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Opthalmic In-Situ Gelling System

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Abstract

Amongst the various routes of drug delivery, the field of ocular drug delivery is one of the most interesting and challenging endeavors facing the pharmaceutical scientist for past 10-20 years. As an isolated organ, eye is very difficult to study from a drug delivery point of view. Despite these limitations, improvements have been made with the objective of maintaining the drug for an extended period. Within the last few years, in response to the advent of potent and versatile therapeutic agents, the diversity of conventional ophthalmic formulations has gradually evolved, extending well beyond simple solutions, suspensions and ointments, now includes a variety of types of drug administration. In most recent publications, authors have broadened the notion of conventional ophthalmic delivery systems to encompass more than simple solutions and suspensions. While not strictly 'conventional', the ready availability of several commonly used drug vehicles suggests they have achieved acceptance, have been elevated to the category of conventional, and will be considered in this comparison. In this article, we have summarized the different types of commonly used ophthalmic formulations with the advanced novel formulation in many respects like their applicability, acceptance, characteristics and utility. Poor bioavailability of ophthalmic solutions caused by dilution and drainage from the eye can be overcome by using in-situ-forming ophthalmic drug delivery systems prepared from polymers that exhibit reversible liquid-gel phase transition.

Key-Words: Opthalmic Solution, In-Situ, Hydrogel, Liquid-gel transition.

Introduction

The eye is a sensory organ that converts light to an electric signal that is treated and interpreted by the brain. Briefly, the eye ball is covered by three layers: an outer fibrous protective layer (sclera and cornea), a middle vascular layer(choroid),and an inner nervous layer (retina). The cornea is a clear, transparent, thin avascular tissue that is composed of five layers: epithelium, bowmans's layer, stroma, Descemet's membrane and endothelium. The stroma is the only hydrophilic layer. The eye is generally divided into two parts: the anterior and the posterior segments. The anterior segment includes the cornea, sclera, ciliary body, and the lens; these structures delimit a cavity: the anterior chamber filled with the aqueous humor. The posterior segment includes all the structures between the lens and the optic nerve that delimit a cavity: the vitreous filled with an aqueous gel (the vitreous humor).

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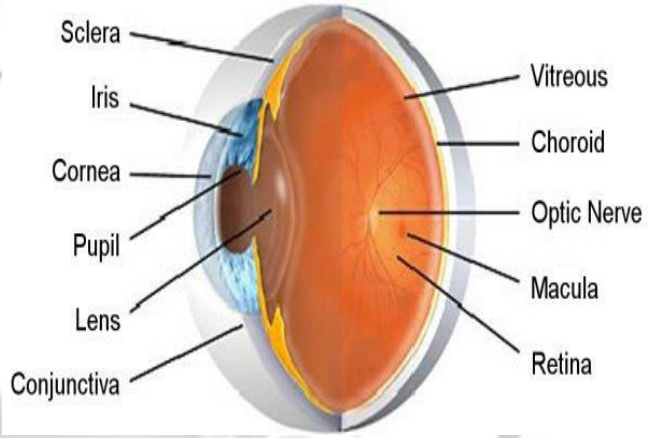

Fig. 1: Anatomy of human eye

The eye possesses efficient protective mechanisms like reflex blinking, lachrymation, and drainage, while lid closure protects the eye from external aggression. Tears permanently wash the surface of the eye and exert an anti-infectious activity through the lysozyme and immunoglobulins they contain. Finally, the lachrymal fluid is drained down the nasolacrimal pathways. All these protective mechanisms are responsible for te rapis and extensive procorneal loss of topically applied ophtalmic drugs [1-5].

Ophthalmic Disorders

Eye Infections

Eyes can get infections from bacteria, fungi or viruses. Eye infections can occur in different parts of the eye and can affect just one eye or both. Common eye infections are Conjunctivitis, Corneal ulcers & Endophthalmitis.

Conjunctivitis

Conjunctivitis is swelling (inflammation) or infection of the membrane lining the eyelids (conjunctiva).It is characterized by cellular infiltration and exudation. Staphylococcus aureus is the most common cause of bacterial conjunctivitis and blepharo-conjunctivitis. Many other organisms like Haemophilus influenza, Streptococcus pneumoniae also cause conjunctivitis. Conjunctivitis can be classified as (1) Infective – Acute, Sub acute & Chronic (2) Allergic conjunctivitis.

Corneal ulcers / Keratitis

Inflammation of cornea (Keratitis) is characterized by corneal edema, cellular infiltration & ciliary congestion. Being the most anterior part of eyeball, cornea is exposed to atmosphere & hence prone to get infected easily.Bacterial corneal ulcers are the most commonly caused by virulent organism. Common bacteria associated with corneal ulceration are Staphylococcus *aureus, Pseudomonas pyocyanea, E.coli, Proteus* etc.

Endophthalmitis

It is severe form of intraocular inflammation (purulent uveitis) involving ocular cavities & inner coats of eyeball. Causative organisms include *Streptococci, E.coli, Pseudomonas,* etc. Accordingly, the armamentarium of available antimicrobials used in the prevention and treatment of these infections includes antivirals, antifungals, and antibacterials. Common topical antibacterials used in the treatment of ocular infectious diseases include sulfonamides, aminoglycosides, polymyxin-based combinations, and **Fluoroquinolones**.

The fluoroquinolones represent an expanding class of broad-spectrum antibacterials which cover a host of Gram-negative and anaerobic species responsible for ocular infections. These antibacterials have gained popularity in the ophthalmology field since they have been shown to be equivalent to combination therapy in the treatment of many ocular infections.

Fluoroquinolones are also effective against a variety of Gram-positive organisms, including *Streptococcal* and *Staphylococcal* species; however, resistance is emerging among some of these organisms. [6, 7, 8]The classification (Table 1.1) and mechanism of action of fluoroquinolones are given below.

Table 1.1: Commonly Used Fluoroquinolones in Ophthalmic Delivery [9]

Antibiotic generation	Example	Activity
1 ST GENERATION	· Nalidixic acid	Have limited activity against gram negative & gram positive organism.
2 ND GENERATION	· Oxolinic acid · Cinoxacin · Pipemic acid	Improvement in gram negative coverage including antipseudomonal activity. Shows limited activity against gram positive organism.
3 RD GENERATION	· Norfloxacin · Ciprofloxacin · Levofloxacin · Ofloxacin	• Having antipseudomonal activity against gram negative bacilli.
4 TH GENERATION	· Ciprofloxacin · Moxifloxacin	• Having dual mechanism of action in gram positive bacteria in addition reducing efflux from the bacterial cell and Improved spectrum of activity.

Mechanism of action of Fluoroquinolones

Fluoroquinolones act by inhibiting two enzymes involved in bacterial DNA synthesis, both of which are DNA topoisomerases that human cells lack and that are essential for bacterial DNA replication, thereby enabling these agents to be both specific and bactericidal. [10]

DNA topoisomerases are responsible for separating the strands of duplex bacterial DNA, inserting another strand of DNA through the break, and then resealing the originally separated strands. DNA gyrase introduces negative superhelical twists in the bacterial DNA doublehelix ahead of the replication fork, thereby catalyzing the separation of daughter chromosomes. This activity is essential for initiation of DNA replication and allows for binding of initiation proteins. Topoisomerase IV is responsible for decatenation that is, removing the interlinking of daughter chromosomes thereby allowing segregation into two daughter cells at the end of a round of replication. Fluoroquinolones interact with the enzyme-bound DNA complex (i.e., DNA gyrase with bacterial DNA or topoisomerase IV with bacterial DNA) to create conformational changes that result in the inhibition of normal enzyme activity. As a result, the new drug– enzyme–DNA complex blocks progression of the replication fork, thereby inhibiting normal bacterial DNA synthesis and ultimately resulting in rapid bacterial cell death.[11]

Older fluoroquinolones exhibit a relatively consistent pattern with respect to specificity of enzyme inhibition in different types of bacteria. The newer fourth generation fluoroquinolones like moxifloxacin, gatifloxacin have a dual-binding mechanism of action, inhibiting both DNA gyrase and topoisomerase IV, in Grampositive species. [12]

OCULAR DRUG DELIVERY SYSTEM

Eye is most interesting organ due to its drug disposition characteristics. Generally, topical application of drugs is the method of choice under most circumstances because of its convenience and safety for ophthalmic chemotherapy. [13]

A significant challenge to the formulator is to circumvent (bypass) the protective barriers of the eye without causing permanent tissue damage. Development of newer, more sensitive diagnostic techniques and novel therapeutic agents continue to provide ocular delivery systems with high therapeutic efficacy. Conventional ophthalmic formulations like solution, suspension, and ointment have many disadvantages which result into poor bioavailability of drug in the ocular cavity. The specific aim of designing a therapeutic system is to achieve an optimal concentration of a drug at the active site for the appropriate duration. [14]

Ocular disposition and elimination of a therapeutic agent is dependent upon its physicochemical properties as well as the relevant ocular anatomy and physiology. A successful design of a drug delivery system, therefore, requires an integrated knowledge of the drug molecule and the constraints offered by the ocular route of administration. The various approaches that have been attempted to increase the bioavailability and the duration of the therapeutic action of ocular drugs can be divided into two categories. The first one is based on the use of sustained drug delivery systems, which provide the controlled and continuous delivery of ophthalmic drugs. The second involves maximizing corneal drug absorption and minimizing precorneal drug loss. [15]

Ideal ophthalmic drug delivery must be able to sustain the drug release and to remain in the vicinity of front of the eye for prolong period of time. Consequently it is imperative to optimize ophthalmic drug delivery; one of the ways to do so is by addition of polymers of various grades, development of in situ gel or colloidal suspension or using erodible or non erodible insert to prolong the precorneal drug retention. [16]

Routes of ocular drug delivery

There are several possible routes of drug delivery into the ocular tissues. (Figure 2) The selection of the route of administration depends primarily on the target tissue.

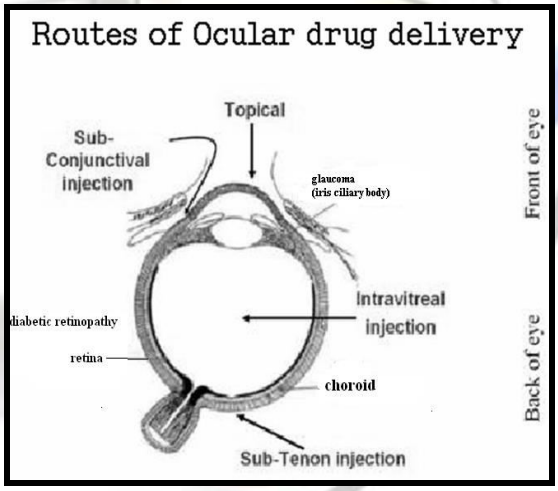

Fig. 2: Routes of ocular drug delivery

A. Topical route

Typically topical ocular drug administration is accomplished by eye drops, but they have only a short contact time on the eye surface. The contact, and thereby duration of drug action, can be prolonged by formulation design (e.g. gels, gellifying formulations, ointments, and inserts).

B. Subconjunctival administration

Traditionally subconjunctival injections have been used to deliver drugs at increased levels to the uvea. Currently this mode of drug delivery has gained new momentum for various reasons. The progress in materials sciences and pharmaceutical formulation have provided new exciting possibilities to develop controlled release formulations to deliver drugs to the posterior segment and to guide the healing process after surgery.

C. Intravitreal administration

Direct drug administration into the vitreous offers distinct advantage of more straightforward access to the vitreous and retina. It should be noted; however that delivery from the vitreous to the choroid is more complicated due to the hindrance by the RPE (Retinal Pigment Epithelium) barrier. Small molecules are able to diffuse rapidly in the vitreous but the mobility of large molecules, particularly positively charged, is restricted. [17]

Mechanism of ocular drug absorption

Drugs administered by instillation must penetrate the eye and do so primarily through the cornea followed by the non-corneal routes. These non-corneal routes involve drug diffusion across the conjunctiva and sclera and appear to be particularly important for drugs that are poorly absorbed across the cornea. (Figure 3)

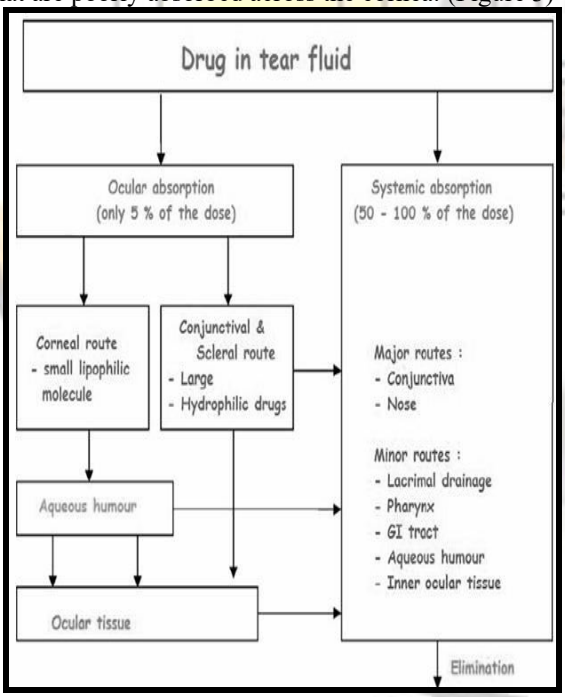

Fig. 3: Ocular Drug Absorption

A.Corneal permeation

The permeation of drugs across the corneal membrane occurs from the precorneal space. Thus, the mixing and the kinetic behavior of drug disposition in tears have a direct bearing on efficiency of drug absorption into the inner eye. The productive absorption of most ophthalmic drugs results from diffusion process across corneal membrane.

The efficiency of absorption process is a function of rate and extent at which the transport processes occur. The flux of any drug molecule across the biological membrane depends on the physicochemical properties of the permeating molecule and its interaction with the membrane. The extent to which the transport or absorption process occurs is also function of physiological mechanism of precorneal fluid drainage or turnover. In terms of transcorneal drug permeation, the cornea can be considered to consist of three primary layers (epithelium, stroma and endothelium).

The epithelium and endothelium contain on the order of a 100 fold greater amount of lipid material than the stroma. Consequently, depending on the physicochemical properties of a diffusing drug, the resistance offered by the individual layers varies greatly. Epithelium, being lipoidal, represents a diffusion barrier offering high resistance to ionic or other aqueous soluble or polar species. In contrast, compounds with relatively low polarity encounter a greater diffusional resistance in the hydrophilic stroma layer. This frequently cited concept of drug permeation across the corneal membrane is referred to as "differential solubility concept".

B. Non-corneal permeation

Primary mechanism of drug permeation is the sclera is likely to be diffusion across the intercellular aqueous media in the case of structurally similar corneal stroma. Therefore the possibility of partitioning mechanism cannot be eliminated. Although like cornea, the conjunctiva is composed of an epithelial layer covering an underlying stroma, the conjunctival epithelium offers substantially less resistance than the corneal epithelium. [18]

Interests of novel ophthalmic drug delivery

Ophthalmic drug delivery is one of the most interesting and challenging endeavors facing the pharmaceutical scientist. [19]

The landscape of ophthalmic drug delivery is highly competitive and rapidly evolving. New classes of pharmaceuticals and biologics are fueling the demand for novel drug delivery. The main aim of pharmacotherapeutics is the attainment of effective drug concentration at the site of action for the sufficient period of time to elicit a response.

The challenge is to provide a system with improved ocular drug bioavailability and prolonged duration of activity, but still with a minimum risk of ocular

complications. A major problem of ophthalmic drug delivery is not the lack of efficient drugs but the attainment of their optimal concentration at the site of their optimal concentration at the site of action.[20]

The emergence of new and innovative means for improving therapeutic efficacy suggests that a greater choice of dosage forms will be provided to physicians and patients in the next decade. Most of the formulation efforts aim at maximizing ocular drug absorption through prolongation of the drug residence time in the cornea and conjunctival sac, as well as to slow drug release from the delivery system and minimize precorneal drug loss. Various ophthalmic formulations and their residence time period in the ocular cavity are given below. (Figure 4)[21]

Fig. 4: Duration of action of ocular drug delivery systems

Various factors affecting poor bioavailability of ocular drugs

Bioavailability of drugs administered to the eye is an important consideration. There are physiological factors, which can affect a drug's bioavailability including protein binding, drug metabolism and lachrymal drainage.

Protein bound drugs are incapable of penetrating the corneal epithelium due to the size of the protein drug complex. Because of the brief time in which an ophthalmic solution may remain present in the eye (due to lachrymal drainage), protein binding of a drug substance could quickly negate its therapeutic value by rendering it unavailable for absorption.

One of the major problems encountered with conventional ophthalmic solutions is the rapid and extensive elimination of drugs from the precorneal lachrymal fluid. It must be noted that this high drainage rate is due to the tendency of the eye to maintain its residence volume at 7–10 µL permanently, whereas volumes topically instilled range from 20–50 µL.

In fact it has been demonstrated in vivo that 90% of the dose was cleared within 2 min for an instilled volume of 50 µL and, within 4 min for an instilled volume of 10 µl. Consequently, the ocular residence time of conventional solutions is limited to a few minutes, and the overall absorption of a topically applied drug is limited to 1–10%. As in the case with other biological fluids, tears contain enzymes (such as lysozymes) capable of the metabolic degradation of the drug substance.

In addition to the physiological factors affecting ocular bioavailability, other factors as the physicochemical properties of the drug substance, and product formulation are important. Because the cornea is a membrane-barrier containing both hydrophilic and lipophilic layers, it is permeated most effectively by drug substances having both hydrophilic and lipophilic characteristics. [18] It is advantageous for corneal penetration to adjust the pH of the solution to increase the proportion of unionized drug in the instilled dose. Drugs, which are highly water insoluble, do not readily permeate the cornea.

Nasolacrymal drainage system

The nasolachrymal drainage system consists of three parts: the secretory system, the distributive system and the excretory system. The secretory system consists of basic secretors that are stimulated by blinking and temperature change due to tear evaporation and reflex secretors that have an efferent parasympathetic nerve supply and secrete in response to physical or emotional stimulation.

The distributive system consists of the eyelids and the tear meniscus around the lid edges of the open eye, which spread tears over the ocular surface by blinking, thus preventing dry areas from developing.

The excretory part of the nasolachrymal drainage system consists of: the lachrymal puncta, the superior, inferior and common canaliculi; the lachrymal sac; and the nasolachrymal duct. In humans, the two puncta are the openings of the lachrymal canaliculi and are situated on an elevated area known as the lachrymal papilla. It is thought that tears are largely absorbed by

the mucous membrane that lines the ducts and the lachrymal sac; only a small amount reaches the nasal passage. [22]

Barriers for ocular delivery

A. Drug loss from the ocular surface

After instillation, the flow of lacrimal fluid removes instilled compounds fromthe surface of the eye. Even though the lacrimal turnover rate is only about 1 µl/min the excess volume of the instilled fluid is flown to the nasolacrimal duct rapidly in a couple of minutes. Another source of non-productive drug removal is its systemic absorption instead of ocular absorption. Systemic absorption may take place either directly from the conjunctival sac via local blood capillaries or after the solution flow to the nasal cavity.

B. Lacrimal fluid-eye barriers

Corneal epithelium limits drug absorption from the lacrimal fluid into the eye. The corneal epithelial cells form tight junctions that limit the paracellular drug permeation. Therefore, lipophilic drugs have typically at least an order of magnitude higher permeability in the cornea than the hydrophilic drugs. In general, the conjunctiva is leakier epithelium than the cornea and its surface area is also nearly 20 times greater than that of the cornea.

C. Blood-ocular barriers

The eye is protected from the xenobiotics in the blood stream by blood-ocular barriers. These barriers have two parts: blood-aqueous barrier and blood-retina barrier. The anterior blood-eye barrier is composed of the endothelial cells in the uvea (The middle layer of the eye beneath the the sclera. It consists of the iris, ciliary body, and choroid).

This barrier prevents the access of plasma albumin into the aqueous humor, and also limits the access of hydrophilic drugs from plasma into the aqueous humor. The posterior barrier between blood stream and eye is comprised of retinal pigment epithelium (RPE) and the tight walls of retinal capillaries. Unlike retinal capillaries the vasculature of the choroid has extensive blood flow and leaky walls. Drugs easily gain access to the choroidal extravascular space, but thereafter distribution into the retina is limited by the RPE and retinal endothelia.[17,23].

Polymeric drug delivery for ophthalmic

Hydrogels are one of the upcoming classes of polymer-based controlled release drug delivery systems. [24] Polymeric drug delivery systems have been extensively studied in order to solve the potential problems associated with drugs or bioactive molecules including toxicity, site dependence, low effectiveness, poor solubility, short half life, rapid degeneration and rapid clearance from the body. Considering various properties such as flexibility, structure, biocompatibility, and hydrophilicity, three dimensional matrices, hydrogels, are being extensively used as drug delivery carriers. [25]

Advantages of polymeric drug delivery

- Reduce toxic effects on the healthy tissue and reach sites that are conventionally Inaccessible due to the presence of various barriers[17] by targeted drug delivery.
- Increase the half-life of drugs, preventing their rapid degradation, and reduce the rate of elimination, thus maintaining drug concentration within a therapeutically effective window.
- Reduce the amount of drug required to achieve therapeutic efficacy.
- Cut down the number of repeated invasive dosage required for certain conditions and thus helps to improve patient's compliance and offers better living.

A. In situ hydrogels

Hydrogels are polymeric networks that absorb large quantities of water while remaining insoluble in aqueous solutions due to chemical or physical crosslinking of individual polymer chains. They resemble natural living tissue more than any other class of synthetic biomaterials due to their high water content; furthermore, the high water content of the materials contributes to their biocompatibility. [26]

Hydrogels show minimal tendency to adsorb proteins from body fluids because of their low interfacial tension. Further, the ability of molecules of different sizes to diffuse into (drug loading) and out of (drug release) hydrogels allows the possible use of dry or swollen polymeric networks as drug delivery systems for oral, nasal, buccal, rectal, vaginal, ocular and parenteral routes of administration. [27]

These are polymers endowed with an ability to swell in water or aqueous solvents and induce a liquid–gel transition.[28]

Currently; two groups of hydrogels are distinguished, namely preformed and in situ forming gels. Preformed hydrogels can be defined as simple viscous solutions which do not undergo any modifications after administration. The use of preformed hydrogels still has drawbacks that can limit their interest for ophthalmic drug delivery or as tear substitutes. They do not allow accurate and reproducible administration of quantities of drugs and, after administration; they often produce blurred vision, crusting of eyelids, and lachrymation.

Thus in situ hydrogels can be instilled as eye drops and undergo an immediate gelation when in contact with the eye. In situ-forming hydrogels are liquid upon instillation and undergo phase transition in the ocular

cul-de-sac to form viscoelastic gel and this provides a response to environmental changes. Three methods have been employed to cause phase transition on the surface: change in temperature, pH, and electrolyte composition.

Increase in solution viscosity by using polymers improves retention of product on the corneal surface. More recently, the approach to improve precorneal retention is based on the use of mucoadhesive polymers. The principle for use of bioadhesive vehicles relies on their ability to interact with the mucin-coating layer present at the eye surface. The polymers chosen to prepare ophthalmic hydrogels should meet some specific rheological characteristics. It is generally well accepted that the instillation of a formulation should influence tear behavior as little as possible. Because tears gave a pseudoplastic behavior, pseudoplastic vehicles would be more suitable as compare to Newtonian formulations, which have a constant viscosity independent of the shear rate, whereas pseudoplastic solution exhibit decreased viscosity with increasing shear rate, thereby offering lowered viscosity during blinking and stability of the tear film during fixation. [29, 30]

Drug release from hydrogels:

As discussed in the previous sections, hydrogels have a unique combination of characteristics that make them useful in drug delivery applications. Due to their hydrophilicity, hydrogels can imbibe large amounts of water. Therefore, the molecule release mechanisms from hydrogels are very different from hydrophobic polymers. Both simple and sophisticated models have been previously developed to predict the release of an active agent from a hydrogel device as a function of time. These models are based on the rate limiting step for controlled release and are therefore categorized as diffusion, swelling & chemically controlled mechanism (Fig. 5).

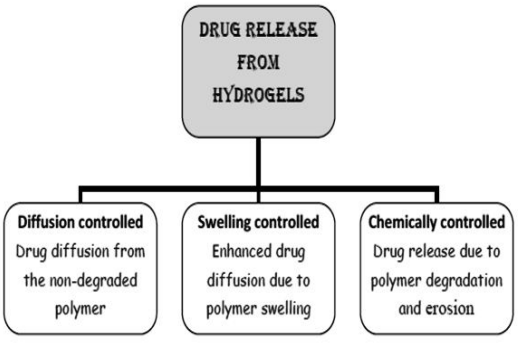

Fig. 5: Drug release from hydrogel [31]

B. Smart hydrogels

"Smart" hydrogels, or stimuli-sensitive hydrogels, are very different from inert hydrogels in that they can "sense" changes in environmental properties such as pH and temperature and respond by increasing or decreasing their degree of swelling. The volume-changing behavior of 'smart' hydrogels is particularly useful in drug delivery applications as drug release can be triggered upon environmental changes.

These "intelligent" or "smart" polymers play important role in drug delivery since they may dictate not only where a drug is delivered, but also when and with which interval it is released. The stimuli that induce various responses of the hydrogel systems include physical (temperature) or chemical (pH, ions) ones. [29]

In this, polymers may undergo phase transition in presence of various ions. Gellan gum commercially available as Gelrite® is an anionic polysaccharide that undergoes *in situ* gelling in the presence of mono- and divalent cations, including Ca^{2+}, Mg^{2+}, K^{+} and Na^{+}. Gelation of the low-methoxy pectins can be caused by divalent cations, especially Ca^{2+}. Mechanism and examples of stimuli sensitive hydrogels are given in Table 1.3. [32, 33, 34]

Table 1.3: Stimuli sensitivity of hydrogels

External stimuli	Mechanism	Examples
Temperature	Formulation is liquid at room temperature (20° – 25° C) which undergoes gelation with contact to body fluids (35° – 37°C) Temperature increases the degradation of polymer chain which leads to formation of hydrophobic domains and transition of an aqueous liquid to hydrogel network	Poloxamer/Pluronics Co-polymers of polyethylene oxide PEO Co-polymers of polypropylene oxide PPO Polyester Xyloglucan Cellulose derivatives

Ionic interactions	Formulation undergoes liquid-gel transition under influence of an increase in ionic strength Gel formation takes place because of complexation with polyvalent cations (like Ca^{+2}) in lacrimal fluid	Chitosan Gallen gum Alginates
pH change	Sol to gel transition when pH raised from 4.2 – 7.4 (eye pH) At higher pH polymer forms hydrogen bonds with mucin which leads to formation of hydrogel networks	Pseudolatexes Acrylates (Carbopols) Cellulose acetate phthalate (CAP) Polyox

References:

1. Khurana AK, Khurana I. Anatomy & physiology of Eye; 2nd ed. CBS publishers & Dist. 2007.280-298.
2. Khurana AK. Comprehensive ophthalmology; 4th ed. Age International (P) Ltd Pub. 2007.114-119.
3. Snell RS, Michel A. Clinical Anatomy of the eye; 2nd ed. Cemp. Blackwell science.112-120.
4. http://www.ivy-rose.co.uk/HumanBody/Eye/Anatomy_Eye.php.
5. Hosoyaa K, Vincent HL, Kim KJ .Roles of the conjunctiva in ocular drug delivery: a review of conjunctival transport mechanisms and their regulation; Eur J Pharm Biopharm 2005; 60:227–240.
6. Meqi SA, Deshpande SG. Ocular drug delivery: Controlled and novel drug delivery. New delhi:CBS Publishers; 2002. 82-84.
7. Eva M, Amo D, Urtti A. Current and future ophthalmic drug delivery systems. A shift to the posterior segment. Drug Discov Today 2004; 13:135-143.
8. Blondeau JM. Fluoroquinolones: Mechanism of Action, Classification, and Development of Resistance; Surv Ophthalmol 2004; 49:S73-S78.
9. Martinez M¸ McDermott P, Walker R. Pharmacology of the fluoroquinolones: A perspective for the use in domestic animals; The Veterinary Journal 2006; 172:10– 28.
10. Gupta P, Vermani K and Garg S. Hydrogels: from controlled release to pHresponsive drug delivery; Drug Discov Today 2002; 7:569-579.
11. Desai PN. Synthesis and characterization of polyionic hydrogels, Bachelors of Homoeopathic Medicine and Surgery; LMF's Homoeopathic Medical College, India, 2005.
12. He C, Kim SW, Lee DS. In situ gelling stimuli-sensitive block copolymer hydrogels for drug delivery, J Control Release 2008; 127:189–207.
13. Sasaki H, Yamamura K, Nishida K, Nakamurat J, Ichikawa M. Delivery of drugs to the eye by topical application. Progress in Retinal and Eye Research 1996; 15(2):553-620.
14. Macha S, Mitra AK. Ophthalmic drug delivery systems; 2nd edition revised and expanded. Chapter 1 Overview of Ocular Drug Delivery. 1-3.
15. Mundada AS, Avari JG, Mehta SP, Pandit SS, Patil AT. Recent advances in ophthalmic drug delivery system; Pharm Rev 2008; 6(1).
16. Wagh VD, Inamdar B, Samanta MK. Polymers used in ocular dosage form & drug delivery system; Asian J Pharm 2008;2(1):12-17.
17. Urtti A. Challenges and obstacles of ocular pharmacokinetics and drug delivery; Adv Drug Deliv Rev 2006; 58:1131–1135.
18. Jtirvinena K, Tomi J, Urttia SA. Ocular absorption following topical delivery; Adv Drug Deliv Rev 1995; 16:3-19.
19. Eva M, Amo D, Urtti A. Current and future ophthalmic drug delivery systems; A shift to the posterior segment. Drug Discov Today 2004; 13:135-143.
20. Blondeau JM. Fluoroquinolones: Mechanism of Action, Classification, and Development of

Resistance; Surv Ophthalmology 2004; 49:S73-S78.

21. Martinez M¸ McDermott P, Walker R. Pharmacology of the fluoroquinolones: A perspective for the use in domestic animals; The Veterinary Journal 2006; 172:10– 28.
22. Nanjawade BK, Manvi FV, Manjappa AS. In situ-forming hydrogels for sustained ophthalmic drug delivery; J Control Release 2007; 122:119–134.
23. Cross JT. Fluoroquinolones *Seminars* in Pediatric Infectious Diseases; 2001;12:211-223.
24. Satish CS, Satish KP, Shivkumar SG. Hydrogels as a controlled drug delivery system: Synthesis, cross linking, water and drug transport mechanism; Indian J Pharm Sci 2006; 03:133-141.
25. Bourlais CL, Acar L, Zia H, Sado PA, Needham T, Levergc R. Ophthalmic drug delivery systems recent advances; Progress in Retinal and Eye Research 1998;17(1):33-58.
26. Netland PA. Glaucoma Medical Therapy, Principles & Management; 2nd Ophthalmic Monograph 13:11-14.
27. Franzesi GT, Ni B, Ling Y, Khademhosseini A. A Controlled-Release Strategy for the Generation of Cross-Linked Hydrogel Microstructures; J Am Chem Soc 2006; 128:64-65.
28. Fang JY, Chen JP, Wang HY. Characterization & Evaluation of Silk protein hydrogels for drug delivery; Chem Pharm Bull 2006; 44(55):373-377.
29. Masteikova R, Chalupova Z, Sklubalova Z. Stimuli-sensitive hydrogels in controlled and sustained drug delivery; MEDICINA 2003; 39:19-24.
30. Eag CM, Kandukuri JM, Allenki V, Yamsani MR. In-situ gels -a novel approach for ocular drug delivery; Der Pharmacia Lettre 2009;1(1):21-33.
31. Zignani M, Tabatabay C, Gurny R. Topical semi-solid drug delivery: kinetics and tolerance of ophthalmic hydrogels; Adv Drug Deliv Rev 1995; 16:51-60.
32. Lin C, Metters AT. Hydrogels in controlled release formulations: Network design and mathematical modelling. Adv Drug Deliv Rev 2006; 58:1379–1408.
33. Gariepy ER, Leroux GC. In situ-forming hydrogels—review of temperaturesensitive systems; Eur J Pharm Biopharm 2004; 58:409–426.
34. Tomme SRV, Storm G, Hennink EW. *In situ* gelling hydrogels for pharmaceutical and biomedical application; Int J Pharm 2008; 355:1–18.