

Original Research

Received 25 May 2026

Revised 12 July 2026

Accepted 27 July 2026

Natural Resources for Human
Health 2026, Vol. 6 (13s): 1032–1053

KEYWORDS:

Charantin; Gymnemic acid;
dissolving microneedles;
transdermal drug delivery; beeswax
mould; polyvinyl alcohol;
polyvinylpyrrolidone K30;
polyethylene glycol 400; factorial
design; transdermal flux.

Design and Development of Charantin and Gymnemic Acid Microneedle Based Transdermal Drug Delivery System for Diabetes Management.

Quraishi Inshrah fatema^{1*}, Mirza Shahed Baig²

¹ Research scholar, Y.B. Chavan College of Pharmacy, Chatrapati Sambhajinagar, Maharashtra, India.

² Associate Professor, Y.B.Chavan College of Pharmacy . Chatrapati Sambhajinagar, Maharashtra, India.

*Corresponding author: Quraishi Inshrah fatema, Y.B.Chavan College of Pharmacy, Chatrapati Sambhajinagar, Maharashtra, India. Email: inshrahfatema456@gmail.com

ABSTRACT: The present study focused on the formulation and optimization of dissolving microneedle-assisted transdermal patches containing the phytoconstituents Charantin and Gymnemic acid using a pharmaceutical-grade beeswax mould. Hydrophilic polymers, including polyvinyl alcohol (PVA), polyvinylpyrrolidone K30 (PVP K30), hydroxypropyl methylcellulose E15, and sodium alginate, were screened for their suitability in microneedle fabrication. Preliminary formulations were prepared using PVA, PVP K30, and polyethylene glycol 400 (PEG 400), and evaluated for appearance, microneedle morphology, ease of demoulding, mechanical integrity, flexibility, uniformity of microneedle formation, drug content, swelling behaviour, in vitro drug release, and transdermal flux.

Five preliminary trial batches were prepared with varying concentrations of Charantin and Gymnemic acid. Among these, TB2, containing 5 mg of each phytoconstituent, demonstrated the most satisfactory overall characteristics, including sharp and uniformly formed microneedles, good mechanical strength, flexibility, drug content uniformity of $99.12 \pm 0.86\%$, and cumulative drug release of $96.84 \pm 0.92\%$ after 24 hours. Further optimization was performed using a 2^3 full factorial design to investigate the effects of PVA, PVP K30, and PEG 400 concentrations on cumulative drug release and transdermal flux.

The optimized formulation exhibited satisfactory physicochemical and mechanical properties. The factorial batches showed drug release values ranging from $84.36 \pm 1.42\%$ to $98.64 \pm 0.78\%$, while the transdermal flux ranged from 162.84 ± 3.12 to $218.76 \pm 2.48 \mu\text{g}/\text{cm}^2/\text{h}$. Formulation B8, containing 20% PVA, 10% PVP K30, and 15% PEG 400, demonstrated the highest cumulative drug release and transdermal flux. The findings indicate that beeswax mould-based dissolving microneedle patches may provide a promising transdermal delivery platform for Charantin and Gymnemic acid.

INTRODUCTION

The oral administration of phytoconstituents may be associated with several limitations, including poor aqueous solubility, variable gastrointestinal absorption, enzymatic degradation, first-pass metabolism, and difficulty in maintaining consistent plasma drug concentrations. These limitations can reduce the therapeutic efficiency of bioactive plant constituents and may necessitate frequent administration. Transdermal drug delivery provides an alternative route capable of bypassing the

gastrointestinal tract and hepatic first-pass metabolism while offering the possibility of sustained drug release and improved patient compliance (1-3).

However, the stratum corneum, the outermost layer of the skin, represents a significant barrier to the transport of most therapeutic molecules. Conventional transdermal patches are generally suitable only for potent, low-molecular-weight drugs with adequate lipophilicity and skin permeability. Phytoconstituents with limited permeability or unfavourable physicochemical properties may therefore require a method capable of temporarily disrupting the skin barrier without causing substantial tissue damage (4-6).

Microneedle technology has emerged as an effective approach for enhancing transdermal delivery. Microneedles are micron-sized projections capable of creating temporary aqueous microchannels within the stratum corneum, thereby facilitating drug transport into the skin. Dissolving microneedles are particularly advantageous because they are composed of water-soluble or biodegradable polymers that dissolve after insertion, eliminating the need for sharps disposal and reducing the possibility of needle retention. Their minimally invasive nature may also improve patient acceptability compared with conventional hypodermic injections (7-10). Charantin and Gymnemic acid are important phytoconstituents associated with the therapeutic potential of *Momordica charantia* and *Gymnema sylvestre*, respectively. Their incorporation into a microneedle-assisted transdermal system may improve drug administration by promoting controlled delivery and reducing limitations associated with conventional routes. Nevertheless, successful microneedle fabrication requires careful selection of polymers and plasticizers to obtain sufficient mechanical strength, sharp needle tips, uniform mould filling, rapid dissolution, and effective drug release (11-15).

In the present study, PVA, PVP K30, and PEG 400 were selected as the principal formulation components. PVA and PVP K30 provide film-forming, hydrophilic, and structural properties, whereas PEG 400 acts as a plasticizer that improves flexibility and reduces brittleness. A beeswax mould method was employed to fabricate the microneedle arrays because it offers a simple, economical, and adaptable approach for producing negative microneedle cavities. Preliminary trial batches were prepared to identify a suitable drug loading and polymeric composition. Subsequently, a 2^3 full factorial design was used to investigate the individual and interactive effects of PVA, PVP K30, and PEG 400 on cumulative drug release and transdermal flux. The objective of this investigation was therefore to develop and optimize dissolving microneedle transdermal patches containing Charantin and Gymnemic acid using a beeswax mould-based fabrication technique and to evaluate their physicochemical, mechanical, drug-release, and permeation characteristics (17-20).

MATERIALS AND METHODS

Selection of Polymers for Microneedle-Assisted Transdermal Patches of Charantin and Gymnemic acid [15,16]

The hydrophilic polymers are to be screened in individual instances and in suitable combinations- polyvinyl alcohol (PVA), polyvinylpyrrolidone (PVP K30), hydroxypropyl methylcellulose (HPMC E15), and sodium alginate. Polymer solutions of varying concentrations are to be prepared in distilled water under continuous magnetic stirring at either room temperature or at 40-60°C, depending on the polymer, until a homogeneous solution is obtained. The prepared polymer solutions shall be evaluated for clarity, viscosity, pH, homogeneity, ease of mould filling, film-forming ability. It is the conclusion of the present study, based on these preliminary evaluations, that the polymer or polymer blend exhibiting optimum physicochemical properties, adequate mechanical strength, and suitable processing characteristics should be selected for further study.

Table 1: Data for Selection of Polymer Blend

Trial Batch	PVA (% w/v)	PVP K30 (% w/v)	HPMC E15 (% w/v)	Sodium Alginate (% w/v)	PEG 400 (% w/w of total polymer)
T1	15	15	—	—	10
T2	15	—	—	—	10
T3	15	—	5	—	10
T4	15	—	—	3	10
T5	10	10	2	2	10

Preparation of Beeswax Mold

The beeswax must be of the highest pharmaceutical grade and be melted in a stainless-steel beaker at a temperature of 65–70°C using a water bath. The molten beeswax is then poured into a clean Petri dish, resulting in a wax layer with a thickness of approximately 5–8 mm. Prior to complete solidification, a stainless-steel microneedle master template (or an array of stainless-steel acupuncture needles/insect pins arranged uniformly) is to be vertically pressed into the softened beeswax to produce uniformly distributed conical cavities. Following a cooling period to ambient temperature, the master template is to be meticulously removed, thereby leaving a negative beeswax mould containing microneedle cavities. Prior to utilisation, a visual inspection of the mould is required to ensure the uniformity of the cavity dimensions and the absence of any defects.

Fabrication of Microneedle-Assisted Transdermal Patches

PVA (15% w/v) is dissolved in distilled water at 80–90°C with stirring and cooled to room temperature. PVP K30 (15% w/v) is then added gradually, followed by PEG 400 (10% w/w of total polymer) as a plasticizer. Accurately weighed charantin and gymnemic acid are dissolved/dispersed in methanol and incorporated into the polymer solution with continuous stirring for 20–30 minutes. The formulation is allowed to stand or vacuum-treated to remove air bubbles. The drug-loaded solution is poured into the beeswax mould and gently tapped or centrifuged at 3000–3500 rpm for 10–15 minutes to ensure complete cavity filling. The mould is dried at $25 \pm 2^\circ\text{C}$ for 24 hours or at $35\text{--}40^\circ\text{C}$ for 8–10 hours. The dried microneedle patch is carefully removed and packed in laminated aluminium pouches with silica gel, then stored at $25 \pm 2^\circ\text{C}$ until evaluation (21–22).

Preliminary trial batches for Microneedle-Assisted Transdermal Patches of Charantin and Gymnemic acid

Initial trial batches were prepared to identify a suitable polymer composition for dissolving microneedle patches with adequate mechanical strength, flexibility, rapid dissolution, and satisfactory drug release. Based on literature review, preliminary trials, and polymer properties, the polymer composition was fixed at PVA 15% w/v, PVP K30 15% w/v, and PEG 400 10% w/w. Different drug loadings will then be evaluated using this fixed polymer composition to determine the optimum drug concentration for well-formed and mechanically stable microneedles.

Table 2: Preliminary trial batches of Micro-needle-Assisted Transdermal Patches

Ingredients	TB1	TB2	TB3	TB4	TB5
Charantin (mg)	2.5	5.0	7.5	10.0	12.5
Gymnemic acid (mg)	2.5	5.0	7.5	10.0	12.5
PVA (15% w/v) (g)	1.5	1.5	1.5	1.5	1.5
PVP K30 (15% w/v) (g)	1.5	1.5	1.5	1.5	1.5
PEG 400 (10% w/w of total polymer) (mg)	300	300	300	300	300
Distilled Water	q.s. to 10 mL	q.s. to 10 mL	q.s. to 10 mL	q.s. to 10 mL	q.s. to 10 mL

Evaluation of trial batches of Micro-needle-Assisted Transdermal Patches

The preliminary trial batches of dissolving micro-needle-assisted transdermal patches were evaluated in order to identify the most suitable polymeric composition for subsequent optimization by factorial design. An examination was conducted on the prepared patches to ascertain their physical appearance, microneedle formation, mechanical integrity, flexibility, and preliminary drug release characteristics.

2. Microneedle Morphology

The prepared microneedles were then subjected to a thorough observation using an optical microscope. This evaluation encompassed several parameters, including the morphology of the needles, the sharpness of the tips, the uniformity of the needle arrangement, the completeness of the cavity filling, and the presence of broken or deformed needles. The formulation that exhibited well-defined and uniformly distributed microneedles was deemed suitable for further optimization.

3. Ease of Demoulding

The prepared patches were then meticulously extracted from the beeswax mould using sterile forceps, after which they were subjected to a thorough evaluation to ascertain their ease of demoulding. The patches were then subjected to rigorous scrutiny to ascertain the presence of any needle breakage, deformation, or adherence to the mould surface. Formulations that could be removed without causing damage were considered acceptable.

4. Mechanical integrity

The mechanical strength of the microneedles was assessed manually by applying gentle thumb pressure to the patch surface. Microneedles that remained intact without bending or fracture were considered mechanically acceptable for skin insertion.

5. Patch Flexibility

The flexibility of the prepared patches was evaluated by subjecting the backing layer to a gentle bend of approximately 180°. The patches were subjected to a thorough examination to ascertain the presence of any cracks, brittleness, or permanent deformation. Flexible patches that did not exhibit visible cracks were deemed suitable for further investigation.

6. Uniformity of micro needle formation

The entire micro needle array was examined under an optical microscope to determine the uniformity of needle height, base diameter, spacing, and tip formation. Microneedles exhibiting uniform formation and complete cavity filling were deemed to be acceptable.

7. Preliminary investigation of drug content uniformity

Preliminary drug content uniformity will be evaluated to determine the uniform distribution of charantin and gymnemic acid in the microneedle matrix. Three patches of identical size ($2 \times 2 \text{ cm}^2$) will be randomly selected from each trial batch, cut into small pieces, and transferred to a 100 mL volumetric flask containing 70 mL of methanol:phosphate buffer pH 7.4 (70:30, v/v). The samples will be sonicated for 30 minutes, diluted to volume with the same solvent, and filtered through a $0.45 \mu\text{m}$ membrane filter. Suitable dilutions will be prepared, and the drug content will be determined simultaneously using the validated UV method. The percentage drug content will be calculated as:

$$\text{Drug Content (\%)} = (\text{Actual amount of drug} / \text{Theoretical amount of drug}) \times 100$$

8. The preliminary in vitro drug release study

The preliminary in vitro drug release study will be performed using a Franz diffusion cell to evaluate the release of charantin and gymnemic acid from the microneedle matrix. A pre-hydrated cellulose acetate membrane (MWCO 12–14 kDa) will be placed between the donor and receptor compartments. The receptor compartment will contain phosphate buffer (pH 7.4) with 20% v/v ethanol and will be maintained at $37 \pm 0.5^\circ\text{C}$ with stirring at 500 rpm. The microneedle patch will be placed on the membrane with the microneedle side facing the membrane. Samples (1 mL) will be withdrawn at 0.5, 1, 2, 4, 6, 8, 12, and 24 hours and replaced with an equal volume of fresh, preheated medium. Samples will be filtered through a $0.45 \mu\text{m}$ membrane filter and analysed using the validated UV method. Cumulative drug release will be calculated with volume-replacement correction and plotted against time. The trial batch showing satisfactory and uniform drug release will be selected for further optimization using a 2^3 factorial design (22-25).

Selection of an Optimized Trial Batch

Following a comprehensive evaluation of the formulation, including physical appearance, microneedle morphology, mechanical integrity, ease of demoulding, flexibility, drug content uniformity, and preliminary in vitro drug release, it was determined that the formulation demonstrating optimum performance should be selected as the preliminary optimized trial batch. This formulation was subsequently employed as the centre-point formulation for the 2^3 full factorial design to optimise the concentrations of PVA, PVP K30, and PEG 400 with respect to cumulative drug release and transdermal flux.

Factorial Design for Micro-needle-Assisted Transdermal Patches of Charantin and Gymnemic acid

The formulation variables affecting the performance of dissolving micro-needle-assisted transdermal patches containing Charantin and Gymnemic acid were optimised using a 2^3 full factorial design. The experimental design was generated using

Design-Expert® software (Version 13.0, Stat-Ease Inc.). A full factorial design was selected to evaluate the individual and interaction effects of three independent formulation variables on the critical quality attributes of the microneedle patches.

Table 3: Experimental design batches of Micro-needle-Assisted Transdermal Patches

		Factor 1	Factor 2	Factor 3	Response 1	Response 2
Std	Run	A:Concentration of Polyvinyl Alcohol (PVA)	B:Concnetration of Polyvinylpyrrolidone K 30 (PVP K30)	C:Concentration of Polyethylene Glycol 400 (PEG 400)	Cumulative drug release	Flux
		%	%	%	(%)	($\mu\text{g}/\text{cm}^2/\text{h}$)
1	1	10	10	5		
5	2	10	10	15		
12	3	15	15	10		
8	4	20	20	15		
7	5	10	20	15		
13	6	15	15	10		
4	7	20	20	5		
6	8	20	10	15		
10	9	15	15	10		
3	10	10	20	5		
2	11	20	10	5		
9	12	15	15	10		
11	13	15	15	10		

The experimental runs were subjected to randomisation by the software programme in order to minimise systematic bias and the influence of uncontrolled variables. The centre-point formulations were distributed throughout the experimental sequence rather than being performed consecutively. This enabled estimation of pure experimental error, assessment of process reproducibility, and detection of curvature in the response surface.

Table 4: Experimental factors for design batches

Factor	Name	Units	Type	SubType	Minimum	Maximum	Code Low	Code High	Mean	Std. Dev.
A	Concentration of Polyvinyl Alcohol (PVA)	%	Numeric	Continuous	10.00	20.00	-1 ↔ 10.00	+1 ↔ 20.00	15.00	4.08
B	Concnetration of Polyvinylpyrrolidone K 30 (PVP K30)	%	Numeric	Continuous	10.00	20.00	-1 ↔ 10.00	+1 ↔ 20.00	15.00	4.08
C	Concentration of Polyethylene Glycol 400 (PEG 400)	%	Numeric	Continuous	5.00	15.00	-1 ↔ 5.00	+1 ↔ 15.00	10.00	4.08

Evaluation of Designed Micro-needle Transdermal Patches

The physicochemical, mechanical, morphological, and drug delivery characteristics of dissolving micro-needle-assisted transdermal patches containing Charantin and Gymnemic acid are to be evaluated using a factorial design batches approach.

1. Determination of thickness of formulated patch

The thickness of the microneedle patches is to be measured using a calibrated digital micrometer at five different locations (centre and four corners). The mean thickness was calculated and expressed in millimetres.

2. Weight Variation test

A total of ten microneedle patches are to be selected at random, and subsequently weighed individually using an analytical balance. The mean weight, standard deviation, and percentage deviation was calculated.

3. Determination of Folding Endurance

The flexibility of the backing layer is determined by repeatedly folding the patch at the same location until visible cracks appear or the patch breaks. The number of folds required to produce failure is to be recorded as the folding endurance.

4. Determination of Surface pH

The microneedle patches, which have been prepared in advance, are to be applied to the agar gel (prepared with distilled water) for a period of two hours. Thereafter, the surface pH was measured using a calibrated digital pH meter, with the electrode being gently placed in contact with the surface of the hydrated patch.

5. Determination of Drug Content Uniformity

The content of the Charantin and Gymnemic acid determined by the dissolution of a known area of the microneedle patch in a methanol/ethanol, followed by sonication. The solution is to be filtered through a 0.45 μm membrane filter and analysed using the validated UV method for simultaneous estimation of Charantin and Gymnemic acid. The percentage of the theoretical drug content was expressed as the drug content.

6. Determination of the swelling index

Micro-needle-assisted transdermal patches measuring 1 cm^2 from each batch was weighed and placed on a pre-weighed cover slip. The specimen was placed in a petri dish, after which 10 mL of distilled water was added. Following a period of 10 minutes, the cover slip was detached and the excess water was meticulously removed using a tissue paper. The object was then weighed. The weight increase resulting from the absorption of water and subsequent swelling of the patch was determined through a meticulous calculation of the difference between the initial and final weights. The percentage swelling index was calculated using the following equation

$$\% \text{ Swelling index} = (W_t - W_o / W_o) \times 100$$

Where W_t is the final weight of the swollen film after time t , and W_o is the initial weight of the film at zero time.

7. In Vitro Drug Release

The in vitro drug release study is to be conducted using above process (given in preliminary in vitro dissolution study) a Franz diffusion cell containing phosphate buffer (pH 7.4) as the receptor medium. Samples are to be withdrawn at predetermined intervals up to 24 hours and analysed by UV- visible Spectrophotometer. The cumulative percentage drug release shall be calculated and plotted against time.

8. Determination of Flux

Excised goat abdominal skin will be mounted between the donor and receptor compartments of a Franz diffusion cell, with the stratum corneum facing the donor side. The receptor compartment will contain phosphate buffer (pH 7.4) with 20% v/v ethanol and will be maintained at $37 \pm 0.5^\circ\text{C}$ with stirring at 500 rpm. The microneedle patch will be applied to the skin, and 1 mL samples will be withdrawn at 0.5, 1, 2, 4, 6, 8, 12, and 24 hours, replacing each sample with an equal volume of fresh, preheated medium. Samples will be filtered through a 0.45 μm membrane filter and analysed using the validated UV-Visible

spectrophotometric method. The cumulative amount of drug permeated will be calculated and expressed as $\mu\text{g}/\text{cm}^2$ based on the effective diffusion area of the Franz diffusion cell.

$$J_{ss} = dQ/dt / A$$

Where,

J_{ss} = Steady-state flux ($\mu\text{g}/\text{cm}^2/\text{h}$)

dQ/dt = Steady-state permeation rate (slope of the linear portion of the cumulative amount permeated vs. time plot) ($\mu\text{g}/\text{h}$)

A = Effective diffusion area of the Franz diffusion cell (cm^2)

Table 5: Optimized batch Final formula

Ingredients	TB1
Charantin (mg)	5
Gymnemic acid (mg)	5
PVA (20% w/v) (g)	2.0
PVP K30 (10% w/v) (g)	1.0
PEG 400 (15% w/w of total polymer) (mg)	450
Distilled Water	q.s. to 10 mL

Preliminary trial batches for Microneedle-Assisted Transdermal Patches of Charantin and Gymnemic acid

Five preliminary trial batches (T1-T5) of microneedle-assisted transdermal patches containing Charantin and Gymnemic acid were prepared using the beeswax mould casting method to identify an appropriate polymeric composition for further optimization. The formulations were prepared by varying the concentrations of Polyvinyl Alcohol (PVA), Polyvinylpyrrolidone K30 (PVP K30), and Polyethylene Glycol 400 (PEG 400), while maintaining a constant concentration of both phytoconstituents. The prepared patches were then subjected to a comprehensive evaluation that encompassed various parameters, including visual appearance, ease of demoulding, Microneedle Morphology, mechanical integrity, patch flexibility, uniformity of micro needle formation, preliminary drug content uniformity, and preliminary in vitro drug release.



Figure 1: Bees wax mold for the formulation of Microneedle-Assisted Transdermal Patches

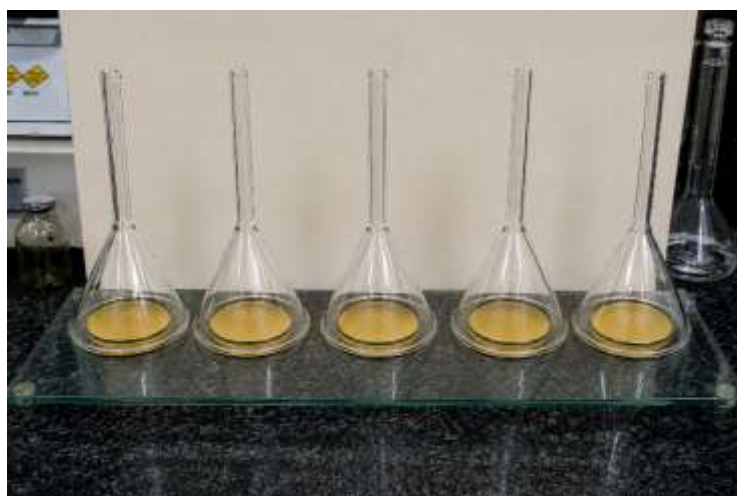


Figure 2: Preliminary trial batches of Microneedle-Assisted Transdermal Patches of Charantin and Gymnemic acid

Evaluation of trial batches of Micro-needle-Assisted Transdermal Patches

1) Visual Inspection

It was determined through visual inspection that an increase in the concentration of Charantin and Gymnemic acid resulted in a gradual intensification of the colour of the patches, accompanied by a slight reduction in transparency at higher drug loadings. The polymeric matrix composed of PVA, PVP K30, and PEG 400 effectively maintained the structural integrity and flexibility of the patches across all formulations.

2) Microneedle Morphology

The morphological evaluation indicated that the selected polymeric matrix possessed excellent mouldability and consistently produced well-defined microneedle arrays. The increase in drug concentration from 2.5 mg to 12.5 mg had only a minor influence on needle morphology. Although formulations containing higher drug concentrations (TB4 and TB5) exhibited slight reductions in tip sharpness and surface uniformity, complete needle formation and structural integrity were maintained. TB2 exhibited the optimal overall morphology, characterised by sharp needle tips, uniform geometry, and exceptional structural integrity, thus establishing itself as the most suitable formulation for subsequent factorial optimisation.

3) Ease of Demoulding

All preliminary trial batches showed satisfactory demoulding, with minimal adhesion to the beeswax mould. PEG 400 improved polymer flexibility and facilitated clean removal of the patches. Although TB4 and TB5 required slightly greater force due to their higher drug loading, all patches retained their structural integrity and complete microneedle arrays. TB2 showed the best overall demoulding characteristics and was therefore selected for further factorial optimization.

4) Mechanical integrity

The mechanical integrity of the microneedles was mainly attributed to the PVA–PVP K30 matrix, while PEG 400 improved flexibility and reduced brittleness. Drug loading from 2.5 to 7.5 mg showed little effect on mechanical strength, whereas higher drug loading (10.0 and 12.5 mg) slightly reduced needle rigidity, possibly due to increased solid content and reduced polymer continuity. However, all formulations maintained their structural integrity without fracture or collapse. TB2 provided the best balance between drug loading and mechanical strength and was therefore selected for further optimization.

5) Patch Flexibility

The flexibility of the patches was attributed to the combined effect of PVA, PVP K30, and PEG 400, which provided adequate elasticity for handling and application. Drug loading from 2.5 to 7.5 mg had minimal effect on flexibility, while higher loading (10–12.5 mg) slightly increased patch stiffness, possibly due to reduced polymer chain mobility. However, no formulation showed cracking or structural damage during bending. TB2 demonstrated the best balance of flexibility, elastic recovery, handling, and drug loading and was therefore selected for further optimization.

6) Uniformity of micro needle formation

Microneedle uniformity was mainly influenced by the flow and viscosity of the drug–polymer solution during casting. The selected polymer system provided adequate viscosity for proper mould filling and consistent needle formation. Drug loading up to 5.0 mg had little effect on needle formation, whereas higher loading (10–12.5 mg) slightly reduced needle height and tip sharpness due to increased viscosity. Nevertheless, all formulations maintained complete and defect-free microneedle arrays. TB2 showed the most uniform needle geometry and was selected for further optimization.

7) Preliminary investigation of drug content uniformity

The preliminary drug content study demonstrated that the selected polymeric matrix, consisting of PVA, PVP K30, and PEG 400, provided uniform incorporation of Charantin and Gymnemic acid throughout the microneedle patches. The drug content of Charantin and Gymnemic acid was analysed at 216 nm by using uv visible spectrophotometer. Although an increase in drug concentration from 2.5 mg to 12.5 mg resulted in a slight increase in variability in drug content, the differences were not substantial enough to affect formulation quality.

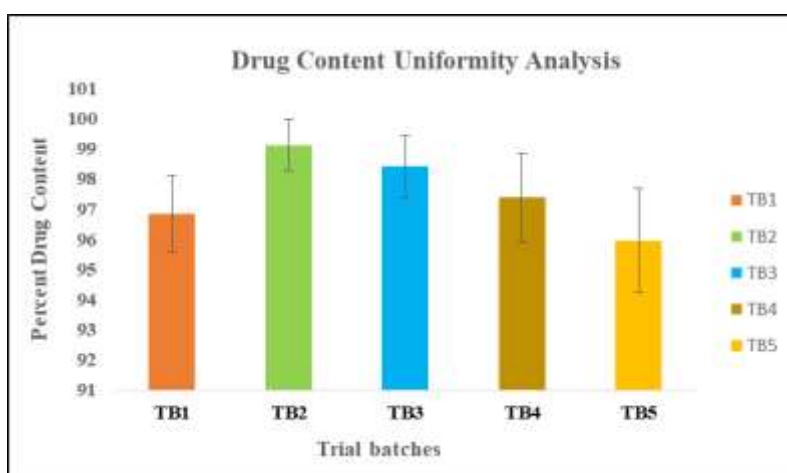


Figure 3: Drug Content Uniformity analysis of trial batches

Table 6: Preliminary Drug Content Uniformity of Trial Batches

Trial Batch	Theoretical Drug Content (mg/patch)*	Drug Content (%)
TB1	5.0	96.84 ± 1.25
TB2	10.0	99.12 ± 0.86
TB3	15.0	98.41 ± 1.03
TB4	20.0	97.38 ± 1.48
TB5	25.0	95.96 ± 1.72

(Mean ± SD, n = 3) *

The highest drug content uniformity observed in TB2 suggests that this formulation provided the most homogeneous distribution of both phytoconstituents while maintaining excellent physical characteristics. Consequently, TB2 was selected as the optimized preliminary formulation for further factorial optimization.

8) The preliminary in vitro drug release study

The preliminary in vitro drug release study was performed using a Franz diffusion cell with phosphate buffer (pH 7.4) containing 20% v/v ethanol as the receptor medium. Samples were withdrawn at predetermined intervals and analysed by UV-Visible spectrophotometry at 216 nm. All formulations showed gradual and sustained drug release. TB1 (2.5 mg each) showed the lowest release ($84.52 \pm 1.36\%$), while TB2 (5.0 mg each) showed the highest release ($96.84 \pm 0.92\%$) with a

uniform profile. The higher release from TB2 may be attributed to better drug distribution and hydration of the hydrophilic polymer matrix. TB3 (7.5 mg each) showed $93.76 \pm 1.18\%$ release, while TB4 (10.0 mg each) showed $90.38 \pm 1.44\%$. The gradual reduction in release at higher drug loading may be due to increased matrix density and reduced hydration and diffusion pathways. Overall, TB2 demonstrated the most satisfactory release profile and was selected for further optimization.

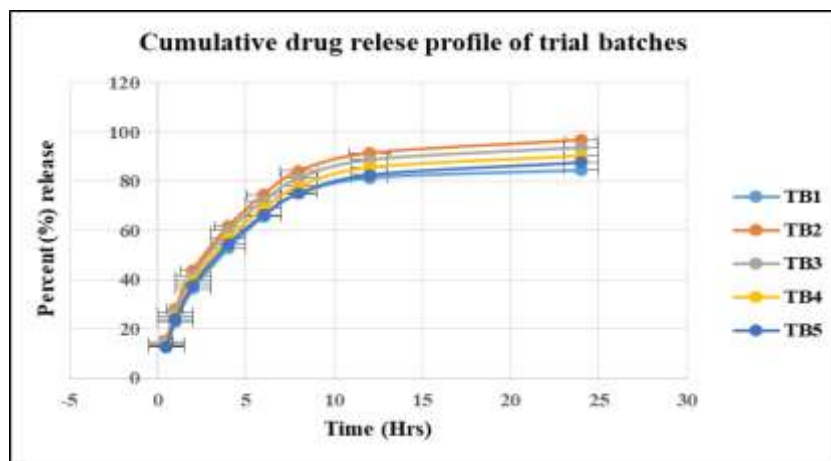


Figure 4: Cumulative percent drug release profile of trial batches

Table 7: Preliminary In Vitro Drug Release Profile of Trial Batches

Time (h)	TB1 (%)	TB2 (%)	TB3 (%)	TB4 (%)	TB5 (%)
0.5	12.46 ± 0.42	15.28 ± 0.36	14.62 ± 0.41	13.85 ± 0.39	12.94 ± 0.45
1	22.84 ± 0.56	27.96 ± 0.48	26.74 ± 0.52	25.13 ± 0.58	23.86 ± 0.61
2	36.15 ± 0.74	43.82 ± 0.67	41.56 ± 0.71	39.44 ± 0.73	37.62 ± 0.79
4	52.63 ± 0.88	61.74 ± 0.81	59.92 ± 0.84	56.86 ± 0.87	54.48 ± 0.91
6	65.74 ± 1.02	74.58 ± 0.95	71.86 ± 0.97	68.92 ± 1.01	66.35 ± 1.05
8	74.96 ± 1.18	84.27 ± 1.06	81.34 ± 1.12	77.95 ± 1.14	75.18 ± 1.19
12	81.38 ± 1.29	91.46 ± 1.15	88.76 ± 1.21	85.64 ± 1.24	82.58 ± 1.31
24	84.52 ± 1.36	96.84 ± 0.92	93.76 ± 1.18	90.38 ± 1.44	87.64 ± 1.61

(Mean $\pm$ SD, n = 3)

In the present study, formulation TB5, containing 12.5 mg of each Charantin and Gymnemic acid, demonstrated the lowest release rate among the higher drug-loaded formulations ($87.64 \pm 1.61\%$). The comparatively slower drug release may be due to partial drug aggregation and reduced wettability of the polymeric matrix, resulting in slower diffusion of the phytoconstituents.

Selection of an Optimized Trial Batch

The five trial batches (TB1–TB5) were compared based on appearance, microneedle morphology, demoulding, mechanical strength, flexibility, needle uniformity, drug content, and in vitro drug release. Among the formulations, TB2 showed the most satisfactory overall performance, with well-formed microneedles, good handling properties, uniform drug distribution, and the highest drug release. Therefore, TB2 was selected as the optimized preliminary batch and used as the baseline for further optimization of polymer concentrations using a 2^3 full factorial design.

Factorial Design for Micro-needle-Assisted Transdermal Patches of Charantin and Gymnemic acid

Following the selection of TB2, a 2^3 full factorial design was used to optimize the formulation variables of the microneedle patches. The three independent variables were PVA concentration (X_1), PVP K30 concentration (X_2), and PEG 400

concentration (X_3). Cumulative drug release (Y_1) and transdermal flux (Y_2) were selected as the responses. A total of 13 experimental runs, including eight factorial points and five centre points, were generated using Design-Expert software. The design was used to study the individual and interaction effects of the formulation variables and to identify the optimized formulation.

RESULTS AND DISCUSSIONS

9. Determination of thickness of formulated patch

The thickness of the microneedle-assisted transdermal patches (F1–F13) was measured at three locations using a calibrated digital micrometer, and the mean was calculated. Patch thickness ranged from 0.42 ± 0.01 to 0.61 ± 0.02 mm, indicating good uniformity among the formulations. Minor variations were attributed to differences in PVA, PVP K30, and PEG 400 concentrations. Higher PVA and PVP K30 concentrations increased film thickness due to greater solution viscosity and polymer matrix density, whereas higher PEG 400 concentrations produced thinner, more flexible patches owing to its plasticising effect, which enhances polymer chain mobility and reduces matrix rigidity.



Figure 5: Determination of thickness of formulated patch

Table 8: Thickness of Formulated Microneedle-Assisted Transdermal Patches

Formulation	Thickness (mm)
F1	0.42 ± 0.01
F2	0.45 ± 0.02
F3	0.48 ± 0.01
F4	0.58 ± 0.02
F5	0.46 ± 0.01
F6	0.52 ± 0.02
F7	0.56 ± 0.01
F8	0.61 ± 0.02
F9	0.48 ± 0.01
F10	0.47 ± 0.01
F11	0.49 ± 0.02
F12	0.48 ± 0.01
F13	0.48 ± 0.01

(Mean $\pm$ SD, n = 3)

Thickness evaluation showed that all microneedle-assisted transdermal patches had good uniformity, with only minor batch-to-batch variations, confirming the reproducibility of the beeswax mould casting technique. Variations in thickness were mainly attributed to differences in PVA, PVP K30, and PEG 400 concentrations, which influenced polymer solution viscosity and polymer deposition during solvent evaporation. All formulations remained within an acceptable thickness range, providing adequate mechanical strength, flexibility, and uniform drug distribution. Among the factorial batches, B8 exhibited a desirable balance between structural integrity and flexibility with consistent dimensions, indicating good manufacturing reproducibility. However, the final optimized formulation was selected subsequently based on numerical optimization and desirability function analysis using Design-Expert® software.

10. Weight Variation test

Weight variation was evaluated to assess the uniformity and reproducibility of the microneedle-assisted transdermal patches (F1–F13). Each formulation was weighed in triplicate using a calibrated analytical balance, and the mean weight and standard deviation were calculated. The mean patch weight ranged from 145.26 ± 2.18 to 176.82 ± 2.46 mg, indicating satisfactory batch uniformity. Minor variations were attributed to differences in PVA, PVP K30, and PEG 400 concentrations. Higher PVA and PVP K30 concentrations resulted in greater patch weight due to increased polymer content and deposition during solvent evaporation, whereas lower polymer concentrations produced lighter patches. PEG 400 had a minimal effect on patch weight, primarily functioning as a plasticiser without substantially altering the total solid content.

Table 9: Weight Variation of Formulated Microneedle-Assisted Transdermal Patches

Formulation	Weight (mg)
F1	145.26 ± 2.18
F2	149.84 ± 1.94
F3	153.72 ± 2.11
F4	170.56 ± 2.38
F5	151.63 ± 1.86
F6	160.48 ± 2.05
F7	167.35 ± 2.29
F8	176.82 ± 2.46
F9	158.74 ± 1.32
F10	156.92 ± 1.58
F11	159.46 ± 1.47
F12	158.74 ± 1.32
F13	158.36 ± 1.41

(Mean $\pm$ SD, n = 3)

The weight variation study demonstrated satisfactory uniformity across all microneedle-assisted transdermal patches, with minimal batch-to-batch variation, confirming the reproducibility of the beeswax mould casting technique. Minor differences in weight were attributed to variations in PVA, PVP K30, and PEG 400 concentrations, which influenced the overall solid content of the polymeric matrix. Among the factorial batches, B8 showed the lowest weight variability, indicating uniform mould filling, consistent polymer distribution, and good process reproducibility. However, the final optimized formulation was selected subsequently through numerical optimization and desirability analysis using Design-Expert® software.

11. Determination of Folding Endurance

Folding endurance was evaluated to assess the flexibility and mechanical strength of the microneedle-assisted transdermal patches (B1–B13). Each patch was repeatedly folded at the same point until cracking or breakage occurred. All formulations showed good folding endurance, ranging from 245 ± 4 to 296 ± 3 folds, indicating that the polymeric films could withstand repeated handling without damage. Higher PEG 400 concentrations generally improved flexibility by increasing polymer chain mobility, while PVA and PVP K30 contributed to the strength and integrity of the polymer matrix. Overall, the combined polymers produced flexible and mechanically stable patches suitable for transdermal application.

Table 10: Determination of Folding Endurance

Formulation	Folding Endurance (Number of Folds)
F1	245 ± 4
F2	252 ± 5
F3	270 ± 4
F4	278 ± 3
F5	255 ± 4
F6	286 ± 3
F7	289 ± 4
F8	296 ± 3
F9	271 ± 4
F10	260 ± 5
F11	274 ± 4
F12	272 ± 3
F13	273 ± 4

(Mean $\pm$ SD, n = 3)

Among the factorial batches, B8 demonstrated the highest folding endurance (296 ± 3 folds), indicating superior flexibility and mechanical robustness. The optimal polymeric network, capable of withstanding repeated bending without forming cracks or fractures, was achieved through a balanced combination of higher concentrations of PVA, PVP K30, and PEG 400. The centre point batches also exhibited satisfactory folding endurance, thus confirming the reproducibility of the formulation process. However, the performance of B8 was comparatively superior among the factorial formulations.

12. Determination of Surface pH

The surface pH of the microneedle-assisted transdermal patches (B1–B13) was evaluated to assess their suitability for skin application. The patches showed pH values ranging from 5.62 ± 0.04 to 6.41 ± 0.03 , which falls within the physiological pH range of human skin (approximately 4.5–6.5), indicating good skin compatibility. Minor variations were attributed to differences in PVA, PVP K30, and PEG 400 concentrations and their effects on matrix hydration. PEG 400 had minimal influence on pH, while PVA and PVP K30 helped maintain a stable polymeric matrix. The presence of Charantin and Gymnemic acid did not cause any significant pH alteration, suggesting good compatibility with the selected excipients.

Table 11: Surface pH of Formulated Microneedle-Assisted Transdermal Patches

Formulation	Surface pH
F1	5.62 ± 0.04
F2	5.74 ± 0.03

F3	5.93 ± 0.05
F4	6.08 ± 0.04
F5	5.79 ± 0.04
F6	6.05 ± 0.03
F7	6.10 ± 0.04
F8	6.12 ± 0.03
F9	5.98 ± 0.04
F10	5.86 ± 0.03
F11	6.02 ± 0.05
F12	6.00 ± 0.04
F13	6.01 ± 0.03

(Mean ± SD, n = 3)

Among the factorial batches, B8 demonstrated a surface pH of 6.12 ± 0.03 , which was closest to the physiological skin pH and therefore deemed the most suitable for transdermal application. The pH values of the formulations were found to be within the acceptable range, indicating that the prepared patches are not likely to cause skin irritation or discomfort during application.

13. Determination of Drug Content Uniformity

Drug content uniformity of the microneedle-assisted transdermal patches (B1–B13) was evaluated to assess the distribution of Charantin and Gymnemic acid within the polymeric matrix. The drug content was determined by dissolving accurately weighed patches in the optimized extraction solvent, followed by UV-visible spectrophotometric analysis. All formulations showed satisfactory drug content, ranging from $94.86 \pm 1.42\%$ to $99.74 \pm 0.68\%$, with low variability, indicating good drug incorporation and uniform distribution. The film-forming properties of PVA and PVP K30 supported homogeneous drug dispersion, while PEG 400 improved flexibility and spreading during mould filling. Overall, balanced concentrations of PVA, PVP K30, and PEG 400 contributed to consistent drug content and reproducible patch preparation.

Table 12: Drug Content Uniformity of Formulated Microneedle-Assisted Transdermal Patches

Formulation	Drug Content (%)
F1	94.86 ± 1.42
F2	95.78 ± 1.24
F3	97.46 ± 0.94
F4	98.18 ± 0.86
F5	95.92 ± 1.15
F6	98.64 ± 0.79
F7	99.12 ± 0.74
F8	99.74 ± 0.68
F9	97.82 ± 0.88
F10	96.54 ± 1.03

F11	98.06 $\pm$ 0.82
F12	97.95 $\pm$ 0.85
F13	98.12 $\pm$ 0.81

(Mean $\pm$ SD, n = 3)

Among the factorial batches, B8 exhibited the highest drug content uniformity (99.74 $\pm$ 0.68%), indicating excellent incorporation and homogeneous distribution of both phytoconstituents within the polymeric matrix. The combination of higher concentrations of PVA, PVP K30, and PEG 400 in B8 produced a stable polymeric network that effectively minimised drug loss during casting and drying. The centre point batches also demonstrated satisfactory drug content with minimal variability, thus confirming the reproducibility of the formulation process. However, B8 demonstrated the most optimal overall performance among the factorial formulations.

14. Determination of the swelling index

The swelling index of the microneedle-assisted transdermal patches (B1–B13) was evaluated by immersing pre-weighed patches in phosphate buffer (pH 7.4) at 37 $\pm$ 0.5°C and measuring the increase in weight after hydration. The swelling index ranged from 162.84 $\pm$ 3.26% to 228.73 $\pm$ 2.94%, indicating good water absorption by the polymeric matrix. The hydrophilic nature of PVA and PVP K30 promoted water uptake and matrix hydration, while PEG 400 improved flexibility and water penetration. Swelling was influenced by polymer concentration; higher PVA and PVP K30 levels generally increased water uptake, although excessively dense polymer networks slightly restricted swelling. Overall, the formulations showed adequate hydration, supporting microneedle dissolution and drug diffusion.



Figure 6: Swelling index analysis of Patches

Table 13: Determination of the swelling index

Formulation	Swelling Index (%)
F1	162.84 $\pm$ 3.26
F2	171.52 $\pm$ 3.14
F3	192.36 $\pm$ 2.88
F4	214.18 $\pm$ 3.05
F5	176.94 $\pm$ 3.32
F6	219.64 $\pm$ 2.96

F7	223.52 ± 3.18
F8	228.73 ± 2.94
F9	196.84 ± 3.06
F10	183.48 ± 3.24
F11	206.18 ± 3.01
F12	201.84 ± 2.97
F13	204.26 ± 3.08

(Mean ± SD, n = 3)

Among the factorial batches, B8 demonstrated the highest swelling index ($228.73 \pm 2.94\%$), indicating superior hydration capacity and optimum polymer interaction with the aqueous medium. The combination of higher concentrations of PVA, PVP K30, and PEG 400 produced a polymeric matrix that was both highly hydrophilic and mechanically stable. This facilitated efficient water absorption without compromising patch integrity. The centre point formulations also demonstrated satisfactory swelling behaviour, thus confirming the reproducibility of the formulation process. Nevertheless, B8 demonstrated the most favourable swelling characteristics among all the factorial formulations.

15. *In- Vitro* Drug Release study

The *in vitro* drug release of Charantin and Gymnemic acid from the microneedle-assisted transdermal patches (B1–B13) was evaluated using a Franz diffusion cell with phosphate buffer (pH 7.4) at $37 \pm 0.5^\circ\text{C}$. Samples were withdrawn at predetermined intervals and analysed by UV-visible spectroscopy. All formulations showed controlled and progressive drug release over 24 h, with cumulative release ranging from $84.36 \pm 1.42\%$ to $98.64 \pm 0.78\%$. An initial rapid release was attributed to hydration and dissolution of the hydrophilic polymers, followed by sustained release through the hydrated polymer network. Higher PVA concentrations formed a denser matrix and slightly slowed drug diffusion, whereas PVP K30 promoted hydration and pore formation, enhancing release. PEG 400 further improved water penetration and microneedle dissolution through its plasticising effect. Thus, the combined polymer composition influenced the overall drug release behaviour of the formulations.





Figure 7: *In-vitro* drug release study of Formulated Microneedle-Assisted Transdermal Patches

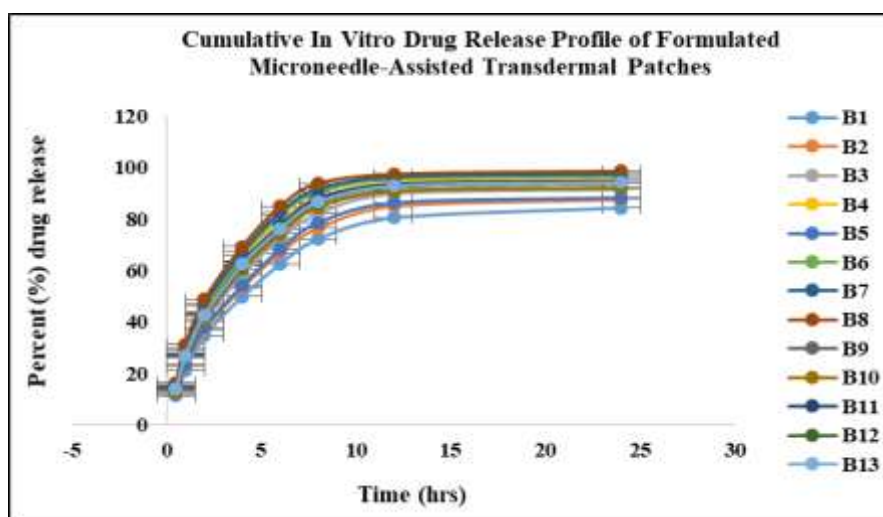


Figure 8: Cumulative In Vitro Drug Release Profile of Formulated Microneedle-Assisted Transdermal Patches

Table 14: Data for Cumulative In Vitro Drug Release Profile of Formulated Microneedle Transdermal Patches

Time (hrs)	B1	B2	B3	B4	B5	B6
0.5	11.26±0.28	12.18±0.26	13.52±0.24	14.86±0.22	12.34±0.27	15.24±0.21
1	21.34±0.34	22.86±0.31	25.68±0.29	27.92±0.27	23.46±0.32	29.36±0.26
2	34.62±0.42	36.84±0.39	40.26±0.36	43.78±0.34	37.84±0.40	45.96±0.32
4	49.84±0.56	53.18±0.52	58.42±0.48	63.74±0.45	54.16±0.53	65.82±0.43
6	62.48±0.68	66.82±0.63	72.46±0.58	78.84±0.54	68.24±0.65	81.36±0.50
8	72.18±0.82	76.34±0.76	82.18±0.71	88.26±0.66	78.54±0.78	90.46±0.60
12	80.46±1.02	84.92±0.94	89.84±0.88	94.36±0.82	86.28±0.96	95.68±0.72
24	84.36±1.42	87.54±1.26	91.42±1.08	95.64±0.92	88.28±1.18	96.84±0.84

(Mean ± SD, n = 3)

Time (hrs)	B7	B8	B9	B10	B11	B12	B13
0.5	15.82±0.20	16.46±0.18	13.84±0.23	12.72±0.25	14.38±0.23	13.96±0.22	14.12±0.22
1	30.12±0.25	31.44±0.24	26.42±0.28	24.18±0.30	27.36±0.28	26.84±0.27	27.04±0.27
2	46.82±0.31	48.74±0.29	41.36±0.35	39.14±0.38	43.26±0.35	42.54±0.34	42.86±0.34
4	67.24±0.41	69.36±0.39	60.46±0.46	56.82±0.50	63.18±0.46	61.82±0.45	62.36±0.45
6	82.74±0.48	84.92±0.45	74.18±0.56	70.42±0.60	77.34±0.55	75.82±0.54	76.18±0.54
8	91.68±0.57	93.84±0.54	84.72±0.68	81.34±0.72	87.96±0.66	86.28±0.64	86.64±0.65
12	96.46±0.69	97.42±0.64	90.84±0.84	88.16±0.89	93.54±0.82	92.48±0.80	92.84±0.81
24	97.52±0.82	98.64±0.78	92.18±1.02	89.74±1.16	94.82±0.98	93.96±0.94	94.24±0.91

(Mean ± SD, n = 3)

Among the factorial batches, B8 demonstrated the most significant cumulative drug release ($98.64 \pm 0.78\%$) after 24 hours. The enhanced release behaviour of B8 can be ascribed to the optimal equilibrium between the concentrations of PVA, PVP K30, and PEG 400, which resulted in the formation of a polymeric matrix with sufficient mechanical strength, expeditious hydration, and efficient diffusion pathways for the incorporated phytoconstituents. Formulations containing comparatively lower polymer concentrations exhibited faster initial hydration but slightly reduced total drug release because of lower matrix stability. In contrast, excessively dense matrices slowed drug diffusion. The centre point batches exhibited consistent release profiles, thus confirming the reproducibility of the formulation process. However, B8 demonstrated the best overall release characteristics among the factorial formulations.

16. Determination of Flux

The transdermal flux of Charantin and Gymnemic acid from the microneedle-assisted patches (B1–B13) was evaluated using a Franz diffusion cell. Steady-state flux was determined from the slope of the linear portion of the cumulative drug permeation versus time plot and ranged from 162.84 ± 3.12 to 218.76 ± 2.48 $\mu\text{g}/\text{cm}^2/\text{h}$. Variations in flux were mainly influenced by PVA, PVP K30, and PEG 400 concentrations, which affected matrix hydration, swelling, microneedle dissolution, and drug diffusion. PVA and PVP K30 promoted hydration and the formation of diffusion pathways, while PEG 400 enhanced polymer flexibility and permeability. Lower polymer concentrations resulted in reduced flux due to weaker matrix integrity, whereas excessively dense matrices could restrict drug diffusion. Overall, an appropriate balance between mechanical strength and polymer hydration was essential for achieving efficient drug permeation.



Figure 9: Flux study by using Franz diffusion cell

Table 15: Data for Flux study

Formulation	Flux ($\mu\text{g}/\text{cm}^2/\text{h}$)
F1	162.84 ± 3.12
F2	168.52 ± 2.96
F3	181.74 ± 2.84
F4	198.36 ± 2.72
F5	171.86 ± 3.04
F6	207.42 ± 2.61
F7	213.58 ± 2.54
F8	218.76 ± 2.48
F9	186.42 ± 2.81
F10	176.64 ± 2.94
F11	201.82 ± 2.66
F12	194.76 ± 2.74
F13	197.34 ± 2.69

(Mean $\pm$ SD, n = 3)

Among the factorial batches, B8 demonstrated the highest steady-state flux ($218.76 \pm 2.48 \mu\text{g}/\text{cm}^2/\text{h}$), indicating the most efficient permeation of Charantin and Gymnemic acid across the membrane. The enhanced permeation performance of B8 can be ascribed to the balanced concentrations of PVA, PVP K30, and PEG 400, which resulted in microneedles characterised by optimal mechanical integrity, rapid hydration, controlled dissolution, and efficient drug diffusion. The centre point batches also demonstrated satisfactory permeation profiles with minimal variability, thus confirming the reproducibility of the formulation process. Nevertheless, B8 exhibited the most favourable permeation characteristics among the factorial formulations.

Experimental design process

The formulation variables and responses obtained from the 2³ full factorial design with five centre-point runs are presented in the design matrix. PVA (10–20%), PVP K30 (10–20%), and PEG 400 (5–15%) were selected as independent variables, while cumulative drug release and flux were the measured responses. The results showed that changes in polymer and plasticizer concentrations influenced both drug release and permeation, with cumulative release ranging from 84.36% to 98.64% and flux from 162.84 to 218.76 µg/cm²/h. Among the factorial batches, B8 (20% PVA, 10% PVP K30, and 15% PEG 400) showed the highest drug release (98.64%) and flux (218.76 µg/cm²/h), suggesting a favourable balance between matrix hydration, microneedle dissolution, and drug diffusion. Comparable responses among the centre-point batches confirmed the reproducibility of the experimental design. Overall, the factorial design provided a reliable basis for response surface analysis and numerical optimization using Design-Expert® software.

Table 16: Experimental design batches of Micro-needle-Assisted Transdermal Patches with obtained response

		Factor 1	Factor 2	Factor 3	Response 1	Response 2
Std	Run	A:Concentration of Polyvinyl Alcohol (PVA)	B:Concnetration of Polyvinylpyrrolidone K 30 (PVP K30)	C:Concentration of Polyethylene Glycol 400 (PEG 400)	Cumulative drug release	Flux
		%	%	%	(%)	(µg/cm ² /h)
1	1	10	10	5	84.360	162.84
5	2	10	10	15	87.540	168.52
12	3	15	15	10	91.420	181.74
8	4	20	20	15	95.640	198.36
7	5	10	20	15	88.280	171.86
13	6	15	15	10	96.840	207.42
4	7	20	20	5	97.520	213.58
6	8	20	10	15	98.640	218.76
10	9	15	15	10	92.180	186.42
3	10	10	20	5	89.740	176.64
2	11	20	10	5	94.820	201.42
9	12	15	15	10	93.960	194.76
11	13	15	15	10	94.240	197.34

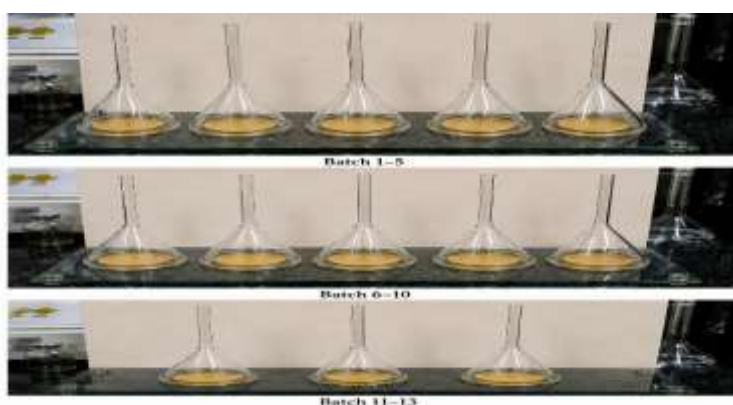


Figure 10: B1-B13 batches of Microneedle-Assisted Transdermal Patch formulation

Evaluation of experimentally Designed Micro-needle-Assisted Transdermal Patches

CONCLUSION

Dissolving microneedle-assisted transdermal patches containing Charantin and Gymnemic acid were successfully developed using a beeswax mould casting method. The combination of PVA, PVP K30, and PEG 400 produced patches with satisfactory microneedle morphology, mechanical integrity, flexibility, drug content uniformity, swelling capacity, and in vitro drug-release characteristics.

Among the preliminary trial batches, TB2, containing 5 mg each of Charantin and Gymnemic acid, demonstrated the most favourable balance of drug loading, microneedle formation, demoulding behaviour, mechanical strength, flexibility, drug content uniformity, and cumulative drug release. Consequently, TB2 was selected as the preliminary optimized formulation for further formulation development.

The [2³] factorial design demonstrated that changes in the concentrations of PVA, PVP K30, and PEG 400 influenced the release and permeation behaviour of the microneedle patches. Formulation B8, containing 20% PVA, 10% PVP K30, and 15% PEG 400, produced the highest cumulative drug release of $98.64 \pm 0.78\%$ and the highest transdermal flux of $218.76 \pm 2.48 \mu\text{g}/\text{cm}^2/\text{h}$. It also exhibited favourable thickness, folding endurance, surface pH, drug content uniformity, and swelling characteristics.

Overall, the results suggest that the optimized beeswax mould-based dissolving microneedle patch is a promising platform for the transdermal delivery of Charantin and Gymnemic acid. Further studies, including skin irritation testing, stability studies, pharmacokinetic evaluation, and in vivo efficacy assessment, are required to confirm its safety, stability, and therapeutic performance.

CONFLICT OF INTEREST Declared None

REFERENCES

- [1] Badhe RV, Adkine D, Godse A. Development of polylactic acid and bovine serum albumin-layered-coated chitosan microneedles using novel bees wax mould. *Turk J Pharm Sci.* 2021;18(3):367-375. doi:10.4274/tjps.galenos.2020.47897.
- [2] Mugo S, Robertson S, Lu W. Transdermal microneedle patch for antioxidants release. *Discov Mater.* 2025;5:64. doi:10.1007/s43939-025-00240-8.
- [3] Dharadhar S, Majumdar A, Dhoble S, Patravale V. Microneedles for transdermal drug delivery: a systematic review. *Drug Dev Ind Pharm.* 2019;45(2):188-201. doi:10.1080/03639045.2018.1539497.
- [4] Waghule T, Singhvi G, Dubey SK, Pandey MM, Gupta G, Singh M, et al. Microneedles: a smart approach and increasing potential for transdermal drug delivery system. *Biomed Pharmacother.* 2019;109:1249-1258. doi:10.1016/j.biopha.2018.10.078.
- [5] Aich K, Singh T, Dang S. Advances in microneedle-based transdermal delivery for drugs and peptides. *Drug Deliv Transl Res.* 2022;12(7):1556-1568. doi:10.1007/s13346-021-01056-8.
- [6] Dalvi M, Kharat P, Thakor P, Bhavana V, Singh SB, Mehra NK. Panorama of dissolving microneedles for transdermal drug delivery. *Life Sci.* 2021;284:119877. doi:10.1016/j.lfs.2021.119877.
- [7] Halder J, Gupta S, Kumari R, Gupta GD, Rai VK. Microneedle array: applications, recent advances, and clinical pertinence in transdermal drug delivery. *J Pharm Innov.* 2021;16(3):558-565. doi:10.1007/s12247-020-09460-2.
- [8] Jana BA, Wadhwani AD. Microneedle: future prospect for efficient drug delivery in diabetes management. *Indian J Pharmacol.* 2019;51(1):4-10. doi:10.4103/ijp.IJP_16_18.
- [9] Roy SK, Ali KA, Biswas M, Dey A, Bishal A, Kuiry A. Microneedles: an efficient technique to increase transdermal drug delivery system. *Curr Drug Deliv.* 2025;22(7):878-894. doi:10.2174/0115672018301931240624072453.
- [10] Ansari Yaasir Ahmed, Farooque Mohd, Ahmad Anwar, Azhruddin Md, Sayyed M. Phytochemical Investigation Studies of Anti-Diabetic Phytochemicals of Plant *Murraya Koenigii* (Linn.) Spreng Belonging to Family Rutaceae. *International Journal of Research in Advent Technology.* 2019;7(4S):45-49.

- [11] Deore Hemant Vinayak, Bhandari Harshal S, Ahire Vinod S, Deshmukh Swapnil B, Patil Jayashree A, Gomase Pravin V, Touseef Begum, Qazi Shoeb, Meman Rahil, Ansari Yaasir, Ansari Vaseem Ahamad, Ansari Mohd Razi. Evaluation of Hepatoprotective activity of *Caesalpinia bonduc* (L.) Roxb. On experimentally induced liver damage in animals. *Journal of Chemical Health Risks*. 2024;14(1):119-124.
- [12] Huang X., Wang L., Guo H., et al. Macrophage membrane-coated nanovesicles for dual-targeted drug delivery to inhibit tumor and induce macrophage polarization. *Bioactive Materials*. 2023;23():69-79. doi:10.1016/j.bioactmat.2022.09.027
- [13] Deore Hemant Vinayak, Deore Harshalkumar Vinayak, Pawar Pallavi Pandit, Qazi Shoeb, Makrani Shaharukh, Farooq Syed Umar, Ansari Mohd Razi, Ansari Yaasir. The effect of *Hydnocarpus Laurifolia* Seeds extract on Blood Glucose in Streptozotocin-induced Diabetic Rats. *Bulletin of Environment, Pharmacology and Life Sciences*. 2023;12(10):217-221.
- [14] Priya S, Tomar Y, Desai VM, Singhvi G. Enhanced skin drug delivery using dissolving microneedles: a potential approach for the management of skin disorders. *Expert Opin Drug Deliv*. 2023;20(6):721-738. doi:10.1080/17425247.2023.2190095.
- [15] Dabholkar N, Gorantla S, Waghule T, Rapalli VK, Kothuru A, Goel S, et al. Biodegradable microneedles fabricated with carbohydrates and proteins: revolutionary approach for transdermal drug delivery. *Int J Biol Macromol*. 2021;170:602-621. doi:10.1016/j.ijbiomac.2020.12.177.
- [16] Prabhu A, Baliga V, Shenoy R, Dessai AD, Nayak UY. 3D printed microneedles: revamping transdermal drug delivery systems. *Drug Deliv Transl Res*. 2025;15(2):436-454. doi:10.1007/s13346-024-01679-7.
- [17] Biswas AA, Dhondale MR, Agrawal AK, Serrano DR, Mishra B, Kumar D. Advancements in microneedle fabrication techniques: artificial intelligence assisted 3D-printing technology. *Drug Deliv Transl Res*. 2024;14. doi:10.1007/s13346-023-01510-9.
- [18] Devangan S, Varghese B, Johnny E, Gurram S, Adela R. The effect of *Gymnema sylvestre* supplementation on glycemic control in type 2 diabetes patients: a systematic review and meta-analysis. *Phytother Res*. 2021;35(12):6802-6812. doi:10.1002/ptr.7265.
- [19] Pothuraju R, Sharma RK, Chagalamarri J, Jangra S, Kavadi PK. A systematic review of *Gymnema sylvestre* in obesity and diabetes management. *J Sci Food Agric*. 2014;94(5):834-840. doi:10.1002/jsfa.6458.
- [20] Khan J, Sheoran S, Khan W, Panda BP. Metabolic differentiation and quantification of gymnemic acid in *Gymnema sylvestre* (Retz.) R.Br. ex Sm. leaf extract and its fermented products. *Phytochem Anal*. 2020;31(4):488-500. doi:10.1002/pca.2912.
- [21] Ahamad J, Amin S, Mir SR. Simultaneous quantification of gymnemic acid as gymnemagenin and charantin as β -sitosterol using validated HPTLC densitometric method. *J Chromatogr Sci*. 2015;53(7):1203-1209. doi:10.1093/chromsci/bmu166.
- [22] Nagella P, Dandin VS, Murthy HN. Production of gymnemic acid from cell suspension cultures of *Gymnema sylvestre*. *Methods Mol Biol*. 2016;1391:229-239. doi:10.1007/978-1-4939-3332-7_16.
- [23] Kahksha, Alam O, Naaz S, Sharma V, Manaitiya A, Khan J, et al. Recent developments made in the assessment of the antidiabetic potential of gymnema species—from 2016 to 2020. *J Ethnopharmacol*. 2022;286:114908. doi:10.1016/j.jep.2021.114908.
- [24] Grover JK, Yadav SP. Pharmacological actions and potential uses of *Momordica charantia*: a review. *J Ethnopharmacol*. 2004;93(1):123-132. doi:10.1016/j.jep.2004.03.035.
- [25] Kaushik U, Joshi SC. Formulation and evaluation of *Momordica charantia* fruit extract based transdermal drug delivery against diabetes. *Biomed Ther Lett*. 2024;1(1):905. doi:10.62110/sciencein.btl.2024.v11.905.
- [26] Rizvi SI, Mishra N. Traditional Indian medicines used for the management of diabetes mellitus. *J Diabetes Res*. 2013;2013:712092. doi:10.1155/2013/712092.