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Original Research Article

Formulation of Transfersome Containing *Kaempferia galanga* for Transdermal Patch Application

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ABSTRACT

Kaempferia galanga L. is commonly used for its analgesic and anti-inflammatory properties. However, its therapeutic effectiveness is hindered by low solubility and poor skin permeability, which necessitates improvements in drug delivery systems. This study focused on formulating a transfersome containing *Kaempferia galanga* extract (KGE) by varying the ratios of phospholipid and surfactant, and incorporating it into a transdermal patch. Transfersomes were prepared using the thin film hydration method with different soy lecithin-to-Tween 80 ratios and were characterized to identify the optimal formulation. This formulation was then integrated into a polymeric transdermal patch through the solvent casting method. Comprehensive evaluations, including physicochemical characterization and *ex vivo* skin permeation studies, were performed on the KGE transfersome-loaded patch. The optimal transfersome, with a soy lecithin-to-Tween 80 ratio of 80:20, exhibited a particle size of 152.85 ± 6.88 nm, good deformability at 98.76 ± 2.14%, high entrapment efficiency of 89.33 ± 3.35%, and a spherical morphology. In the *ex vivo* skin permeation study, the KGE transfersome-loaded patch demonstrated a higher cumulative drug permeation of ethyl p-methoxycinnamate (EPMC), the primary active component of *Kaempferia galanga*, compared to the KGE-loaded patch. These results indicate that the KGE transfersome-loaded patch has potential as an effective alternative for transdermal delivery.

Keywords: Deformability, *Kaempferia galanga*, Osteoarthritis, Transdermal, Transfersome

Introduction

Kaempferia galanga, commonly known as kencur, is a medicinal plant extensively used in Indonesia for its analgesic, anti-inflammatory, and antipyretic properties. Its primary bioactive compound, ethyl p-methoxycinnamate (EPMC), contributes to these pharmacological effects.[1] Recent clinical research indicates that *Kaempferia galanga* may be beneficial in treating osteoarthritis (OA), a degenerative joint disease marked by chronic inflammation and decreased mobility. In a double-blind, randomized clinical trial, participants who took 160 mg/day of *Kaempferia galanga* extract for ten days reported significant improvements in pain, stiffness, and physical function, with results comparable to those achieved with meloxicam.[2] As of 2020, the global prevalence of osteoarthritis was estimated at 7.6%, according to the Global Burden of Disease Study, and this figure is expected to rise, particularly among older adults.[3] Currently, OA management is largely dependent on non-steroidal anti-inflammatory drugs (NSAIDs), which, while moderately effective, can lead to serious side effects such as gastrointestinal, cardiovascular, and renal complications, especially in elderly patients with existing health issues.[4] These safety concerns highlight the urgent need for safer, well-tolerated alternatives, such as herbal therapies with established anti-inflammatory properties.

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The therapeutic potential of *Kaempferia galanga* extract is limited by its low solubility and poor skin permeability, which pose challenges for systemic delivery. The stratum corneum, with its dense lipid matrix, serves as a significant barrier to drug permeation, restricting the transdermal delivery of many bioactive compounds.[5] To address these limitations, alternative delivery systems are necessary to enhance bioavailability and improve therapeutic outcomes. Transfersomes, or elastic liposomes, present a promising solution for increasing dermal penetration. Made from phospholipids and surfactants, transfersomes are highly flexible, allowing them to navigate the narrow intercellular spaces of the stratum corneum. This makes them particularly effective for delivering therapeutic agents aimed at alleviating inflammation and pain associated with osteoarthritis. Transfersomes also offer higher bioavailability compared to traditional oral routes.[6,7] Their biocompatibility, ability to encapsulate various drug types, and improved skin penetration position them as ideal carriers for transdermal therapy.[8,9]

The efficiency of transfersomes is influenced by the lipid bilayer composition, particularly the ratio of phospholipids to surfactants, which affects vesicle stability, deformability, and drug entrapment.[10,] To improve usability and therapeutic adherence, transfersomes can be incorporated into transdermal patches, creating an innovative drug delivery platform for OA treatment. These transfersome-based patches facilitate sustained drug release, targeted delivery, and enhanced patient compliance.[12] Recent studies highlight the clinical effectiveness of traditional transdermal systems, such as the Thai herbal patch (Ya-Pok-Dud-Pid), which has been shown to reduce pain and inflammatory markers in knee OA patients.[13] A similar approach using niosomal vesicle-based transdermal patches loaded with lornoxicam has demonstrated improved drug entrapment and superior permeation compared to conventional patches, indicating its potential as a novel drug delivery option for OA management.[14] Furthermore, magnetophoresis-enhanced transdermal patches delivering ibuprofen have shown faster and more significant reductions in pain and functional impairment in individuals with knee osteoarthritis, while minimizing adverse effects and improving user tolerability.[15]

Consequently, transfersome patches could enhance skin permeability and therapeutic efficacy while addressing critical safety concerns in long-term OA management. This study aims to optimize the transfersome formulation of *Kaempferia galanga* extract by adjusting the lecithin-to-surfactant ratio, characterize its physicochemical properties, and develop it into a transdermal patch for potential OA therapy.

Materials and Methods

Plant Collection and Identification

The *Kaempferia galanga* rhizomes used in this study were sourced from cultivated plants grown in agroforestry systems in Blitar, East Java, Indonesia, consistent with our previous research (GPS: 8°08'44.3"S 112°20'13.2"E), which was collected in June 2022.[16] The botanical identity of the plant material was verified by the UPT Laboratory of Herbal Balai Materia Medika in Batu, Indonesia, under voucher number 074/400/102.20-A/2022 by Achmad Mabrur, S.KM., M.Kes, and deposited at the Balai Materia Medika, Batu.

Reagents and Instruments

Reagents and excipients were procured from CV Duta Jaya in Malang, Indonesia, including soy lecithin, Tween 80, aquadest, chloroform, and phosphate-buffered saline (PBS). Additional materials included EPMC (Cat. No. M1204) from Tokyo Chemical Industry (Tokyo, Japan), Water for Injection (WFI) from PT. Ikapharmindo Putramas (Jakarta, Indonesia), HPLC-grade methanol (Cat. No. A4524) and acetonitrile (Cat. No. A998-4) from Fisher Scientific (Fair Lawn, NJ, USA), a 0.45 µm nylon syringe filter (Cat. No. S13NY045E) from Microlab Scientific Co., Ltd. (Yueqing, China), and a 0.2 µm membrane filter (Cat. No. A351860101) from Meissner Filtration Products, Inc. (Camarillo, CA, USA).

The instruments utilized in this study included an overhead stirrer (RW 20 digital, IKA, Germany), rotary evaporator (RV 10 basic, IKA, Germany), oven (ED 53, Binder, Germany), sonicator (M2800, Branson Ultrasonic, Emerson, Japan), pH meter (HM-30R, TOA-DKK, Japan), scanning electron microscope (SEM; FEG 650, FEI Quanta, USA), centrifuge (Fresco 21, Thermo Scientific, USA), digital caliper (RoHS), analytical balance (OHAUS PioneerTM, USA), moisture analyzer (Simadzu MOC 63, Japan), digital force gauge (ZP-200N, Imada, Japan), particle size analyzer (Flex 11, Microtrac Japan), and high-performance liquid chromatography (HPLC; LC-2030C 3D Plus, Shimadzu, Japan).

Plant Extraction

The extract was prepared through maceration with 96% ethanol as the solvent. Three hundred grams of dried *Kaempferia galanga* powder were macerated in 3 liters of 96% ethanol. The mixture was stirred for 30 minutes at 340-350 rpm using an overhead stirrer, then left covered to macerate at room temperature for 24 hours. After this period, the mixture was filtered, and two additional maceration cycles were performed using the same method to enhance extraction efficiency. All macerates were combined and concentrated in a rotary evaporator at 40°C and 50 rpm until a thick extract was formed. The concentrate was then dried in a hot air oven at 50°C for three days to produce the final extract. The extract is stored in the refrigerator until needed, yielding approximately 10.89% of *Kaempferia galanga* extract (KGE).

Preparation of Transfersome

Transfersomes were prepared using a modified thin film hydration method, with some modifications from a previous study,[17] as shown in Figure 1. Soy lecithin, serving as the phospholipid, was dissolved in chloroform, while Tween 80, the surfactant, was dissolved in an equal volume of methanol (1:1). These components were mixed in three different ratios, detailed in Table 1. The extract was added to a beaker containing the lecithin and surfactant mixture. This mixture was then transferred to a dry round-bottom flask, and the organic solvent was evaporated using a rotary evaporator under reduced pressure, starting at 25 rpm and increasing to 250 rpm to create a thin lipid film. Any remaining solvent was removed by allowing the flask to sit at room temperature for 24 hours. The dried lipid film was hydrated with 50 mL of PBS (pH 7.4) by rotating at 100 rpm at room temperature for 30 minutes. The resulting vesicles were allowed to swell for 2 hours at room temperature. For further size reduction, the transfersome suspension was sonicated for 30 minutes and then stored in refrigeration at 4°C for subsequent characterization. Each transfersome formulation was prepared in triplicate.

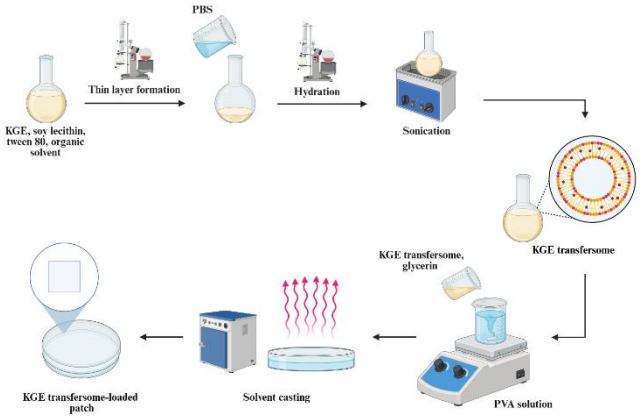


Figure 1: Formulation of KGE transfersome-loaded transdermal patch by the thin-film hydration method and the solvent casting method. The image was created using Biorender.

Table 1: The Formula of KGE-loaded Transfersome

Formula Code	Concentration (mg)		
	Soy Lecithin	Tween 80	KGE
F1 (90:10)	540	60	200
F2 (85:15)	510	90	200
F3 (80:20)	480	120	200

KGE: *Kaempferia galanga* extract

Characterization of Transfersomes

pH Measurement

The pH of the transfersome suspension was measured with a digital pH meter. After calibrating the instrument, the electrode was immersed in the suspension to obtain the pH readings for each formula, which were then recorded.[9]

Particle Size, Polydispersity Index, and Zeta Potential

Transfersomes were characterized for particle size, polydispersity index (PDI), and zeta potential using a particle size analyzer at Yogyakarta State University, Indonesia. All tests were conducted in triplicate.[11]

Vesicle Morphology

The vesicle morphology test was performed using a Scanning Electron Microscope (SEM).[7] A drop of the sample was applied to a cover glass, which was then dried and examined under the SEM at an accelerating voltage of 10 kV.

Deformability Index

The deformability index was measured using the extrusion method. A total of 5 mL of transfersome suspension was passed through a polycarbonate membrane with a pore size smaller than the suspension (100 nm) using a syringe while applying constant manual pressure. Vesicle size was assessed before and after extrusion using particle size analyzer. The deformability index of the transfersome suspension was calculated using the following equation:[18,19]

$$\text{Deformability index } (D) = J\left(\frac{rv}{rp}\right)^2 \quad \ldots (1)$$

Where D represents the deformability index, J is the volume of

suspension extruded in 15 minutes, *rv* is the vesicle size after extrusion, and *rp* is the membrane pore size.

$$Deformability\ (\%) = \frac{Vesicle\ size\ after\ extrusion}{Vesicle\ size\ before\ extrusion}\ x\ 100\% \quad \ldots (2)$$

Drug Entrapment Efficiency (EE) and Drug Loading
Entrapment efficiency (EE) refers to the amount of drug encapsulated in the bilayer of the vesicles, while drug loading (DL) indicates the total drug amount retained in the vesicles relative to the total lipid used. Both EE and DL were assessed using an indirect method. The transfersome solution was centrifuged at 14,000 rpm for 30 minutes at 4°C to separate the vesicles from any unentrapped drug. The supernatant, containing the free drug, was analyzed using HPLC. Quantification was based on ethyl p-methoxycinnamate (EPMC), the primary active ingredient in KGE. All measurements were performed in triplicate. The percentages of EE and DL were calculated using the following equations:[20]

$$\%EE\ = \frac{Total\ drug\ concentration - Total\ free\ drug\ concentration}{Total\ drug\ concentration} x\ 100\ . \quad (3)$$

$$\%DL\ = \frac{Total\ drug\ concentration - Total\ free\ drug\ concentration}{Total\ components\ transfersome\ formula}\ x\ 100 \ldots (4)$$

Formulation of Transdermal Patch
Single-layer transdermal patches were prepared using the solvent evaporation method, as shown in Figure 1. The formula for the KGE transfersome-loaded patch contained transfersome equivalent to 0.2% w/v extract, polyvinyl alcohol (PVA), glycerin, and distilled water. The polymer solution was created by dissolving 5% w/v PVA in hot distilled water (80-100 °C) while continuously stirring at 500 rpm for 10 minutes using a magnetic stirrer. Glycerin, acting as a plasticizer, and transfersome were gradually added to the solution while stirring, and the mixture was maintained at room temperature for 10 minutes. It was then sonicated for 15 minutes to eliminate air bubbles. A volume of 20 mL of the mixture was poured into a petri dish (16 cm diameter, 1 cm depth) and dried in an oven at 40°C for 12 hours. The patch was subsequently stored in a desiccator until further evaluation. The KGE-loaded patch was prepared using the same procedure, substituting the transfersome formulation with 0.2% w/v extract as the active ingredient. Each formulation was prepared in triplicate, and the resulting patches were cut into smaller sections for further evaluation.

Characterization of Transdermal Patch
Thickness Variation
The thickness of each transdermal patch was measured using a digital caliper at six different locations. The average thickness of the patch was then calculated.[21]

Weight Variation
Weight variations among the formulated transdermal patches were assessed using an analytical balance. Each patch was weighed individually, with three replicates conducted for accuracy. The average weight of the patches was then calculated.[21]

Moisture Content
The transdermal patches were cut into 2 cm x 2 cm pieces and weighed individually prior to drying. Each piece was placed on the moisture analyzer's pan and dried separately. After drying, the patches were reweighed, and this process was repeated three times.[21]

Moisture Loss
Each formula patch was cut and weighed individually at 100 mg. The patches were then stored in a desiccator with activated silica at room temperature for 24 hours. After storage, the patches were removed and reweighed to determine their final weight. The percentage of moisture loss was calculated by subtracting the final weight from the initial weight, dividing by the initial weight, and multiplying by 100.[21] The average percentage of moisture loss was obtained from three repetitions.

$$\%\ Moisture\ Loss\ = \frac{Initial\ weight\ of\ the\ patch - Final\ weight\ of\ the\ patch}{Initial\ weight\ of\ the\ patch}\ x\ 100 \ldots (5)$$

Folding Endurance
Folding endurance was assessed by repeatedly folding the patch at the same location until it cracked or broke. The maximum number of folds completed without cracking or breaking was recorded as the folding endurance value.[22]

pH Measurement
The pH measurement was conducted using a pH meter. A 100 mg patch was placed in a beaker containing 20 mL of distilled water and left for 15 minutes at room temperature. The glass electrode was submerged, and the pH was recorded after a one-minute stabilization period. This measurement was repeated three times.

Tensile Strength and Percent Elongation Break
Tensile strength and elongation at break were assessed with a digital force gauge. Patches from each formula were cut to dimensions of 5 cm x 1 cm. Each patch was secured in the device with 1 cm clamped at both the top and bottom. The upper end was attached to the upper clamp of the tensile tool, while the lower end was secured in the lower clamp. Weight was gradually added until the tensile force caused the patch to break. The force required for the breakage is defined as tensile strength, measured in kg/cm². The following equation is used to calculate the tensile strength of the patch:[21]

$$Tensile\ Strength\ = \frac{F}{[a.b\ (1+\frac{L}{I})]} \quad \ldots (6)$$

Where *F* represents the force needed to break the patch, *a* is the width of the patch in centimeters, *b* is the thickness in centimeters, *L* is the length in centimeters, and *I* is the elongation in centimeters before breaking or cracking occurs. The percentage elongation is calculated using the following equation:[21]

$$\%\ Elongation\ = \frac{(Lf - Li)}{Li}\ x\ 100 \quad \ldots (7)$$

Here, *Lf* is the length of the patch at the point of breaking in centimeters, and *Li* is the initial length of the patch in centimeters.

Ex vivo Skin Permeation Study
Ex-vivo permeation studies were performed using abdominal rat skin, with all procedures approved by the Ethics Committee of the Faculty of Medicine, Universitas Brawijaya, Indonesia (Approval Number 50/EC/KEPK/02/2025). Healthy Wistar Albino rats were selected and euthanized. Their abdominal hair was carefully shaved, and the skin was excised into square pieces (3 cm x 3 cm) using a scalpel. The skin samples were washed multiple times with PBS (pH 7.4), individually wrapped in aluminum foil, and stored at -20°C until needed. Prior to use, the skin was thawed and hydrated in PBS (pH 7.4) at room temperature for at least one hour.
The *ex vivo* permeation study utilized a Franz diffusion cell to evaluate drug permeability through the transdermal system. The rat abdominal skin was placed in the receptor compartment with the dermal side in contact with the receptor fluid and the stratum corneum side facing the donor compartment. A square patch (3 cm x 3 cm) was adhered to the stratum corneum, and both were secured with clamps. The receptor compartment was filled with 22 mL of PBS (pH 7.4) and stirred continuously at 500 rpm. The temperature of the system was maintained at 37°C ± 1°C using a hot plate magnetic stirrer. The diffusion study lasted for 10 hours, with samples collected from the receptor compartment at 1, 2, 4, 8, and 10 hours. A total of 5 mL of sample was withdrawn and replaced with an equivalent volume of PBS (pH 7.4) to preserve the sinking condition. The samples were prepared and analyzed using HPLC at a wavelength of 308 nm.[16]

Statistical Analysis
All data are presented as mean ± standard deviation (SD). Statistical analysis of the transfersome data was conducted using one-way

ANOVA, followed by Tukey's post hoc test with IBM SPSS software to identify significant differences among the variables. A p-value of less than 0.05 was deemed statistically significant. For the patch data, an unpaired t-test was utilized to compare differences between groups.

Results and Discussion

Characterization of Transfersome

pH

The pH value is crucial for the stability, solubility, and skin compatibility of the transfersome formulation, affecting drug entrapment and permeation capacity.[9] The measured pH values were as follows: 7.030 ± 0.057 for F1, 6.994 ± 0.025 for F2, and 7.028 ± 0.022 for F3. All values fell within the acceptable range for topical application. Statistical analysis revealed no significant differences among the formulations. These pH values are consistent with the Indonesian National Standard for skin products, which allows a pH range of 4.5 to 8.0.[23] The near-neutral pH of the formulations is likely due to the use of PBS at pH 7.4 during the hydration step, which is isotonic, biocompatible, and non-toxic.[24] Overall, the results suggest that all transfersome formulations are suitable for dermal application without the risk of irritation or pH-induced instability.

Particle size, Polydispersity Index, and Zeta Potential

Particle size is a critical factor affecting the ability of transfersomes to penetrate the skin barrier. For effective transdermal delivery, vesicle sizes should be below 300 nm to facilitate transport through the stratum corneum.[25] As shown in Table 2, all formulations met the required particle size criteria, with F3 (lecithin-to-Tween 80 ratio of 80:20) displaying the smallest average vesicle size. No statistically significant differences were found among the formulations. Higher surfactant concentrations correlated with smaller particle sizes, likely due to improved interfacial stabilization and decreased vesicle aggregation.[26] In this study, F3 had the smallest particle size due to its higher surfactant concentration. However, concentrations exceeding 20% may adversely affect vesicle structure, resulting in rigidity and decreased drug permeability.[27]

Table 2: Vesicle size, Polydispersity Index, and Zeta Potential

Formula (*Lecithin* : Tween 80)	Vesicle Size (nm)	PDI	Potential Zeta (mV)
F1 (90:10)	191.45 ± 12.52	0.25 ± 0.051	4.56 ± 2.63
F2 (85:15)	183.12 ± 17.39	0.44 ± 0.057	-5.59 ± 2.71
F3 (80:20)	152.85 ± 6.88	0.39 ± 0.045	-5.80 ± 1.36

The polydispersity index (PDI) quantifies the uniformity of a sample based on particle size.[28] PDI values below 0.5 indicate a monodisperse sample, while values above 0.7 suggest polydispersity.[29] All formulations in this study exhibited monodispersity, with PDI values below 0.5. Among these, formulation F1 (90:10) had the lowest PDI, indicating the most consistent particle size distribution, in contrast to F2. The PDI can be influenced by surfactant concentration; higher concentrations typically lead to lower PDI values due to reduced interfacial tension. However, excessively high surfactant concentrations can increase PDI values, resulting from micelle formation or structural instability.[30] Lecithin also plays a role in PDI, with an optimal concentration that minimizes PDI to reduce aggregation and narrow the size distribution.[31]

Zeta potential measurements were conducted to evaluate surface charge and predict colloidal stability. The zeta potential values ranged from +4.56 ± 2.63 mV (F1) to -5.80 ± 1.36 mV (F3), indicating relatively low surface charges across all formulations, with no statistically significant differences observed. Typically, zeta potential values above ±30 mV suggest strong electrostatic repulsion and good stability. However, the use of a non-ionic surfactant (Tween 80) indicates that steric stabilization, rather than electrostatic repulsion, primarily governs the stability of these systems.[32] Despite the low zeta potential values, all formulations were visually stable and showed no signs of aggregation, supporting their colloidal stability.

Vesicle Morphology

SEM images were captured for the optimal formulation, F3, which has a lecithin-to-Tween 80 ratio of 80:20, as indicated in Table 2. Figure 2 shows that the transfersomes are spherical with a smooth surface. This spherical shape is advantageous as it enhances entrapment efficiency, stability, and skin permeation. Additionally, vesicles with a smooth surface and uniform morphology are less likely to fuse or degrade during storage, contributing to the formulation's long-term stability.[9,33] These results align with the expected characteristics of transfersomes and confirm that the preparation method effectively produced vesicles suitable for transdermal application.

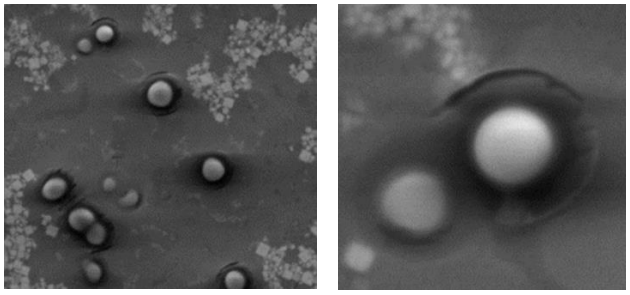
Deformability Test

The deformability test assessed the flexibility and elasticity of transfersome vesicles, crucial for their ability to traverse the narrow pores of the stratum corneum.[9] In this study, the deformability test involved extruding the vesicles through a 100 nm polycarbonate membrane. Measurements of vesicle size before and after extrusion were used to calculate the deformability index and percent deformability. A 100 nm pore size was chosen because the transfersomes had particle sizes below 200 nm. As detailed in Table 3, all formulations demonstrated high deformability, with percent deformability values exceeding 85%.

Table 3: Deformability Test Results

Formula (*Lecithin* : Tween 80)	Vesicle Size After Extrusion (nm)	Deformability Index	Deformability (%)
F1 (90:10)	185.83 ± 13.22	17.11 ± 2.34	97.10 ± 3.86
F2 (85:15)	160.03 ± 7.31	12.83 ± 1.15	87.84 ± 4.74
F3 (80:20)	150.99 ± 8.13	11.43 ± 1.21	98.76 ± 2.14

The F3 formulation exhibited the highest percent deformability, indicating greater vesicle flexibility or elasticity. This enhanced flexibility is likely attributed to the higher concentration of Tween 80, a membrane-softening agent known to increase vesicle flexibility.[6] Increased surfactant concentration correlates with a higher percent deformability, enhancing the transfersome's elasticity and their ability to return to their original shape after passing through membrane pores. However, a statistically significant difference in the deformability index was observed between F1 and F3. The deformability index decreases with increasing surfactant concentration because a higher surfactant level can reduce transfersome size. Consequently, the ability of the vesicles to change shape diminishes as their size approaches that of the membrane pores. Conversely, excessive surfactant use can negatively impact deformability by leading to the formation of rigid mixed micelles.[34] The high deformability of the transfersomes may enhance their ability to penetrate the skin barrier, potentially improving transdermal delivery. This possibility was further investigated in the *ex vivo* skin permeation study.



(a) (b)

Figure 2: Vesicle morphology. (a) 2.000x magnification, scale bar 10 µm; (b) 10.000x magnification, scale bar 2 µm.

Entrapment Efficiency and Drug Loading

Entrapment efficiency (EE) measurements were conducted to assess the amount of drug encapsulated in vesicles relative to the total drug used, with a desirable range of 80 to 100%.[35] Drug loading (DL) refers to the amount of drug encapsulated in vesicles compared to the total amount of vesicle-forming components. DL values were calculated using standard EE results.[9] As shown in Table 4, all formulations achieved EE values exceeding 80%, with formulation F1 demonstrating the highest entrapment efficiency and drug loading capacity. The consistently high EE across all formulations indicates the effectiveness of the transfersome system in encapsulating the active compound. Factors such as formulation composition and preparation methods can influence EE. Optimal concentrations of phospholipids and edge activators (surfactants) enhance bilayer flexibility and improve the vesicles' encapsulation capacity.[36] These findings suggest that the transfersomes developed in this study effectively incorporate EPMC, making them suitable carriers for transdermal delivery.

Table 4: Entrapment Efficiency Results

Formula (*Lecithin* : Tween 80)	Entrapment Efficiency (%)	Drug Loading (%)
F1 (90:10)	89.64 ± 6.81	12.13 ± 0.92
F2 (85:15)	86.33 ± 1.86	11.68 ± 0.25
F3 (80:20)	89.33 ± 3.35	12.09 ± 0.45

Characterization of Transdermal Patch

Physical Evaluation

Transdermal patches are non-invasive drug delivery systems designed to deliver therapeutic agents through the skin. They provide several benefits, including ease of use, improved patient adherence, and reduced gastrointestinal side effects associated with oral medications.[37,38] The release of drugs from transdermal patches can be adjusted by modifying parameters such as surface area, dosage, and application frequency. Additionally, the formulation characteristics determine whether the effect is local or systemic.[39]

Table 5 presents the physical properties of the KGE-loaded patch and the KGE transfersome-loaded patch. No statistically significant differences were found between the two formulations across all evaluated parameters. The KGE transfersome-loaded patch was thicker and heavier than the KGE-loaded patch, likely due to the presence of vesicles in the formulation. Both patches exhibited acceptable moisture content and flexibility, with folding endurance values significantly exceeding the minimum requirement. The pH of both patches fell within the physiologically acceptable range for skin application. In mechanical tests, the KGE-loaded patch demonstrated higher tensile strength and greater elongation at break than the transfersome-loaded patch, indicating better mechanical integrity.

Table 5: Evaluation of Transdermal Patch

Parameters	KGE-loaded patch	KGE transfersome-loaded patch
Thickness (mm)	0.01 ± 0.000	0.200 ± 0.000
Weight (gram)	0.140 ± 0.006	0.179 ± 0.013
Moisture Content (%)	13.558± 0.49	13.110 ± 2.22
Moisture Loss (%)	13.344 ± 1.057	8.832 ± 1.563
Folding Endurance	>500	>500
pH	7.33 ± 0.02	7.38 ± 0.03
Tensile strength (kg/cm^2)	76.33 ± 28.66	52.04 ± 11.96
Elongation break (%)	93.33 ± 3.33	74.44 ± 5.09

The KGE-loaded patch was thinner than the KGE transfersome-loaded patch. This difference in thickness may result from the incorporation of transfersomes, which increases the viscosity and density of the casting solution, resulting in a thicker film. Conversely, the ethanolic KGE extract in the non-vesicular patch dispersed more readily, creating a thinner matrix. Uniform patch thickness indicates even distribution of the patch solution and drug on the petri dish surface.[40] Weight variation evaluation ensures consistency in the manufacturing process, as it can influence drug dosage uniformity. The slightly higher mass of the transfersome formulation may stem from additional components like phospholipids and surfactants. Both patches exhibited low standard deviation values, indicating consistent composition and uniform casting. The low standard deviation of the transdermal patch suggests a uniform weight, reflecting even distribution of the drug and polymer.[41] Moisture content assessment determines the amount of water absorbed by the patch. This test is crucial to prevent the patch from becoming brittle, bulky, or prone to microbial contamination in humid environments. Acceptable moisture content typically ranges from 2% to 10%. Although slightly above the recommended range, the observed values are still acceptable and do not indicate instability. Moisture loss measurement assesses the liquid lost during the drying process, preventing brittleness or toxicity due to residual solvent.[42] These results suggest that the transfersome-based matrix may retain moisture more effectively, likely due to its vesicular structure that modulates water distribution. Folding endurance tests analyze the patch's brittleness and its ability to withstand repeated folding.[43] This test counts the number of folds the patch can endure before cracking or tearing, ensuring that the patch remains durable during daily activities. Both patches demonstrated excellent flexibility, with folding endurance exceeding 500 cycles, surpassing the typical threshold of 300. This flexibility can be attributed to the plasticizing effect of glycerin and the film-forming properties of PVA. pH measurement is crucial for the patch's stability and skin safety, ideally close to the skin's pH of 4.1 to 5.8. However, patch pH can range from 6.1 to 7.4 due to varying physiological conditions.[9,44] Tensile strength and percent elongation at break are used to evaluate mechanical strength and elasticity under stress. Patches with higher tensile strength typically exhibit lower elongation values, indicating greater rigidity. A tensile strength above 4.0 MPa (40.789 kg/cm^2) indicates an elastic patch.[45] Based on the results, the KGE-loaded patch demonstrated suitable tensile strength, exceeding 40.789 kg/cm^2.

Ex vivo Permeation Study

The permeation test evaluates a patch's ability to release active substances through the skin, typically using animal skin *in an ex vivo* simulation. This test employs a Franz diffusion cell filled with saline buffer media at pH 7.4. The patch is secured in place, and drug content is measured periodically over a 10-hour period.[46] Figure 3 illustrates the cumulative permeation of ethyl p-methoxycinnamate (EPMC) from both the KGE-loaded patch and the KGE transfersome-loaded patch over 10 hours. The transfersome-loaded patch consistently demonstrated higher drug permeation than the conventional patch at all time points. Both formulations displayed a continuous upward trend throughout the study, indicating an ongoing drug release process that had not yet plateaued. This improvement in delivery performance can be attributed to the structural benefits of transfersomes over non-vesicular systems. Transfersomes, made of phospholipids and edge activators like Tween 80, form ultra-deformable vesicles that can navigate the narrow intercellular pathways of the stratum corneum, enhancing transdermal absorption.[6,9] Transfersomes serve as both drug carriers and penetration enhancers. Their bilayer deformability and lipid compatibility facilitate interaction with skin lipids, promoting deeper penetration and sustained release.[47] Comparative studies have shown that transfersome patches outperform conventional patches in skin permeation and pharmacokinetic results. For instance, Chabru *et* al.[48] demonstrated that benztropine mesylate-loaded transfersomes achieved significantly higher drug permeation and systemic exposure compared to traditional pressure-sensitive adhesive patches. Similarly, Majukar *et* al.[22] found that aceclofenac-loaded transfersome patches exhibited improved anti-inflammatory effects and followed a non-Fickian drug release pattern, consistent with this study's finding.

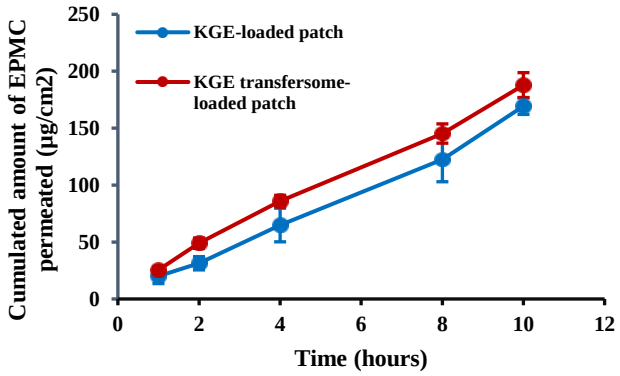


Figure 3: *Ex vivo* permeation profile of EPMC from KGE-loaded patch and KGE transfersome-loaded patch.

The lecithin-to-surfactant ratio is crucial for vesicle formation and function. This study identified an optimal 80:20 ratio, producing vesicles characterized by favorable size, low polydispersity, and high deformability, correlating with the enhanced performance of the transfersome patch. A higher surfactant content increases vesicle flexibility by disrupting lipid packing, thus improving bilayer fluidity and skin penetration.[11,26] However, excessive surfactant can destabilize vesicles by promoting micelle formation, emphasizing the need for careful ratio optimization. Clinically, using a transfersome patch for osteoarthritis therapy offers several advantages. Transdermal delivery targets the site of inflammation while reducing gastrointestinal side effects often associated with oral NSAIDs. Additionally, the inclusion of *Kaempferia galanga* extract, rich in EPMC, complements this delivery strategy. Its anti-inflammatory efficacy is comparable to meloxicam in clinical studies,[2] and its enhanced bioavailability through vesicle-based systems supports its development as a phytotherapeutic alternative.[47] In conclusion, the KGE transfersome-loaded patch demonstrated superior permeation compared to the conventional patch. These findings highlight the significance of formulation design, particularly the choice of vesicle type and composition, in optimizing transdermal delivery systems for anti-inflammatory herbal compounds. Future research should explore the use of other polymers, such as ethyl cellulose (EC) and hydroxypropyl methylcellulose (HPMC), both individually and in combination. Additionally, optimizing penetration enhancer concentrations may improve patch integrity and drug permeation. Complementary *in vitro* drug release studies alongside *ex vivo* testing are recommended to better predict release kinetics and optimize formulation parameters. Finally, extending the current *ex vivo* permeation study from 10 to 24 hours is advisable to capture a complete release profile and assess the potential for prolonged drug delivery.

Conclusion

This study successfully formulated a transfersome containing *Kaempferia galanga* extract with an optimal lecithin-to-surfactant ratio of 80:20, which was then incorporated into a transdermal patch. The transfersome patch demonstrated improved characteristics and greater skin permeation compared to a traditional extract patch. These results indicate its potential as a transdermal delivery system. Future research should investigate alternative polymers, optimize penetration enhancers, and extend the duration of permeation evaluations to enhance delivery performance.

Conflict of Interest

The authors declare no conflict of interest

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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