Published in final edited form as:

Cornea. 2010 October ; 29(10): 1075–1085. doi:10.1097/ICO.0b013e3181d103bb.

Forty Years in Search of the Perfect Contact Lens

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Abstract

Purpose—To identify the pathophysiological changes produced by contact lens wear that predispose the cornea to infection and search for prospective, modifiable risk factors that could reduce the incidence of this critical complication in millions of patients worldwide.

Methods—Significant experimental and clinical publications are reviewed and the results of ongoing studies are presented.

Results—*Pseudomonas aeruginosa* (PA) is the most common pathogen causing lens-related infectious keratitis over three decades. Contact lens wear can increase the risk of infection by increasing surface cell PA binding thereby promoting invasion between broken tight junctions (TJs), and initiating direct intracellular invasion mediated by lens-induced membrane lipid rafts. Prevention of upregulation of specific surface binding receptors for PA with concomitant increase in infection risk, is a Zero Damage Game where independent interactions between lens type, mode of wear, oxygen transmissibility, polymer and toxic effects of associated care solutions ideally should collectively produce no increased ability for PA to attach and/or to invade, thus minimizing the risk for lens-associated infections. The specific hypothesis tested is: “*no increased epithelial surface damage...no increased PA binding or invasion...no increased risk for infection.*” Testing of this new paradigm has been performed in vitro, and in animal and human clinical trials and correlated clinically with relative risk results from robust current epidemiological studies. Results to date clearly support the use of lens-related increases in PA binding (bench) as a non-invasive clinical predictor of risk for lens-related infection in subsequent large scale population studies (bedside). Currently, results suggest that use of common commercial multi-purpose care solutions (MPS) with soft lenses may alone significantly increase infection risk by enhancing lens-related PA binding as compared to use of non-preserved solutions (hydrogen peroxide). Clinical testing also shows that only peroxide solutions show significant disinfection capability against amoebic cysts. Further case-control studies to examine relative risk for infection by lens type and lens care solution are urgently needed.

Conclusions—Millions of patients are dependent on contact lenses for vision worldwide; and, over three decades lens use has increased while risk for lens-related infection has remained stubbornly unchanged. Unfortunately, recent introduction of a new generation of hyper oxygen transmissible lenses used with traditional MPS solutions has not lowered overall risks for lens-related infections; however, similar lenses used with non-preserved care solutions (peroxide) recently demonstrated no significant increases in PA binding in a one-year clinical trial. Collectively, these findings along with the urgent need for amoebic cysticidal disinfection, have led to a current recommendation to patients to use non-preserved (hydrogen peroxide) care solutions in soft lens wear.

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Disclosure: Drs. Robertson, Petroll, and Jester (none); Dr. Cavanagh is a consultant for Menicon Ltd., Nagoya, Japan.

Keywords

Contact Lens; Hypoxia; Corneal infection; Lens Care Solutions; Corneal Epithelium; *Pseudomonas*

Over the past forty years, contact lenses have gained increasing popularity for the correction of refractive errors of the eye. At this time, about 140 million patients wear contact lenses world wide, with some 38 million exposed to the device in the United States, and a similar 10–12% of the population of Great Britain (Richard Weisbarth, personal communication). Importantly, for several decades contact lens use has continued to grow at a variable annual rate of 5–15%; thus, sight threatening complications while uncommon, adversely afflict significantly increased numbers of patients each year (Richard Weisbarth, personal communication). If, as recent epidemiological studies continue to confirm, the rates of lens-related microbial keratitis in daily wear remain 1/2500 and 1/500 in overnight or extended wear, then there will be increasing tens of thousands of new lens wearers at risk world wide every year.^{1–4}

Given this challenge, it is critical to identify the biological mechanisms that predispose the cornea to infection in lens use, and search for modifiable risks that could reduce this sight-threatening complication. The purpose of this Castroviejo Lecture is to review significant clinical and basic science publications as well as ongoing studies to achieve the safest possible contact lens wear.

Contact Lens-related Microbial Keratitis

Lens-related corneal infection has been and remains the major, sight-threatening complication of contact lens use. All major epidemiological studies have consistently shown that the most predominant pathogen is *Pseudomonas aeruginosa* (PA).^{1–5} In order to establish infection however, PA must first “stick” to the corneal epithelial surface. Without attachment infection can not occur. Adhesion is mediated by specific molecular receptors expressed on the outer plasma cell membrane: Lipopolysaccharide (LPS, endotoxin), asialo-ganglioside M-1, N-acetyl-mannosamine, and sialic acid.^{6–11} Up regulation of these lectin surface receptors is associated with increased ability of PA to bind to a cornea exposed to contact lens wear;^{12–19} thus, such lens-related upregulation of binding should be a prospective, measureable outcome measure predictive of increased risk for clinical infection. It is critical to understand however, that lens-induced increases in PA adherence to the corneal epithelium can be driven by many independent but highly interactive factors such as: lens oxygen transmission, lens type (rigid gas permeable (RGP) vs. soft), lens polymer (hydrogel, silicone hydrogel), wearing schedule (daily, occasional overnight use, extended wear), lens fit (alignment vs. Orthokeratology), and the potential adverse effects of lens-care solutions on corneal epithelial cells and on the integrity of ZO-1- mediated, tight cell-to-cell corneal surface junctions (TJ).^{20–22} The prevention of microbial keratitis is thus a Zero Damage Game (Figure 1), where ideally none of these interactive components alone or in combination over decades of wear should alter the corneal epithelium to enhance PA surface adhesion or promote intercellular invasion through breakdown down of TJ surface barriers. Unfortunately, the use of daily *disposable* soft contact lenses has consistently failed to show an overall reduction in risk for PA lens-related infection over the past two decades.^{1–34}

There is also a second pathophysiological pathway which is as yet unappreciated by most clinicians.²³ Notably, PA has recently been shown to invade the corneal epithelium *intracellularly* through lipid-raft-mediated endocytosis during contact lens wear.^{24–27} Lipid rafts are aggregated cholesterol and glycopospholipid (GM-1)–enriched domains in the corneal epithelial cell plasma membrane which form and transport PA to the cell interior. Rafts can be stained with fluorescently labeled antibodies to the beta sub-unit of cholera-toxin and

imaged dynamically in vitro and in vivo by laser scanning confocal microscopy.^{24, 25} Monolayer or air-lifted cultures of human corneal epithelial cells readily demonstrate raft-mediated PA invasion. Importantly however, in vivo rabbit model studies²⁶ reveal that: (1) there are no rafts present in the living corneal epithelium and none are inducible by exposure alone to differing stains of invasive PA in high concentration (10^9); (2) by contrast, wear of a rigid test contact lens that induces hypoxia causes rafts to form with subsequent PA internalization restricted to the *mid-peripheral* and *central* corneal epithelium (Figure 2). No rafts or internalization are seen in the para-limbal, limbal or conjunctival epithelium, even though positive staining is present for beta cholera toxin indicating a potential for this process.²⁶ (Figure 3) The message here is clear: the normal cornea can be exposed to high numbers of invasive PA without attachment or invasion; the presence of a lens with low oxygen transmission is required to initiate the potential pathogenesis of intracellular PA infection through rafts.

The importance of the recognition of this intracellular pathway to corneal invasion is that it can easily be prevented by both in vitro and in vivo use of non-toxic beta cyclo-methyl dextrins (statins) or topical filipin (anti-cholesterol membrane drugs),^{24–27} thus offering a new paradigm both for the treatment of established PA corneal infection or the possible prevention of the same. For example, future case-control studies of contact lens-related PA infection should investigate a potential risk reduction in contact lens patients routinely using concomitant daily statins (Lipitor, Mevacor, Zocor, etc) to lower serum cholesterol. Furthermore, since commonly used antimicrobial agents such as aminoglycosides can not penetrate intracellularly, concomitant treatment with suitable fluoroquinolones which can kill intracellular PA in susceptible strains is also required to eradicate established corneal infection.

Relationship between Lens Oxygen Transmission, Lens Type, and Enhanced PA Binding

In vivo rabbit studies

Using the rabbit model, Imayasu et al examined the effects of 24 hours of rigid, hydrogel, silicone hydrogel or silicone lens wear in the corneal epithelium.²⁸ Lens-induced surface cell damage was assessed by in vivo confocal microscopy, and correlated with PA-binding to the entire corneal epithelium by the colony forming unit method.²⁸ The results are summarized in Figure 4. Findings from this work demonstrated that at the same oxygen transmission level, RGP lenses produce significantly *more* epithelial surface damage, but manifest significantly *less* PA binding than soft lenses with a “stagnant” posterior lens tear volume. This provided for the first time a strong biological rationale for why rigid lens wear (daily or extended) has always been found to manifest a lower clinical relative risk for microbial keratitis (1–2 per 10,000 users) than soft lenses.^{1–4} Thus, even as lens oxygen transmission levels are increased to higher levels associated with decreased induced surface damage,²⁸ the hypothesis of less lens damage, less PA binding, less risk for infection should continue to favor the RGP lens in future clinical epidemiological studies.

In vivo human studies

In a series of recent papers,^{14, 16–18} Ren et al have established an important correlation between PA binding to corneal surface cells exfoliated by gentle washing from the contact lens exposed cornea (Figure 5) and total corneal epithelial PA binding as measured by colony forming unit analysis ($P < 0.01$).^{14, 17, 29} This finding opened the way to non-invasive measurement of PA binding to the human cornea in clinical trials with differing test lens types, polymers, associated lens care solutions, and modalities of fit and wearing time. In a prospective, randomized, double-masked, single center clinical trial, Ren, et al also established the now classic finding that lens oxygen transmission parameters directly regulate PA binding ($p < 0.0001$; $R = 0.392$;

power 0.985, Figure 6).¹⁴ Thus, if all other interactive causes of increasing PA binding were neutralized, a new generation of hyper-oxygen transmitting RGP and silicone hydrogel lenses could potentially demonstrate an overall less risk for lens-associated microbial keratitis.

In placing into clinical perspective the effects of lens oxygen, it is also critical to note a companion study by Ren et al which demonstrated that even when the cornea is swollen by 7% in human volunteers wearing eye goggles under anoxic conditions (100% nitrogen; 95% nitrogen and 5% CO₂) for six hours, exfoliated corneal surface epithelial cells did not exhibit increased PA binding but did exfoliate at a decreased rate.¹⁶ Thus, as with raft formation, hypoxia alone is not sufficient to increase PA binding, the presence of a lens is required. Open eye hypoxia alone however, is sufficient to reduce surface cell desquamation.

Effects of Lens Wear on Corneal Epithelial Homeostasis

The importance of preventing initial adherence of PA to surface corneal cells is heightened by the consistent clinical observations that *all* lens wear of any lens type or duration shuts down normal apoptotic-driven central surface cell exfoliation, gradually increasing the size of the “retained” cells, and resulting in a centrally thinned epithelial layer.^{13–15} Parallel animal studies in the rabbit confirm these effects, and also demonstrate a significant reduction in basal epithelial mitosis as well as a slowing of vertical differentiation.^{30–34} These effects appear to be mediated by lens-induced retention of the nuclear regulatory oncogene Bcl2 in non-phosphorylated form; interestingly, the reduced shedding effect is seen with both daily and extended wear, and also appears to be independent of lens oxygen transmission.^{35–37} Overall lens wear thus creates a “stagnant” corneal surface which can not spontaneously exfoliate a cell with adherent PA; thus all lens wear of any duration or lens type reduces corneal defenses against PA invasion by prevention of normal host sloughing of infected cells.

Effects of Multipurpose Lens Care Solutions on PA Binding and Inter-Epithelial Cell Invasion

In vitro studies

Effects of multipurpose lens care solutions (MPS) on cultured corneal epithelial cells have been studied by several authors in different test cell lines.^{20, 21, 38} Recent studies by Imayasu et al revealed that some test solutions, especially those containing borate buffers, induced breakdown of TJ's as measured by decreased ZO-1 staining, electron microscopy, and increased PA adhesion specifically targeting “unzippered” open junctions as well overall decreased electrical monolayer resistance.²⁰ (Figure 7). Increased PA adhesion was quantitated by direct PA binding counting and polymerase chain reaction (PCR) analysis.²¹ (Figure 8) Identical changes in MPS-induced breakdown of TJ's were also seen by Yi, et al when LPS (endotoxin) alone was added to the same cultured human corneal epithelial cell line or by direct hypoxic challenge to cultured cells.²² In contrast, Lim et al have recently reported lesser toxic effects of MPS solutions on TJ integrity in monolayer cell layer culture using an alternative cell line with decreased or no toxic effects seen in multi-layered, air-lifted constructs.³⁹ This latter study however did not confirm a normal, differentiated multi-layered corneal phenotype by staining with corneal epithelial-specific cytokeratan markers and reported findings that differ significantly from other recent reports.^{20–22, 38} Taken together, the obvious demonstration of increased PA binding with invasion of broken TJ's in vitro produced by many MPS solutions suggests an urgent need for correlative data from human studies. (Figure 9)

In Vivo human studies of MPS effects on PA binding – No Lens

Li et al performed a prospective, randomized, double-masked, single center, cross-over clinical trial examining the effects of several commercial MPS products on surface cell desquamation

rates and PA binding to exfoliated cells (Figures 10, 11).⁴⁰ Results shown in figures 12 and 13 show that *all* solutions tested elevated PA binding ($P<0.02$) but also decreased surface cell exfoliation rates ($P<0.004$). Taken together, these effects are the same as those produced by human¹⁴ or rabbit²⁸ wear of a low oxygen transmissible soft contact lens using preserved lens care solutions (Renu Multiplus).

Another potential explanation for MPS-driven increases in PA binding to corneal epithelial cells in vitro is the recent intriguing finding of Imayasu et al that three MPS containing boric acid significantly reduced gene activation of corneal mucins (MUC1, MUC4, and MUC16) and also reduced specific expression of MUC16 (Masaki Imayasu, personal communication). These preliminary results suggest the possibility that prolonged use of MPS containing boric acid by soft lens wearers may produce decreased expression of ocular surface mucins leading to increased PA binding and associated risk of microbial infection. Further studies are clearly needed to confirm and extend these novel findings.

As a result of these studies, the next important question is whether MPS solutions have effect (s) on lens-related PA binding in human clinical trials.

MPS effects on PA Binding in Clinical Trials with Varying Lenses and Wearing Schedules

Two prospective, randomized, 13 month, single center, double-masked parallel treatment group clinical trials have been carried out at UT Southwestern Medical Center utilizing identical protocols except for associated lens care solutions: Renu Multiplus^{14, 15} versus Clear Care (preservative-free peroxide).¹⁹ The comparative results are compelling. Utilizing the preserved MPS (shown to open tight junctions and increase PA binding, Figure 9)^{20, 21}, the first month of daily wear was associated with a significant rise in PA binding for both the test hydrogel (Acuvue II) and silicone hydrogel (Pure Vision) soft lenses but not for the test RGP lens (Menicon Z) which utilized non-preserved, sterile Saline;^{14,15} by contrast, when non-preserved Clear Care (peroxide) was used, no rise in PA binding was seen in daily wear by any test silicone hydrogel lens over 1 year.¹⁹ (Figure 14) ^{14–15} These results are clinically significant because half to two-thirds of all lens-related PA infections occur in *daily* wear.^{1–4}

The results seen in extended wear (6 versus 30 nights for Purevision; 30 nights for RGP extended-wear, and 6 night wear of Acuvue II) using Renu Multiplus are shown in figure 15. There is a significant rise in PA binding seen over the first 3 months of wear for both test soft lenses (greater as expected for the lower oxygen transmissible lens, Acuvue II) with a slow decline to baseline at 6 months, versus no elevation of PA binding seen over 13 months of wear of the RGP lens (one month daily wear, 30 night, 12 month extended wear) using non-preserved saline care solution¹⁴. Significantly, the results shown for extended wear of a variety of hyper-oxygen transmitting silicone hydrogel lenses using non-preserved Clear Care (hydrogen peroxide) solution also showed no rise in PA binding in either 6 or 30 nights wear over one year (Figure 16).

Importantly, the results of the one-year clinical trial by Ren et al using Renu Multipurpose (MP) care solution was the first to show *adaptive* effects of lens wear by the corneal epithelium.¹⁴ Quantitative masked findings showed increased PA binding during the first three months for both the hyper and high oxygen transmissible test soft lenses, with a gradual return to baseline by 6–12 months ($p<0.05$). Similar *adaptive* effects were seen over 12 months for central epithelial corneal thickness, superficial cell area, and epithelial surface cell exfoliation (data not shown).¹⁴ Initially, the corneal epithelium showed central thinning; enlargement of surface cell area, and the expected decrease in surface cell exfoliation ($p<0.05$). Subsequently,

epithelial central thickness and cell shedding rates showed partial adaptive recovery, but not surface cell size.¹⁴

Based upon our hypothesis that increases in PA binding to the epithelium induced by contact lens wear at the cellular level should be a surrogate predictor for risk for clinical infectious keratitis as seen in parallel epidemiological studies, the Ren et al data¹⁴ in 2001 made some important novel and testable clinical predictions: (1) less than 6 months wearers (non-adapted) using MPS should manifest a higher relative risk for PA keratitis; (2) using new hyper-oxygen transmissible silicone hydrogel lenses and MPS, there should be no difference in relative risk for PA keratitis between 6 consecutive versus 30 consecutive nights of extended wear; (3) RGP daily or extended wear should have the safest risk profile for lens use versus increasing risk with daily wear soft lenses > extended wear soft lenses.¹⁻⁴ Taken together with supporting results from previous parallel animal studies,^{18, 28} the clinical data for the first-time provided a biological rationale for the long-known relative risk hierarchy for lens-related infections keratitis. Unfortunately ignoring potential confounding MPS effects on PA binding with possible resulting increasing risk, the results were optimistically interpreted at that time to hope that widespread clinical introduction of a new generation of hyper-oxygen transmissible lenses (RGP, Silicone hydrogel) might have the potential to lower overall rates of lens-related microbial keratitis.¹⁴

Epidemiology Studies

Recently, Stapleton et al have comprehensively reassessed the absolute risk of contact lens microbial keratitis, the incidence of vision loss, and risk factors for disease in a robust, prospective, 12-month population based surveillance study in Australia. Disappointingly, the overall incidence rates for microbial keratitis with soft contact lenses remained similar to these previously reported;²⁻⁴ however, the majority of wearers surveyed in Australia used preserved, MPS lens care solutions with lens wear. Importantly however, this study also included a multi-variable risk analysis for all presumed keratitis in extended wear for: (1) age; (2) gender; (3) lens material; (4) nights of continuous wear; (5) socio-economic class of wearers; and (6) for the first time assessed risks for lens wear (greater or less than 6 months use). Validating the Ren et al prediction,¹⁴ the largest single increased risk factor of all variables analyzed in either daily or extended wear was *less than six months of lens wear*. (Odds ratio 4.42; 95% CI 1.31–14.92; p=0.016). The study also validated the Ren et al prediction that 30 night extended wear of soft contact lenses versus 6 night wearing schedules posed no increased risk for microbial keratitis.¹⁴ (P=0.139) Taken together, these results provide strong confirmation for the use of *changes* (increased, decreased) in PA binding produced by lens wear as a maskable, quantitative, non-invasive outcome measure in clinical trials that is predictive for subsequent results of large population studies of risks for microbial keratitis. Specifically, elimination of adverse surface epithelial effects and TJ breakdown caused by use of preserved MPS care solutions, should lower overall risks. The *flat* PA binding curves over one year with peroxide, seen in both daily and extended wear of silicone hydrogel hyper-oxygen transmissible lenses, are identical to those seen in non-preserved care solution use in 30 night extended wear of hyper-oxygen transmissible RGP lenses worn for one year which manifest the lowest rate of daily wear infection (1.1 per 10,000).¹⁻⁴ Comparative results of the second one-year clinical trial by Robertson et al,¹⁹ 2008 using non-preserved MPS solution (Clear Care, hydrogen peroxide) but an investigative protocol otherwise the same as Ren et al¹⁴ who used MPS (Renu Multiplus), supply important support for this concept and predict that when non-preserved care solutions are utilized, the risk for microbial keratitis may become similar for both daily and de novo extended soft lens wear.

In support of verifying these new predictions, Stapleton has most recently reanalyzed prior epidemiological data from the United Kingdom where the overall penetrance of peroxide use

among lens wearers was 14%; and has found that combining one- and two-step solution data showed a *decreased* relative risk for *all* microbial keratitis with peroxide use of about 2.5 times (95% CI, 0.22–0.75, OR 41, referent $p < 0.004$) and also for *severe* infections (90% CI, 0.19–0.91, OR 0.41, referent $p < 0.028$; Fiona Stapleton, personal communication). These preliminary but supportive univariate analysis results make a strong and urgent case for multivariate, case control studies which can further validate these findings, and which could significantly reduce the risk for lens-related corneal infection world wide.

A final important factor in deciding which lens care solutions to recommend to patients to minimize lens-related corneal infection is the important associated question of lens care solution- related increased risk for causing amoebic keratitis. Due to the ongoing lack of *any* current U.S. Food and Drugs Administration (FDA) testing standards for amoeba disinfection of contact lenses (which remain the most common cause of that rare but devastating infection), results of a recent study by Johnson et al⁴¹ from the Communicable Diseases Center in Atlanta, Georgia show that: “the data generated with *A. castellanii*, polyphagia, and *hachetti* (genotype 4) isolated from the recent outbreak⁴²... suggest that only the two commercial contact lens solutions containing hydrogen peroxide... showed disinfecting ability against amoebic cysts.”^{40,41} Taken together then, the currently available paradigm to achieve the safest soft contact lens wear for all patients for the prevention of lens-related, corneal microbial infection (amoeba or *pseudomonas*) is the convergent clinical use of non-preserved, hydrogen peroxide care solutions with lenses of the highest possible oxygen transmission recognizing that the RGP lens type will continue to manifest the lowest possible relative risk compared to soft lenses.

Summary

In summary, how then does the cornea withstand the risk of infection while wearing a foreign body, (contact lens) for varying intervals over many decades with use of solutions that can independently cause surface damage? Two effects of all lens wear appear physiologically non-negotiable: (1) all lens-mediated reduction of surface cell apoptosis; and, (2) the stagnant tear lake under soft lenses which even with lenses of the highest oxygen transmission and no preserved care solution use, cause higher PA binding per unit area of cornea in a thin, stagnant, posterior lens tear film than the surface mechanical effects of their freely movable rigid counterparts.²⁸ The best strategy to minimize future prospective risk appears to be to exploit the paradigm of: “*no lens induced microbial binding...no infection*”, applying this test independently and comprehensively to lens type, wearing schedules, lens polymers, fit, lens oxygen-transmission and commercial care solutions. The goal is to have all contact lens wear, produce no increases in PA or other microbial binding to exfoliated surface epithelial cells versus non lens-wearing controls. The current best way to achieve this is clearly a hyper-oxygen transmissible RGP lens cleaned and then rinsed with sterile preservative-free saline. Unfortunately, approximately 85–90% of patients prefer the comfort of soft contact lenses, and hence will continue to accept increased risk for lens-related microbial keratitis. Results seen in the recent study by Robertson et al 2008 however predict that there should be a good possibility that infection rates in the soft lens wear risk group can finally be reduced overall if hyper-oxygen transmissible hydrogel or silicone hydrogel soft lenses are used in association with non-preserved (hydrogen peroxide) care solutions.¹⁹ This prediction is testable, and robust, multivariate case-control studies are urgently needed. Likewise, the only commercially available care solutions that have amoebic cysticidal activity are also those with hydrogen peroxide. The convergent take home message is then: hyper-oxygen transmissible soft lenses used with hydrogen peroxide care solutions should currently be preferentially recommended by clinicians to our patients for any modality of lens wear.

Acknowledgments

Supported in part by National Eye Institute Grants EY 10738 (HDC), EY 018219, and Infrastructure Grant EY 016664, and an unrestricted grant from Research to Prevent Blindness, Inc., New York, New York. Dr. Cavanagh is a Research to Prevent Blindness Senior Scientist Awardee.

I would like to express my sincere appreciation to the officers and Board of Directors of the Corneal Society for inviting me to present this 35th Lecture in honor of Dr. Ramon Castroviejo. I would also like to acknowledge my debt to all the students, residents and fellows who assisted with the studies that are cited in this lecture with Special Thanks to: Hideji Ichijima, Masaki Imayasu, Patrick Ladage, Susanna Li, David Ren, Kazuaki Yamamoto, Naoka Yamamoto, Nobutaka Yamamoto, and Takashashi Yamamoto. I would also respectfully like to dedicate this Lecture to my old friend and long companion on the quest for the perfect contact lens, Mr. Kyoichi Tanaka of Nagoya, Japan.

References

1. Stapleton F, Keay L, Edwards K, et al. The incidence of contact lens-related microbial keratitis in Australia. *Ophthalmology* 2008;115:1655–1662. [PubMed: 18538404]
2. Schein OD, Glynn RJ, Poggio EC, Seddon JM, Kenyon KR. The relative risk of ulcerative keratitis among users of daily-wear and extended-wear soft contact lenses. A case-control study. *N Engl J Med* 1989;321:773–778. [PubMed: 2671733]
3. Poggio EC, Glynn RJ, Schein OD, et al. The incidence of ulcerative keratitis among users of daily-wear and extended-wear soft contact lenses. *N Engl J Med* 1989;321:779–783. [PubMed: 2770809]
4. Cheng KH, Leung SL, Hoekman HW, et al. Incidence of contact-lens-associated microbial keratitis and its related morbidity. *Lancet* 1999;354:181–185. [PubMed: 10421298]
5. Pachigolla G, Blomquist P, Cavanagh HD. Microbial keratitis pathogens and antibiotic susceptibilities: a 5-year review of cases at an urban county hospital in North Texas. *Eye Contact Lens* 2007;33:45–49. [PubMed: 17224678]
6. Reichert R, Stern G. Quantitative adherence of bacteria to human corneal epithelial cells. *Arch Ophthalmol* 1984;102:1394–1395. [PubMed: 6433864]
7. Stern G, Lubniewski A, Allen C. The interaction between *Pseudomonas aeruginosa* and the corneal epithelium. *Arch Ophthalmol* 1985;103:1221–1225. [PubMed: 3927878]
8. Hazlett LD, Moon M, Berk RS. In vivo identification of sialic acid as the ocular receptor for *Pseudomonas aeruginosa*. *Infect Immun* 1986;51:687–689. [PubMed: 3080374]
9. Hazlett LD, Moon MM, Strejc M, Berk RS. Evidence for N-acetylmannosamine as an ocular receptor for *P. aeruginosa* adherence to scarified cornea. *Invest Ophthalmol Vis Sci* 1987;28:1978–1985. [PubMed: 3119512]
10. Fletcher EL, Fleiszig SMJ, Brennan NA. Lipopolysaccharide in adherence of *Pseudomonas aeruginosa* to the cornea and contact lenses. *Invest Ophthalmol Vis Sci* 1993;34:1930–1936. [PubMed: 8491546]
11. Fenton MJ, Golenbock DT. LPS-binding proteins and receptors. *J Leukoc Biol* 1998;64:25–32. [PubMed: 9665271]
12. Latkovic S, Nilsson SE. The effect of high and low Dk/L soft contact lenses on the glycocalyx layer of the corneal epithelium and on the membrane associated receptors for lectins. *CLAO J* 1997;23:185–191. [PubMed: 9240830]
13. Cavanagh HD, Ladage PM, Li SL, et al. Effects of daily and overnight wear of a novel hyper oxygen-transmissible soft contact lens on bacterial binding and corneal epithelium: a 13-month clinical trial. *Ophthalmology* 2002;109:1957–1969. [PubMed: 12414399]
14. Ren DH, Yamamoto K, Ladage PM, et al. Adaptive effects of 30-night wear of hyper-O₂ transmissible contact lenses on bacterial binding and corneal epithelium. A 1-year clinical trial. *Ophthalmology* 2002;109:27–40. [PubMed: 11772575]
15. Ladage PM, Yamamoto K, Ren DH, et al. Effects of rigid and soft contact lens daily wear on corneal epithelium, tear lactate dehydrogenase, and bacterial binding to exfoliated epithelial cells. *Ophthalmology* 2001;108:1279–1288. [PubMed: 11425688]
16. Ren DH, Petroll WM, Jester JV, Ho-Fan J, Cavanagh HD. Short-term hypoxia downregulates epithelial cell desquamation in vivo, but does not increase *pseudomonas aeruginosa* adherence to exfoliated human corneal epithelial cells. *CLAO J* 1999;2:73–79. [PubMed: 10344293]

17. Ren DH, Petroll WM, Jester JV, Ho-Fan J, Cavanagh HD. The relationship between contact lens oxygen permeability and binding of *Pseudomonas aeruginosa* to human corneal epithelial cells after overnight and extended wear. *CLAO J* 1999;25:80–100. [PubMed: 10344294]
18. Ren H, Petroll WM, Jester JV, et al. Adherence of *Pseudomonas aeruginosa* to shed rabbit corneal epithelial cells after overnight wear of contact lenses. *CLAO J* 1997;23:63–68. [PubMed: 9001775]
19. Robertson DM, Petroll WM, Cavanagh HD. The effects of nonpreserved care solutions on 12 months of daily and extended silicone hydrogel contact lens wear. *Invest Ophthalmol Vis Sci* 2008;49:7–15. [PubMed: 18172068]
20. Imayasu M, Shimizu H, Shimada S, Suzuki T, Cavanagh HD. Effects of multipurpose contact-lens care solutions on adhesion of *Pseudomonas aeruginosa* to corneal epithelial cells. *Eye Contact Lens* 2009;2:98–104. [PubMed: 19265332]
21. Imayasu M, Shiraishi A, Ohashi Y, Shimada S, Cavanagh HD. Effects of multipurpose solutions on corneal epithelial tight junctions. *Eye Contact Lens* 2008;34:50–55. [PubMed: 18180685]
22. Yanai R, Ko J-A, Morishige N, Chikama T-I, Ichijima H, Nishida T. Disruption of zonula occludens-1 localization in the rabbit corneal epithelium by contact lens-induced hypoxia. *Invest Ophthalmol Vis Sci* 2009;50:4605–4610. [PubMed: 19474401]
23. Fleiszig SMJ, Zaidi TS, Pier GB. *Pseudomonas aeruginosa* invasion of and multiplication within corneal epithelial cells in vitro. *Infect Immun* 1995;63:4072–4077. [PubMed: 7558321]
24. Yamamoto N, Yamamoto N, Petroll WM, Cavanagh HD, Jester JV. Internalization of *Pseudomonas aeruginosa* is mediated by lipid rafts in contact lens-wearing rabbit and cultured human corneal epithelial cells. *Invest Ophthalmol Vis Sci* 2005;46:1348–1355. [PubMed: 15790901]
25. Yamamoto N, Yamamoto N, Petroll WM, Jester JV, Cavanagh HD. Regulation of *Pseudomonas aeruginosa* internalization after contact lens wear in vivo and in serum-free culture by ocular surface cells. *Invest Ophthalmol Vis Sci* 2006;47:3430–3440. [PubMed: 16877413]
26. Yamamoto N, Yamamoto N, Jester JV, Petroll WM, Cavanagh HD. Prolonged hypoxia induces lipid raft formation and increases *Pseudomonas* internalization in vivo after contact lens wear and lid closure. *Eye Contact Lens* 2006;32:114–120. [PubMed: 16702863]
27. Zaidi T, Bajmoczy M, Zaidi T, Golan DE, Pier GB. Disruption of CFTR-dependent lipid rafts reduces bacterial levels and corneal disease in a murine model of *Pseudomonas aeruginosa* keratitis. *Invest Ophthalmol Vis Sci* 2008;49:1000–1009. [PubMed: 18326723]
28. Imayasu M, Petroll WM, Jester JV, Patel SK, Ohashi J-I, Cavanagh HD. The relation between contact lens oxygen transmissibility and binding of *Pseudomonas aeruginosa* to the cornea after overnight wear. *Ophthalmology* 1994;101:371–388. [PubMed: 8115159]
29. Fleiszig SMJ, Efron N, Pier GB. Extended contact lens wear enhances *Pseudomonas aeruginosa* adherence to human corneal epithelium. *Invest Ophthalmol Vis Sci* 1992;33:2908–2916. [PubMed: 1526741]
30. Yamamoto K, Ladage PM, Ren DH, et al. Effect of eyelid closure and overnight contact lens wear on viability of surface epithelial cells in rabbit cornea. *Cornea* 2002;21:85–90. [PubMed: 11805514]
31. Li L, Ren DH, Ladage PM, et al. Annexin V binding to rabbit corneal epithelial cells following overnight contact lens wear or eyelid closure. *CLAO J* 2002;28:48–54. [PubMed: 11838990]
32. Ren DH, Petroll WM, Jester JV, Cavanagh HD. The effects of rigid gas permeable contact lens wear on proliferation of rabbit corneal and conjunctival epithelial cells. *CLAO J* 1999;25:136–141. [PubMed: 10444048]
33. Ladage PM, Yamamoto K, Ren DH, et al. Proliferation rate of rabbit corneal epithelium during overnight rigid contact lens wear. *Invest Ophthalmol Vis Sci* 2001;42:2804–2812. [PubMed: 11687521]
34. Ladage PM, Jester JV, Petroll WM, Bergmanson JPG, Cavanagh HD. Vertical movement of epithelial basal cells toward the corneal surface during use of extended-wear contact lenses. *Invest Ophthalmol Vis Sci* 2003;44:1056–1063. [PubMed: 12601029]
35. Yamamoto K, Ladage PM, Ren DH, et al. Bcl-2 expression in the human cornea. *Exp Eye Res* 2001;73:247–255. [PubMed: 11446775]
36. Yamamoto K, Ladage PM, Ren DH, et al. Effects of low and hyper Dk rigid gas permeable contact lenses on Bcl-2 expression and apoptosis in the rabbit corneal epithelium. *CLAO J* 2001;27:137–143. [PubMed: 11506438]

37. Yamamoto K, Ladage PM, Ren DH, Li L, Jester JV, Cavanagh HD. Epitope variability of Bcl-2 immunolocalization in the human corneal epithelium. *CLAO J* 2001;27:221–224. [PubMed: 11725986]
38. Yi X-J, Wang Y, Yu F-SX. Corneal epithelial tight junctions and their response to lipopolysaccharide challenge. *Invest Ophthalmol Vis Sci* 2000;41:4093–4100. [PubMed: 11095601]
39. Lim MJ, Hurst RK, Konynenbelt BJ, Ubels JL. Cytotoxicity testing of multipurpose contact lens solutions using monolayer and stratified cultures of human corneal epithelial cells. *Eye Contact Lens* 2009;35:287–296. [PubMed: 19726996]
40. Li SL, Ladage PM, Yamamoto T, Petroll WM, Jester JV, Cavanagh HD. Effects of contact lens care solutions on surface exfoliation and bacterial binding to corneal epithelial cells. *Eye Contact Lens* 2003;29:27–30. [PubMed: 12769153]
41. Johnson SP, Sriram R, Qvarnstrom Y, et al. Resistance of *Acanthamoeba* cysts to disinfection in multiple contact lens solutions. *J Clin Microbiol* 2009;47:2040–2045. [PubMed: 19403771]
42. Verani JR, Lorick SA, Yoder JS, et al. National outbreak of *Acanthamoeba* keratitis associated with use of a contact lens solution, United States. *Emerg Infect Dis* 2009;15:1236–1242. [PubMed: 19751585]

Prevention of microbial keratitis: A Zero Damage Game

GOAL: None of the above should change the corneal epithelium and enhance the microbial agent's ability to stick or invade.

Figure 1.

Prevention of microbial keratitis: a zero damage game. Figure adapted from Robertson DM, Petroll WM, Cavanagh HD. The effects of nonpreserved care solutions on 12 months of daily and extended silicone hydrogel contact lens wear. *Invest Ophthalmol Vis Sci* 2008;49:7–15 (Copyright © Association for Research in Vision and Ophthalmology).

Figure 2.

Propidium Iodide (PI) staining of corneal epithelial nuclei (red) and PA (red), FITC-conjugated beta cholera toxin staining of lipid rafts (green). A: Normal rabbit cornea (no lens); B: 24 hours of PMMA lens wear, no PA; C: 24 hours of PMMA lens wear, 30 minutes after PA infection; D: 24 hours of PMMA lens wear; 1 hour after PA infection. Figure adapted from Yamamoto N, Yamamoto N, Petroll WM, Cavanagh HD, Jester JV. Internalization of *Pseudomonas aeruginosa* is mediated by lipid rafts in contact lens-wearing rabbit and cultured human corneal epithelial cells. *Invest Ophthalmol Vis Sci* 2005;46:1348–1355 (Copyright © Association for Research in Vision and Ophthalmology).

Figure 3.

PI staining of corneal epithelial nuclei (red) and FITC-conjugated beta cholera toxin staining of lipid rafts (green). A: Note the presence of rafts in the conjunctival and limbal epithelium in the control eye, no rafts were noted in the central or peripheral corneal epithelium in the non-lens wearing condition. B: Following 24 hours of PMMA lens wear, rafts appeared in the peripheral and central corneal epithelium; PA preferentially localized to these rafts. No PA adherence was seen with rafts localized to the conjunctival or limbal epithelium (arrows). Figure adapted from Yamamoto N, Yamamoto N, Petroll WM, Cavanagh HD, Jester JV. Internalization of *Pseudomonas aeruginosa* is mediated by lipid rafts in contact lens-wearing rabbit and cultured human corneal epithelial cells. *Invest Ophthalmol Vis Sci* 2005;46:1348–1355 (Copyright © Association for Research in Vision and Ophthalmology).

In Vivo Rabbit Studies

Figure 4.

Results from animal studies show that at the same level of oxygen transmissibility, rigid lenses produce inherently greater epithelial surface damage but induce significantly less PA adhesion than their soft lens counterparts. ($p < 0.05$). Figure adapted from Imayasu et al, 1994, with permission.

Corneal Epithelial Cell Collection

Total surface corneal PA binding correlates with PA binding to exfoliated corneal epithelial cells (rabbit model, $p < 0.01$)

Figure 5.

An eye irrigation chamber was used to wash exfoliated epithelial cells from the corneal surface using warmed sterile saline. Following epithelial cell collection, cells were stained with acridine orange and visualized under an epifluorescent microscope, allowing for quantitation of the number of cells collected and the number of adherent PA per cell. Figure adapted from Ren et al, 1997, with permission.

Test Lenses:

Rigid gas permeable
lenses (n=3):
Dk 40, 66, 146

Soft lenses (n=7):
Dk 10, 23, 33, 51,
64, 83*, 96*
*Silicone hydrogels

Randomized,
prospective,
masked clinical
trial; 110 patients

Ren et al. *CLAO J*
1999; 25: 80-100

Figure 6.

The clinical relationship between lens oxygen parameters and increased PA binding to exfoliated corneal cells. Adapted from Ren et al, 1999, with permission.

Multipurpose solution	Preservative	Surfactant	Buffer
MenCare S	PHMB	Macrogolglycerol hydroxystearate	None
Renu MP	PHMB	Poloxamine	Boric acid
Optifree P.	POLYQUAD	Poloxamine	Boric acid

Figure 7.

Summary table detailing MPS components: preservative, surfactant, and buffer, utilized by three commercially available MPS solutions. Note that both Renu MP and Optifree P. contain Boric acid as a buffering agent. Table adapted from Imayasu et al, 2008, with permission.

Quantitative PA Binding Results

The 2 test solutions containing boric acid buffers showed greater PA adhesion rates assessed by direct count and confirmed by real-time PCR

Figure 8.

Quantitative analysis of PA adhesion to corneal epithelial cells following treatment with the three MPS outlined in Figure 7. Importantly, both solutions containing Boric acid showed the largest increase in PA adhesion compared to the control ($p < 0.05$). Figure adapted from Imayasu et al, 2009, with permission

ReNu MP

OptiFree P

Figure 9.

Scanning electron micrograph showing PA selectively adhering and migrating toward areas of tight junction breakdown in monolayer epithelial cell cultures treated with boric acid containing MPS. Figure adapted from Imayasu et al, 2009, with permission.

Chemically Preserved Contact Lens Care Solutions: NO LENS

- Li, S et al. *Eye & Contact Lens* 2003; 29: 27-30.
- Prospective, double-masked, randomized cross-over clinical trial.
- N=20 patients (10 male; 10 female).
- 4 lens care solutions: ReNu Multiplus; Optifree Express; Complete Blink-n-Clean, and Lens Plus Rewetting drops.
- Outcome measures: PA binding to exfoliated surface epithelial cells; shedding rates.

Figure 10.

Study outline for a clinical trial investigating the effects of chemically preserved care solutions on the surface of the eye in the absence of a contact lens. PA adhesion rates and surface epithelial exfoliation rates were assessed as previously described.

Test Solutions

Test Solutions

1. Renu MP
2. Optifree
3. Blink-N-Clean
4. Lens Plus

Preservatives

1. Dymed
2. Polyquad
3. PHMB
4. Boric Acid

- 6 times a day/4 days/tested 5th day
- 1 week recovery/next solution tested
- Randomized to all 4 solutions
- Double-masked as to test solution and binding assays

Figure 11.

Study outline for a clinical trial investigating the effect of topically applied chemically preserved care products in the absence of a contact lens. Figure adapted from Li et al, 2003, with permission.

Solution Effects on PA Binding

Figure 12.

All four chemically preserved solutions demonstrated an increase in PA adhesion when applied directly to the eye in the absence of a contact lens ($p < 0.02$). Figure adapted from Li et al, 2003, with permission.

Solution Effects on Cell Shedding

Figure 13.

All four solutions concomitantly decreased epithelial surface cell shedding in the absence of a confounding lens effect ($p < 0.004$). Figure adapted from Li et al, 2003, with permission.

PA Binding: 1 Month Lens Wear

Figure 14.

Results from a one year clinical trial demonstrate that silicone hydrogel lenses, worn daily or extended wear over a one month period, failed to increase PA adhesion when used in conjunction with a preservative free (peroxide) solution. Acuvue 2 data shown here as an historical control for comparison. Figure adapted from Robertson DM, Petroll WM, Cavanagh HD. The effects of nonpreserved care solutions on 12 months of daily and extended silicone hydrogel contact lens wear. *Invest Ophthalmol Vis Sci* 2008;49:7–15 (Copyright © Association for Research in Vision and Ophthalmology).

Figure 15.

Results from a one year clinical trial showing an increase in PA binding associated with lens wear and use of a chemically preserved MPS (Renu MP). PA rates were stratified by lens type and showed an adaptive return to baseline during the first 6 months of lens wear. Figure adapted from Ren et al, 2002.

PA Binding: Peroxide

Figure 16.

In contrast to Figure 15, silicone hydrogel lenses in combination with a preservative free solution (peroxide) failed to show an increase in PA adhesion when followed over one year. Acuvue 2 data shown here as an historical control for comparison. Figure adapted from Robertson DM, Petroll WM, Cavanagh HD. The effects of nonpreserved care solutions on 12 months of daily and extended silicone hydrogel contact lens wear. *Invest Ophthalmol Vis Sci* 2008;49:7–15 (Copyright © Association for Research in Vision and Ophthalmology).