RESEARCH PAPER

Design, Optimization and Evaluation of a Thermo responsive Hydrogel Loaded with Berberine Hydrochloride for Enhanced Wound Healing Therapy

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ABSTRACT

Background: Wound healing is a multifaceted biological process that requires an optimal microenvironment to facilitate cell proliferation, angiogenesis, and tissue regeneration while preventing infection. Chronic wounds, including diabetic ulcers and burns, present challenges due to persistent inflammation and delayed healing. In this study, a thermoresponsive hydrogel loaded with berberine hydrochloride was designed, optimized, and evaluated for enhanced wound healing therapy.

Methods: Pre-formulation studies confirmed the identity, solubility, and stability of berberine hydrochloride, with λ max at 345 nm and a melting point of 204 ± 0.1 °C. Hydrogels were prepared using Carbopol 940 P and propylene glycol via a simple polymer dispersion technique, with formulation optimization carried out using a central composite design. Viscosity and in-vitro drug release were selected as key response variables, and the optimized hydrogel exhibited a viscosity of 10,969 cP and 97.56% drug release over 270 minutes.

Results: Physicochemical characterization demonstrated suitable pH, homogeneity, spreadability, extrudability, and stability over three months under accelerated conditions. The optimized thermoresponsive hydrogel maintained a sustained release of berberine hydrochloride, providing a moist wound environment and potential antimicrobial, anti-inflammatory, and regenerative benefits.

Conclusion: These results suggest that the developed thermoresponsive hydrogel is a promising candidate for effective wound healing therapy.

Keywords: Thermoresponsive hydrogel, Berberine hydrochloride, wound healing, central composite design, Drug delivery, In-vitro drug release, Carbopol 940 P, Optimization, Chronic wounds

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INTRODUCTIONS

Wound healing is a complex and dynamic biological process involving overlapping phases including haemostasis, inflammation, proliferation, and tissue remodelling. Effective wound management requires an ideal environment that promotes cell proliferation, angiogenesis, and extracellular matrix deposition while preventing infection and excessive inflammation. However, chronic wounds such as diabetic ulcers, pressure sores, and burn injuries often exhibit delayed healing due to persistent inflammation, microbial contamination, and impaired tissue regeneration. Therefore, the development of advanced drug delivery systems capable of providing sustained therapeutic action and maintaining a moist wound environment has become an important area of pharmaceutical research (Boateng *et al*., 2008).

Hydrogels have emerged as promising wound dressing materials due to their high water content, biocompatibility, and ability to mimic the natural extracellular matrix. In particular, temperature-sensitive hydrogels have attracted considerable attention because they undergo sol–gel transition in response to temperature changes. These systems remain in a liquid state at room temperature, allowing easy application, and subsequently form a gel at physiological temperature, enabling localized and sustained drug delivery at the wound site (Klouda & Mikos, 2008). Such thermoresponsive systems can improve drug retention, increase therapeutic efficacy, and minimize systemic side effects.

Berberine hydrochloride is a natural isoquinoline alkaloid isolated from plants such as Berberis species and has been widely investigated for its diverse pharmacological activities including antimicrobial, anti-inflammatory, antioxidant, and wound healing properties. Several studies have reported that berberine can promote fibroblast proliferation, enhance collagen deposition, and accelerate re-epithelialization, thereby facilitating wound repair. Additionally, its antimicrobial activity against various pathogenic microorganisms helps prevent infection in the wound microenvironment (Imenshahidi & Hosseinzadeh, 2019).

MATERIAL AND METHODS

Berberine hydrochloride are purchase from Kshipra Biotech Private Limited, Dewas, Carbopol 940, Triethanolamine (TEA), Propyl glycol, Methyl paraben, Propyl paraben, Propyl paraben, Potassium dihydrogen phosphate, Sodium hydroxide are obtained from Loba Chemie Pvt Ltd Mumbai, MH, Dialysis membrane 70 are purchase from HI Media Pvt Ltd Thane, MH.

PREFORMULATION STUDY

1. Identifications and Authentication

a. UV Spectrophotometric Studies

Accurately weighed 10 mg of the drug was dissolved in 10 mL of water in a volumetric flask. An aliquot of 0.2 mL of this solution was further diluted to 10 mL with the same solvent to obtain a suitable dilution. The prepared solution was scanned in the 200–400 nm range using a UV–Visible spectrophotometer and compared with the standard. (Imenshahidi, M., & Hosseinzadeh, H. 2019).

Table No. 1: U.V Spectrum Peaks of Berberine

S. No	Peaks (λmax)	Abs	Description
1	345 nm	0.363	Berberine HCL
2	262 nm	0.399	Other peak
3	288 nm	0.417	Other peak

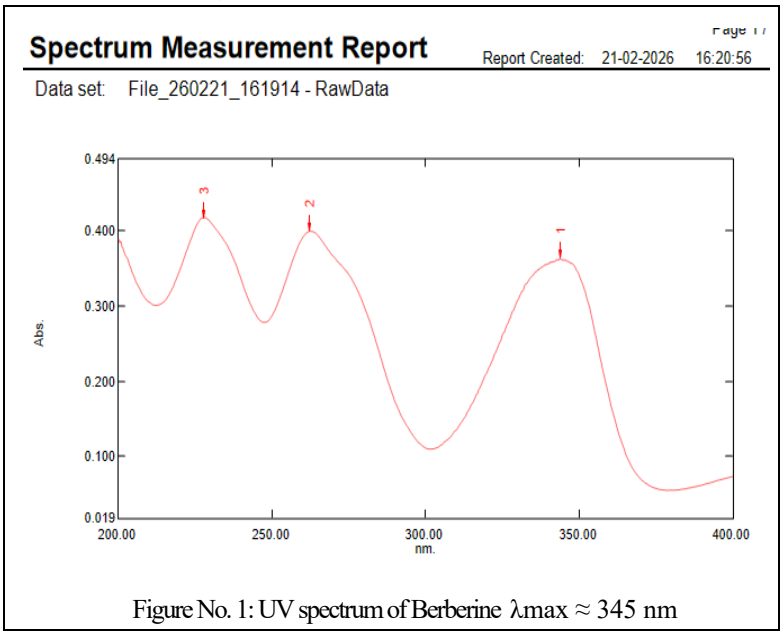


Figure No. 1: UV spectrum of Berberine λmax ≈ 345 nm

2. Physical Descriptions

Berberine hydrochloride is a yellow to bright yellow crystalline powder with a characteristic bitter taste and is generally odorless. The drug is slightly hygroscopic and stable under normal storage conditions, but it should be protected from light and moisture to prevent degradation. It is soluble in water, phosphate buffer pH 7.4 and polar organic solvents such as methanol and ethanol, while showing poor solubility in non-polar solvents. (Imenshahidi, M., & Hosseinzadeh, H. 2019).

Figure No. 2: Physical Appearance of Drug

3. Solubility Analysis

The solubility of Berberine hydrochloride was determined using the equilibrium shake-flask method. An excess amount of drug was added to 10 mL of different solvents and the mixtures were shaken at 25 ± 1 °C for 24 h to reach equilibrium. The solutions were then centrifuged and filtered, and the concentration of dissolved drug was analyzed using a UV–Visible spectrophotometer at λmax ≈ 345 nm after suitable dilution. The solubility was calculated from the calibration curve and expressed in mg/ml. (Higuchi, T., & Connors, K. A. 1965).

- **Determinations of Solubility of drug in various solvents**

Table No. 2: Solubility of Berberine hydrochloride in Different Solvents (UV–Visible Method)

S. No.	Solvent	Solubility (mg/ml)	Solubility Description
1	Water	1.0 ± 0.05	Soluble
2	Methanol	15.4 ± 0.60	Freely soluble
3	Ethanol	5.2 ± 0.30	Soluble
4	Dimethyl sulfoxide	22.5 ± 0.85	Freely soluble
5	Acetone	0.18 ± 0.02	Slightly soluble
6	Dichloromethane	0.12 ± 0.01	Slightly soluble
7	Chloroform	<0.05	Insoluble
8	Ethyl acetate	0.25 ± 0.03	Slightly soluble

9	Phosphate buffer pH 6.8	1.6 ± 0.08	Moderately soluble
10	Phosphate buffer pH 7.4	3.4 ± 0.07	Good soluble
11	HCL buffer pH 1.2	2.3 ± 0.10	Good soluble

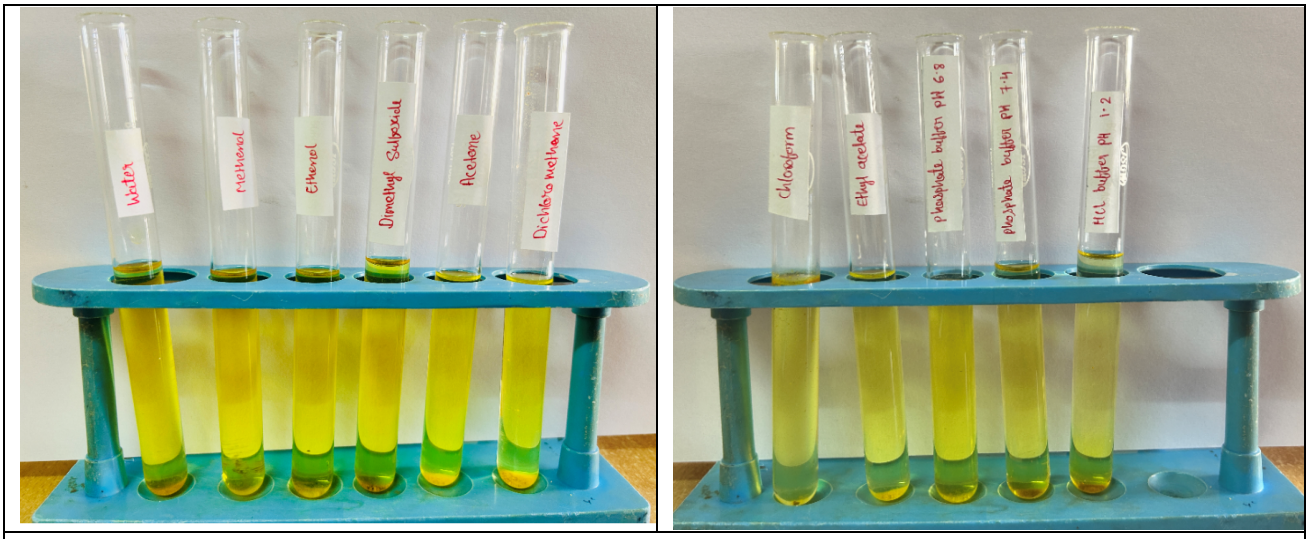


Figure No. 3. Solubility study of Berberine hydrochloride in different solvents.

4. Melting Point Range The melting point of Berberine hydrochloride was determined using the capillary tube method. A small quantity of finely powdered drug was filled into a capillary tube and placed in a melting point apparatus, and the temperature was gradually increased until the sample completely melted. The observed melting point was recorded and compared with reported literature values to confirm drug purity (Aulton & Taylor, 2018; Indian Pharmacopoeia).

Table No. 3: Melting point of Berberine hydrochloride (n=3)

S. No.	Parameter	Observation
1	Method Used	Open Capillary Method
2	Reported Melting Point	204 - 208 °C
3	Observed Melting Point	204 ± 0.1 °C

5. Calibration Curve by U.V Spectrophotometer

A primary stock solution (1000 µg/mL) was prepared by dissolving 10 mg of Berberine hydrochloride in 10 mL of phosphate buffer (pH 7.4). From this solution, a secondary stock solution (100 µg/mL) was prepared by diluting 10 mL to 100 mL with the same buffer. Appropriate aliquots were further diluted to obtain working concentrations of 10, 20, 30, 40, and 50 µg/ml, and the absorbance of each solution was measured at λmax ≈ 345 nm using a UV–Visible spectrophotometer. A calibration curve was plotted between absorbance and concentration, which demonstrated good linearity within the selected concentration range. (Beckett, A. H., & Stenlake, J. B. 2004).

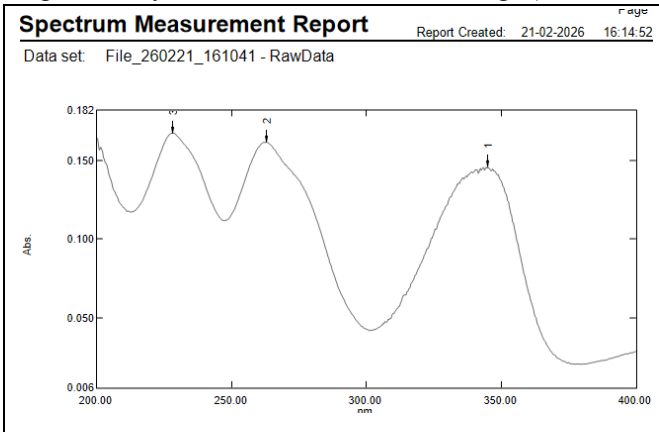


Figure No. 4: UV spectrum of 10 µg/ml

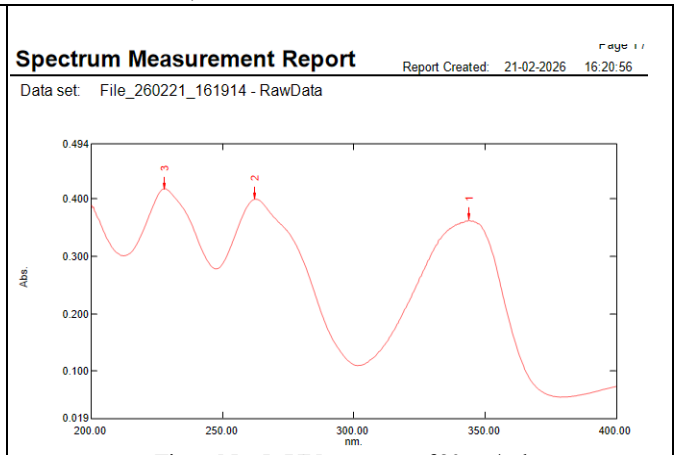


Figure No. 5: UV spectrum of 20 µg/ml

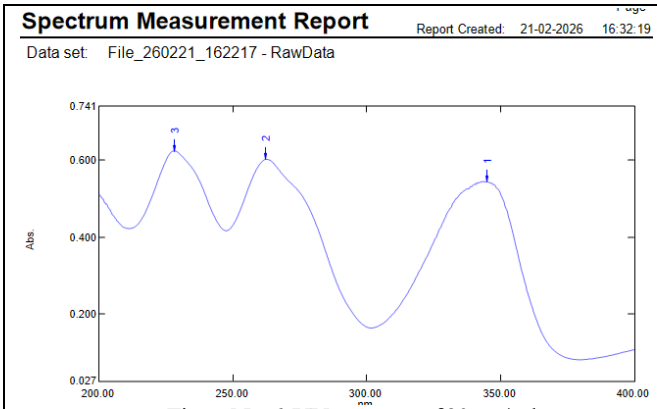


Figure No. 6: UV spectrum of 30 µg/ml

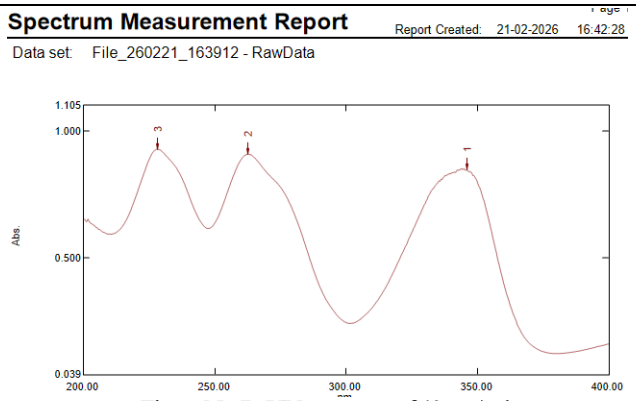


Figure No.7: UV spectrum of 40 µg/ml

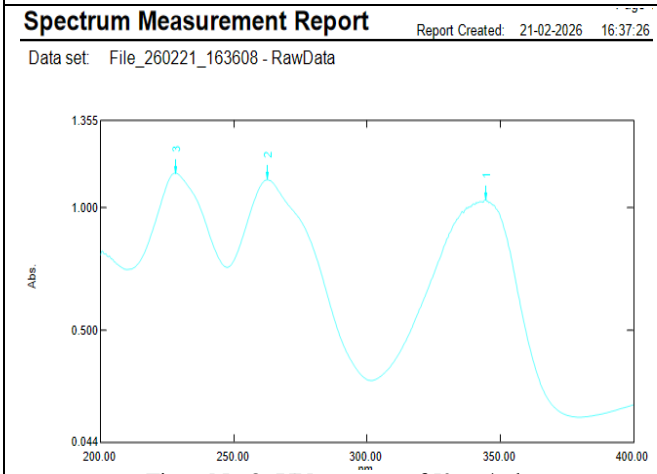


Figure No. 8: UV spectrum of 50 µg/ml

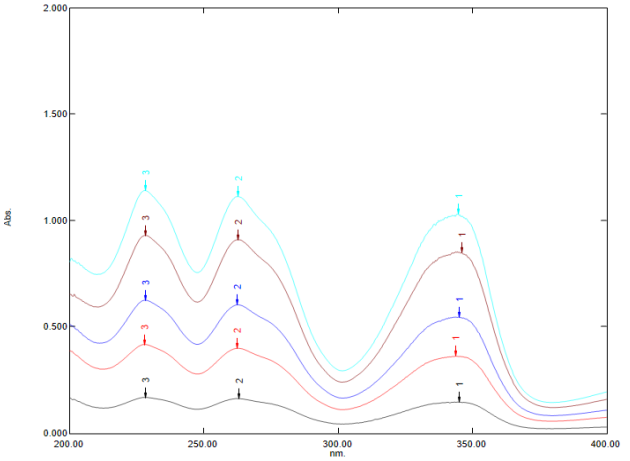


Figure No. 9: UV Overlay spectrum of 10, 20, 30, 40, 50 µg/ml

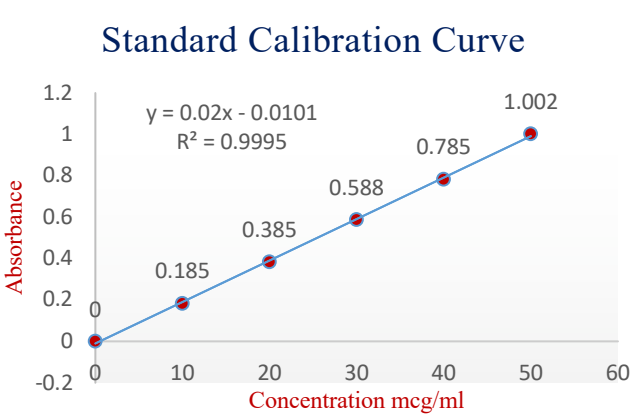


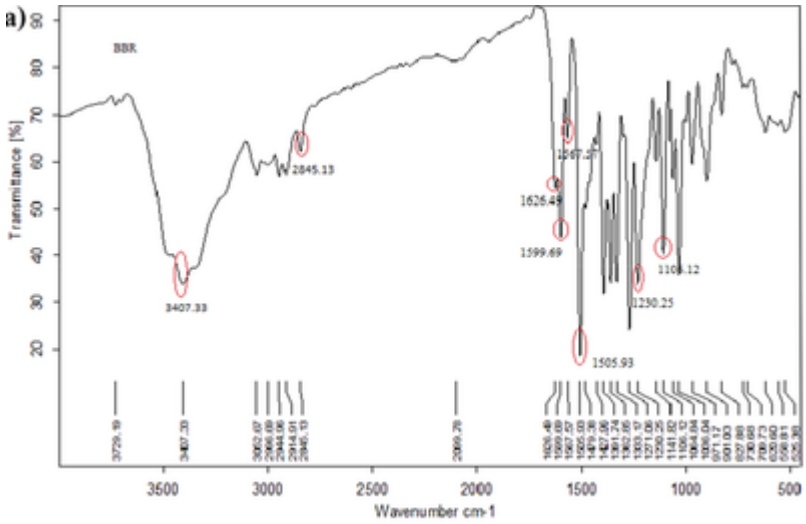
Table No. 4: Calibration Curve of Berberine hydrochloride

S. No.	Concentration (µg /ml)	Mean Absorbance
1.	10	0.140
2.	20	0.363
3.	30	0.575
4.	40	0.848
5.	50	1.033

Figure No. 10: Standard Calibration Graph

6. **DEC Study by using FTIR**

Figure No. 11: FTIR Spectra of Berberine Hydrochloride



The FTIR spectrum of Berberine hydrochloride was recorded using the KBr pellet method in the range of 4000–400 cm^{-1} using an FTIR spectrophotometer. A small quantity of the drug was mixed with dry potassium bromide, compressed into a transparent pellet, and placed in the sample holder. The obtained spectrum was analyzed to identify the characteristic functional groups present in the drug molecule. (Silverstein, R. M. *et al*., 2014).

The FTIR spectrum of Berberine hydrochloride shows several characteristic absorption peaks corresponding to its functional groups. Major peaks are typically observed around ~3400 cm^{-1} (O–H stretching), ~3050 cm^{-1} (aromatic C–H stretching), ~1600 cm^{-1} (C=C aromatic stretching), ~1500–1450 cm^{-1} (C=N stretching of isoquinoline ring), and ~1260–1050 cm^{-1} (C–O stretching of methoxy groups). These peaks confirm the presence of aromatic and methoxy functional groups in the berberine structure.

FORMULATIONS AND OPTIMIZATIONS

Optimizations Methodologies

A central composite design was employed to optimize formulation variables using 3D response surface plots generated by the Design-Expert (DOE) software 8 version. The observed responses, viscosity (Y_1) and in-vitro % drug release (Y_2), ranged from 12,800–16,520 cP and 85.5–95.22%, respectively. Response models were developed using coded factor levels to evaluate the effect of formulation variables on viscosity and drug release. (Singh B, *et al*., 2005)

Table No. 5: Selected independent factor and their levels

S. No.	Name of factors	Levels (Concentration of factor)	
		Low	High
1	X1: Conc. of Carbopol 940 P (w/w %)	0.5 %	2%
2	X2: Conc. of Propylene glycol (w/w %)	5 %	10 %

Table 6: Dependent response variable

S. No.	Name of Response	Unit
1	Viscosity	cP
2	*In-vitro* % release	%

Table No.7: Composition of hydrogel of Berberine hydrochloride

Ingredients	F1	F2	F3	F4	F5	F6	F7	F8	F9
Berberine hydrochloride (mg)	50	50	50	50	50	50	50	50	50
Carbopol 940 P (mg)	625	250	250	1000	1000	625	250	625	1000
Propylene glycol (ml)	2.4	4.8	2.4	4.8	2.4	4.8	3.6	3.6	3.6
Methyl paraben (mg)	10	10	10	10	10	10	10	10	10
Propyl paraben (mg)	100	100	100	100	100	100	100	100	100
Triethanolamine (ml)	Q. S	Q. S	Q. S	Q. S	Q. S	Q. S	Q. S	Q. S	Q. S

Distilled water (ml)	100	100	100	100	100	100	100	100	100

Experimental Design

A typical CCD consists of three types of experimental points: **factorial points, axial (star) points, and center points**. The factorial points represent combinations of the independent variables at their high and low levels and are used to estimate the main effects and interactions of the factors. The axial or star points extend beyond the factorial design space and allow the estimation of curvature in the response surface. Center points represent the midpoint of all factors and are usually replicated several times to estimate experimental error and assess the reproducibility of the design. (Bezerra MA, *et al*., 2008)

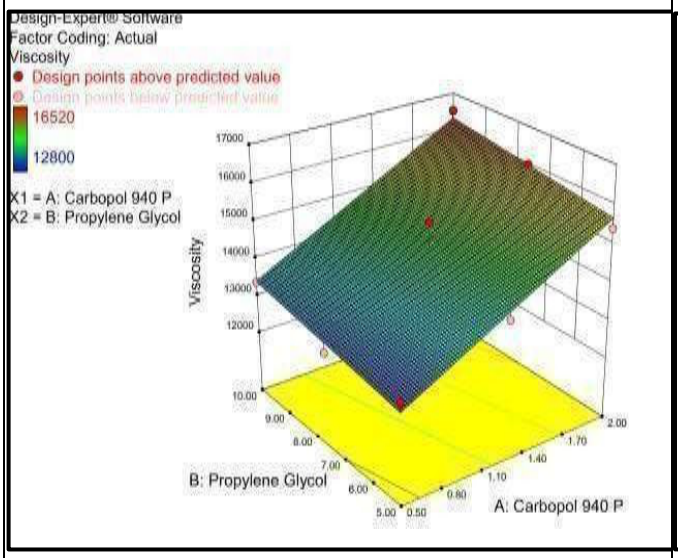


Figure No. 12: 3D response surface plot for evaluating the influence of carbopol 940 P (X1) and propylene glycol (X2) on viscosity.

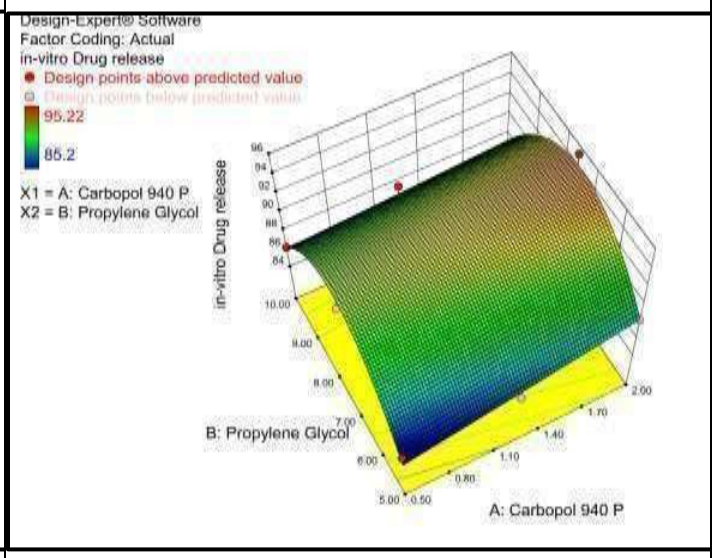


Figure No. 13: 3D response surface plot for evaluating the influence of carbopol 940 P for evaluating the influence of carbopol 940 P (X1) and propylene glycol (X2) in-vitro % drug release.

Effect of independent variable on in - vitro % drug release and viscosity

The effect of carbopol 940 P and Propylene glycol on the viscosity and *in-vitro* % drug release was evaluated and documented. The results indicated that viscosity and *in-vitro* % drug release increased with higher concentrations of both carbopol 940 P and propylene glycol as shown in **table no. 8**.

Table No. 8: Observed response in central composite design for Berberine hydrochloride hydrogel (n=3)

Formulation Code	Independent Variable		Dependent variable	
	Carbopol 940 P (%)	Propylene glycol (%)	Viscosity (cP)	*In-vitro* drug release (%)
F1	1.25	5.00	14000	85.90±1.83
F2	0.50	10.00	13380	86.53±1.60
F3	0.50	5.00	13000	84.77±1.40
F4	2.00	10.00	16250	85.93±1.46
F5	2.00	5.00	15400	85.87±1.38
F6	1.25	10.00	14460	87.57±0.70
F7	0.50	7.50	12800	90.73±0.97
F8	1.25	7.50	15280	92.67 ± 0.6
F9	2.00	7.50	16000	94.66±0.98

All formulations were analyzed using design expert software to determine their fit to various models. It was found that the linear and quadratic

model provided the best fit, with the detailed ANOVA results presented in **table no. 9 and 10**.

Table No. 9: ANOVA for linear model for viscosity

Source	Sum of squares	DF	Mean square	F-value	p-value
Model (Surface Linear Model)	1.337E+007	2	6.686E+006	39.70	0.0003 (significant)
A-Carbopol 940 P	1.273E+007	1	1.273E+007	75.60	0.0001
B-Propylene Glycol	6.403E+005	1	6.403E+005	3.80	0.0991
Residual	1.010E+006	6	1.684E+005	-	-
Cor Total	1.438E+007	8	-	-	-

Table No. 10: ANOVA for quadratic model of *in-vitro* % drug release

Source	**Sum of squares**	**DF**	**Mean square**	**F-value**	**p-value**
Model (Quadratic)	93.05	5	18.61	10.81	0.0390 (Significant)
A-Carbopol 940	10.45	1	10.45	6.07	0.0906
B- Propylene Glycol	0.24	1		0.14	0.7337
AB	1.96	1	1.96	1.14	0.3643
A2	8.000E-004	1	8.000E-004	4.645-004	0.9842
B2	90.39	1	90.39	46.68	0.0064
Residual	5.17	3	1.72	-	-
Cor Total	98.21	8	-	-	-

Data Analysis

The R^2 values for viscosity and in-vitro % drug release were 0.9297 and 0.9474, respectively, indicating a strong model fit. The significance and reliability of the model were evaluated using ANOVA to determine the effect of individual variables and their interactions on the model diagnostic factor (MDF). The obtained F-values for viscosity and in-vitro % drug release were 39.70 and 10.81, respectively, confirming the robustness of the model.

Optimized formulation of Hydrogel

The final optimized formula was obtained from above data which was prepared by taking different concentration of factors X1 and X2 is shown in **table no. 5**. The response of optimized batch that is viscosity (Y1) and in-vitro drug permeation (Y2) was recorded in triplicates is shown in **table no.8** and was validated by confirmation **table no. 11** obtained for optimized batch.

Table No. 11: Final optimized formula for Berberine hydrochloride hydrogel

Carbopol940 P (mg)	Propylene Glycol (ml)	Triethanol amine (ml)	Propyl Paraben(mg)	Drug (mg)	Methyl Paraben (mg)	Water (ml)
1000 mg	2.4	Q.S.	100 mg	50 mg	10 mg	Q.S.

Methods of Preparation

The hydrogel was prepared using the simple polymer dispersion technique. Carbopol 940 P was dispersed in a measured quantity of distilled water under continuous stirring at room temperature until a uniform dispersion was obtained. Propylene glycol was added as a humectant and co-solvent to enhance polymer hydration and drug solubilization. The accurately weighed amount of berberine hydrochloride was then incorporated into the polymer dispersion with continuous stirring to ensure uniform distribution. Methyl paraben and propyl paraben were added as preservatives, and the pH of the formulation was adjusted to neutral (~6.8–7.0) using triethanolamine, which also facilitated gel formation by neutralizing the Carbopol

polymer. The final hydrogel was mixed thoroughly until it convert into homogeneous, smooth gel and allowed to equilibrate at room temperature before further physicochemical evaluation. (Ahmed EM., 2015)

Formulations of Hydrogel

The Berberine hydrochloride hydrogel was formulated using the simple polymer dispersion technique. The prepared hydrogel was Yellowish in color, with a smooth and homogeneous texture. The formulation appeared stable, without any phase separation or visible aggregates, and exhibited a semi-solid consistency suitable for topical application are shown in **figure 14 (a) and (b)**.

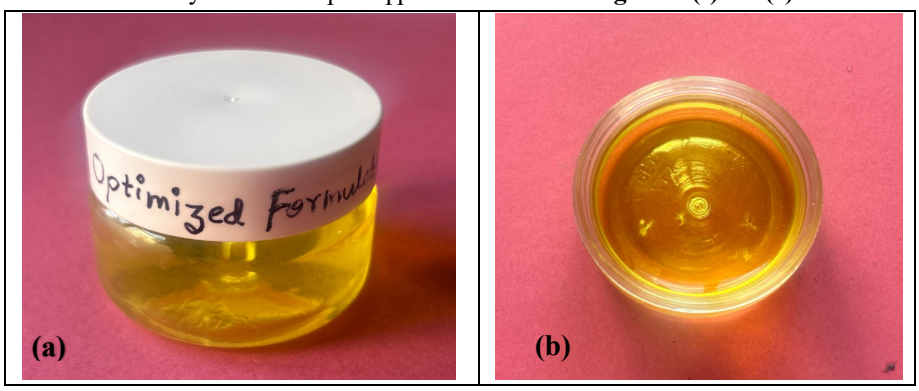


Figure No. 14: Berberine hydrochloride hydrogel (a) side view and (b) top view.

EVALUATION PARAMETER

1. Physical characteristic the prepared hydrogel formulations were inspected visually for their pH, colour, odor homogeneity, consistency, texture and phase separation, as presented in **table no.12**.(Sharma A. *et al*., 2025)

2. Determination of pH- pH of the formulation was determined by using Digital pH meter. The two readings should agree to within the accuracy limits of the meter. The samples were analyzed in triplicate. If slight deviations in pH were note down , it was adjusted to skin pH using drop wise addition of tri-ethanolamine solution (McGlynn, W. 2003)

3. Determination of Viscosity- The viscosity of the hydrogel formulations was determined using Brookfield viscometer with spindle no. 61 at 100 rpm at the temperature of 25 ^{0}C (Monica and Gautami 2014).

4. Determination of Spreadability Two glass slides of standard dimensions (6×2) were selected. The hydrogel formulation selected who have high spreadability and to be determined by placed over one of the slides.

$$\textbf{Spreadability} = m \times l / t$$

Where, S = Spreadability (gcm/sec), m = weight tied to the upper slide (20 grams), l = length of glass slide (6cms), t = time taken in seconds. (Sandeep, D. S. 2020).

5. Washability Study Formulations were applied on the skin and then ease and extent of washing with water were checked manually

6. Extrudability Study The hydrogel formulations were filled into collapsible metal tubes or aluminium collapsible tubes. The tubes were pressed to extrude the material and the extrudability of the formulation was evaluated.

7. Drug Content Drug content analysis was conducted to evaluate the uniform distribution of the drug within the formulations. The results indicated good homogeneity, with drug content values ranging from 87.67 ± 1.53% to 98.50 ± 0.02%, as shown in **table no.13** (Sharma A. *et al*., 2025).

Table No. 12: Physical parameter of formulations and Optimize Formulation (OF)

S. No.	Color and Texture	Odor	Homogeneity	Consistency	Phase Separation	Washability
F1	Yellow & Uniform	Odorless	+++	Excellent	None	+++
F2	Yellow & Uniform	Odorless	+++	Good	None	+++
F3	Yellow & Uniform	Odorless	+++	Average	None	+++
F4	Yellow & Uniform	Odorless	+++	Average	None	+++

F5	Yellow & Uniform	Odorless	+++	Average	None	+++
F6	Yellow & Uniform	Odorless	+++	Good	None	+++
F7	Yellow & Uniform	Odorless	+++	Good	None	+++
F8	Yellow & Uniform	Odorless	+++	Good	None	+++
F9	Yellow & Uniform	Odorless	+++	Excellent	None	+++
OF	Yellow & Uniform	Odorless	+++	Excellent	None	+++

Table No. 13: Characterization of hydrogel formulations and OF (Viscosity, pH, Extrudability, Spreadability, and Drug Content)

S. No.	Viscosity (cP)	Spreadability g.cm/sec	pH at (25-30 ^{0}C)	Drug Content (%)	Extrudability
F1	14000	19.62±0.016	5.94 ± 0.01	97.92± 0.05	+++
F2	13980	20.42 ± 0.62	5.99 ± 0.01	98.50 ± 0.02	++
F3	13000	19.62±0.001	5.07 ± 0.01	96.90 ± 0.01	+++
F4	16520	13.99 ± 0.01	5.54 ± 0.01	97.00 ± 0.10	+++
F5	15400	14.06 ± 0.01	5.42 ±0.01	98.03 ± 0.06	+++
F6	14460	21.92 ± 0.59	5.30 ± 0.02	87.67 ± 1.53	+++
F7	12800	18.25 ± 0.02	5.28 ± 0.01	90.03 ± 0.15	+++
F8	15280	23.90 ± 0.01	5.23 ± 0.01	88.57 ± 0.02	+++
F9	16000	21.20±0.02	6.02 ± 0.01	91.33 ± 0.59	++
OF	10969	24.20 ± 0.10	6.00 ± 0.01	98.29 ± 0.03	+++

8. *In- Vitro* drug release studies

1. The in-vitro % drug release from the prepared hydrogel was tested using a Franz diffusion cell. A dialysis membrane-70, pre-soaked before use, was placed between the donor and receptor compartments. The temperature was maintained at 37 ± 0.5 °C using a circulating water jacket. The hydrogel was applied in the donor compartment, while the receptor compartment was filled with phosphate buffer pH 7.4 to simulate body conditions. At fixed time intervals, small samples (1–3 ml) were withdrawn and replaced with fresh buffer of the same volume to maintain sink conditions. The collected samples were analyzed using a UV–Visible spectrophotometer at 345 nm to determine the amount of drug released over time (Biswas and Majee, 2016). As shown in **table no.14**. (Khoshbakht et al. , Kumar et al., 2019)

Table No. 14: In-vitro % drug release data of hydrogel of berberine hydrochloride (F1-F9) and Optimize formulations (n=3)

S. No.	Time (min)	F1	F2	F3	F4	F5	F6	F7	F8	F9	OF
1	30	14.92	17.90	19.61	26.96	28.09	26.85	16.47	18.42	26.01	26.22
2	60	23.83	29.52	32.95	33.85	38.43	31.19	33.83	24.23	40.73	40.09
3	90	35.71	37.67	40.28	46.51	42.10	35.16	42.25	43.23	43.47	43.50
4	120	47.15	48.57	48.09	47.62	51.61	42.38	49.68	48.52	50.11	56.80
5	150	57.63	56.65	57.81	60.81	62.30	57.77	60.76	59.27	60.84	62.23
6	180	64.10	65.72	61.38	66.47	66.68	66.79	69.77	64.52	68.09	70.25

7	210	71.07	73.79	69.52	71.80	75.71	76.66	77.82	73.62	77.41	78.96
8	240	77.60	73.80	69.52	77.60	86.64	77.82	88.56	88.46	88.07	90.23
9	270	85.90	86.53	84.77	85.93	85.87	87.57	90.73	92.60	94.66	97.56

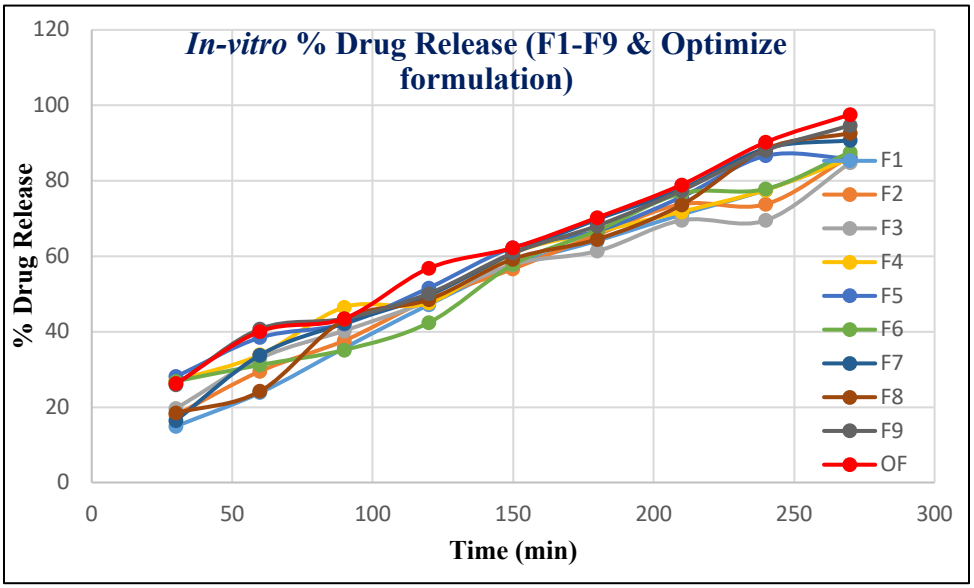


Figure No. 15: In-vitro % Drug Release Profile F1-F9 & Optimize formulation

Validation of optimized batch of hydrogel

The validation data of optimized formulation of hydrogel of Berberine hydrochloride is shown in **table no. 15**, results indicate the closeness with expected values suggested by the central composite design.

Table No. 15: Validation of optimized formula of hydrogel of Berberine hydrochloride

Responses	Result of responses expected	Result of responses found
Viscosity	10971.6	10969.8
In-vitro % Drug Release	90.30 ± 0.02	97.56 ± 0.02

Stability Studies

Stability studies of the hydrogel formulation were performed in according to ICH guidelines. The formulation was stored at 40 ± 2 °C and 75 ± 5 % relative humidity for a period of three months. During the study, parameters including pH, drug content, viscosity, spreadability, and in-vitro drug release were periodically evaluated, as shown in table no.16. (Qindeel *et al.,* 2019).

Table No.16: Stability study of Optimize formulation (OF)

S. No.	Formulation	Months	Spreadability	pH (At 25^{0}C)	Drug Content (%)	Viscosity	% Drug Release
1	Optimize Formulation (OF)	0	24.20 ± 0.10	6.00 ± 0.01	98.29 ± 0.03	10969	97.56
2		1	23.23 ± 0. 23	6.14 ± 0.20	97.34 ± 0.54	11559	96.98
3		2	22.01 ± 0.51	6.52 ± 0.67	97.07 ± 0.44	12349	96.22
4		3	22.57 ± 0.71	6.71 ± 0.77	96.45 ± 0.08	12339	95.77

RESULT AND DISCUSSION

Berberine hydrochloride exhibited characteristic UV absorption at λ max = 345 nm, confirming its identity and suitability for UV–Visible analysis. The drug appeared as a yellow crystalline powder, soluble in water, methanol, ethanol, and phosphate buffer pH 7.4, with a melting point of 204 ± 0.1 °C consistent with literature values. FTIR analysis confirmed the presence of key functional groups (O–H, C–H, C=N, and C–O), indicating chemical integrity of the drug molecule. The calibration curve demonstrated linearity ($R^2 = 0.998$), enabling accurate quantification for in-vitro release studies.

Hydrogels were successfully formulated using the **simple polymer dispersion technique**, producing a smooth, homogeneous, yellowish gel suitable for topical application. Central Composite Design (CCD) was employed to optimize polymer (Carbopol 940 P) and co-solvent (propylene glycol) concentrations. Viscosity ranged from 12,800–16,520 cP, and in-vitro drug release varied from 84.77–94.66%, demonstrating that both independent variables significantly influenced hydrogel properties. Higher Carbopol concentration increased viscosity and reduced drug diffusion, while moderate propylene glycol levels enhanced solubilization and drug release, as supported by 3D response surface plots and ANOVA analysis. The optimized formulation (Carbopol 940 P 1000 mg, propylene glycol 2.4 ml, berberine 50 mg) showed a viscosity of 10,969 cP, pH 6.0, and spreadability 24.2 g·cm/sec, with uniform drug content (98.29 ± 0.03%) and sustained in-vitro release (97.56 ± 0.02% over 270 min). The observed responses closely matched predicted values, validating the optimization process.

All hydrogel formulations were physically stable, with no phase separation, smooth texture, and odorless appearance. pH values were compatible with skin (5.2–6.0), and extrudability and spreadability were satisfactory for topical application. The in-vitro release profile indicated an initial burst followed by sustained release, demonstrating effective drug retention and potential for prolonged therapeutic action. Viscosity and drug release trends confirme the suitability of Carbopol 940 P and propylene glycol concentrations for achieving a balance between mechanical strength and drug diffusion.

Optimized hydrogel exhibited excellent stability under accelerated conditions (40 ± 2 °C, 75 ± 5% RH) for three months. Slightly increases in viscosity and pH were observed, but drug content and release profile remained above 95%, confirming formulation robustness and suitability for practical use.

CONCLUSIONS

The study demonstrates that **thermoresponsive hydrogels** containing **berberine hydrochloride** can be successfully prepared using simple polymer dispersion. The combination of Carbopol 940 P and propylene glycol allows precise tuning of viscosity and drug release. Sustained in-vitro release, uniform drug content, and physical stability indicate the hydrogel's potential as a topical wound healing system. The findings align with previous reports on polymeric hydrogels for enhanced drug retention and therapeutic efficacy.

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