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Original Article

Quality by Design Development of a Thermosensitive *in situ* Gel for Enhanced Ocular Delivery of Carvedilol

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ABSTRACT

Introduction: This research focuses on formulating a thermoresponsive *in situ* gel containing Carvedilol for ocular delivery, employing a Quality by Design (QbD) strategy to improve therapeutic effectiveness in glaucoma management. Carvedilol, known for its β- and α1-blocking effects along with antioxidant activity, holds potential in reducing Intraocular Pressure (IOP) and protecting retinal cells. However, its therapeutic performance is hindered by poor retention and low bioavailability when delivered via standard eye drops. Objectives: To overcome these limitations, a sol-to-gel system was developed, which respond to ocular temperature and improve drug residence time. **Materials and Methods:** A 3^2 factorial design was implemented to assess the influence of polymer concentrations like Pluronic F127 and Pluronic F68 on key formulation parameters such as viscosity and gelation temperature. **Results:** The optimized formulation displayed ideal pH (6.8±0.2), high drug content (97.82±0.4%), and immediate gel formation at eye temperature. *In vitro* studies showed a controlled drug release profile of 97.62% over 8 hr, consistent with Higuchi kinetics. Safety evaluations including Draize and HET CAM tests confirmed the formulation's non-irritant behavior. Stability assessment under ICH Q1A(R2) guidelines indicated the formulation remained stable for at least 30 days. **Conclusion:** The developed *in situ* gel offers a promising, patient-friendly approach to glaucoma treatment by enhancing its ocular delivery.

Keywords: Carvedilol, Glaucoma, *In situ* gel, Ocular delivery, Quality by Design, Sustained release, Thermoresponsive gel.

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INTRODUCTION

Glaucoma is a progressive and irreversible eye condition that affects millions worldwide, primarily due to elevated intraocular pressure (IOP).[1] Effective management of this disease is critical to prevent optic nerve damage and permanent vision loss.[2] Among various therapeutic options, Carvedilol a non-selective beta-blocker with additional alpha-1 blocking and antioxidant properties has emerged as a promising agent for ocular therapy.[3] Despite its potential, the conventional use of Carvedilol through eye drops is challenged by the eye's natural barriers, including rapid tear turnover, blinking, and nasolacrimal drainage, which drastically limit drug retention and absorption at the target site.[4]

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Overview of Ocular Drug Delivery

Successful ocular therapy demands a drug delivery system that ensures prolonged retention at the site of action, enhances permeability, reduces administration frequency, and offers patient comfort.[5] Traditional forms such as solutions and suspensions suffer from low bioavailability, typically less than 5%, owing to anatomical and physiological constraints of the eye.[6] Hence, innovative delivery strategies are needed to overcome these limitations and achieve better therapeutic efficiency.

In situ Gel Systems

In situ gel systems offer a transformative solution in ocular drug delivery. These systems are applied as low-viscosity liquids and transform into gels upon exposure to specific physiological triggers such as pH, temperature, or ionic strength.[7] The transition from sol to gel enhances the drug's contact time with the ocular surface, allowing for sustained release and reduced drainage losses.[8]

Common Trigger Mechanisms

- **pH-Sensitive Gels:** Utilize polymers like Carbopol that remain in liquid form at acidic pH but gel at tear fluid pH (~7.4), enabling localized and prolonged action.[9]
- **Thermosensitive Gels:** Employ polymers such as Poloxamers (e.g., Pluronic F127) that gel at ocular surface temperatures, enhancing drug residence time post-instillation.[10]
- **Ion-Sensitive Gels:** Utilize ionic interactions with tear fluid cations (e.g., Na^+, Ca^{2+}) to initiate gelation using polymers like gellan gum and alginate.[11]

These systems not only enhance drug retention and controlled release but also improve patient adherence by minimizing the need for frequent dosing and reducing systemic side effects.

Rationale for Carvedilol in *in situ* Gels

Carvedilol's multifunctional pharmacological profile β-adrenergic blockade, α1-antagonism, and antioxidant activity makes it particularly suitable for managing glaucoma.[12] It reduces IOP by decreasing aqueous humour production and improving ocular blood flow. Moreover, its antioxidant potential may offer neuroprotective benefits by preventing retinal ganglion cell degeneration, a major concern in glaucoma progression. However, its therapeutic potential is underutilized in traditional dosage forms due to poor solubility and low precorneal retention.

Incorporating Carvedilol into a thermosensitive *in situ* gel can overcome these challenges by

- **Enhancing Ocular Bioavailability:** Extended retention time improves corneal absorption.[13]
- **Reducing Dosing Frequency:** Sustained drug release ensures prolonged IOP control.[14]
- **Minimizing Systemic Absorption:** Localized action reduces adverse effects.[15]

Schematic Mechanism of *in situ* Gel Functionality

- **Pre-Administration:** The formulation exists as a low-viscosity solution for easy instillation.[16]
- **Post-Administration:** Goes under transition sol-to-gel.
- **Thermosensitive:** Gels at ocular temperature (~35-37°C).[17]
- **pH-Sensitive:** Transforms at physiological pH (~7.4).[18]
- **Ion-Sensitive:** Reacts with electrolytes in tear fluid to form gel.[19]
- **Drug Release:** The gel adheres to the ocular surface, enabling sustained drug diffusion.[20]
- **Gel Dissolution:** Eventually breaks down and clears naturally, avoiding residue build up.

QbD Benefits in *in situ* Gel Development

- Increases process understanding and formulation consistency.[21]
- Reduces development costs and time.
- Ensures product safety, stability, and regulatory compliance.
- Enhances patient-centric design with predictable therapeutic outcomes.[22]

MATERIALS AND METHODS

Materials

The materials used in the formulation included Carvedilol, sourced from Med Chem Express, India. Pluronic F127 was obtained from Sigma Aldrich, Mumbai, while both Pluronic F68 and HPMC E15 LV were procured from Loba Chemicals, Mumbai. Benzalkonium Chloride, serving as a preservative, was also supplied by Loba Chemicals. All excipients and reagents used were of pharmaceutical or analytical grade.

Methods

Preformulation Study

Preformulation studies were conducted to evaluate the physicochemical properties, stability, and excipient compatibility of Carvedilol for effective dosage form development.[23]

Organoleptic properties

This includes testing of properties like color, odor, texture.[24]

Melting Point Determination

The melting point of Carvedilol was determined using the open capillary tube method to confirm its identity and purity, with the average value from three trials compared to the standard reference.[25]

Solubility

Solubility of Carvedilol was assessed in various solvents by dissolving the drug in polymer solutions, incubating at 25°C for 72 hr, centrifuging, and analyzing the clear supernatant using a spectrophotometer to determine drug content.[26]

Spectroscopic Analysis

To determine the maximum absorbance wavelength (λ_{max}) of Carvedilol artificial tear fluid, a 1000 µg/mL stock solution was made by dissolving 10 mg of Carvedilol in 10 mL of fluid. A secondary stock of 100 µg/mL was prepared through dilution. Standard solutions ranging from 2-10 µg/mL were then created

and analyzed at 272 nm to construct the calibration curve. The artificial tear fluid was prepared using NaCl, KCl, $NaHCO_3$, Na_2HPO_4, and NaH_2PO_4, with pH adjusted to 7.4. All absorbance measurements were carried out using a UV spectrophotometer. These studies ensured accurate drug quantification in both buffer and tear fluid media.[27]

Compatibility studies

X-ray Diffraction (XRD) analysis was performed to assess the crystalline nature of Carvedilol.[28] The powdered drug sample exhibited sharp and distinct peaks at specific 2θ angles, confirming its crystalline structure, which plays a crucial role in solubility, stability, and bioavailability. Fourier Transform Infrared (FTIR) spectroscopy was conducted using Agilent Cary 630 to identify functional groups and detect any chemical changes. The spectrum showed characteristic peaks consistent with the known structure of Carvedilol, confirming the absence of degradation or incompatibility with formulation excipients.[29] Thermogravimetric Analysis (TGA) evaluated thermal stability by monitoring weight changes with increasing temperature, helping determine moisture content and decomposition profile.[30] Differential Scanning Calorimetry (DSC) was used to investigate melting behavior, crystallinity, and possible interactions. The thermal events observed in the DSC thermogram confirmed the drug's purity and stability, with no significant shifts indicating interaction with excipients.

Quality by Design (QbD)-Based Formulation and Optimization Approach

The Quality by Design (QbD) approach was applied to systematically develop and optimize the Carvedilol-loaded *in situ* gel formulation.[31] This strategy enabled a deeper understanding of the formulation variables and process parameters, ensuring robust product performance. By incorporating QbD principles, the formulation was refined for enhanced stability, efficacy, and consistent quality throughout development.

Quality Target Product Profile

The Quality Target Product Profile (QTPP) was designed to guide the development of Carvedilol *in situ* gel by outlining key attributes such as dosage form, route of administration, and performance criteria.[32] This ensured the formulation met therapeutic goals, regulatory standards, and patient compliance needs.

Critical Quality Attributes (CQA) for Carvedilol Loaded *in situ* Gel

ICH Q8(R2) defines Critical Quality Attributes as key properties controlled to ensure safety and efficacy, with specific targets set for Carvedilol *in situ* gel.[33] is presented in Table 1.

Critical Material Attributes (CMAs) and Process Parameters (CPPs)

The formulation variables for the *in situ* gel were optimized following the Quality by Design (QbD) approach.[34] guided by preliminary research and literature. The critical material attributes (CMAs) identified were:

- Concentration of Pluronic F127 at 16%, 18%, and 20% w/w.
- Concentration of Pluronic F68 at 3%, 4%, and 5% w/w.
- Carvedilol concentration fixed at 10 mg w/v.
- Use of a phosphate buffer system with pH ranging between 6.5 and 6.8.

Critical Process Parameters (CPPs) considered included:

- Stirring speed between 300 and 500 rpm.
- Hydration time ranging from 1 to 2 hr.

These factors were carefully selected to optimize the gel formulation's performance and stability.

Optimization with Experimental Design

A 3^2 full factorial design was employed to systematically optimize the ocular *in situ* gel formulation of Carvedilol.[34] This design allowed the evaluation of the influence of two independent variables concentrations of Pluronic F127 (X1) and Pluronic F68 (X2) on key formulation responses. The dependent variables assessed included the gelling temperature (Y1) and gelling time (Y2), both critical for ensuring suitable gelation under ocular conditions. The study was conducted in the presence of simulated tear fluid to closely mimic physiological conditions. The Design of Experiments (DoE) matrix along with the corresponding results for formulations A1 to A9 is presented in Table 2.

The *in situ* gel was assembled entirely under chilled conditions to protect the temperature-sensitive polymers. First, Pluronic F68 (2% w/v) was weighed and dissolved in about 75 mL of distilled water kept at 4-8°C, stirred gently until clear. Once fully hydrated,

Table 1: Critical Quality Attributes (CQA) Elements.

CQAs	Target value
pH	6.5-7.2
Gelling Temperature	35-37°C
Gelling Time	>10 Sec
Viscosity	50-5000 cP
Drug Content	95-105%
% Drug Release	≥80%
Clarity	Clear
Sterility	No contamination

Table 2: Experimental Design.

Factors	Levels		
Independent Variables	Low (-1)	Medium (0)	High (+1)
X1- Pluronic F127 (%w/v)	16.0	18.0	20.0
X2-Pluronic F68 (%w/v)	3.0	4.0	5.0
Dependent Variables		Goals	
Y1-Gelling Temperature		In range	
Y2-Gelling Time		Minimize	

Pluronic F127 (16-20% w/v, as dictated by the factorial design) was added portion-wise to the cold solution, and the dispersion was left stirring overnight in the refrigerator to obtain a uniform, transparent blend. Meanwhile, Carvedilol was solubilized in a small volume of PEG solution when needed then slowly introduced into the chilled Pluronic mixture with continuous stirring. To raise viscosity and prolong ocular residence, HPMC E15 LV (0.5-1.0% w/v) was sprinkled in gradually, taking care to prevent clumping. After thorough mixing, the batch volume was topped up to 100 mL with cold distilled water, giving a pourable preparation that was stored under refrigeration. At room temperature the formulation remained a free-flowing liquid, but it converted into a soft, viscoelastic gel when warmed to physiological temperature (≈ 35-37°C) shown in Table 3.

Evaluation of Carvedilol *in situ* Gel

Organoleptic Properties

The organoleptic evaluation of the developed ophthalmic *in situ* gels included assessment of appearance, color, odor, and consistency.[35] Clarity was checked under natural light, and odor was evaluated by direct sniffing. Consistency was compared in both liquid form at room temperature and gel form after exposure to simulated ocular conditions (pH 7.4 and tear electrolytes like Na^+ and Ca^{2+}). Any physical instability such as precipitation, phase separation, or visual changes was carefully observed and recorded.

Visual Appearance and Clarity

The appearance and clarity of the samples were evaluated by observing color and checking for any visible particles. Assessments were done against both black and white backgrounds to enhance particle detection white for dark particles and black for light or translucent ones. Color uniformity was also examined to identify any signs of contamination or instability.[36]

Determination of pH

The pH determination of the *in situ* gel was carried out to assess its acidity or alkalinity, which is critical for its stability, efficacy, and compatibility with biological systems.[37] For the test, 10 mL of the gel was transferred into a clean beaker. The electrode of a calibrated digital pH meter was then carefully immersed into the gel to ensure full contact. After allowing the reading to stabilize, the pH value was recorded.

Gelling Capacity Test

The gelling capacity was tested by mixing the formulation with 2 mL freshly prepared artificial tear fluid at 37°C, simulating ocular conditions, and visually observing the sol-to-gel transition and gel stability to assess its suitability for sustained release and retention.[38]

Gelling Temperature Test

The gelling temperature was determined by gradually heating 2 mL of the formulation from 20°C in a controlled water bath at 1°C per minute.[39] The temperature at which the solution visibly thickened and transitioned into a gel was recorded as the gelation point.

Viscosity of Formulated Gel

The viscosity of the gel was measured at room temperature using a Brookfield viscometer with an S-62 spindle, suited for medium to high viscosity samples.[40] The spindle's resistance to rotation within the gel provided viscosity values in centipoise, reflecting the formulation's flow properties before gelation.

In vitro Drug Release Study

The sustained release behaviour of the *in situ* gel was studied using a dialysis method under conditions mimicking the eye. A measured quantity of gel was placed in a dialysis bag (MWCO 12,000-14,000 Da) and suspended in 100 mL of artificial tear fluid (pH 7.4) at 37±0.5°C, stirred at 50 rpm. At hourly intervals for 8 hr, 1 mL samples were withdrawn and replaced with fresh, pre-warmed medium to maintain sink conditions. Drug content in each sample was analyzed using UV spectrophotometry to determine the release profile and evaluate the gel's potential for sustained ocular drug delivery.[41]

Mathematical Modeling and Kinetic Evaluation of *In vitro* Drug Release

The *in vitro* release profile of the carvedilol-loaded *in situ* gel was evaluated using various kinetic models Zero-order, First-order, Higuchi, and Korsmeyer-Peppas. These models helped determine the release kinetics and underlying drug diffusion mechanism.

The cumulative release data were fitted to each model to better understand the drug release behavior from the gel matrix.

Comparative *in vitro* Drug Release Study of Marketed Eye Drop

To evaluate the drug release performance, a comparative *in vitro* drug release study was conducted between the optimized Carvedilol *in situ* gel formulation (Batch A7) and a marketed eye drop formulation. The study was performed under identical conditions using the dialysis bag diffusion technique to ensure consistency. A dialysis membrane was soaked overnight in distilled water prior to use. One milliliter (1 mL) of the marketed eye drop was accurately placed into the dialysis bag, which was then sealed and suspended in 50 mL of simulated tear fluid (pH 7.4) maintained at 37±0.5°C with constant stirring at 50 rpm. At predetermined time intervals (1, 2, 3, 4, 5, 6, 7, and 8 hr), 1 mL of sample was withdrawn and replaced with fresh, pre-warmed simulated tear fluid to maintain sink conditions. The samples were analyzed spectrophotometrically at 272 nm, and the cumulative percentage drug release was calculated and compared with the release profile of the optimized *in situ* gel.

Determination of Drug Content

1 mL of the formulation was diluted with artificial tear fluid (pH 7.4), followed by further dilution to achieve a measurable concentration.[42] The final solution was analyzed using a UV spectrophotometer at 272 nm (λ_{max}), and drug content was determined by comparing absorbance with a standard calibration curve.

In vivo Ocular Irritation Study

The Draize test on New Zealand White rabbits involved instilling 50 µL of the *in situ* gel into the right eye, with saline in the left as control. Ocular parameters were observed at 1, 24, 48, and 72 hr No signs of irritation or abnormalities were noted, indicating the formulation was non-irritating.[43]

HET-CAM Test (Hen Egg Test Chorioallantoic Membrane)

Healthy chicken eggs (<7 days old, 50-60 g) were incubated at 35°C and 58% RH, with regular turning. On day 9, viable eggs had their air cell drained for CAM exposure. NaCl (0.9%) and NaOH (0.1 N) served as negative and positive controls, respectively.[44] The test sample was applied to the CAM, and reactions like hemorrhage, lysis, and coagulation were observed for up to 300 sec. The Irritation Score (IS) was calculated using a standard formula and classified as non, slight, moderate, or severe irritant. This method offers a reliable, animal-free alternative for irritation testing

Table 3: Formulation Table of *in situ* Gel.

Ingredients (unit)	A1	A2	A3	A4	A5	A6	A7	A8	A9
Carvedilol (%w/v)	10	10	10	10	10	10	10	10	10
Pluronic F127 (g)	16	16	16	18	18	18	20	20	20
Pluronic F68 (g)	3	4	5	3	4	5	3	4	5
HPMC E15 LV (g)	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
PBS (q.s. to 100 mL)	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.
Benzalkonium Chloride (0.02%)	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02

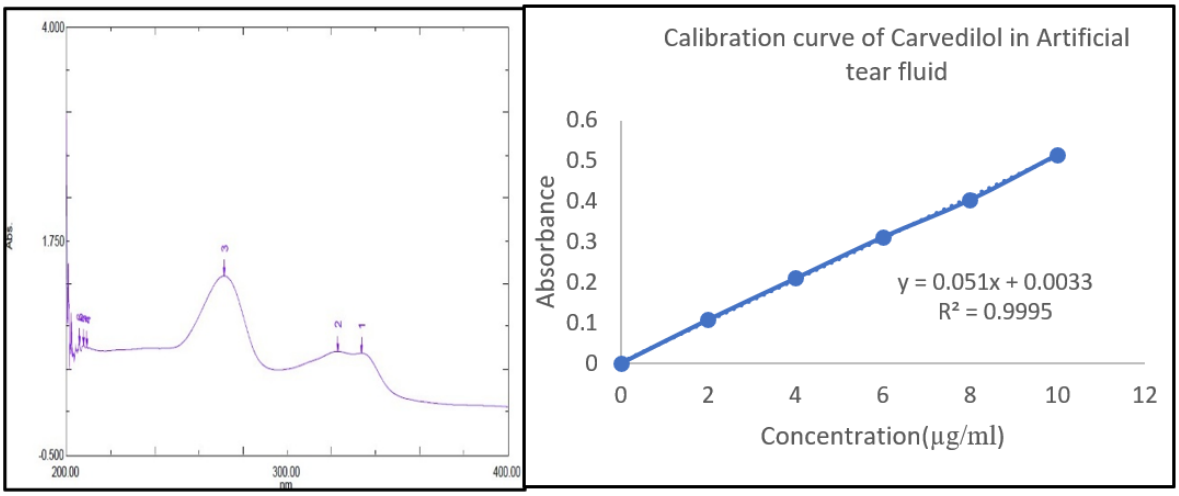


Figure 1: a) Carvedilol λ_{max} in Artificial Tear solution (pH 7.4). b) Calibration curve of Carvedilol.

Sterility Testing

Sterility was evaluated using the direct inoculation method as per Indian Pharmacopoeia. A 2 mL sample was added to 20 mL each of Fluid Thioglycolate Medium (FTM) and Soybean-Casein Digest Medium (SCDM). FTM was incubated at 30-35°C for bacterial growth, and SCDM at 20-25°C for fungi and aerobic microbes. After 14 days of incubation, no turbidity or growth was observed, confirming the formulation's sterility and suitability for ophthalmic use.[45]

Accelerated Stability Study

As per ICH Q1A (R2) guidelines, the optimized *in situ* gel was stored at 40±2°C and 75±5% RH for one month. Samples were evaluated on days 0, 15, and 30 for physical appearance, clarity, pH, gelling ability, and *in vitro* drug release. No significant changes were observed, confirming the formulation's stability.[46]

RESULTS

Preformulation Study

The organoleptic evaluation of carvedilol confirmed it as an off-white, odourless crystalline powder, in line with British Pharmacopoeia standards. Its melting point was 115.60°C, within the standard range of 114-116°C, indicating good purity. Carvedilol has poor water solubility and limited solubility in ethanol but is more soluble in DMF and DMSO. To enhance its solubility for ocular use without irritation, co-solvents like propylene glycol, hydroxypropyl-β-cyclodextrin, and PEG 400 were selected for their safety and compatibility.

Spectroscopic Analysis

The λ_{max} of carvedilol in artificial tear fluid was found to be 272.10 nm, closely matching the standard value of 272 nm, confirming drug identity as shown in Figure 1. The calibration curve followed Beer-Lambert's law in the 2-10 µg/mL range, showing linearity with an R^2 value of 0.9995.

X-ray Diffraction (XRD) Study

The X-ray Diffraction (XRD) distinct peaks at various 2θ angles, confirming the crystalline nature of pure carvedilol shown in Figure 2A. The presence of sharp and intense peaks, particularly at 10.1°, 16.3°, and 20.2°, indicates high crystallinity and suggests that the drug sample is of high purity.

FTIR of Carvedilol

The FTIR spectrum of carvedilol was recorded and analyzed by comparing observed peaks with typical functional group frequencies. Key absorptions included broad O-H stretch around 3427 cm^{-1}, aromatic and aliphatic C-H *stretch* near 2925 cm^{-1} and 2858 cm^{-1}, amide-related N-H bending or C=O *stretching* at 1696 cm^{-1}, and aromatic C=C *stretch* between 1545 and 1610 cm^{-1}, confirming the drug's characteristic functional groups as shown in Figure 2B.

Drug Excipient Compatibility Study

The drug-excipient compatibility study compares the FTIR spectra of carvedilol and the polymers used in the formulation. The presence of key peaks from the drug in the combined spectra indicates no interaction or incompatibility between carvedilol and the excipients. Observed peaks include O-H/N-H *stretch* at 3377 and 3531 cm^{-1}, alkyne *stretch* around 2346 cm^{-1}, aliphatic C-H at 2887 cm^{-1}, aromatic C=C *stretch* near 1623 and 1703 cm^{-1}, imidazole/amine C-N *stretch* at 1343 cm^{-1}, and ether C-O-C *stretch* between 1146 and 1278 cm^{-1}, confirming the presence of characteristic functional groups from both drug and excipients without any chemical incompatibility as shown in Figure 2C.

Thermogravimetric Analysis (TGA) and Differential Scanning Calorimetry (DSC) Analysis

The thermal profile of Carvedilol was evaluated using DSC and TGA. A minor endothermic peak at 115.60°C likely indicates the loss of surface-bound moisture or volatiles, with a possible glass transition. A sharp melting peak at 311.61°C (enthalpy

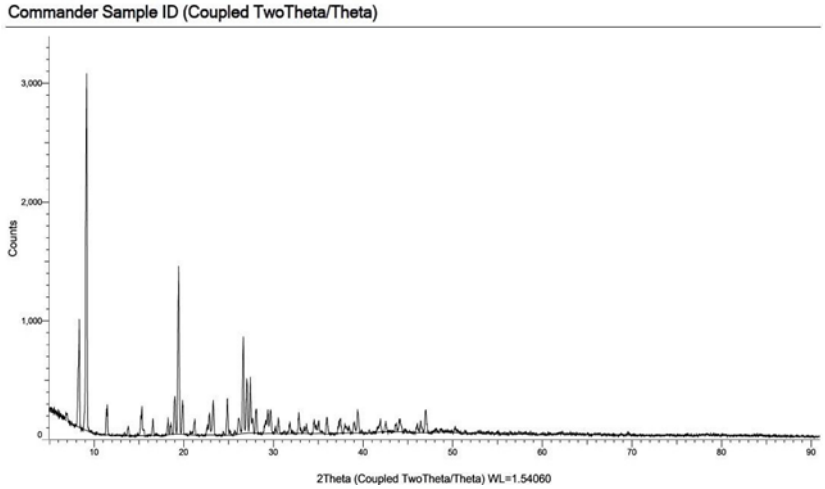


Figure 2A: X-ray Diffraction (XRD) Study of Carvedilol.

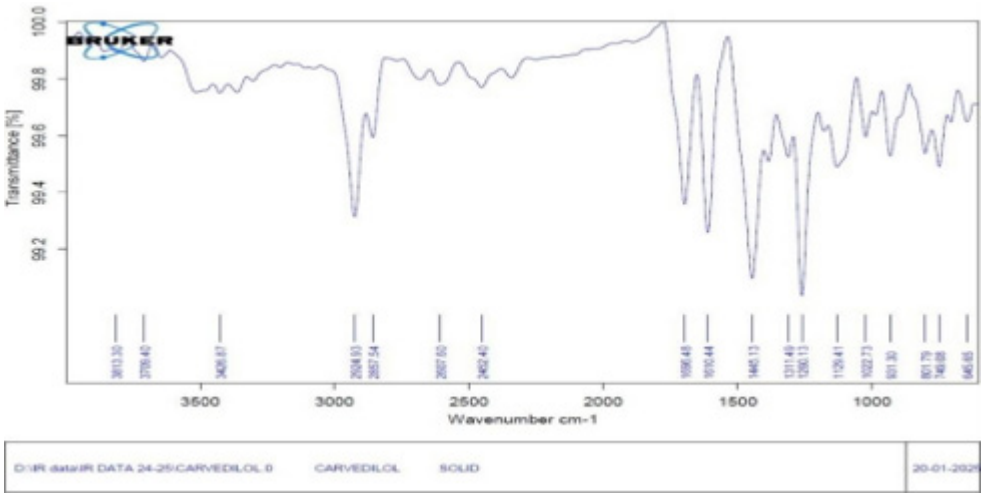


Figure 2B: FTIR of Carvedilol.

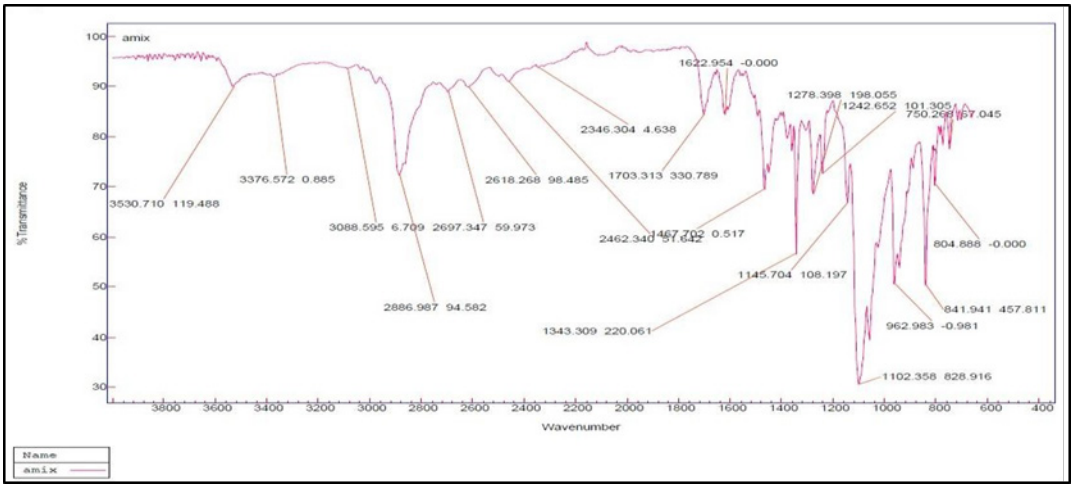


Figure 2C: Drug Excipient Compatibility Study.

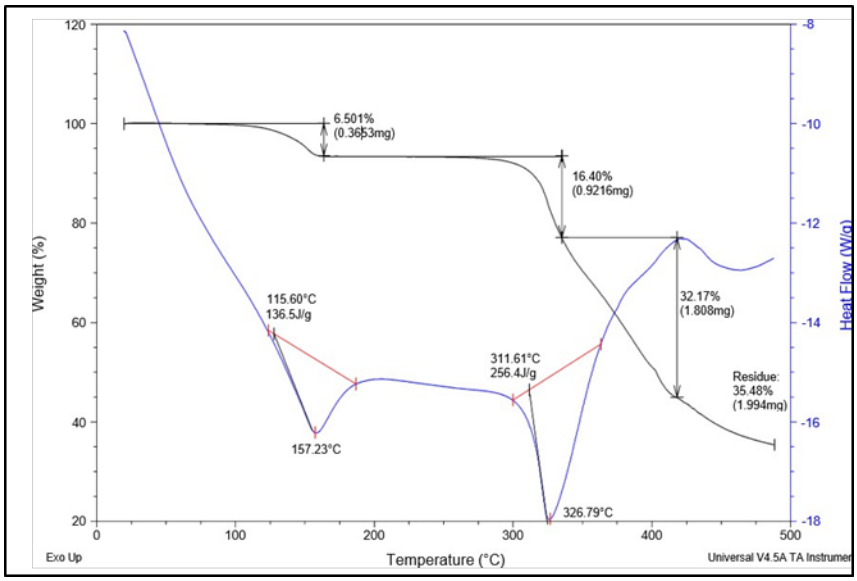


Figure 2D: Thermogravimetric Analysis (TGA) and Differential Scanning Calorimetry (DSC) Analysis of Carvedilol.

256.4 J/g) confirms the crystalline and pure nature of the drug. Decomposition appears to begin around 326.79°C.TGA revealed three major weight loss steps:

- 6.50% below 150°C (moisture/volatile loss).
- 16.40% between 150°C and 330°C (organic degradation).
- 32.17% beyond 330°C (complete breakdown).

A residue of 35.48% remained, likely from inorganic or carbonaceous content. No major degradation occurred below 250°C, indicating good thermal stability for formulation purposes as shown in Figure 2D.

QbD-Driven Strategy for Formulation and Process Optimization

Quality Target Product Profile

Based on the Intended use the QTTP Elements were defined. The Quality Target Product Profile (QTPP) is a critical element in the development of an ophthalmic *in situ* gel formulation, ensuring the product meets therapeutic objectives and patient acceptability. The selected dosage form, an ophthalmic *in situ* gel, is designed to prolong precorneal residence time and enhance ocular bioavailability. Administered through the ocular route, it facilitates site-specific drug delivery to the anterior chamber of the eye. Aesthetic clarity and a near-neutral pH (~7.0) are essential to ensure patient comfort and prevent ocular irritation by mimicking natural tear fluid. Optimal gelling time and a suitable gelling temperature are vital for ease of application and consistent usability. The viscosity of the formulation is optimized to allow for smooth instillation while ensuring efficient gelation. Accurate and reproducible drug content guarantees therapeutic efficacy, while a sustained drug release profile aids in prolonging the therapeutic effect. Furthermore, non-irritant behavior in both *in vitro* and *in vivo* irritation studies confirms ocular compatibility and safety. Lastly, the formulation's stability over its shelf life assures maintained product quality and effectiveness.

- Critical Quality Attributes (CQA) For Carvedilol Loaded *in situ* Gel.
- Critical Quality Attributes (CQAs) for the optimized *in situ* gel formulation were chosen based on their impact on product performance, stability, and antiglaucoma efficacy.
- Critical Material Attributes (CMAs) and Process Parameters (CPPs).

Optimization of *in situ* gel was done by considering Critical Material Attributes (CMAs) such as Concentration of Pluronic F127 and Pluronic F68 as independent variables and Critical Quality Attributes (CQAs) such as Gelling time and Gelling temperature.

Experimental Design

A 3^2 full factorial design was used to study the effect of two key variables Pluronic F127 (X1) and Pluronic F68 (X2) on the behavior of the Carvedilol-based *in situ* gel. As dependent Y1 Gelling time Y2 gelling temperature. Pluronic F127 acted as the main thermosensitive polymer, while Pluronic F68 served as a co-polymer. Their concentrations were tested at 16%, 18%, and 20% w/v for F127 and 3%, 4%, and 5% w/v for F68. HPMC E15LV was included at a fixed 0.5% w/v in all batches to improve viscosity and mucoadhesion.

Formulation Responses

Nine batches (A1-A9) were developed using varying concentrations of Pluronic F127 (16-20% w/v) and Pluronic F68 (3-5% w/v). and constant Carvedilol concentration. The observed gelling temperature ranged from 35.6°C to 45.9°C, and the gelling time varied between 22 and 45 sec. This experimental design aided in identifying suitable polymer combinations for rapid gelation and effective performance at ocular temperatures.

Table 4: Evaluation of pH, gelling capacity, gelling temperature, and viscosity of different gel batches.

Batches	pH	Gelling Capacity	Gelling Temperature	Viscosity
A1	6.57± 0.24	-	42.3	123±3 cps
A2	7.23±0.16	+	45.9	356±6 cps
A3	6.38±0.69	++	38.6	459±9 cps
A4	6.59±0.32	+	40.6	473±3 cps
A5	6.45±0.16	+	38.9	667±7 cps
A6	6.64±0.35	-	38.5	1563±13 cps
A7	7.16 ± 0.04	+++	36.7	1132±12 cps
A8	6.22±0.47	+	38.2	1965±15 cps
A9	6.09±0.60	+	35.6	2456±16 cps

Gelling capacity is denoted as follows: "-" Delayed gelation; unstable, "+" Immediate gelation; slightly less stable or moderate stability, "++" Gelation after a few seconds; stable, "+++" Immediate gelation; strong and stable gel formation.

Gelling Temperature

The ANOVA results demonstrated that model was statistically significant (<0.05), confirming validity and reliability of experimental design.

ANOVA for Quadratic model

The ANOVA results confirm the quadratic model is significant (F=25.59, p=0.0115). Pluronic F127 (A), Pluronic F68 (B), and A^2 significantly affect the response, while AB and B^2 are not significant. The low residual error (1.76) supports the model's good fit

Final Equation in Terms of Coded Factors

Gelling Temperature +37.66-2.45 *A+1.75*B-0.7750*AB+3.02A^2-0.2833 B^2

Pluronic F127 (A) lowers gelling temperature, while Pluronic F68 (B) raises it. Their interaction (AB) slightly reduces temperature. Non-linear effects from A^2 and B^2 suggest a balanced mix is crucial for optimal gelation. The predicted vs. actual plot for gelling temperature shows a close alignment of data points along the diagonal, indicating strong model accuracy with minimal error. The contour plot demonstrates the effect of Pluronic F127 and F68 on gelling temperature, where higher concentrations of A increase the temperature, and B shows a peak effect around 4% as shown in Figure 3A. The 3D surface plot further illustrates this interaction, with gelling temperature rising with A and a dip at mid-level B, confirming their combined influence on the formulation's thermal behaviour as shown in Figure 3B.

Gelling Time

The ANOVA results demonstrated that model was statistically significant (p<0.05), confirming validity and reliability of experimental design

ANOVA for linear model

The ANOVA results confirm a statistically significant linear model (F=5.32, p=0.0469). Pluronic F127 (A) has a significant effect on gelling time (p =0.0328), while Pluronic F68 (B) is not significant (p=0.1334). The model shows a fair fit, with some unexplained variability (residual SS=160.72 out of 445.56).

Final Equation in Terms of Coded Factors

Gelling time +30.22-5.83 *A+3.67*B

The equation shows that increasing Pluronic F127 (A) reduces gelling time, promoting faster gelation, while increasing Pluronic F68 (B) delays gelation by increasing gelling time.

The Predicted vs. Actual plot shows strong agreement between observed and predicted gelling times, indicating the model's accuracy. The contour plot illustrates that increasing Pluronic F127 (A) shortens gelling time, while higher Pluronic F68 (B) prolongs it. The shortest gelling times occur at high A and low B, and the longest at low A and high B as shown in Figure 3C. The 3D surface plot confirms this linear relationship, with gelling time decreasing as A increases and increasing as B rises, supported by experimental data points as shown in Figure 3D.

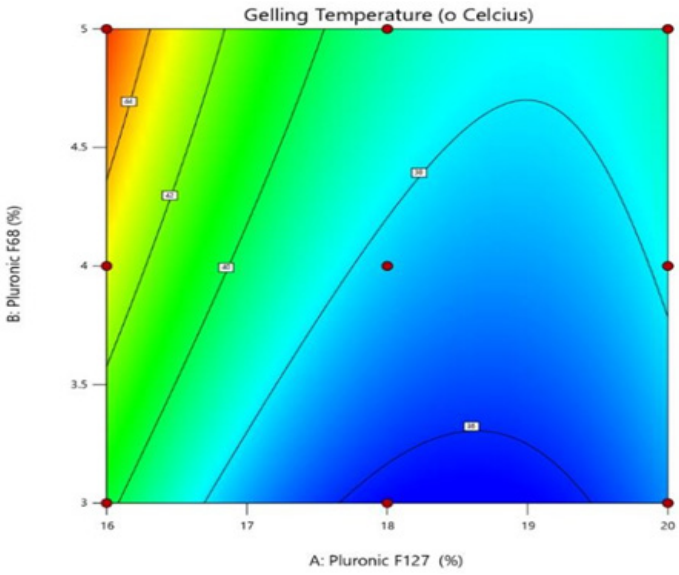


Figure 3A: Contour Plot for Gelling Temperature.

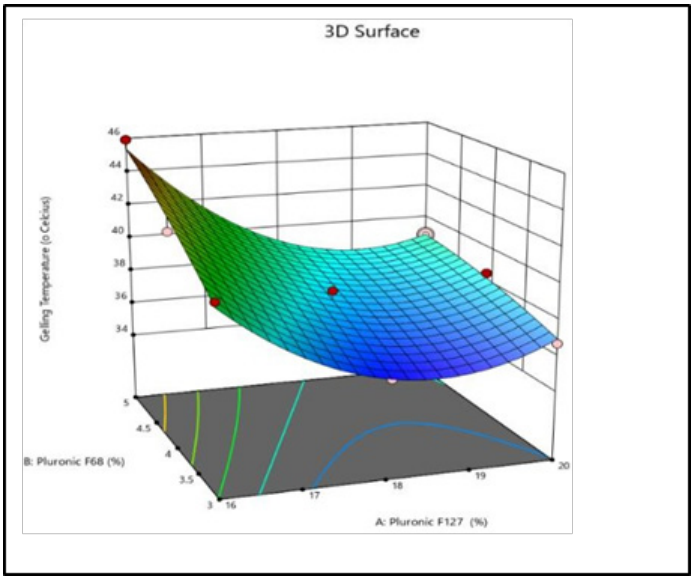


Figure 3B: 3D Surface Plot for Gelling Temperature.

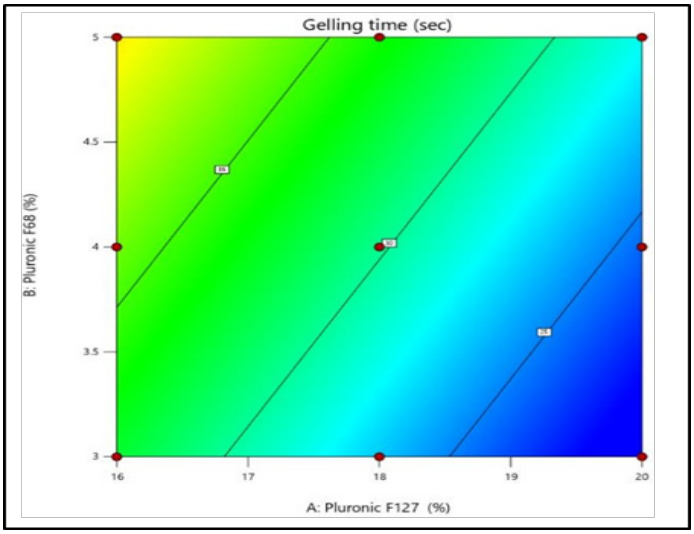


Figure 3C: Contour Plot for Gelling Time.

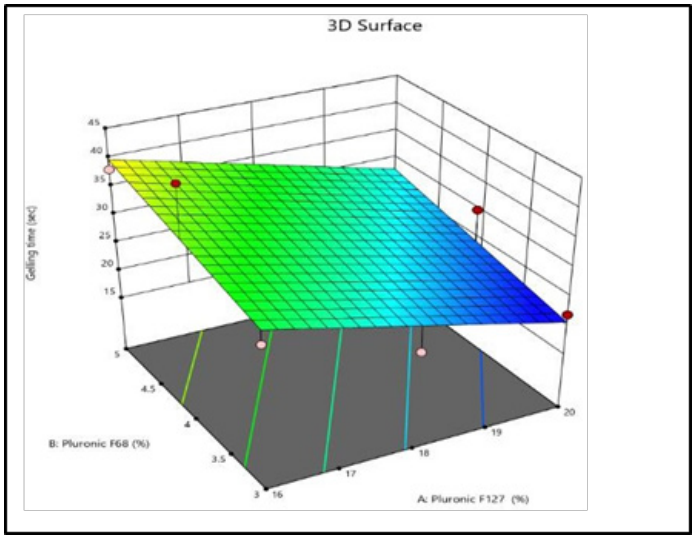


Figure 3D: 3D Surface Plot for Gelling Time.

Evaluation of Prepared *in situ* Gel

Organoleptic Properties

Visual Appearance and Clarity

Ophthalmic *in situ* gel formulations were visually examined under natural light against black and white backgrounds for clarity before and after gelation with simulated tear fluid (pH 7.4). Formulations showing clear or slightly translucent appearance without visible particles or phase separation were considered acceptable.

Determination of pH

pH of *in situ* gel solution was found to be around 6.0-7.39 as shown in Table 4 for all formulations. The formulation A7 has pH 7.16 which is an acceptable range for ophthalmic preparations.

Gelling Capacity Test

Gelling capacity of prepared formulation was determined by mixing 2 mL of the gel with 2 mL of artificial tear fluid at a physiological temperature of 37°C, and it was visually observed for gelling time. It was graded as follows: +; gel after few minutes dissolves rapidly, ++; immediate gelation remains for few mins, +++; immediate gelation remains for nearly an hour. The gelling capacity of all the 9 batches as given in Table 4 where Formulation A7 shows immediate gelation and for extended period shown in Figure 4.

Gelling Temperature Test

Gelling temperature was determined by gradually heating 2 mL of formulation from 20°C at 1°C/min in a water bath. The sol-to-gel transition point was identified by observing a visible increase in viscosity and gel formation. The gelling temperature of all the 9 batches were determined as given in Table 4 where, A7 batch showed Ideal, very close to ocular temperature.

Viscosity of Preformed Gel

The table shows viscosity values of various *in situ* gel batches. Batches A1 to A4 exhibited low to moderate viscosity, while A5 and A7 showed acceptable viscosity suitable for ocular use. Batches A6, A8, and A9 had high viscosity, which may hinder ease of installation as given in Table 4. Batch A7 was identified as optimal due to its balance between viscosity and performance.

In vitro Drug Release Study

Percentage drug release in case of *in situ* gel of Carvedilol was found to be 97.62% release in 8 hr of optimized batch A7. Thus, the *in vitro* diffusion test indicated the sustained release nature of *in situ* gel of Carvedilol.

Mathematical Model

The optimized Carvedilol *in situ* gel showed 97.62% drug release over 8 hr. Release data were fitted to various kinetic models, and the model with the highest correlation coefficient (R^2) was identified to determine the drug release mechanism The *in vitro* release data of the optimized Carvedilol *in situ* gel (Batch A7) best fit the Korsmeyer-Peppas model (R^2=0.9872), indicating diffusion-controlled release as shown in Figure 5. The Zero-order model also showed a good fit (R^2=0.9801), suggesting sustained release. First-order and Higuchi models had lower R^2 values, implying the release is governed by both diffusion and matrix erosion mechanisms The drug release profile of the optimized Carvedilol A7 *in situ* gel was best described by the Korsmeyer-Peppas model (R^2=0.9872), with the regression equation y=0.9837x + 1.1263 and a release exponent n=0.9837. According to the standard interpretation for cylindrical matrices (Fickian: n<0.45; anomalous: 0.45<n<0.89; Case II: n>0.89), this n value indicates Case II transport (zero-order-like release) governed predominantly by polymer relaxation/swelling rather than purely Fickian diffusion. Consistently, the zero-order

model also showed a high correlation (R^2=0.9801). Thus, the sustained release observed (97.62% over 8 hr) is attributable to polymer-relaxation-controlled kinetics with a constant-rate release profile (Figure 5).

Comparative *in vitro* Drug Release Study of Marketed Eye Drop

The *in vitro* drug release profile of the marketed Carvedilol eye drop showed a rapid release pattern, with 15.84% drug release at 1 hr, 31.38% at 2 hr, 34.05% at 3 hr, 50.49% at 4 hr, 62.97% at 5 hr, 84.95% at 6 hr, 93.26% at 7 hr, and 99.20% at 8 hr. In contrast, the optimized *in situ* gel formulation exhibited a comparatively sustained release pattern, with 13.06% at 1 hr, 28.91% at 2 hr, 38.41% at 3 hr, 47.02% at 4 hr, 62.97% at 5 hr, 89.00% at 6 hr, 93.16% at 7 hr, and 97.62% at 8 hr. as shown in Figure 6.

Determination of Drug Content

Drug content of the optimized batch (A7) was found to be 93.76%. This indicates that the amount of drug in the formulation is almost complete and within the acceptable range.

In vivo Ocular Irritation Study

An acute eye irritation study was conducted on New Zealand White rabbits following OECD 405 guidelines. A single dose of 0.1 mL of optimized *in situ* gel (Sample A7) was instilled into the left eye, with the right eye serving as the control as shown in Figure 7. Observations at 1, 24, 48, and 72 hr showed no signs of corneal opacity, iris damage, conjunctival redness, or chemosis as shown in Figure 7. All Draize scores remained zero throughout the test The test formulation was classified as non-irritant and safe for ocular use.

- No signs of irritation or corrosion observed at any time point.
- No ocular discharge, corneal opacity, chemosis, or redness detected.
- Animals remained clinically normal and active throughout the study.
- Visual documentation (below) supports the Draize scoring.
- Hen Egg Test Chorioallantoic Membrane (HET-CAM) Test.

The HET-CAM test was performed to evaluate ocular irritation of the optimized Carvedilol *in situ* gel. Compared to the marketed eye drop, saline (negative control), and irritants like SLS/NaOH (positive control), the *in situ* gel showed only slight haemorrhage with no lysis or coagulation. Irritation Scores (IS) were low for the *in situ* gel (0.32) and the marketed product (0.28), classifying them as non-irritants. The negative control showed no irritation (IS=0.0), while the positive control confirmed severe irritation (IS=18.92), validating the test. These results confirm the ocular safety of the formulation were shown in Figure 8.

Sterility Testing

The sterility test for the optimized batch A7 of Carvedilol was performed using Soya Casein Digest Medium and Fluid Thioglycolate Medium as per Indian Pharmacopoeia guidelines. After 14 days of incubation, no microbial growth or turbidity was observed, confirming the formulation's sterility and the effectiveness of the sterilization process. The positive control

Figure 4: Gelling Capacity of *in situ* Gel (A7 Batch).

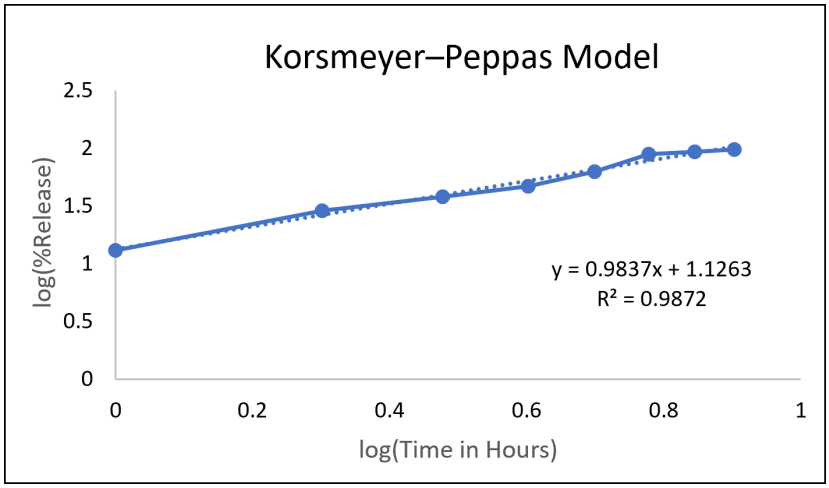


Figure 5: Korsmeyer-Peppas model fit for the optimized batch (A7): y=0.9837x + 1.1263; n=0.9837; R^2=0.9872. For cylindrical matrices, n>0.89 indicates Case II transport (zero-order-like release) consistent with polymer-relaxation-controlled kinetics.

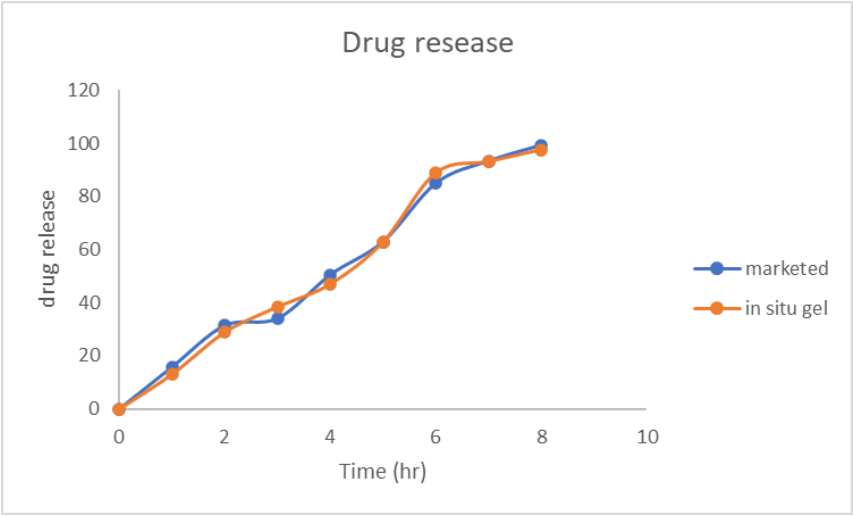


Figure 6: Comparative *in vitro* Drug Release Study of Marketed Eye Drop.

showed expected growth, validating the media's suitability. Thus, Batch A7 passed the sterility test successfully.

Accelerated Stability Study

Accelerated stability study was carried out at 40±2°C at 75±5% RH for 1 month using stability chamber. Samples were an analyzed periodically one very week, and found that there are no changes in visual appearance, clarity, pH and gelation.

DISCUSSION

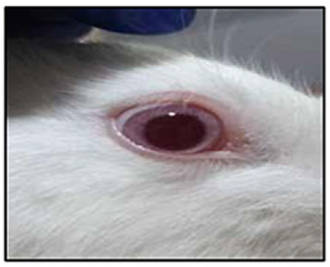
The present study was designed to develop a thermosensitive *in situ* gel containing Carvedilol for ocular administration, using a Quality by Design (QbD) approach to ensure enhanced formulation efficiency and reproducibility. The primary objective was to improve the ocular bioavailability of Carvedilol, a drug with established efficacy in glaucoma management, by addressing its inherent limitations such as poor retention, rapid drainage, and low corneal permeability in conventional eye drop formulations. The preformulation studies confirmed the physicochemical stability and compatibility of Carvedilol with selected excipients, an essential prerequisite for successful formulation. Spectroscopic and thermal analyses validated the purity and stability of the drug throughout the process. The application of Pluronic F127 and F68 in varying concentrations effectively leveraged thermosensitive gelation behaviour, transforming the formulation from a liquid to a gel at ocular surface temperature (~35-37°C), thus prolonging drug residence and minimizing precorneal loss. A 3^2 factorial design facilitated a systematic exploration of the effects of Pluronic concentrations on two critical quality attributes gelling temperature and gelling time. The optimized batch (A7) demonstrated excellent gelation characteristics, with an ideal gelling temperature of 36.7°C and rapid gel formation, indicating its suitability for ocular application. Viscosity measurements confirmed that the formulation maintained a flowable consistency at room temperature and formed a stable gel upon administration,

ensuring patient comfort and ease of instillation. *In vitro* drug release studies showed that the optimized formulation provided sustained release of Carvedilol (97.62% over 8 hr), aligning well with the requirements of long-acting ocular therapy. Comparative evaluation with the marketed eye drop further highlighted the advantage of the *in situ* gel system. The marketed product exhibited a faster release (99.20% at 8 h), with nearly half of the drug released within the first 4 hr (50.49%), whereas the *in situ* gel provided a slower and more controlled release (47.02% at 4 hr) while reaching 97.62% at 8 h. This sustained release pattern confirms the potential of the gel system to prolong ocular residence time, reduce dosing frequency, and maintain therapeutic drug levels for extended periods.

Kinetic modeling confirmed a predominant Case II transport mechanism for the optimized batch (A7), as the Korsmeyer-Peppas release exponent was found to be $n \approx 0.98$. For cylindrical matrices, values of $n>0.89$ correspond to Case II transport, indicating a zero-order-like release profile governed primarily by polymer chain relaxation and swelling processes rather than simple Fickian diffusion ($n<0.45$) or anomalous/non-Fickian transport ($0.45<n<0.89$). This observation is consistent with the strong correlation to the zero-order model ($R^2=0.9801$), supporting that the release from the *in situ* gel followed a relaxation-controlled mechanism with a nearly constant drug release rate. Such a mechanism is particularly desirable for ophthalmic delivery, as it ensures sustained drug levels at the ocular site while minimizing dosing frequency.

The safety profile of the optimized formulation was established through Draize and HET-CAM tests. No signs of irritation, redness, or vascular damage were observed, confirming the non-irritant nature of the gel. Additionally, sterility testing validated the aseptic integrity of the formulation, making it suitable for ophthalmic use. The accelerated stability study further indicated that the product retained its physical, chemical, and functional attributes over 30 days under stress conditions, reinforcing its formulation robustness.

Overall, the developed *in situ* gel system demonstrated strong potential as an improved ocular delivery platform for Carvedilol.

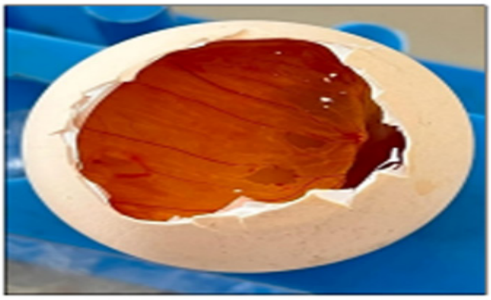


In vivo Ocular Irritation Study (Control Eye).

In vivo Ocular Irritation Study (Test Eyes)

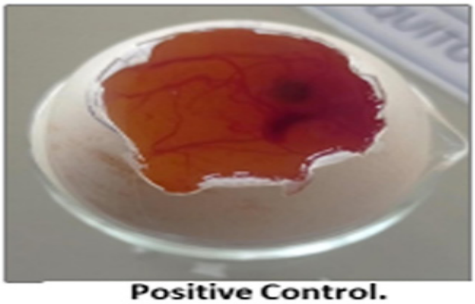
Figure 7: *In vivo* Eye Irritation Test.

Carvedilol *In situ* Gel Formulation.



Marketed formulation

Negative Control.



Positive Control.

Figure 8: HET CAM test.

By integrating QbD principles with thermoresponsive polymeric systems, the study successfully addressed the limitations of conventional ocular drug delivery, offering a more effective, patient-friendly alternative for the management of glaucoma.

CONCLUSION

The developed thermoresponsive *in situ* gel of Carvedilol shows strong potential for glaucoma management by overcoming the drawbacks of conventional eye drops. Guided by QbD principles, the optimized formulation achieved suitable gelling temperature, viscosity, pH, and drug content, with sustained drug release up to 8 hr. Safety studies confirmed its non-irritant and sterile nature, while stability testing demonstrated consistent performance. Overall, this novel *in situ* gel offers an effective, patient-friendly, and reliable platform for enhancing ocular delivery and therapeutic efficacy.

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ABBREVIATIONS

QbD: Quality by Design; **FTIR:** Fourier Transform Infrared Spectroscopy; **DSC:** Differential Scanning Calorimetry; **TGA:** Thermogravimetric Analysis; **XRD:** X-ray Diffraction; **UV:** Ultraviolet; **RPM:** Revolutions Per Minute; **ICH:** International Council for Harmonisation; **PBS:** Phosphate Buffered Saline; **SLS:** Sodium Lauryl Sulfate; **STF:** Simulated Tear Fluid; **HET-CAM:** Hen's Egg Test on Chorioallantoic Membrane; **OECD:** Organisation for Economic Co-operation and Development; **SCDM:** Soybean Casein Digest Medium; **FTM:** Fluid Thioglycollate Medium; **CQA:** Critical Quality Attributes; **QTPP:** Quality Target Product Profile; **CMAs:** Critical Material Attributes; **CPPs:** Critical Process Parameters; **cP:** Centipoise; **IS:** Irritation Score.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study was approved by the Institutional Animal Ethics Committee (IAEC) of Ashokrao Mane College of Pharmacy, Peth-Vadgaon, Maharashtra, India. All experimental protocols were conducted in accordance with CCSEA guidelines and institutional ethical standards.

HUMAN AND ANIMAL RIGHTS

The study involved animal experiments but no human subjects. All animal procedures strictly followed the CCSEA and institutional guidelines for animal care and use.

SUMMARY

This study developed a thermoresponsive *in situ* gel of Carvedilol for glaucoma therapy using Quality by Design (QbD) principles. A 3^2 factorial design optimized key parameters including gelling temperature, pH, viscosity, and drug release. The optimized formulation exhibited sustained release up to 8 hr (97.62%), with a more controlled profile compared to a marketed eye drop (99.20%). Safety was confirmed through Draize and HET-CAM tests, while sterility and stability were maintained under accelerated conditions. The developed *in situ* gel demonstrated improved ocular retention, reduced dosing frequency, and enhanced patient compliance, offering a promising alternative to conventional eye drops for effective glaucoma management.

REFERENCES

1. Lee SS, Mackey DA. Glaucoma-risk factors and current challenges in the diagnosis of a leading cause of visual impairment. Maturitas. 2022;163:15-22.
2. Guier C, Kaur K, Stokkermans T. Optic neuritis. StatPearls. 2025.
3. Hafez MA, Mahmoud MF, Ibrahim IA. Carvedilol: New pharmacological approaches. Zagazig Journal of Pharmaceutical Sciences. 2024: 1-21.
4. Chandel A, Kandav G. Insights into ocular therapeutics: a comprehensive review of anatomy, barriers, diseases and nanoscale formulations for targeted drug delivery. Journal of Drug Delivery Science and Technology. 2024: 105785.
5. Achouri D, Alhanout K, Piccerelle P, Andrieu V. Recent advances in ocular drug delivery. Drug development and industrial pharmacy. 2013;39(11):1599-617.
6. Jacob S, Nair AB, Shah J. Emerging role of nanosuspensions in drug delivery systems. Biomaterials research. 2020;24(1):3.
7. Pandey M, Choudhury H, binti Abd Aziz A, Bhattamisra SK, Gorain B, Su JS, Potential of stimuli-responsive *in situ* gel system for sustained ocular drug delivery: Recent progress and contemporary research. Polymers. 2021;13(8):1340.
8. Agrawal AK, Das M, Jain S. *In situ* gel systems as 'smart'carriers for sustained ocular drug delivery. Expert opinion on drug delivery. 2012;9(4):383-402.
9. Cassano R, Di Gioia ML, Trombino S. Gel-based materials for ophthalmic drug delivery. Gels. 2021;7(3):130.
10. Kassaee SN, Nili-Ahmadabadi A, Mahboobian MM. Fabrication of poloxamer based besifloxacin thermosensitive *in situ* gelling nanoemulsions for ophthalmic delivery. Journal of Bioactive and Compatible Polymers. 2023;38(4):298-310.
11. Yoshida T, Shakushiro K, Sako K. Ion-responsive drug delivery systems. Current Drug Targets. 2018;19(3):225-38.
12. Almutairy BK, Khafagy ES, Abu Lila AS. Development of Carvedilol Nanoformulation-Loaded Poloxamer-Based *in situ* Gel for the Management of Glaucoma. Gels. 2023;9(12):952.
13. Shatz W, Aaronson J, Yohe S, Kelley RF, Kalia YN. Strategies for modifying drug residence time and ocular bioavailability to decrease treatment frequency for back of the eye diseases. Expert opinion on drug delivery. 2019;16(1):43-57.
14. Al-Qaysi ZK, Beadham IG, Schwikkard SL, Bear JC, Al-Kinani AA, Alany RG. Sustained release ocular drug delivery systems for glaucoma therapy. Expert Opinion on Drug Delivery. 2023;20(7):905-19.
15. El-Boghdadly K, Pawa A, Chin KJ. Local anesthetic systemic toxicity: current perspectives. Local and regional anesthesia. 2018: 35-44.
16. Ma C, Cui W, Liu X, Ding Y, Wang Y.C. *In situ* preparation of gel polymer electrolyte for lithium batteries: Progress and perspectives. InfoMat. 2022;4(2):e12232.
17. Bhowmik M, Das S, Chattopadhyay D, Ghosh LK. Study of thermo-sensitive *in situ* gels for ocular delivery. Scientia Pharmaceutica. 2011;79(2):351.
18. Guan X, Li Y, Jiao Z, Chen J, Guo Z, Tian H, Chen X. A pH-sensitive charge-conversion system for doxorubicin delivery. Acta Biomaterialia. 2013;9(8):7672-8.
19. Li Q, Cao Y, Wang P. Recent advances in hydrogels for the diagnosis and treatment of dry eye disease. Gels. 2022;8(12):816.
20. Mylonaki I. Sustained release systems for the perivascular administration of atorvastatin to prevent vein graft failure. J. Control. Release. 2016;232:93-102.

21. Beg S, Hasnain MS, Rahman M, Swain S. Introduction to quality by design (QbD): fundamentals, principles, and applications. In Pharmaceutical quality by design 2019: 1-17. Academic Press.
22. Taticek R, Liu J. Definitions and scope of key elements of QbD. Quality by design for biopharmaceutical drug product development. 2015: 31-46.
23. Khan D, Kirby D, Bryson S, Shah M, Mohammed AR. Development of an age-appropriate mini orally disintegrating carvedilol tablet with paediatric biopharmaceutical considerations. Pharmaceutics. 2021;13(6):831.
24. Alves JM, Prado LD, Rocha HV. Evaluation and correlation of the physicochemical properties of carvedilol. Pharmaceutical development and technology. 2016;21(7):856-66.
25. Sharapova AV, Ol'khovich MV, Blokhina SV. Thermodynamic consideration of dissolution and distribution behavior of carvedilol in pharmaceutical significant media. The Journal of Chemical Thermodynamics. 2024;190:107207.
26. Ravikumar AA, Kulkarni PK, Osmani RA, Hani U, Ghazwani M, Fatease AA, Carvedilol precipitation inhibition by the incorporation of polymeric precipitation inhibitors using a stable amorphous solid dispersion approach: Formulation, characterization, and *in vitro In vivo* evaluation. Polymers. 2022;14(22):4977.
27. Abdelmonem R, Elhabal SF, Abdelmalak NS, El-Nabarawi MA, Teaima MH. Formulation and characterization of acetazolamide/carvedilol niosomal gel for glaucoma treatment: *in vitro*, and *in vivo* study. Pharmaceutics. 2021;13(2):221.
28. Mohamed JM, Alqahtani A, Menaa F, Kayarohanam S, Fatease AA, Alqahtani T. *In vitro* physical characterizations and docking studies on carvedilol nanocrystals. Crystals. 2022;12(7):988.
29. Borba PA, Vecchia DD, Riekes MK, Pereira RN, Tagliari MP, Silva MA, Pharmaceutical approaches involving carvedilol characterization, compatibility with different excipients and kinetic studies. Journal of thermal analysis and calorimetry. 2014;115:2507-15.
30. Alrashdan M, Shraideh ZA, Abulateefeh SR. Optimizing formulation parameters for the development of carvedilol injectable *in situ* forming depots. Pharmaceutical Development and Technology. 2023;28(9):865-76.
31. Kapoor DU, Kukkar RR, Gaur M, Prajapati BG, Malviya R. The Potential of Quality Target Product Profile in the Optimization of Nanoemulsions. Current Nanomedicine. 2025;15(1):4-17.
32. Sipos B, Rajab F, Katona G, Csóka I. Current insights into polymeric micelles for nasal drug delivery. Expert Opinion on Drug Delivery. 2025
33. Dos Santos Simões JR. Development of Semi-Solid formulations according to the Quality by Design approach.
34. Salem HF, Nafady MM, Eissa EM, Abdel-Sattar HH, Khallaf RA. Assembly of *in situ* gel containing nano-spanlastics of an angiotensin II inhibitor as a novel epitome for hypertension management: factorial design optimization, *In vitro* gauging, pharmacokinetics, and pharmacodynamics appraisal. AAPS PharmSciTech. 2024;25(5):115.
35. Deka MR, Ahmed AB, Chakraborty JA. Development, evaluation and characteristics of ophthalmic *in situ* gel system: a review. Int J Curr Pharm Res. 2019;11(4):47-53.
36. Otten R, Schrepp M, Thomaschewski J. Visual clarity as mediator between usability and aesthetics. In Procedings of Mensch und Computer 2020 2020; 11-15.
37. Kurniawansyah IS, Rusdiana T, Sopyan I, Desy Arya IF, Wahab HA, Nurzanah D. Comparative study of *in situ* gel formulation based on the physico-chemical aspect: Systematic review. Gels. 2023;9(8):645.
38. Nair AB, Shah J, Jacob S, Al-Dhubiab BE, Sreeharsha N, Morsy MA. Experimental design, formulation and *in vivo* evaluation of a novel topical *in situ* gel system to treat ocular infections. PloS one. 2021;16(3):e0248857.
39. Gupta H, Jain S, Mathur R, Mishra P, Mishra AK, Velpandian T. Sustained ocular drug delivery from a temperature and pH triggered novel *in situ* gel system. Drug delivery. 2007;14(8):507-15.
40. Dunn M, Barbano DM, Drake M. Viscosity changes and gel formation during storage of liquid micellar casein concentrates. Journal of Dairy Science. 2021;104(12):12263-73.
41. Swain GP, Patel S, Gandhi J, Shah P. Development of Moxifloxacin Hydrochloride loaded *in situ* gel for the treatment of periodontitis: *In vitro* drug release study and antibacterial activity. Journal of Oral Biology and Craniofacial Research. 2019;9(3):190-200.
42. Tayel SA, El-Nabarawi MA, Tadros MI, Abd-Elsalam WH. Promising ion-sensitive *in situ* ocular nanoemulsion gels of terbinafine hydrochloride: design, *in vitro* characterization and *In vivo* estimation of the ocular irritation and drug pharmacokinetics in the aqueous humor of rabbits. International journal of pharmaceutics. 2013;443(1-2):293-305.
43. Laddha UD, Kshirsagar SJ. Formulation of nanoparticles loaded *in situ* gel for treatment of dry eye disease: *In vitro*, *ex vivo* and *in vivo* evidences. Journal of Drug Delivery Science and Technology. 2021;61:102112.
44. Smail SS. *Ex vivo* Irritation Evaluation of a Novel Brimonidine Nanoemulsion Using the Hen's Egg Test on Chorioallantoic Membrane (HET-CAM). Cureus. 2024;16(8).
45. Raghav N, Parveen S, Kaur S, David SA, Kong H, Kenney JL. A rapid and simple sterility test method based on solid culture medium containing blood. Biologicals. 2024; 85:101722.
46. Galgatte UC, Kumbhar AB, Chaudhari PD. Development of *in situ* gel for nasal delivery: design, optimization, *in vitro* and *in vivo* evaluation. Drug delivery. 2014;21(1):62-73.

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