

DEVELOPMENT OF A NOVEL HUMAN ORAL TISSUE MODEL OF GONORRHOEA

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Background: Oropharyngeal *Neisseria gonorrhoeae* (NG) infections are common, increasing and have higher treatment failure compared with other infection sites. Antimicrobial resistant (AMR) NG is a global public health threat as available treatments remain scarce due to AMR. Little is known about where NG colonizes in the oral mucosa and therefore, where antibiotics need to be distributed to cure infection. In April 2022 we started to create an in-vitro co-culture model for NG strains with human oral epithelial cells to understand NG growth in the mouth and later examine antibiotic uptake by oral cell types supporting NG growth.

Methods: FA1090 and WHO-X NG strains were grown on Chocolate agar with IsovitaleXTM and in Fastidious broth media in optimised conditions. NG cell numbers were determined using a colony counter (Scan[®] 1200, Interscience technology). Viability of NG cultured cells was assessed at 7 early timepoints (0/1/3/6/9/12/24 hours) in 3 different oral keratinocytes culture medias co-culture with 3 human oral keratinocytes cell lines - hTERT TIGKs CRL-3397TM, #2610, #2560, and OKF6, isolated from the alveolar process, buccal mucosa, tonsil and floor of the mouth, respectively. Keratinocytes viability was measured using MTS cell proliferation assay, and absorbance measured at 490-500nm at timepoints 0/24/48/72 hours. Intra- and extra-cellular bacteria were quantified and their spatial distribution was assessed with confocal microscopy and immunofluorescence. Host cell viability in response to gonococcal infection was measured with LIVE/DEAD Assays, and Annexin V Assay.

Results: We created the first-of-its-kind in-vitro model for NG oral infection demonstrating that is possible to co-culture NG with oral derived cells. We uncovered the dynamics of NG colonization of the oral cavity and present an alternative model to investigate novel therapeutics against infection.

Conclusion: Our presented model can explore the interactions of NG with oral tissues and investigate current and new therapeutics against oropharyngeal gonorrhoea.

Disclosure of Interest Statement: This project is funded by an ARC Industry Transformation Research Hub to Combat Antimicrobial Resistance (Project ID IH190100021; Kirby Institute, UNSW). All authors declare no competing interests.