INTRODUCTION
Contact lens wear is a risk factor for the development of microbial keratitis (MK). A variety of microorganisms have been implicated in MK (Figure 1), such as Pseudomonas aeruginosa and Staphylococcus aureus. Other bacterially driven adverse events during contact lens wear include contact lens-induced acute red eye, contact lens-induced peripheral ulcer and infiltrative keratitis. Previous studies have confirmed that surface attached antimicrobial peptides (AMP) such as melimine or cathelicidin (LL-37) show high biocidal activity. LL-37 is a naturally occurring AMP that has been found in tears. Melimine is a synthetic peptide and a chimera of the active regions of protamine (from salmon sperm) and melittin (from bee venom).

PURPOSE
Development of antimicrobial contact lenses may reduce the rate of contact lens related adverse events. The purpose of this study was to compare LL-37 and melimine when bound to contact lenses for their activity against P. aeruginosa and S. aureus.

METHODS
• Minimum inhibitory concentration (MIC) of LL-37 and melimine against P. aeruginosa ATCC 19660, P. aeruginosa 6294, S. aureus ATCC 29213 and S. aureus 31 were determined following an established procedure.
• Peptides were attached individually onto the surface of contact lenses (Etafilcon A; ACUVUE® 2®) using a previously described method.
• Varying concentrations of peptides were covalently bound to contact lenses.
• The amount of peptide associated with the lens was quantified using amino acid analysis (AAA).
• Antimicrobial activity of peptide coated contact lenses were evaluated against P. aeruginosa 6294 and S. aureus 31.
• Biocidal activity was evaluated by viable plate count of bacteria after exposure to the coated lenses.
• The adhesion data were log10(x+1) transformed prior to data analysis where x is the number of adherent bacteria in CFU mm⁻². A linear mixed model ANOVA was used for statistical analysis and significance was set at 5% level.

RESULTS
Table 1. represent the MIC of LL-37 and melimine against P. aeruginosa and S. aureus strains.

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>LL-37 (µg ml⁻¹)</th>
<th>Melimine (µg ml⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>P. aeruginosa ATCC 19660</td>
<td>3.9</td>
<td>500</td>
</tr>
<tr>
<td>P. aeruginosa 6294</td>
<td>62</td>
<td>250</td>
</tr>
<tr>
<td>S. aureus ATCC 29213</td>
<td>3.9</td>
<td>250</td>
</tr>
<tr>
<td>S. aureus 31</td>
<td>250</td>
<td>125</td>
</tr>
</tbody>
</table>

• Figures 2 and 3 show quantification of LL-37 and melimine associated with contact lenses. In addition the figures show the antimicrobial activity of the respective peptides attached to the contact lenses.

DISCUSSION
• Both LL-37 and melimine showed high antimicrobial activity in solution against P. aeruginosa and S. aureus, the activity varies with different strains of bacteria.
• However, probably due to the lower level of cationic amine groups on LL-37 (11) compared to melimine (16), much less LL-37 was able to be bound to the contact lenses.
• This lower level of LL-37 resulted in no activity against S. aureus, although there was good activity against P. aeruginosa even with only 18 µg lens.

CONCLUSION
Covalent surface attachment of AMPs offers excellent potential for development as an antimicrobial coating for contact lenses and thus for other biomaterials. Biocidal mechanisms of various AMPs vary with their innate structure and therefore AMP specific surface attachment techniques may be necessary for optimal outcomes.

ACKNOWLEDGEMENTS
• The authors thank Prof. Alison McDermott, University of Houston for helping data collection with valuable suggestions. This presentation was supported by University of New South Wales PRSS travel grant.

REFERENCES

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ANTIMICROBIAL ACTIVITY OF MELIMINE OR CATHELICIDIN BOUND TO CONTACT LENSES

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