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Historical Perspective

Strategies to prolong the residence time of drug delivery systems on ocular surface

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ABSTRACT

Ocular diseases may be treated via different routes of administration, such as topical, intracameral, intravitreal, oral and parenteral. Among them the topical route is most accepted by patients, although it provides in many cases the lowest bioavailability. Indeed, when a topical formulation reaches the precorneal area, i.e., the drug absorption and/or action site, it is rapidly eliminated due to eye protection mechanisms such as blinking, basal and reflex tearing, and naso-lacrimal draining. To avoid this and to reduce the frequency of dosing, various strategies have been developed to prolong drug residence time after topical administration. These strategies include the use of viscosity increasing and mucoadhesive excipients as well as combinations thereof. From the drug delivery system point of view, liquid and semisolid formulations are preferred over solid formulations such as ocular inserts and contact lenses. Furthermore, liquid and semisolid formulations can contain nano- and microcarrier systems that contribute to a prolonged residence time. Within this review an overview about the different types of excipients and formulations as well as their performance in valid animal models and clinical trials is provided.

Contents

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1. Introduction

Quality of life is considerably affected by visual impairment caused by the advent of ocular diseases. Although there are potent drugs available to cure most of these diseases, the therapeutic efficacy of many of these drugs is limited due to a low bioavailability that is often less than 5%. Chronic ocular diseases, such as ageing related macular degeneration and diabetic retinopathy, require long term therapies. Many commercial products require frequent daily administration to maintain therapeutic drug concentrations, which may cause side effects such as endophthalmitis, increased intraocular pressure, hemorrhage, even detachment of retina if the administration has to be done via repeated injections [1,2].

As a relatively isolated organ with strong barriers the eye is from the drug delivery point of view a challenging target. Systemic administration of drugs into the eye is highly restricted by the blood-ocular barrier system formed by two main barriers: the blood-retinal barrier [3] and the blood-aqueous humor barrier [4]. Consequently ocular medications for treatment of in particular the cornea, conjunctiva, and anterior chamber are administered topically although cornea and conjunctiva are major epithelial barriers. Furthermore, drugs are rapidly eliminated via blinking, baseline and reflex lachrymation, and drainage from ocular surfaces even before they can penetrate these tissues in effective quantities. The major challenges in ocular drug delivery through topical administration are therefore poor permeability of mucosal tissues and short mucosal residence time. Among the various drug delivery strategies aiming to improve topical ocular bioavailability are delivery systems providing a prolonged residence time on cornea and conjunctiva. Due to such a prolonged drug residence time there is more time for drugs available to locally act on these mucosal surfaces or to penetrate into deeper ocular tissues reaching their target. Being aware of the great potential of drug delivery systems providing a prolonged residence time on ocular surfaces, formulation scientists are working since many decades on concepts addressing this issue.

First successful attempts to prolong ocular residence time date back to the 1980s and led to the development of mucoadhesive polymers providing adhesion to the ocular surface and reducing the draining rate by increasing the viscosity of eye drops [5,6]. First generation of mucoadhesive polymers were able to form just weak non-covalent bonds such as hydrogen bonds to the mucus gel layer covering ocular surfaces, whereas their second generation binds even covalently to mucus glycoproteins via the formation of disulfide bonds [7]. With the hype of nanomedicine since the late 1990s first nanocarrier systems remaining for a prolonged period of time on ocular surface were pioneered [8].

In parallel, it was shown that even comparatively very large dosage forms such as drug releasing ocular inserts and contact lenses adhere to the ocular surface for numerous hours up to days. Since then, the concept of increasing drug residence time on the precorneal area to increase its bioavailability after topical administration has been utilized for numerous drug delivery systems, resulting in a great variety of commercial products.

Within this review we provide an overview about all these different formulations and techniques to evaluate the potential of drug delivery systems to prolong ocular residence time. In addition, the concept behind each of these drug delivery systems is discussed in detail and an outlook on new developments is given.

2. Methods to evaluate ocular residence time

2.1. In vitro studies

As interactions of drug delivery systems with the mucus gel layer covering ocular surfaces are involved in all types of formulations providing a prolonged ocular residence time, most in vitro studies investigate the performance of excipients or formulations on or in mucus.

Generally there are numerous techniques for such evaluations available including rheological studies of polymeric excipients with mucus [9], tensile studies of polymers and formulations on freshly excised mucosal tissues [10,11], determination of size, zeta-potential, and turbidity changes of nano- and microcarriers caused by the interaction with mucins [12–14] as well as diffusivity measurements via single and multiple particle tracking [15]. As a comprehensive overview about these methods is already provided in other reviews [9,16]- [17], just ocular specific test methods that are of relevance for the evaluation of the ocular residence time of drug delivery systems are discussed herein.

Most ocular specific methods are simple “washability” tests of formulations, such as particles or gels, from ocular surfaces. These ocular surfaces are either monolayers of certain ocular epithelial cells or freshly excised ocular tissues from various species. Although mono- and multilayers formed by ocular epithelial cell lines are very similar to in vivo conditions, essential components such as a covering mucus gel layer and the lipid layer are missing. In fact, models of co-cultures with mucus-secreting cells have been proposed [18], but, to our knowledge, none has yet been used to estimate the ocular residence time of drug delivery systems. In most studies not the amount of delivery systems remaining on the ocular surface, but the amount of remaining drug, that is from the therapeutic point of view much more important, is quantified. Consequently, these results are a combination of at least two parameters: prolonged ocular residence time and sustained drug release. In cell-based models also cellular drug uptake has to be taken into account that does only to a minor extent correlate with the in vivo absorption behavior. A sound interpretation of the obtained data can therefore only be done when additional data about the drug release behavior of the specific formulation are available. Without this additional information the ocular residence time of the formulation cannot be properly determined. A highly adhesive formulation providing a strongly prolonged residence time, for instance, could not be recognized at all, when the incorporated drug is immediately released after application. The quantification of the remaining delivery system instead of the drug would allow focusing just on the ocular residence time of the vehicle. However, such approaches are from an analytical point of view challenging.

The likely simplest ocular specific test systems are based on ocular epithelial cell lines. For example, Hafner et al. have proposed a method whereby nanoparticles (NPs) containing melatonin are incubated with human corneal epithelium cell monolayers. The adhesion of NPs is assessed from the decrease of melatonin content in NP suspension following incubation. In reality, the sum of melatonin released by the NPs and that present in the NPs unbound to the cell monolayer are determined [19]. Li et al. have used multilayer corneal epithelial cells to simulate the corneal barrier and to study the drug residence time in precorneal area. For their purpose they used an apparatus simulating the processes of tear generation and elimination, like the one shown in Fig. 1. Every 10 min an aliquot of the tear fluid eliminated was withdrawn and analyzed for drug content [20].

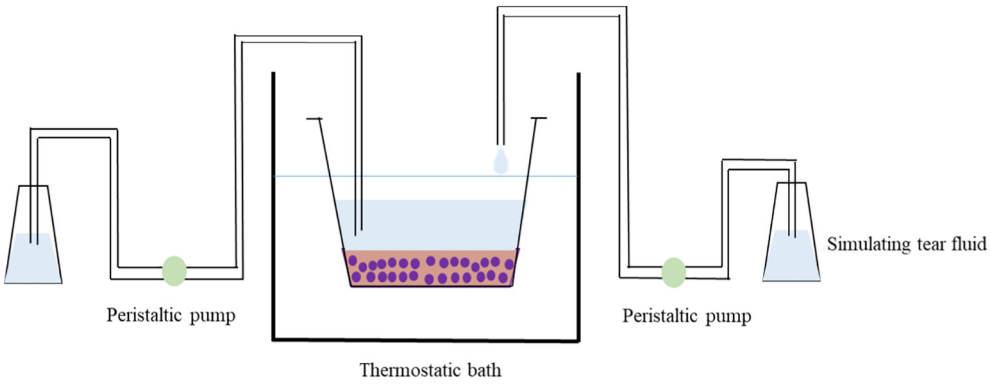

Fig. 1. Schematic diagram of the in vitro tear-turnover apparatus. Illustration adapted from [20].

In case of freshly excised ocular tissues an intact mucus gel layer is available that is essential for the evaluation of mucoadhesive delivery systems. One of the first ex vivo methods to determine drug corneal residence time was described by Bonferoni et al. [21]. The excised cornea was placed between the donor and acceptor compartments of an adequately modified Franz cell. Water was filled in the acceptor compartment with the sole function to thermostate the cornea. After placing the system under study on the corneal surface, the tissue was rinsed with simulated tear fluid. At pre-settled time points an aliquot of fluid eliminated was withdrawn and analyzed for drug content [21]. The above method was subsequently modified to obtain a model more predictive for drug precorneal residence time in vivo. For example, Chaiyasan et al. developed a method according to which "buttons" of excised cornea of 6 mm side were pasted on a glass slanting plane and subsequently incubated with the formulation. The drug fraction adherent to the mucosa was calculated as the percent difference between the drug content in the formulation incubated with the tissue and that in the formulation fallen from the slanting plane [22].

2.2. In vivo studies

The residence time of drugs and/or excipients in precorneal area is currently considered a measure of the mucoadhesivity of an ocular drug delivery system. The most used animal model is the New Zealand albino rabbit, since its eyes have several anatomic and physiologic similarities with the human eyes [23]. In vivo studies to evaluate the drug residence time in precorneal area have been carried out on the awaked rabbit, placed in a restraining box. After administration of the system under study, at pre-established intervals the tear fluid is withdrawn and analyzed for the drug [20]. The tear fluid is usually withdrawn from the lower eyelid rhyme by a one ml capillary. The concentration in tear fluid (C_{TF}) vs. time profiles are used to calculate the mean drug residence time in tear fluid (MRT) according to the relevant non-compartmental technique [24]. This parameter is calculated from the ratio between the area under moment curve, C_{TF} t vs. time (AUMC) and the area under curve C_{TF} vs. time (AUC) [25]. Tear fluid can also be withdrawn by a piece of blotting paper. The weight difference between wet and dry paper has been considered a measure of the tear fluid absorbed. The wet paper is then dried and the drug contained in it is extracted with methanol and quantified by HPLC [26]. Techniques different from that described above have also been used to determine the time of drug disappearance from precorneal area. The rabbit ocular surface, for instance, has been analyzed by gamma scintigraphy after administering formulations having been labelled with technetium (Tc-99) [27]. The anatomy of the rabbit eye, however, differs from that of the human eye in the following properties: presence of nictitating membrane (third eyelid); larger conjunctival sacs; thinner corneas and a relatively larger ocular surface. Such differences can result in longer retention times of the drug delivery system on the ocular surface of rabbits [28]. Also, it is essential to note that the human blink frequency is around 8–21 blinks per minute, while in rabbits it is only around 2–3 per minute. Two different types of fluorophores were applied in the rabbit and human eyes, and the relevant difference in fluorescence of precorneal tear film was assessed [29]. The resulting AUC of the fluorophores concentration vs time graph was 3 times greater in rabbits than in humans. As a consequence, the transcorneal permeation of a drug can be overestimated. However, according to these researchers, the drug partition coefficient between corneal epithelium and tear fluid should be considered. In case of a high drug partition coefficient, the differences in drug residence time in the precorneal area would become irrelevant [29], also because it has been shown that in these cases the drug absorption takes place during the first 2–3 min from instillation [30]. Although a very rapid partition might decrease the effect of loss through blinking, the around 10-fold higher blink frequency of humans strongly limits the validity of ocular residence time studies in rabbits.

Apart from rabbits, some ocular residence time studies are also performed in other animal models such as mice and rats. Pai and coworkers, for instance, assessed the ocular mucoadhesion on Sprague Dawley rat eyes by means of confocal microscopy. After 30 min from the administration of fluorescent dispersions, rats were sacrificed, and the eyes were surgically enucleated. Sagittal sections 5 μm thick were taken and observed by the confocal microscope to evaluate the fluorescence intensity [15].

2.3. Clinical studies

In order to evaluate the residence time and pharmacokinetic of drugs on the ocular surface tear fluid can be collected at predetermined time points after administration using for instance Schirmer test strips placed over the junction of the temporal and medial margins of the lower third of the eyelid followed by HPLC or LC-MS analyses [31] [32]. This approach provides valuable information about the behavior of the drug on the ocular surface. As it does not provide information about the residence time of the vehicle, however, this method is of just limited value for the development of new ocular delivery systems providing a prolonged ocular residence time. Scintigraphic analyses quantifying the ocular residence time of isotopes such as technetium 99 m chelated with diethylenetriaminepentaacetic acid (DTPA) instead of a drug [33] provide additional information about the distribution of the isotope over the ocular surface over time but do not provide valid information about the residence time of the vehicle as well. Studies with low molecular mass fluorescence markers such as sodium fluorescein using clinical fluorometers offer the advantage of a simplified analytic [34]. As these low molecular mass markers can penetrate into the tissue in the same way as drugs it is in many cases impossible to discriminate

between the fluorescence marker remaining on the ocular surface because of the vehicle and that remaining there because of a penetration into the tissue. Using non-penetrating fluorescent probes such as FITC-dextrans of high molecular mass above 70 kDa avoids this problem [35] [36]. Moreover, such high molecular mass tracers are to a much higher extent anchored in particular in polymeric vehicles as they can form numerous hydrogen bonds with viscosity increasing and mucoadhesive polymers. In comparison to all other markers described above FITC-dextrans provide therefore likely most accurate data about the fate of polymeric vehicles on the ocular surface. More recently optical coherence tomography (OCT) was introduced as a fast infrared-light-based imaging modality for ocular residence time studies in real time. Napoli et al., for instance, evaluated the adhesive properties of a 0.5% sodium carboxymethylcellulose solution on the ocular surface by analyzing the dynamic behavior of the precorneal tear film after instillation. The polymer solution was detected by OCT as a two-layered structure localized onto the epithelial surface of the cornea. The ocular residence time and thickness of the two-layered structure onto the epithelial surface of the cornea was considered an index of the adhesive properties of this formulation [37]. In another study the central tear film thickness was determined by OCT over time following instillation of eye drops and an eye gel [38]. In contrast to all other methods described above OCT enables the direct analysis of the adhesive properties of polymeric excipients on the ocular surface.

3. Excipients

3.1. Viscosity increasing polymers

One of the most applied approaches to prolong ocular drug residence time is to increase the viscosity of the vehicle. This increase in viscosity is achieved with synthetic polymers such as polyacrylates and polyvinylalcohols, natural polymers such as hyaluronic acid and alginate and derivatives of natural polymers such as cellulose derivatives. Ludwig and Ooteghem, for instance, investigated the impact of viscosity on the ocular residence time of sodium fluorescein in humans demonstrating that due to the addition of hydroxyethylcellulose increasing the viscosity up to 25 mPa s the ocular residence of this marker is substantially prolonged. Results of this study are illustrated in Fig. 2.

Eye drops of high viscosity, however, may increase reflex tearing, thus causing a faster drug removal from the surface. Furthermore,

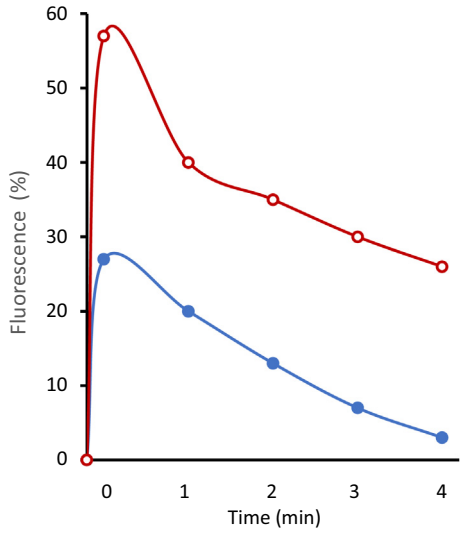

Fig. 2. Fluorescence decay curves of phosphate buffer only (•) and 1.7% hydroxyethylcellulose solution exhibiting a viscosity of 25 mPa s (o) both containing 0.05% sodium fluorescein in humans; adopted from Ludwig and Ooteghem [34].

high viscous eye drops are irritating for many patients, do not always allow a reproducible drug dosing and, in addition, cause a blurred vision following administration. An optimal viscosity range of 15–55 mPa is therefore recommended [39] providing on the one hand a prolonged residence time and on the other hand avoiding side effects as described above. Alternatively, in situ gelling polymers that can be instilled in form of lone viscous eye drops in accurate and reproducible doses undergoing an immediate gelation once in contact with the precorneal environment can be used. Indeed, such systems are well tolerated on administration, and can give rise to a sol-gel transition if subjected to specific stimuli [40].

Generally in situ gelling polymers can be divided into three categories, based on the phase transition properties: temperature sensitive (i), pH sensitive (ii) and ionic strength sensitive (iii) [41] [42]. Thermosensitive polymers include block copolymers and poloxamers; pH sensitive polymers include phthalates, cellulose acetates and acrylic acid polymers; the more used polymers sensitive to ionic strength are sodium alginate and gellan gum [43].

Thermosensitive polymers are the most studied for administration. A thermogelling polymer to be acceptable must have a gelling temperature in the range of 32–36 °C, so that they are liquid at room temperature undergoing an instantaneous transition to gel on the ocular surface [43]. Such polymers are both synthetic and natural polymers. Among the synthetic ones the most frequently used and commercially available polymer is Poloxamer 407 [44]. It consists of polyethylene oxide (PEO) and polypropylene oxide (PPO) units with general formula PEOx–PPOy–PEOx [45]. PEO has prevailing hydrophilic properties, PPO hydrophobic. These, once combined, result in amphiphilic characteristics which can vehicle either hydrophilic or hydrophobic active principles [46]. PPO tends to reduce the gelling temperature while PEO tends to increase it. Hence, polymers with different transition temperatures can be obtained by modulating the PEO:PPO ratio [47].

In order to improve drug residence time in the precorneal area, the combination of in situ gelling and mucoadhesive polymers seems advantageous providing both high cohesive and mucoadhesive properties [40] such as poloxamer/chitosan [48] and poloxamer/polycarbophil [49], poloxamer/carboxymethylcellulose [50] poloxamer/polyacrylate [51], poloxamer/gellan gum/polyacrylate [52] or poloxamer/hydroxypropylmethycellulose [53]. As an example for commercially available eye drops containing such a combination, the DuraSite® platform seems appropriate [54]. It exhibits a temperature-dependent sol-gel transition because of poloxamers and high mucoadhesive properties because of a polyacrylate. More recently, a second version of this system (DuraSite 2®) has been developed. This latest version of eye drops to which a positively charged polymer was added guaranteed an even longer residence time of the drug in the precorneal area. These platforms were used for azithromycin eye drops (AzaSite®) and besifloxacin eye drops (Besivance®). More recently, bromfenac eye drops for pain and inflammation associated with eye surgery have also been released. They demonstrated significantly greater anti-inflammatory activity in the rabbit eye than other traditional eye drops containing the same drug [55].

As an example for a pH-triggered in situ gelling systems the pilocarpine hydrochloride containing ophthalmic gel Pilopine HS® being based on the crosslinked polyacrylic acid Carbopol 940 should be mentioned [56].

Polymers sensitive to the ionic strength show a sol-gel transition due to interactions between anionic polymer substructures and cations, in particular Ca^{2+}, present in tear fluid [57]. The advantage of these systems consists in a temperature and pH independent gelation. Among such polymers in particular gellan gum is of high relevance [58,59]. It is an anionic hetero-polysaccharide able to interact with the cations found in tear fluid (Na^{+}, Ca^{2+}, Mg^{3+}, etc.). Formulations containing levofloxacin and this polymer were well tolerated and showed more than a 3.5 fold improvement of the $AUC_{0\text{-}24}$ and a 3-fold improvement of the mean residence time compared to the commercial solution

Levotop PF® [60]. Among other examples of products currently on the market are Timoptic XE® eye drops. Containing deacetylated gellan gum as in situ gelling polymer for a prolonged ocular residence time these eye drops show a 2-fold higher efficacy than a simple timolol solution [61,62]. A similar system for the ocular delivery of timolol has been marketed under the brand name Timoptol-LA® [56].

3.2. Mucoadhesive polymers

The mucus gel layer protecting the ocular surface consists of mucins, a family of at least 20 O-glycosyl proteins of anionic character. On conjunctival and corneal surfaces mucins form a layer called glycocalyx [63]. Excipients providing adhesion to this mucus gel layer exhibit a prolonged ocular residence time [64]. High mucoadhesive properties are provided by the following structural characteristics: strong positive or negative charges favoring ionic interactions (i); groups able to form strong hydrogen bonds such as carboxyls, hydroxyls, amino and sulfate groups (ii); high molecular mass and chain flexibility favoring the interpenetration of mucoadhesive polymers into the mucus gel layer followed by chain entanglements (iii); surface energy conducive to spread on mucus (iv) [65]. Polymeric excipients fulfilling these characteristics may be classified according to their charges into: cationic, anionic, nonionic and amphoteric [65,66].

A frequently used strategy is the incorporation of synthetic or natural mucoadhesive polymers in formulations resulting in liquid gels that can be applied in form of eye drops. In recent years attention has been paid to polymers of natural origin as they are biodegradable and biocompatible, in addition to viscosity increasing and mucoadhesive characteristics [67]. Mucoadhesive polymers have to adhere on the one hand on the ocular mucus gel layer and have on the other hand to release the drug being attached to the polymer chains in a sustained manner as illustrated in Fig. 3.

If the drug is not bound to the mucoadhesive polymer at all, drug ocular residence time will not be prolonged. In Table 1 an overview about mucoadhesive excipients and their binding mechanisms to mucus and to drugs is provided.

Numerous formulations based on mucoadhesive polymers of polysaccharide origin are commonly commercialized as artificial tears to treat the dry eye syndrome. They often contain xyloglucan (the polysaccharide from tamarind seeds (TPS)), hyaluronic acid (HA), cellulose derivatives, guar and xanthan gums. Clinical studies carried out with a commercial product based on HA and TSP, for instance, have shown the ability to reduce ocular surface damage and symptoms of discomfort in patients with dry eye syndrome [90]. These results can be attributed to a synergistic effect between the two polymers present in the eye drops. It was found that the mucoadhesion of the TSP/HA mixture (3:2) was stronger than that of each of the unmixed polymers or of the TSP/HA mixtures of different composition, thus ensuring a prolonged residence time of the lubricant in the precorneal area [91]. In addition, TSP is supposed to increase tear fluid stability, thus increasing drug mean residence time [28].

Methylcellulose was the first cellulose polymer introduced over 60 years ago. Subsequently numerous cellulose-ethers have been employed for eye drops as viscosity-enhancing vehicles, as well as for their wetting properties able to increase the contact time due to their film forming properties [64]. Cellulose derivatives have widely been used in ophthalmic preparations, such as eye drops, contact lenses, and artificial tears. They are endowed with high thickening properties, good ocular tolerance, compatibility with most drugs, stability of tear film and stability to autoclave sterilization. Among nonionic cellulose derivatives hydroxyethylcellulose has been the most used mucoadhesive excipient.

HA is a glycosaminoglycan found in numerous tissues as a major component of the extracellular matrix. As it can bind water molecules by hydrogen bonding and it can stabilize the tear film thereby reducing the winking reflex. By the way a severe limitation with ophthalmic formulations based on HA consists in a short ocular residence time of less than 10 min even if molecular mass has an important impact on mucoadhesiveness. It has in fact been found that the greater the molecular mass, the greater the mucoadhesiveness [92]. Salzillo et al. evaluated six commercial HA based formulations, comparing them with newly developed HA-based eye drops. The products analyzed were: Bluyal®, Blugel®, Hyabak®, Artelac Splash MDSC®, Hyalistil Bio®, and Octilia Natural®. Between these formulations Bluegel® had the most optimized drainage based on its zero-shear viscosity (24.2 mPas); therefore, this clinically accepted viscosity was chosen as the target value for the following optimization studies [92].

Chitosan is a biocompatible, non-toxic, biodegradable polymer, obtained by deacetylation of chitin. Chitosan has been widely employed in pharmaceutical technology by virtue of its polycationic nature. As it contains reactive amino groups, able to interact with cornea and conjunctiva, it is of great potential as excipient for ophthalmic vehicles. Unfortunately, non-modified chitosan is insoluble at physiologic pH, more precisely, at pH > 5, and this drastically limits its topic ocular application [75]. Therefore numerous studies have been carried out on chitosan derivatives soluble at physiologic pH such as *N*-carboxymethylchitosan [93] and quaternary ammonium-chitosans like *N*-trimethylchitosan (TMC) [77] or N,O-[*N*,*N*-diethylaminomethyl(diethyldimethylene ammonium)$_n$]methyl chitosan [76].

Primary amino groups left free from quaternization were subsequently thiolated to obtain a multifunctional chitosan derivative containing quaternary ammonium and thiol groups [78]. The synergism of quaternary ammonium and thiol groups has led to increased pre-corneal residence time of drugs [78].

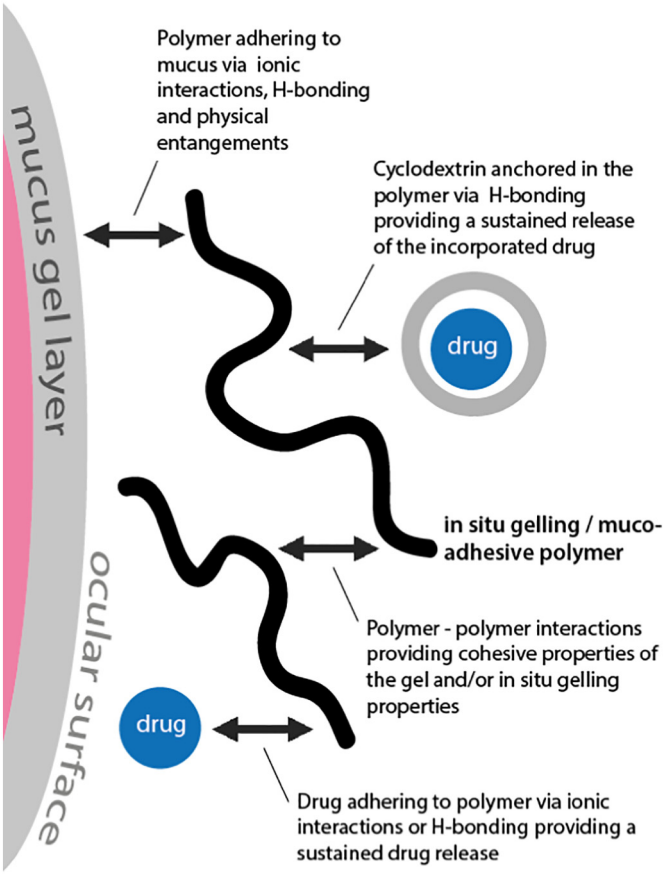

Fig. 3. Illustration of interactions between in situ gelling/mucoadhesive polymers with the mucus gel layer and with the drug; ideally these interactions provide a prolonged ocular residence time as long as the drug is released from the polymer.

Table 1
Overview about mucoadhesive excipients used for ocular drug delivery.

Mucoadhesive excipients	Type of adhesion to mucus	Type of drug binding	Mucoadhesive properties	References
In situ gelling polymer and mucoadhesive polymers				
Polaxamer complex with mucoadhesive polymers	Ionic interactions; hydrogen bonding; chain entanglements	Hydrogen bonding; amphiphilic interaction	++	[48–50,52]
Gelatin-chitosan complex	Ionic interactions; hydrogen bonding; chain entanglements	Intermolecular crosslinks; ionic interactions; hydrogen bonding	++	[68]
Hexanoylglycol-chitosan	Ionic interactions; hydrogen bonding; chain entanglements	Intermolecular crosslinks; ionic interactions; hydrogen bonding	++	[69]
Chitosan-thioethylamide	Disulfide bond formation; ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+++	[70,71]
Gellan gum and derivatives	Cationic interactions; hydrogen bonding	Ionic interactions; hydrogen bonding	+	[58,59,62,72]
Mucoadhesive polymers				
Tamarind seed polysaccharide	Ionic interactions; hydrogen bonding	Ionic interactions; hydrogen bonding	+	[25]
Xanthan and guar gums	Ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+	[39]
Cellulose derivatives	Ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+	[39]
Sialic acid or collagen type 1 functionalized hyaluronic acid	Ionic interactions; hydrogen bonding	Ionic interactions; hydrogen bonding	++	[73]
Hyaluronic acid	Ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+	[74]
Chitosan	Ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	++	[75]
Quaternary ammonium-chitosan	Ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	++	[76,77]
Quaternary ammonium thiolated chitosan	Disulfide bond formation; ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+++	[78]
Polyvinylpyrrolidone	Ionic interactions.	Hydrophobic interactions	+	[79]
Carbosilane dendrimer	Ionic interactions; hydrogen bonding	Hydrophobic interactions	+++	[80]
Thiomers	Disulfide bond formation; ionic interactions; hydrogen bonding; chain entanglements	Ionic interactions; hydrogen bonding	+++	[81]
Cyclodextrins				
Cyclodextrin with mucoadhesive polymers	Ionic interactions; hydrogen bonding	Incorporation; hydrophobic interactions	+	[82–84]
Quaternary ammonium-chitosan grafted with cyclodextrins	Ionic interactions; hydrogen bonding; chain entanglements	Incorporation; hydrophobic interactions	++	[85]
Thiolated cyclodextrins	Disulfide bond formation	Incorporation, hydrophobic interactions	+++	[86–89]

Bravo-Osuna et al. described the synthesis of a new water-soluble dendrimer having quaternary ammonium groups, the carbolsilane, to be used as an excipient in eye drops medicated with acetazolamide (ACZ) [80]. The dendrimer, when added to the eye drops administered to the rabbit eyes produced a more rapid and extended hypotensive effect, leading to a significant efficacy increase with respect to the eye drops containing the sole ACZ. Such an increase was ascribed to the dendrimer ability to interact with the transmembrane mucins present on ocular surface and its surfactant properties [80].

Thiolated polymers-designated thiomers- are an innovative class of mucoadhesive polymers able to form disulfide bonds with cysteine residues of mucins. These interactions are stronger than Van der Waals, hydrogen bridges or dipole-dipole interactions that other polymers can form with the mucosa [94]. However, thiomers undergo in aqueous environment at pH >5 oxidative degradation limiting their topical ocular application. Hintzen et al. developed a method to synthesize a thiolated mucoadhesive polymer, stable also at pH values higher than 5. It is a thiomer of pectin fully S-protected with mercaptonicotinic acid with increased stability and mucoadhesive and cohesive properties [95]. When the same S-protection method was used with a thiomer from HA promising results were obtained. In particular, the polymer showed a prolonged stability of up to 4 weeks under oxidative stress at pH 7.4. The mucoadhesive properties of HA thiolated S-protected were found to be 52 times greater than those of non thiolated HA [81].

Using microPET technology the ocular residence time and biodistribution of a topically applied thiolated polymer - chitosan-*N*-acetylcysteine conjugate (chitosan-NAC) - was evaluated in rabbits and compared with that based on unmodified chitosan [96]. As shown in Fig. 4 after application of 0.3% and 0.5% chitosan-NAC formulations, the radioactivity distribution detected on the ocular surface during the entire imaging period (up to 22 h) was homogeneous, whereas after application of the 0.5% chitosan-NAC formulation highest activity levels were detected in the inner canthus. This finding was interpreted as a proof of a better performance of the chitosan-NAC formulation compared to unmodified chitosan, likely due to a higher mucoadhesiveness of the former. These results together with a high safety profile of thiolated polymers [7] led to the development of chitosan-NAC-based eye drops called Lacrimera® [97]. In particular, based on corneal staining data, it was found that the percentage of patients with intact corneas increased from 12 to 64% after one month of treatment with Lacrimera® [98].

3.3. Cyclodextrins

A large fraction (90%) of actives approved since 1995 have poor solubility. Among the agents able to increase drug solubility, cyclodextrins have been in focus of numerous research works for decades. Indeed, they solubilize sparingly soluble actives, due to the formation of inclusion complexes [99]. Cyclodextrins are cyclic glycopyranose oligosaccharides and their ability to increase drug bioavailability or solubility, intensity or duration of their therapeutic activity, permeability properties and physico-chemical stability and to reduce their toxicity/irritation on tissues has widely been demonstrated by literature reports. As native cyclodextrins are unable to prolong drug residence in precorneal area, their possible enhancing effect on the efficacy of ocular systems containing them should mainly be ascribed to the solubility increase of hydrophobic drugs in tear fluid. In addition, Morrison et al. demonstrated that

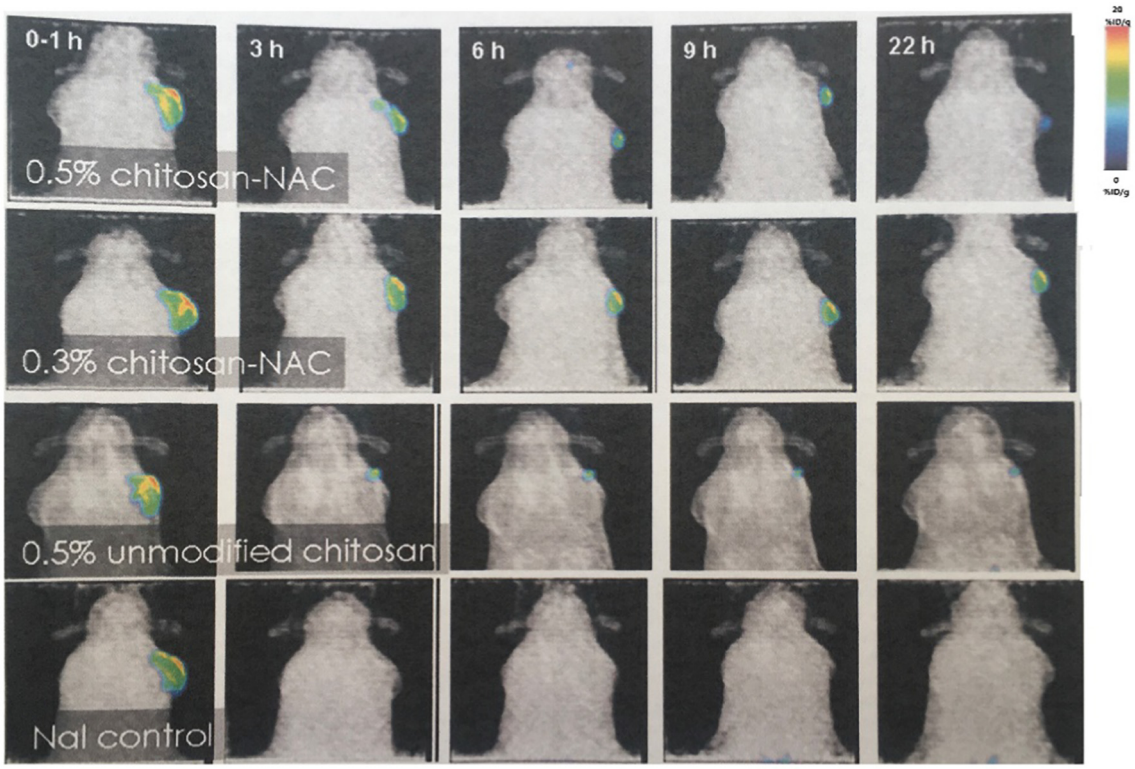

Fig. 4. MicroPET images of rabbit eyes after administration of ^{124}I-labelled 0.5% and 0.3% chitosan-*N*-acetylcysteine (chitosan-NAC), 0.5% unmodified chitosan and NaI (control) solutions. Imagine taken from [96].

β-cyclodexstrin enhances the corneal permeability. This effect, however, was shown to be caused by the extraction of cholesterol from corneal cells, thus making them more permeable [100].

In order to prolong the precorneal residence time of drugs complexed by cyclodextrins mucoadhesive polymers were used as vehicle for liquid and semisolid formulation as illustrated in Fig. 3 [82,84]. Others have used mucoadhesive polymers to prepare films containing the drug complexed by cyclodextrins [101,102].

Via their hydroxyl groups cyclodextrins can be covalently attached to mucoadhesive polymers [85,103]. A quaternary ammonium chitosan derivative, for instance, was conjugated with methyl-β-cyclodextrin. The cyclodextrin is able to solubilize highly hydrophobic drugs while the quaternized chitosan provides high mucoadhesion through electrostatic interactions with mucins [103]. Budai-Szucs et al. synthesized a cyclodextrin-thiolated-poly-aspartic-acid conjugate for ocular administration of the anti-inflammatory drug prednisolone [104]. Compared to the physical mixture of cyclodextrin and polymer the conjugate showed a more sustained prednisolone release [104]. Another example is sulphobuthyether-β-cyclodextrin (SBE-β-CD) [105,106]. The inclusion complex of chloramphenicol with SBE-β-CD, for instance, prolonged the drug ocular residence time and provided a significant increase of *endo*-ocular bioavailability [106].

Recently the substitution of –OH with thiol groups has led to a new class of cyclodextrins, —namely thiolated cyclodextrins- with pronounced mucoadhesive properties due to their ability to form disulfide bonds with mucus glycoproteins [87,88]. The thiolation of CD can improve their potential, bestowing mucoadhesive properties on them, while maintaining their drug solubilizing properties and ability to mask drug-related irritations.

Ijaz et al. pioneered thiolated CDs by the use of α-cyclodextrin conjugated with cysteamine (α-CD-Cys) for the administration of the sparingly soluble drug cetirizine. α-CD-Cys showed a mucoadhesivity 52 times higher than that of α-CD, and the ability to mask irritations caused by cetirizine. Moreover the conjugate was completely invisible [89]. This preliminary study has been the premise of a more recent one in which 6 hydroxyls of α-CD were replaced with thiol groups, thus yielding an α-CD having a higher substitution degree with thiol groups. This showed promise for the mucosal administration of BCS class IV drugs [86].

Another approach is focusing on the encapsulation of cycodextrin in nanosystems. A representative study was carried out by Jóhannsdóttir et al. [107]. They modulated the ratio between α-CD-cyclosporine A and γ CD, used as aggregating agent, and succeeded in preparing NPs medicated with an immunosuppressive drug for the treatment of dry eye syndrome [107]. On their part Wang et al. proposed the preparation of nanoliposomes encapsulating a brinzolamide-hydroxypropyl-β-cyclodextrin complex. The nanoliposomes had a size of 86 nm and a brinzolamide entrapment efficiency higher than 90%. Such nanosystems were able to reduce the *endo*-ocular pressure in rabbits significantly in comparison to commercial drug suspensions [108].

4. Drug delivery systems

4.1. Liquid and semisolid formulation

Eye drops are currently the liquid formulation of choice for the topical treatment of ocular diseases, mainly due to their fair compliance, although bioavailability is low. Indeed binkling, basal and reflex tearing as well as nasolacrimal drainage concur to shorten drug ocular residence time, and as a result frequent administrations are required [109]. The need of increasing instillation frequency is usually associated not only with a decrease in patient compliance but also with substantial side effects if an excess of drug is drained via the nasolacrimal duct. In order to improve this dosage form, viscosity increasing and mucoadhesive polymers as described above are used.

As highly viscous aqueous solutions can lead to an improvement in drug ocular residence time, semisolid formulations are advantageous. Ointments and gels such as the dibrompropamidine eye ointment

Goldeneye® or the dextran-hypromellose containing gel Lubri-Tears® are therefore commonly used for topical drug delivery. However, due to their high viscosity causing a blurred vision, they are mostly limited to night use. Recent reviews are available on this topic [110,111].

4.1.1. Polymeric nanoparticles

Both liquid and semisolid formulation can contain nano- and microcarrier systems that can provide per se a prolonged ocular residence time by their entrapment in the mucus gel layer and unspecific interactions of their surface with mucus glycoproteins. According to recent studies, the tear film is extremely thin, varying from a minimum of 2 μm to a maximum of 5.5 μm on the corneal region [112]. Traditionally, the tear film is described by the three-layer model proposed by Wolff [113] where the mucus layer is the innermost layer less than 500 nm thick [112]. The mucus layer is responsible for the mucoadhesiveness of ocular systems and, unlike the mucus layer that lines the intestinal epithelium, it can hardly be considered as a diffusion a barrier for nanoparticles, since these nanocarriers exhibiting a mean size between 10 and 100 nm can even exceed the thickness of the mucus layer as shown in Fig. 5.

Different biodegradable polymers have been extensively used for the design of nanoparticles. Depending on the characteristics and properties of the polymer and drug used NPs are prepared by different techniques, such as coacervation, ionotropic gelation and emulsification followed by solvent removal [114]. Since drugs can be incorporated in the polymeric network their sustained release can be provided. Furthermore, they are well accepted by patients as most of them do not cause any blurred vision.

If NPs neither are entrapped nor interact with the mucus layer, however, they are eliminated from the precorneal area at exactly the same rate as conventional eye drops [114]. Therefore, NPs based on mucoadhesive polymers, have been studied to prolong their residence time. Among mucoadhesive polymers used for this purpose are poly(3-hydroxybutyrate-*co*-3-hydroxyvalerate) [115], galactomannan derivatives [116] and poly(D,L-lactide), dextran functionalized with phenylboronic acid (PLA-b-Dex-PBA) [117] and chitosan [118]. In Fig. 6 a schematic illustration of the self-assembly process to form cyclosporine A loaded PLA-b-Dex-g-PBA nanoparticles is shown. In particular, while the commercial cyclosporine A formulation Restasis®, if administered three times daily has shown just a reduction of inflammatory processes after a week, a single administration of a PLA-b-Dex-PBA-NPs formulation has eliminated the inflammatory processes and restored the full health of the eyes in a week's time [117].

NPs based on chitosan and lecithin were found to be a sound tool to prolong the ocular residence of natamycin, an antifungal contained in the commercial product Natamet® [119]. Lecithin is a natural phospholipid mix which, being lipophilic, can increase the entrapment efficiency of nanosystems and sustain the release of entrapped lipophilic drugs. In particular, in vivo on rabbit eyes mucoadhesive NPs increased 1.47 times the AUC of the drug concentration in tear fluid vs. time profile in comparison to the commercially available suspension. This allowed a reduction of the instillation frequency from 6 to 3 times in 10 h [119].

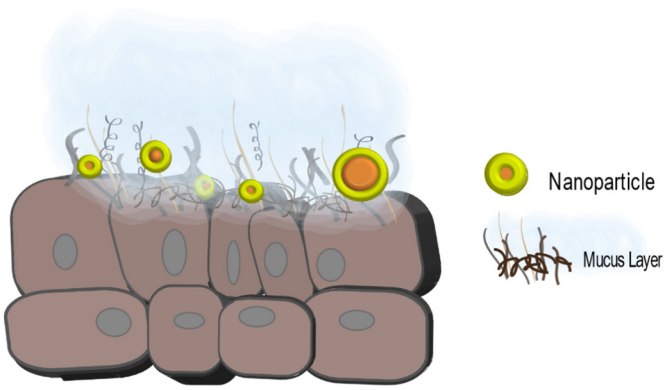

Fig. 5. Graphical representation of the NPs interaction with the mucus layer.

Although PLGA is not a mucoadhesive polymer it has widely been used in the ophthalmic field to prepare NPs [120,121]. Nevertheless, just because of the poor mucoadhesivity of PLGA the coating of the relevant pre-formed NPs with PEG has been proposed [122]. The authors compared the reducing effect of a melatonin solution on intraocular pressure with that of melatonin containing PLGA NPs and of the same coated with PEG. They observed a pressure reduction for a longer time in the case of the coated NPs. This was explained with the ability of PEG chains to interpenetrate the mucus glycoproteins, and also, with a reduction of the negative charge on the surface of the coated NPs. Indeed, since cornea and conjunctiva are charged negatively a less negative zeta-potential of NPs could strengthen NP interaction with tissue [122].

In order to increase their mucoadhesivity NPs based on PLGA have also been coated with chitosan. It was found that the chitosan coating not only increased NP mucoadhesivity, and consequently precorneal residence time but also affected the drug release kinetics. Indeed the coating, once in contact with the tear fluid swells without dissolving, because of the neutral pH of this fluid, thus giving rise to a hydrogel layer, surrounding the NPs, hampering drug diffusion out of the particles [123].

Following this principle, chitosan NPs (Ch NP) containing the antitumor drug 5-fluorouracil (5-FU) have been introduced in a thermogel (TSOH) of pH 7.4. The thermogel containing Ch NPs provided a constant concentration of the antitumor drug in the aqueous phase up to 7 h from instillation [124]. As shown in Fig. 7, instillation of the TSOH containing positively (QA-Ch NP) or negatively (SB-Ch-NP) charged NPs in rabbit eyes led to a plateau of the drug in the aqueous vs time diagram in the 1–10 h range. The negative charges on the SB-Ch NP surfaces slowed down 5-FU release from thermogel, whereas the positive ones on the QA-Ch NPs increased contact with the negatively charged eye surface [125].

4.1.2. Lipid-based nanocarriers

Lipid-based nanocarriers include microemulsions, self-emulsifying drug delivery systems (SEDDS), liposomes, nanoliposomes, archeosomes, solid lipid NPs (SLN), nanostructured lipid carriers (NLC) and micelles [126,127]. They are of high relevance because of their tunable properties and capability to overcome solubility problems of lipophilic drugs that are poorly soluble or insoluble in tear fluid. Without an appropriate surface decoration, however, the mucoadhesive properties of all these lipid-based nanocarriers are comparatively low. A somewhat prolonged ocular residence time of these delivery systems can be explained by their entrapment in the mucus gel layer and unspecific interactions of their surface with mucus glycoproteins. A promising strategy to prolong their precorneal residence time is the introduction of a cationic surface (i), able to interact with the negatively charged mucus layer or epithelium. Another strategy is based on the coating of lipid-based nanocarriers with mucoadhesive polymers (ii). Furthermore, due to the immobilization of thiol groups on the surface of these nanocarriers (iii), they adhere to the mucus gel layer via the formation of disulfide bonds with cysteine-rich subdomains of mucus glycoproteins.

4.1.2.1. Microemulsions. Microemulsions are thermodynamic stable dispersions of an oil and aqueous phase with a droplet size of less than around 300 nm. They are transparent systems exhibiting a high spreading capability onto the ocular surface. Ma et al. designed a cationic microemulsion in order to prolong the corneal retention behavior of this lipophilic delivery system [128]. In other studies a chitosan-coated cationic microemulsion was designed for treatment in case of chronic uveitis providing high mucoadhesive properties. The developed

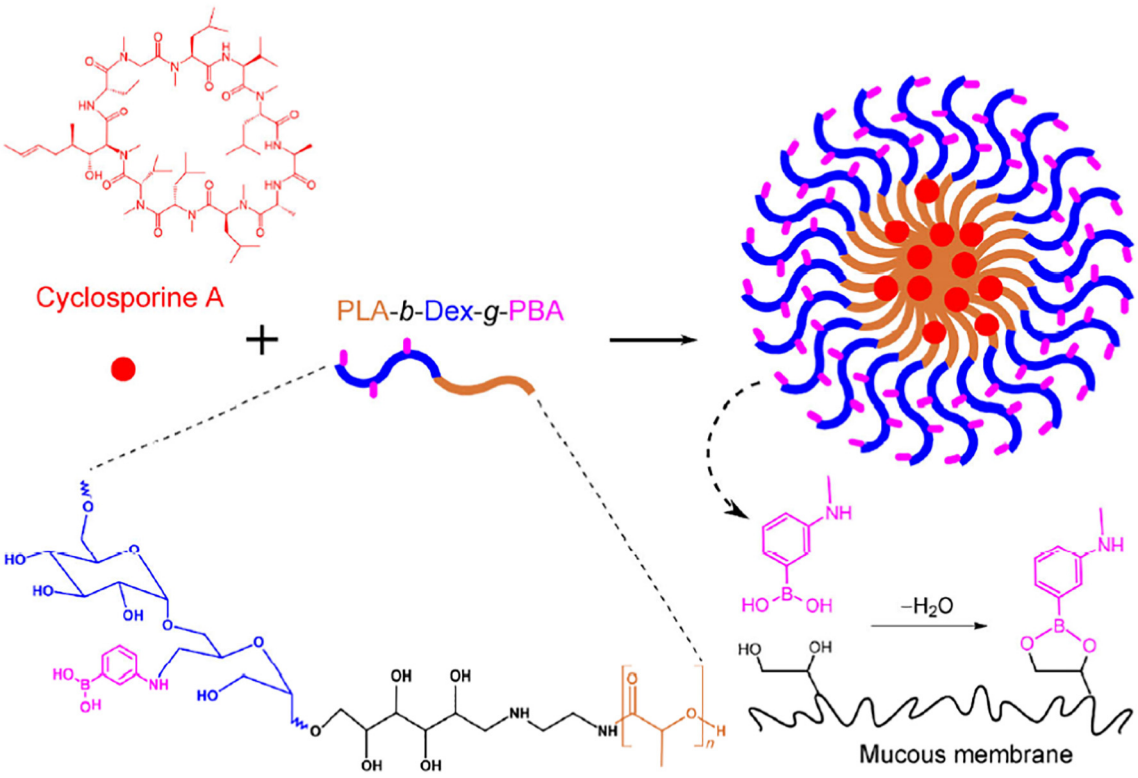

Fig. 6. Schematic illustration of the self-assembly process to form Cyclosporine A loaded PLA-b-Dex-g-PBA nanoparticles, and the mucoadhesion mechanism of the grafted PBA. Illustration taken from [117].

microemulsion significantly improved the therapeutic effects of the incorporated drug in vivo [129,130]. Ibrahim et al. designed microemulsions in order to enhance corneal residence time and penetration of ribavirin. A comparison of microemulsions containing the mucoadhesive polymers alginate and crosslinked polyacrylate (Carbopol 981) showed significantly higher adhesive properties of those containing the polyacrylate [131].

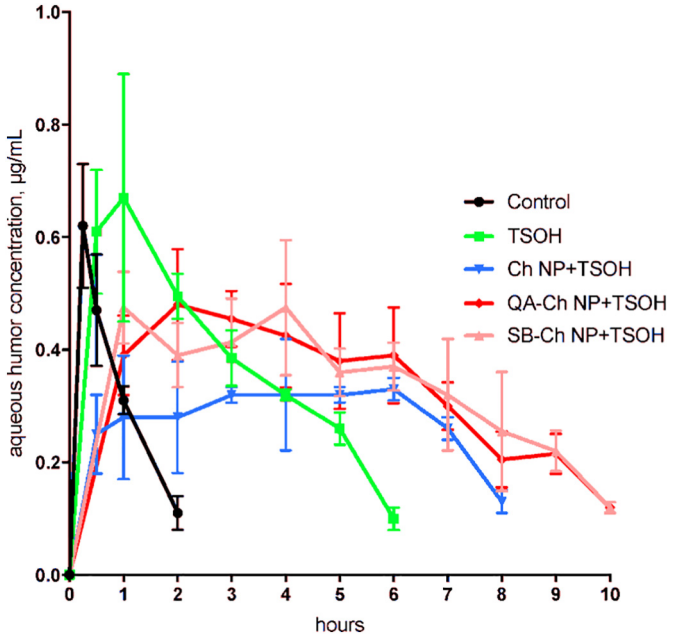

Fig. 7. Pharmacokinetics in the aqueous following instillation of ophthalmic drops in rabbit. Figure taken from [125].

4.1.2.2. Self-emulsifying drug delivery systems (SEDDS). SEDDS are one of the more promising vehicles for poorly soluble ocular drugs. They are made up of isotropic mixtures of oils, surfactants and co-surfactants that spontaneously form emulsions when brought into contact with an aqueous medium [132]. They are rather stable in view of their low critical micellar concentration (CMC). In water they consist of a hydrophobic core in a hydrophilic shell, whereby the core can be loaded with drug while the surrounding polymer chains interact with the biologic substrate. Recently, thiolated and S-protected Eudragit®L100-55 has been embedded in SEDDS for the ocular administration of econazole. As thiomers are oxidized in aqueous solutions at pH >5, they partly lose their mucoadhesivity. For this reason, the protection of the sulfhydryl group by the formation of a disulfide bond with an aromatic leaving group has been proposed. The reducing environment of the ocular surface allows setting the thiol groups free to bind to the cysteine residues of mucus. According to in vitro studies this system taking advantage of the thiomers and SEDDS synergy, showed high mucoadhesivity and provided a controlled drug release [133].

4.1.2.3. Liposomes. Liposomes are appropriate for the entrapment of both hydrophilic and hydrophobic drugs [134]. Although readily internalized by corneal cells, conventional liposomes are unable to prolong drug residence in precorneal area, and drugs are consequently rapidly drained away from the absorption site [135]. In order to address this shortcoming, cationic liposomes were designed for the ophthalmic administration of ibuprofen [136]. The delivery system provides a prolonged drug precorneal residence time, an enhanced transcorneal permeation and an improved bioavailability in aqueous humor. Furthermore, a higher T_{max} than that for the ibuprofen solution was observed, indicating that the liposome based formulation was able to maintain a higher

drug concentration on the cornea [136]. Coating with mucoadhesive polymers has been a further strategy to prolong liposome residence in precorneal area. Lin et al. coated conventional liposomes with hyaluronic acid [137]. Following instillation in rabbit eyes, these liposomes showed a longer retention time than the bare liposomes. Indeed, the analysis of the ocular surface by confocal microscope showed that the liposomes coated with hyaluronic acid displayed a longer residence in precorneal area than that of the non-modified liposomes [137]. A similar strategy was adopted by Tan et al., who coated liposomes with chitosan [138]. Hydroxypropylmethyl cellulose (HPMC) is well tolerated and largely used in eye drops as a thickener and, exhibits pronounced mucoadhesive properties. The synergistic action of liposomes and HPMC has allowed reducing the instillation frequency needed to maintain an adequate timolol concentration in precorneal area and to enhance the lowering effect on the intraocular pressure of rabbits with or without glaucoma [139]. Mustafa et al. incorporated liposomes containing fluconazole into a hyaluronic hydrogel [106]. An increased drug corneal permeability was observed leading to a drug concentration in the aqueous phase above the MIC, maintained for up to 24 h from administration. This allowed a reduction of the frequency of eye drops instillation from 3 to 4 times down to once a day, thus improving patient compliance [106]. Phua et al. incorporated nanoliposomes into a thermosensitive hydrogel based on Pluronic to prolong the residence on cornea of senicapoc, a natural active for the treatment of alcaline corneal burns [107]. The residence time of formulations as observed in Sprague-Dawley rat eyes, as can be seen in Fig. 8, was increased up to 12 times with the hydrogel-containing formulations (60 min) compared to the liposomes alone (5 min) [107].

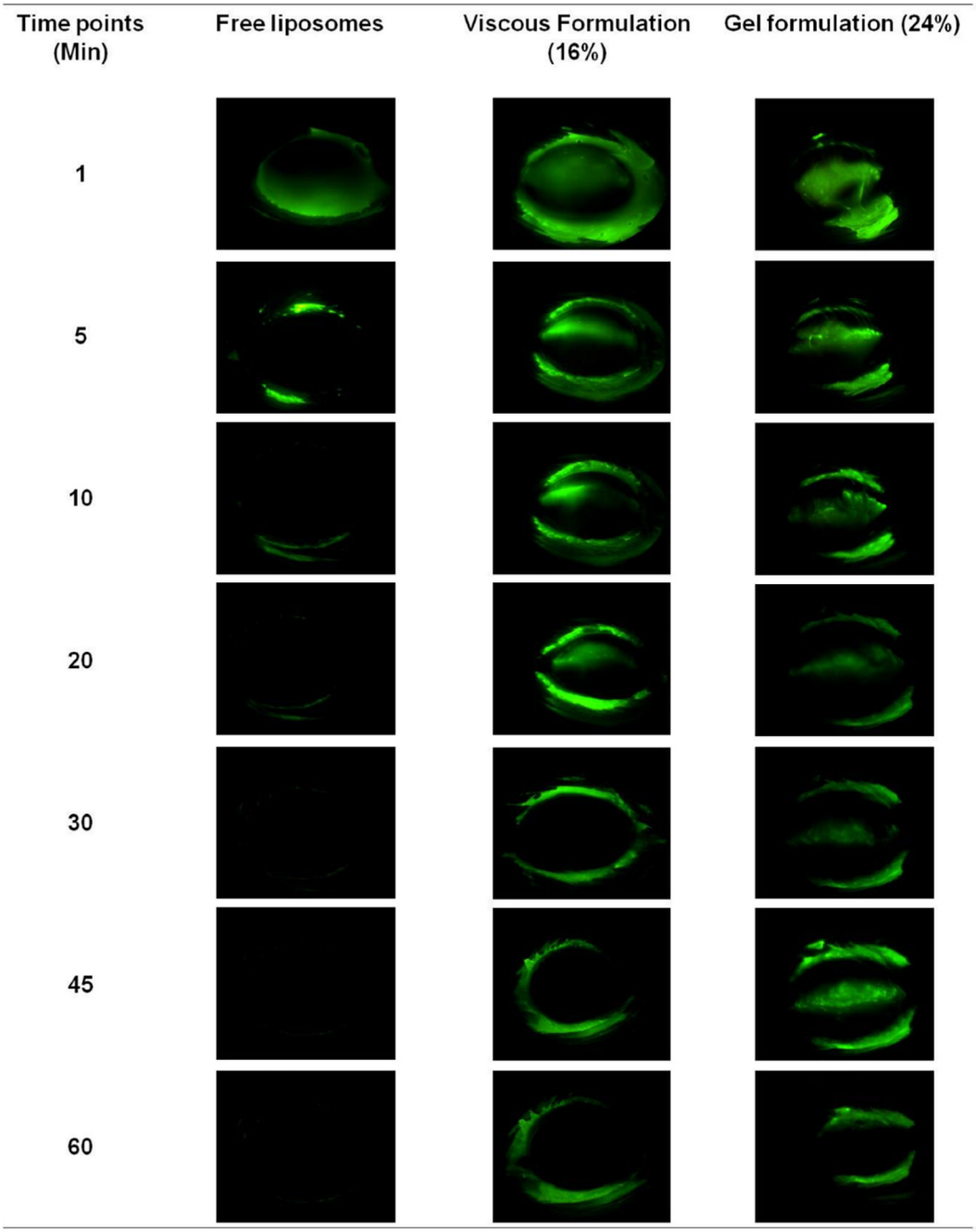

Fig. 8. Residence images (Micron IV Imaging) of free fluorescein-tagged liposomes and hydrogel formulations in eyes of anesthetized Sprague-Dawley rats. Figure taken from [140].

4.1.2.4. Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC). SLN and NLC are lipid based colloidal systems prepared by emulsifying a mixture of melted lipids, drug and surfactant in aqueous media, followed by cooling. In SLN the drug is suspended in the structured solid lipid, whereas in NLC the drug is entrapped in a non-structured solid and liquid lipid mix [141]. These particles show higher stability than liposomes, and can be prepared by more straightforward techniques. The matrix being made up of physiological lipids is the big advantage of SLN and NLC, since the risk of acute and chronic toxicity is reduced. Many emulsifying agents have been proposed to stabilize the above mentioned systems by preventing NPs aggregation [142]. These systems have mainly been proposed to promote the ocular bioavailability of lipophilic drugs, sparingly soluble in tear fluid, and also, barely tolerated by the patient if administered in suspension [143]. SLN have shown the ability to promote the absorption of encapsulated drugs thanks to a prolonged drug retention in precorneal area [144]. Many research groups have focused on the design of SLN and NLC able to prolong drug residence time in precorneal area. A widely applied strategy in this respect is the introduction of lipids carrying positively charged polar heads providing ionic interactions with the negatively charged mucus gel layer [145,146]. A further strategy to prolong the retention of the nanosystem on the cornea has been to coat it with mucoadhesive polymers [147–149]. For example SLN coating with polyethylene glycol (PEG) in addition to chitosan oligosaccharide was proposed [150]. It is known, indeed, that the PEG chains can interpenetrate the mucin chains and stabilize molecules and particles in physiological fluids. Chitosan and PEG coated SLN were found to raise the ofloxacin concentration in the tear fluid up to about 3-fold compared to commercial eye drops [150]. The possibility to establish covalent bonds with specific ligands on the ocular surface can strengthen NP mucoadhesivity and prolong ocular residence. For this purpose, dexamethasone containing NLC have been functionalized with a derivative of boronic acid, (3-aminomethylphenyl) boronic acid conjugated with chondroitin sulphate meant to form affinity complexes with sialic acid residues on mucins [151]. Furthermore, NLC were made more mucoadhesive by coating with polymers carrying thiol groups forming disulfide bonds with cysteine residues present on mucus glycoproteins [73,152,153]. NLC have also been introduced into in situ gelling thermosensitive gels [154]. The liquid vehicle, composed of Pluronic and carboxymethyl chitosan, becomes a gel on the corneal surface, thus increasing the ocular retention time of NLC and the encapsulated drug [154].

4.1.3. Polymeric microparticles

Polymeric microparticles are effective ocular drug delivery systems. Because of a higher size up to 25 μm microparticles are more stable and allow a more accurate control of release. Moreover, their industrial production is simpler. Taking the internalization mechanisms by cells, namely, phagocytosis, macropinocytosis, clathrin-mediated endocytosis and caveolae-mediated endocytosis into consideration however, it should be mentioned that NPs can be taken up by cells via all these mentioned mechanisms, whereas microparticles can be taken up only by the first two of them. Therefore, microparticles are less penetrant [155]. The ophthalmic use of microparticulate systems is mostly recommended for drugs employed in pathologies, such as glaucoma or dry eye syndrome, requiring a constant drug concentration for a prolonged time in precorneal area or in anterior chamber. Such systems must ensure both a slow release and a prolonged residence time in precorneal area [156]. Furthermore, their mean particle size must be less than 25 μm to prevent the feeling of foreign body in the eye, which would reduce patient compliance. [157]. For example, Choy et al. prepared poly(lactic-*co*-glycolic acid)-based microparticles coated with the mucoadhesive polymer poly(ethylene glycol). The mucoadhesion of the latter particles was greater in vitro than those of poly(lactic-co-glycolic acid) alone. Furthermore, these microparticles were found in the eye of rabbits even 30 min after administration [158].

4.2. Solid formulations

4.2.1. Ocular inserts

Ocular inserts are solid systems intended for administration of drugs with local action or acting in the anterior chamber of the eye. Although ensuring an extensively prolonged drug residence time in precorneal area, they are just rarely used as they can move around the ocular surface causing discomfort, irritation, or easy loss unless they are mucoadhesive [159]. Furthermore, accidents such as traffic collisions having been attributed to ocular inserts because of a limited vision and irritation as well as poor patient compliance have directed the focus of pharmaceutical companies to other formulations.

An ocular insert currently on the market and in common use is Mydriasert®, releasing phenylephrine and tropicamide, used exclusively in hospitals to induce preoperative mydriasis. With a single application of the ocular insert a constant release of the drug to the ocular surface for two hours preceding surgery is provided without any personal assistance for this time period. Other marketed inserts are pilocarpine ophthalmic inserts (Ocusert®) and hydroxypropyl cellulose ophthalmic inserts (Lacrisert®).

Ocular inserts are mainly prepared with polymers. They are either reservoir systems where a membrane controls release, or monolithic matrices. They are either disk or elliptic shaped so as to be inserted in the superior or inferior conjunctival sac [160]. They can provide a controlled drug release and a constant drug concentration in tear fluid for a prolonged time, thus avoiding concentration peaks that could bring about side effects. In addition, these systems, being solid, are endowed with remarkable stability and often do not require preservatives [159].

One of the first solid inserts proposed was based on polyethylene oxide (PEO) [161]. When this insert is brought into contact with the tear fluid it is rapidly converted into an erodible hydrogel. PEO can form hydrogen bonds with water molecules. When PEO solid matrices come into contact with aqueous media the polymer hydrates and a superficial gel is formed that is eroded as the polymer is dissolved. Thus drug release is controlled by the swelling and/or erosion of polymer [161]. With erodible ocular inserts drug bioavailability is maximized if release is only controlled by insert erosion, since parallel mechanisms can increase drug release, and consequently, clearance. It should also be considered that in the case of erosion-controlled release the drug release kinetics would be of zero order, the release rate would be constant over time and, if release is more resisted than drug permeation across cornea, also the intraocular absorption would be of zero order. This was in fact the case for ophthalmic inserts based on gelatin, medicated with ciprofloxacin. Ciprofloxacin eye drops need administration of 1–2 drops every 15–20 min for the treatment of acute infections. The use of a gelatin based ocular inserts allowed limiting the administration frequency, while maintaining a constant ciprofloxacin concentration in the eye [160].

Within the scope of employing mucoadhesive polymers a mucoadhesive ocular insert based on thiolated polyacrylic acid was developed and compared with a non-thiolated one. The vehicle effectiveness was tested in human volunteers [159]. Fig. 9 shows the behavior of that mucoadhesive insert in the human eye. As expected, the thiolated inserts showed good adhesive properties. The concentration in tear fluid of fluorescein, used as a model for hydrophilic drugs, turned out to be constant for 8 h, whereas it decreased as rapidly as with either the eye drops or non-thiolated ocular insert, used as controls. Due to the prolonged drug residence time in precorneal area these mucoadhesive inserts could be appropriate for the prevention and treatment of post-surgical *endo*-ocular infections, as they ensure eye protection, also during night time [159].

4.2.2. Contact lenses

The attempt by pharmaceutical technologists to prolong the drug residence time in the precorneal area has also led to systems such as ocular films and contact lenses. Contact lenses are separated from cornea

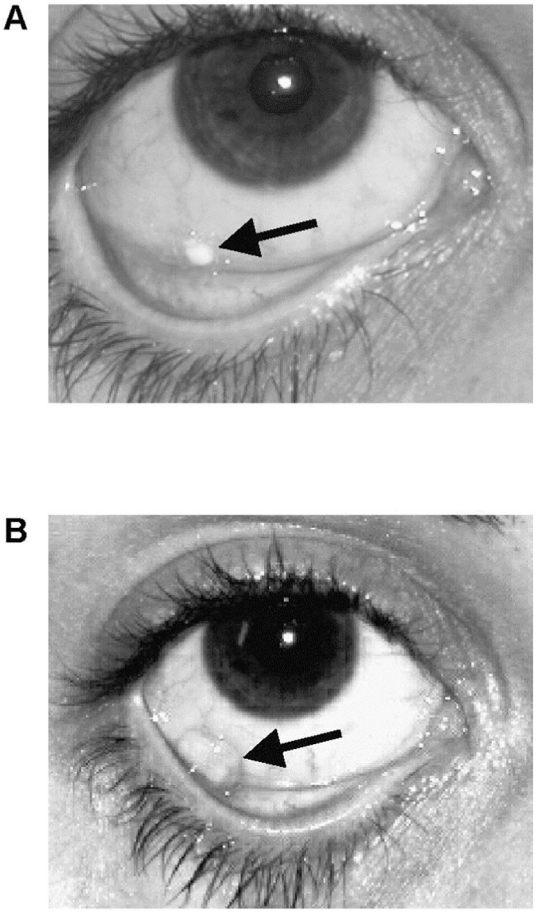

Fig. 9. (A) Mucoadhesive insert based on thiolated polyacrylic acid immediately after application; (B) 8 h after application. Figure taken from [159].

by a fluid layer defined post lens tear film. The clearance of post lens tear film is about 30 min that is clearly longer than the contact time of drugs administered by eye drops being in the range of a few minutes. A further important aspect concerning contact lenses is their bandaging properties. Under certain circumstances such as post-surgical treatment, ensuring eye protection is a necessity. Contact lenses while on the one hand controlling drug release, form on the other hand a physical barrier, so that optimal ocular conditions are restored [162].

The first publication that proposed contact lenses as drug vehicles dates back to 1960. In most of these first studies the inserts were medicated by incubation with a drug solution. Since then, numerous in vitro and in vivo studies have testified the effectiveness of the soak and release method. Yet these systems were shown to be unable to provide a sustained release for more than two hours [163]. This is probably the reason why after five decades just a limited number of products has reached the market [164]. With whatever vehicle drug release can be slowed down by creating a barrier to drug diffusion, it is rather complex to apply to contact lenses, as they would be expected both to guarantee a controlled release and to preserve transparency of device and permeability of ions and oxygen across it.

Chauhan et al. suggested loading of timolol into silicone contact lenses pre-treated with vitamin E, for the treatment of glaucoma [165]. Vitamin E was meant to oppose a physical barrier to drug diffusion. This approach was found to be particularly useful for hydrophilic drugs such as timolol and fluconazole exhibiting in their ionized form a negligible solubility in vitamin E [165,166]. In vivo studies showed an ability of this type of systems, loaded with timolol, to reduce intraocular pressure for four consecutive days, during which the same lenses were worn [167].

A further approach was to encapsulate the drug in NPs and subsequently immobilize them in contact lenses. By this means the release can be adjusted by modulating NP characteristics. Drug loaded NPs, immobilized in contact lenses as nano-reservoir systems, showed a 24–48 h sustained drug release [163] which was subsequently brought up to 48–72 h [168]. Highly crosslinked spherical NPs were prepared from monomers with multivinyl functions, such as glycerol propoxylate triacylate and ethylene glycol dimethacrylate. NPs of about 3.5 nm containing timolol were loaded in commercial contact lenses. In vitro tests showed a slow timolol release from NPs over four weeks, by a mechanism dependent on hydrolysis of ester bonds between timolol and the particle matrix that had been formed during the NP formation process. As the release depended also on temperature, the temperature change following lens insertion triggered drug release [163].

Maulvi et al. described yet another example of NPs loaded in contact lenses [169]. The authors suggested implanting a ring loaded with ethylcellulose-based NPs containing timolol maleate into contact lenses that provided a controlled drug release of therapeutic concentrations without prejudicing the critical properties of lenses. In vivo studies of pharmacokinetics in rabbit tear fluid showed a significantly higher mean residence time and AUC for tear fluid with contact lenses compared to eye drops therapy. In vivo pharmacodynamic data obtained with the rabbit model showed a marked reduction of intraocular pressure for 192 h [169].

A similar strategy has recently been proposed by Xu et al. [170]. In this case micelles prepared from the monomer hydroxyethylmethacrylate loaded with timolol and latanoprost were introduced in contact lenses. This is the first study ever where two drugs were released concurrently from micelles inserted in contact lenses [170]. Actually the system released timolol and latanoprost in simulated tear fluid for 144 h and 120 h, respectively. The pharmacokinetic study with rabbit eyes showed the presence of timolol and latanoprost in tear fluid for up to 120 h and 96 h, respectively. The mean residence in precorneal area for timolol or latanoprost was prolonged significantly (79.6 or 122.2 times) and the *endo*-ocular bioavailability was increased (2.2 or 7.3 times) in contact lenses compared to commercial eye drops. Considering the transparency and ion diffusion characteristics these lenses loaded with micelles have been found particularly interesting for the concurrent administration of two ocular drugs [170].

5. Concluding remarks

Within this review we have summarized the different strategies to prolong the residence time of drug delivery systems on ocular surface. From the excipients point of view in particular viscosity increasing and mucoadhesive polymers can essentially contribute to a prolonged drug residence time. Ideally, the release of drugs from these polymers should last as long as these auxiliary excipients remain on the ocular surface. Such a sustained release can be provided via ionic interactions between the drug and oppositely charged polymeric excipients, via hydrophobic interactions or via hydrogen bonding. Furthermore, cyclodextrins providing a sustained release of hydrophobic drugs being incorporated in their cavity can be embedded in such polymers. From the formulation point of view nano- and microcarrier systems such as polymeric nanoparticles and mircoparticles as well as lipid-based nanocarriers have shown high potential as they can be designed in a way that they exhibit both mucoadhesive and controlled drug release properties. Generally, nano- and microcarrier systems are instilled in form of liquid dispersions. Furthermore, nano- and microcarrier systems that provide a sustained drug release but do not exhibit sufficient mucoadhesive properties can be incorporated in in situ gelling and/or mucoadhesive polymeric gels. Apart from these liquid and semisolid formulations that allow to prolong the ocular residence time of many drugs from a few minutes to several hours, solid formulations like ocular inserts and contact lenses can remain on the ocular surface even for

days. As the compliance of solid formulations is limited, however, such delivery systems are not first choice.

The design of new excipients providing both high mucoadhesive and drug release controlling properties like thiolated cyclodextrins forming disulphide bonds with cysteine-rich subdomains of mucus glycoproteins on the ocular surface and providing a sustained release of drugs having been incorporated in their hydrophobic cavity will definitely shape the future landscape in this field. Moreover, the design of even more potent nano- and microcarrier systems in particular in combination with in situ gelling and mucoadhesive polymers will contribute to more efficient systems.

Declaration of Competing Interest

None.

References

[1] Andreoli CM, Miller JW. Anti-vascular endothelial growth factor therapy for ocular neovascular disease. Curr Opin Ophthalmol 2007;18:502–8.
[2] Li H, Palamoor M, Jablonski MM. Poly(ortho ester) nanoparticles targeted for chronic intraocular diseases: ocular safety and localization after intravitreal injection. Nanotoxicol 2016;10:1152–9.
[3] Cunha-Vaz J, Bernardes R, Lobo C. Blood-retinal barrier. Eur J Ophthalmol 2011;21 (Suppl. 6):S3–9.
[4] Lee J, Pelis RM. Drug transport by the blood-aqueous humor barrier of the eye. Drug Metab Dispos 2016;44:1675–81.
[5] Lee VH, Robinson JR. Topical ocular drug delivery: recent developments and future challenges. J Ocul Pharmacol 1986;2:67–108.
[6] Davies NM, Farr SJ, Hadgraft J, Kellaway IW. Evaluation of mucoadhesive polymers in ocular drug delivery. I Viscous solutions. Pharm Res 1991;8:1039–43.
[7] Leichner C, Jelkmann M, Bernkop-Schnürch A. Thiolated polymers: bioinspired polymers utilizing one of the most important bridging structures in nature. Adv Drug Deliv Rev 2019;151–152:191–221.
[8] de la Fuente M, Raviña M, Paolicelli P, Sanchez A, Seijo B, Alonso MJ. Chitosan-based nanostructures: a delivery platform for ocular therapeutics. Adv Drug Deliv Rev 2010;62:100–17.
[9] Rossi S, Vigani B, Bonferoni MC, Sandri G, Caramella C, Ferrari F. Rheological analysis and mucoadhesion: a 30 year-old and still active combination. J Pharm Biomed Anal 2018;156:232–8.
[10] Richoz O, Kling S, Zandi S, Hammer A, Spoerl E, Hafezi F. A constant-force technique to measure corneal biomechanical changes after collagen cross-linking. PLoS One 2014;9:e105095.
[11] Prüfert F, Hering U, Zaichik S, Le NN, Bernkop-Schnürch A. Synthesis and in vitro characterization of a preactivated thiolated acrylic acid/acrylamide-methylpropane sulfonic acid copolymer as a mucoadhesive sprayable polymer. Int J Pharm 2020; 583:119371.
[12] Da Silva SB, Ferreira D, Pintado M, Sarmento B. Chitosan-based nanoparticles for rosmarinic acid ocular delivery–in vitro tests. Int J Biol Macromol 2016;84:112–20.
[13] Verma A, Sharma G, Jain A, Tiwari A, Saraf S, Panda PK, et al. Systematic optimization of cationic surface engineered mucoadhesive vesicles employing Design of Experiment (DoE): a preclinical investigation. Int J Biol Macromol 2019;133:1142–55.
[14] Bonferoni MC, Sandri G, Ferrari F, Rossi S, Larghi V, Zambito Y, et al. Comparison of different in vitro and ex vivo methods to evaluate mucoadhesion of glycol-palmitoyl chitosan micelles. J Drug Deliv Sci Technol 2010;20:419–24.
[15] Pai RV, Monpara JD, Vavia PR. Exploring molecular dynamics simulation to predict binding with ocular mucin: an in silico approach for screening mucoadhesive materials for ocular retentive delivery systems. J Control Release 2019;309:190–202.
[16] Davidovich-Pinhas M, Bianco-Peled H. Mucoadhesion: a review of characterization techniques. Expert Opin Drug Deliv 2010;7:259–71.
[17] Roy S, Prabhakar B. Bioadhesive polymeric platforms for transmucosal drug delivery systems–a review. Trop J Pharm Res 2010;9.
[18] Argüeso P, Tisdale A, Spurr-Michaud S, Sumiyoshi M, Gipson IK. Mucin characteristics of human corneal-limbal epithelial cells that exclude the rose bengal anionic dye. Invest Ophthalmol Vis Sci 2006;47:113–9.
[19] Hafner A, Lovrić J, Romić MD, Juretić M, Pepić I, Cetina-Čižmek B, et al. Evaluation of cationic nanosystems with melatonin using an eye-related bioavailability prediction model. Eur J Pharm Sci 2015;75:142–50.
[20] Li J, Tian S, Tao Q, Zhao Y, Gui R, Yang F, et al. Montmorillonite/chitosan nanoparticles as a novel controlled-release topical ophthalmic delivery system for the treatment of glaucoma. Int J Nanomedicine 2018;13:3975–87.
[21] Bonferoni MC, Rossi S, Ferrari F, Caramella C. A modified Franz diffusion cell for simultaneous assessment of drug release and washability of mucoadhesive gels. Pharm Dev Technol 1999;4:45–53.
[22] Chaiyasan W, Srinivas SP, Tiyaboonchai W. Mucoadhesive chitosan-dextran sulfate nanoparticles for sustained drug delivery to the ocular surface. J Ocul Pharmacol Ther 2013;29:200–7.
[23] Worakul N, Robinson JR. Ocular pharmacokinetics/pharmacodynamics. Eur J Pharm Biopharm 1997;44:13.
[24] Gibaldi M, Perrier D. Pharmacokinetics, 2nd ed (New York) ; 1982.
[25] Di Colo G, Zambito Y, Zaino C, Sansò M. Selected polysaccharides at comparison for their mucoadhesiveness and effect on precorneal residence of different drugs in the rabbit model. Drug Dev Ind Pharm 2009;35:941–9.
[26] Huang Y, Tao Q, Hou D, Hu S, Tian S, Chen Y, et al. A novel ion-exchange carrier based upon liposome-encapsulated montmorillonite for ophthalmic delivery of betaxolol hydrochloride. Int J Nanomedicine 2017;12:1731–45.
[27] Zzafar A, Imam SS, Abbas Bukhari SN, Ahmad J, Ali A. Formulation and optimization of levofloxacin loaded chitosan nanoparticle for ocular delivery: in-vitro characterization, ocular tolerance and antibacterial activity. Int J Biol Macromol 2018;108: 650–9.
[28] Destruel PL, Zeng N, Maury M, Mignet N, Boudy V. In vitro and in vivo evaluation of in situ gelling systems for sustained topical ophthalmic delivery: state of the art and beyond. Drug Discov Today 2017;22:638–51.
[29] Maurice D. The effect of the low blink rate in rabbits on topical drug penetration. J Ocul Pharmacol Ther 1995;11:297–304.
[30] Makoid MC, Sieg JW, Robinson JR. Corneal drug absorption: an illustration of parallel first-order absorption and rapid loss of drug from absorption depot. J Pharm Sci 1976;65:150–3.
[31] Raizman MB, Rubin JM, Graves AL, Rinehart M. Tear concentrations of levofloxacin following topical administration of a single dose of 0.5% levofloxacin ophthalmic solution in healthy volunteers. Clin Ther 2002;24:1439–50.
[32] Torkildsen G, O'Brien TP. Conjunctival tissue pharmacokinetic properties of topical azithromycin 1% and moxifloxacin 0.5% ophthalmic solutions: a single-dose, randomized, open-label, active-controlled trial in healthy adult volunteers. Clin Ther 2008;30:2005–14.
[33] Snibson GR, Greaves JL, Soper ND, Prydal JI, Wilson CG, Bron AJ. Precorneal residence times of sodium hyaluronate solutions studied by quantitative gamma scintigraphy. Eye (Lond) 1990;4(Pt 4):594–602.
[34] Ludwig A, Van Ooteghem M. The evaluation of viscous ophthalmic vehicles by slit lamp fluorophotometry in humans. Int J Pharm 1989;54:95–102.
[35] Meadows DL, Paugh JR, Joshi A, Mordaunt J. A novel method to evaluate residence time in humans using a nonpenetrating fluorescent tracer. Invest Ophthalmol Vis Sci 2002;43:1032–9.
[36] Paugh JR, Nguyen AL, Ketelson HA, Christensen MT, Meadows DL. Precorneal residence time of artificial tears measured in dry eye subjects. Optom Vis Sci 2008;85: 725–31.
[37] Napoli PE, Coronella F, Satta GM, Zucca IA, Fossarello M. A novel OCT technique to measure in vivo the corneal adhesiveness for sodium carboxymethylcellulose in humans and its validity in the diagnosis of dry eye. Invest Ophthalmol Vis Sci 2014;55:3179–85.
[38] Gagliano C, Papa V, Amato R, Malaguarnera G, Avitabile T. Measurement of the retention time of different ophthalmic formulations with ultrahigh-resolution optical coherence tomography. Curr Eye Res 2018;43:499–502.
[39] Račić A, Čalij B, Milić J, Milašinović N, Krajišnik D. Development of polysaccharide based mucoadhesive ophthalmic lubricating vehicles: the effect of different polymers on physicochemical properties and functionality. J Drug Deliv Sci Technol 2018;49:50–7.
[40] Zahir-Jouzdani F, Wolf JD, Atyabi F, Bernkop-Schnürch A. In situ gelling and mucoadhesive polymers: why do they need each other? Expert Opin Drug Deliv 2018;15:1007–19.
[41] Fakhari A, Corcoran M, Schwarz A. Thermo-gelling properties of purified poloxamer 407. Heliyon 2017;3:e00390.
[42] Edsman K, Carfors J, Petersson R. Rheological evaluation of polxamer as an in-situ gel for ophthalmic use. Eur J Pharm Sci 1998;6:105–12.
[43] Zhu M, Wang J, Li N. A novel thermo-sensitive hydrogel-based on poly(N-isopropylacrylamide)/hyaluronic acid of ketoconazole for ophthalmic delivery. Artif Cells Nanomed Biotechnol 2018;46:1282–7.
[44] Wang Q, Zuo Z, Cheung CKC, Leung SSY. Updates on thermosensitive hydrogel for nasal, ocular and cutaneous delivery. Int J Pharm 2019;559:86–101.
[45] Vadnere M, Amidon G, Lindenbaum S. Thermodynamic studies on the gel-sol transition of some pluronic polyols. Int J Pharm 1984;2:207–18.
[46] Nijenhuis KT. Thermo-reversible networks: viscoelastic properties and structure of gels. Adv Polym Sci 1997;130:160–93.
[47] Bohorquez M, Koch C, Trygstad T, Pandit N. A study of the temperature-dependent Micellization of Pluronic F127. J Colloid Interface Sci 1999;216:34–40.
[48] Gratieri T, Gelfuso GM, de Freitas O, Rocha EM, Lopez RF. Enhancing and sustaining the topical ocular delivery of fluconazole using chitosan solution and poloxamer/chitosan in situ forming gel. Eur J Pharm Biopharm 2011;79:320–7.
[49] Huang W, Zhang N, Hua H, Liu T, Tang Y, Fu L, et al. Preparation, pharmacokinetics and pharmacodynamics of ophthalmic thermosensitive in situ hydrogel of betaxolol hydrochloride. Biomed Pharmacother 2016;83:107–13.
[50] Üstündağ-Okur N, Yozgatl V, Okur ME, Yolta A, Siafaka PI. Improving therapeutic efficacy of voriconazole against fungal keratitis thermo sensitive in situ gels as ophthalmic drug carriers. J Drug Deliv Sci Technol 2019;49:323–33.
[51] Li J, Liu H, Liu LL, Cai CN, Xin HX, Liu W. Design and evaluation of a brinzolamide drug-resin in situ thermosensitive gelling system for sustained ophthalmic drug delivery. Chem Pharm Bull (Tokyo) 2014;62:1000–8.
[52] Gupta C, Juyal V, Nagaich U. Formulation and optimization of thermosensitive in situ gel of moxifloxacin hydrochloride for ocular drug delivery. Int J Appl Pharm 2018;10:123–30.
[53] Vodithala S, Khatry S, Shastri N, Sadanandam M. Development and evaluation of thermoreversible ocular gels of ketorolac tromethamine. Int J Biopharm 2010;1: 39–45.
[54] Si EC, Bowman LM, Hosseini K. Pharmacokinetic comparisons of bromfenac in DuraSite and Xibrom. J Ocul Pharmacol Ther 2011;27:61–6.

[55] Sheppard JD, Cockrum PC, Justice A, Jasek MC. In vivo pharmacokinetics of Bromfenac ophthalmic solution 0.075%, bromfenac ophthalmic solution 0.07%, and Nepafenac/Amfenac ophthalmic suspension 0.3% in rabbits. Ophthalmol Therapy 2018;7:157–65.
[56] Wu Y, Liu Y, Li X, Kebebe D, Zhang B, Ren J, et al. Research progress of in-situ gelling ophthalmic drug delivery system. Asian J Pharm Sci 2019;14:1–15.
[57] Choi H, Jung JH, Ryu JM. Development of in situ gelling and mucoadhesive acetaminophen liquid suppository. 165. Int J Pharm 1998;165:33–44.
[58] Rupenthal ID, Green CR, Alany RG. Comparison of ion-activated in situ gelling systems for ocular drug delivery. Part 1: physicochemical characterisation and in vitro release. Int J Pharm 2011;411:69–77.
[59] Meng YC, Hong LB, Jin JQ. A study on the gelation properties and rheological behavior of Gellan gum. Appl Mech Mater 2013;284-287:20–4.
[60] Bhalerao H, Koteshwara KB, Chandran S. Levofloxacin hemihydrate in situ gelling ophthalmic solution: formulation optimization and in vitro and in vivo evaluation. Pharm Sci Tech 2019;20:272.
[61] Hommer A, Nowak A, Huber-Spitzy V. Multicenter double-blind study with 0.25% timolol in Gelrite (TG) once daily vs. 0.25% timolol solution (TS) twice daily. German Study Group. Ophthalmologe 1995;92:546–9.
[62] Shedden A, Laurence J, Tipping R, T.-X.S. Group. Efficacy and tolerability of timolol maleate ophthalmic gel-forming solution versus timolol ophthalmic solution in adults with open-angle glaucoma or ocular hypertension: a six-month, double-masked, multicenter study. Clin Ther 2001;23:440–50.
[63] Gipson IK. Distribution of mucins at the ocular surface. Exp Eye Res 2004;78:379–88.
[64] Ludwig A. The use of mucoadhesive polymers in ocular drug delivery. Adv Drug Deliv Rev 2005;57:1595–639.
[65] Khutoryanskiy VV. Advances in mucoadhesion and mucoadhesive polymers. Macromol Biosci 2011;11:748–64.
[66] Brannigan RP, Khutoryanskiy VV. Progress and current trends in the synthesis of novel polymers with enhanced mucoadhesive properties. Macromol Biosci 2019;19:e1900194.
[67] Lee D, Lu Q, Sommerfeld SD, Chan A, Menon NG, Schmidt TA, et al. Targeted delivery of hyaluronic acid to the ocular surface by a polymer-peptide conjugate system for dry eye disease. Acta Biomater 2017;55:163–71.
[68] Cheng YH, Tsai TH, Jhan YY, Chiu AW, Tsai KL, Chien CS, et al. Thermosensitive chitosan-based hydrogel as a topical ocular drug delivery system of latanoprost for glaucoma treatment. Carbohydr Polym 2016;144:390–9.
[69] Shi H, Wang Y, Bao Z, Lin D, Liu H, Yu A, et al. Thermosensitive glycol chitosan-based hydrogel as a topical ocular drug delivery system for enhanced ocular bioavailability. Int J Pharm 2019;570:118688.
[70] Kafedjiiski K, Föger F, Werle M, Bernkop-Schnürch A. Synthesis and in vitro evaluation of a novel chitosan-glutathione conjugate. Pharm Res 2005;22:1480–8.
[71] Krauland AH, Hoffer MH, Bernkop-Schnürch A. Viscoelastic properties of a new in situ gelling thiolated chitosan conjugate. Drug Dev Ind Pharm 2005;31:885–93.
[72] Yadav S, Ahuja M, Kumar A, Kaur H. Gellan–thioglycolic acid conjugate: Synthesis, characterization and evaluation as mucoadhesive polymer. Carbohydrate Polymers 2014;99:601–7.
[73] Li J, Tan G, Cheng B, Liu D, Pan W. Transport mechanism of chitosan-N-acetylcysteine, chitosan oligosaccharides or carboxymethyl chitosan decorated coumarin-6 loaded nanostructured lipid carriers across the rabbit ocular. Eur J Pharm Biopharm 2017;120:89–97.
[74] Postorino EI, Rania L, Aragona E, Mannucci C, Alibrandi A, Calapai G, et al. Efficacy of eyedrops containing cross-linked hyaluronic acid and coenzyme Q10 in treating patients with mild to moderate dry eye. Eur J Ophthalmol 2018;28:25–31.
[75] Zambito Y, Di Colo G. Chitosan and its derivatives as intraocular penetration enhancers. J Drug Deliv Sci Technol 2010;20:45–52.
[76] Zambito Y, Zaino C, Burchielli S, Carelli V, Serafini MF, Colo GD. Novel quaternary ammonium chitosan derivatives for the promotion of intraocular drug absorption. J Drug Del Sci Tech 2007;17:19–24.
[77] Di Colo G, Burgalassi S, Zambito Y, Monti D, Chetoni P. Effects of different N-trimethyl chitosans on in vitro/in vivo ofloxacin transcorneal permeation. J Pharm Sci 2004;93:2851–62.
[78] Zambito Y, Di Colo G. Thiolated quaternary ammonium-chitosan conjugates for enhanced precorneal retention, transcorneal permeation and intraocular absorption of dexamethasone. Eur J Pharm Biopharm 2010;75:194–9.
[79] Wang H, Li X, Yang H, Wang J, Li Q, Qu R, et al. Nanocomplexes based polyvinylpyrrolidone K-17PF for ocular drug delivery of naringenin. Int J Pharm 2020;578:119133.
[80] Bravo-Osuna I, Vicario-de-la-Torre M, Andrés-Guerrero V, Sánchez-Nieves J, Guzmán-Navarro M, de la Mata FJ, et al. Novel water-soluble mucoadhesive carbosilane dendrimers for ocular administration. Mol Pharm 2016;13:2966–76.
[81] Pereira de Sousa I, Suchaoin W, Zupančič O, Leichner C, Bernkop-Schnürch A. Totally S-protected hyaluronic acid: evaluation of stability and mucoadhesive properties as liquid dosage form. Carbohydr Polym 2016;152:632–8.
[82] Mazet R, Choisnard L, Levilly D, Wouessidjewe D, Gèze A. Investigation of combined cyclodextrin and hydrogel formulation for ocular delivery of dexamethasone acetate by means of experimental designs. Pharmaceutics 2018;10.
[83] Bozkir A, Denli ZF, Basaran B. Effect of hydroxypropyl-beta-cyclodextrin on the solubility, stability and in-vitro release of ciprofloxacin for ocular drug delivery. Acta Pol Pharm 2012;69:719–24.
[84] Bíró T, Horvát G, Budai-Szűcs M, Csányi E, Urbán E, Facskó A, et al. Development of prednisolone-containing eye drop formulations by cyclodextrin complexation and antimicrobial, mucoadhesive biopolymer. Drug Des Devel Ther 2018;12:2529–37.
[85] Piras AM, Fabiano A, Chiellini F, Zambito Y. Methyl-β-cyclodextrin quaternary ammonium chitosan conjugate: nanoparticles vs macromolecular soluble complex. Int J Nanomedicine 2018;13:2531–41.
[86] Asim MH, Nazir I, Jalil A, Laffleur F, Matuszczak B, Bernkop-Schnürch A. Per-6-thiolated cyclodextrins: a novel type of permeation enhancing excipients for BCS class IV drugs. ACS Appl Mater Interfaces 2020;12:7942–50.
[87] Asim MH, Jalil A, Shahzadi I, Khan M, Matuszczak B, Bernkop-Schnürch A. Mucoadhesive S-protected thiolated cyclodextrin-iodine complexes: a promising strategy to prolong mucosal residence time of iodine. Future Microbiol 2019;39:693–6.
[88] Asim MH, Moghadam A, Ijaz M, Mahmood A, Götz RX, Matuszczak B, et al. S-protected thiolated cyclodextrins as mucoadhesive oligomers for drug delivery. J Colloid Interface Sci 2018;531:261–8.
[89] Ijaz M, Ahmad M, Akhtar N, Laffleur F, Bernkop-Schnürch A. Thiolated alpha-cyclodextrin: the invisible choice to prolong ocular drug residence time. J Pharm Sci 2016;105:2848–54.
[90] Versura P, Profazio V, Balducci N, Campos EC. Efficacy of two-month treatment with Xiloial eyedrops for discomfort from disposable soft contact lenses. Clin Ophthalmol 2010;4:1035–41.
[91] Uccello-Barretta G, Nazzi S, Zambito Y, Di Colo G, Balzano F, Sansò M. Synergistic interaction between TS-polysaccharide and hyaluronic acid: implications in the formulation of eye drops. Int J Pharm 2010;395:122–31.
[92] Salzillo R, Schiraldi C, Corsuto L, D'Agostino A, Filosa R, De Rosa M, et al. Optimization of hyaluronan-based eye drop formulations. Carbohydr Polym 2016;153:275–83.
[93] Di Colo G, Zambito Y, Burgalassi S, Nardini I, Saettone MF. Effect of chitosan and of N-carboxymethylchitosan on intraocular penetration of topically applied ofloxacin. Int J Pharm 2004;273:37–44.
[94] Bernkop-Schnürch A. Muco-Adhesive Polymers, Use Thereof and Method for Producing the Same (United States) ; 1999.
[95] Hintzen F, Hauptstein S, Perera G, Bernkop-Schnürch A. Synthesis and in vitro characterization of entirely S-protected thiolated pectin for drug delivery. Eur J Pharm Biopharm 2013;85:1266–73.
[96] Dangl D, Hornof M, Hoffer M, Kuntner C, Wanek T, Kvaternik H. In vivo evaluation of ocular residence time of ^{124}I-labelled thiolated chitosan in rabbits using MicroPET technology. ARVO Annual Meeting Abstract; 2009.
[97] Messina M, Dua HS. Early results on the use of chitosan-N-acetylcysteine (Lacrimera®) in the management of dry eye disease of varied etiology. Int Ophthalmol 2018.
[98] Nepp J, Knoetzl W, Prinz A, Hoeller S, Prinz M. Management of moderate-to-severe dry eye disease using chitosan-N-acetylcysteine (Lacrimera®) eye drops: a retrospective case series. Int Ophthalmol 2020;40:1547–52.
[99] Jacob S, Nair AB. Cyclodextrin complexes: perspective from drug delivery and formulation. Drug Dev Res 2018;79:201–17.
[100] Morrison PW, Connon CJ, Khutoryanskiy VV. Cyclodextrin-mediated enhancement of riboflavin solubility and corneal permeability. Mol Pharm 2013;10:756–62.
[101] Calles JA, Mora MJ, Onnainty R, Tartara LI, Granero GE, Longhi MR, et al. Cross-linked hyaluronan films loaded with acetazolamide-cyclodextrin-triethanolamine complexes for glaucoma treatment. Ther Deliv 2018;9:205–20.
[102] Nanda A, Sahoo RN, Pramanik A, Mohapatra R, Pradhan SK, Thirumurugan A, et al. Drug-in-mucoadhesive type film for ocular anti-inflammatory potential of amlodipine: effect of sulphobutyl-ether-beta-cyclodextrin on permeation and molecular docking characterization. Colloids Surf B: Biointerfaces 2018;172:555–64.
[103] Piras AM, Zambito Y, Burgalassi S, Monti D, Tampucci S, Terreni E, et al. A water-soluble, mucoadhesive quaternary ammonium chitosan-methyl-β-cyclodextrin conjugate forming inclusion complexes with dexamethasone. J Mater Sci Mater Med 2018;29:42.
[104] Budai-Szűcs M, Kiss EL, Szilágyi B, Szilágyi A, Gyarmati B, Berkó S, et al. Mucoadhesive cyclodextrin-modified thiolated poly(aspartic acid) as a potential ophthalmic drug delivery system. Polymers (Basel) 2018;10.
[105] Järvinen T, Järvinen K, Urtti A, Thompson D, Stella VJ. Sulfobutyl ether beta-cyclodextrin (SBE-beta-CD) in eyedrops improves the tolerability of a topically applied pilocarpine prodrug in rabbits. J Ocul Pharmacol Ther 1995;11:95–106.
[106] Xu Y, Zhang C, Zhu X, Wang X, Wang H, Hu G, et al. Chloramphenicol/sulfobutyl ether-β-cyclodextrin complexes in an ophthalmic delivery system: prolonged residence time and enhanced bioavailability in the conjunctival sac. Expert Opin Drug Deliv 2019;16:657–66.
[107] Jóhannsdóttir S, Kristinsson JK, Fülöp Z, Ásgrímsdóttir G, Stefánsson E, Loftsson T. Formulations and toxicologic in vivo studies of aqueous cyclosporin a eye drops with cyclodextrin nanoparticles. Int J Pharm 2017;529:486–90.
[108] Wang F, Bao X, Fang A, Li H, Zhou Y, Liu Y, et al. Nanoliposome-encapsulated Brinzolamide-hydropropyl-β-cyclodextrin inclusion complex: a potential therapeutic ocular drug-delivery system. Front Pharmacol 2018;9:91.
[109] Anshul S, Renu S. A review on levofloxacin in situ-gel formulation. Asian J Pharm Clin Res 2014;8:37–41.
[110] Al-Kinani AA, Zidan G, Elsaid N, Seyfoddin A, Alani AWG, Alany RG. Ophthalmic gels: past, present and future. Adv Drug Deliv Rev 2018;126:113–26.
[111] Souto EB, Dias-Ferreira J, López-Machado A, Ettcheto M, Cano A, Camins Espuny A, et al. Advanced formulation approaches for ocular drug delivery: state-of-the-art and recent patents. Pharmaceutics 2019;11.
[112] Willcox MD, Argüeso P, Georgiev GA, Holopainen JM, Laurie GW, Millar TJ, et al. TFOS DEWS II tear film report. Ocul Surf 2017;15:366–403.
[113] Wolff E. The muco-cutaneous junction of the lidmargin and the distribution of the tear fluid. Trans Ophthalmol Soc UK 1946;66:291–308.

[114] Nagarwal RC, Kant S, Singh PN, Maiti P, Pandit JK. Polymeric nanoparticulate system: a potential approach for ocular drug delivery. J Control Release 2009;136:2–13.
[115] Alvarez-Alvarez L, Barral L, Bouza R, Farrag Y, Otero-Espinar F, Feijóo-Bandín S, et al. Hydrocortisone loaded poly-(3-hydroxybutyrate-co-3-hydroxyvalerate) nanoparticles for topical ophthalmic administration: preparation, characterization and evaluation of ophthalmic toxicity. Int J Pharm 2019;568:118519.
[116] Mittal N, Kaur G. Leucaena leucocephala (Lam.) galactomannan nanoparticles: optimization and characterization for ocular delivery in glaucoma treatment. Int J Biol Macromol 2019;139:1252–62.
[117] Liu S, Chang CS, Verma MS, Hileeto D, Muntz A, Stahl U, et al. Phenylboronic acid modified mucoadhesive nanoparticle drug carriers facilitate weekly treatment of experimentally induced dry eye syndrome. Nano Res 2015;8:621–35.
[118] Fabiano A, Chetoni P, Zambito Y. Mucoadhesive nano-sized supramolecular assemblies for improved pre-corneal drug residence time. Drug Dev Ind Pharm 2015;41:2069–76.
[119] Bhatta RS, Chandasana H, Chhonker YS, Rathi C, Kumar D, Mitra K, et al. Mucoadhesive nanoparticles for prolonged ocular delivery of natamycin: in vitro and pharmacokinetics studies. Int J Pharm 2012;432:105–12.
[120] Sánchez-López E, Esteruelas G, Ortiz A, Espina M, Prat J, Muñoz M, et al. Dexibuprofen biodegradable nanoparticles: one step closer towards a better ocular interaction study. Nanomaterials (Basel) 2020;10.
[121] Gonzalez-Pizarro R, Silva-Abreu M, Calpena AC, Egea MA, Espina M, García ML. Development of fluorometholone-loaded PLGA nanoparticles for treatment of inflammatory disorders of anterior and posterior segments of the eye. Int J Pharm 2018;547:338–46.
[122] Musumeci T, Bucolo C, Carbone C, Pignatello R, Drago F, Puglisi G. Polymeric nanoparticles augment the ocular hypotensive effect of melatonin in rabbits. Int J Pharm 2013;440:135–40.
[123] Yoncheva K, Vandervoort J, Ludwig A. Development of mucoadhesive poly(lactide-co-glycolide) nanoparticles for ocular application. Pharm Dev Technol 2011;16:29–35.
[124] Fabiano A, Bizzarri R, Zambito Y. Thermosensitive hydrogel based on chitosan and its derivatives containing medicated nanoparticles for transcorneal administration of 5-fluorouracil. Int J Nanomedicine 2017;12:633–43.
[125] Fabiano A, Piras AM, Guazzelli L, Storti B, Bizzarri R, Zambito Y. Impact of different mucoadhesive polymeric nanoparticles loaded in thermosensitive hydrogels on transcorneal administration of 5-fluorouracil. Pharmaceutics 2019;11.
[126] Danaei M, Dehghankhold M, Ataei S, Hasanzadeh Davarani F, Javanmard R, Dokhani A, et al. Impact of particle size and polydispersity index on the clinical applications of lipidic nanocarrier systems. Pharmaceutics 2018;10.
[127] Bonferoni MC, Sandri G, Dellera E, Rossi S, Ferrari F, Zambito Y, et al. Palmitoyl glycol chitosan micelles for corneal delivery of cyclosporine. J Biomed Nanotechnol 2016;12:231–40.
[128] Ma SW, Gan Y, Gan L, Zhu CL, Zhu JB. Preparation and in vitro corneal retention behavior of novel cationic microemulsion/in situ gel system. Yao Xue Xue Bao 2008;43:749–55.
[129] Kesavan K, Kant S, Singh PN, Pandit JK. Mucoadhesive chitosan-coated cationic microemulsion of dexamethasone for ocular delivery: in vitro and in vivo evaluation. Curr Eye Res 2013;38:342–52.
[130] Kesavan K, Pandit JK, Kant S, Muthu MS. Positively charged microemulsions of dexamethasone: comparative effects of two cosurfactants on ocular drug delivery and bioavailability. Ther Deliv 2013;4:1385–95.
[131] Ibrahim MM, Maria DN, Wang X, Simpson RN, Hollingsworth TJ, Jablonski MM. Enhanced corneal penetration of a poorly permeable drug using bioadhesive multiple microemulsion technology. Pharmaceutics 2020;12.
[132] Elkasabgy NA. Ocular supersaturated self-nanoemulsifying drug delivery systems (S-SNEDDS) to enhance econazole nitrate bioavailability. Int J Pharm 2014;460:33–44.
[133] Elbahwy IA, Lupo N, Ibrahim HM, Ismael HR, Kasem AA, Caliskan C, et al. Mucoadhesive self-emulsifying delivery systems for ocular administration of econazole. Int J Pharm 2018;541:72–80.
[134] Torchilin VP. Recent advances with liposomes as pharmaceutical carriers. Nat Rev Drug Discov 2005;4:145–60.
[135] Meisner D, Mezei M. Liposome ocular delivery systems. Adv Drug Deliv Rev 1995;16:75–93.
[136] Gai X, Cheng L, Li T, Liu D, Wang Y, Wang T, et al. In vitro and in vivo studies on a novel bioadhesive colloidal system: cationic liposomes of ibuprofen. AAPS PharmSciTech 2018;19:700–9.
[137] Lin J, Wu H, Wang Y, Chen Q, Zhu X. Preparation and ocular pharmacokinetics of hyaluronan acid-modified mucoadhesive liposomes. Drug Deliv 2016;23:1144–51.
[138] Tan G, Yu S, Pan H, Li J, Liu D, Yuan K, et al. Bioadhesive chitosan-loaded liposomes: a more efficient and higher permeable ocular delivery platform for timolol maleate. Int J Biol Macromol 2017;94:355–63.
[139] Zhang HH, Luo QH, Yang ZJ, Pan WS, Nie SF. Novel ophthalmic timolol meleate liposomal-hydrogel and its improved local glaucomatous therapeutic effect in vivo. Drug Deliv 2011;18:502–10.
[140] Phua JL, Hou A, Lui YS, Bose T, Chandy GK, Tong L, et al. Topical delivery of senicapoc nanoliposomal formulation for ocular surface treatments. Int J Mol Sci 2018;19.
[141] Srinivasarao DA, Lohiya G, Katti DS. Fundamentals, challenges, and nanomedicine-based solutions for ocular diseases. Wiley Interdiscip Rev Nanomed Nanobiotechnol 2019;11:e1548.
[142] Severino P, Szymanski M, Favaro M, Azzoni AR, Chaud MV, Santana MH, et al. Development and characterization of a cationic lipid nanocarrier as non-viral vector for gene therapy. Eur J Pharm Sci 2015;66:78–82.
[143] Lütfi G, Müzeyyen D. Preparation and characterization of polymeric and lipid nanoparticles of pilocarpine HCl for ocular application. Pharm Dev Technol 2013;18:701–9.
[144] Cavalli R, Gasco MR, Chetoni P, Burgalassi S, Saettone MF. Solid lipid nanoparticles (SLN) as ocular delivery system for tobramycin. Int J Pharm 2002;238:241–5.
[145] Abul Kalam M, Sultana Y, Ali A, Aqil M, Mishra AK, Chuttani K, et al. Part II: enhancement of transcorneal delivery of gatifloxacin by solid lipid nanoparticles in comparison to commercial aqueous eye drops. J Biomed Mater Res A 2013;101:1828–36.
[146] Nirbhavane P, Sharma G, Singh B, Begum G, Jones MC, Rauz S, et al. Triamcinolone acetonide loaded-cationic nano-lipoidal formulation for uveitis: evidences of improved biopharmaceutical performance and anti-inflammatory activity. Colloids Surf B: Biointerfaces 2020;190:110902.
[147] Pai RV, Vavia PR. Chitosan oligosaccharide enhances binding of nanostructured lipid carriers to ocular mucins: effect on ocular disposition. Int J Pharm 2020;577:119095.
[148] Üstündağ-Okur N, Gökçe EH, Bozbıyık D, Eğrilmez S, Ozer O, Ertan G. Preparation and in vitro-in vivo evaluation of ofloxacin loaded ophthalmic nano structured lipid carriers modified with chitosan oligosaccharide lactate for the treatment of bacterial keratitis. Eur J Pharm Sci 2014;63:204–15.
[149] Luo Q, Zhao J, Zhang X, Pan W. Nanostructured lipid carrier (NLC) coated with chitosan oligosaccharides and its potential use in ocular drug delivery system. Int J Pharm 2011;403:185–91.
[150] Eid HM, Elkomy MH, El Menshawe SF, Salem HF. Development, optimization, and in vitro/in vivo characterization of enhanced lipid nanoparticles for ocular delivery of ofloxacin: the influence of pegylation and chitosan coating. AAPS PharmSciTech 2019;20:183.
[151] Tan G, Li J, Song Y, Yu Y, Liu D, Pan W. Phenylboronic acid-tethered chondroitin sulfate-based mucoadhesive nanostructured lipid carriers for the treatment of dry eye syndrome. Acta Biomater 2019;99:350–62.
[152] Shen J, Deng Y, Jin X, Ping Q, Su Z, Li L. Thiolated nanostructured lipid carriers as a potential ocular drug delivery system for cyclosporine a: improving in vivo ocular distribution. Int J Pharm 2010;402:248–53.
[153] Shen J, Wang Y, Ping Q, Xiao Y, Huang X. Mucoadhesive effect of thiolated PEG stearate and its modified NLC for ocular drug delivery. J Control Release 2009;137:217–23.
[154] Yu Y, Feng R, Li J, Wang Y, Song Y, Tan G, et al. A hybrid genipin-crosslinked dual-sensitive hydrogel/nanostructured lipid carrier ocular drug delivery platform. Asian J Pharm Sci 2019;14:423–34.
[155] Halasz K, Kelly SJ, Iqbal MT, Pathak Y, Sutariya V. Micro/nanoparticle delivery systems for ocular diseases. Assay Drug Dev Technol 2019;17:152–66.
[156] Rodríguez Villanueva J, Rodríguez Villanueva L. Turning the screw even further to increase microparticle retention and ocular bioavailability of associated drugs: the bioadhesion goal. Int J Pharm 2017;531:167–78.
[157] Gavini E, Bonferoni MC, Rassu G, Sandri G, Rossi S, Salis A, et al. Engineered microparticles based on drug-polymer coprecipitates for ocular-controlled delivery of Ciprofloxacin: influence of technological parameters. Drug Dev Ind Pharm 2016;42:554–62.
[158] Bin Choy Y, Park JH, Prausnitz MR. Mucoadhesive microparticles engineered for ophthalmic drug delivery. J Phys Chem Solids 2008;69:1533–6.
[159] Hornof M, Weyenberg W, Ludwig A, Bernkop-Schnürch A. Mucoadhesive ocular insert based on thiolated poly(acrylic acid): development and in vivo evaluation in humans. J Control Release 2003;89:419–28.
[160] Mundada AS, Shrikhande BK. Formulation and evaluation of ciprofloxacin hydrochloride soluble ocular drug insert. Curr Eye Res 2008;33.
[161] Di Colo G, Burgalassi S, Chetoni P, Fiaschi MP, Zambito Y, Saettone MF. Gel-forming erodible inserts for ocular controlled delivery of ofloxacin. Int J Pharm 2001;215:101–11.
[162] Guzman-Aranguez A, Fonseca B, Carracedo G, Martin-Gil A, Martinez-Aguila A, Pintor J. Dry eye treatment based on contact lens drug delivery: a review. Eye Contact Lens 2016;42:280–8.
[163] Jung HJ, Chauhan A. Temperature sensitive contact lenses for triggered ophthalmic drug delivery. Biomaterials 2012;33:2289–300.
[164] Bengani LC, Hsu KH, Gause S, Chauhan A. Contact lenses as a platform for ocular drug delivery. Expert Opin Drug Deliv 2013;10:1483–96.
[165] Peng CC, Burke MT, Carbia BE, Plummer C, Chauhan A. Extended drug delivery by contact lenses for glaucoma therapy. J Control Release 2012;162:152–8.
[166] Peng CC, Burke MT, Chauhan A. Transport of topical anesthetics in vitamin E loaded silicone hydrogel contact lenses. Langmuir 2012;28:1478–87.
[167] Peng CC, Ben-Shlomo A, Mackay EO, Plummer CE, Chauhan A. Drug delivery by contact lens in spontaneously glaucomatous dogs. Curr Eye Res 2012;37:204–11.
[168] Fazly Bazzaz BS, Khameneh B, Jalili-Behabadi MM, Malaekeh-Nikouei B, Mohajeri SA. Preparation, characterization and antimicrobial study of a hydrogel (soft contact lens) material impregnated with silver nanoparticles. Cont Lens Anterior Eye 2014;37:149–52.
[169] Maulvi FA, Lakdawala DH, Shaikh AA, Desai AR, Choksi HH, Vaidya RJ, et al. In vitro and in vivo evaluation of novel implantation technology in hydrogel contact lenses for controlled drug delivery. J Control Release 2016;226:47–56.
[170] Xu J, Ge Y, Bu R, Zhang A, Feng S, Wang J, et al. Co-delivery of latanoprost and timolol from micelles-laden contact lenses for the treatment of glaucoma. J Control Release 2019;305:18–28.