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REVIEW

Unleashing the Potential of Cubosomes to Overcome Ocular Barriers in Precision Drug Delivery

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ABSTRACT

The complex structure of the human eye presents significant challenges for administering drugs, particularly in the treatment of disorders affecting the anterior and posterior segments. Traditional ocular formulations, which involve eye drops, are limited by low absorption, rapid tear turnover, and anatomical barriers, necessitating repeated administration and thereby limiting therapeutic efficacy. Recent advances in nanotechnology have led to the development of new pharmaceutical delivery methods, with cubosomes emerging as a promising approach. Cubosomes are nanostructured lipid carriers with a unique honeycomb-like cubic lattice composed of bicontinuous lipid bilayers and aqueous channels. Due to their unique structure, they are capable of encapsulating amphiphilic, hydrophobic, and hydrophilic compounds, resulting in controlled and extended drug release. Furthermore, their small particle size, bioadhesive properties, and biocompatibility increase corneal retention and permeability, making them a promising approach for ocular therapy. This review focuses on the advantages of cubosomes in overcoming ocular barriers, discussing their preparation methods, which include both top-down and bottom-up approaches, as well as spray drying and microfluidic methods. Additionally, it explores optimization strategies that target particle size, surface charge, and viscosity to enhance efficacy. Though cubosomes offer transformative potential, challenges such as scalability, excipient-induced pain, and regulatory hurdles persist.

1 | Introduction

The eye is a highly intricate organ with very distinct anatomy and physiology. It is subdivided into two segments: the anterior, which comprises around one-third of the eye, and the posterior, which comprises the rest. Tissues like the conjunctiva, cornea, ciliary body, lens, and iris together make up the anterior segment of the eye, and tissues like the retinal pigment epithelium, choroid, sclera, vitreous humor, optic nerve, and neural retina make up the posterior portion of the eye [1]. Numerous vision-threatening diseases affect these two segments of the eye. Topical instillation is the primary non-invasive therapeutic method for diseases affecting the anterior region [2]. A total of 90% of the marketed ophthalmic formulations are traditional dosage forms, with ophthalmic eye drops dominating the market due to their ease of use and patient compliance.

Abbreviations: GMO, Glyceryl monooleate; PPO, Polypropylene oxide; P407, Poloxamer 407; PPO, Polyethylene oxide; Re, Reynolds number; TEM, Transmission electron microscopy; PDI, Polydispersity index; DSC, Differential scanning calorimetry; Jss, Steady state flux; AUC, Area under curve; TM, Timolol maleate; VCZ, Voriconazole; EE, Entrapment efficiency; MIC, Minimum inhibitory concentration; LLCs, Lyotropic liquid crystals; MRT, Mean residence time; Kp, permeability coefficient.

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Despite the widespread use and patient-friendly nature of topical instillation, ocular bioavailability remains a major challenge [3]. Several anatomical and physiological constraints, including nasolachrymal drainage, dynamic barriers, reflex blinking, ocular static, and tear turnover, impede deeper ocular penetration, allowing less than 5% of the administered dosage to reach deeper tissues after topical application. Concerns regarding the above-mentioned barriers also make it challenging to achieve therapeutic drug concentrations following topical eye drop administration into the posterior segment [4, 5]. Alternatively, drug administration to the posterior segment can be achieved through various modes of administration, including periocular injections, systemic administration, and intravitreal injections. Yet, due to the relatively small size of the eye in comparison to other organs and tissues of the body, systemic delivery may not be feasible. Intravitreal injection is commonly advised for treating posterior ocular diseases. However, it can lead to recurrent eye punctures, causing hemorrhage, endophthalmitis, retinal detachment, and poor patient tolerance [6]. Several approaches have been employed to date to enhance ocular bioavailability and overcome delivery barriers in ocular drug delivery systems. Moreover, topical instillation in the lower precorneal pocket is the most preferred route owing to its patient compliance [7]. Conventional delivery forms, such as penetration enhancers, cyclodextrins, and prodrugs, have gained wide acceptance due to their stability, cost-effectiveness, enhanced drug product efficacy, and improved patient acceptability. Despite these advantages, conventional forms have limitations, including frequent administration, drug loss through lachrymal fluid, tears, and low absorption [8]. Only 20% of the administered dosage stays in the precorneal pocket, with the remainder lost owing to reflux and blinking. The drug in the precorneal region undergoes passive diffusion across the cornea. Therefore, efficient ocular delivery requires high corneal penetration and extended drug contact time.

For this purpose, nanotechnology is being utilized for administering drugs in the ophthalmic field due to its low irritation, compatibility with eye tissue, and improved bioavailability [9]. Numerous nanocarriers, including niosomes, nanosuspensions, nanoparticles, micelles, cubosomes, dendrimers, and liposomes, have shown promising results for overcoming these barriers. These novel carriers can overcome ocular obstacles and associated issues with traditional forms of treatment. Furthermore, these innovative formulations have a high precorneal residence duration, negligible irritation, prolonged drug release, and enhanced ocular bioavailability [10]. They are also biocompatible, have well-established scale-up technology, and offer controlled and sustained drug release. Although these novel carriers have promising potential, their clinical use is limited due to stability issues, difficulty delivering drugs through the eye's inner structure via excipient-induced irritation, sterilization concerns, local infusion complications, and high costs. For instance, liposomes undergo physical and chemical instability due to the oxidative degradation of phospholipids, which is a drawback that involves the use of costly raw materials [11]. Niosomes, while having a similar structure to liposomes, tend to experience stability issues. They are susceptible to aggregation and fusion, which results in drug leakage and decreased shelf-life, restricting their use in drug delivery systems [12]. Nanosuspensions are beneficial for enhancing the aqueous solubility of poorly water-soluble drugs; however, they are susceptible to physical instability issues, such as particle aggregation and sedimentation. These types of stability problems can compromise drug efficacy and complicate storage and administration. Micelles have been reported to increase the solubility of hydrophobic drugs; however, they exhibit faster release rate of drug due to less diffusional path and thus illustrated less therapeutic duration [13, 14]. Cubosomes, as third-generation lipid-based nanocarriers, display a distinctive bicontinuous cubic phase structure that allows the simultaneous encapsulation of hydrophilic and hydrophobic drugs, providing advantages such as high surface area, biocompatibility, and sustained release [15]. Compared to liposomes, cubosomes exhibit greater structural stability and more effective drug entrapment, especially for poorly soluble compounds like curcumin [16]. Polymeric micelles have already achieved clinical success, with several formulations like Genexol-PM (paclitaxel-loaded micelles) approved for cancer therapy. However, micelles often suffer from poor therapeutic efficacy which can be overcome by cubosomal formulation due to tortuous diffusional path and targeting capacity [17, 18]. Dendrimers, with their highly branched structures and adjustable surface functionalities, have demonstrated potential in improving the solubility and delivery of hydrophobic drugs such as camptothecin and methotrexate. However, unmodified dendrimers, like non-PEGylated PAMAM, have been associated with higher haemolytic activity, indicating possible cytotoxicity. This toxicity can be mitigated through surface modifications such as PEGylation, which enhances their biocompatibility and makes them more suitable for therapeutic use. Nonetheless, their clinical application remains limited due to concerns over cytotoxicity, complex synthesis, and high production costs [19].

Given the aforementioned setbacks of different nanocarriers, cubosomes have shown tremendous potential for ocular drug delivery [20]. They are minuscule dispersions of lipid bicontinuous cubic phases in water composed of a lipid interior and aqueous surroundings arranged in a cubic lattice together with hydrophilic molecules [21]. The bicontinuous cubic phases offer a significant advantage due to their capability to autonomously adjust their membrane curvature without being limited by the size of nanoparticles over extensive lengths. The system's capacity to tune and adapt is due to its intrinsic bilayer membrane structure. The membrane forms a lattice network of linked water channels [22]. Surfactants form 3D bilayers with limited surface area and densely packed structures. These structures feature bicontinuous zones of lipid and water, creating a honeycomb-like structure. Cubosomes are formed by disrupting the cubic lipid-water phase in a three-phase area [23]. The formation of cubosomes depends on the phenomenon of lipid self-assembly, where a lipid mixture spontaneously arranges itself into a bicontinuous cubic phase when combined with loading proteins, stabilizers, and the desired chemical entity [24]. Cubosomes possess several advantages that make them attractive for use in ocular drug delivery, including a small particle size with a large surface area, biodegradable properties, and good adhesivity. Furthermore, they can extend corneal retention time and improve corneal permeability.

Recent progress in lipid-based depot systems, such as Brixadi and Buvidal for opioid use disorder and Oczyesa for acromegaly, highlighted the translational promise of injectable lipid nanocarriers [25, 26]. The Camurus FluidCrystal platform has facilitated several long-acting injectables advancing to Phase 3 trials and

gaining regulatory approval, confirming the viability of lipid-based sustained-release systems in clinical practice [27]. These successes suggest that cubosomes could also have future applications in ophthalmology, provided that manufacturing and safety issues are effectively addressed.

This review discusses the various advantages, methodologies, and characterization of cubosome formulations. Moreover, different barriers in ocular drug delivery are also highlighted through the exploration of various drugs utilized after loading into cubosomes.

2 | Challenges in Ocular Drug Delivery

Owing to the presence of complex barriers in the eye, the ocular delivery of drug molecules is compromised as these barriers limit the entry of therapeutic substances. A significant portion of the administered dose is eliminated from the eye through systemic absorption via the conjunctival sac or through nasolacrimal drainage after topical application, potentially leading to suboptimal therapeutic outcomes for the drug.

The ocular absorption of the drug is hindered by the conjunctival and corneal epithelial cells. The blood-ocular barrier restricts the entry of hydrophilic macromolecules into the systemic circulation and inhibits xenobiotics from entering the eye through systemic routes [28]. The blood-ocular barrier consists of two components: the anterior blood-aqueous barrier and the posterior blood-retinal barrier. The blood-aqueous barrier consists of non-fenestrated vascular endothelial cells covering the iris blood vessels and tight junctions (Zonula occludens) connecting epithelial cells of the non-pigmented ciliary body. The blood-retinal barrier consists of two components: the inner retinal microvascular endothelium and the outer retinal blood barrier created by retinal pigment epithelial cells [29]. These physiological barriers maintain retinal homeostasis by controlling the movement of electrolytes, nutrients, osmotic water flows, peptides, and proteins into and out of the retina. Alterations in the blood-retinal barrier are associated with prevalent posterior segment retinal disorders, including diabetic retinopathy and age-related macular degeneration. The aqueous tear film consists of lipocalin, an aqueous layer enriched with lysozyme, an outer lipid layer, and an innermost mucin layer. Tear film also acts as a protective layer between the external environment and the corneal layer. The anterior segment is affected by various diseases, which include anterior uveitis, cataracts, glaucoma, and allergic conjunctivitis [30]. Traditional ophthalmic formulations showed poor bioavailability (less than 5%) due to physiological constraints that restrict their effectiveness. This low bioavailability implies frequent dosage administration, which restricts patient compliance. Therefore, innovative strategies are required to improve the therapeutic outcome of the ocular conditions. The primary challenge in ocular drug delivery lies in overcoming these barriers to achieve a therapeutically effective concentration of the drug with enhanced and prolonged bioavailability at the target ocular site. In addition to the problem of reduced bioavailability, conventional drug formulations, as mentioned in (Table 1), namely ointments, solutions, suspensions, etc., administrated topically require frequent dosing to show the desired therapeutic effect at the target site.

To overcome these challenges, there has been an increasing focus on novel drug delivery systems such as nanoparticles, liposomes, hydrogels, nanoemulsions, niosomes, micelles, and dendrimers [31]. However, they face certain constraints, including possible toxicity, limited drug loading capacity, and drainage from the ocular surface. Under some circumstances, nanoparticles may be harmful to eye tissues, therefore posing safety considerations [32]. Despite being biocompatible and capable of encapsulating both hydrophilic and hydrophobic drugs, liposomes have stability and degradation issues [33]. Nanoemulsions offer increased drug solubility and absorption, but they are prone to instability and rapid clearance. Controlled drug release properties are shown by dendrimers but are limited by biocompatibility concerns and probable toxicity at higher concentrations [34]. Micelles solubilize hydrophobic drugs, yet they show poor stability in ocular surroundings, which causes drug leakage [35]. Although hydrogels are well-known for their high hydration and continuous drug release, they have little mechanical strength and are prone to bacterial contamination. Similarly, niosomes have limited encapsulation efficiency and fast ocular clearance, even if they are more stable than liposomes [36]. These limitations highlight the need for advanced nanosystems, such as lyotropic liquid crystals (LLCs). These are self-assembled structures formed from amphiphilic molecules that include surfactants and lipids. They possess a distinctive mesophase that blends the ordered structure of crystalline solids with the fluidity of liquids, making them highly effective for applications in drug delivery. These characteristics are especially beneficial for ocular drug delivery, where extended retention times and sustained drug release are essential for enhancing their therapeutic efficacy.

3 | Cubosomes are a Promising Approach to Overcome Challenges with the Ocular Delivery of Bioactive

Despite the promising potential of these nanocarriers, several challenges limit their practical application, including limited drug loading, the risk of aggregation, physical instability, and a short residence time. In light of the aforementioned issues, cubosomes have demonstrated notable success in achieving wide applications for ocular drug delivery. Thus, the use of cubosomes as a drug carrier could significantly reduce the drawbacks, which would result in significant advancements in ocular treatment. Cubosomes are liquid crystalline nanostructured with nano dimensionality being biocompatible in nature, which consist of amphiphilic lipids in certain ratios that form the self-assembled matrices of the bicontinuous cubic phase nanoparticulate colloidal dispersions. They represent the honeycomb structures comprising curved bicontinuous lipid bilayers organized in three dimensions to produce an unbroken, regular, cubic, symmetrical surface. It has two internal aqueous channels that may be used for bioactive substances like synthetic drugs, proteins, and peptides. Cubosomes are a promising carrier due to their thermodynamic stability, capacity to encapsulate various compounds, bioadhesion, and controlled release after functionalization [18].

Given that they have a large surface area and more interior sections, they exhibit higher entrapment efficiency and slower release of therapeutic substances compared to lamellar liposomes. Lipid bilayers in the cubosomes' structure serve as a

TABLE 1 | Different approaches have been used for various ocular diseases.

S.no	Delivery	Drug	Disease	Challenges in drug delivery	References
Conventional delivery systems					
1	Eye drops	Gatifloxacin	Conjunctivitis	Frequent Administration, Low residence time on the conjunctival surface	[37]
2	Ocular inserts	Pilocarpine Hydrochloride	Glaucoma	Proper placement of the insert in the conjunctival sac is difficult for patients	[38]
3	Hydrogels, emulsions, and ointments	Tetracyclines, macrolides, omega fatty acids, NSAIDs, Cyclosporine A	Dry eye syndrome	Cytotoxicity in the case of emulsions, ointments may cause blurred vision	[39]
4	Eye drops	Bendazac, Aldose reductase inhibitors, Calpain inhibitors	Cataract	Poor penetration into the lens, limited ocular absorption	[40]
5	Intravitreal implant	Corticosteroids, Antimetabolites, T-cell inhibitors, Alkylating agents	Uveitis	Invasive approach, compromised patient compliance, risk of elevation of intraocular pressure	[41]
6	Intravitreal injection	Corticosteroids, Anti-VEGF agents	Diabetic Retinopathy	Need for frequent injections, tachyphylaxis, and elevation in intraocular pressure	[42]
Novel delivery systems					
7	NLCs	Dasatinib	Corneal neovascularization	Chances of aggregation, inconsistent penetration efficiency, and limited drug loading	[43]
8	Liposomes	Fluconazole	Fungal keratitis	Short residence time, poor stability, low entrapment efficiency, and early drug leakage	[44]
9	Niosomes	Brimonidine tartrate	Glaucoma	Aggregation, drug leakage, and physical instability	[45]

protective barrier, limiting degradation and improving drugs' stability. Furthermore, the cubic lattice form allows for control over drug release kinetics by adjusting the lipid content and surface properties. This enables the formation of sustained or triggered drug release systems that improve therapeutic efficacy while minimizing negative effects [46]. Cubosomes can be functionalized by conjugating the surface with targeting ligands or chemical moieties, therefore avoiding systemic toxicity by targeting specific sites of interest. The two most commonly employed lipids involved in the preparation of cubosomes are glyceryl monooleate (GMO), also known as monoolein, and phytantriol, a non-ionic surfactant. GMO is a synthetic product of glycerides of oleic acid and other monooleate fatty acids, which fall under the family of amphiphilic lipids. Due to the presence of a hydroxyl group in the head region and a hydrocarbon chain in the tail region, GMO exhibits both hydrophilic and hydrophobic characteristics simultaneously [47]. Phytantriol serves as a GMO alternative in cubosome formulations. It can establish a bicontinuous cubic structure in aqueous environments under physiological temperatures and conditions. Due to its unique chemical stability, increased skin penetration, and ability to retain moisture, phytantriol has gained more limelight in the biomedical fields. Additionally, phytantriol can maintain the release of different drug molecules, particularly drugs with hydrophilic characteristics [48]. Besides amphiphilic lipids, surfactants play an essential role in providing colloidal stability to cubosomes, thus averting their re-coalescence in the bulk cubic phase. The most widely used surfactant in the preparation of cubosomes is Poloxamer 407 (P407), a triblock copolymer with polypropylene oxide (PPO) chains exposed to the surrounding aqueous phase, whereas polyethylene oxide (PEO) portions are located at the surface of cubosomes or within bilayer structures [49]. P407 is usually used at a concentration of up to 20% w/w, dependent on the dispersed phase. Monoglyceride polymer mixtures, on the other hand, have concentrations ranging from 2.5% to 10% w/w, depending on the total dispersion weight. The surfactant's stabilizing action is due to the adsorption of hydrophobic portions on the surface of particles. The steric shielding is provided by the hydrophilic portion extending into aqueous media [50].

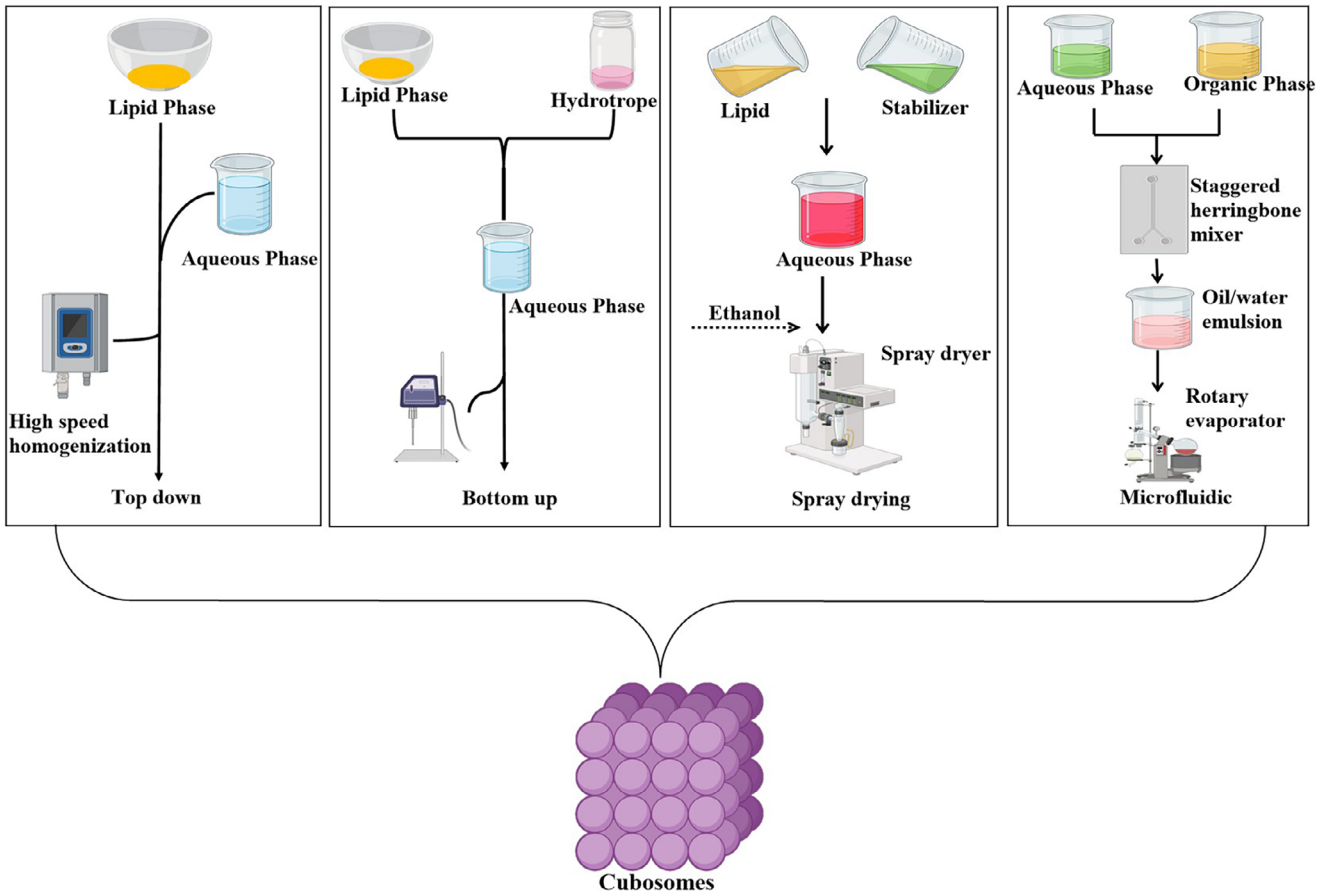

FIGURE 1 | Methods of preparation of cubosomes.

4 | Methods of Preparation for Cubosomes and Their Characterization

Cubic phases typically consist of three macroscopic phases: precursor, bulk gel, and particle dispersion. The precursor form occurs as a solid or liquid that transitions to a cubic phase upon stimulation, such as contact with a liquid. Bulk cubic phase gel is an optically isotropic, rigid, solid-like substance in equilibrium with water that may be distributed into particles known as Cubosomes, and these can be produced via different techniques such as (i) the Top-down method, (ii) the Bottom-up method, (iii). Spray drying approach (iv) Microfluidics approach (Figure 1) [51].

4.1 | Top-Down Method

The top-down technique starts with an adequate starting material and then shapes the functionality from that material. This approach first generates a bulk cubic phase, which is then dispersed into cubosome nanoparticles by high-energy processing [52]. The bulk cubic phase resembles a transparent, rigid gel formed of water-saturated, cross-linked polymer chains. However, cubic phases are unique in that they feature a periodic liquid crystalline structure. The eruption of these cubic phases transpires in a direction aligned with the shear direction, and the energy necessitated is directly related to the number of tubular network branches that shatter. Cubic phase rupture occurs when a bilayer breaks under shear stresses and transforms along slip planes. The yield stress in cubic phases increases with the addition of more bilayer-forming surfactants and oils and is inversely proportional to the size of the crystalline unit cell. At modest shear rates, dispersed liquid crystalline particles arise, whereas a defect-free bulk phase is reconstituted at higher speeds. At high oscillation frequencies, cubic phases show remarkable flexibility [53].

4.2 | Bottom-Up Method

This approach generates the nanostructure building blocks and then integrates them into the finished material. This approach facilitates the development and crystallization of cubosomes at the molecular length scale from precursors [54]. An example of this technique is the solvent exchange method, often referred to as solvent shifting and nanoprecipitation. The first phase of this process involves the formulation of a water-like precursor solution by the dissolution of the lipid (e.g. phytantriol or monoolein) and stabilizer (e.g. Pluronic F127) in a water-miscible solvent, such as ethanol (act as hyrotropic solvent). Subsequently, the precursor solution is injected into the water phase, leading to the dilution of the hydrotropic solvent, faster solvent exchange occurs between ethanol and water, and a rapid reduction in lipid solubility. As a result, these lipid molecules spontaneously self-assemble into a bicontinuous cubic phase driven by the hydrophobic effect and their molecular shape (lipid packing parameters). Subsequently, ethanol is evaporated by a mild evaporation process, and transient

vesicular intermediates reorganize into a thermodynamically stable cubosome. Pluronic F127 forms a thin layer, preventing aggregation and providing stability to the structure.

The internal architecture of cubosomes features a periodic lipid bilayer network with interwoven aqueous channels, reducing free energy and enabling the encapsulation of both hydrophilic and hydrophobic substances [52, 55, 56]. Hence, the main advantage of the solvent exchange method over other methods, especially the top-down approach, is that it results in the formation of stable particles with a small size and a narrow size distribution. This technique's little energy input preserves the integrity of temperature-sensitive molecules [57, 58].

4.3 | Spray Drying Method

In this process, a powder form of Cubosomes is characterized by organic solvents that evaporate on exposure to air. A wave of hot air is used to atomize the lipid-surfactant solvent mixture, leading to the rapid evaporation of the solvent and the formation of a dry powder of cubosomes. The lipid and stabilizer are then dissolved in ethanol (alternatively, binary solutions can be used instead of ethanol). A second aqueous phase containing a hydrophilic solid carrier, such as sorbitol or dextran, is then mixed with the previously prepared lipid mixture [59, 60].

4.4 | Microfluidics Approach

Microfluidics is an advanced method used for the preparation of cubosomes. In particular, the staggered herringbone mixer device is employed to prepare cubosomes. The device has two inlets, where the lipid dissolved in ethanol is filled in the first inlet, and the aqueous phase is filled in the second inlet with a syringe pump [61]. The overall flow rate and the ratio of the flow rate of the lipid and aqueous phases need to be tuned to generate the specified size of cubosomes. The herringbone-patterned microchannels create chaotic advection in the ethanol and aqueous phases, enhancing mixing efficiency. After attaining these conditions, a nanoemulsion is formed, with a lipid layer coating the interface between the two solvent forms. The formed nanoemulsion is collected from the outlet and then subjected to a rotary evaporator, where the solvent (ethanol) evaporates. Ultimately, a cubosome dispersion with a narrow size distribution and small particle size is achieved [62].

After employing specific methods for cubosome preparation, it is critical to assess their structural, physicochemical, and functional features to verify their appropriateness for the intended use. Characterization approaches give insights into cubosomes' stability, shape, size distribution, internal nanostructure, and encapsulation efficiency, allowing a thorough knowledge of their performance and potential as given in Table 2.

5 | Factors Affecting Cubosomes' Interaction with Ocular Tissues

Topical drug administration to the cul-de-sac occurs either by the non-corneal channel, whereby the drug diffuses across the sclera and conjunctiva, or through the corneal membrane into intraocular tissues. The topical route for drug delivery is preferred for treating superficial infections and inflammation, as well as anterior segment diseases, including glaucoma and uveitis, due to its benefits, including avoidance of systemic absorption, bypassing hepatic metabolism, and convenience of delivery [73]. However, topical therapies have only 5%–10% ocular bioavailability, which makes it challenging to attain therapeutic drug concentrations, particularly in the posterior part of the eye. Most therapeutic drugs are absorbed via transcellular transport via the corneal epithelial membrane and stroma, as opposed to non-corneal absorption. Corneal permeability is affected by the active molecule's physicochemical properties, such as surface area, diffusivity, pKa, log *p* value, and concentration gradient, which are consistent with other biological membranes. To overcome these limitations, nanocarriers like cubosomes have been developed to enhance ocular drug delivery [74]. The various factors that affect their interaction with the ocular tissues are discussed as follows (Figure 2).

5.1 | Lipid

The polar lipid GMO in cubosomes enhances permeability by mimicking the biological membrane and fusing with the lipophilic corneal epithelial membrane, which is a substantial ocular barrier [75]. Cubosomes arranged in a cubic honeycomb structure enable longer retention on the corneal surface, thereby allowing extended time for drug permeation through the lipophilic corneal epithelium. Kaul and colleagues reported that tobramycin-loaded cubosomes showed a 3.6-fold higher Papp and a 3.7-fold higher Jss than Tobrex eye drops. This improvement was attributed to their bioadhesive properties and extended retention on the cornea [76, 77]. Furthermore, a suitable lipid-to-surfactant ratio provides better stability to the cubosomes formulated than other lipid/surfactant-based vesicles by virtue of a large lipid bilayer ratio and strong electric repulsion forces [78]. Phytantriol lipids mimic biological membranes that facilitate greater adhesion with the corneal epithelium, and thereby drug permeability and residence time on the ocular surface. This is complemented by the mucoadhesive characteristics of cubosomes that allow them to adhere to the mucin layer in the eye and maximize contact time and reduce the frequency of administration. Additionally, phytantriol cubosomes allow the encapsulation and prolonged release of hydrophilic as well as hydrophobic drugs, offering prolonged therapeutic activity [79]. Their efficiency in drug delivery of drugs like latanoprost and timolol maleate for glaucoma therapy with increased bioavailability and minimal cytotoxicity has been demonstrated by studies [78, 80].

5.2 | Viscosity

Viscosity is an essential factor that tends to improve the ocular residence time and, hence, the ocular bioavailability of the administered ophthalmic formulation, as these systems are more resistant to drainage to the nasolacrimal duct and rapid tear dilution [81]. The transparent cubic phase is physically stable in contact with the excess water because of its high viscosity. Furthermore, these structural properties enhance the adherence of cubosomal systems to the conjunctival sac and ciliary muscle, thus prolonging retention time [78, 82].

TABLE 2 | A summary of characterization techniques employed for the Cubosomes.

S. No	Evaluation Parameter	Characterization Technique	Significance	References
1	Direct	Electron microscopy	Shape	[63, 64]
		X-Ray scattering	Crystal arrangement	[65, 66]
			Internal nanostructure, determination of critical packing parameter	[67, 68]
2	Indirect	Dynamic light scattering	Particle size and polydispersity index	[69]
		Zeta potential	Charge of particles	[70]
		Entrapment efficiency	Percentage of drug encapsulated in carriers	[71]
		Differential scanning calorimetry (DSC)	Thermotropic behaviour, transition temperatures, and enthalpies	[72]

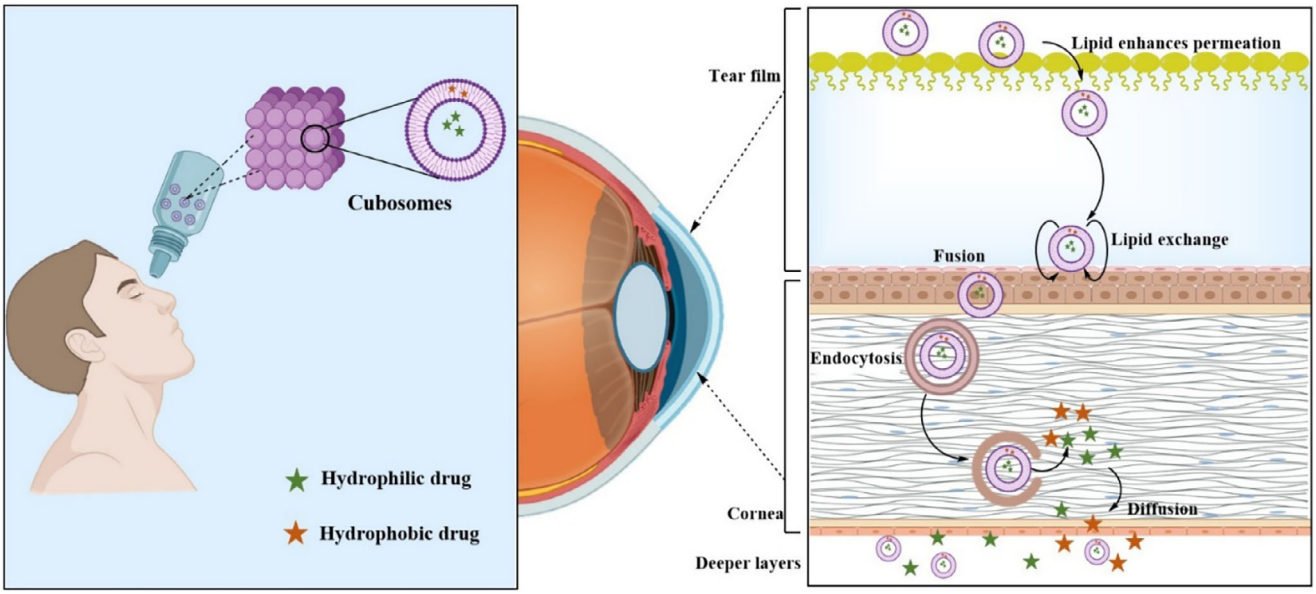

FIGURE 2 | Role of lipid factors in affecting the cubosome interaction with the ocular tissues.

5.3 | Surface Tension

The enhancement in the transcorneal permeation of the prepared cubosomes through the corneal epithelium is attributed to the decreased surface tension due to increased miscibility with the lipid layer of the precorneal tear film and spreadability over the lipophilic corneal epithelium [83]. Therefore, lowering the surface tension of the cubosomes allows better wetting of the lipophilic surface of the corneal epithelium [84, 85].

5.4 | Particle and Pore Size

The particle size is considered an important factor influencing the dispersion of the ophthalmic dosage form and ocular bioavailability of the loaded drug. Usually, the particle size of cubosomes <200 nm is preferred so that it can easily permeate through the corneal surface [86]. In addition to small particles, the small pore size of the cubosomes also contributes to achieving the controlled release of the drug through the regular channels of the cubosomal structure. The release rate of the entrapped hydrophobic drug is curtailed due to its strong affinity toward the hydrophobic region of the cubic phase. In contrast, the entrapped hydrophilic drugs encounter immediate drug release due to increased contact surface area with the surrounding aqueous environment [87].

5.5 | pH

The effect of pH on ocular drug permeability is closely linked to the drug's ionization properties. Acidic drugs like ciprofloxacin are more negatively charged at high pH, causing electrostatic repulsion from the negatively charged corneal membrane and decreasing permeability. Conversely, at low pH, these drugs become unionized or positively charged, which increases permeability through electrostatic attraction, as demonstrated by Alharbi and colleagues Hosny [88]. In contrast, basic drugs such as ketoconazole exhibit increased permeability at low pH

due to their cationic form, which interacts favourably with the corneal or skin tissue membrane. Rapalli et al. found that ketoconazole-loaded cubosomes showed much greater skin permeation (92.73%) than the marketed cream (18.05%) in an ex vivo study using phosphate buffer saline at pH 7.4 [89]. Amphoteric drugs, like brimonidine tartrate, display more complex behavior due to their ability to carry either positive or negative charges depending on the pH. Eldeeb et al. demonstrated that brimonidine tartrate formulated in cubosomes exhibited a 1.6-fold increase in corneal permeability at physiological pH, suggesting that its amphoteric nature, combined with lipid-based nanocarriers, allows for favourable interactions and sustained drug delivery across different pH conditions [90].

6 | Ocular Drugs Loaded into Cubosomes

6.1 | Sertaconazole

Sertaconazole, an imidazole fungistatic agent, mediates its antifungal activity through the inhibition of 1,4-α demethylase enzyme. Furthermore, it impedes the formation of the fungal cell membrane by preventing the synthesis of ergosterol and promotes cellular permeability, resulting in the leakage of cellular components [91, 92]. Though sertaconazole's ocular tolerance makes it an effective agent against fungal keratitis, its poor aqueous solubility poses a major challenge for its formulation as conventional eye drops. A study with the aim of overcoming the solubility issue of sertaconazole was conducted through the development of sertaconazole-loaded cubosomes to produce enhanced transcorneal permeation and adequate stability. A 3^3 central composite face-centred design was utilized to develop and optimize sertaconazole-loaded cubosomes and examine the impact of formulation factors on dependent variables. The optimised formulation demonstrated promising mucoadhesive properties and maintained its physicochemical characteristics after terminal sterilisation with gamma irradiation, indicating its suitability for ocular use. The statistical analysis showed that the cubosomal formulation significantly improved transcorneal drug delivery, with increased steady-state flux (Jss) and permeability coefficient (Kp), resulting in a 2.4-fold increase in cumulative drug permeation at 24 h (Q24h) compared to a conventional sertaconazole suspension. Furthermore, in vivo studies indicated superior corneal uptake and excellent ocular tolerability, supporting the potential of cubosomes as an effective ocular delivery system for sertaconazole [93].

6.2 | Acetazolamide

Acetazolamide primarily lowers the intraocular pressure and is particularly used in the management of glaucoma. Moreover, it exhibits its action through the inhibition of the activity of carbonic anhydrase in the eye, resulting in the alleviation of the production of aqueous humor [94]. To date, no such topical formulation for acetazolamide has been commercially available because of its limited corneal permeability and poor aqueous solubility, resulting in low ocular bioavailability and causing an inadequate quantity of the drug to reach the ciliary body. Several research studies have made attempts to enhance the ocular bioavailability of acetazolamide through various topical formulations, and then cubosomes were chosen as a more suitable option to enhance drug solubility and penetration through the cornea. In a study, different acetazolamide cubosomes were developed, and the optimized formulation displayed a particle size of 359.5±2.8 nm and a polydispersity index of 0.18±0.03 [90, 95]. In addition, a 4-fold increase was observed in the results of ex vivo permeation studies. A superior therapeutic efficacy was demonstrated by optimized formulation representing 38.22% decrease in intraocular pressure in comparison to 21.99% and 31.14% decrease for commercial Cidamex tablets and Azopt eye drops respectively; the effect persisting for a longer time was identified by mean residence time (MRT). Hence, acetazolamide-loaded cubosomes offered a niche method for topical drug delivery for glaucoma management, extending the intraocular pressure-lowering activity for more than 9 h [96].

6.3 | Fluorometholone

Fluorometholone, a topical anti-inflammatory and corticosteroid, is used to reduce inflammation after eye surgery and treat keratoconjunctivitis. While passing through the cornea, the drug transforms into dehydrofluorometholone by undergoing various metabolic processes, thus reducing its efficacy [97, 98]. Its lipophilic properties and low solubility in water further limit its application in the pharmaceutical field [99]. Hence, novel drug delivery systems have the potential to effectively address the limitations associated with the use of eye drops. To overcome the issue of low drug solubility, enhanced penetration, longer retention time, and fewer side effects, the drug was encapsulated in GMO-based cubosomes using a surfactant Pluronic F127. The optimized cubosomal formulation demonstrated a particle size ranging between 125.10 and 434.75 nm. Further, the in vitro release analysis showed that the drug was completely released after 12 h, resulting in a reduction in drug administration. Additional studies on the accelerated stability confirmed that there is no difference in the release profile of the drug in vitro and no change in the physical properties of the formulation, thereby showing formulation stability. Therefore, cubosomes proved to be suitable carriers for the delivery of Fluorometholone to the eye [100].

6.4 | LM22A-4

LM22A-4, a small molecular neurotrophic that specifically activates tropomyosin receptor kinase B, the cognate receptor of BDNF. However, it presents a clinical difficulty due to its short half-life, which necessitates its frequent intravitreal administration, making it impractical for continued glaucoma treatment [101, 102]. Through the use of Annexin V-conjugated cubosomes, Ding and colleagues encapsulated LM22A-4 to enable its targeted delivery to injured retinal ganglion cells. According to cryo-TEM images (Figure 3A), 9%–33% of cubosomes displayed a distinct inner periodicity with widths of roughly 100 nm. Further, confocal images revealed that rhodamine B-labelled Annexin V-conjugated cubosomes and LM22A-4 loaded cubosomes reached the central optic nerve through the retina as demonstrated by red fluorescence, following intravitreal injection (Figure 3B). Therefore, the findings show that both Annexin V-conjugated cubosomes and LM22A-4-loaded cubosomes had a positive

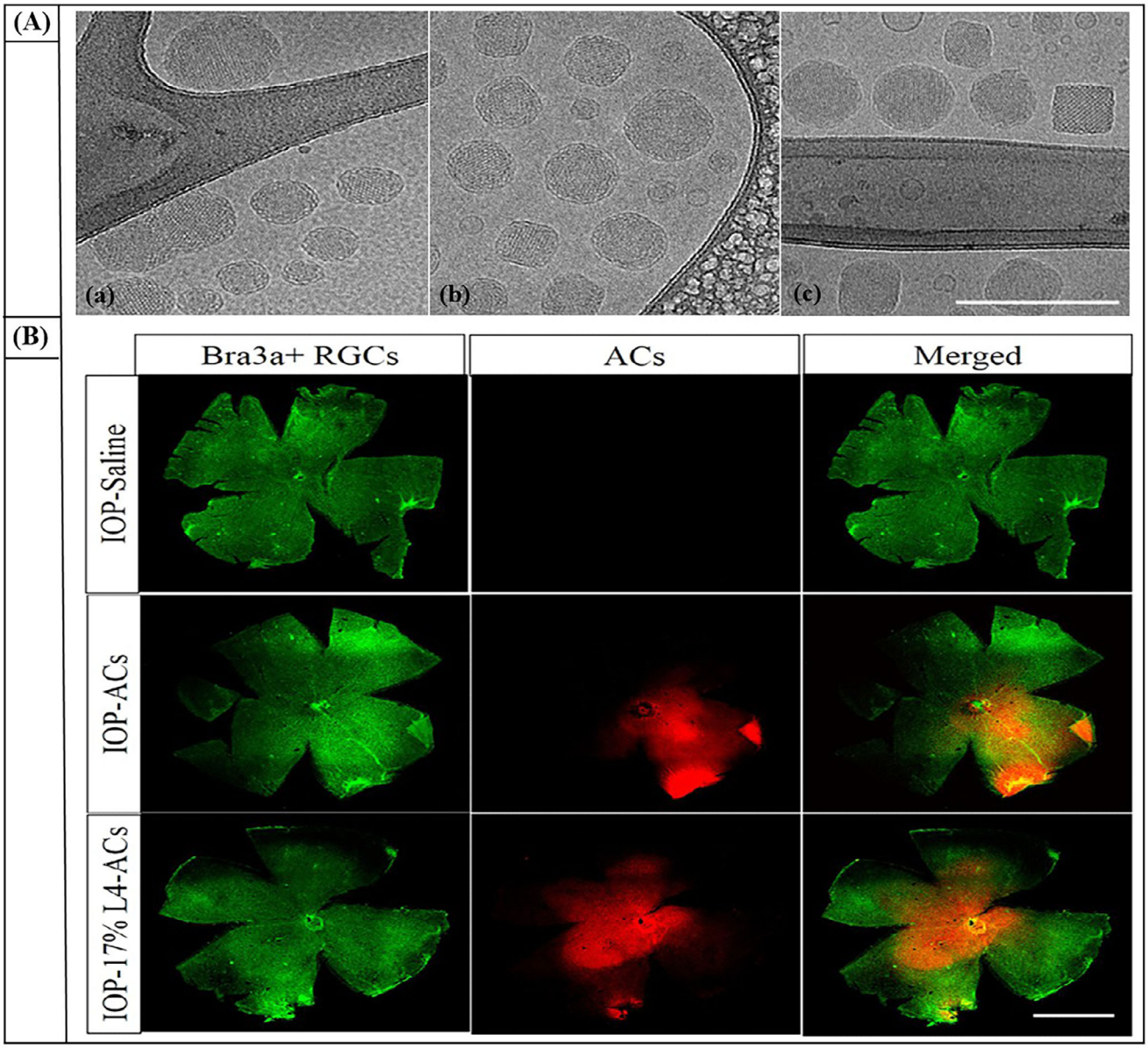

FIGURE 3 | **(A)** Cryo-TEM images of LM22A-4-loaded Annexin V Conjugated cubosomes at different drug loading percentages (a;9%, b;17% and c;33%) showing internal nanostructures, (B) Confocal microscopy images of the retina subjected to elevated intraocular pressure (IOP) injury, following intravitreal administration of rhodamine B-labelled Annexin V-conjugated cubosome (ACs)s and LM22A-4-loaded cubosomes (17% LA-ACs). Alexa Fluor 488-conjugated Bra3a+ (green) was used to label RGCs (IOP-treated retina). The red fluorescence indicated effective retinal penetration and targeted delivery to the central optic nerve region [103]. Copyright 2021, Acta Biomaterialia.

effect on delaying apoptosis in retinal ganglion cells. However, LM22A-4-loaded cubosomes had a greater therapeutic effect on preventing cell death [103].

6.5 | Brimonidine Tartrate

Brimonidine, categorized as an $\alpha 2$ adrenergic agonist, exhibits a dual mechanism of action in the reduction of intraocular pressure. It decreases the production of aqueous humor, as well as increases the outflow of uvo-scleral pathway through the increased synthesis of prostaglandins [104, 105]. Hence, the development of a sustained ocular delivery system became a significant priority for individuals with glaucoma. In a study conducted on cubosomes loaded with Brimonidine, a D-optimal design was used to predict the optimised formulation resulted in improved ocular bioavailability of Brimonidine along with prolonged intraocular pressure-lowering effect. The particle size exhibited a range of 347 nm to 250.3 nm, indicating that with an increase in the dispersed phase percentage from 5% to 10%, a notable increase was observed in the particle size. On the contrary, with an increase in concentration of surfactant from 0.5% to 2.5%, a reduction in particle size was observed. Further, a decrease in particle size was observed with an increase in the GMO: Poloxamer ratio from 2:1 to 6:1, and subsequently, an increase in the ratio from 6:1 to 10:1 resulted in increased particle size. Morphology obtained from transmission electron microscopy depicted cubic nanostructures of optimised cubo-

somes. Additionally, ex vivo corneal permeation demonstrated a 1.6-fold increase in comparison to Alphagan, whereas the results of in vivo pharmacodynamics studies displayed a considerable increase of 4.6 times in $AUC_{0\text{-}24}$ and 9.1 times in the mean residence duration in contrast to the commercially available formulation [45].

6.6 | Fenticonazole Nitrate

An imidazole antifungal belonging to the azole class, fenticonazole nitrate (FCN), has concentration-dependent pharmacological activity. At low concentrations, it inhibits the synthesis of ergosterol, and at high concentrations, it disrupts the fungal cytoplasmic cell membrane and acts as a fungicidal agent [106, 107]. Using the hot-emulsification process, FCN was encapsulated inside a cubosomal dispersion enhanced with limonene as a penetration enhancer in accordance with a full factorial design. The optimised cubosomal formulation demonstrated a high negative zeta potential of 34.25±0.64 mV, particle size of 249.75±1.48 nm, and encapsulation efficiency of 87.94±0.7% (Figure 4A). The ocular penetration of rhodamine band (Rhb)-Cubosomes in comparison to Rhb solution was assessed using confocal laser scanning microscopy (Figure 4B). The cubosomal formulation demonstrated better penetration into corneal layers, measuring 55 µm, as opposed to the solution's 25 µm. Histopathology studies also confirmed the results of in vivo permeation studies (Figure 4C). Microscopic assessment revealed intact tissue structures in the negative control and cubosome groups, with no evidence of inflammation, edema, or necrosis. The drug suspension group exhibited mild vacuolation in the ciliary body, whereas the positive control displayed significant inflammatory cell infiltration, vacuolation, and epithelial necrosis. Therefore, cubosomes are a promising vehicle for improving FCN's bioavailability and ocular administration [108].

6.7 | Tobramycin

Tobramycin sulphate, derived from cultures of *Streptomyces tenebrarius* is a hydrophilic cationic aminoglycoside used against a variety of bacterial infections, especially Gram-negative microbes such as strains of *Pseudomonas*. Like all other aminoglycosides, it is not suitable for oral administration due to its limited absorption via the digestive tract. Instead, it is administered through intramuscular, intravenous, or ocular route [109]. In the current study, cubosomes encapsulating tobramycin were formulated for increased permeation, ameliorated therapeutic effect, curtailed dosing frequency, enhanced stability, and prolonged residence time. From the corneal penetration studies, it can be noted that the value of the apparent permeation coefficient of the drug-loaded cubosomes was found to be 3.6 times higher than that of the marketed eye drops. Further, in vivo analysis indicated that the bacterial keratitis was reduced by day 3, followed by its alleviation by day 5 with tobramycin cubosomes in comparison to marketed eye drops. The severity of the disease was completely mitigated following 72 and 120 h. Pharmacokinetic profiling showcased the peak concentration and area under the curve of optimized cubosomes as 3.3 and 3.1-fold, respectively. Therefore, tobramycin encapsulated in cubosomes resulted in enhanced retention time and increased corneal permeability [76].

6.8 | Timolol Maleate (TM)

Timolol, a non-selective beta-blocker, is a primary medication used in the medical treatment of glaucoma [110]. Timolol decreases intraocular pressure (IOP) by inhibiting the production of aqueous humor through the blockade of sympathetic nerve terminals in the ciliary epithelium [111]. Topically instilled TM eye drops have limited corneal permeability following drainage through the nasolacrimal duct [112, 113]. In addition, TM eye drops necessitate frequent administration (twice daily), which results in poor compliance among patients. In order to address these drawbacks, Huang et al. developed timolol maleate cubosomes that were functionalized and stabilized using glycerol monooleate and pluronic F127, which indicated their ability to possess significant potential as an ocular drug delivery system for glaucoma. The formulated cubosomes exhibited a mean particle size of 142 nm and a PDI value of 0.107, indicating good homogeneity and a significant propensity for corneal transportation. Furthermore, the zeta potential was observed to be −6.27 mV. The cubosomes exhibited an encapsulation efficiency of 86.4 ± 4.6%, indicating that a significant proportion of the drug was successfully enclosed within the cubosomes, corresponding to the drug loading of 4.1 ± 0.5%. The mean duration of retention of commercial eyedrop in the aqueous humor was 43.5 min. However, the TM cubosome eye drops were quantifiable in concentration for a duration of 360 min, with an AUC of 123.9 µg/mL min, thus suggesting higher drug bioavailability. Further, the cubosomes exhibited a peak time of 30 min, surpassing that of commercially available eye drops. Presumably, GMO and Pluronic F127 encapsulated TM effectively inhibited the rapid release of TM and achieved a sustained release of it in the cornea. In addition, the assessment of the toxic effects on rabbit corneal cells and the evaluation of tissue structure indicated that the cubosomes exhibited biocompatibility and were devoid of any signs of toxicity [78].

6.9 | Gemifloxacin

Gemifloxacin (GEM), a fourth-generation fluoroquinolone, is a promising broad-spectrum antibiotic against both Gram-negative and Gram-positive bacteria. The suppression of bacterial DNA gyrase and topoisomerase II is the basis for its bactericidal action [114, 115]. Silver nanoparticles, nanoparticles, and chitosan nanoparticles are examples of advanced drug delivery systems that may offer improved bioavailability and increased drug corneal exposure. However, the primary drawback of these systems, according to earlier research, is their low antibacterial efficacy against Gram-positive bacteria compared to Gram-negative bacteria. The research conducted by El-Emam and colleagues investigated the potential of GEM-cubosomes loaded in in situ gel for the management of corneal lesions and inflammation. The optimized GEM-cubosome micrographs revealed tiny particles primarily in a cubic polyangular shape, along with small hexagonal vesicles (Figure 5A). Through in vivo studies, it was found that GEM-cubosomes decreased infection in rats as compared to GEM in situ gel. This was illustrated (Figure 5B) by the decrease in the area percentage of corneal opacity in the rats given GEM-cubosomes in situ gel, in contrast to the group that had GEM in situ gel treatment [116].

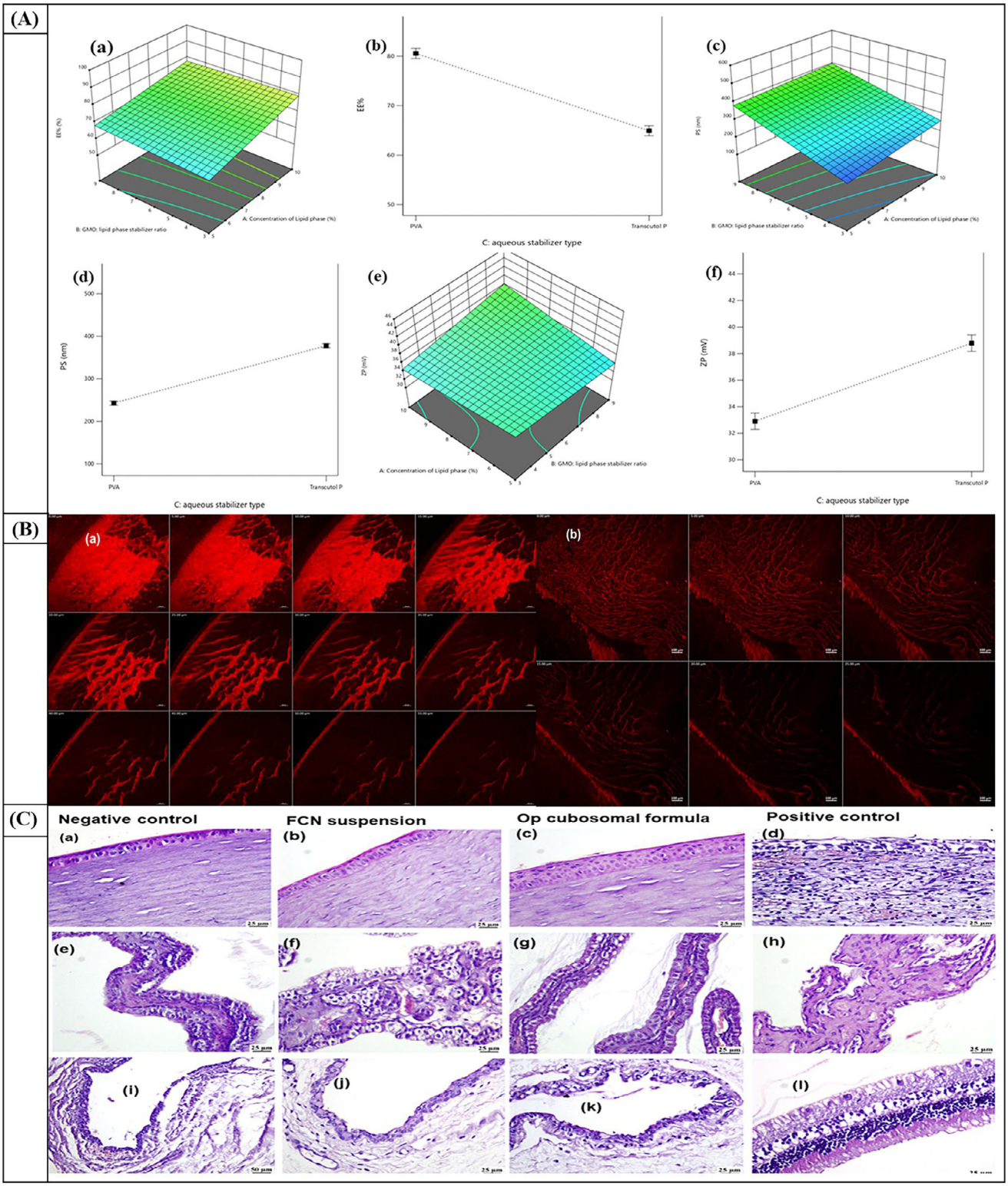

FIGURE 4 | (A) Response surface plots and line graphs illustrating the effect of formulation variables; Effect of lipid phase concentration and GMO: Poloxamer 188 ratio on encapsulation efficiency (a), particle size (c), Zeta potential (e); Effect of aqueous phase surfactant type on encapsulation efficiency (b), particle size (d), Zeta potential (f). (B) Confocal microscopy images comparing corneal penetration of rhodamine B-loaded cubosomes (a; penetration measured upto 55 μm) with rhodamine B solution (b; penetration measured upto 25 μm), illustrating a deeper fluorescence signal achieved through cubosomal delivery. (C) Histopathological analysis of rabbit eye tissues after topical application of various formulations showed that corneal sections (first row), the ciliary body (second row), and conjunctiva (third row) were examined across four groups; negative control with normal saline (a, e, i), drug suspension (b, f, j), optimized cubosomal formulation (c, g, k), and positive control with isopropyl alcohol (d, h, l) [108]. Copyright 2024, Journal of Drug Delivery Science and Technology.

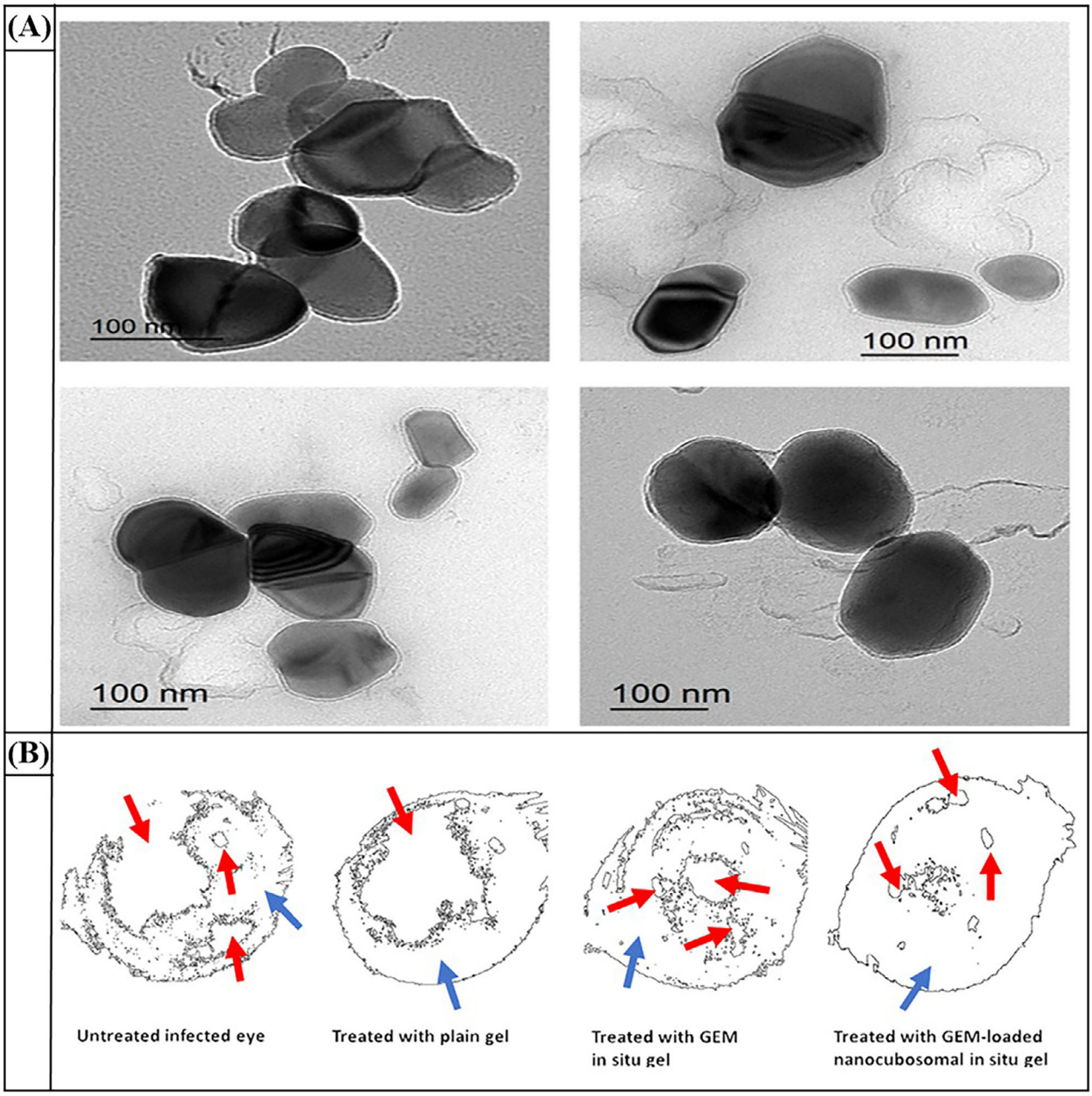

FIGURE 5 | (A) TEM images of GEM cubosomes depicting cubic shape along with small hexagonal vesicles, (B) Images showing MRSA-infected eyes in varied groups indicating the effect of different formulations on the area percentage of corneal opacity, which is a direct indicator of the severity of the infection and focal lesions [116]. Copyright 2023, Journal of Drug Delivery Science and Technology.

6.10 | Voriconazole

A second-generation hydrophobic antifungal drug, voriconazole (VCZ), is active against dimorphic and filamentous fungi with low minimum inhibitory concentration. Besides its efficacy in the prevention of diseases in comparison to other antifungals, its low solubility limits its application [117]. For instance, through VCZ loading in cubosomes, a controlled release was achieved along with increased bioavailability. Cubosomes prepared using GMO and PF127 were statistically optimized with face-centered central composite design to attain a composition with minimum particle size, maximum zeta potential, entrapment efficiency, and drug content with a sustained release profile. Small particle size is highly desirable as it offers decreased ocular discomfort along with enhancement of corneal permeation of VCZ; therefore, the particle size of the formulated VCZ cubosomes ranged between 109 and 243 nm. The PDI of the formulated cubosomes was reported between 0.12 and 0.40 for different formulations. Furthermore, ANOVA demonstrated that GMO and PF127 were crucial model terms, indicating an increase in drug entrapment with an increase in GMO and a decrease in PF127, resulting in EE ranging from 21.90% to 90.60%. Due to the dissolution of the drug, which was adsorbed at the surface, led to an initial burst release in the first 0.50 h, followed by sustained release for 24 h as water diffused slowly from the cubosomal channels [118].

6.11 | Fluconazole

Fluconazole, a broad-spectrum bis-triazole antifungal compound, is effective against several fungal species, including *Candida albicans* [119, 120]. It is reasonably formulated as a topical ocular drug, given its high penetration into the aqueous humor and low toxicity [121]. Though a topically instilled drug solution is effective in the treatment of *Candida Keratitis*, its usage is limited because of low bioavailability as a result of poor permeability of the drug through the cornea and rapid precorneal drainage [122, 123]. The research conducted by Nasr et al.al. investigated the potential of cubosomes encapsulating Fluconazole, which would result in greater penetration, improved bioavailability, and reduced ocular irritancy in the treatment of fungal keratitis. A full factorial design was employed to formulate fluconazole-loaded cubosomes. The optimized formulation displayed an average particle size of 48.17±0.65 nm with an encapsulation efficiency of 85.70±2.56%. Through ex vivo permeation studies, a 2-fold enhancement in permeation of fluconazole was observed in comparison to fluconazole aqueous solution. Further in vivo analysis indicated the safety and efficacy of fluconazole-loaded cubosomes [124].

6.12 | Ciprofloxacin

Ciprofloxacin, a quinolone antibiotic, treats ocular diseases such as conjunctivitis and corneal ulcers [125]. Current ciprofloxacin products encounter two main problems such as low solubility in lacrimal fluid and poor medication adherence due to high dose frequency, ultimately causing ocular irritation. Alharbi and Hosny developed ciprofloxacin cubosomes loaded in situ gel to prolong ocular retention time, improve eye permeation, and enhance antimicrobial activity. The optimization of cubosomes was carried out through Box-Behnken design, which resulted in cubosomes with a particle size of 121 nm, 75% of entrapment efficiency, and 2.54 times antimicrobial activity in comparison to commercial eye drops. Further, increased levels of ciprofloxacin were detected above the minimum inhibitory concentration (MIC) after 4 h, which lasted at 12 h within a range of 2–3.75 μg/ml. Thus, the data demonstrated that once-a-day administration can produce desired efficacy [88].

6.13 | Bromofenac

Bromofenac, an effective COX-2 inhibitor, reduces inflammation caused by eye dryness in people with dry eye syndrome. Shoman et al. formulated Bromofenac-loaded cubosomes via an emulsification method using a full factorial design. The optimized cubosome formulation had a particle size of 149.30 nm and 61.66% entrapment efficiency with a zeta potential of −30.80 mV. Further, the ex vivo permeation studies were conducted using goat cornea, which showed increased drug deposition and permeability parameters. Additionally, the in vivo studies revealed a mean residence time of 3.16 h and a higher $AUC_{0\text{-last}}$ of about 18.88 μg.h/ml in comparison to marketed eye drops [126]. Various other literature instances of ocular drug-loaded cubosomes are reported in Table 3.

7 | Preclinical Evaluation of Ocular Tolerability and Safety of Cubosome-Based Formulation

Preclinical models have extensively assessed the ocular tolerability, ocular safety, and cytotoxicity of cubosome-based drug delivery systems. For instance, Gaballa et al. evaluated the ocular safety of beclomethasone dipropionate-loaded cubosomes and cubo gel using both the bovine corneal opacity and permeability (BCOP) assay and histological examination. The BCOP scores for cubosomes and Cubo-gel formulations remained consistently below 0.5, thus suggesting minimal to no irritation potential. Histological examination of rabbit corneas showed intact epithelial and stromal layers, with no signs of swelling, tissue damage, or cell detachment, confirming ocular safety. Furthermore, the study assessed inflammatory markers in a rabbit model of endotoxin-induced uveitis. In comparison to control and Beclomethasone Dipropionate suspension groups, eyes treated with cubosomes showed significant reductions in leukocyte counts, nitric oxide levels, and prostaglandin E2, indicating not only safety but also anti-inflammatory effectiveness. These results were supported by histopathological examination of the iris and retina, which revealed decreased edema and inflammatory infiltration in the cubosome-treated groups [129].

Han and colleagues conducted a study to assess the ocular irritation potential of flurbiprofen-loaded cubosomes through the Draize test and histological examination in rabbits. The formulations showed excellent ocular tolerance, with irritation scores below 2, classifying them as non-irritant. Histological analysis confirmed that the corneal epithelium and stroma remained intact, with no evidence of inflammation or tissue damage, even at higher drug concentrations [127]. Shoman et al. evaluated the ocular safety of hyaluronan-enriched cubosomes loaded with bromfenac sodium through histopathological analysis in rabbit models. The optimized formulation caused no irritation or damage to the corneal tissues after multiple topical doses. Histological examination showed that the epithelium remained intact, the endothelial layers were preserved, and the stroma was well-organized, similar to the saline-treated control group. Therefore, these results demonstrated the cubosomal formulation's excellent ocular tolerability and biocompatibility, indicating its promise for safe topical ophthalmic application [126].

8 | Recent Patents on Cubosomes for Ocular Delivery

Many researchers have investigated the possible applications of cubosomes in various application. Recent patent applications have been filed as a result of the successful use of cubosomes in ocular delivery systems (Table 4**)**.

9 | Conclusion and Future Perspectives

This review article emphasizes the cubosomes' transformational potential in addressing several issues associated with ocular drug delivery. Cubosomes serve to be a very viable platform for both anterior and posterior ocular treatment techniques due to

TABLE 3 | Literature instances of drug-loaded cubosomes for ocular diseases.

S.no	Active ingredient	Disease	Excipients used	Method of preparation	Particle size	Drug entrapment efficiency	Zeta potential	Remarks	References
1	Flurbiprofen	Ocular Inflammation	GMO, Poloxamer 407, Glycerol	Hot High-pressure Homogenization	130-150 nm	>98%	−15mV	Low ocular irritancy, enhanced transcorneal permeation of flurbiprofen	[127]
2	Dexamethasone	Uveitis	GMO, Poloxamer 407	Emulsification technique	F1- 214.1±41.1 nm F2-226.3±55.6nm	>98%	—	Enhanced permeability coefficient of dexamethasone, Increased retention time, and ocular bioavailability	[77]
3	Tropicamide	Eye examination	Monoolein, Pluronic F127	Ultrasonic fragmentation, High-speed homogenization	54.52 nm	96.5%	−8.76mV	Faster onset of action and higher mydriatic action intensity	[128]
4	Beclomethasone Dipropionate	Uveitis	GMO, Poloxamer 407, Solulan C24	Top-down technique	Poloxamer 407- 104.6 to 263.2 nm Solulan C24- 121.7 to 278.5 nm	Poloxamer 407- (94.3%–98.1%) Solulan C24 – (94.2%–96.5%)	−16.3 to −43.7 mV	Enhanced transcorneal permeation, Controlled release	[129]
5	Latanoprost	Glaucoma	Phytantriol, Pluronic F127	Top-down technique	Unloaded- 215.3 nm Loaded- 207.9 to 217.8 nm	87.4%–94.0%	Unloaded- (−24.7 mV) Loaded- (−23.3 to 24.7 mV)	Sustained release, reduced potential side effects of Latanoprost, reduced dosage frequency	[80]
6	Moxifloxacin Hydrochloride	Conjunctivitis	GMO, P407	Top-down approach	306.5–714.2nm	69%–85%	−27.9 mV to −39.8mV	Sustained release, non-irritating	[130]
7	Natamycin	Fungal Disease	Phytantriol, PolyMulse	—	46–222 nm	51%–91%	—	Prolonged residence time, enhanced corneal permeation, reduced ocular irritancy	[131]
8	Gatifloxacin	Bacterial keratitis	GMO, P407	Emulsification technique	197.4nm	52.8%	−21.9 mV	High corneal permeation, a 4-fold reduction in the minimum inhibitory concentration of gatifloxacin	[132]
9	Tetrandrine	Cataract, Chronic keratitis, Glaucoma, Retinopathy	GMO, P407	—	170.0 nm	95.4%	29.3 mV	Increased retention time and transcorneal permeation, improved ocular bioavailability	[133]
10	Riboflavin	Keratoconus	GMO, P407	Emulsification and homogenization process	145 nm	46%	—	Improved ocular bioavailability and preocular retention	[134]
11	Pirfenidone	Ocular chemical burns	Monoolein, oleic acid, P407	Top-down approach	Stored at 4°C-247.3 nm Stored at 25°C- 257.5 nm	35.9%	Stored at 4°C- −33.60 mV Stored at 25°C-−46.00 mV	Sustained release of the drug, corneal re-epithelialization time after chemical burning was reduced	[135]

TABLE 4 | Recent patents on cubosomes for ocular delivery.

S.no	Patent number	Patent title	Year of grant	Highlights	Reference
1	CN106619573B	Timolol maleate cubic liquid crystal nano eyedrop and preparation method thereof	2019	Increases the corneal osmosis content of timolol maleate, reduces intraocular pressure, and prolongs the retention time	[136]
2	CN108309930A	A kind of Ofloxacin gel with liquid crystal structure nano eyedrop and preparation method thereof	2020	Prolongs residence time, improves bioavailability, is cost-effective, and easy to scale up	[137]
3	CN101502485A	Nanocubic liquid crystal dexamethasone preparation for the eye and preparation method thereof	Pending	Non-irritant administration improves ophthalmic bioavailability, high comfort level	[138]

their unique structural qualities, which include bicontinuous lipid bilayers, high bioadhesion, and the capability to encapsulate multiple kinds of drugs. Cubosomes outperform traditional methods in terms of corneal retention, drug's bioavailability, and sustained release, making them ideal for treating a wide range of ocular conditions. Looking ahead, the combination of stimuli-responsive mechanisms and optimization might move cubosome formulations toward tailored, on-demand drug delivery systems. Furthermore, moving these achievements from lab to clinical practice would require addressing challenges such as excipient safety, scalable production, and regulatory approvals. With continuing multidisciplinary cooperation and technical advancement, cubosomes have the potential to reinvent ocular therapies, providing patient-friendly, precise, and highly successful therapy options for vision-related illnesses.

Author Contributions

N.D. and H.N. collected the data and drafted the manuscript. T.G.S. discussed the concepts of the manuscript. P.K., C.P., and S.D. commented, read, and critically revised the work. All authors have read and approved the article. The authors confirm that no paper mill or artificial intelligence was used.

Conflicts of Interest

There are no conflicts of interest.

Data Availability Statement

All source data for this work (or generated in this study) are available upon reasonable request.

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Biographies

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