Performance of Cationic Peptide Mel4 as an Antimicrobial Agent and Contact Lens Coating

Debarun Dutta, Ajay K Vijay, Janelle Tong, Wilson Luu, Timothy Zhao, Kai Bing Cheah and Mark DP Willcox

School of Optometry and Vision Science, The University of New South Wales, Sydney, Australia

INTRODUCTION

There remains a need to reduce microbial contamination of contact lenses, and thus the rate of adverse events during wear. We have earlier shown that the antimicrobial peptide melimine retains activity against various ocular pathogens.[1] Although melimine-coated lenses retained antimicrobial activity following human wear, they were associated with occasional corneal staining.[2]

PURPOSE

To determine activity of a melimine-derived antimicrobial peptide Mel4 against drug resistant and clinical isolates of bacteria. In addition, the in vitro activity and clinical performance of Mel4 as an antimicrobial contact coating was evaluated in a human clinical trial.

METHODS

- ✓Antimicrobial activity of Mel4 (K-N-K-R-K-R-R-R-R-R-R-G-G-R-R-R-R; >90% purity) was determined against bacteria (Table 1) by evaluating minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) using a modified broth microdilution assay.
- ✓Etafilcon A lenses were coated by covalently binding the Mel4 peptide to the surface via EDC (1-ethyl-3-[3-dimethylaminopropyl]carbodimide hydrochloride) coupling.[1]
- ✓Antimicrobial activity of Mel4-coated lenses against the *P. aeruginosa* and *S. aureus* strains were evaluated by viable plate count.
- ✓A prospective, randomised, double-masked, clinical trial of one-week daily contralateral contact lens wear with 17 human subjects was conducted to assess the Mel4 coated lens safety and performance.
- ✓For overnight lens storage Biotrue multipurpose solution and lens cases were used. Ethics approval was received from UNSW human research ethic committee.
- ✓Clinical signs were monitored on Days 1, 2 and 7 of lens wear and 1 week and 3 weeks following study lens wear discontinuation.

Table 1: Bacterial strains and resistance profile

Bacterial strain	Isolation site	Resistant to
S. marcescens ATCC 13880	Pond water	Not determined
Drug-resistant organisms		
P. aeruginosa 31	Microbial keratitis	GEN, TOB, PRL, NOR, OFX, MXF, CIP
P. aeruginosa 37	Microbial keratitis	GEN, TOB, PRL, NOR, OFX, MXF, CIP
S. aureus 60	Hospital strain	PCN, MET, TET, GEN, ERY, CIP
S. aureus 110	Microbial keratitis	MET, TOB, ERY ,CIP

GEN: gentamicin, TOB: tobramycin, PRL: piperacillin, NOR: norfloxacin, OFX: ofloxacin, MXF: moxifloxacin, CIP: ciprofloxacin, PCN: penicillin, MET: methicillin, TET: tetracycline, ERY: erythromycin

METHODS (cont..)

- ✓The participants' comfort, dryness and lens awareness with lenses and corneal health were evaluated were recorded by questionnaire on Day 7.

RESULTS

- ✓Table 2 lists the MIC and MBC for all the bacteria tested. Highest MIC and MBC were determined for *S. marcescens* ATCC 13880.
- ✓The Mel4-coated lenses showed more than 1.5 log inhibition of adhesion for *P. aeruginosa* and *S. aureus* (Figure 1).
- ✓All participants successfully completed the trial, 8 male and 9 female, with an average age of 22.5 ± 1.4 years.
- ✓No significant difference in fluorescein staining in any of the five corneal areas were observed between control and Mel4 coated lenses during this study (Figure 2; $p > 0.05$).

Table 2.

Bacterial strains	MIC (nmol ml^{-1})	MBC (nmol ml^{-1})
S. marcescens ATCC 13880	1056	2113
P. aeruginosa 31	66	66
P. aeruginosa 37	132	132
S. aureus 60	4	4
S. aureus 110	8	16

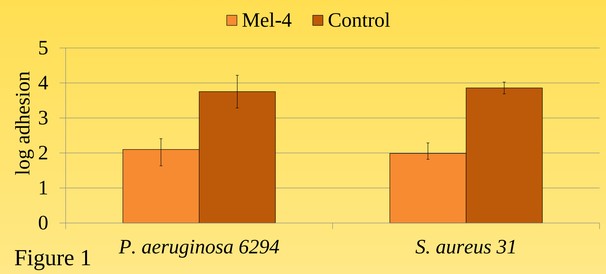

Figure 1

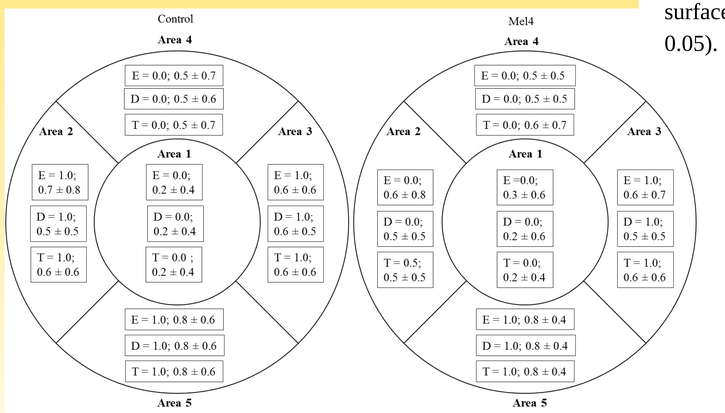

Figure 2. Fluorescein staining scores (median; mean ± SD) in peptide-coated and control contact lens corneal areas.

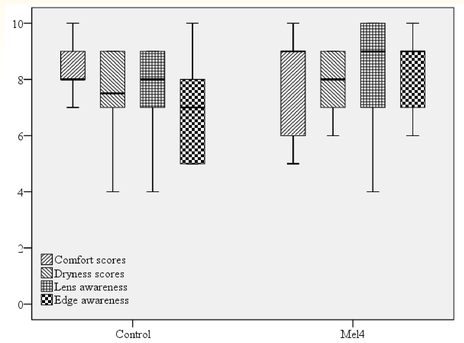

Figure 3. Questionnaire scores for comfort, dryness, lens and edge awareness over one week of wear. Data is represented as a box plot showing the median and 25th and 75th percentile ranges

RESULTS (cont.)

- ✓ No significant difference in subjective responses in comfort, dryness, lens awareness and edge awareness were noted (Figure 3; $p > 0.05$).
- ✓Nearly 60% of study participants preferred the Mel4 coated (Figure 4; $p > 0.05$).
- ✓No significant difference observed in conjunctival or limbal hyperaemia in any quadrant between control and Mel4 coated lenses ($p > 0.05$).
- ✓No differences observed in lens particulates such as front surface wetting, and front and back surface deposits ($p > 0.05$).

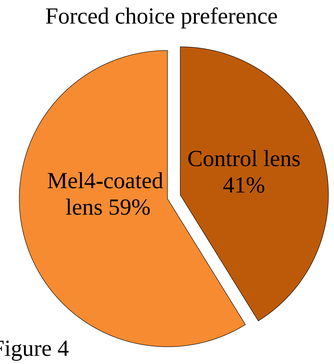

Figure 4

CONCLUSION

Mel4 has high antimicrobial activity against drug resistant bacteria in addition to presenting no adverse effects for human eyes as a contact lens coating, offering excellent potential for development as an antimicrobial agent and contact lens coating.

REFERENCES

1. Dutta et al. Invest Ophthalmol Vis Sci 2013;54:175-82
2. Dutta et al. Optom Vis Sci 2014;91:570-81.
3. Hancock RE. Hancock Laboratory Methods 1999.

Contact: debarun.dutta@unsw.edu.au