Ciprofloxacin resistant bacteria in the waterways surrounding Peradeniya, Sri Lanka

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Introduction and Objective: The presence of antibiotic-resistant bacteria (ARB) and antibiotic resistance genes (ARGs) in waterways has raised considerable concerns. This is an indication of the escalating potential for the development and dissemination of antibiotic-resistant pathogens, which pose significant challenges to both the environment and human health. The aim of the current study was to detect the presence of ciprofloxacin resistant bacteria (CRB) in the waterways in Peradeniya, Sri Lanka, which are contaminated by surrounding dwellings and hospital waste.

Methods: Water samples were randomly collected from 10 waterways located in Peradeniya, Sri Lanka, which are contaminated by waste from surrounding dwellings and hospitals. Samples were cultured in 0.25 μ g/ μ L and 0.5 μ g/ μ L concentrations of ciprofloxacin-containing Brain Heart Infusion (BHI) media at 37 °C overnight. Bacterial colonies were isolated, and purification was done in the same concentration of culture media. Species of purified colonies were identified using biochemical identification methods and further confirmed through Sanger sequencing. DNA were extracted using boiling-lysis method and subjected to PCR to detect quinolone resistant genes (qnrB, qnrS and qnrA).

Results: Six distinct bacterial colonies were isolated from three water samples using ciprofloxacin-containing BHI at two concentrations: $0.25 \mu g/\mu L$ (3 colonies) and $0.5 \mu g/\mu L$ (3 colonies). Through Sanger sequencing, five were identified as *Escherichia coli*, while one colony was identified as *Bordetella* spp. Isolated bacteria tested negative for *qnrB*, *qnrS* and *qnrA* genes.

Conclusions: CRB was detected in the waterways that can survive in concentrations of both $0.25\mu g/\mu L$ and $0.5\mu g/\mu L$ of ciprofloxacin. Their existence in the environment is important, as they could pose a significant threat to both human and animal health. However, the commonly abundant quinolone-resistant genes qnrB, qnrS, and qnrA were not detected. Despite the absence of these genes, the bacteria still exhibited phenotypic resistance to ciprofloxacin. Thus, further analysis is required to elucidate the underlying molecular mechanisms contributing to the resistance phenotype in the bacteria under investigation. Additionally, it is essential to test the isolated bacteria for resistance to other antibiotics that are of critical importance in human medicine.

Keywords: Ciprofloxacin resistant bacteria, waterways, environmental contamination, antibiotic resistance genes

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