Fluoroquinolone resistance in the environment and the human gut

Analysis of bacterial DNA sequences to explore the underlying genetic mechanisms

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FAKULTETSOPPONENT
Professor Morten O. A. Sommer
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AVHANDLINGEN BASERAS PÅ FÖLJANDE DELARBETEN

I **Acquired genetic mechanisms of a multiresistant bacterium isolated from a treatment plant receiving wastewater from antibiotic production**
Anna Johnning, Edward R. B. Moore, Liselott Svensson-Stadler, Yogesh S. Shouche, D. G. Joakim Larsson & Erik Kristiansson
Applied and Environmental Microbiology 79.23 (2013): 7256-7263.

II **Isolation of novel broad host fluoroquinolone resistance plasmids from an antibiotic-polluted lake**
Carl-Fredrik Flach, Anna Johnning, Ida Nilsson, Kornelia Smalla, Erik Kristiansson & D. G. Joakim Larsson
Manuscript

III **Resistance mutations in *gyrA* and *parC* are common in bacterial communities of both pristine and fluoroquinolone-polluted environments**
Anna Johnning, Erik Kristiansson, Jerker Fick, Birgitta Weijdegård & D.G. Joakim Larsson
Submitted

IV **International travel affects the abundance of chromosomal quinolone resistance mutations in the human gut microbiome**
Anna Johnning, Erik Kristiansson, Martin Angelin, Nachiket Marathe, Yogesh S. Shouche, Anders Johansson & D.G. Joakim Larsson
Submitted

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Abstract

Fluoroquinolones (FQs) are synthetic, broad-spectrum antibiotics that target type II topoisomerases. High-level resistance is often caused by mutations in the target genes of FQs, especially in *gyrA* and *parC*. In contrast, plasmid-mediated resistance genes, such as *qnr*, often confer moderate levels of resistance. Several sites near Patancheru, India, have been previously shown to be severely contaminated with FQs. To study how environmental bacteria adapt to this extreme environment, we first used whole-genome sequencing (454) of a highly multi-drug resistant strain of *Ochrobactrum intermedium*. The strain was isolated from a wastewater treatment plant (WWTP) in Patancheru that treats industrial effluent from pharmaceutical production. The strain was considerably more resistant to tetracyclines, sulphonamides, and FQs than to other *O. intermedium* strains, and it had, accordingly, acquired a tetracycline efflux pump, a sulphonamide resistance gene, and mutations in the target genes for FQs. In the second study, sequencing (Illumina) was used to characterise horizontally transferrable resistance plasmids captured from bacterial communities sampled from a lake with a history of FQ pollution, near Patancheru. All transconjugants had acquired *qnr* genes and this is, to the best of our knowledge, the first time *qnrVC1* has been described on a conjugative plasmid. Furthermore, the bacteria from the lake sediments were significantly more resistant to FQs and sulphonamides compared to bacteria from Indian and Swedish reference lakes.

In the third study, the *Escherichia* communities inhabiting a stream in Patancheru receiving WWTP effluent with high levels of FQs were tested for resistance mutations in *gyrA* and *parC* using amplicon sequencing (454). A stream receiving municipal WWTP effluent in Skövde, Sweden, and a remote highland lake were included as references. To our surprise, all communities showed high abundances of FQ resistance mutations, suggesting that these mutations are not associated with a fitness cost in the studied environments. The same method was utilised in the fourth study, on faecal samples collected from Swedish students before and after travel to India. The abundance of the amino acid substitution S83L in GyrA increased significantly, and the number of observed genotypes decreased after travel. This finding shows that international travel contributes to the spread of bacteria carrying chromosomal resistance mutations. Taken together, the development and spread of antibiotic resistance from antibiotic-polluted environments is a concern for everyone.

Keywords: Antimicrobial resistance, quinolones, pollution, next generation sequencing, bioinformatics
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