Urobiome screening for antimicrobial peptides against Uropathogenic *Escherichia coli*

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BACKGROUND: Urinary tract infections (UTIs) are one of the most common bacterial infections worldwide. As global incidences of UTIs caused by multidrug resistant bacteria continue to increase, so too does the demand for novel antimicrobial therapies.

Due to its relatively understudied nature, the urinary microbiome represents a niche with an untapped source of potentially novel antimicrobials (e.g., bacteriocins). Improvements to bacterial culturing and sequencing techniques have highlighted these potential alternative treatments and control strategies to target antibiotic resistant uropathogenic *E. coli* (UPEC).

METHODS: Expanded quantitative urine culture (EQUC) was used to culture bacterial isolates from mid-stream urine samples. Urinary isolates were then tested for their ability to inhibit a bank of five clinically relevant UPEC strains using deferred antagonism assays. Biochemical characterisation, together with genomic analysis, was used to identify and characterise the isolates to species level, and to identify the putative antimicrobial agents.

RESULTS:

A large bank of 260 bacterial isolates from mid-stream urine samples were screened against five clinically relevant UPEC strains resulting in 24 shortlisted isolates displaying antimicrobial activity. Further analysis and characterisation for bacteriocin production resulted in 4 shortlisted isolates.

Preliminary bioinformatic screening in BAGEL4 for bacteriocin gene clusters (BGCs) resulted in the discovery of 15 putative bacteriocin operons. After manual annotation five were considered for further investigation. These included putative genes for microcin, colicin and colicin E1bacteriocin production. Further investigation indicated that these putative bacteriocin associated areas of interest (AOIs) revealed two microcin precursor peptides (McmA *and* MchB*),* three S-type pyocin domain-containing proteins and two colicin E1 proteins with the potential to inhibit UPEC growth.

CONCLUSION: Given that UTIs caused by UPEC are becoming more difficult to treat due to increasing antibiotic resistance, the putative antimicrobials described herein represent a viable potential alternative to antibiotics for the control and prevention of UTIs.