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Using a conjugative CRISPR-Cas9 tool to mitigate antimicrobial resistance in Neisseria gonorrhoeae

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Background

Neisseria gonorrhoeae (Ng) is the causative agent of the sexually transmitted infection ‘gonorrhoea’ and generates a significant global disease burden. Having developed or acquired antimicrobial resistance (AMR) mechanisms to all classes of known antimicrobials, Ng poses a significant threat to human health. With both multidrug-resistant and extensively drug-resistant Ng strains presently in circulation, new therapeutic approaches are required immediately. Horizontal gene transfer of AMR genes commonly occurs via the exchange of AMR plasmids between bacteria, and prior studies have identified that plasmid mediated resistance in Ng forms a key component of the pathogen’s overall resistance. Therefore, either removing AMR plasmids or blocking their uptake may prove an effective strategy to mitigate AMR gene prevalence or resensitise AMR bacteria to presently ineffective therapeutics. Previous work has demonstrated that recombinant conjugative plasmid pKJK5::csg, which expresses CRISPR-Cas9, may be used as a barrier to AMR plasmid uptake in a variety of different bacterial species, as well as remove AMR plasmids from a target strain.

Aim/Methods

In this study, we engineer pKJK5::csg to target Escherichia-Neisseria shuttle vector pLES2.

Results

We show that this vector can readily be removed from E. coli by applying an unrelated donor strain carrying pKJK5::csg. We aim to assess how this system can be similarly used to remove or block the uptake of pLES2 in Ng.

Conclusions

This work will effectively identify whether resensitisation is an appropriate pathway for innovative therapeutic approaches targeting AMR in Ng and highlight any associated bottlenecks for this treatment style. By evaluating the use of a plasmid-delivered resensitisation strategy, this work will assist future studies in the development of innovative therapeutics.