

Ocular surface damage by ophthalmic compounds

Flavio Mantelli^a, Laura Tranchina^b, Alessandro Lambiase^a and Stefano Bonini^a

^aDepartment of Ophthalmology, Campus Bio-Medico, University of Rome and ^bDepartment of Biopathology, Ophthalmology Division, UOSD Glaucoma, University of Rome 'Tor Vergata', Rome, Italy

Correspondence to Professor Stefano Bonini, MD, Department of Ophthalmology, Campus Bio-Medico, University of Rome, Via Alvaro del Portillo 28, 00121 Rome, Italy
Tel: +39 06 225419185; fax: +39 06 22541456;
e-mail: s.bonini@unicampus.it

Current Opinion in Allergy and Clinical Immunology 2011, 11:000–000

Purpose of review

To describe the changes of the ocular surface following chronic use of eye drop therapies. The possible pathogenetic mechanisms responsible for specific signs and symptoms are described and discussed.

Recent findings

Topical treatments for ocular diseases may trigger an inflammatory response in predisposed patients, with local activation of immune cells and ocular surface damage. The resulting clinical picture may vary substantially, from mild to severe, sharing a red eye as a hallmark presentation. Recent in-vitro, in-vivo, and clinical data suggest that these detrimental effects are not solely related to eye drop preservatives and may be caused by the medication itself, especially for patients with preexisting ocular diseases. However, no specific tests are currently available to make a clear cut diagnosis between what is caused by the disease and what is the effect of its therapy. Patients' history and clinical features remain essential to hypothesize the underlying pathogenetic mechanism.

Summary

Topical therapies may induce ocular surface allergic reactions, dry eye-like reactions, and epithelial damage. Patients in need of chronic therapies are at higher risk of facing these detrimental effects of eye drop therapies and should be treated with unpreserved compounds.

Keywords

allergic conjunctivitis, dry eye, glaucoma, ocular toxicity, preservatives

Curr Opin Allergy Clin Immunol 11:000–000
© 2011 Wolters Kluwer Health | Lippincott Williams & Wilkins
1528-4050

Introduction

Patients under treatment with topical eye drops for severe long-standing ocular diseases may experience the occurrence of signs and symptoms of inflammation of the ocular surface [1,2^{••}]. In these patients, adverse reactions to topical ophthalmic medications are expressed with a common clinical feature, the 'red eye'. This condition is a common finding in many clinical settings and among the most common and nonspecific ocular signs encountered in ophthalmology practice today [3–6]. In fact, the ocular surface reacts aspecifically to a wide range of external insults, from environmental factors such as pollution, to infectious agents and allergens, to topical eye drop treatments [2^{••},3–10]. Therefore, identifying the exact pathogenetic mechanisms behind the detrimental effects of topical eye drop preparations could prove beneficial to provide novel approaches to treatment for patients in need of chronic therapies or at higher risk of ocular surface damage.

Moreover, as there are no specific tests available to make a definitive and immediate differential diagnosis between what is caused by a reaction to topical treatments and what is caused by a preexisting disease, with

this review we also aim to define specific aspects of ocular surface damage induced by prolonged eye drop therapies.

Diagnosis and management of ocular surface damage

Ocular signs and symptoms associated with a red eye and caused by local side-effects to topical ophthalmic medications may vary substantially, and are often responsible for substantial discomfort leading to discontinuation of the prescribed eye drop therapy or to unnecessary additional prescriptions that initiate a detrimental loop of eye drop toxicity [11].

As the exact pathogenetic mechanisms are often only hypothesized based on the clinical history and accompanying clinical features, these conditions are generally referred to as ocular allergic reactions to the eye drops [7]. However, a specific differential diagnosis remains mandatory to allow a proper management of the patients. The presence of conjunctival papillae or follicles, the type of conjunctival secretion, the involvement of the cornea, and the prevalence of specific symptoms, such as itching, burning, photophobia, tearing, or dryness, may help in the differential diagnosis [12].

The detrimental role of topical therapies on the ocular surface and the thin border between their therapeutic and toxic effects has been attributed for a long time to the preservatives contained in the eye drops [2^{••}]. However, although it remains a rule that preserved eye drops are far more toxic than unpreserved eye drops, it is now evident both preserved and unpreserved eye drop preparations may induce alterations of the ocular surface related to dose, frequency, and duration of treatment [13–17]. These negative effects appear to be more easily elicited in predisposed patients, such as hyperreactive patients with underlying ocular allergy, elderly patients with dry eye or glaucoma, and patients affected by severe ocular diseases in need of chronic therapy [18[•]].

Although signs and symptoms of ocular surface damage are variable and often shared by all these conditions, recent in-vitro, in-vivo, and clinical studies suggest that ocular surface reactions triggered by eye drop treatments are primarily mediated by three different specific pathogenetic mechanisms [14,19[•],20–26]:

- (1) allergic reactions induced by hypersensitivity to preservatives and/or medications;
- (2) dry eye-like reactions induced by alteration of the tear film by preservatives; and
- (3) direct epithelial damage induced by a chronic and/or cumulative use of topical treatments.

In any of the above cases, except for the red eye that represents a constant finding, the clinical picture resulting from ocular toxicity can vary dramatically, with nonspecific signs that overlap with each other and range from mild to severe, including eyelid dermatitis and blepharitis, conjunctival secretion, subconjunctival fibrosis, superficial punctate keratitis, filamentary keratitis, erosions and/or corneal ulcers, and corneal limbal stem cell deficiency [1,27[•],28,29].

Not only are the clinical pictures of ocular surface toxicity often variable and overlapping; there is also a lack of specific laboratory tests and evaluations to address towards a correct diagnosis through the identification of the exact pathogenetic mechanism. For example, an increase in the number of inflammatory-mediated cells and inflammatory mediators – such as interleukins – in the conjunctiva is constantly found [30,31], as well as histologic changes in the corneal and conjunctival epithelium, with squamous metaplasia of various degrees and goblet cells' alterations ranging from loss to hyperplasia [32–34]. However, these findings are common to both allergic reactions, dry eye-like reactions, and ocular surface reactions induced by direct epithelial damage.

Key points

- A red eye is a common finding in patients under chronic eye drop treatment.
- Adverse reactions to eye drop treatments can be induced by both preservatives and active compounds in predisposed patients.
- Mechanisms of ocular surface damage by topical ophthalmic medications include allergic reactions, dry eye-like reactions, and direct effects on the epithelia.

Allergic reactions

The most frequent clinical picture of ocular surface damage by ophthalmic compounds is the result of an allergic reaction to preservatives, additives, and/or drugs contained in ophthalmic preparations. These reactions may take minutes to hours to manifest themselves and generally require a sensitization time to develop ranging from days to weeks after exposure to the sensitizing agent.

The allergic reaction is characterized at the ocular surface by conjunctival hyperemia and chemosis, lid swelling, eczema of the periorbital skin, tearing, photophobia, and intense itching caused by an IgE-mediated (type I) or by a delayed (type IV) hypersensitivity reaction [7]. Itching can actually be considered a pathognomonic symptom of ocular allergy [35]; however, it can also be found in other conditions affecting the ocular surface [12]. Therefore, a careful evaluation of the clinical history and of the accompanying signs and symptoms is mandatory to make a proper differential diagnosis.

As previously mentioned, other characteristics signs of the ocular allergic reaction are hyperemia and chemosis. In chronic ocular allergy sufferers, vasodilation appears superficial and the conjunctival hyperemia tends to appear pink rather than a deep red, as is the case in acute allergic reactions and other acute inflammations of the ocular surface. Chemosis (swelling of the conjunctiva) can be present in both acute and chronic allergic reactions: although it peaks within 15–30 min after exposure to the allergen or to the toxic compound, it tends to dissipate slowly and is often visible on biomicroscopic examination for a longer period, making the differential diagnosis more difficult [35].

Although there is plenty of literature showing an increased risk of developing an allergic reaction to preservatives in eye drops [2^{••},7,36,37], a similar response may also be elicited by hypersensitivity to the medical compound itself. In fact, it has been described with both in-vitro, in-vivo, and clinical studies that the different antiallergic eye drops have different toxic effects on the ocular surface [20]. For instance, in a recent study [21]

ketotifen seemed to affect epithelial cell viability *in vitro* more than olopatadine or levocabastine, although they contained a slightly greater concentration of benzalkonium chloride (BAK).

It is evident that an allergic reaction to a topical ophthalmic compound could affect any patient, but those who are at greater risk of developing an allergic reaction to eye drop therapies are ocular allergy sufferers [7]. The most frequent tendency of allergic patients to develop a conjunctival reaction to ophthalmic preparations may be also because of the higher frequency of conjunctival hyperreactivity to nonspecific stimuli among allergic patients [38]. Therefore, especially for these patients, it has become of primary importance to discriminate when an ocular surface reaction is caused by allergic conjunctivitis and when it is a consequence of a topical eye drop therapy.

Nevertheless, it must be underlined that ocular surface damage induced by allergic reactions to topical medications is not uncommon in patients with no history of ocular allergy. Specifically, it has been described that allergic reactions to topical ocular medications can also develop in patients who have already been under chronic eye drop therapies as the result of switching from prescription to generic drugs or from an old preservative (such as BAK) to a new generation preservative [39]. In fact, although it is a fact that the type of preservative is crucial for developing a dry eye-like toxic reaction and/or direct damage to the ocular surface epithelia, it must be always remembered that this is not true for the allergic reactions, which can also develop for the medical compound itself.

Dry eye-like reactions

Dry eye, or keratoconjunctivitis sicca, is a multifactorial disease of the ocular surface that results in symptoms of discomfort, visual disturbance, and tear film instability, with potential damage to the ocular surface [40].

It is recognized as a disturbance of the lacrimal functional unit, an integrated system comprising the lacrimal glands, ocular surface (cornea, conjunctiva, and meibomian glands) and lids, and the sensory and motor nerves that connect them [40]. An alteration in any of these structures can lead to the development of dry eye because of an alteration of the lacrimal functional unit homeostasis.

Chronic topical therapies for other diseases may be responsible for the development of such an alteration, especially in elderly patients who are already at greater risk of developing dry eye [41]. A typical example is the need for glaucoma medications: in fact, glaucoma's prevalence is higher in the elderly as is dry eye [41,42]. Therefore, the need for chronic pressure-lowering

medications to control glaucoma results in a dramatic increase in dry eye signs and symptoms to up to 78% [43]. The very same dry eye patients can encounter a worsening of their disease caused by the very same topical medications chronically used to control ocular surface symptoms. Nevertheless, the development of a drug-induced dry eye is often seen also in younger patients such as perennial allergy sufferers [21,41].

Considering all these side-effects of chronic therapies at the ocular surface, it can be said that a critical advance in the treatment of dry eye came with the elimination of preservatives, such as BAK, from ocular surface medications and lubricants (the so-called artificial tears) [44]. Nonpreserved single-unit dose vials as well as multidose vials with vanishing preservatives or air-tight constructions have, therefore, led to a great improvement of the quality of life of patients who became less limited by the number of instillations [44]. Moreover, with an increasing prevalence in the worldwide population, the dry eye market has seen in recent years a tremendous intake of novel products. Specifically, novel anti-inflammatory compounds have been developed and there has been a tremendous increase in the number of topical lubricants with various viscosity agents used to mimic the properties of ocular surface mucins [44–46].

Cell-surface-associated (transmembrane) mucins, a group of high molecular weight glycoproteins lining the apical cell surface of the ocular surface epithelia, constitute a major component of their glycocalyx [47]. Their expression, distribution, and glycosylation are known to be altered in dry eye disease [48–50]. Concomitant to the widely described alteration in transmembrane mucins, namely MUC1, MUC4, and MUC16, another feature of dry eye disease is the reduction in goblet cell number and their major secretory product the gel forming mucin MUC5AC, which has been linked to loss of lubrication and epithelial damage [19[•],51].

Alteration in the distribution of mucins and their glycosylation in dry eye disease may result in adherence and damage to the epithelial surface by abrasive stress during blinking or sleeping [52]. In this perspective, it is evident how chronic therapies with preserved eye drops can have a detrimental effect at the ocular surface, especially for those patients requiring frequent instillations and polytherapies: in fact, it has been proven that exposure to preservatives such as BAK is capable of altering mucin expression [53]. Recently, a novel dry eye animal model resembling human disease has also been developed by topical administration of BAK, which resulted in higher TNF- α expression, increased inflammatory infiltration in the cornea, fewer MUC5AC-positive conjunctival cells, and more apoptotic cells in the corneal basal epithelium [19[•]].

Therefore, preservatives contained in the eye drops remain one of the main reasons for developing a dry eye-like toxicity by ocular compounds. Nevertheless, it must be noted that there is plenty of literature showing that this type of toxic reaction is not observed only for preserved eye drops. A recently published meta-analysis of seven double-masked prospective, controlled clinical trials comparing the incidence of punctate keratitis (a typical sign of dry eye) among 1694 patients assigned to treatment with BAK-containing antiglaucoma medications indicates that BAK does not produce significant corneal toxicity in the vast majority of patients under monotherapy at the concentrations commercially used [27[•]].

Direct epithelial damage

Epithelial cells at the ocular surface are continuously faced with the critical challenge of forming a protective apical barrier that prevents cellular damage and adhesion during blinking or sleeping, while allowing the exchange of molecules with the extracellular milieu. Maintenance of an intact mucosal barrier is, therefore, critical to preventing damage to corneal and conjunctival epithelia. Loss of barrier function is ascribed to numerous mucosal pathologies including those mentioned previously, dry eye and ocular surface hypersensitivity, and can also be a consequence of chronic topical therapies [54^{••}].

Contrary to dry eye-like and allergic reactions, the detrimental role of topical therapies on the epithelial barrier is not only to be considered an adverse side-effect of therapy. In fact, a disruption of the epithelial barrier is partly sought after to increase permeability of the drugs [55]. A typical example is antiglaucoma medications, in which preservatives such as BAK have been widely used for decades not only to stabilize the drugs but also to increase their permeability and, therefore, their effect on lowering intraocular pressure [56,57].

As previously mentioned, BAK is known to alter mucin expression and distribution at the ocular surface. Although it was believed that this effect was responsible itself for ocular toxicity and epithelial damage induced by BAK, recent evidence has shown that the protective mechanisms of the ocular surface are far more complex and mucins interact with carbohydrate-binding proteins in order to provide a further layer of protection that is harder to breach. Specifically, a seminal paper by Argüeso *et al.* [54^{••}] has provided direct evidence of the role of mucins in maintaining mucosal integrity through a carbohydrate-dependent interaction with a molecule known as galectin-3 on apical membranes to promote the formation of an epithelial barrier. Both mucins and galectin-3 are among the most highly expressed molecules at the ocular surface, making the epithelial barrier a major component of the ocular surface that must be considered while

evaluating toxicity of topical ophthalmic compounds. In fact, understanding the importance of the corneal and conjunctival epithelial barrier not only has potential applications to the understanding of barrier dysfunction in epithelial diseases but also allows investigation of whether a transient disruption of the mucin–galectin interaction could prove beneficial to the enhancement of drug delivery into mucosal surfaces for therapeutic purposes without the need for adding toxic preservatives in ocular medications [54^{••}].

In fact, although the epithelial barrier made by mucin–galectin interaction is hard to breach, the cumulative use of polytherapy for glaucoma over time is still likely to increase the incidence of toxic and adverse events at the ocular surface [43]. The ocular hypertension treatment study [58] demonstrated that 40% of patients initially diagnosed with ocular hypertension were using more than one drug after 5 years. Similarly, the Collaborative Initial Glaucoma Treatment Study [59] showed that 75% of patients were using two or more drugs within 2 years of initial glaucoma treatment. These as well as many other studies show that the effects of polytherapy on the epithelial barrier can be substantial, with a disruption of the protective layers resulting in direct epithelial damage [22]. Specifically, combination therapies have been widely proven to induce a significant reduction in the percentage of live corneal and conjunctival epithelial cells both *in vitro* and *in vivo*. Interestingly, this proapoptotic effect appears to be related not only to the increased concentration of preservatives, but also to the increased concentration of the active compound regardless of the type of preservative used [60].

Overall, these evidences confirm that once the epithelial barrier is breached (through the use of preservatives or for any other cause), a direct epithelial damage is easier to obtain and the development of new approaches to prevent ocular surface alterations in patients under chronic topical medications is mandatory. This could be achieved both by aiming at restoring a healthy epithelial barrier after the ocular surface has been compromised or by developing therapeutic strategies that require a lower number of instillation, possibly in monotherapy and without the use of preservatives.

Discussion

In this review, we show that chronic topical therapies may induce ocular surface damage through three separate but strictly connected pathways: allergic reactions, dry eye-like reactions, and direct epithelial damage through epithelial barrier disruption.

Preservatives used to avoid contamination of topical ophthalmic compounds or to enhance their permeability

Table 1 Examples of eye drop preservatives' effects at the ocular surface

Active component	Positive effect	Side-effect
Benzalkonium chloride (BAK)	Antimicrobial activity [61] Enhancement of drug penetration [55]	Conjunctival cell cytotoxicity <i>in vitro</i> [62] Increase in TNF-alpha in immortalized human corneal and conjunctival epithelial cells [63] Overexpression of ICAM-1 in human conjunctival cells [64] Increase in interleukins in tears [31] Decrease in goblet cells [64]
Purite (stabilized oxychloro complex)	Enhancement of drug penetration [56]	Inflammatory cells' infiltration in conjunctiva [24] Ocular allergic reaction [65] Intolerance in irritated eyes [66]
SofZia (preservative buffered system)	Enhancement of drug penetration [57]	Conjunctival hyperemia [57] In-vitro conjunctival cell cytotoxicity [67] Corneal epithelial cell damage and conjunctival lymphocyte infiltration in animal models [68] Alteration in goblet cells in animal models [68]
Polyquad (polyquaternium-1)	Disinfectant in contact lens, good safety and tolerance profile [69]	Cytotoxicity in animal models [70] In-vitro cytotoxicity [22]

ICAM, intercellular adhesion molecule.

remain the main causative agents of these detrimental effects (Table 1) [22,24,31,55–57,61–70]. However, in recent years plenty of evidence has shown that they are not the only players of the inflammatory cascade triggered at the ocular surface by chronic topical therapies. The medical compounds themselves may induce ocular damage in predisposed patients in a cumulative effect related to dosage and duration of therapy or preexisting diseases (Table 2) [14,21,71–82]. Therefore, although the advent of preservative-free topical ophthalmic compounds has certainly represented a significant innovation in ophthalmic drug development, the use of preserved eye drops does not need to be banned *a priori*. The use of unpreserved compounds should be considered, when available, for every patient, but it should become mandatory only for those patients who have already

developed side-effects to topical therapies, or who are at greater risk of ocular surface damage for preexisting conditions in need of chronic therapies, or for those who are affected by severe ocular surface diseases.

In fact, an alteration to any component of the ocular surface may result in a complex clinical picture in which it is hard to determine what is caused by a preexisting disease and what is caused by its therapy. In these cases, making a definitive and immediate differential diagnosis remains a challenge as, to date, no specific tools are available. A clear understanding of the exact pathogenetic mechanism behind ocular surface damage induced by ophthalmic compounds could lead to a better management of patients in need of chronic eye drop treatments or patients who have already presented an ocular surface reaction to topical therapies.

Table 2 Examples of detrimental effects induced by topical ophthalmic drugs

Active component	Ocular surface adverse effect
Agonist α -adrenergic	Topical allergy-like syndrome [71]
Beta-blockers	Allergic conjunctivitis [72] Punctate keratitis [72] Tear film instability [73]
Prostaglandins	Conjunctival hyperemia [74] Reactivation of herpes simplex keratitis [75]
Antiallergic compounds	Anterior segment inflammation [76] Cytotoxicity on human corneal endothelial cells <i>in vitro</i> [21] Corneal epithelial breakdown [77] Overexpression of ICAM-1 on human conjunctiva-derived cells [78]
Nonsteroidal anti-inflammatory compounds	Hyperemia, burning, and stinging [79] Corneal anesthesia [80] Epitheliopathy [14] Corneal ulcer and perforation [81] Corneal melting [82]

ICAM, intercellular adhesion molecule.

Acknowledgements

Conflicts of interest

None declared.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- of special interest
- of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (pp. 000–000).

- 1 Servat JJ, Bernardino CR. Effects of common topical antiglaucoma medications on the ocular surface, eyelids and periorbital tissue. *Drugs Aging* 2011; 28:267–282.
 - 2 Baudouin C, Labbé A, Liang H, *et al*. Preservatives in eyedrops: the good, the bad and the ugly. *Prog Retin Eye Res* 2010; 29:312–334.
- In this review, a large body of evidence is presented showing that ocular surface alterations induced by long-term use of preserved eye drops are not restricted only to allergic reactions and are often difficult to identify. The authors suggest that it would be advisable to use preservative-free solutions in patients in need of prolonged treatments or suffering from preexisting ocular surface diseases.

- 3 Granet D. Allergic rhinoconjunctivitis and differential diagnosis of the red eye. *Allergy Asthma Proc* 2008; 29:565–574.
- 4 Novaes P, Saldiva PH, Matsuda M, *et al.* The effects of chronic exposure to traffic derived air pollution on the ocular surface. *Environ Res* 2010; 110:372–374.
- 5 Novaes P, do Nascimento Saldiva PH, Kara-José N, *et al.* Ambient levels of air pollution induce goblet-cell hyperplasia in human conjunctival epithelium. *Environ Health Perspect* 2007; 115:1753–1756.
- 6 Wirbelauer C. Management of the red eye for the primary care physician. *Am J Med* 2006; 119:302–306.
- 7 Baudouin C. Allergic reaction to topical eyedrops. *Curr Opin Allergy Clin Immunol* 2005; 5:459–463.
- 8 Rummien VT, Matsumoto Y, Dogru M, *et al.* Tear cytokine and ocular surface alterations following brief passive cigarette smoke exposure. *Cytokine* 2008; 43:200–208.
- 9 Ueta M, Kinoshita S. Ocular surface inflammation mediated by innate immunity. *Eye Contact Lens* 2010; 36:269–281.
- 10 Tarabishy AB, Jeng BH. Bacterial conjunctivitis: a review for internists. *Cleve Clin J Med* 2008; 75:507–512.
- 11 Sacchetti M, Lambiase A, Coassin M, *et al.* Toxic corneal ulcer: a frequent and sight-threatening disease. *Eur J Ophthalmol* 2009; 19:916–922.
- 12 Mantelli F, Lambiase A, Bonini S. A simple and rapid diagnostic algorithm for the detection of ocular allergic diseases. *Curr Opin Allergy Clin Immunol* 2009; 9:471–476.
- 13 Ayaki M, Iwasawa A, Inoue Y. Toxicity of antiglaucoma drugs with and without benzalkonium chloride to cultured human corneal endothelial cells. *Clin Ophthalmol* 2010; 21:1217–1222.
- 14 Ayaki M, Iwasawa A, Soda M, *et al.* Cytotoxicity of five fluoroquinolone and two nonsteroidal anti-inflammatory benzalkonium chloride-free ophthalmic solutions in four corneal conjunctival cell lines. *Clin Ophthalmol* 2010; 4:1019–1024.
- 15 Ayaki M, Iwasawa A. Cytotoxicity of prostaglandin analog eye drops preserved with benzalkonium chloride in multiple corneal conjunctival cell lines. *Clin Ophthalmol* 2010; 4:919–924.
- 16 Uusitalo H, Chen E, Pfeiffer N, *et al.* Switching from a preserved to a preservative-free prostaglandin preparation in topical glaucoma medication. *Acta Ophthalmol* 2010; 88:329–336.
- 17 Higginbotham EJ. Considerations in glaucoma therapy: fixed combinations versus their component medications. *Clin Ophthalmol* 2010; 4:1–9.
- 18 Stewart WC, Stewart JA, Nelson LA. Ocular surface disease in patients with ocular hypertension and glaucoma. *Curr Eye Res* 2011; 36:391–398. The authors show that ocular surface alterations are common in patients under chronic eye drop treatment for glaucoma and result in symptoms and signs that may impact their quality of life.
- 19 Lin Z, Lui X, Zhou T, *et al.* A mouse dry eye model induced by topical administration of benzalkonium chloride. *Mol Vis* 2011; 17:257–264. The development of a new animal model of dry eye using benzalkonium chloride suggests that preservatives significantly increase the risk of dry eye-like adverse effects of chronic eye drop therapies.
- 20 Ayaki M, Iwasawa A, Yaguchi S, Koide R. Preserved and unpreserved 12 antiallergic ophthalmic solutions and ocular surface toxicity: in vitro assessment in four cultured corneal and conjunctival epithelial cell lines. *Biocontrol Sci* 2010; 15:143–148.
- 21 Ayaki M, Iwasawa A, Yaguchi S, Koide R. In vitro assessment of the cytotoxicity of antiallergic eye drops using 5 cultured corneal and conjunctival cell lines. *J Oleo Sci* 2011; 60:139–144.
- 22 Ammar DA, Noecker RJ, Kahook MY. Effects of benzalkonium chloride-preserved, polyquad-preserved, and sofZia-preserved topical glaucoma medications on human ocular epithelial cells. *Adv Ther* 2010; 27:837–845.
- 23 Whitson JT, Cavanagh HD, Lakshman N, Petroll WM. Assessment of corneal epithelial integrity after acute exposure to ocular hypotensive agents preserved with and without benzalkonium chloride. *Adv Ther* 2006; 23: 663–671.
- 24 Noecker RJ, Herrygers LA, Anwaruddin R. Corneal and conjunctival changes caused by commonly used glaucoma medications. *Cornea* 2004; 23:490–496.
- 25 Guenoun JM, Baudouin C, Rat P, *et al.* In vitro study of inflammatory potential and toxicity profile of latanoprost, travoprost, and bimatoprost in conjunctiva-derived epithelial cells. *Invest Ophthalmol Vis Sci* 2005; 46:2444–2450.
- 26 Becquet F, Goldschmidt M, Moldovan MS, *et al.* Histopathological effects of topical ophthalmic preservatives on rat corneal conjunctival surface. *Curr Eye Res* 1998; 17:419–425.
- 27 Trocme S, Hwang LJ, Bean GW, Sultan MB. The role of benzalkonium chloride in the occurrence of punctate keratitis: a meta-analysis of randomized, controlled clinical trials. *Ann Pharmacother* 2010; 44:1914–1921. The authors demonstrate with a meta-analysis that the most commonly used eye drop preservative does not produce significant corneal toxicity in the majority of patients. This study highlights that the use of preservative-free eye drops is advisable in every patient, but is mandatory only for patients with preexisting ocular surface disease or at greater risk of developing side-effects.
- 28 Pisella PJ, Pouloquen P, Baudouin C. Prevalence of ocular symptoms and signs with preserved and preservative free glaucoma medication. *Br J Ophthalmol* 2002; 86:418–423.
- 29 Jaenen N, Baudouin C, Poiliqueu P, *et al.* Ocular symptoms and signs with preserved and preservative-free glaucoma medications. *Eur J Ophthalmol* 2007; 17:341–349.
- 30 Sherwood MB, Grierson I, Millar L, Hitchings RA. Long-term morphologic effects of antiglaucoma drugs on the conjunctiva and Tenon's capsule in glaucomatous patients. *Ophthalmology* 1989; 96:327–335.
- 31 Manni G, Centofanti M, Oddone F, *et al.* Interleukin-1 β tear concentration in glaucomatous and ocular hypertensive patients treated with preservative-free nonselective β -blockers. *Am J Ophthalmol* 2005; 139:72–77.
- 32 Broadway D, Hitchings R. Conjunctival damage induced by long-term topical antiglaucoma therapy. *Acta Ophthalmol Scand* 1996; 74:97.
- 33 Brandt JD, Wittmann JR, Katz LJ, *et al.* Conjunctival impression cytology in patients with glaucoma using long-term topical medication. *Am J Ophthalmol* 1991; 112:297–301.
- 34 Arici MK, Arici DS, Topalkara A, Güler C. Adverse effects of topical antiglaucoma drugs on the ocular surface. *Clin Exp Ophthalmol* 2000; 28:113–117.
- 35 Ono SJ, Abelson MB. Allergic conjunctivitis: update on pathophysiology and prospects for future treatment. *J Allergy Clin Immunol* 2005; 115:118–122.
- 36 Hong J, Bielory L. Allergy to ophthalmic preservatives. *Curr Opin Allergy Clin Immunol* 2009; 9:447–453.
- 37 Baines MG, Cai F, Backman HA. Ocular hypersensitivity to thimerosal in rabbits. *Invest Ophthalmol Vis Sci* 1991; 32:2259–2265.
- 38 Ciprandi G, Buscaglia S, Pesce GP, *et al.* Ocular challenge and hyperresponsiveness to histamine in patients with allergic conjunctivitis. *J Allergy Clin Immunol* 1993; 91:1227–1230.
- 39 Sullivan-Mee M, Pensyl D, Alldredge B, *et al.* Brimonidine hypersensitivity when switching between 0.2% and 0.15% formulations. *J Ocul Pharmacol Ther* 2010; 26:355–360.
- 40 The definition and classification of dry eye disease: report of the Definition and Classification Subcommittee of the International Dry Eye Workshop (2007). *Ocul Surf* 2007; 5:75–92.
- 41 The epidemiology of dry eye disease: report of the Epidemiology Subcommittee of the International Dry Eye Workshop (2007). *Ocul Surf* 2007; 5:93–107.
- 42 Quigley HA. Glaucoma. *Lancet* 2011; 377:1367–1377.
- 43 Leung E, Medeiros FA, Weinreb RN. Prevalence of ocular surface disease in glaucoma patients. *J Glaucoma* 2008; 17:350–355.
- 44 Management and therapy of dry eye disease: report of the Management and Therapy Subcommittee of the International Dry Eye Workshop (2007). *Ocul Surf* 2007; 5:163–178.
- 45 Ceulemans J, Vinckier I, Ludwig A. The use of xanthan gum in an ophthalmic liquid dosage form: rheological characterization of the interaction with mucin. *J Pharm Sci* 2002; 91:1117–1127.
- 46 Paugh JR, Nguyen AL, Ketelson HA, *et al.* Precorneal residence time of artificial tears measured in dry eye subjects. *Optom Vis Sci* 2008; 85:725–731.
- 47 Mantelli F, Argüeso P. Functions of ocular surface mucins in health and disease. *Curr Opin Allergy Clin Immunol* 2008; 8:477–483.
- 48 Gipson IK, Hori Y, Argüeso P. Character of ocular surface mucins and their alteration in dry eye disease. *Ocul Surf* 2004; 2:131–148.
- 49 Paulsen F, Jäger K, Worlitzsch D, *et al.* Regulation of MUC16 by inflammatory mediators in ocular surface epithelial cell lines. *Ann Anat* 2008; 190:59–70.
- 50 Mantelli F, Schaffer L, Dana R, *et al.* Glycogene expression in conjunctiva of patients with dry eye: downregulation of Notch signaling. *Invest Ophthalmol Vis Sci* 2009; 50:2666–2672.
- 51 Argüeso P, Balaram M, Spurr-Michaud S, *et al.* Decreased levels of the goblet cell mucin MUC5AC in tears of patients with Sjögren syndrome. *Invest Ophthalmol Vis Sci* 2002; 43:1004–1011.
- 52 Sumiyoshi M, Ricciuto J, Tisdale A, *et al.* Antiadhesive character of mucin O-glycans at the apical surface of corneal epithelial cells. *Invest Ophthalmol Vis Sci* 2008; 49:197–203.

- 53 Chung SH, Lee SK, Cristol SM, *et al.* Impact of short-term exposure of commercial eyedrops preserved with benzalkonium chloride on precorneal mucin. *Mol Vis* 2006; 12:415–421.
- 54 Argüeso P, Guzman-Arangué A, Mantelli F, *et al.* Association of cell surface mucins with galectin-3 contributes to the ocular surface epithelial barrier. *J Biol Chem* 2009; 284:23037–23045.
- This elegant article provides the first evidence of an extrinsic mucosal barrier lining the corneal and conjunctival epithelia, opening new avenues of investigation on the pathogenesis of ocular surface disease. The authors also suggest that their results have potential applications for the enhancement of drug delivery with eye drop preparations.
- 55 Ramselaar JA, Boot JP, van Haeringen NJ, *et al.* Corneal epithelial permeability after instillation of ophthalmic solutions containing local anaesthetics and preservatives. *Curr Eye Res* 1988; 7:947–950.
- 56 Dong JQ, Babusis DM, Welty DF, *et al.* Effects of the preservative purite on the bioavailability of brimonidine in the aqueous humor of rabbits. *J Ocul Pharmacol Ther* 2004; 20:285–292.
- 57 Lewis RA, Katz GJ, Weiss MJ, *et al.* Travoprost 0.004% with and without benzalkonium chloride: a comparison of safety and efficacy. *J Glaucoma* 2007; 16:98–103.
- 58 Kass MA, Heuer DK, Higginbotham EJ, *et al.* The Ocular Hypertension Treatment Study: a randomized trial determines that topical ocular hypotensive medication delays or prevents the onset of primary open-angle glaucoma. *Arch Ophthalmol* 2002; 120:701–713.
- 59 Lichter PR, Musch DC, Gillespie BW, *et al.* Interim clinical outcomes in the Collaborative Initial Glaucoma Treatment Study comparing initial treatment randomized to medications or surgery. *Ophthalmology* 2001; 108:1943–1953.
- 60 Ammar DA, Kahook MY. The effects of combination glaucoma medications on ocular surface epithelial cells. *Adv Ther* 2009; 26:970–975.
- 61 Charnock C. Are multidose over-the-counter artificial tears adequately preserved? *Cornea* 2006; 25:432–437.
- 62 Ammar DA, Noecker RJ, Kahook MY. Effects of benzalkonium chloride- and polyquad-preserved combination glaucoma medications on cultured human ocular surface cells. *Adv Ther* 2011; 28:501–510.
- 63 Epstein SP, Ahdoot M, Marcus E, Asbell PA. Comparative toxicity of preservatives on immortalized corneal and conjunctival epithelial cells. *J Ocul Pharmacol Ther* 2009; 25:113–119.
- 64 Pisella PJ, Debbasch C, Hamard P, *et al.* Conjunctival proinflammatory and proapoptotic effects of latanoprost and preserved and unpreserved timolol: an ex vivo and in vitro study. *Invest Ophthalmol Vis Sci* 2004; 45:1360–1368.
- 65 Kim CY, Hong S, Seong GJ. Brimonidine 0.2% versus brimonidine purite 0.15% in Asian ocular hypertension. *J Ocul Pharmacol Ther* 2007; 23:481–486.
- 66 Mundorf T, Wilcox KA, Ousler GW 3rd, *et al.* Evaluation of the comfort of Alphagan P compared with Alphagan in irritated eyes. *Adv Ther* 2003; 20:329–336.
- 67 Baudouin C, Riancho L, Warnet JM, Brignole F. In vitro studies of anti-glaucomatous prostaglandin analogues: travoprost with and without benzalkonium chloride and preserved latanoprost. *Invest Ophthalmol Vis Sci* 2007; 48:4123–4128.
- 68 Kahook MY, Noecker R. Quantitative analysis of conjunctival goblet cells after chronic application of topical drops. *Adv Ther* 2008; 25:743–751.
- 69 Lipener C; Contact Lens Advisory in Scientific Studies (CLASS) group. A randomized clinical comparison of OPTI-FREE EXPRESS and ReNu MultiPLUS multipurpose lens care solutions. *Adv Ther* 2009; 26:435–446.
- 70 Labbé A, Pauly A, Liang H, *et al.* Comparison of toxicological profiles of benzalkonium chloride and polyquaternium-1: an experimental study. *J Ocul Pharmacol Ther* 2006; 22:267–278.
- 71 Manni G, Centofanti M, Sacchetti M, *et al.* Demographic and clinical factors associated with development of brimonidine tartrate 0.2%-induced ocular allergy. *J Glaucoma* 2004; 13:163–167.
- 72 Zimmerman TJ, Boger WP 3rd. The beta-adrenergic blocking agents and the treatment of glaucoma. *Surv Ophthalmol* 1979; 23:347–362.
- 73 Ishibashi T, Yokoi N, Kinoshita S. Comparison of the short-term effects on the human corneal surface of topical timolol maleate with and without benzalkonium chloride. *J Glaucoma* 2003; 12:486–490.
- 74 Astin M, Stjernschantz J, Selén G. Role of nitric oxide in PGF2 alpha-induced ocular hyperemia. *Exp Eye Res* 1994; 59:401–407.
- 75 Kroll DM, Schuman JS. Reactivation of herpes simplex virus keratitis after initiating bimatoprost treatment for glaucoma. *Am J Ophthalmol* 2002; 133:401–403.
- 76 Warwar RE, Bullock JD, Ballal D. Cystoid macular edema and anterior uveitis associated with latanoprost use: experience and incidence in a retrospective review of 94 patients. *Ophthalmology* 1998; 105:263–268.
- 77 Nakashima M, Nakamura T, Teshima M, *et al.* Breakdown evaluation of corneal epithelial barrier caused by antiallergic eyedrops using an electrophysiologic method. *J Ocul Pharmacol Ther* 2008; 24:43–51.
- 78 Pauly A, Brignole-Baudouin F, Guenoun JM, *et al.* Comparative study of topical antiallergic eye drops on human conjunctiva-derived cells: responses to histamine and IFN gamma and toxicological profiles. *Graefes Arch Clin Exp Ophthalmol* 2007; 245:534–546.
- 79 Flach AJ. Topical nonsteroidal antiinflammatory drugs in ophthalmology. *Int Ophthalmol Clin* 2002; 42:1–11.
- 80 Gaynes BI, Fiscella R. Topical nonsteroidal anti-inflammatory drugs for ophthalmic use: a safety review. *Drug Saf* 2002; 25:233–250.
- 81 Guidera AC, Luchs JI, Udell U. Keratitis, ulceration, and perforation associated with topical nonsteroidal anti-inflammatory drugs. *Ophthalmology* 2001; 108:936–944.
- 82 Flach AJ. Corneal melts associated with topically applied nonsteroidal anti-inflammatory drugs. *Trans Am Ophthalmol Soc* 2001; 99:205–210.