Development of Halogenated Phenazine Prodrugs and Antibiotic Conjugates as Antibacterial Therapeutics

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Introduction

Bacterial biofilms are surface-attached communities of antibiotic-tolerant bacteria that pose a significant threat to human health as they are innately tolerant to every class of antibiotic currently used in the clinic.[1] Each there are 17 million new biofilm-associated bacterial infections and 550,000 deaths resulting from biofilms. Biofilms house metabolically dormant, non-dividing persister cells encased within a protective extracellular polymeric matrix of biomolecules and display tolerance to antibiotics as a result of their non-replicative phenotype. Despite the urgent need for clinical agents to effectively kill persistent biofilms, no biofilm-eradicating therapeutic exists. *Advancing the first biofilm-eradicating therapeutic agent to the clinic would be one of the most critical biomedical breakthroughs of the twenty-first century.*

Experimental

Solid stock of chemically synthesized halogenated phenazine (HP) prodrug/antibiotic conjugates are dissolved in ~500 µL of deuterated chloroform or DMSO containing 0.03% trimethylsilane. NMR experiments will be conducted on 600 MHz NMRs at AMRIS providing critical information of multiple stereochemically complex compounds (i.e., HP-erythromycin conjugates). During this award period, these experiments have been carried out at the AMRIS facility at the University of Florida.

Results and Discussion

Our lab has discovered that select halogenated phenazine analogues are capable of eradicating the bacterial biofilms.[2,3] Halogenated phenazines are synthetically tunable and we have turned to the development of a diversity of prodrug compounds for translational purposes. Prodrugs are inactive versions of a drug that requires specific biochemical reactions to liberate the active, free drug molecule.

We have initiated work to develop new prodrug versions of our halogenated phenazine compounds that have included multiple antibiotic conjugates linked through various prodrug moieties (i.e., quinones, ureas) that undergo various bacteria-specific reactions to release free drug, killing biofilms. The antibiotics we are using as conjugates include erythromycin- and tetracycline-based (Fig. 1), both of which are complex natural products and require high end NMR for full characterization. During this award period, we have synthesized several new halogenated phenazine scaffolds which have been fully characterized in the AMRIS facility. We are finalizing one manuscript detailing initial prodrug advances and aim to submit within the next few months. We plan on submitting another manuscript for publication detailing the synthesis of a HP-erythromycin conjugate within the next 6 months.

Conclusions

In conclusion, we have developed a new series of HP-prodrugs, including a potent HP-erythromycin conjugate, which we aim to develop for therapeutic purposes. Continuing synthesis efforts are underway in our labs and will require the use of high-end NMR facilities, such as the AMRIS.

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References

[1] Garrison, A. T., *et al., Curr. Top. Med. Chem.*, *17*, 1954-1964 (2017).
[2] Garrison, A. T. et al., *Angew. Chem. Int. Ed., 54*, 14819-14823 (2015).
[3] Yang, H., et al. *Sci. Rep. 7*, 2003 (2017).