The Effect of Nonpreserved Care Solutions on 12 Months of Daily and Extended Silicone Hydrogel Contact Lens Wear

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Purpose. To determine the effects of nonpreserved care solutions on human corneal epithelium in long-term daily wear (DW) compared with overnight (extended) wear (EW) of hyper–oxygen-permeable silicone hydrogel contact lenses.

Methods. This was a prospective, randomized, double-masked, single-center, parallel treatment group clinical trial (NCT 00344643). One hundred twenty-one patients completed the 13 month study: (1) Lotrafilcon A (30 night EW, $n = 29$; DW, $n = 32$); (2) Galyfilcon A (DW, $n = 20$); and (3) Lotrafilcon B (6 night EW, $n = 20$; DW, $n = 21$). Irrigation chamber collection of corneal surface cells (OD) and confocal microscopy (OS) were performed at baseline, 1 week; and 1, 3, 6, 9, and 12 months of EW. The main outcome measures were: (1) *Pseudomonas aeruginosa* (PA) binding to exfoliated corneal surface cells; (2) central epithelial thickness (CET); and (3) epithelial surface cell exfoliation rate (desquamation).

Results. DW had no significant effect on CET; there was a decrease in CET with EW that recovered (adapted) over 1 year (Lotrafilcon B, $P < 0.05$). All lens wear (DW, EW) decreased desquamation with adaptive effects over 1 year ($P < 0.001$). There was no significant difference in PA binding between lenses or modality of wear.

Conclusions. PA binding to corneal epithelial cells is a prerequisite for infection, and no binding indicates no lens-enhanced risk of infection. In contrast to prior studies of preserved lens-care products, the absence of a change in the PA binding data results predict that the risk for PA CTL-keratitis should be similar for daily and extended silicone hydrogel lens wear over 1 year when preservative-free care solutions are used. (ClinicalTrials.gov number, NCT00344643.) (*Invest Ophthalmol Vis Sci.* 2008;49:7–15) DOI:10.1167/iovs.07-0940

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Microbial keratitis (MK) is a potentially blinding disease characterized clinically by the presence of a light-blocking infiltrate with an overlying epithelial defect. Risk factors for MK include trauma, preexisting ocular disease, and commonly, contact lens wear, with *Pseudomonas aeruginosa* (PA) as the primary causative organism.[1–3] In the 1980s, the use of hydrogel contact lenses on an extended-wear basis led to an alarming increase in MK. Hallmark studies at that time reported the incidence of MK in hydrogel lens wearers as approximately 4.1 cases per 10,000 persons per year for daily wear (DW) and 20.9 per 10,000 persons per year for extended wear (EW) in the United States,[4,5] and later as 2.16 per 10,000 persons per year for DW and 10.0 per 10,000 persons per year for EW of disposable soft lenses in Sweden.[6] Irrespective of differences in the incidence rates reported, all studies concluded that EW of hydrogel contact lenses was associated with an increased risk of MK. Additional studies evaluating the risk of MK in overnight wear estimated an 8.25-fold excess risk with EW[7] and that continuous wear for periods longer than 6 days led to an increase in morbidity of the disease.[8] In a study in the Netherlands in the late 1990s, the investigators re-examined the incidence of MK, again estimating rates of 3.5 per 10,000 persons per year for DW and 20.0 per 10,000 persons per year for EW, suggesting that despite advances in disposable lens materials and the identification of potential risk factors, MK rates remained unchanged.[9]

The cumulative results of these studies led to the development of new lens materials to address the needs of the ocular surface and ultimately achieve a more biocompatible lens–cornea relationship. Oxygen was proposed as a key mediator of corneal epithelial swelling and in maintaining the homeostasis of the epithelium,[10–13] and lens-oxygen transmissibility was shown to have an important role in induction of increased PA binding to corneal epithelial cells, with bacterial binding being a prerequisite for infection.[14–16] Despite the introduction of increased oxygen-transmissible silicone hydrogel contact lenses into general clinical use, however, more recent epidemiological data on incident rates of MK still show persistently unchanged rates of MK after EW.[17–19] These latter findings suggest that additional factor(s) besides lens oxygen transmission play a critical role in the pathogenesis of MK. Such factors include the mechanical properties of the lens, alterations in the biochemistry of the tear film, tear stagnation beneath the lens, ocular surface inflammation, and solution toxicity from the use of chemically preserved multipurpose lens-care solutions (MPSs). Significantly, recent reports on the use of MPSs suggest an important relationship between chemically preserved multipurpose lens-care solutions, ocular surface inflammation, and corneal infiltrative events, including MK.[20–22] The purpose of this study was to evaluate the effects of nonpreserved care solutions on central corneal

Table 1. Inclusion and Exclusion Criteria

All lenses	Age: 21–38 years
	Myopia: −1.00 to −6.00 D with degree of regular astigmatism <0.75 D
	Any gender, race, or national origin accepted; minority participation encouraged
	No history of ocular allergies
	No lens wear 1 month before entry into study
	Visual acuity of 20/30 or better with the test lens
	Normal ocular surface, cornea, and conjunctiva
	Schirmer test above 3.00 mm wetting in 3 minutes
	No blepharitis or other eyelid problems
	Intraocular pressure less than 21 mm Hg
	Normal crystalline lens
	Normal appearance of retina and optic nerve
	No current use of ocular or systemic medication
	Females not pregnant or with plans to become pregnant

epithelial thickness, surface cell shedding rates, and PA binding to shed corneal epithelial cells in highly oxygen-transmissible silicone hydrogel contact lenses worn in daily and continuous mode over 12 months and to compare these results to identical previous clinical trials of MPSs.

Material and Methods

Study Population

One hundred eighty-eight patients who met the inclusion criteria (Table 1) were enrolled in this study. All patients underwent an initial comprehensive ocular examination and were required to undergo a 30-day washout period during which they abstained from contact lens wear. Patient characteristics were similar for age, race, gender, and refractive error among test groups (Table 2). All patients signed an informed consent approved by the Institutional Review Board of the University of Texas Southwestern Medical Center at Dallas. One hundred twenty-one patients completed the study; reasons for exit are listed in Table 3. The primary reason for exit was loss to follow-up. There were no significant adverse reactions reported.

Study Protocol

The study design is the same as that reported in previous studies[23–25] and meets the Helsinki Standards for Clinical Trials. The study was registered with the National Clinical Trial Registry (NCT 00344643). The overall design is described in detail in Table 4. Patients were assessed at baseline and asked to return to the clinic at 1 week and 1, 3, 6, 9, and 12 months. At each visit, contact lens fit was assessed by biomicroscopy. Immediately after lens removal, ocular irrigation was performed on the right eye, and in vivo confocal microscopy was performed on the left eye, to test the following outcome measures: (1) binding to exfoliated cells; (2) surface epithelial cell desquamation; and (3) central corneal epithelial thickness.

Contact Lenses

Sample size calculations to detect significant differences were derived from data obtained in previous studies.[23–25] To detect a significant change in PA binding of one half the magnitude compared with baseline control with a power of ≥0.8, a minimum of 20 subjects per lens group was required. Studies have confirmed that bacterial binding is indirectly related to lens oxygen, making it more difficult to detect changes in binding rates as the oxygen transmissibility increases, thus a higher number of subjects were used in the highest oxygen-transmissible lens group to ensure adequate sensitivity in detection of potential alterations in binding rates. Additional patients were enrolled to compensate for potential dropouts throughout the 12 month study. In total, 42 patients were fit in Lotrafilcon A (CIBA Vision, Duluth, GA) 30 night (30N) EW; 50 patients in Lotrafilcon A DW; 31 patients in Lotrafilcon B 6 night (6N) EW; 31 patients in Lotrafilcon B DW; and 34 patients in Galyfilcon A DW (Johnson & Johnson Vision Care, Jacksonville, FL). Patients were randomized to a contact lens and wearing modality with a random number table (Excel; Microsoft Corp., Redmond, WA). The specifications for the highly oxygen-transmissible silicone hydrogel test contact lenses are listed in Table 5. Hyper–oxygen-transmissible contact lenses were chosen for this study as they are now the standard of care in clinical contact lens practice. For the DW modality, all study patients were instructed to insert contact lenses in the morning on awakening and to remove lenses in the evening before bed (10–16 hours of lens wear per day). For extended-wear modality, all study patients were instructed to remove their lenses on awakening on day 7 or 32 and to resume lens wear after 24 hours. To eliminate confounding solution preservative artifacts, all patients were dispensed a hydrogen peroxide–based system (Clear Care; CIBA Vision) and nonpreserved saline.

Outcome Measures

For all outcome measures, observers collecting clinical data were masked from knowledge of test lens type, wearing modality, and time point. All clinical data, including outcome measures, were coded with a combination of letters and numbers, to mask laboratory personnel. After the conclusion of the study, the code was broken to allow for analysis of the data. As established in the previous studies,[23–25] to control for daily circadian fluctuations in surface epithelial cell desquamation rates, all samples were collected between 9 and 12 AM.

Surface Epithelial Cell Desquamation Rates and Bacterial Adherence

To assess surface epithelial cell desquamation rates, we collected corneal epithelial cells from the right eye of each patient noninva-

Table 2. Patient Characteristics

	Lotrafilcon A 30 Night EW	Lotrafilcon A DW	Lotrafilcon B 6 Night EW	Lotrafilcon B DW	Galyfilcon A DW
Age (y) Mean ± SD	28.3 ± 6.3	28.3 ± 6.8	29.2 ± 5.4	29.2 ± 7.1	28.8 ± 5.3
Gender					
Female	33	34	27	25	23
Male	17	8	4	6	11
Race					
Asian	7	2	7	3	6
Black	13	16	8	6	8
Hispanic	12	2	8	9	8
White	18	18	8	13	12
Contact lens power					
Mean ± SD	−3.39 ± 1.77	−3.17 ± 1.62	−2.94 ± 1.60	−3.44 ± 1.64	−2.67 ± 1.09

TABLE 3. Enrollment

	Lotrafilcon A 30 Night EW	Lotrafilcon A DW	Lotrafilcon B 6 Night EW	Lotrafilcon B DW	Galyfilcon A DW
Enrolled	42	50	31	31	34
Total completed 12 months	29	31	20	21	20
% Completing 12 months	69.0	62.0	64.5	67.7	58.8
Exited before baseline	4	1	4	0	1
% Exiting before baseline	9.5	2.0	12.9	0	2.9
Reasons for exit					
Loss to follow-up	5	9	2	5	4
Pregnancy	0	0	0	0	1
CL Intolerance	1	0	0	1	1
GPC	0	0	1	0	0
Dry Eye	0	1	1	0	0
Allergic conjunctivitis	1	0	0	1	0
Noncompliance	1	0	0	0	0
Solution irritation	0	1	0	0	1
Trauma	0	0	0	0	0
Study terminated*	1	7	3	3	6

* Additional patients were enrolled to compensate for potential dropouts throughout the 12-month study. Once 20 patients completed 12 months of lens wear in all groups, the study was terminated, and the remaining patients exited.

sively in vivo by using a corneal irrigation chamber.[26] Briefly, 9 mL of saline was used to irrigate the front surface of the cornea gently over a period of 1 minute. Exfoliated cells were then incubated in PA, strain ATCC 27853 (American Type Culture Collection, Manassas VA), which is fully infectious to the cornea and has been used as the standard test organism in experimental rabbit[15,27] and human[23–25] bacterial binding studies. The validity of correlating PA binding to exfoliated corneal epithelial cells to total corneal epithelial PA binding has been established.[15] Exfoliated cells were further stained with acridine orange (Sigma-Aldrich, St. Louis, MO) and visualized with an epifluorescence microscope (Diaplan; Leitz, Wetzlar, Germany), to (1) examine the total number of bound PA bacteria per corneal epithelial cell (PA binding), and (2) to count the total number of desquamated corneal epithelial cells (exfoliation rate/min) under an oil immersion lens at 630× magnification. As in previous studies, rare corneal epithelial cells adherent to the contact lens were not included in the analysis.[28]

Corneal Epithelial Thickness

In vivo tandem scanning confocal microscopy was performed on the left eye of each patient to determine the thickness of the central corneal epithelium, according to previously published methods.[23–25] Measurements of direct central epithelial thickness were made with confocal microscopy through focusing (CMTF) software.[29]

Statistical Analysis

As reported in three previous publications,[23–25] the results of all outcome measures were compared for each contact lens group separately to the prelens, baseline values with a one-way repeated-measures analysis of variance (ANOVA), and a subsequent post hoc multiple comparison test (Student's Newman-Keuls test; SigmaStat ver. 3.1; Systat Software, Inc., San Jose, CA). Statistical significance was set at $P < 0.05$. In addition, differences between test groups were compared by using a two-way repeated-measures analysis of variance, holding wearing modality and time as the independent variables. Data are presented as the mean ± SD. An appropriate general linear model was used to handle the occasional missing data point.

RESULTS

PA Binding

Figure 1A and Table 6 demonstrate PA adherence (bacteria/cell) for all test groups at baseline and after 1 month of lens

TABLE 4. Overall Study Design

Visit	Time Interval	Procedure
1	0	Informed consent/HIPAA authorization, comprehensive eye examination; glasses ordered
2	0	Randomization, contact lens trial fitting, order contact lenses; at least 1 month without contact lens wear; data collection, visit 1: prelens baseline of the three outcome measures
3	1 Week	Contact lens check-up
4	1 Month	Data collection OD: PA binding, CDR/OS: TSCM
5	3 Months	Data collection OD: PA binding, CDR/OS: TSCM
6	6 Months	Data collection OD: PA binding, CDR/OS: TSCM
7	9 Months	Data collection OD: PA binding, CDR/OS: TSCM
8	12 months	Data collection OD: PA binding, CDR/OS: TSCM

PA, *Pseudomonas aeruginosa;* CDR, cell desquamation rate; TSCM, tandem scanning confocal microscopy.

TABLE 5. Silicone Hydrogel Test Contact Lenses

Material	Trade Name*	% Water	Dk_{total}†	Dk/t_{total}‡	Base Curve
Lotrafilcon A	CIBA Focus Night & Day	24	140	175	8.4/8.6
Galyfilcon A	Acuvue Advance	47	60	85	8.3/8.7
Lotrafilcon B	CIBA O_2 Optix	33	110	135	8.6

* CIBA Vision, Duluth, GA; Acuvue, Johnson & Johnson Vision Care, Jacksonville, FL.
† Dk_{total} oxygen permeability, unit: $\times 10^{-11}$ (cm^2/sec)(mL O_2/mL mmHg).
‡ Dk/t_{total} oxygen transmissibility, unit $\times 10^{-9}$ (cm/sec)(mL O_2/mL mmHg).

wear. There was no change in PA adherence for any silicone hydrogel test lens, regardless of mode of wear. Previously published data are included for PA adherence after DW of an Acuvue 2 hydrogel lens as a historical control showing the typical increase in binding during the initial month of lens wear ($P < 0.001$, one-way ANOVA).[24] Figure 1B and Table 6 demonstrate PA adherence for all test groups over 12 months of lens wear. Similar to the 1-month data, a one-way ANOVA demonstrated no effect on PA adherence for any test lens or mode of wear. Again, previous data for PA adherence after Acuvue 2 hydrogel EW are included as a historical control, which showed an initial increase in PA adherence at 1 and 3 months of lens wear ($P < 0.05$, one-way ANOVA), followed by an apparent adaptive return to baseline values.[25] A two-way analysis of variance further showed that there was no difference between individual lens groups, regardless of modality of wear over 12 months ($P = 0.051$), and there was no significant interaction between lens and visit ($P = 0.959$).

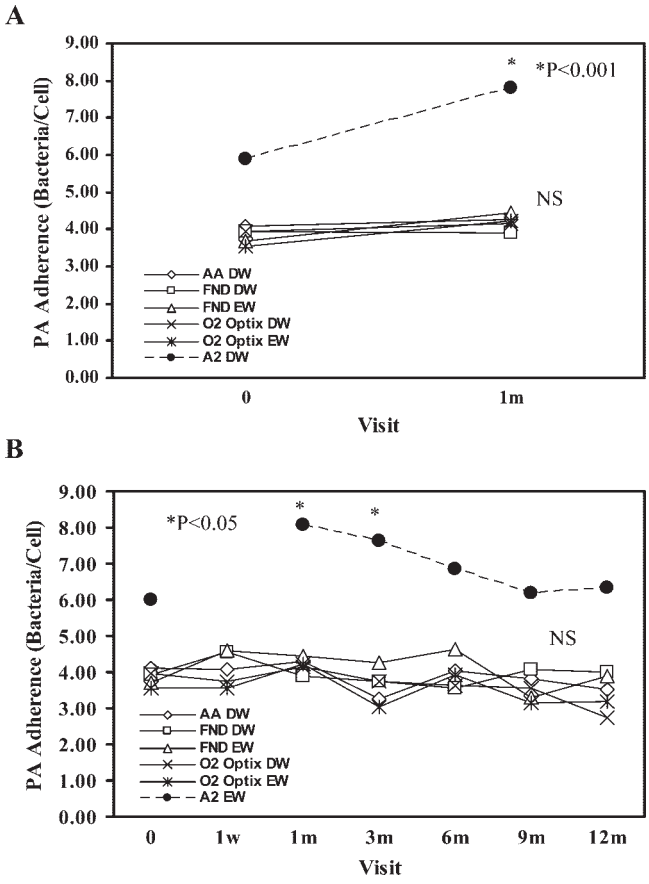

FIGURE 1. PA adherence to desquamated corneal epithelial cells for all test lenses and mode of wear after 1 (**A**) and 12 (**B**) months of lens wear. NS, not significant. Previous data for the Acuvue 2 lens were included as a historical control in DW (**A**, $^*P < 0.001$, one-way ANOVA) and EW (**B**, $^*P < 0.05$) modes.[24,25] For clarity, only the means of the data are represented. Results ± SD are shown in Table 6.

Corneal Epithelial Thickness

CMTF measurements of corneal central epithelial thickness (CET) demonstrated a significant decrease after Lotrafilcon B 6N EW at 1 month of lens wear ($P = 0.026$, one-way ANOVA; Fig. 2A, Table 7). While the Lotrafilcon A 30N EW appeared to decrease thickness, this change was not statistically significant. None of the lenses worn in DW mode demonstrated a significant long-term effect on corneal epithelial thickness. This finding was similar to previously reported studies demonstrating no significant effect on CET after 1 month of Acuvue 2 or silicone hydrogel lens DW.[24] These data are included in Figure 2A as the historical control. When observed over 12 months of lens wear, Lotrafilcon B 6N EW significantly decreased CET at 1, 3, and 6 months of lens wear, with a partial, adaptive recovery ($P < 0.05$, one-way ANOVA; Fig. 2B, Table 7). Daily lens wear had no significant effect on thickness over time. Again, included as a historical control, the Acuvue 2 lens worn in 6N EW mode showed a persistent reduction in epithelial thickness over 12 months ($P < 0.0001$, one-way ANOVA) with minimum recovery.[25]

Surface Epithelial Cell Desquamation

Measurements of surface epithelial desquamation demonstrated a decrease in the rate of desquamation from baseline to 1 month of lens wear ($P < 0.001$, two-way ANOVA, Fig. 3A, Table 8); however, there was no difference between lens type or mode of wear ($P = 0.602$). From previous studies, the Acuvue 2 hydrogel lens also demonstrated a reduction in desquamation rates compared with baseline ($P < 0.0001$, one-way ANOVA).[24] In a 12-month follow-up, there was a similar decrease in epithelial desquamation rates among visits ($P < 0.001$, two-way ANOVA). Again, there was no difference between lens type or mode of wear. Similarly, the Acuvue 2 lens worn in extended wear, included in Figure 3B as an historical control, showed a reduction in desquamation at all time points ($P < 0.05$, one-way ANOVA).[25] There was no interaction between lens type and visit ($P = 0.845$).

DISCUSSION

It has long been established that chemically preserved contact lens solutions have adverse effects on the corneal epithelium,[30,31] with a significant correlation between preservative-induced epithelial toxicity and lactate dehydrogenase release.[32] With the current influx of increased reports of contact-lens–related MK associated with preserved contact lens solution usage, the effects of these care products on the ocular surface is rapidly becoming an area of heightened scrutiny. When combined

TABLE 6. PA Adherence as a Function of Time for Each Lens Group and Mode of Wear

	Baseline	1 Week	1 Month	3 Months	6 Months	9 Months	12 Months
Acuvue Advance DW	4.10 ± 2.29	4.07 ± 2.55	4.28 ± 1.77	3.25 ± 1.95	4.02 ± 2.34	3.82 ± 2.43	3.51 ± 1.76
	$n = 19$	$n = 16$	$n = 17$	$n = 20$	$n = 17$	$n = 17$	$n = 18$
		$P = 0.700$	$P = 0.532$	$P = 0.228$	$P = 0.823$	$P = 0.500$	$P = 0.581$
Focus Night and Day DW	3.94 ± 2.22	4.55 ± 2.29	3.90 ± 2.00	3.74 ± 1.64	3.56 ± 2.35	4.07 ± 2.03	4.00 ± 2.01
	$n = 31$	$n = 26$	$n = 30$	$n = 28$	$n = 28$	$n = 27$	$n = 29$
		$P = 0.233$	$P = 0.899$	$P = 0.816$	$P = 0.408$	$P = 0.594$	$P = 0.705$
Focus Night and Day EW	3.80 ± 1.64	4.57 ± 2.59	4.34 ± 2.22	4.15 ± 2.20	4.64 ± 2.06	3.43 ± 1.48	3.84 ± 2.05
	$n = 27$	$n = 21$	$n = 26$	$n = 24$	$n = 26$	$n = 26$	$n = 27$
		$P = 0.542$	$P = 0.416$	$P = 0.774$	$P = 0.157$	$P = 0.396$	$P = 0.687$
O_2 Optix DW	3.78 ± 2.22	3.55 ± 1.96	3.99 ± 2.35	3.66 ± 2.04	3.62 ± 2.28	3.54 ± 1.77	2.73 ± 1.74
	$n = 18$	$n = 17$	$n = 19$	$n = 21$	$n = 19$	$n = 18$	$n = 18$
		$P = 0.565$	$P = 0.739$	$P = 0.828$	$P = 0.692$	$P = 0.894$	$P = 0.107$
O_2 Optix EW	3.56 ± 2.57	3.54 ± 1.74	4.27 ± 2.17	3.05 ± 1.55	3.75 ± 2.41	3.08 ± 2.22	3.24 ± 1.41
	$n = 18$	$n = 17$	$n = 18$	$n = 19$	$n = 18$	$n = 19$	$n = 17$
		$P = 0.562$	$P = 0.191$	$P = 0.751$	$P = 0.646$	$P = 0.590$	$P = 0.904$

Data are the number of bacteria/cell. The probabilities represents a one-way ANOVA and multiple comparison Student-Newman-Keuls test comparing each time point to the baseline value. Data are expressed as the mean ± SD. (Acuvue Advance, Johnson & Johnson Vision Care, Jacksonville, FL; Focus Night and Day and O_2 Optix, CIBA Vision, Duluth, GA.)

* Two-way repeated measures analysis of variance:
Focus Night & Day DW vs. EW: $P = 0.498$ (lens); $P = 0.567$ (visit).
O_2 Optix DW vs. EW: $P = 0.836$ (lens); $P = 0.364$ (visit).
Among three hyper-Dk lens groups, daily wear: $P = 0.216$ (lens); $P = 0.531$ (visit).
Among two hyper-Dk lens groups, extended wear: *$P = 0.011$ (lens); $P = 0.173$ (visit).
Among five hyper-Dk lens groups at 1 month: $P = 0.964$ (lens); $P = 0.282$ (visit).
Among five hyper-Dk lens groups at 12 months: $P = 0.051$ (lens); $P = 0.131$ (visit).

with silicone hydrogel lens wear, specific MPSs have been shown to induce different levels of corneal staining, suggesting a nonoptimal contact lens–solution relationship.[33,34] Further, the effects of the corneal toxicity of MPSs have been suggested as an increased risk factor for corneal infiltrative events (CIEs),[21] and corneal staining has been shown to increase the risk of a CIE sevenfold, compared with the use of nonpreserved care solutions.[35]

The results from this present study represent the first prospective clinical data correlating the use of nonpreserved care solutions with bacterial binding to desquamated corneal epithelial cells after daily and continuous wear of silicone hydrogel lenses. Significantly, the bacterial binding data for all silicone hydrogel lenses worn in DW mode using nonpreserved care solutions failed to show an increase in PA binding at any time point. This finding markedly differs from our previous PA binding data generated after DW of the same and similar high- and hyper– oxygen-transmissible silicone hydrogel lenses in prior studies of preserved solutions, in which a significant initial increase in PA binding was seen as early as 2 weeks after the initiation of daily lens wear with both high– and hyper–oxygen-transmissible lenses and 24 hours after the initiation of

TABLE 7. CET for Each Lens Group and Mode of Wear

	Baseline	1 Week	1 Month	3 Months	6 Months	9 Months	12 Months
Acuvue Advance DW	49.15 ± 5.52	49.61 ± 3.23	49.34 ± 5.58	49.97 ± 2.99	49.81 ± 5.29	49.82 ± 5.19	48.65 ± 4.91
	$n = 19$	$n = 20$	$n = 20$	$n = 20$	$n = 20$	$n = 20$	$n = 20$
		$P = 0.704$	$P = 0.928$	$P = 0.725$	$P = 0.574$	$P = 0.701$	$P = 0.791$
Focus Night and Day DW	49.40 ± 2.73	49.32 ± 3.08	48.03 ± 3.96	48.73 ± 3.44	48.15 ± 3.63	47.73 ± 4.71	49.40 ± 3.83
	$n = 31$	$n = 29$	$n = 30$	$n = 29$	$n = 30$	$n = 29$	$n = 29$
		$P = 0.890$	$P = 0.106$	$P = 0.382$	$P = 0.131$	$P = 0.333$	$P = 0.932$
Focus Night and Day EW	49.34 ± 3.03	48.92 ± 3.27	48.51 ± 3.98	48.35 ± 3.53	48.92 ± 2.84	47.81 ± 5.12	46.95 ± 6.70
	$n = 26$	$n = 25$	$n = 26$	$n = 27$	$n = 24$	$n = 28$	$n = 25$
		$P = 0.955$	$P = 0.368$	$P = 0.270$	$P = 0.628$	$P = 0.170$	$P = 0.313$
O2 Optix DW	48.82 ± 2.54	48.15 ± 2.67	48.60 ± 3.40	48.46 ± 2.51	48.26 ± 4.88	48.73 ± 4.39	48.48 ± 4.90
	$n = 21$	$n = 20$	$n = 21$	$n = 21$	$n = 20$	$n = 18$	$n = 20$
		$P = 0.256$	$P = 0.840$	$P = 0.657$	$P = 1.000$	$P = 0.602$	$P = 0.382$
O2 Optix EW	50.36 ± 3.17	50.00 ± 3.11	47.49 ± 4.38	47.03 ± 5.86	47.75 ± 4.16	48.16 ± 4.50	48.32 ± 3.37
	$n = 19$	$n = 19$	$n = 20$	$n = 20$	$n = 17$	$n = 20$	$n = 17$
		$P = 0.728$	$P = 0.026$	$P = 0.036$	$P = 0.039$	$P = 0.085$	$P = 0.071$

The probabilities represents a one-way analysis of variance and multiple comparison Student-Newman-Keuls test comparing each time point to the baseline value. Data are in mean micrometers ± SD.

* Two-way repeated measures analysis of variance. Lens manufacturers are as shown in Table 6
Focus Night & Day DW vs. EW: $P = 0.634$ (lens); $P = 0.354$ (visit).
O_2 Optix DW vs. EW: $P = 0.941$ (lens); $P = 0.376$ (visit).
Among three hyper-Dk lens groups, daily wear: $P = 0.076$ (lens); $P = 0.995$ (visit).
Between two hyper-Dk lens groups, extended wear: $P = 0.924$ (lens); $P = 0.084$ (visit).
Among five hyper-Dk lens groups at 1 month: $P = 0.963$ (lens); $P = 0.052$ (visit).
Among five hyper-Dk lens groups at 12 months: $P = 0.147$ (lens); $P = 0.283$ (visit).

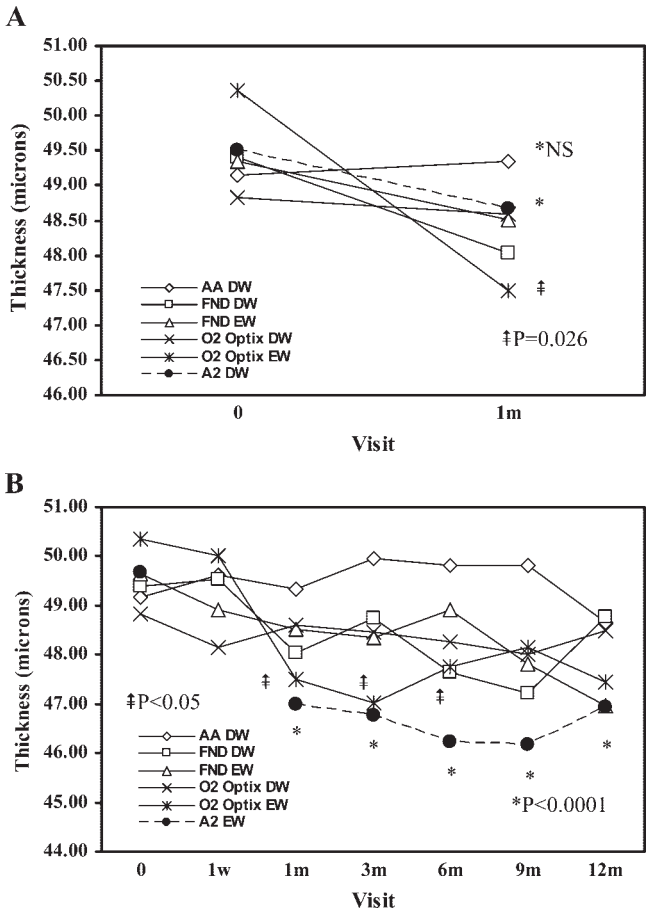

FIGURE 2. CET for all test lenses and mode of wear. The Lotrafilcon B lens showed significant thinning after 1 month (**A**; $P = 0.026$, one-way ANOVA) with partial recovery at 12 months (**B**; $P < 0.05$) of lens wear. Previous data for the Acuvue 2 lens in DW (**A**, *NS) and EW (**B**; $^*P < 0.0001$, one-way ANOVA) modes were included as a historical control.[24,25] For clarity, only the means of the data are represented. Results ± SD are shown in Table 7.

extended wear of the hyper–oxygen-transmissible lens, followed by adaptation and decreased binding over the next 6 months. Moreover, using the data from our previous studies,[23–25] we were able to correlate PA-binding results for each lens type with estimates of relative risk of MK derived from epidemiologic studies within the lens-wearing population (rigid gas-permeable [RGP] DW < soft DW < soft EW lenses).[4,5,9] In addition, the validity of using lens-related increases in PA binding to corneal cells to estimate risk for MK is further supported by an unexpected adaptive decrease in PA binding seen over the initial 6 months of extended wear. These data prospectively predicted for the first time that MK risk could be correlated to duration of lens wear and was subsequently validated in more recent epidemiologic studies (Stapleton F, personal communication, 2007). Since identical study protocols were used in the current and past studies, the disparity in PA binding in silicone hydrogel lens wear arises from the use of nonpreserved and chemically preserved contact lens care solutions, respectively, suggesting that chemically preserved solutions induce epithelial surface damage that leads to an upregulation of PA-specific binding receptors. This finding has also been demonstrated in a prospective, double-masked, randomized, crossover clinical trial evaluating the effects of preserved contact lens care solutions and PA binding in non–contact-lens wearers.[22] The findings in the latter study demonstrated that the use of chemically preserved solutions on the corneal surface result in a significant decrease in central epithelial cell desquamation, with a corresponding increase in PA binding to exfoliated corneal epithelial cells. By contrast, comparison of extended wear of hyper–oxygen-transmissible silicone hydrogel lenses compared with DW in the present study of nonpreserved lens care solutions did not significantly increase PA binding. As PA binding is the initial inciting event in infection, absence of an increase in PA binding suggests that there is no lens-enhanced risk of infection. This paradigm has already explained the relative risk rate of MK and predicted before the epidemiologic data were available that less than 6 months of contact lens wear would have a higher MK rate. Thus, the results of the present study predict no difference in MK rates between daily and overnight wear in the absence of concomitant solution-induced corneal surface damage.

MK is the result of a multifactorial process, and to date, the absence of a standard animal model in contact lens wear has limited our understanding of the pathogenesis of this condition. It has been established that initial PA binding to corneal epithelial cells is a prerequisite for infection to occur and extended wear of low oxygen-transmissible soft contact lenses increases PA binding to the corneal surface.[36,37] Further, once bound to the plasma membrane, PA has demonstrated the

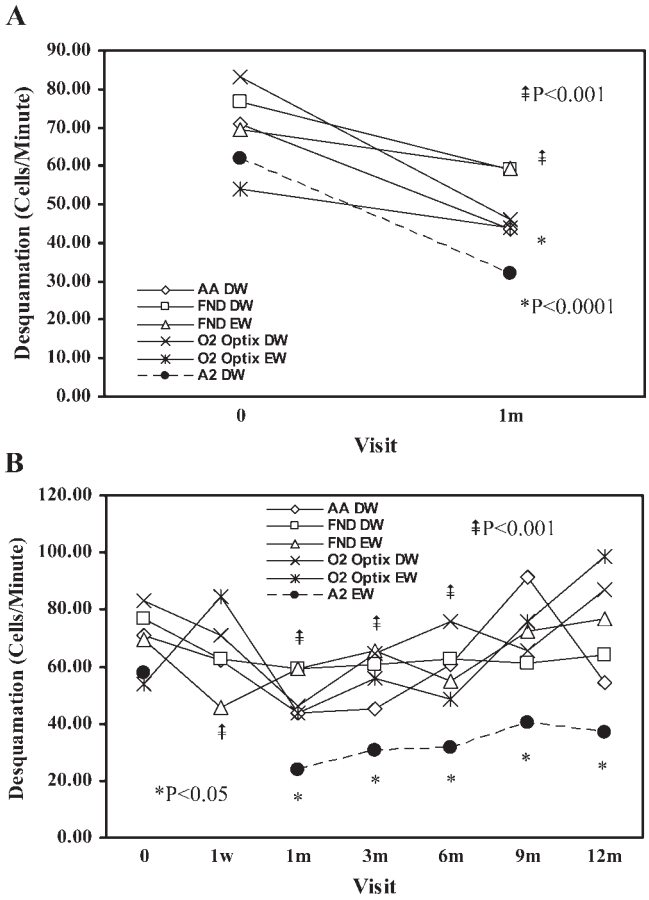

FIGURE 3. Corneal epithelial cell desquamation for all test lenses and mode of wear was reduced at 1 month (**A**; $P < 0.001$, two-way ANOVA) with partial recovery at 12 months (**B**; $P < 0.001$, two-way ANOVA) of lens wear. Previous data for the Acuvue 2 lens in DW (**A**, $^*P < 0.0001$, one-way ANOVA) and EW (**B**, $^*P < 0.05$) modes were included as a historical control.[24,25] For clarity, only the means of the data are represented. Results ± SD are shown in Table 8.

TABLE 8. Corneal Epithelial Desquamation as a Function of Time for Each Lens Group and Mode of Wear

	Baseline	1 Week	1 Month	3 Months	6 Months	9 Months	12 Months
Acuvue Advance DW	70.94 ± 47.93	62.20 ± 65.63	43.58 ± 33.97	45.00 ± 40.54	49.82 ± 48.77	68.38 ± 48.52	54.50 ± 43.11
	$n = 18$	$n = 20$	$n = 19$	$n = 20$	$n = 17$	$n = 13$	$n = 16$
		$P = 0.139$	$P = 0.043$	$P = 0.043$	$P = 0.077$	$P = 0.561$	$P = 0.222$
Focus Night and Day DW	76.56 ± 56.69	57.24 ± 50.67	54.62 ± 43.70	50.41 ± 40.92	55.96 ± 36.38	61.29 ± 38.08	57.35 ± 36.86
	$n = 27$	$n = 29$	$n = 29$	$n = 27$	$n = 28$	$n = 24$	$n = 26$
		$P = 0.131$	$P = 0.090$	$P = 0.075$	$P = 0.325$	$P = 0.521$	$P = 0.364$
Focus Night and Day EW	68.54 ± 51.54	47.25 ± 41.19	51.52 ± 45.06	56.73 ± 56.35	54.19 ± 47.71	59.96 ± 43.25	77.20 ± 57.38
	$n = 24$	$n = 24$	$n = 25$	$n = 26$	$n = 26$	$n = 24$	$n = 25$
		$P = 0.044$	$P = 0.087$	$P = 0.093$	$P = 0.188$	$P = 0.485$	$P = 0.582$
O2 Optix DW	83.94 ± 57.94	49.88 ± 49.59	44.63 ± 29.04	47.67 ± 51.47	70.07 ± 52.59	65.78 ± 50.02	66.85 ± 50.70
	$n = 17$	$n = 17$	$n = 19$	$n = 15$	$n = 15$	$n = 18$	$n = 13$
		$P = 0.146$	$P = 0.212$	$P = 0.142$	$P = 0.685$	$P = 0.639$	$P = 0.391$
O2 Optix EW	55.18 ± 44.26	63.38 ± 52.23	44.68 ± 54.34	45.83 ± 31.88	49.87 ± 24.52	77.11 ± 64.87	90.44 ± 55.02
	$n = 17$	$n = 16$	$n = 19$	$n = 18$	$n = 15$	$n = 18$	$n = 16$
		$P = 0.871$	$P = 0.093$	$P = 0.874$	$P = 0.806$	$P = 0.597$	$P = 0.027$

The probability represents a one-way analysis of variance and multiple comparison Student-Newman-Keuls test comparing each time point to the baseline value. Data are in mean cells/minute ± SD. Lens manufacturers are as shown in Table 6

* Two-way repeated measures analysis of variance (*denotes significance):

Focus Night & Day DW vs. EW: $P = 0.955$ (lens); $P = 0.183$ (visit).

O_2 Optix DW vs. EW: $P = 0.959$ (lens); $^*P = 0.038$ (visit).

Among three hyper-Dk lens groups, daily wear: $P = 0.724$ (lens); $^*P = 0.009$ (visit).

Between two hyper-Dk lens groups, extended wear: $P = 0.536$ (lens); $^*P = 0.035$ (visit).

Among five hyper-Dk lens groups at 1 month: $P = 0.602$ (lens); $^*P < 0.001$ (visit).

Among five hyper-Dk lens groups at 12 months: $P = 0.940$ (lens); $^*P < 0.001$ (visit).

ability to internalize into corneal epithelial cells through the formation of cholesterol-enriched microdomains known as lipid rafts.[38–40] After internalization, the normal host response to limit the spread of infection is for the cell to undergo apoptosis, which in the corneal epithelium would result in desquamation or sloughing of the infected epithelial cell from the surface of the cornea; however, studies have shown that all contact lens wear inhibits the desquamation process.[11,23,25] The reduction in desquamation has been attributed to oxygen availability, the use of chemically preserved solutions, and the mechanical presence of the lens itself.[22,41,42] Consistent with these findings, the 1-month lens wear of test silicone hydrogel lenses showed a reduction in desquamation across all lens types and mode of wear, which recovered to baseline values after 12 months of lens wear; however, these findings were not significant. Of note, the Galyfilcon A lens worn in DW mode, which has the lowest oxygen transmissibility of all the test lenses, had the greatest effect on desquamation in any mode of wear, which was significant. Thus, with high levels of oxygen and the use of nonpreserved solutions, the residual decrease in central epithelial cell desquamation is probably due to the mechanical pressure of the lens on the cornea or the elimination of shear forces from the eyelids, which are shielded by the presence of the lens during blinking.[43]

In addition to desquamation, homeostasis of the epithelium is further characterized during contact lens wear by the inhibition of basal cell proliferation measured by BrdU-labeling and a delay in the vertical migration of epithelial cells toward the corneal surface, producing a thinned, stagnant epithelium.[44,45] In the present study, the greatest degree of central epithelial thinning was seen in contact lenses worn on an EW basis, with significant thinning after 6N EW of the Lotrafilcon B lens. These results are in agreement with our previously published clinical studies demonstrating a decrease in CET after extended contact lens wear and suggest that the effects on epithelial thickness are independent of lens oxygen and lens care solutions.[24,25] The actual significance of thinning on epithelial barrier function is unknown; however, EW of soft contact lenses has also been attributed to increased epithelial permeability.[46]

Significantly, the results of this study also suggest that independent of lens oxygen transmissibility, continuous wear of silicone hydrogel contact lenses disrupts the normal homeostatic renewal mechanisms of the corneal epithelium; however, wear of these lenses does not effect PA adherence when used with nonpreserved care solutions and/or when worn on an extended-wear basis as summarized in Figure 4. One final consideration regarding the ongoing risk of MK in daily and continuous wear of silicone hydrogel lenses, seen in current epidemiologic studies, may also be inadequate patient selection and poor lens hygiene compliance. Although the combination of hyper– oxygen-transmissible lens materials and hydrogen-peroxide–based solutions may offer the best lens solution combination to reduce corneal surface damage and optimize ocular health, the length of time the lens is worn, exposure to waterborne pathogens while swimming, inadequate tear flushing behind the lens result-

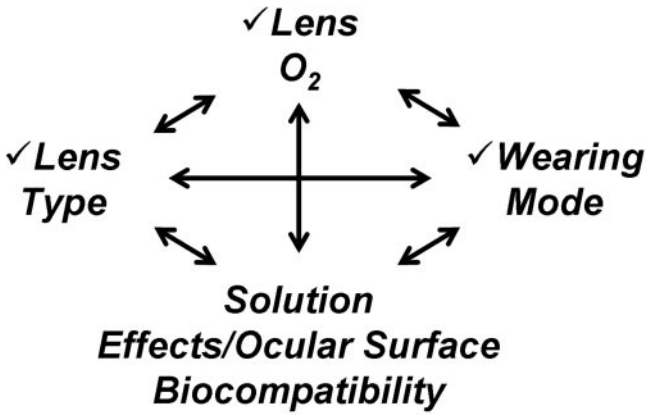

FIGURE 4. Four risk factors for MK associated with contact lens wear. Having reached the maximum oxygen needed for safe contact lens wear, PA binding data predict the same risk independent of wearing mode and lens type. The confounding variables remaining are solution-induced corneal damage and ocular surface biocompatibility, which may play critical roles in mediating contact-lens–related MK.

ing in debris and potential microbial pathogens trapped in the postlens tear film, lens spoilage, deposits, and poor patient hygiene remain predisposing factors for MK and may further account for the persistent MK rates noted over two decades. In comparison, patients enrolled in a contact lens clinical trial are carefully screened for preexisting ocular disease; seen in a compulsory manner for contact lens follow-up visits; and continuously dispensed new contact lenses and care solutions, including nonpreserved saline to rinse lenses as needed. They also undergo extensive patient education and training. These realizations emphasize the critical importance of careful patient selection and education for initiation of all lens wear, especially those individuals electing to pursue overnight or continuous lens wear.

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