© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

Formulation, Optimization and Evaluation of an Ion-Activated Ocular In-Situ Gel of Timolol Maleate Using Natural Gelling Agents (Sodium Alginate and Gellan Gum) for Sustained Glaucoma Therapy

PRINCE PANDEY[1], VISHAL KUMAR SINGH[2]
[1]*Research Scholar (M. Pharm), (Department of Pharmaceutics, SHEAT College of Pharmacy, Varanasi)*
[2]*Assistant Professor, (Department of Pharmaceutics, SHEAT College of Pharmacy, Varanasi)*

Abstract- ***Conventional ophthalmic eye drops suffer from poor ocular bioavailability, typically below five percent, owing to rapid precorneal drainage, tear turnover and nasolacrimal clearance, which shorten the residence time of the drug and necessitate frequent dosing. The present study was undertaken to formulate, optimize and evaluate an ion-activated ocular in-situ gel of the antiglaucoma β-blocker timolol maleate using the naturally derived, ion-sensitive gelling polymers sodium alginate and gellan gum, with the aim of prolonging precorneal residence time, sustaining drug release and reducing dosing frequency. Nine formulations (F1–F9) were prepared by the cold (dispersion) method with the drug fixed at 0.5% w/v and varying polymer and calcium chloride concentrations. Preformulation FTIR and DSC studies confirmed the absence of any drug–polymer interaction. All formulations were transparent, of ocular-compatible pH (6.6–7.3), exhibited pseudoplastic flow and underwent immediate sol-to-gel transition on contact with simulated tear fluid, with viscosity rising sharply after gelation. Drug content was uniform (96.2–101.4%). In-vitro release across F1–F9 was sustained over 8–12 hours; the optimized batch F7 released 92% of drug in 12 hours without any burst effect, best fitting the Korsmeyer–Peppas model (R^2 = 0.989, n = 0.67) indicative of non-Fickian (anomalous) transport. Ex-vivo corneal permeation of F7 was gradual and sustained, reaching 82.16% at 8 hours, markedly outperforming a marketed solution. The optimized formulation remained physically and chemically stable over three months under ICH long-term and accelerated conditions. The developed natural-polymer ion-activated in-situ gel is a promising, patient-friendly alternative to conventional timolol eye drops for glaucoma management.***

Keywords: ***Ocular In-Situ Gel, Timolol Maleate, Sodium Alginate, Gellan Gum, Ion-Activated Gelation, Glaucoma, Sustained Release, Precorneal Residence Time.***

I. INTRODUCTION

The eye is among the most challenging organs for drug delivery, being a uniquely isolated and well-protected structure with a series of effective anatomical and physiological barriers. Topical instillation remains the most widely used and best-accepted route for treating anterior-segment disorders, yet the bioavailability achieved with conventional ophthalmic solutions is notoriously low, frequently less than five percent of the instilled dose. This is a direct consequence of the rapid pre-corneal loss processes that operate at the ocular surface: reflex lachrymation, constant tear turnover, rapid nasolacrimal drainage, non-productive absorption across the conjunctiva and the low permeability of the corneal epithelium all combine to clear the drug from the eye within minutes of instillation [1, 2].

These limitations reduce therapeutic efficacy and oblige the patient to instil the drug several times a day, which in turn undermines adherence—a particular problem in chronic, asymptomatic conditions such as glaucoma. Glaucoma is the leading cause of irreversible blindness worldwide and is often described as the “silent thief of sight” because it progresses without symptoms until substantial and irreversible visual-field loss has occurred. An estimated 76 million people were affected globally in 2020, a figure projected to rise to approximately 111.8 million by 2040 as populations age, with the burden falling disproportionately on Asia and Africa [3, 4]. Since lowering intraocular pressure (IOP) is the only proven means of arresting glaucomatous optic-nerve damage, and since this

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

requires lifelong, uninterrupted therapy, there is a compelling need for ocular delivery systems that maintain effective drug levels while minimising the dosing burden.

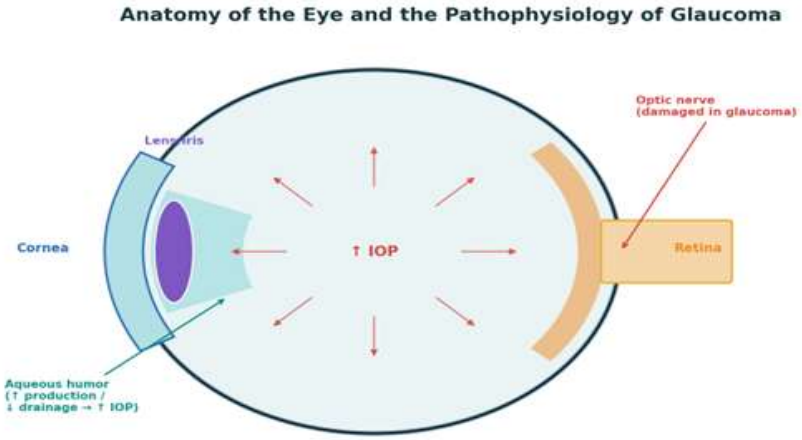

Figure 1. Anatomy of the eye and the pathophysiology of glaucoma. Impaired drainage or over-production of aqueous humor raises intraocular pressure (IOP), which progressively damages the optic nerve and causes irreversible vision loss.

In-situ gelling systems have emerged as an elegant solution to the shortcomings of conventional eye drops. These are low-viscosity liquid formulations that can be instilled as drops but undergo a rapid sol-to-gel transition in the physiological environment of the eye, in response to a change in temperature, pH or ionic concentration. Once gelled, the formulation adheres to the ocular surface, resists the pre-corneal clearance mechanisms, prolongs the contact time of the drug with the cornea and releases it in a sustained manner, thereby improving ocular bioavailability and reducing the frequency of administration [5, 6]. Among the several triggering mechanisms, ion-activated gelation is especially attractive for ophthalmic use because it exploits the divalent cations (chiefly Ca^{2+}) naturally present in tear fluid and does not depend on large temperature or pH changes.

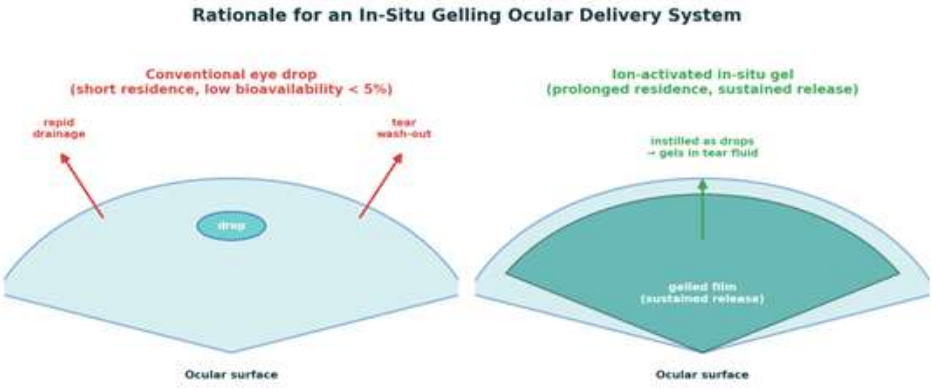

Figure 2. Rationale for an in-situ gelling ocular system. Conventional drops are rapidly lost from the ocular surface, whereas an ion-activated in-situ gel forms a retentive film that sustains drug release and prolongs residence time.

The choice of polymer is central to the performance of an in-situ gel. Natural, ion-sensitive polymers such as sodium alginate and gellan gum are particularly desirable because they are biocompatible,

biodegradable, non-toxic, non-immunogenic and economical, and because they gel efficiently in the presence of the cations of the tear film. Sodium alginate, a seaweed-derived anionic polysaccharide of mannuronic and guluronic acid residues, cross-links with calcium ions through its guluronate blocks, while gellan gum, a microbial tetrasaccharide-repeat polysaccharide, gels in the presence of mono- and divalent cations. Both are established pharmaceutical excipients well suited to ophthalmic in-situ gelling systems [7, 8].

1.1 Ocular Barriers and Routes of Drug Absorption

A rational appreciation of ocular drug delivery begins with the anatomy of the eye and its formidable barriers. The eye is organised into three layers: an outer fibrous coat of sclera and cornea, a middle vascular layer of choroid, ciliary body and iris, and an inner neural retina. For topically applied drugs the cornea is the principal route to the anterior chamber, but its outermost layer, the corneal epithelium, is the chief rate-limiting barrier because its tightly packed cells and tight junctions strongly restrict permeation. Hydrophilic drugs are limited mainly by the lipophilic epithelium, whereas lipophilic drugs are limited by the hydrophilic stroma, so that a balanced partitioning behaviour favours corneal transport. A drug at the ocular surface can be absorbed transcellularly or paracellularly across the cornea, or across the more permeable but highly vascular conjunctiva, where much of the dose is lost to the systemic circulation.

Superimposed on these permeability barriers are the dynamic pre-corneal factors that so drastically shorten residence time. The pre-corneal tear volume is only about 7–10 μL, and a single blink expels most of any instilled excess; tear turnover, reflex lachrymation and rapid nasolacrimal drainage then clear the remaining drug within a few minutes. The tear film itself is slightly alkaline (pH ≈ 7.4), of low viscosity (≈ 3 mPa·s) and mildly hypertonic (osmolality ≈ 310–350 mOsm/kg). It is precisely these clearance mechanisms that reduce the bioavailability of conventional drops to a few percent and that an in-situ gelling system is designed to counteract by increasing viscosity and adhering to the ocular surface immediately after instillation.

1.2 Approaches to In-Situ Gelling Systems

In-situ gelling systems are classified by the physiological stimulus that triggers the sol-to-gel transition. Temperature-triggered systems, typically based on poloxamers, remain liquid at room temperature and gel at the ocular surface temperature of about 35 °C; they are simple but can be affected by dilution and by ambient temperature. pH-triggered systems employ weakly acidic or basic polymers such as cellulose acetate phthalate or Carbopol that swell and gel as the pH shifts toward the slightly alkaline tear film; they depend on a sufficient pH change and can require relatively high polymer concentrations. Ion-triggered (osmotically activated) systems, exemplified by gellan gum and alginate, gel in response to the mono- and divalent cations of the tear fluid, chiefly Ca^{2+}. The ion-activated approach adopted in this study is especially suitable for ophthalmic use because it does not rely on a large temperature or pH change, gels rapidly and reproducibly under physiological conditions, and can be achieved with low, well-tolerated concentrations of natural polymers, minimising the risk of irritation or blurred vision. Compared with ointments and inserts, in-situ gels also offer less visual disturbance, more accurate and reproducible dosing, and greater patient comfort and compliance [5, 6].

The model drug selected for this study, timolol maleate, is a non-selective β_1/β_2-adrenergic receptor antagonist that lowers intraocular pressure by reducing the secretion of aqueous humor by the ciliary body, and it has been a first-line topical agent for open-angle glaucoma and ocular hypertension for decades. Conventionally administered as a 0.25% or 0.5% eye drop once or twice daily, it is limited by the same rapid pre-corneal loss that afflicts all ophthalmic solutions, together with the risk of systemic β-blockade following nasolacrimal absorption [9, 10]. Incorporating timolol maleate into a natural-polymer, ion-activated in-situ gel therefore offers a rational strategy to prolong its ocular residence, sustain its release and reduce both dosing frequency and systemic exposure. The present work describes the formulation, optimization and comprehensive in-vitro and ex-vivo evaluation of such a system built on sodium alginate and gellan gum.

The rationale for combining two natural polymers rather than relying on a single gelling agent deserves emphasis. Sodium alginate provides robust calcium-induced gelation through its guluronate blocks and contributes mucoadhesion via its carboxyl groups, while gellan gum forms clear, strong gels at very low concentration and tolerates the ionic environment of the tear film. Used together, they allow the total polymer load—and hence any risk of irritation, blurred vision or foreign-body sensation—to be kept low while still achieving rapid, strong gelation and a sustained release profile. Natural polymers additionally carry advantages of biodegradability, ready availability, low cost and an established regulatory acceptance in ophthalmic products, which is important for eventual translation. The study was therefore designed not merely to demonstrate feasibility but to identify, through a systematic variation of polymer type and concentration and cross-linker level across nine batches, the specific composition that best balances gel strength, sustained release, corneal permeation and stability.

II. MATERIALS AND METHODS

2.1 Materials

Timolol maleate (ophthalmic grade) was received as a gift sample. Sodium alginate (medium-viscosity grade) and gellan gum (low-acyl grade) were procured from HiMedia, Mumbai, and used as the ion-sensitive gelling polymers. Calcium chloride (cross-linker), benzalkonium chloride (preservative), sodium chloride (tonicity modifier) and sodium hydroxide / hydrochloric acid (pH modifiers) were of analytical reagent grade. Timolol maleate (molecular formula $C_{17}H_{28}N_4O_7S$; molecular weight 432.49 g/mol) is a white crystalline powder that is freely soluble in water, in keeping with its suitability for an aqueous ophthalmic system. Its key physicochemical and pharmacological attributes are summarised in Table 1.

Table 1. Physicochemical and pharmacological profile of timolol maleate.

Parameter	Value / description
Molecular formula	$C_{17}H_{28}N_4O_7S$
Molecular weight	432.49 g/mol
Category	Non-selective β_1/β_2-adrenergic blocker
Mechanism	↓ aqueous humor secretion → ↓ intraocular pressure
Solubility	Freely soluble in water; soluble in methanol/ethanol
λ_max	294 nm (phosphate buffer pH 7.4)
Ophthalmic dose	0.25–0.5% w/v
Indication	Open-angle glaucoma; ocular hypertension

2.2 Preformulation Studies

Drug–excipient compatibility was assessed by Fourier-transform infrared (FTIR) spectroscopy and differential scanning calorimetry (DSC). For FTIR, samples of pure drug and 1:1 physical mixtures of drug with each polymer were analysed as KBr pellets over 4000–400 cm^{-1}, and the spectra were compared for any shift, disappearance or emergence of peaks. For DSC, accurately weighed samples of drug, polymer and physical mixture were heated from 30 to 300 °C at 10 °C/min under a nitrogen atmosphere in aluminium pans, and the thermograms were examined for changes in the characteristic melting endotherm of the drug.

Solubility was determined by equilibrating an excess of timolol maleate in distilled water, phosphate buffer (pH 6.8 and 7.4) and simulated tear fluid (STF, pH 7.4) in capped vials shaken for 24 hours at 25 ± 1 °C, followed by filtration and spectrophotometric assay. For construction of a calibration curve, a 100 µg/mL stock solution in phosphate buffer (pH 7.4) was scanned over 200–400 nm to identify the wavelength of maximum absorbance (λ_max), and dilutions of 2–10 µg/mL were assayed by UV–visible spectrophotometry at that wavelength.

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

2.3 Formulation of the Ocular In-Situ Gel

The in-situ gels were prepared by the cold (dispersion) method. The required quantity of sodium alginate or gellan gum was slowly dispersed in distilled water under continuous magnetic stirring and hydrated overnight in a cool environment to obtain a fully swollen, lump-free dispersion. Timolol maleate was separately dissolved in a small volume of distilled water, and calcium chloride was added in a controlled manner to serve as the cross-linker without provoking premature gelation. The drug solution was then blended into the hydrated polymer dispersion under stirring, benzalkonium chloride (0.01% w/v) was added as preservative, sodium chloride was incorporated to achieve isotonicity, and the pH was adjusted to 6.8–7.4 with 0.1 N NaOH or 0.1 N HCl. The volume was made up with distilled water, and the preparation was sterilised by filtration through a 0.22-µm membrane and filled aseptically into ophthalmic vials [11, 12]. Nine batches (F1–F9) were formulated with the drug fixed at 0.5% w/v while varying the type and concentration of gelling polymer and the cross-linker level, as shown in Table 2.

Table 2. Composition of the ocular in-situ gel formulation batches (F1–F9).

Code	Timolol (%)	Na alginate (%)	Gellan gum (%)	$CaCl_2$ (%)	BKC (%)	NaCl (%)
F1	0.5	0.5	–	0.1	0.01	0.9
F2	0.5	1.0	–	0.1	0.01	0.9
F3	0.5	1.5	–	0.1	0.01	0.9
F4	0.5	–	0.25	0.05	0.01	0.9
F5	0.5	–	0.5	0.05	0.01	0.9
F6	0.5	–	0.75	0.05	0.01	0.9
F7	0.5	0.5	0.25	0.075	0.01	0.9
F8	0.5	1.0	0.5	0.075	0.01	0.9
F9	0.5	1.5	0.75	0.075	0.01	0.9

The gelation of the system is ion-activated: on instillation, the divalent calcium ions of the tear fluid cross-link the carboxylate groups of the guluronate residues of sodium alginate and the acyl-linked chains of gellan gum, transforming the low-viscosity solution into a viscoelastic three-dimensional gel network (Figure 3). Because the mechanism relies on the ions of the tear film rather than on a large pH or temperature change, gelation occurs rapidly and reproducibly on the ocular surface.

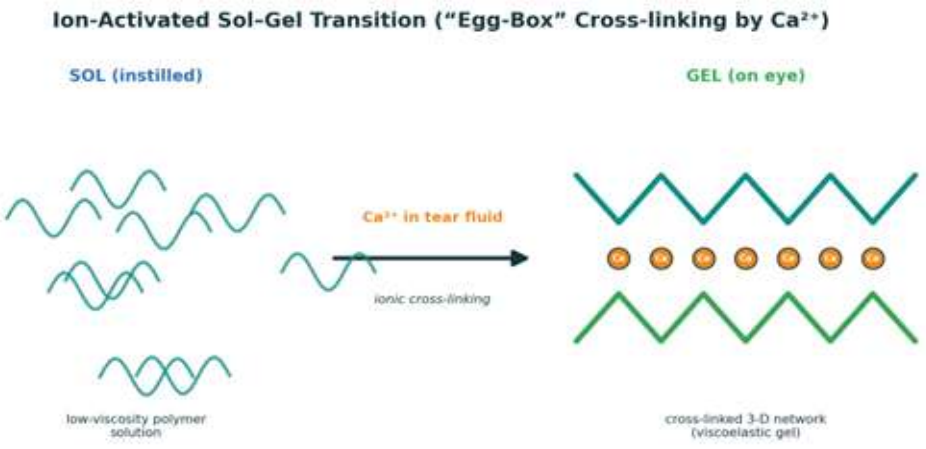

Figure 3. Mechanism of ion-activated sol–gel transition. Calcium ions in tear fluid cross-link the guluronate blocks of sodium alginate and gellan gum through the "egg-box" arrangement, converting the instilled solution into a retentive viscoelastic gel.

2.4 Evaluation of the In-Situ Gels

All formulations were evaluated in triplicate (n = 3). Clarity and appearance were assessed by visual inspection against a black-and-white background. The pH of each formulation was measured with a calibrated digital pH meter at 25 ± 2 °C. Viscosity was determined with a Brookfield viscometer (spindle 62) at 25 ± 1 °C both before gelation (sol state) and after gelation in simulated tear fluid, and the rheological behaviour was characterised. Gelling capacity was assessed by adding a drop of formulation to simulated tear fluid (pH 7.4) at 37 ± 1 °C and visually grading the speed and duration of gel formation. Drug content was determined by diluting a measured volume of formulation in STF (pH 7.4), filtering and assaying spectrophotometrically at 294 nm against the calibration curve, with acceptable content defined as 95–105% of the label claim.

In-vitro drug release was studied in a vertical Franz diffusion cell fitted with a cellophane membrane pre-soaked in simulated tear fluid, separating the donor from a receptor compartment filled with STF (pH 7.4) maintained at 37 ± 0.5 °C and stirred at 50 rpm. One-millilitre samples were withdrawn at 0.5, 1, 2, 4, 6, 8, 10 and 12 hours, replaced with fresh medium to maintain sink conditions, and assayed at 294 nm. The release data were fitted to zero-order, first-order, Higuchi and Korsmeyer–Peppas models, and the model with the highest regression coefficient (R^2) was taken as the best fit; the Korsmeyer–Peppas release exponent (n) was used to identify the release mechanism [13, 14].

Ex-vivo corneal permeation was evaluated using freshly excised goat/bovine cornea obtained from a local abattoir, cleaned in cold normal saline and mounted on a Franz diffusion cell with the epithelial side facing the donor compartment. The receptor compartment contained STF (pH 7.4) at 37 ± 2 °C under constant stirring; samples were withdrawn at intervals and assayed at 294 nm, and the cumulative amount permeated per unit area was plotted against time. The steady-state flux (J) was obtained from the slope of the linear portion, and the apparent permeability coefficient was calculated from the flux and the donor concentration [15]. The optimized formulation was subjected to stability studies as per ICH guidelines at 25 ± 2 °C / 60 ± 5% RH (long-term) and 40 ± 2 °C / 75 ± 5% RH (accelerated) for three months, with periodic evaluation of appearance, pH, viscosity, drug content and release. All data are expressed as mean ± SD; differences between formulations were assessed by one-way ANOVA, with $p < 0.05$ taken as significant.

III. RESULTS AND DISCUSSION

3.1 Preformulation and Compatibility Studies

The FTIR spectrum of pure timolol maleate displayed its characteristic absorption bands: a broad O–H stretch near 3350 cm^{-1}, an N–H stretch near 3200 cm^{-1}, a sharp C=O stretch of the maleate group at about 1715 cm^{-1}, aromatic C=C stretching between 1600 and 1650 cm^{-1}, and a C–O stretch at about 1100 cm^{-1}. In the overlay spectra of the drug with sodium alginate and gellan gum, all of these principal peaks were retained without any appreciable shift, disappearance or emergence of new bands, indicating the absence of chemical interaction and confirming the compatibility of timolol maleate with the selected natural gelling agents [16].

The DSC thermogram of pure timolol maleate showed a sharp endothermic melting peak at approximately 199–203 °C, consistent with its crystalline nature. In the physical mixtures the same endotherm was observed with only a slight reduction in intensity, attributable to the dilution effect of the polymer, and with no temperature shift or new thermal events. The retention of the melting endotherm confirms that neither a drug–polymer interaction nor a change in crystallinity occurred, and that the drug remains stable within the polymer matrix. The solubility studies confirmed the high aqueous solubility of timolol maleate in water, phosphate buffer and simulated tear fluid, supporting its suitability for an aqueous ion-activated gel in which the hydrophilic drug is retained and released in

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

a controlled fashion rather than being lost by immediate drainage.

3.2 Determination of λ_max and Calibration Curve

Scanning of the drug solution over 200–400 nm gave a wavelength of maximum absorbance of 294 nm, which was used for all subsequent quantification. The calibration curve constructed over 2–10 µg/mL was linear, obeying the regression equation y = 0.059x − 0.0021 with a regression coefficient (R^2) of 0.9999, confirming excellent adherence to the Beer–Lambert law across the working range (Figure 4) and validating the analytical method for the assay of drug content, release and permeation.

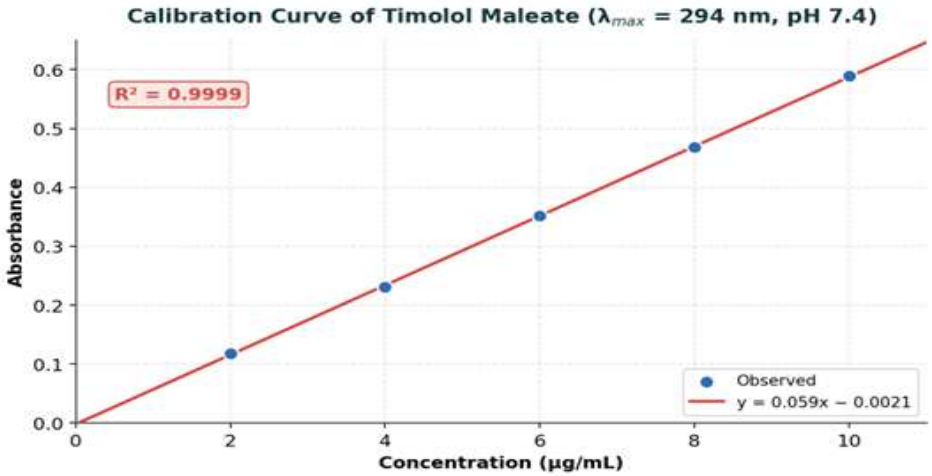

Figure 4. Calibration curve of timolol maleate at 294 nm in phosphate buffer (pH 7.4). The plot is linear over 2–10 µg/mL (y = 0.059x − 0.0021; R^2 = 0.9999).

3.3 Physicochemical Evaluation of the Formulations

All nine formulations (F1–F9) were clear and transparent, free of particulate matter and showed no turbidity, precipitation or phase separation on preparation or short-term storage, confirming proper solubilisation of the drug, absence of incompatibility and suitability for ophthalmic use. The pH of the formulations ranged from 6.60 ± 0.05 to 7.30 ± 0.04, lying comfortably within the physiological ocular range (6.5–7.5) and thus expected to cause minimal irritation and to be compatible with the lacrimal fluid; the differences between batches were not statistically significant ($p > 0.05$), indicating that polymer concentration did not markedly influence pH.

Rheological measurement showed that all formulations exhibited pseudoplastic (shear-thinning) flow, which is desirable for ophthalmic systems because it permits easy instillation under the shear of blinking while retaining viscosity at rest. The viscosity was low before gelation, ensuring pourability and accurate dropwise dosing, and rose sharply after gelation in simulated tear fluid, confirming the ion-triggered sol-to-gel transition; both values increased in proportion to polymer concentration (Figure 5). This behaviour is advantageous: low viscosity during instillation minimises discomfort, while the high post-gelation viscosity prolongs pre-corneal residence. Statistically significant differences ($p < 0.05$) between low- and high-polymer batches confirmed the concentration-dependent rheology. Gelling capacity mirrored this trend, with lower-polymer batches showing moderate gelation (++) and higher-polymer batches strong, prolonged gelation (+++). Drug content across all batches ranged from 96.2 ± 0.8% to 101.4 ± 0.6%, within pharmacopeial limits, with low standard deviations confirming uniform drug distribution and batch-to-batch reproducibility ($p > 0.05$). The complete physicochemical data are presented in Table 3.

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

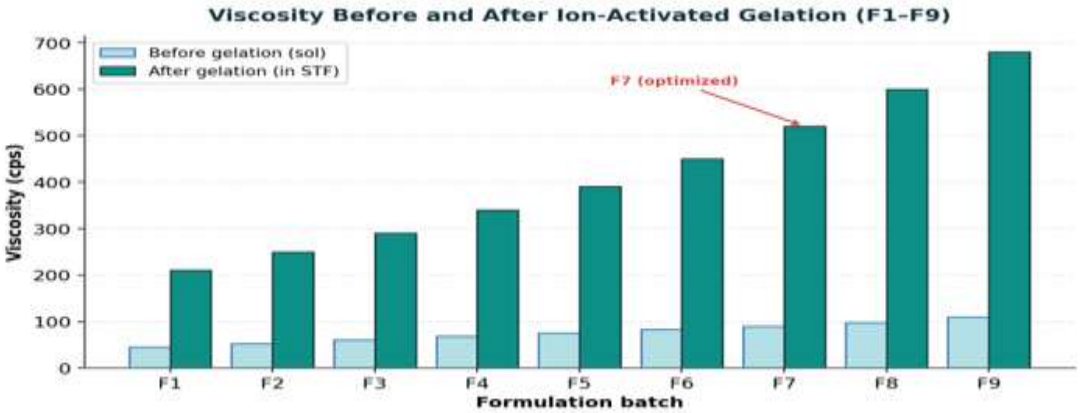

Figure 5. Viscosity of the formulations before and after ion-activated gelation. Viscosity rises sharply on gelation and increases with polymer concentration, confirming the concentration-dependent sol–gel transition.

Table 3. Physicochemical evaluation of the ocular in-situ gel formulations (F1–F9).

Batch	pH (±SD)	Visc. before (cps)	Visc. after (cps)	Gelling	Drug content (%)
F1	6.60 ± 0.05	45 ± 2.1	210 ± 5.4	++	96.2 ± 0.8
F2	6.70 ± 0.04	52 ± 1.8	250 ± 6.2	++	97.5 ± 0.6
F3	6.80 ± 0.06	60 ± 2.5	290 ± 7.1	++	98.1 ± 0.9
F4	6.90 ± 0.05	68 ± 2.3	340 ± 8.5	++	99.0 ± 0.7
F5	7.00 ± 0.04	75 ± 2.0	390 ± 9.4	+++	99.6 ± 0.5
F6	7.10 ± 0.03	82 ± 2.6	450 ± 10.2	+++	100.2 ± 0.6
F7	7.20 ± 0.04	90 ± 2.8	520 ± 11.3	+++	100.8 ± 0.4
F8	7.20 ± 0.05	98 ± 3.1	600 ± 12.5	+++	101.0 ± 0.5
F9	7.30 ± 0.04	110 ± 3.4	680 ± 14.2	+++	101.4 ± 0.6

3.4 In-Vitro Drug Release and Kinetics

In-vitro release from all batches was sustained over 8–12 hours, with no formulation exhibiting an undesirable initial burst (Figure 6). The rate of release was clearly governed by polymer concentration: the lower-polymer batches (F1–F3) released the drug relatively rapidly, reaching about 85–95% within 8–10 hours through their comparatively loose, easily hydrated matrices, whereas the higher-polymer batches (F8, F9) released most slowly owing to their dense, highly cross-linked networks that present a greater diffusional barrier. The optimized batch F7 achieved the most desirable profile, releasing 92% of the drug over a full 12 hours in a controlled manner without any burst, reflecting an ideal balance between gel strength and drug diffusivity. The complete release data are given in Table 4.

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

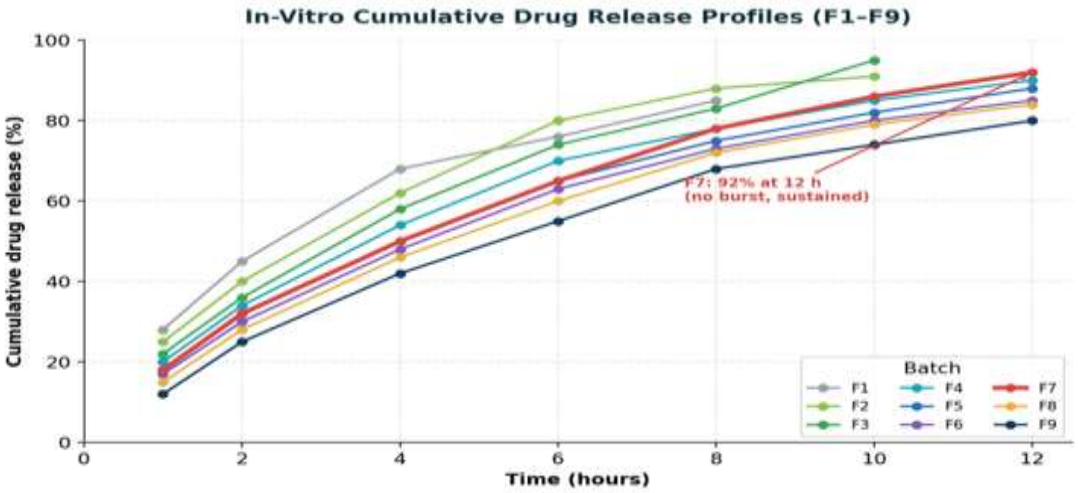

Figure 6. In-vitro cumulative drug-release profiles of formulations F1–F9. Release is sustained over 8–12 hours and slows with increasing polymer concentration; the optimized batch F7 releases 92% at 12 hours without burst.

Table 4. In-vitro cumulative percentage drug release of F1–F9.

Time (h)	F1	F2	F3	F4	F5	F6	F7	F8	F9
1	28	25	22	20	18	17	18	15	12
2	45	40	36	34	32	30	32	28	25
4	68	62	58	54	50	48	50	46	42
6	76	80	74	70	65	63	65	60	55
8	85	88	83	78	75	73	78	72	68
10	–	91	95	85	82	80	86	79	74
12	–	–	–	90	88	85	92	84	80

To elucidate the mechanism of release, the data for the optimized batch F7 were fitted to four kinetic models (Figure 7 and Table 5). The zero-order model gave a moderate fit (R^2 = 0.945) and the first-order model a weaker one (R^2 = 0.912), indicating that release was neither strictly constant nor purely concentration-dependent. The Higuchi model fitted well (R^2 = 0.978), pointing to a substantial diffusion-controlled component, but the highest correlation was obtained with the Korsmeyer–Peppas model (R^2 = 0.989), which was therefore taken as the best-fit model. Its release exponent, n = 0.67, lies between 0.45 and 0.89, denoting non-Fickian (anomalous) transport. The overall release mechanism is thus governed by a combination of drug diffusion through the hydrated matrix and relaxation/swelling of the cross-linked polymer chains, consistent with an ion-cross-linked network that both retards diffusion and swells on hydration in the tear fluid.

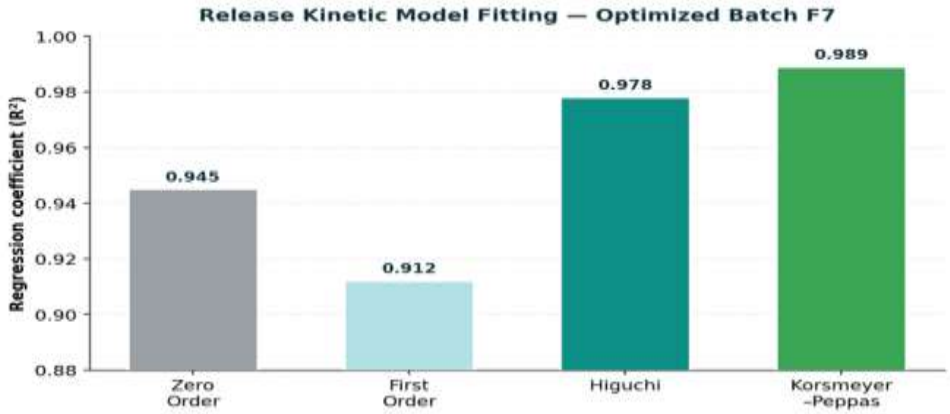

Figure 7. Kinetic model fitting for the optimized batch F7. The Korsmeyer–Peppas model gives the best fit (R^2 = 0.989); the release exponent n = 0.67 indicates non-Fickian (anomalous) transport.

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

Table 5. Release kinetic model fitting for the optimized batch F7.

Model	R^2 value	Interpretation
Zero order	0.945	Near-constant release
First order	0.912	Partially concentration-dependent
Higuchi	0.978	Diffusion-controlled release
Korsmeyer–Peppas	0.989	Best fit (n = 0.67, non-Fickian)

3.5 Ex-Vivo Corneal Permeation

The ex-vivo permeation of the optimized gel F7 through excised cornea was gradual and sustained throughout the eight-hour study, reaching a cumulative permeation of 82.16 ± 1.85% (41.08 µg/cm²) at 8 hours (Table 6). In contrast, a marketed timolol solution showed a rapid initial permeation with a steep early slope followed by an early plateau, reflecting rapid diffusion accompanied by pre-corneal elimination (Figure 8). The steady-state flux of F7, obtained from the linear portion of the permeation profile, was markedly higher and more sustained than that of the lower-polymer batches, and the difference in cumulative permeation was statistically significant ($p < 0.05$).

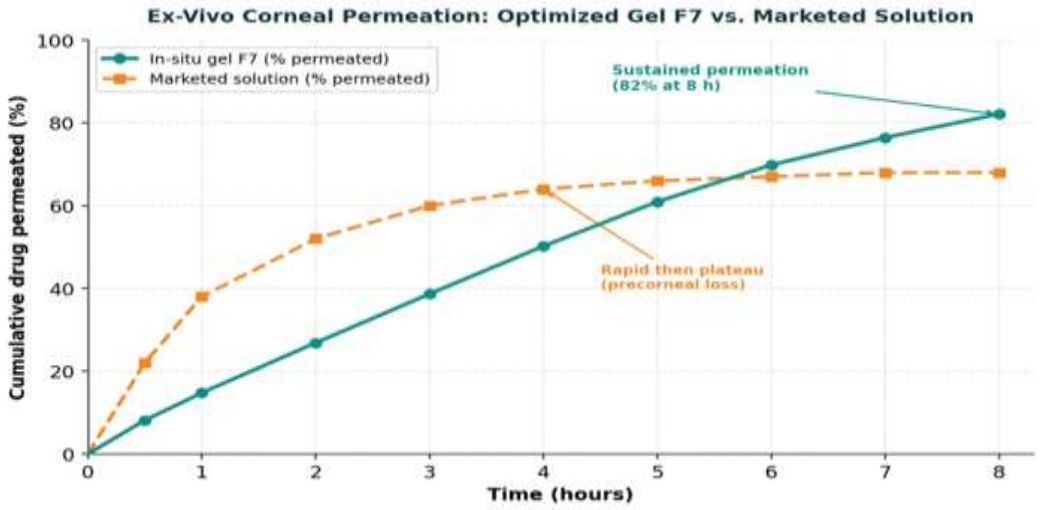

Figure 8. Ex-vivo corneal permeation of the optimized in-situ gel F7 compared with a marketed solution. F7 shows gradual, sustained permeation reaching 82% at 8 hours, whereas the solution permeates rapidly then plateaus.

Table 6. Ex-vivo corneal permeation of the optimized batch F7.

Time (h)	Cumulative % permeated (±SD)	Amount (µg/cm²)
0	0.00 ± 0.00	0.00
0.5	8.12 ± 0.42	4.06
1	14.75 ± 0.63	7.37
2	26.84 ± 0.91	13.42
3	38.67 ± 1.12	19.33
4	50.21 ± 1.35	25.10
5	60.94 ± 1.44	30.47
6	69.83 ± 1.58	34.92
7	76.45 ± 1.72	38.22
8	82.16 ± 1.85	41.08

This enhanced and sustained permeation can be attributed to several formulation-related factors acting together. The ion-activated gelation of sodium alginate and gellan gum raised the viscosity on instillation and minimised rapid tear wash-out, prolonging pre-corneal residence; the carboxyl groups of the two polymers conferred a mucoadhesive interaction with corneal mucin, enhancing drug–tissue contact; the increased gel

© JUL 2026 | IRE Journals | Volume 10 Issue 1 | ISSN: 2456-8880
DOI: https://doi.org/10.64388/IREV10I1-1719986

consistency reduced nasolacrimal drainage and widened the absorption window; and the cross-linked polymeric network modulated drug release and maintained a sustained concentration gradient across the cornea. Together these effects strongly support improved ocular bioavailability of timolol maleate from the optimized in-situ gel relative to a conventional solution, with the potential for reduced dosing frequency and better therapeutic outcomes in glaucoma.

3.6 Stability Studies

The optimized batch F7, packed in amber ophthalmic dropper bottles, remained physically and chemically stable over three months under both ICH long-term (25 °C / 60% RH) and accelerated (40 °C / 75% RH) conditions (Figure 9). The formulation stayed clear throughout, with only negligible changes in pH (7.20 to 7.10–7.15), a minor rise in viscosity (520 to 528–535 cps), and drug content remaining well within pharmacopeial limits (from 100.8% to 99.2% long-term and 98.6% accelerated). The 12-hour release was essentially unchanged (92% to 91.2% long-term and 89.5% accelerated). The absence of significant change confirms the robustness of the ion-activated gel system and supports the projected shelf life and suitability of the formulation for further scale-up.

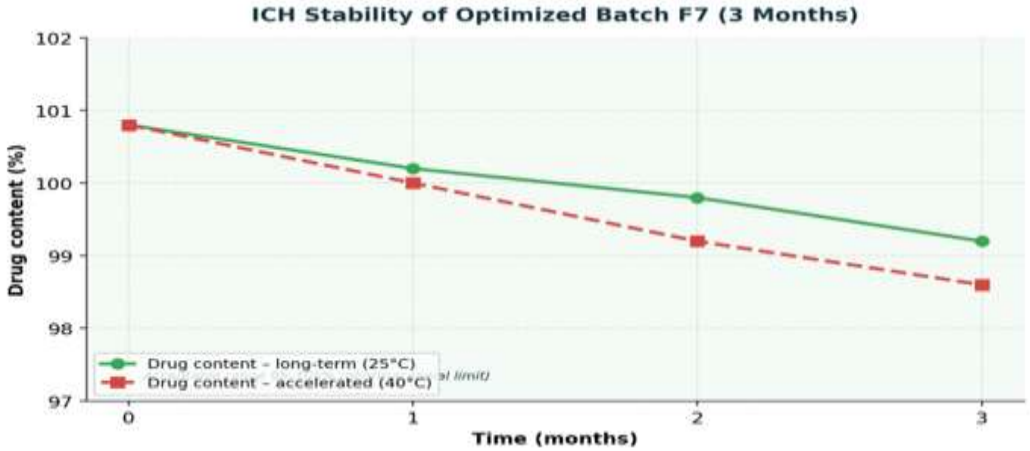

Figure 9. ICH stability of the optimized batch F7 over three months. Drug content remains well within the acceptable 95–105% range under both long-term and accelerated conditions, confirming formulation stability.

Taken together, the physicochemical, release, permeation and stability data identify F7—containing 0.5% sodium alginate and 0.25% gellan gum with 0.075% calcium chloride—as the optimized formulation. It combines an ocular-compatible pH, appropriate pseudoplastic rheology, immediate and strong gelation, uniform drug content, a burst-free sustained 12-hour release governed by non-Fickian transport, superior sustained corneal permeation and satisfactory stability, thereby fulfilling all the design objectives of the study.

3.7 Comparison with Previous Studies

The behaviour of the present system is consistent with, and in several respects extends, the published literature on ocular in-situ gels. El-Kamel reported that a Pluronic F127 in-situ gel of timolol maleate raised ocular bioavailability roughly 2.5-fold over conventional drops, establishing the principle that prolonging pre-corneal residence improves the delivery of this drug; the present ion-activated system achieves a similar prolongation while avoiding the temperature-dependence and relatively high polymer loads of poloxamer gels [1]. Mandal and colleagues, and later Makwana and co-workers, showed that sodium alginate combined with a viscosity enhancer forms an ion-activated ophthalmic gel giving sustained release and prolonged residence, and Sultana and colleagues demonstrated that gellan gum (Gelrite) gels efficiently with the ions of the lacrimal fluid to sustain drug release for around twelve hours—observations directly paralleled by the sustained 8–12-hour release seen here [27, 26, 36].

The choice of a natural-polymer, ion-triggered platform also addresses gaps identified in several recent reports. A number of published glaucoma in-situ gels have relied predominantly on synthetic polymers such as poloxamers and Carbopol, or on complex vesicular carriers, and several have lacked natural-polymer alternatives or quantified residence-time and permeation data [20, 21, 22]. By combining two complementary natural ion-sensitive polymers, the present formulation attains strong immediate gelation at low polymer concentration and adds a mucoadhesive interaction with corneal mucin that further prolongs contact. The non-Fickian release mechanism identified for F7 mirrors that commonly reported for cross-linked polysaccharide matrices, in which drug diffusion is accompanied by polymer swelling and relaxation. The enhanced and sustained ex-vivo corneal permeation, together with three-month ICH stability, indicates that the system is not only mechanistically sound but also pharmaceutically robust, supporting its progression to in-vivo intraocular-pressure evaluation.

IV. CONCLUSION

An ion-activated ocular in-situ gel of timolol maleate was successfully formulated using the natural, ion-sensitive gelling polymers sodium alginate and gellan gum for the sustained management of glaucoma. The system was designed to be instilled as a low-viscosity solution that gels rapidly on contact with the calcium ions of the tear fluid, thereby prolonging pre-corneal residence and sustaining drug release. Preformulation FTIR and DSC studies confirmed the compatibility and stability of the drug within the polymer matrix, and a validated UV method (λ_max 294 nm; R^2 = 0.9999) supported accurate quantification. All nine formulations were clear, of ocular-compatible pH, pseudoplastic, uniform in drug content and capable of immediate ion-triggered gelation, with viscosity, gel strength and release rate all increasing with polymer concentration. The optimized batch F7 exhibited an ideal balance of properties—strong immediate gelation, a burst-free sustained release of 92% over 12 hours following non-Fickian (Korsmeyer–Peppas) kinetics, markedly enhanced and sustained ex-vivo corneal permeation reaching 82% at 8 hours, and full physical and chemical stability over three months under ICH conditions. The developed natural-polymer, ion-activated in-situ gel of timolol maleate is therefore a promising, patient-friendly and cost-effective alternative to conventional eye drops, offering prolonged precorneal residence, sustained drug release, reduced dosing frequency and the potential for improved patient compliance and therapeutic efficacy in glaucoma therapy. Further in-vivo intraocular-pressure and pharmacokinetic studies are warranted to confirm these advantages and to advance the formulation toward clinical translation.

REFERENCES

[1] El-Kamel AH. In vitro and in vivo evaluation of Pluronic F127-based ocular delivery system for timolol maleate. Int J Pharm. 2002;241(1):47–55.

[2] Varshosaz J, Tabbakhian M, Salmani Z. Designing of a thermosensitive chitosan/poloxamer in situ gel for ocular delivery of ciprofloxacin. Open Drug Deliv J. 2008;2:61–70.

[3] Tham YC, Li X, Wong TY, Quigley HA, Aung T, Cheng CY. Global prevalence of glaucoma and projections of glaucoma burden through 2040: a systematic review and meta-analysis. Ophthalmology. 2014;121(11):2081–90.

[4] Global prevalence and burden of glaucoma: leading cause of irreversible blindness (76 million in 2020; 111.8 million projected by 2040). J Glob Health. 2024;14:04252.

[5] Sonawane SD, Patil RY, et al. In situ gel: a novel approach in sustained ophthalmic drug delivery. Int J Pharm Sci Res. 2013.

[6] Patil S, Kumar A. Ocular in situ gels: development, evaluation and advantages. Int J Pharm Investig. 2014.

[7] Paul S, et al. Sodium alginate: a versatile ion- and pH-triggered biopolymer for in situ gelling drug delivery. 2017.

[8] Stubb J, et al. Gelation properties and pharmaceutical applications of alginate and gellan gum. 2019.

[9] Timolol maleate ophthalmic solution: prescribing information (TIMOPTIC 0.25% and 0.5%). Merck / FDA DailyMed.

[10] Timolol maleate: non-selective β-adrenergic blocker for glaucoma – pharmacology and clinical use. Goodman & Gilman's Pharmacological Basis of Therapeutics. 2018.

[11] Balasubramaniam J, Pandit JK. Ion-activated in situ gelling systems for sustained ophthalmic delivery of ciprofloxacin. Drug Deliv. 2003;10(3):185–91.

[12] Patil S, et al. Formulation and evaluation of ocular in situ gel by cold method. 2015.

[13] Higuchi T. Mechanism of sustained-action medication: rate of release of solid drugs dispersed in solid matrices. J Pharm Sci. 1963;52:1145–49.

[14] Korsmeyer RW, Gurny R, Doelker E, Buri P, Peppas NA. Mechanisms of solute release from porous hydrophilic polymers. Int J Pharm. 1983;15:25–35.

[15] Rathore KS. In situ gelling ophthalmic drug delivery system: an overview. Int J Pharm Pharm Sci. 2010–2011.

[16] Jagdale S, et al. Compatibility study of timolol maleate with polymers by FTIR and DSC. 2016.

[17] Erol S, Karatas A. Development of a triple-polymer ophthalmic in situ gelling system of berberine HCl. 2025.

[18] Ozlem C, et al. Thermosensitive mucoadhesive timolol maleate ophthalmic in situ gel using poloxamers with chitosan/CMC. 2024.

[19] Xu Y, et al. Ion-sensitive in situ ophthalmic gel of brimonidine tartrate based on gellan gum: enhanced ocular retention and bioavailability. 2023.

[20] Lokapur V, et al. Proniosomal in situ gel of timolol maleate for glaucoma. 2022.

[21] Bharath S, et al. pH-triggered in situ gel of brimonidine tartrate using Carbopol/HPMC. 2020.

[22] Kurniawansyah IS, et al. Optimization of ophthalmic in situ gel using 3^2 factorial design (Poloxamer 407/HPMC). 2020.

[23] Prabhu P, Koland M. Thermosensitive in situ gel of ofloxacin using Poloxamer 407 and HPMC/PVA. 2019.

[24] Al-Bazzaz FY, et al. Thermosensitive in situ ocular gel of ciprofloxacin using poloxamer 407 and HPMC. 2018.

[25] Nagesh C, et al. pH-sensitive in situ ocular gel of ciprofloxacin. 2017.

[26] Makwana SB, et al. Formulation and characterization of in situ ophthalmic gel of ciprofloxacin hydrochloride using sodium alginate and HPMC. 2015.

[27] Mandal S, et al. In situ gel-forming ophthalmic system of ciprofloxacin using sodium alginate (ion-activated). 2012.

[28] Kavitha K, Rajas NJ. Sustained-release in situ ophthalmic gel of levofloxacin hemihydrate using gelrite. 2011.

[29] Cao F, et al. In situ gelling ophthalmic system of azithromycin using poloxamer and Carbopol 974P. 2010.

[30] Gratieri T, et al. Thermosensitive mucoadhesive poloxamer/chitosan in situ gel for prolonged ocular retention. Eur J Pharm Biopharm. 2010.

[31] Qian Y, et al. Thermosensitive in situ gel of methazolamide for ocular delivery. 2010.

[32] Al-Kassas RS, El-Khatib MM. Controlled-release ophthalmic in situ gels of ciprofloxacin using carbopol and alginate. 2009.

[33] Mohan EC, et al. pH-, thermo- and ion-responsive in situ ocular gels of ciprofloxacin. 2009.

[34] Mansour M, et al. Thermosensitive in situ gel of ciprofloxacin hydrochloride using poloxamers. 2008.

[35] Gupta H, et al. Thermo- and pH-triggered in situ gel of timolol maleate using Pluronic F-127 and chitosan. 2007.

[36] Sultana Y, et al. Ion-activated in situ ophthalmic gel of pefloxacin mesylate using gellan gum. 2006.

[37] Nanjawade BK, Manvi FV, Manjappa AS. In situ-forming hydrogels for sustained ophthalmic drug delivery. J Control Release. 2007;122(2):119–34.

[38] Tham YC, et al. Global glaucoma prevalence by region and type (POAG/PACG). Ophthalmology. 2014;121(11):2081–90.