Expert Opinion

1. Introduction
2. Structure of the eye
3. Structure of the retina
4. Drug delivery to the retina
5. Transporters and transporter-mediated drug delivery
6. Conclusion and expert opinion

For reprint orders, please contact:
reprints@ashley-pub.com

Ashley Publications
www.ashley-pub.com

Delivery

Drug delivery to the retina: challenges and opportunities

Sridhar Duvvuri, Soumyajit Majumdar & Ashim K Mitra†
†Division of Pharmaceutical Sciences, School of Pharmacy, University of Missouri-Kansas City, Kansas City, Missouri, 64110-2499, USA

Retinal drug delivery is a challenging area in the field of ophthalmic drug delivery. An ideal drug delivery system for the retina and vitreous humor has not yet been found, despite extensive research. Drug delivery to retinal tissue and vitreous via systemic administration is constrained due to the presence of a blood–retinal barrier (BRB) which regulates permeation of substances from blood to the retina. Although intravitreal administration overcomes this barrier, it is associated with several other problems. In recent years, transporter targeted drug delivery has become a clinically significant drug delivery approach for enhancing the bioavailabilities of drug molecules with poor membrane permeability characteristics. Various nutrient transporters, which include peptide, amino acid, folate, monocarboxylic acid transporters and so on, have been reported to be expressed on the retina and BRB. Prodrug derivatisation of drug molecules which target these transporters could result in enhanced ocular bioavailability. Highlighted in this review are various strategies currently employed for drug delivery to the posterior chamber, and novel opportunities that can be exploited to enhance ocular bioavailability of drugs.

Keywords: blood–retinal barrier, carrier-mediated transport, drug delivery, ocular bioavailability, retina, retinal pigmented epithelium, transporters

Expert Opin. Biol. Ther. (2003) 3(1):45-56

1. Introduction

Drug delivery to the posterior segment of the eye, comprising the retina, choroid, vitreous humor and sclera, is one of the most interesting and challenging tasks facing the pharmaceutical scientist. The choroid is richly perfused with blood vessels lined by endothelial cells which allow easy exchange of molecules between blood and choroid. Such rapid drug uptake allows effective systemic therapy of infections affecting the choroid, such as choroiditis. However, diseases commonly affecting the retina, which include bacterial endopthalmitis and cytomegaloviral (CMV) retinitis, may lead to severe sight threatening pathologies, particularly due to poor penetration of drug molecules into the retina. Two main components of the barrier that prevent entry of molecules from the systemic circulation into the ocular compartments are the blood–retinal barrier (BRB) in the posterior segment, and the blood–aqueous barrier (BAB) in the anterior segment of the eye. Together, the BRB and BAB form the blood–ocular barrier. The BRB prevents free passage of xenobiotics from the choroid into the retina and vitreous. Circumventing this barrier in an efficient way, with no or minimal damage to the tissue, is a significant challenge to an ocular drug delivery scientist. Passage of drugs from the anterior segment to the posterior segment would not be efficient because of the continuous drainage of aqueous humor. Thus, overcoming the BAB may not be an effective approach to enhance drug concentrations in the retina and vitreous. Conventional ophthalmic dosage forms, that is, solutions, ointments and creams, are unable to provide us with an answer to the problem of drug delivery to the retina and vitreous. Development of newer dosage

2003 © Ashley Publications Ltd ISSN 1471-2598

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 1 of 13

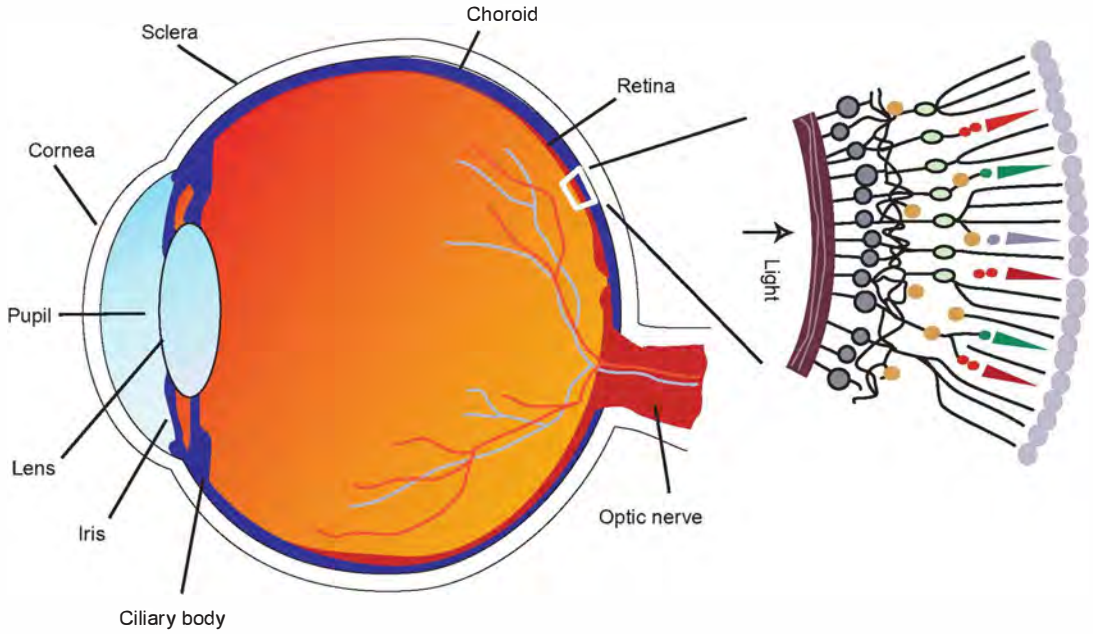


Figure 1. Cross-sectional view of the eye.

forms, like intravitreal injections, implants, subconjunctival implants and so on, have had only limited success. To understand the barriers for drug delivery to the retina, a closer look at the structure and anatomy of the eye and retina is necessary.

2. Structure of the eye

A cross-sectional view of the eye depicts three different layers:

- external layer formed by cornea and sclera
- intermediate layer divided into two parts: anterior (iris–ciliary body) and posterior (choroid)
- internal layer: retina (**Figure 1**)

The eye contains the following fluid chambers:

- aqueous humor – between the cornea and iris
- vitreous humor – between the lens and retina

The globe is held in position in the orbital cavity by various ligaments and muscles. The conjunctiva is a protective layer which covers the eyeball, and is in contact with the outer environment. It consists of a thin mucous membrane layer in the inside of the eyelids and anterior sclera. In common parlance, the anterior segment refers to the cornea, aqueous humor and iris–ciliary body, whereas the vitreous humor, retina, choroid and sclera together comprise the posterior segment of the eye.

So far, a bulk of research activity has been devoted to improving drug penetration into the tissues and fluids of the anterior segment, following systemic and topical administrations. Few such investigations have been carried out on the posterior segment. With the advent of diseases like CMV retinitis, bacterial and fungal endophthalmitis, and diabetic and proliferative vitreal and retinal pathologies, achieving effective drug levels in the retina/choroid and vitreous has gained high clinical significance. Despite the developments achieved in the area of drug delivery to the posterior segment to date, further delineation in drug transport mechanisms and development of novel delivery systems targeted to the retina and vitreous are necessary. A close examination of the local physiology and anatomy of the posterior segment of the eye, particularly the retina and the BRB, will be of importance in understanding the current and new approaches to deliver drugs to the retina and vitreous humor.

3. Structure of the retina

The retina is a richly perfused tissue with a complex cellular organisation. It is a multi-layered membrane of neuro-ectodermal origin, occupying the internal space of the posterior portion of the eye wall. The retina can be divided into two sections: neural retina and retinal pigmented epithelium (RPE). The early optic vesicle invaginates to form a cup, the inner layer of which becomes the neurosensory retina and the outer layer of which becomes the RPE. Subretinal space separates the neural retina from the RPE.

The neural retina is a seven-layered structure (**Figure 2**) which is involved in signal transduction. Light enters the retina through the ganglion cell layer (GCL) and must penetrate all the cell types before reaching the rods and cones. The transduced signal is then conducted out of the retina by various neuronal cells to the optic nerve, which subsequently conducts the signal to the brain, where it is registered and an image is formed.

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 2 of 13

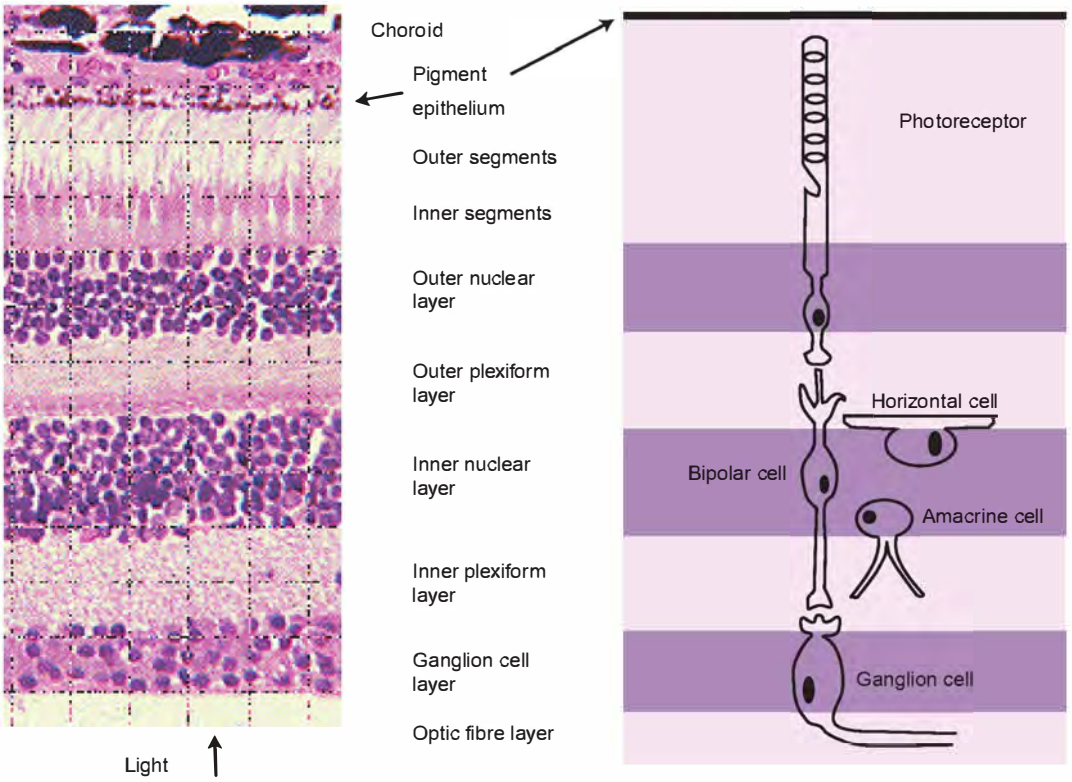


Figure 2. Cross section view of the retina.

The RPE, on the other hand, is a single cell layer structure that separates the outer surface of the neural retina from the choroid [1] and appears as an uniform and continuous layer extending through the entire retina, playing a vital role in supporting and maintaining the viability of the neural retina [2]. The RPE cells are differentiated into an apical portion, which faces the neural retina, and a basolateral portion, facing the choroid. The apical portion contains numerous microvilli, while the basolateral side shows small, convoluted infoldings that increase the surface area for absorption of nutrients from the choroid side. RPE cells are polarised, expressing specific nutrient transport systems on the apical and basolateral sides which maintain the environment of the neural retina [2].

The neural elements of the retina are separated from the blood supply at two levels. An outer level, where the RPE separates the entire neural retina from the vascular layers of the choroid, and an inner level, where endothelial cells of the retinal blood vessels separate the neural retina from the retinal blood supply [3]. Together, these two components constitute the BRB, which regulates the diffusion of substances from the blood into the vitreous and retina.

Blood vessels in the choroid possess large fenestrations that allow free diffusion of substances into and out of the choroidal stroma. Bruch's membrane (basement membrane, separating the choroid from the RPE) acts as a diffusional barrier only to macromolecules like proteins, oligonucleotides and genes. However, the RPE exhibits tight junctions, creating a barrier to the entry of small molecules into the retina from the choroidal stroma [3]. Recently, efflux pumps, such as P-glycoprotein, and multi-drug resistance (MDR) proteins (MRP) have been identified on the RPE [4,5], which may limit the permeation of various xenobiotics and endogenous compounds from the choroidal side into the vitreous. Thus, both transcellular and paracellular passage of molecules across the RPE is restricted and only selected nutrients are exchanged between the neural retina and the choroid. These characteristics result in the formation of the outer BRB (oBRB).

Retinal capillaries may be considered as barrier capillaries. In humans, major blood vessels lie in the innermost retinal layers and can usually be found just beneath the inner limiting membrane (a layer formed by neural retinal cells forming a diffusional barrier between the vitreous and the retina), although in a few animals the capillaries can be observed as overlying the retina, in contact with the vitreous. The ultrastructure of retinal blood capillaries is uniform throughout the retina and consists of a continuous endothelium surrounded by a thick basement membrane. Unlike capillary endothelium in other tissues, retinal endothelial vessels do not exhibit signs of fenestrations, and express tight junctions in a conserved manner, surrounding every endothelial cell [3].

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 3 of 13

Astrocytes are reported to be present in close apposition with the retinal blood vessels, similar to brain capillaries that constitute the blood–brain barrier [6]. A combination of these cellular structures and functions may cause retinal blood vessel endothelial cells to form the inner BRB (iBRB). The recent discovery of a multi-drug efflux pump, P-gp, on the retinal endothelium, adds to the barrier properties of iBRB [7].

Thus, both RPE and retinal endothelial blood vessels collectively form the BRB and act in conjunction to prevent unrestricted exchange of molecules between the systemic circulation and the posterior chamber of the eye.

4. Drug delivery to the retina

Major pathologies affecting the retina include fungal and bacterial endophthalmitis, CMV retinitis and diabetic, glaucomatic and proliferative vitreal retinopathy. Local drug delivery to the posterior segment of the eye, particularly to the retina, is a challenging task due to the presence of the BRB. Various strategies have been employed to deliver drugs to the retina. A transient increase in BRB permeability has been attempted by transient modification of the barrier properties of the RPE and endothelial cells of the retinal blood vessels by intracarotid infusion of a hyperosmotic solution, such as mannitol or arabinose. However, osmotic gradient methods result in non-specific opening of the BRB, and are associated with retinal and nervous system toxicity [8,9]. Chemical modification of drug molecules is more commonly employed to overcome the BRB. This strategy involves the use of lipophilic prodrugs of therapeutic entities, so as to increase their transcellular diffusion into the neural retina [10].

Drug levels attained in the posterior segment of the eye also depend on the mode of administration of the drug. The commonly employed techniques of delivering drugs to the retinal tissue include systemic and topical administration, intravitreal injections, local injections and ocular implants.

4.1 Systemic administration

Systemic administration of drugs to treat ocular conditions has been attempted by various researchers. However, only 1 – 2% of plasma drug concentration is achieved in the vitreous humor. Such low levels warrant frequent administrations of high doses, in order to maintain the drug levels in the ocular tissues above the minimum therapeutic concentrations. In the treatment of acute HCMV (human cytomegalovirus) retinitis, intravenous therapy includes foscarnet sodium or ganciclovir (GCV) i.v. [11]. The advantage of systemic administration is that it controls the spread of infection to other tissues. Disadvantages, which outweigh the advantages of intravenous therapy, include nonspecific absorption by other tissues, which could result in serious toxicities. Taking both advantages and disadvantages into consideration, systemic administration of drugs for the treatment of ocular pathologies is not an ideal strategy.

4.2 Topical route of administration

Topical applications, like ophthalmic solutions and ointments, are efficient drug delivery systems for the anterior segment, which includes the aqueous humor, iris–ciliary body and lens. For a drug to enter the posterior segment upon topical administration, the drug has to traverse through the cornea, aqueous humor and lens. Precorneal constraints include solution drainage, tear turnover and conjunctival absorption. Typically, < 1% of topically applied dose gets absorbed into the aqueous humor [12]. Drug elimination from the anterior segment mainly occurs by the canal of Schlemn or via absorption through the ciliary body and/or via the suprachoroid into the episcleral space [12].

Enzymatic metabolism also takes place resulting in further loss of drug in the anterior chamber before it reaches the posterior segment. Clearly, barriers towards attainment of therapeutic drug levels in the posterior segment following topical administration are formidable and may not be easily overcome.

However, a recent report suggests that therapeutic levels of α-2 adrenergic agent, brimonidine, are achieved in the posterior segment upon topical instillation into rabbit eyes. The drug is indicated in glaucomatous neuropathy for its neuroprotective action and promotion of retinal ganglion cell survival [13]. The authors reported that therapeutic levels of brimonidine were achieved in the retina/choroid and vitreous upon topical instillation of the drug. Vitreal levels confirm effective permeation of the drug across the BRB. Levels achieved in the vitreous humor and retina are relevant for neuroprotection.

Siegner *et al.* [14] reported the entry of verapamil into the posterior segment tissues upon topical instillation in rabbits. The drug concentrations achieved were in the following order: aqueous humor > vitreous humor > serum upon topical administration.

However, many therapeutic agents, including those indicated in treatment of infectious diseases like CMV retinitis and endophthalmitis, do not reach minimum therapeutic levels upon topical instillation.

4.3 Intravitreal injections

Over the past 20 years, intravitreal injections have become a mainstay treatment of posterior segment infections/diseases. Ocular antivirals like GCV, acyclovir (ACV), cidofovir and foscarnet, and antibiotics like cephalexin, cefazolin, ceftazidime, gentamicin and so on, have been administered intravitreally to assess their pharmacokinetic parameters so that proper dose and dosing frequency can be designed to effectively treat CMV retinitis and endophthalmitis [15-20].

Penetration studies of various drugs, particularly antibiotics, from the blood into the ocular fluids, have confirmed that penetration into the vitreous is much more restricted than penetration into the aqueous humor, probably by one order of magnitude [3]. Macha and Mitra [21] studied the penetration of a paracellular marker, fluorescein, into the aqueous and vitreous humors of rabbits. They reported that a tighter barrier surrounds the vitreous, in comparison to aqueous humor. Only

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 4 of 13

about 1 – 2% of the plasma levels of fluorescein can be detected in the vitreous humor. Thus, in order to achieve sufficiently high drug levels in the retina and vitreous, higher plasma levels are required. In an attempt to increase the plasma levels, clinicians increased the amount of dose administered, or administered the drugs by rapid intravenous infusions [22,23]. Even though these procedures resulted in increased ocular levels, long-term systemic toxicity concerns remain [24,25].

Intravitreal injection offers various advantages over systemic and topical routes for drug delivery to the retina and vitreous. It may result in increased drug levels in the posterior segment without causing any systemic side effects. Therapeutic levels may be maintained with lower doses as this route of administration circumvents the BRB. Intravitreal injections are a mainstay therapy in severe cases of endophthalmitis and CMV retinitis [15,26,27].

Intravitreal injections suffer from several major deficiencies, such as patient non-compliance, need for repeated injections (once a week), injection-associated infections like endophthalmitis and retinal detachment, and so on [28-31]. To overcome some of these shortcomings, researchers have attempted to increase the drug residence time in the vitreous, following a single injection. Intravitreal injection of liposomes, microparticles and lipophilic ester prodrugs has been tried and met with mixed results [10,29,32-35]. Administration of particulate substances carries the risk of infection and blurred vision [36]. Macha and Mitra [10] studied the vitreous disposition of GCV and its various monoester prodrugs. The GCV monoester prodrugs generated sustained GCV levels in the vitreous for longer time periods than those from direct injection of the parent drug. Despite all the reported advantages, intravitreal injections are not well-tolerated, due to the associated pain and discomfort.

4.4 Intravitreal implants

Direct intraocular injections overcome the problem of high intravenous dose-related toxicity. However, multiple injections are associated with an increased risk of cataract, astigmatism, endophthalmitis, retinal detachment and vitreous haemorrhage [31]. Intravitreal implants have been developed as an alternative to multiple intravitreal injections. Vitrasert® (Chiron Vision Corp., CA, USA) is a non-biodegradable intraocular GCV implant. It is surgically inserted in the posterior segment of the eye, where it delivers GCV locally over a period of 5 – 8 months. Once the device is depleted of drug, it is replaced with a new device. Data from Chiron Vision Phase III trial demonstrated that disease progression was significantly delayed in patients implanted with Vitrasert compared to patients receiving intravenous doses of GCV [201]. However, the risks associated with intravitreal injections are not completely eliminated. Adverse reactions reported for Vitrasert include loss of visual acuity, vitreous haemorrhage, retinal detachment, cataract formation, lens opacities, macular abnormalities, intraocular pressure spikes, optic disk/nerve changes, hyphemas, uveitis and acute or delayed onset endophthalmitis [37]. Also, repeated surgeries are required for implantation and removal of the implant. Like intravitreal injections, intravitreal implants release drug locally into the vitreous humor and may not be able to stem the spread of infections to the contralateral eye. Thus, systemic administration is recommended in conjunction with intravitreal injections and implants.

4.5 Scleral drug delivery

A major risk associated with intravitreal injections and implants is retinal detachment, which generally occurs as a result of retinal piercing due to surgery or multiple injections. To overcome this injury to the retina, researchers have investigated sustained release devices implanted into the sclera. Previous work has revealed that the sclera does not constitute a significant barrier towards diffusion of drug molecules into the vitreous chamber. It appears to be five times more permeable than the cornea to hydrocortisone [38]. Moreover, the primary mechanism of transscleral drug transport occurs by passive diffusion through the intercellular aqueous pore pathway. As a result, molecular size appears to be the controlling factor in drug transport, rather than lipophilicity of the molecule [38]. Scleral drug delivery to the posterior chamber of the eye has been attempted in two ways:

- scleral plugs and implants
- subconjunctival injections

4.5.1 *Scleral plugs and implants*

To lower the surgery-related risk, biodegradable devices have been developed by Sakurai *et al.* [37]. The inserts were prepared from poly (DL-lactide-co-glycolide) (PLGA) and poly (DL-lactide) (PLA) polymers. Inserts were active up to a period of 3 weeks in HCMV inoculated eyes, and the vitreo-retinal lesions were reduced in comparison to single intravitreal injections of GCV [37]. In another study, various blends of PLA and PLGA were examined for their suitability as vitreal implants [39]. PLA-70,000 and 5000 (80/20) resulted in sustained GCV release for a period of 24 weeks, with drug levels maintained well above the minimum therapeutic levels during the entire study period, without any incidence of sudden burst effect.

In conclusion, sustained drug release could be achieved with scleral implants. The main disadvantages of these implants are repeated operations and patient discomfort due to the presence of a foreign body in the eye.

4.5.2 *Subconjunctival injections*

Subconjunctival injections may be employed to target drugs to the posterior segment of the eye. The primary absorption surfaces available are the sclera and conjunctiva. Sasaki *et al.* [40] studied absorption of tilisolol, a β-blocker, into ocular structures and plasma upon corneal, conjunctival and scleral applications. Scleral application resulted in higher intraocular levels relative to conjunctival application. Reverse was true for plasma concentrations, that is, higher plasma levels of the drug were observed upon conjunctival application. From this study, it may be concluded that conjunctival

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 5 of 13

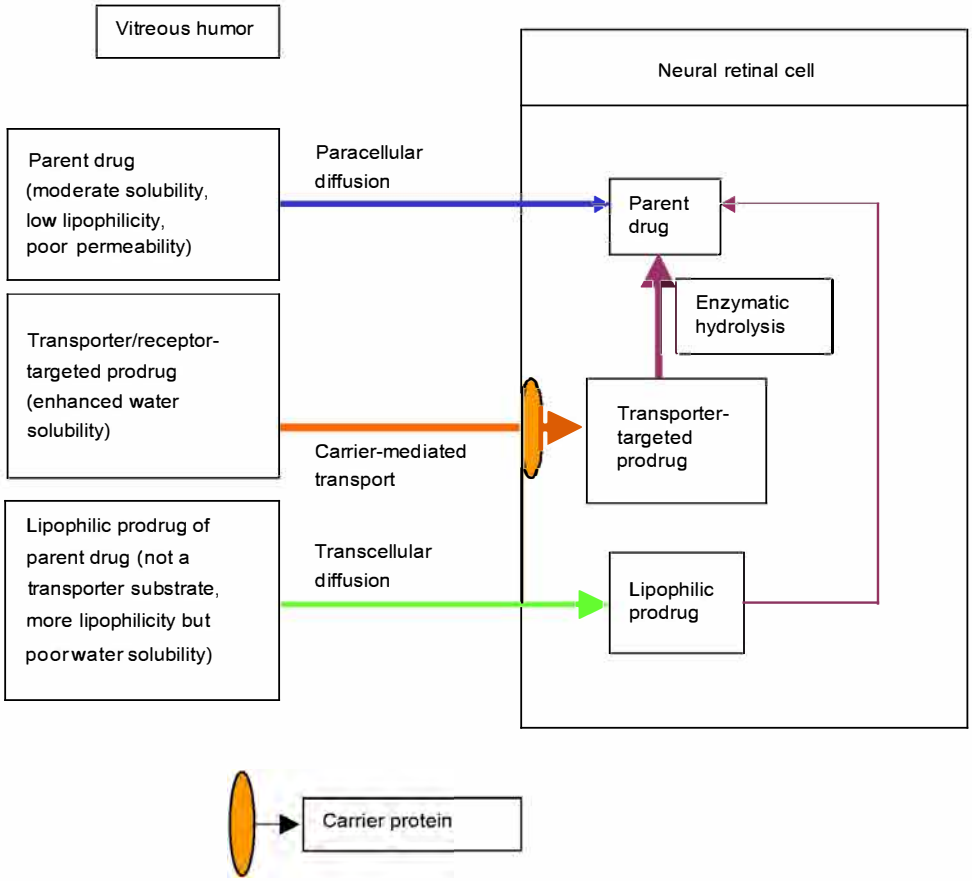


Figure 3a. Schematic representation of retinal drug levels following intravitreal administration of the parent drug, lipophilic prodrug and transporter targeted prodrug.

absorption results in drainage of the drug into systemic circulation. As a consequence, scleral application results in increased drug levels in intraocular tissues, relative to conjunctival administration. Kalsi *et al.* [41] studied ocular pharmacokinetics of an anticancer drug, dacarbazine, following systemic and subconjunctival administrations. Elevated levels of dacarbazine were observed in aqueous and vitreous humors upon subconjunctival injections, relative to intravenous injections.

An attempt was made to achieve sustained drug release upon subconjunctival injections of a polymer-based ophthalmic formulation. Polymers usually employed for such purposes must be biocompatible and biodegradable. Such a material should not induce or cause any inflammatory reactions in the sensitive ocular tissues. The polymer should erode or degrade in a controlled manner such that a zero order release profile can be attained over a prolonged period. Polymers employed in ocular drug delivery include alginates [42], pluronics and poloxamers [43], polyorthoesters [44,45] and polyvinyl alcohol [46]. Efficacy of these polymers in providing sustained ocular drug levels following subconjunctival administration is still under investigation.

In spite of recent progress, retinal drug delivery is still challenging, due to factors like drug lipohilicity, presence of MDR gene products and drug ionisation. In conditions like CMV retinitis, drug entry into retinal cells could be restricted due to their polar and hydrophilic properties (GCV, cidofovir, foscarnet). Membrane transporters have been targeted to gain cellular entry by an active transport process. This approach has met with considerable success in enhancing permeation of several molecules across biological membranes. This strategy can be employed to increase the intracellular drug concentrations in the retina following systemic, intravitreal and transscleral administration. A brief discussion on transporters and transporter-mediated drug delivery is presented in the following sections, prior to the discussion of its application to retinal drug delivery.

5. Transporters and transporter-mediated drug delivery

Transporter-mediated drug delivery involves targeting of drug molecules to the membrane transporters, in order to

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 6 of 13

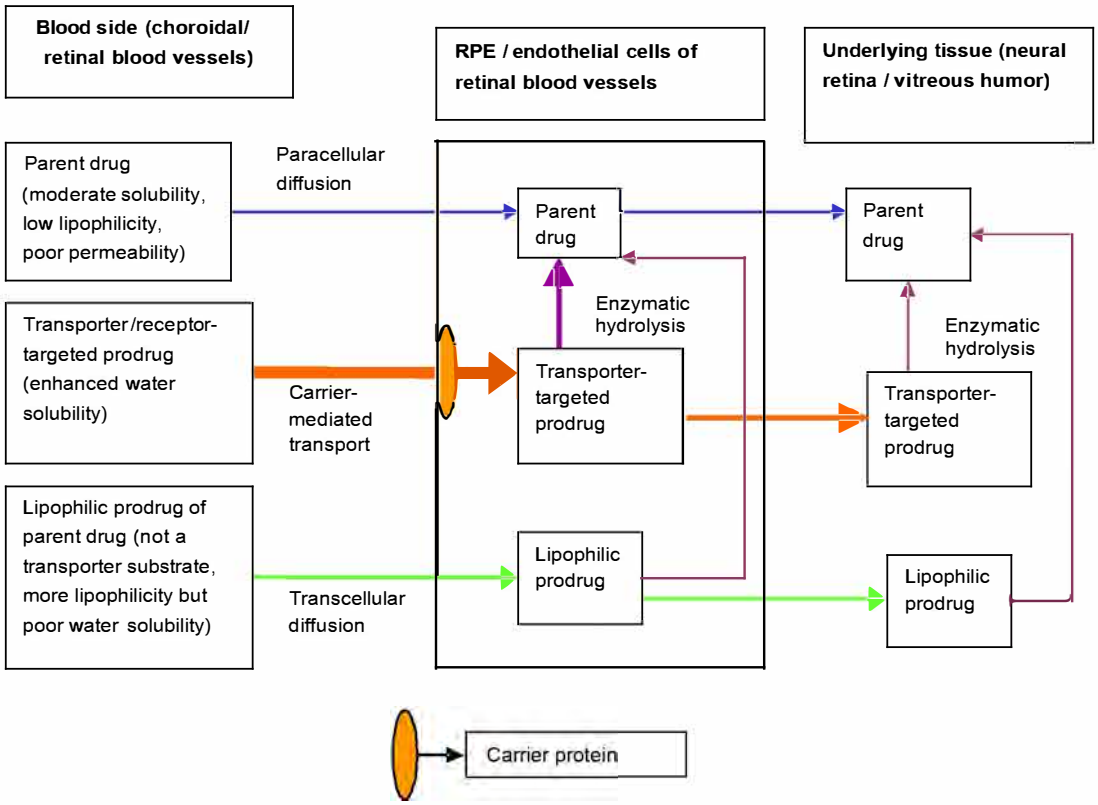


Figure 3b. Schematic representation of retinal drug levels following systemic administration of the parent drug, lipophilic prodrug and transporter-targeted prodrug.
RPE: Retinal pigmented epithelium.

produce effective passage across the cell membrane. Various nutrient transporters are usually targeted, such as peptide, amino acid, monocarboxylic acid, folate, nucleoside and nucleobase, organic anion and organic cation transporters and so on [47]. Parent drugs can be targeted to the membrane transporters by coupling with a promoiety that is an endogenous substrate for the same transporter. The promoiety, is recognised by the transporter, which results in translocation of the entire prodrug molecule across the cell membrane. The parent drug is usually released intracellularly by enzymatic cleavage of the ligand–drug bond. As the ligand is a nutrient, no toxicity concerns arise from the release of the leaving group. This strategy (**Figures 3a** and **3b**) can be employed to target drugs to transporters expressed on the neural retina and RPE, following intravitreal and systemic administration, respectively. Transporter-targeted prodrug design has the advantage of higher solubility and the ability to administer higher doses of prodrugs, when compared with conventional lipophilic prodrug design.

Various nutrient transporters expressed on the retina include peptide [48], glucose [49] and amino acids such as glycine, GABA, glutamine, taurine, arginine, proline, tryptophan [50] and nucleoside-like adenosine [51]. Amongst the transporters identified, the few that could be targeted to enhance retinal drug delivery are discussed in the following sections.

5.1 Peptide transporter

Peptide transporters (PepTs) have been identified on various epithelia [52-58]. Considerable information is available on their mechanism and substrate specificity [59-65]. These transporters, that is, PepT1 and PepT2, have broad substrate specificity, particularly for smaller peptides like di- and tripeptides. Through prodrug modification, a drug molecule may be converted to a substrate for this transporter to be easily ferried across lipophilic membranes [64]. At the same time, the solubility of the compound may be enhanced, depending on the type of amino acids selected in the peptide promoiety. Following its transport, the enzymes, specifically the cholinesterases, dipeptidases and aminopeptidases, present in the vitreal and retinal tissue may cleave the prodrug to regenerate the parent drug. Peptide prodrugs of nucleosides like GCV, zidovudine and ACV, became substrates of PepTs and resulted in a significant increase in the bioavailability of the parent drugs upon oral administration [65,66].

Studies have established the presence of a PepT on the retina side facing the vitreous [48] and on the RPE side facing the

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 7 of 13

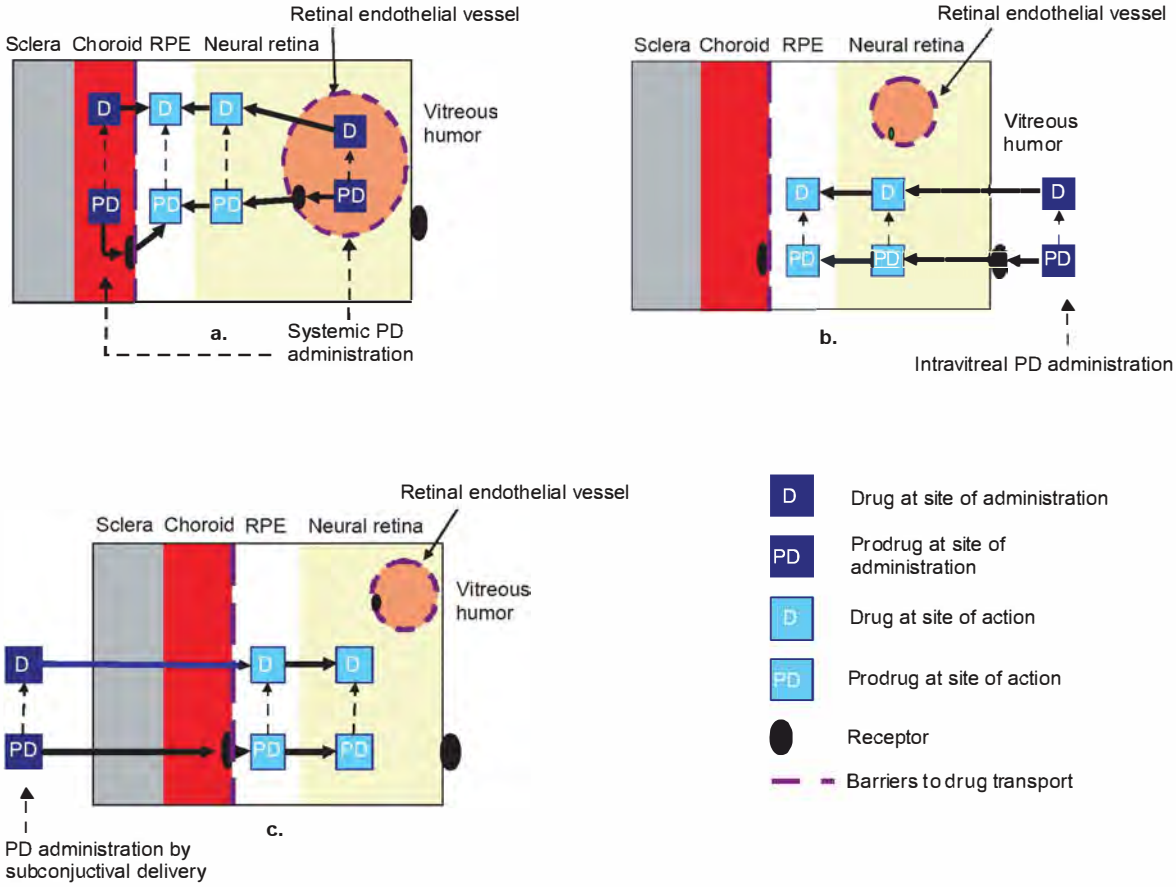


Figure 4. Strategies involved in enhancing drug delivery to the posterior segment by transporter-mediated drug delivery.
a. Transporter-targeted drug delivery following systemic administration. **b.** Transporter-targeted drug delivery following intravitreal administration. **c.** Transporter-targeted drug delivery following transscleral/subconjuctival administration.
D: Drug; PD: Prodrug; RPE: Retinal pigmented epithelium.

blood [67]. The transporter facing the vitreous could be targeted to increase the intracellular drug concentrations following intravitreal administration. On the other hand, targeting to the PepT present on the RPE can enhance retinal and vitreal drug concentrations following systemic administration.

5.2 Amino acid transporters

Amino acid transporters on the blood–brain barrier and intestinal mucosa have been extensively studied [68-73]. These transporters are very substrate specific and lead to the passage of only specific amino acids. Various amino acid transporters, including glutamate, glycine, GABA, tryptophan and proline, have been identified on the retina [50]. Expression of such transport systems on the retina or RPE once again opens up the opportunity of designing prodrugs targeted towards these carrier proteins.

5.3 Monocarboxylic acid transport systems

Mammalian cells take up and excrete lactate, pyruvate and other monocarboxylic acids by means of proton-coupled monocarboxylic acid transporters (MCTs) [74-80]. Various MCT isoforms (MCT1 – 9) have been identified [81]. Monocarboxylic drugs like salicylic acid and pravastatin are reported to be transported via MCTs in the small intestine [82-84]. In the eye, MCTs are expressed on the retina and conjunctiva [85,86]. MCT1 has been reported as present on the apical membrane, and MCT3 on the basolateral membrane, of rat RPE [87]. MCT can thus ferry substrates (carboxylate drugs and prodrugs) from the blood/choroid into the retina, following intravenous and transscleral administrations.

5.4 Folate transport system

Two types of transport process have been identified for folate entry into mammalian cells. One process involves the folate receptors that bind and internalise the bound folate via receptor-mediated endocytosis. A second process utilises the reduced folate transporter, which is a typical transporter protein with multiple membrane spanning domains. The protein interacts with the reduced folate more efficiently than folate

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 8 of 13

itself [88-92]. Folate receptors have been targeted for facilitating anticancer drug entry into cancer cells and also for liposome based gene delivery [93]. Therefore, targeting the folate transporter could result in enhanced drug levels in the vitreous. Since the folate receptor is generally absent in most normal tissues, with the exception of the choroid plexus and placenta, or expressed at very low levels (lung, thyroid and kidney) [93], it may be possible to achieve targeted delivery of the folate ester prodrugs to the retina, following intravenous administration.

5.5 Strategies to improve ocular bioavailability by transporter-mediated drug delivery

Expression of transporters on the retina, particularly the RPE and endothelial cells of the retinal blood vessels, provides us with an opportunity to increase the retinal and vitreal levels of various drugs, thereby increasing their efficacy and decreasing the required dose. The strategies that can be employed to target these transporters on the retina are illustrated in **Figure 4**. Compounds can be targeted to the transporter by delivery through any of the three main routes: i) systemic, ii) intravitreal and iii) subconjunctival.

5.5.1 Systemic administration of prodrugs

As discussed earlier, vitreal drug levels achieved upon systemic administration are around 1 – 2% of plasma levels. If a drug is targeted to a transporter expressed on the BRB, it could result in enhanced concentrations in the retina. Prodrug cleavage in the retinal cells may also generate drug levels in the vitreous humor. The strategy is shown in **Figure 4a**.

The main disadvantage with this strategy is that drug uptake will be nonspecific, due to possible distribution of the transporter at multiple tissues.

5.5.2 Intravitreal administration

In order to avoid the nonspecific absorption of drugs into all the tissues and to avoid systemic toxicity, intravitreal administration of prodrugs may be justified. **Figure 4b** describes this strategy where the prodrug is injected directly into the vitreous humor, thereby targeting it to the transporter expressed on the retinal cell membranes. Prodrug uptake by the neural retina and RPE cells may be more efficient than the parent drug itself.

5.5.3 Subconjunctival administration

Subconjunctival administration usually results in higher vitreal concentrations than systemic administration. Moreover, subconjunctival injections of particulate dosage forms may result in sustained drug release over a prolonged period of time, thereby decreasing the frequency of administration. This route may be used to deliver prodrugs targeted to specific transporters expressed on the basolateral side of the RPE. Following subconjunctival administration, the prodrug first diffuses into the sclera and then into the choroidal circulation, where it interacts with transporters expressed on the RPE. These transporters will then carry the prodrug into the retinal tissue, where it is cleaved into the parent drug. This mechanism is depicted in **Figure 4c**. If the drug is incorporated into a polymeric vehicle, which controls the release of the prodrug, a sustained delivery of the drug to the retina and vitreous humor may be achieved.

6. Conclusion and expert opinion

Drug delivery to the retina has always been a challenge to ophthalmologists. The complex physiology of the retina, coupled with low blood flow, makes it even more difficult. Several routes of administration, including systemic, topical, intravitreal and scleral, have been attempted and have met with varying degrees of success.

The past decade has witnessed rapid developments in the field of membrane transporters and their applications to drug delivery. Many therapeutic entities have utilised these transporters for efficient absorption into systemic circulation from the intestine. Carrier-mediated drug delivery is important for compounds that ionise at physiological conditions and permeate the biological barriers poorly. Drugs like valacyclovir and valganciclovir are success stories for transporter-mediated drug delivery.

Various transporters have been identified and characterised on the retina. Not much work has been done in the area of drug delivery to the retina and posterior segment of the eye by utilising membrane transporters. Targeting these transporters can enhance ocular bioavailability of highly potent, but poorly bioavailable compounds. A small increase in the bioavailability of such drug molecules could result in a significant increase in therapeutic response. These transporters can be targeted by various modes of administration, including intravitreal, intravenous and transscleral modes. Transscleral targeting of the transporters expressed on the RPE could avoid many of the drawbacks and side effects of systemic and intravitreal administration. Moreover, prodrug derivatisation of the parent molecule could significantly increase the solubility and enable administration of higher doses. Further research into this aspect of retinal drug delivery could result in promising results.

In conclusion, drug delivery to the retina and posterior segment of the eye can be significantly enhanced by targeting drugs to the membrane transporters expressed on the ocular tissues. This approach could solve many of the problems that ophthalmologists encounter during the treatment of diseases of the posterior segment of the eye.

Acknowledgements

Supported by National Institute of Health grants R01 EY 09171-08 and R01 10659-07.

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 9 of 13

Bibliography

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

1. RIZZOLO LJ: Polarity and the development of the outer blood-retinal barrier. *Histol. Histopathol.* (1997) **12**:1057-1067.
2. MARMOR MF: Structure, function and disease of the retinal pigment epithelium, in: *The Retinal Pigment Epithelium*. Wolfensberger TJ, Marmor MF (Eds.) Oxford University Press. New York, USA (1998):3-9.
3. CUNHA-VAZ JG: The blood-retinal barriers. *Doc Ophthalmol.* (1976) **41**:287-327.

•• **Detailed review of anatomy and physiology of the BRB.**

4. AUKUNURU JV, SUNKARA G, BANDI N, THORESON WB, KOMPELLA UB: Expression of multidrug resistance-associated protein (MRP) in human retinal pigment epithelial cells and its interaction with BAPSG, a novel aldose reductase inhibitor. *Pharm. Res.* (2001) **18**:565-572.
5. KENNEDY BG, MANJINI NJ: Native and Cultured Human RPE Express P-glycoprotein. *ARVO Annual Meeting, 2002*. (2002).
6. HOLASH JA, STEWART PA: The relationship of astrocyte-like cells to the vessels that contribute to the blood-ocular barriers. *Brain Res* (1993) **629**:218-224.
7. GREENWOOD J: Characterization of a rat retinal endothelial cell culture and the expression of P-glycoprotein in brain and retinal endothelium *in vitro*. *J. Neuroimmunol.* (1992) **39**:123-132.
8. MILLAY RH, KLEIN ML, SHULTS WT, DAHLBORG SA, NEUWELT EA: Maculopathy associated with combination chemotherapy and osmotic opening of the blood-brain barrier. *Am. J. Ophthalmol.* (1986) **102**:626-632.
9. FRANK JA, DWYER AJ, GIRTON M *et al.*: Opening of blood-ocular barrier demonstrated by contrast-enhanced MR imaging. *J. Comput. Assist. Tomogr.* (1986) **10**:912-916.
10. MACHA S, MITRA AK: Ocular disposition of ganciclovir and its monoester prodrugs following intravitreal administration using microdialysis. *Drug Metab. Dispos.* (2002) **30**:670-675.
11. COSTABILE BS: Treatment of cytomegalovirus retinitis with intraocular implants. *AORN J.* (1998) **67**:356-360,362-365,368; quiz 369-372.
12. HUGHES PM, MITRA AK: Overview of Ocular Drug Delivery, in: *Drug Delivery Systems*, Mitra AK (Ed.), Marcell Dekker, Inc., New York, USA (1993).
13. ACHEAMPONG AA, SHACKLETON M, JOHN B *et al.*: Distribution of brimonidine into anterior and posterior tissues of monkey, rabbit, and rat eyes. *Drug Metab. Dispos.* (2002) **30**:421-429.
14. SIEGNER SW, GIOVANONI RL, ERICKSON KA, NETLAND PA: Distribution of verapamil and norverapamil in the eye and systemic circulation after topical administration of verapamil in rabbits. *J. Ocul. Pharmacol. Ther.* (1998) **14**:159-168.
15. MACHA S, MITRA AK: Ocular pharmacokinetics of cephalosporins using microdialysis. *J. Ocul. Pharmacol. Ther.* (2001) **17**:485-498.
16. BEN-NUN J, JOYCE DA, COOPER RL, CRINGLE SJ, CONSTABLE IJ: Pharmacokinetics of intravitreal injection. Assessment of a gentamicin model by ocular dialysis. *Invest. Ophthalmol. Vis. Sci.* (1989) **30**:1055-1061.
17. LOPEZ-CORTES LF, PASTOR-RAMOS MT, RUIZ-VALDERAS R *et al.*: Intravitreal pharmacokinetics and retinal concentrations of ganciclovir and foscarnet after intravitreal administration in rabbits. *Invest. Ophthalmol. Vis Sci.* (2001) **42**:1024-1028.
18. CUNDY KC, LYNCH G, SHAW JP, HITCHCOCK MJ, LEE WA: Distribution and metabolism of intravitreal cidofovir and cyclic HPMPC in rabbits. *Curr Eye Res* (1996) **15**:569-576.
19. HUGHES PM, KRISHNAMOORTHY R, MITRA AK: Vitreous disposition of two acycloguanosine antivirals in the albino and pigmented rabbit models: a novel ocular microdialysis technique. *J. Ocul. Pharmacol. Ther.* (1996) **12**:209-224.
20. WAGA J, NILSSON-EHLE I, LJUNGBERG B *et al.*: Microdialysis for pharmacokinetic studies of ceftazidime in rabbit vitreous. *J. Ocul. Pharmacol. Ther.* (1999) **15**:455-463.
21. MACHA S, MITRA AK: Ocular pharmacokinetics in rabbits using a novel dual probe microdialysis technique. *Exp Eye Res.* (2001) **72**:289-299.
22. LALEZARI JP, FRIEDBERG DN, BISSETT J *et al.*: High dose oral ganciclovir treatment for cytomegalovirus retinitis. *J. Clin. Virol.* (2002) **24**:67-77.
23. LOPEZ-CORTES LF, RUIZ-VALDERAS R, LUCERO-MUNOZ MJ *et al.*: Intravitreal, retinal, and central nervous system foscarnet concentrations after rapid intravenous administration to rabbits. *Antimicrob. Agents Chemother.* (2000) **44**:756-759.
24. HENDERLY DE, FREEMAN WR, CAUSEY DM, RAO NA: Cytomegalovirus retinitis and response to therapy with ganciclovir. *Ophthalmology* (1987) **94**:425-434.
25. JABS DA, NEWMAN C, DE BUSTROS S, POLK BF: Treatment of cytomegalovirus retinitis with ganciclovir. *Ophthalmology* (1987) **94**:824-830.
26. BAUM J, PEYMAN GA, BARZA M: Intravitreal administration of antibiotic in the treatment of bacterial endophthalmitis. III. Consensus. *Surv. Ophthalmol.* (1982) **26**:204-206.
27. DIAMOND JG: Intraocular management of endophthalmitis. A systematic approach. *Arch. Ophthalmol.* (1981) **99**:96-99.
28. MARTIN DF, SIERRA-MADERO J, WALMSLEY S *et al.*: A controlled trial of valganciclovir as induction therapy for cytomegalovirus retinitis. *N. Engl. J. Med.* (2002) **346**:1119-1126.
29. STONE TW, JAFFE GJ: Reversible bull's-eye maculopathy associated with intravitreal fomivirsen therapy for cytomegalovirus retinitis. *Am. J. Ophthalmol.* (2000) **130**:242-243.
30. BAUDOUIN C, CHASSAIN C, CAUJOLLE C, GASTAUD P: Treatment of cytomegalovirus retinitis in AIDS patients using intravitreal injections of highly concentrated ganciclovir. *Ophthalmologica* (1996) **210**:329-335.
31. CANTRILL HL, HENRY K, MELROE NH *et al.*: Treatment of cytomegalovirus retinitis with intravitreal ganciclovir. Long-term results. *Ophthalmology* (1989) **96**:367-374.
32. VELOSO AA JR, ZHU Q, HERRERO-VANRELL R, REFOJO MF: Ganciclovir-loaded polymer microspheres in rabbit eyes inoculated with human cytomegalovirus. *Invest. Ophthalmol. Vis. Sci.* (1997) **38**:665-675.

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 10 of 13

33 PEYMAN GA, KHOOBEHI B, TAWAKOL M *et al.*: Intravitreal injection of liposome-encapsulated ganciclovir in a rabbit model. *Retina* (1987) **7**:227-229.

34 DIAZ-LLOPIS M, MARTOS MJ, ESPANA E *et al.*: Liposomally-entrapped ganciclovir for the treatment of cytomegalovirus retinitis in AIDS patients. Eperimental toxicity and pharmacokinetics, and clinical trial. *Doc. Ophthalmol.* (1992) **82**:297-305.

35 HERRERO-VANRELL R, REFOJO MF: Biodegradable microspheres for vitreoretinal drug delivery. *Adv. Drug Deliv. Rev.* (2001) **52**:5-16.

36 MAURICE D: Review: practical issues in intravitreal drug delivery. *J. Ocul. Pharmacol. Ther.* (2001) **17**:393-401.

37 SAKURAI E, MATSUDA Y, OZEKI H *et al.*: Scleral plug of biodegradable polymers containing ganciclovir for experimental cytomegalovirus retinitis. *Invest. Ophthalmol. Vis. Sci.* (2001) **42**:2043-2048.

38 UNLU N, ROBINSON JR: Scleral permeability to hydrocortisone and mannitol in the albino rabbit eye. *J. Ocul. Pharmacol. Ther.* (1998) **14**:273-281.

39 YASUKAWA T, KIMURA H, TABATA Y, OGURA Y: Biodegradable scleral plugs for vitreoretinal drug delivery. *Adv. Drug Deliv. Rev.* (2001) **52**:25-36.

40 SASAKI H, ICHIKAWA M, KAWAKAMI S *et al.*: *In situ* ocular absorption of tilisolol through ocular membranes in albino rabbits. *J. Pharm. Sci.* (1996) **85**:940-943.

41 KALSI GS, SILVER HK, ROOTMAN J: Ocular pharmacokinetics of dacarbazine following subconjunctival versus intravenous administration in the rabbit. *Can. J. Ophthalmol.* (1991) **26**:247-251.

42 HATEFI A, AMSDEN B: Biodegradable injectable *in situ* forming drug delivery systems. *J. Control Release* (2002) **80**:9-28.

43 GEROSKI DH, EDELHAUSER HF: Transscleral drug delivery for posterior segment disease. *Adv. Drug. Deliv. Rev.* (2001) **52**:37-48.

• **Review of opportunities for transscleral drug delivery.**

44 EINMAHL S, SAVOLDELLI M, D'HERMIES F *et al.*: Evaluation of a novel biomaterial in the suprachoroidal space of the rabbit eye. *Invest. Ophthalmol. Vis. Sci.* (2002) **43**:1533-1539.

45 ZIGNANI M, EINMAHL S, BAEYENS V *et al.*: A poly(ortho ester) designed for combined ocular delivery of dexamethasone sodium phosphate and 5-fluorouracil: subconjunctival tolerance and *in vitro* release. *Eur. J. Pharm. Biopharm.* (2000) **50**:251-255.

46 SMITH TJ, ASHTON P: Sustained-release subconjunctival 5-fluorouracil. *Ophthalmic Surg. Lasers* (1996) **27**:763-767.

47 LEE VH: Membrane transporters. *Eur. J. Pharm. Sci.* (2000) **11**(Suppl. 2):S41-S50.

•• **Review of various transporters involved in transporter-targeted drug delivery.**

48 BERGER UV, HEDIGER MA: Distribution of peptide transporter PEPT2 mRNA in the rat nervous system. *Anat. Embryol. (Berl.)* (1999) **199**:439-449.

49 BAN Y, RIZZOLO LJ: Regulation of glucose transporters during development of the retinal pigment epithelium. *Brain Res. Dev. Brain Res.* (2000) **121**:89-95.

50 POW DV: Amino acids and their transporters in the retina. *Neurochem. Int.* (2001) **38**:463-484.

51 WILLIAMS EF, EZEONU I, DUTT K: Nucleoside transport sites in a cultured human retinal cell line established by SV-40 T antigen gene. *Curr. Eye Res.* (1994) **13**:109-118.

52 SMITH QR: Drug delivery to brain and the role of carrier-mediated transport. *Adv. Exp. Med. Biol.* (1993) **331**:83-93.

53 CHEN L, WRIGHT LR, CHEN CH *et al.*: Molecular transporters for peptides: delivery of a cardioprotective epsilonPKC agonist peptide into cells and intact ischemic heart using a transport system, R(7). *Chem. Biol.* (2001) **8**:1123-1129.

54 PUTNAM WS, PAN L, TSUTSUI K, TAKAHASHI L, BENET LZ: Comparison of bidirectional cephalexin transport across MDCK and caco-2 cell monolayers: interactions with peptide transporters. *Pharm. Res.* (2002) **19**:27-33.

55 GRONEBERG DA, DORING F, THEISS *et al.*: Peptide transport in the mammary gland: expression and distribution of PEPT2 mRNA and protein. *Am. J. Physiol. Endocrinol. Metab.* (2002) **282**:E1172-1179.

56 KRAMER W, DECHENT C, GIRBIG F, GUTJAHR U, NEUBAUER H: Intestinal uptake of dipeptides and beta-lactam antibiotics. I. The intestinal uptake system for dipeptides and beta-lactam antibiotics is not part of a brush border membrane peptidase. *Biochim. Biophys. Acta.* (1990) **1030**:41-49.

57 DANIEL H, ADIBI SA: Transport of beta-lactam antibiotics in kidney brush border membrane. Determinants of their affinity for the oligopeptide/H+ symporter. *J. Clin. Invest.* (1993) **92**:2215-2223.

58 DIECK ST, HEUER H, EHRCHEN J, OTTO C, BAUER K: The peptide transporter PepT2 is expressed in rat brain and mediates the accumulation of the fluorescent dipeptide derivative beta-Ala-Lys- Nepsilon-AMCA in astrocytes. *Glia* (1999) **25**:10-20.

59 DORING F, MARTINI C, WALTER J, DANIEL H: Importance of a small N-terminal region in mammalian peptide transporters for substrate affinity and function. *J. Membr. Biol.* (2002) **186**:55-62.

60 TERADA T, SAITO H, MUKAI M, INUI KI: Identification of the histidine residues involved in substrate recognition by a rat H+/peptide cotransporter, PEPT1. *FEBS Lett.* (1996) **394**:196-200.

61 FEI YJ, LIU W, PRASAD PD *et al.*: Identification of the histidyl residue obligatory for the catalytic activity of the human H+/peptide cotransporters PEPT1 and PEPT2. *Biochemistry* (1997) **36**:452-460.

62 KRAMER W, DURCKHEIMER W, GIRBIG F *et al.*: Influence of amino acid side-chain modification on the uptake system for beta-lactam antibiotics and dipeptides from rabbit small intestine. *Biochim. Biophys. Acta* (1990) **1028**:174-182.

63 MEREDITH D, TEMPLE CS, GUHAN *et al.*: Modified amino acids and peptides as substrates for the intestinal peptide transporter PepT1. *Eur J. Biochem.* (2000) **267**:3723-3728.

64 YANG C, TIRUCHERAI GS, MITRA AK: Prodrug based optimal drug delivery via membrane transporter/receptor. *Expert. Opin. Biol. Ther.* (2001) 1:159-175.

•• **Review of transporter-targeted drug delivery strategies.**

65 HAN H, DE VRUEH RL, RHIE JK *et al.*: 5'-Amino acid esters of antiviral nucleosides, acyclovir, and AZT are absorbed by the intestinal PEPT1 peptide transporter. *Pharm. Res.* (1998) **15**:1154-1159.

66 PESCOVITZ MD, RABKIN J, MERION RM *et al.*: Valganciclovir results in improved oral absorption of ganciclovir

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 11 of 13

in liver transplant recipients. *Antimicrob. Agents Chemother.* (2000) **44**:2811-2815.

67 ATLURI H, ANAND BS, PATEL J, MITRA AK: Carrier mediated transport of a dipeptide across blood retinal barrier following systemic administration. *AAPS Annual meeting* (2002).

68 MUNCK LK, GRONDAHL ML, THORBOLL JE, SKADHAUGE E, MUNCK BG: Transport of neutral, cationic and anionic amino acids by systems B, b(o,+), X(AG), and ASC in swine small intestine. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* (2000) **126**:527-537.

69 KILBERG MS, STEVENS BR, NOVAK DA: Recent advances in mammalian amino acid transport. *Ann. Rev. Nutr.* (1993) **13**:137-165.

70 KANAI Y, SEGAWA H, CHAIROUNGDUA A *et al.*: Amino acid transporters: molecular structure and physiological roles. *Nephrol. Dial. Transplant.* (2000) **15**:9-10.

71 FUKASAWA Y, SEGAWA H, KIM JY *et al.*: Identification and characterization of a Na(+)-independent neutral amino acid transporter that associates with the 4F2 heavy chain and exhibits substrate selectivity for small neutral D- and L-amino acids. *J. Biol. Chem.* (2000) **275**:9690-9698.

72 YANG C, MITRA AK: Nasal absorption of tyrosine-linked model compounds. *J. Pharm. Sci.* (2001) **90**:340-347.

73 HARGREAVES KM, PARDRIDGE WM: Neutral amino acid transport at the human blood-brain barrier. *J. Biol. Chem.* (1988) **263**:19392-19397.

74 ALM A, TORNQUIST P: Lactate transport through the blood-retinal and the blood-brain barrier in rats. *Ophthalmic Res.* (1985) **17**:181-184.

75 TAMAI I, SAI Y, ONO A *et al.*: Immunohistochemical and functional characterization of pH-dependent intestinal absorption of weak organic acids by the monocarboxylic acid transporter MCT1. *J. Pharm. Pharmacol.* (1999) **51**:1113-1121.

76 NORD EP, WRIGHT SH, KIPPEN I, WRIGHT EM: Specificity of the Na+-dependent monocarboxylic acid transport pathway in rabbit renal brush border membranes. *J. Membr. Biol.* (1983) **72**:213-221.

77 KANG YS, TERASAKI T, TSUJI A: Acidic drug transport *in vivo* through the blood-brain barrier. A role of the transport carrier for monocarboxylic acids. *J. Pharmacobiodyn.* (1990) **13**:158-163.

78 TERASAKI T, TAKAKUWA S, MORITANI S, TSUJI A: Transport of monocarboxylic acids at the blood-brain barrier: studies with monolayers of primary cultured bovine brain capillary endothelial cells. *J. Pharmacol. Exp. Ther.* (1991) **258**:932-937.

79 GERHART DZ, LEINO RL, DREWES LR: Distribution of monocarboxylate transporters MCT1 and MCT2 in rat retina. *Neuroscience* (1999) **92**:367-375.

80 BERGERSEN L, JOHANNSSON E, VERUKI ML *et al.*: Cellular and subcellular expression of monocarboxylate transporters in the pigment epithelium and retina of the rat. *Neuroscience* (1999) **90**:319-331.

81 HALESTRAP AP, PRICE NT: The proton-linked monocarboxylate transporter (MCT) family: structure, function and regulation. *Biochem. J.* (1999) **343**(Pt 2):281-299.

82 TAMAI I, TAKANAGA H, MAEDA H *et al.*: Proton-cotransport of pravastatin across intestinal brush-border membrane. *Pharm. Res* (1995) **12**:1727-1732.

83 TAKANAGA H, TAMAI I, TSUJI A: pH-dependent and carrier-mediated transport of salicylic acid across Caco-2 cells. *J. Pharm. Pharmacol.* (1994) **46**:567-570.

84 TAKANAGA H, MAEDA H, YABUUCHI H *et al.*: Nicotinic acid transport mediated by pH-dependent anion antiporter and proton cotransporter in rabbit intestinal brush-border membrane. *J. Pharm. Pharmacol.* (1996) **48**:1073-1077.

85 HOSOYA K, KONDO T, TOMI M *et al.*: MCT1-mediated transport of L-lactic acid at the inner blood-retinal barrier: a possible route for delivery of monocarboxylic acid drugs to the retina. *Pharm. Res.* (2001) **18**:1669-1676.

86 HORIBE Y, HOSOYA K, KIM KJ, LEE VH: Carrier-mediated transport of monocarboxylate drugs in the pigmented rabbit conjunctiva. *Invest. Ophthalmol. Vis. Sci.* (1998) **39**:1436-1443.

87 YOON H, FANELLI A, GROLLMAN EF, PHILP NJ: Identification of a unique monocarboxylate transporter (MCT3) in retinal pigment epithelium. *Biochem. Biophys Res. Commun.* (1997) **234**:90-94.

88 CHANCY CD, KEKUDA R, HUANG W *et al.*: Expression and differential polarization of the reduced-folate transporter-1 and the folate receptor alpha in mammalian retinal pigment epithelium. *J. Biol. Chem.* (2000) **275**:20676-20684.

89 SIROTNAK FM, TOLNER B: Carrier-mediated membrane transport of folates in mammalian cells. *Ann. Rev. Nutr.* (1999) **19**:91-122.

90 ANTONY AC: The biological chemistry of folate receptors. *Blood* (1992) **79**:2807-2820.

91 REDDY JA, LOW PS: Enhanced folate receptor mediated gene therapy using a novel pH- sensitive lipid formulation. *J. Control. Release* (2000) **64**:27-37.

92 SMITH SB, KEKUDA R, GU X *et al.*: Expression of folate receptor alpha in the mammalian retinol pigmented epithelium and retina. *Invest. Ophthalmol. Vis. Sci.* (1999) **40**:840-848.

93 SUDIMACK J, LEE RJ: Targeted drug delivery via the folate receptor. *Adv. Drug Deliv. Rev.* (2000) **41**:147-162.

Websites

101 http://www.centerwatch.com/patient/drugs/drug67.html

102 http://www.ama-assn.org/special/hiv/treatmnt/druginfo/vitraser.htm

Affiliation

Sridhar Duvvuri, Soumyajit Majumdar & Ashim K Mitra†

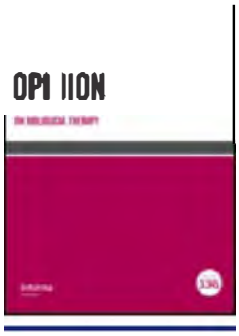
†Author for correspondence

Curator's Professor and Chairman, Division of Pharmaceutical Sciences, School of Pharmacy, University of Missouri-Kansas City, 5005 Rockhill Road, Kansas City, MO 64110, USA

Tel: +1 816 235 1615; Fax: +1 816 235 5190;

E-mail: mitraa@umkc.edu

Regeneron Ex. 2063
Alvotech v. Regeneron
PGR2025-00085
Page 12 of 13



Expert Opinion on Biological Therapy

ISSN: 1471-2598 (Print) 1744-7682 (Online) Journal homepage: https://www.tandfonline.com/loi/iebt20

Drug delivery to the retina: challenges and opportunities

Sridhar Duvvuri, Soumyajit Majumdar & Ashim K Mitra

To cite this article: Sridhar Duvvuri, Soumyajit Majumdar & Ashim K Mitra (2003) Drug delivery to the retina: challenges and opportunities, Expert Opinion on Biological Therapy, 3:1, 45-56, DOI: 10.1517/14712598.3.1.45

To link to this article: https://doi.org/10.1517/14712598.3.1.45

Published online: 03 Mar 2005.

Submit your article to this journal

Article views: 567

View related articles

Citing articles: 12 View citing articles

Full Terms & Conditions of access and use can be found at
https://www.tandfonline.com/action/journalInformation?journalCode=iebt20