug Release and Permeation Dynamics

The *in vitro* release and skin permeation of diclofenac sodium from formulations F1 to F5 was monitored over twenty-four hours using Franz diffusion cells. The cumulative release profiles are shown in Table 6.

Table 6. *In vitro* cumulative percentage drug release of diclofenac sodium from formulation batches F1–F5

Time (hours)	Cumulative Release (%)*				
	F1	F2	F3	F4	F5
0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
0.5	14.2 ± 0.8	16.8 ± 0.9	19.5 ± 1.1	10.4 ± 0.6	12.8 ± 0.7
1.0	25.4 ± 1.2	28.9 ± 1.4	32.4 ± 1.6	18.5 ± 1.1	21.9 ± 1.3
2.0	39.8 ± 1.6	44.1 ± 1.9	49.8 ± 2.2	30.2 ± 1.5	35.4 ± 1.8
4.0	55.1 ± 2.1	61.2 ± 2.3	68.4 ± 2.5	45.8 ± 1.9	51.2 ± 2.2
6.0	68.4 ± 2.4	74.5 ± 2.5	82.1 ± 2.8	58.1 ± 2.2	64.5 ± 2.4
8.0	78.5 ± 2.6	85.6 ± 2.8	92.4 ± 2.9	68.4 ± 2.4	74.8 ± 2.6
12.0	86.4 ± 2.9	91.2 ± 2.7	97.8 ± 1.8	77.5 ± 2.7	83.9 ± 2.8
24.0	92.1 ± 2.4	95.4 ± 1.8	99.1 ± 0.8	85.4 ± 2.1	91.2 ± 1.9

(mean ± SD, n = 3)

All formulations showed a biphasic release profile, characterized by an initial rapid drug release during the first two hours, followed by a sustained and controlled release over the remaining twenty-four hours. The initial rapid release phase is attributed to the dissolution of drug molecules located near the patch surface, while the sustained release phase is controlled by the slow diffusion of drug molecules through the hydrated polymer matrix.

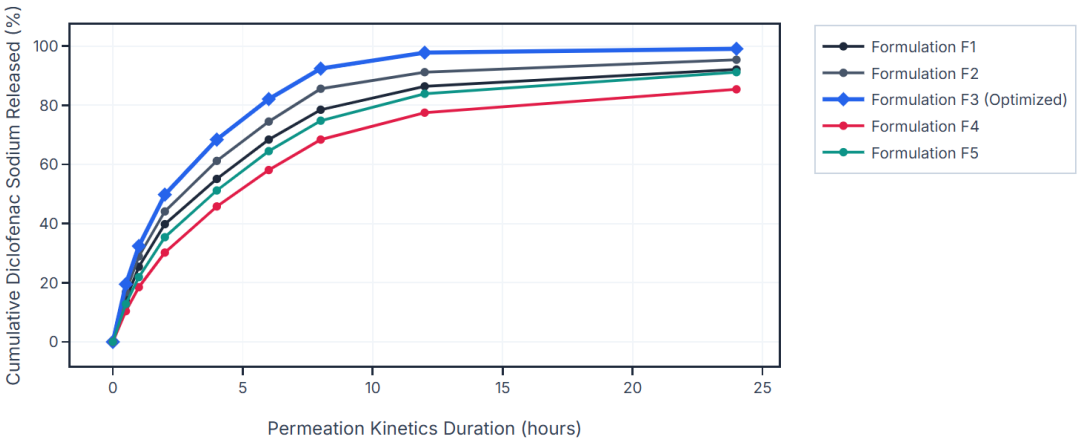

Figure 3. Cumulative permeation curves through synthetic membranes over 24 h using Franz diffusion cell

Formulation F3 showed the most complete drug release, reaching 99.1±0.8% cumulative release at twenty-four hours. This high release rate is due to F3's high proportion of hydrophilic HPMC E15, which rapidly hydrates and swells to form a porous gel layer that allows for fast drug diffusion.

In contrast, formulation F4, which contains a higher proportion of hydrophobic ethyl cellulose, showed a slower release profile, reaching only 85.4±2.1% cumulative release at twenty-four hours. The hydrophobic nature of ethyl cellulose limits water penetration into the patch, reducing the hydration rate of the polymer matrix and slowing down the diffusion of drug molecules. To understand the mechanisms controlling drug release, the in vitro release data were analyzed using several mathematical models. The resulting correlation coefficients (R^2) and diffusion exponents (n) are presented in Table 7.

Table 7. Mathematical modeling of *in vitro* drug release kinetics.

Formulation	Zero-order (R^2)	First-order (R^2)	Higuchi (R^2)	Korsmeyer-Peppas (R^2)	Peppas Exponent (n)	Primary Release Mechanism
F1	0.8845	0.9412	0.9854	0.9912	0.62	Anomalous Transport
F2	0.8612	0.9324	0.9812	0.9935	0.65	Anomalous Transport
F3	0.8415	0.9102	0.9785	0.9954	0.68	Anomalous Transport
F4	0.9125	0.9654	0.9912	0.9921	0.58	Anomalous Transport
F5	0.8954	0.9521	0.9884	0.9942	0.61	Anomalous Transport

For all formulations, the Higuchi and Korsmeyer-Peppas models showed the highest correlation coefficients ($R^2 > 0.97$). This indicates that drug release from these matrix patches is primarily diffusion-controlled. The calculated Korsmeyer-Peppas diffusion exponent (n) values ranged between 0.58 and 0.68. In matrix-type systems, an n value between 0.45 and 0.89 indicates anomalous, non-Fickian transport. This suggests that the drug release is governed by a combination of drug diffusion through the swollen polymer network and the slow relaxation and erosion of the polymer matrix.

3.5. *In Vivo* Biocompatibility and Skin Irritation Studies

The *in vivo* skin irritation and biocompatibility study was conducted on Wistar rats to evaluate the safety of the optimized synergistic transdermal formulation (F3). The skin irritation scoring results over seventy-two hours are presented in Table 8.

Table 8. *In vivo* skin irritation scoring in Wistar rats

Treatment Group	Erythema Score (24 h)*	Edema Score (24 h)*	Erythema Score (48 h)*	Edema Score (48 h)*	Erythema Score (72 h)*	Edema Score (72 h)*
Group I (Untreated Control)	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
Group II (Formalin Standard)	3.2 ± 0.4	2.8 ± 0.3	3.5 ± 0.3	3.1 ± 0.4	2.4 ± 0.5	2.1 ± 0.4
Group III (Optimized Patch F3)	0.0 ± 0.0	0.0 ± 0.0	0.1 ± 0.1	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0

**(mean ± SD, n = 6)*

The Group II rats treated with the standard irritant formalin showed severe erythema and moderate edema at twenty-four and forty-eight hours, indicating a significant inflammatory response. In contrast, Group III rats treated with the optimized synergistic patch F3 showed no signs of edema and only negligible erythema (score of 0.1±0.1) at forty-eight hours, which completely resolved by seventy-two hours. These results indicate that the polymers, plasticizers, and active ingredients used in the transdermal patches are biocompatible and safe for long-term topical application.

3.6. *In Vivo* Pharmacological Efficacy

3.6.1. Anti-Inflammatory Performance in Paw Edema Model

The synergistic anti-inflammatory efficacy of the co-delivered ginger extract and diclofenac sodium from the optimized patch (F3) was evaluated using the carrageenan-induced paw edema model in rats. The paw volume and percentage inhibition results are presented in Table 9.

The inflammatory control group (Group I) showed a rapid and sustained increase in paw volume, reaching 1.56±0.07 mL at five hours post-carrageenan injection.

Table 9. *In vivo* anti-inflammatory performance using carrageenan-induced paw edema in rats

Treatment Group	Paw Volume (0 h, mL)*	Paw Volume (1 h, mL)*	Paw Volume (3 h, mL)*	Paw Volume (5 h, mL)*	Edema Inhibition at 5 h (%)*
Group I (Inflammatory Control)	0.62 ± 0.02	1.15 ± 0.05	1.48 ± 0.06	1.56 ± 0.07	—
Group II (Plain Diclofenac Patch)	0.61 ± 0.03	0.98 ± 0.04	1.12 ± 0.04	1.04 ± 0.05	44.68 ± 1.85
Group III (Plain Ginger Patch)	0.63 ± 0.02	1.04 ± 0.05	1.28 ± 0.05	1.22 ± 0.06	25.53 ± 1.12
Group IV (Synergistic Patch F3)	0.62 ± 0.02	0.85 ± 0.03	0.88 ± 0.03	0.81 ± 0.04	69.15 ± 2.10

**(mean ± SD, n = 6)*

Treatment with patches containing only diclofenac sodium (Group II) or only ginger extract (Group III) resulted in moderate anti-inflammatory activity, achieving 44.68±1.85% and 25.53±1.12% inhibition of paw edema, respectively, at five hours.

In comparison, the group treated with the synergistic combination patch F3 (Group IV) showed a significantly higher reduction in paw volume, achieving 69.15±2.10% inhibition of paw edema at five hours ($p < 0.01$). This enhanced anti-inflammatory activity shows the synergistic effect of combining diclofenac sodium with ginger extract. The gingerols and shogaols in the ginger extract target pro-inflammatory cytokines and oxidative stress pathways, while diclofenac sodium acts via the cyclooxygenase pathway. Together, they provide a more comprehensive inhibition of the inflammatory cascade.

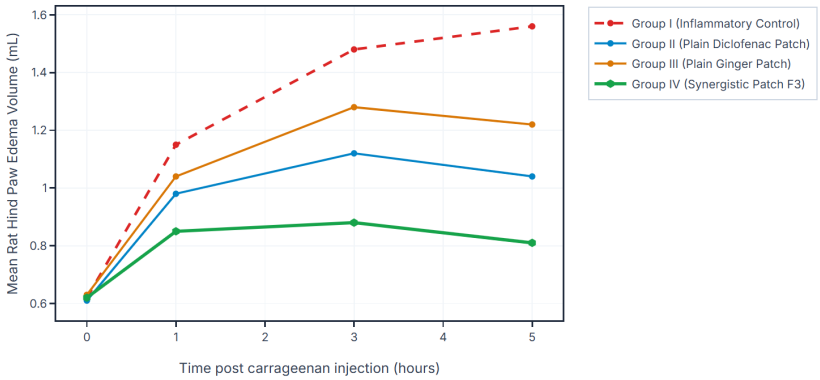

Figure 4. Synergistic *in vivo* anti-inflammatory activity in carrageenan paw edema models

3.6.2. Analgesic Performance in Thermal Nociception Model

The systemic analgesic activity was evaluated using the hot plate method in mice, measuring the delay in reaction time to thermal pain. The latency results are summarized in Table 10.

Table 10. *In vivo* analgesic activity using the hot plate thermal pain model in mice

Treatment Group	Baseline Latency (s)*	Latency at 30 min (s)*	Latency at 60 min (s)*	Latency at 120 min (s)*	Latency at 180 min (s)*
Group I (Control)	4.15 ± 0.25	4.22 ± 0.31	4.18 ± 0.28	4.09 ± 0.33	4.12 ± 0.29
Group II (Plain Diclofenac Patch)	4.12 ± 0.31	5.89 ± 0.42	7.15 ± 0.48	7.85 ± 0.52	7.45 ± 0.50
Group III (Plain Ginger Patch)	4.18 ± 0.28	4.95 ± 0.35	5.62 ± 0.41	6.12 ± 0.39	5.85 ± 0.44
Group IV (Synergistic Patch F3)	4.16 ± 0.29	7.12 ± 0.51	9.85 ± 0.62	11.45 ± 0.71	10.95 ± 0.68

**(mean ± SD, n = 6)*

The untreated control group showed no significant change in latency times throughout the study, remaining at approximately 4.12 - 4.22 seconds. The plain ginger patch (Group III) and plain diclofenac patch (Group II) groups showed moderate increases in latency, reaching maximum values of 6.12±0.39 seconds and 7.85±0.52 seconds , respectively, at one hundred and twenty minutes.

The group treated with the synergistic combination patch F3 (Group IV) showed a rapid onset and a significantly greater increase in pain threshold, with latency times reaching 11.45±0.71 seconds at one hundred and twenty minutes. This marked increase in pain latency confirms that co-delivering ginger extract and diclofenac sodium produces a strong synergistic analgesic effect, providing superior pain relief compared to the individual therapies.

3.7. Accelerated Stability Profiling

The optimized synergistic formulation F3 was subjected to accelerated stability testing at 40±2°C and 75±5% relative humidity for three months. The evaluation results are presented in Table 11.

The stability studies showed that the optimized transdermal patches maintained their physical integrity and performance characteristics over the three-month period. No visible changes, such as color fading, film cracking, polymer degradation, or drug crystallization, were observed.

Table 11. Accelerated stability evaluation of the optimized synergistic transdermal formulation F3

Evaluation Parameter	Baseline (0 Months)	1 Month	2 Months	3 Months
Physical Appearance	Smooth, Transparent	No Change	No Change	No Change
Folding Endurance	290 ± 3	288 ± 4	285 ± 5	282 ± 4
Surface pH	6.7 ± 0.2	6.7 ± 0.1	6.6 ± 0.2	6.6 ± 0.1
Drug Content (%)	98.12 ± 0.5	97.95 ± 0.6	97.42 ± 0.8	96.85 ± 0.7
Cumulative Release at 24 h (%)	99.1 ± 0.8	98.8 ± 1.1	98.2 ± 1.2	97.5 ± 1.4

**(mean ± SD, n = 6)*

The mechanical properties remained stable, with only minor, statistically insignificant decreases in folding endurance (from 290±3 to 282±4 folds). The surface pH and drug content also remained highly stable, with the final drug content at three months being 96.85±0.7%, representing a negligible degradation loss. The *in vitro drug* release profile did not change significantly, showing a cumulative release of 97.5±1.4% at twenty-four hours after three months of storage. These results indicate that the optimized formulation is physically and chemically stable, with a robust shelf-life under accelerated environmental conditions.

4. Conclusion

Stable and flexible matrix-type transdermal patches containing a synergistic combination of diclofenac sodium and standardized *Zingiber officinale* extract were developed using a solvent casting technique. The incorporation of a hydrophilic polymer (HPMC E15) and a hydrophobic polymer (ethyl cellulose) allowed for controlled, sustained drug release over twenty-four hours, governed by anomalous, non-Fickian diffusion. The optimized formulation (F3) showed excellent mechanical properties, uniform thickness, and a skin-compatible pH. *In vivo* pharmacological evaluations in animal models showed that combining the natural anti-inflammatory components of ginger extract with the synthetic NSAID diclofenac sodium produced a significantly higher inhibition of inflammatory paw edema and a greater increase in pain thresholds compared to patches containing only the synthetic drug. This hybrid transdermal delivery system could be a promising, stable, and biocompatible therapeutic platform for the long-term management of chronic inflammatory pain, offering improved therapeutic efficacy while reducing the dosage and systemic side effects of synthetic drugs.

References

[1] Chatterjee S, Banerjee A, Chandra R. Hemidesmus indicus: A rich source of herbal medicine. Medicinal and Aromatic Plants. 2014; 3(4): 169–173.

[2] Chaitanya S, Chippada SS, Volluri SR, Bammidi M. *In-vitro* anti-inflammatory activity of methanolic extract of Centella asiatica by HRBC membrane stabilisation. Biosciences Biotechnology Research Asia. 2011; 8(1): 321–326.

[3] Chou CT. The anti-inflammatory effect of an extract of *Tripterygium wilfordii* Hook F on adjuvant-induced paw edema in rats. Phytotherapy Research. 1997; 11: 152–154.

[4] Khemasili K, Widodo MA, Sanarto S, Setyawati K. *In vitro* and *in vivo* anti-inflammatory activities of *Coptosapelta flavescens* Korth root's methanol extract. Journal of Applied Pharmaceutical Science. 2018; 8(9): 102–108.

[5] Muhammad KN, Swandari P. Membrane stabilization activity as anti-inflammatory mechanisms of *Vernonia amygdalina* leaves extracts. International Conference on Tropical Studies and its Applications. 2017; November: 245–252.

[6] Oyedapo O, Akinpelu A. Red blood cell membrane stabilizing potentials of extracts of *Lantana camara* and its fraction. International Journal of Plant Physiology and Biochemistry. 2010; 2(4): 46–51.

[7] Rajendran V, Lakshmi KS. *In vitro* and *in vivo* anti-inflammatory activity of leaves of *Symplocos cochinchinensis* Moore ssp. laurina. Bangladesh Journal of Pharmacology. 2008; 3: 121–124.

[8] Rosa MD, Giround JP, Willoghby DA. Studies of the acute inflammatory response induced in rats in different sites by carrageenan and turpentine. Journal of Pathology. 1971; 104(1): 15–23.

[9] Sadique J, Al-Rqobah WA, Bughaith MF, El-Gindy AR. The bioactivity of certain medicinal plants on the stabilization of RBC membrane system. Fitoterapia. 1989; 60: 525–532.

[10] Sakat S, Juvekar AR, Gambhire MN. *In vitro* anti-oxidant and anti-inflammatory activity of methanol extract of *Oxalis corniculata* Linn. International Journal of Pharma and Phytochemical Sciences. 2010; 2(1): 146–155.

[11] Shenoy S, Shwetha K, Prabhu K, Maradi R, Bairy KL, Shanbhag T. Evaluation of anti-inflammatory activity of *Tephrosia purpurea* in rats. Asian Pacific Journal of Tropical Medicine. 2010; 3(3): 193–195.

[12] Kumar V, Bhat ZA, Kumar D, Bohra P, Sheela S. *In-vitro* anti-inflammatory activity of leaf extracts of *Basella alba* Linn. var. alba. International Journal of Drug Development and Research. 2011; 3: 124–127.

[13] Yurugasan N, Vember S, Damodharan C. Studies on erythrocyte membrane IV: *In vitro* hemolytic activity of Oleander extract. Toxicology Letters. 1981; 8: 33–38.

[14] Mizushima Y, Kobayashi M. Interaction of anti-inflammatory drugs with serum proteins, especially with some biologically active proteins. Journal of Pharmacy and Pharmacology. 1968; 20: 169–173.

[15] Vane JR, Botting RM. New insights into the mode of action of anti-inflammatory drugs. Inflammation Research. 1995; 44(1): 1–10.

[16] Winter CA, Risely EA, Nuss GW. Carrageenan-induced edema in hind paw of the rat as an assay for anti-inflammatory. Proceedings of the Society for Experimental Biology and Medicine. 1962; 111: 544–547.

[17] Patil KR, Mahajan UB, Unger BS, Goyal SN, Belemkar S, Surana SJ, Ojha S, Patil CR. Animal models of inflammation for screening of anti-inflammatory drugs: Implications for the discovery and development of phytopharmaceuticals. International Journal of Molecular Sciences. 2019; 20: 4367–4378.

[18] Mao QQ, Xu XY, Cao SY, Gan RY, Corke H, Beta T, Li HB. Bioactive compounds and bioactivities of ginger (*Zingiber officinale* Roscoe). Foods. 2019; 8: 185–203.

[19] Black CD, Herring MP, Hurley DJ, O'Connor PJ. Ginger (*Zingiber officinale*) reduces muscle pain caused by eccentric exercise. Journal of Pain. 2010; 11: 894–903.