**Investigating selection for antimicrobial resistance by non-antibiotic drugs in freshwater microbial communities**

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**Lead Supervisor Location/Student Home Institution**

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**Full Project Description**

A growing body of research indicates low concentrations of antibiotics, such as those found in freshwater environments, select for antimicrobial resistance (AMR). A recent Nature study found non-antibiotic drugs (NADs) have antimicrobial effects, and so they may be an emerging risk contributing to the evolution and dissemination of AMR in aquatic systems where they are present as micropollutants. The selective pressure for AMR NADs pose alongside antibiotics in freshwater environments has not been previously studied. The selective NADs identified in the Nature study will be used in an established experimental microcosm system, where freshwater bacterial communities will be exposed to environmentally relevant NAD concentrations.

Objective 1: Identify NAD resistance mechanisms A combination of metagenomics and culture based methods will identify impacts on community diversity and identify genes which confer cross-resistance to NADs and antibiotics. Knock out mutants of resistant colonies and cloning of functional genes will identify novel resistance determinants.

Objective 2: Determine minimal selective concentrations of NADs in isolation and in mixtures Genes identified in Objective 1 will be used as real-time qPCR targets to accurately determine minimal selective concentrations (MSCs) of NADs - the lowest concentration which selects for AMR. Effects of multiple selective pressures will be investigated by testing for additive and other interactive effects of NADs and antibiotics.

Objective 3: Explore selection by NADs in different freshwater contexts To explore potential spatio-temporal factors within freshwater systems, the experimental system will be developed to test different nutrient levels, temperatures and aquatic microbial communities. Chemical quantification of NADs will be performed alongside these experiments with support from Bath collaborators to understand fate and levels of biodegradation by resistant bacterial populations. These results will inform water quality policy on release of NADs into freshwater ecosystems in relation to AMR evolution.

The student will benefit from a multi-disciplinary team of supervisors to gain skills in molecular biology, microbiology, bioinformatics, analytical chemistry, ecotoxicity and environmental risk assessment. The student will be based in state-of-the-art microbiology labs at the Environment & Sustainability Institute, which includes a qPCR machine with 384-well plate capacity. Exeter has an in-house Sequencing Service and the student will have access to a high performance computing cluster to facilitate rapid metagenome analyses. The student will also access mass spectrometry instrumentation for identification and quantification of NADs at Bath.Through collaboration with the AstraZeneca stake holder supervisor and existing contacts within Defra, the waste water industry and the Environment Agency, the student will engage with policy makers and industrial stakeholders.

**Real Life challenges this project will address**

AMR is one of the greatest threats facing society, affecting human and animal health, food sustainability and ecosystem health. Recent research on selection for AMR by antibiotics concludes selection may occur in certain environmental hotspots. This project will determine if NADs contribute to this problem. Findings will inform novel mitigation strategies aimed at reducing the evolution and transmission of AMR.

**What you should know about this project**

This project focuses on the impact of micropollutants on the evolution of antimicrobial resistance (AMR) and the ecology of microbial communities in fresh water ecosystems. Research will inform policy and regulation to mitigate the spread of AMR and to protect ecosystem and human health. The supervisory team comprises of leading experts in environmental AMR, ecotoxicology and environmental risk assessment and microbial evolution with a strong track record of collaboration.

**What expertise you will develop**

The student will master several microbiological techniques, including classical culturing methods and molecular approaches such as PCR, qPCR, knock-out mutant generation and cloning. Computer coding and bioinformatics skills will be developed, enabling interrogation of next-generation sequencing data and design of automated pipelines. Students will gain experience in analytical chemistry and engaging with a range of stakeholders from the pharmaceutical and waste water industries, as well as policy makers and regulators.

**Why this project is novel**

The first paper showing non-antibiotic drugs (NADs) can select for antimicrobial resistance (AMR) in human gut-associated strains has just been published, raising concerns for selection for AMR in vivo. However, NADs are also discharged into freshwater environments from waste water treatment plants, alongside antibiotics and other antimicrobial compounds. How these multiple pressures may interact to select for AMR in freshwater environments and how they impact microbial diversity is a completely novel research area, with the potential to influence policy at an international level and improve fundamental understanding of AMR evolution and aquatic microbial ecology.

**Rest of Supervisory Team:**

**Stakeholder Organisation** AstraZeneca

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