Assessment of Potential Toxicity of Rigid Gas Permeable (RGP) Contact Lens Care Solutions on Cultured Epithelial Cells
May Bakkar, Alshefaa Almasri

**Purpose:** The cytotoxicity of multipurpose contact lens solutions (MPSs) used with rigid gas permeable (RGP) lenses is poorly studied. The aim of this work is to study possible effect of RGP MPSs on cultured epithelial cells and to compare levels of possible cytotoxicity between studied solutions.

**Method:** Cultured Vero epithelial cells were treated with different types of RGP MPSs; polyhexanide 0.0005% and chlorhexidine 0.003% -based MPS (solution A), polyhexanide 0.0002% -based MPS (solution B), polyhexanide 0.0001%-based MPS (solution C), and an appropriate control was used. 

- 3-(4,5-Dimethylthiazol-2-yl)-2,5-Diphenyltetrazolium Bromide (MTT) assay was used to study possible toxic effect caused by MPSs against the cells. Additionally, cell cycle analysis using flowcytometry was used to assess the impact of MPSs on cell cycle profile. Wound healing model was also used to study the effect of different MPSs on cell migration during healing process.

**Results:** The toxic effect on epithelial cultured cells differs between the three tested RGP MPSs after 4 hours and 16 hours of exposure (p<0.05). Solution A showed the most toxic effect on cultured epithelial cells. Solution B showed less toxic effect than solution A. While, solution C showed the least toxic effect against cultured cells. Cell activity was significantly reduced in a dose-response and in a time-response manner for all tested solutions manner. However, some toxic effects seem to be reversible as it is observed after 12 hours of re-growing the cells that exposed to MPS in fresh cell culture media, cells showed higher regeneration rate in Solution C > Solution B > Solution A, (p<0.05).

**Conclusions:** Different toxic effect of RGP MPSs on cultured epithelial cells may be related to different preservatives formula among the studied MPSs. Solution A showed the most toxic effect which may attribute to higher toxicity caused by co-presence of polyhexanide and chlorhexidine in the solution formula.

**Research funding received:** This project was supported by the deanship of research at Jordan University of Science and Technology

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Bacterial growth in soft contact lens containers
Lois van Dinther, Imke Jacobs, Mirjam van Tilborg

**Purpose:** To investigate the bacterial growth in day by day used soft contact lens containers with different hygiene regime of soft contact lens wearers, using Multi-Purpose Solution (MPS).

**Method:** In this explorative research, day by day used soft contact lens containers were collected to be microbiological investigated for the appearance of Staphylococcus Aureus, Staphylococcus Epidermis, Pseudomonas Aeruginosa and Streptococcus Pneumoniae. Only flat soft contact lens containers, used at least one week in combination with a MPS, from contact lens users of 16 years and older were used. The investigation letter was given during a contact lens examination and sent by direct mail, asking the soft contact lens wearers to participate anomalous in this investigation, by donating their contact lens container and fill in a questionnaire of 8 questions. The collection of the containers was restricted to the month September 2017 at two different locations in the south of the
Netherlands. Two new fresh containers were investigated too.

**Results:** In total, there were 68 donated contact lens containers from 24 males (mean age 38.83 sd 15.77) and 44 females (mean age 40.36 sd 13.88). Cultivated growth of bacteria was found in 91.2% of the contact lens containers. In 50% of the containers Streptococcus Pneumoniae was found, followed by Staphylococcus Aureus (39.7%), Staphylococcus Epidermis (32.4%) and Pseudomonas Aeruginosa (2.9%). The unused containers were found positive for all the bacteria investigated except for the Pseudomonas Aeruginosa. Proportional less bacterial growth was seen when the MPS was refreshed more than once a week, the hands were dried after washing hands and the container was not rinsed with water.

**Conclusions:** Better attention for contact lens care and hygiene could have a positive effect to minimize bacterial culture in this specific contact lens container, by using a MPS.

**Research funding received:** Alcon Pharmaceuticals Ltd.

**Paper Number:** 30
**Presentation time:** 13:46-13:54

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**Comparative Antimicrobial Efficacy of a Novel Povidone Iodine Rigid Gas Permeable (RGP) Contact Lens Disinfection System**

*Simon Kilvington, Ritsue Ota, Yohei Mizuno, Yoshiyuki Kitamura, Katsuhide Yamasaki*

**Purpose:** RGP contact lenses (CL) account for approximately 16% of the market in Japan, 13% in the UK and 6% in the USA. As with all CL wear, effective disinfection is essential to safe use in the prevention of microbial keratitis (MK). Here we evaluated the antimicrobial efficacy of a novel povidone iodine (PI) disinfection system for RGP lenses.

**Method:** The PI system was evaluated according to the microbiological requirements of ISO:14729 stand-alone procedure, clinical strains of Pseudomonas aeruginosa, Serratia marcescens and Achromobacter xylosoxidans under planktonic and biofilm growth conditions, herpes simplex virus (HSV-1) and Acanthamoeba. Biofilm disinfection studies were also conducted with commercial RGP care solutions based on 5 ppm PHMB-30 ppm chlorhexidine (MPS-A) and 5 ppm PHMB (MPS-B).

**Results:** The PI system gave a log10 kill of >4 for bacteria and fungi according to ISO:14729 methodology, >4 for clinical bacteria isolates, 3 for HSV-1, and 4 for Acanthamoeba trophozites and 1 for cysts. In biofilm experiments, the PI system gave a 5-log10 reduction in bacteria compared to 3-log10 with MPS-A for PA and SM and 1.4 for AX and <1 for MPS-B after the recommended disinfection time of 5 minutes.

**Conclusions:** The PI system contains 500 ppm PI, surfactant and a separate neutralising and proteolytic enzyme cleaner tablet. Lens and storage case disinfection and neutralisation completes within 20 minutes (indicated by a PI solution colour change from orange to clear). Four hours is necessary to ensure removal of protein and lipids by the enzyme and surfactant cleaner. As the PI is completely neutralised, no residual antimicrobial remains that may cause complications such as corneal staining and infiltrative events observed with other systems. Good compliance and use of an effective disinfection system will help reduce the incidence of MK and improve comfort in RGP and Orthokeratology CL wearers.

**Research funding received:** None

**Paper Number:** 31
**Presentation time:** 13:54-14:02

**Impact on gas-permeable contact lens parameters after storage in a non-neutralized hydrogen peroxide case during 1 to 30 days.**

*Langis Michaud, Anna Zarouk*

**Purpose:** This study was conducted to evaluate the impact of storing large gas permeable lenses in a regular soft contact lens case, filled with non-neutralized...
hydrogen peroxide, during 1 to 30 days. **Method:** Twenty scleral contact lenses, never worn, were used for this study. All lenses were manufactured by the same laboratory, with the same material (Hexafoncon A), the same central thickness (250 microns) and the same diameter (14.9mm). They were soaked in a regular soft contact lens case, filled with a controlled volume of hydrogen peroxide solution (Alcon, Tx, US) for 1, 3, 7 and 30 days. Another set of 20 cases was also filled with peroxide, for a chemical analysis of its content, by iodometric titration with sodium thiosulfate. Lens parameters (power, diameter, base curve) were checked at every step, and the wetting angle was also evaluated at these moments (captive bubble, Image J software analysis).

**Results:** There was no statistical difference in the chemical composition of the hydrogen peroxide over 30 days. Lens parameters were also not affected, except for the wetting angle, which showed a significant decrease over time. There was a statistical difference (F16.64; p<0.001), specifically between day 1 (16.73 deg) and 7 (13.96 deg), 1 and 30 (13.15 deg), 3 (15.50 deg) and 7 and 3 and 30 days (ANOVA for repeated measurements, with Bonferroni posthoc testing).

**Conclusions:** Storing gas permeable lenses in a non-neutralizing case seems to preserve the lens parameters. By nature, the gas permeable material does not absorb more than 1% of the soaking solution, limiting peroxide release when the lens is worn. Longer storage reduces the wetting angle. Hydrogen peroxide keeps its content intact over 30 days, without neutralization and not exposed to air. Consequently, storing scleral lenses in a regular soft lens case, with hydrogen peroxide, is considered a valid option.

**Research funding received:** Lenses used in the study provided by Laboratoires Blanchard (Sherbrooke, Canada)

**Paper Number:** 32
**Presentation time:** 14:02-14:10

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**Can a novel system to assess contact lens wettability predict tear film stability in a clinical setting?**

**Michael Read, Chantal Coles-Brennan, Carole Maldonado-Codina, Philip Morgan**

**Purpose:** Is a novel in vitro contact lens wettability system able to predict clinical tear film stability?

**Method:** A novel laboratory system has been developed to assess contact lens wettability, where a thin saline film is generated on a contact lens surface by automated withdrawal from a heated saline bath. This saline film is then monitored using live image analysis and on detection of film-break-up a new saline film is automatically generated (akin to the blinking action of the eye). Eight contact lens types underwent testing over a 6-hour cycling period (nine lenses for each lens type), with time to film break-up recorded for each cycle. To allow comparison with clinical data, four of the study lens types (lotrafilcon B, etafilcon A & PVP, senofilcon and filcon iv) were assessed in a clinical study (15 subjects, 4-visit, randomized, double-masked, crossover study) which focused on tear film stability (Keeler Tearscope) over 30-minutes of lens wear.

**Results:** Differences in film stability were observed between lens types in both the laboratory (p<0.0001) and clinical (p=0.0007) studies. Laboratory measures of film stability ranged between 22 seconds (filcon iv) and 188 seconds (etafilcon A & PVP). Clinical tear film break-up time at the 30-minute time point ranged between 7.5 seconds (lotrafilcon B) and 11.3 seconds (filcon iv). In both the laboratory and clinical environments film stability was seen to reduce significantly over time (p=0.001 and p=0.052 respectively).

**Conclusions:** Conclusion: This novel laboratory system is able to characterize saline film stability over a prolonged period of lens cycling. Differences in stability are observed between lens types and over time highlighting the influence lens material has on wettling properties.
This instrument shows promise in predicting clinical performance; however further optimisation is required to better model the biologically complex and highly dynamic characteristics of the ocular surface.

**Research funding received:** This study was funded by Johnson & Johnson Vision Inc.

**Paper Number:** 33  
**Presentation time:** 14:10-14:18

**The effect of fluorescein sodium volume on anterior eye surface measurements using the Medmont corneal topographer**  
**Jeroen A Mulder, Mirjam M van Tilborg, Byki Huntjens**  

**Purpose:** This study investigates the effect of fluorescein sodium (NaFl) volume on the regularity of the anterior eye surface using the Medmont corneal topographer measurements.

**Method:** The simulated keratometry values (flat = Kf; steep = Ks), Inferior Superior Index (ISI), Surface Asymmetry Index (SAI), and Surface Regularity Index (SRI) measurements, using the E300 corneal topographer (Medmont International Pty Ltd., Victoria, Australia), were taken three times under three different conditions: baseline (without NaFl), including a single dose NaFl, and a double dose NaFl. Of the 57 participants (males n=23, females n=34) only the right eye was included. Mean age (± SD) was 35.1±15.2 years (range 19 to 65 years); grouped by age (<40 years [n=34] versus ≥40 years [n=23]). There was no history of ocular diseases, contact lens wear, or previously diagnosed dry eye.

**Results:** At baseline, there were no significant differences between the three consecutive measures of Kf (p=0.30), Ks (p=0.71), ISI (p=0.10), SAI (p=0.53) and SRI (p=0.34). This was comparable to any amount of NaFl (p>0.05). Median ISI values following the addition of 1 or 2 doses NaFl significantly decreased compared to baseline (both z=3.2, effect size 0.43; p=0.001), irrespective of age group (p=0.74). No significant differences between the three conditions for any other parameter (p>0.05) were found. The intraclass correlation coefficient (ICC) improved for Kf (0.99), Ks (0.99) and SRI (0.60) with one dose of NaFl, compared to baseline or 2 doses of NaFl. For ISI and SAI, ICC was most reliable without NaFl (0.85 and 0.77 respectively).

**Conclusions:** One drop NaFl increases reliability in corneal topography and regularity measurements using the E300 topographer. Corneal asymmetry measurements (IS and SAI) are more reliable without any ocular dye. It is possible that the addition of NaFl enhances tear film reflection during topography measurements, but the addition of a liquid solution decreases symmetry.

**Research funding received:** None

End of session