

# The Conjugative Plasmid RK2 as a Delivery System for Artificial AnatheriaH Genes: A Novel Synthetic Biology Alternative to Traditional Antibiotics

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Antibiotic resistance is an ever growing threat. Bacterial infections resistant to carbapenems, a “last resort” antibiotic, have been sharply rising and can cause “potentially untreatable infections” according to the CDC. This investigation is the second phase in the development of a synthetic biology treatment that approaches bacterial infection in a completely original manner. The purpose of this research was to use bacterial conjugation as a novel delivery system for an antimicrobial coding gene designed in phase one. The antimicrobial gene was cloned into the broad host range conjugative plasmid RK2 (using PCR cloning) and the plasmid was inserted into donor bacteria. The recipients were transformed with UV8.5, a plasmid coding for a pink protein known as mCherry, so that their growth would be visible. A mating experiment was performed; donor bacteria containing the inserted antimicrobial gene were mixed with recipient bacteria capable of activating transcription of the gene. The recipients’ growth was observable by the presence or absence of pink pigment. The control group showed growth of the recipients while the experimental group did not. This demonstrated that the antimicrobial gene was transferred during conjugation and that it effectively inhibited the growth of the recipient bacteria. This system is an entirely new approach to the antibiotic resistance problem. Within the human body, this technique could be used to transfer the antimicrobial gene into pathogens and eliminate infection. This method could also be used as a technology to deliver other beneficial genes into the human microbiome.

## Awards Won:

Intel ISEF Best of Category Award of \$5,000

First Award of \$5,000