

TITLE: DIVERSITY OF EXTENDED-SPECTRUM B-LACTAMASES IN BACTERIAL ISOLATES FROM ISABELA RIVER IN DOMINICAN REPUBLIC

AUTORS: CADERÓN, V.¹; ROSARIO, C.D.¹; BONNELLY, R.¹; DUARTE, A.¹; CARRERAS, R.¹; RODRIGUEZ, L.¹; RAMOS, R.T.J.²; BARAÚNA, R.²; PERDOMO, O.¹; FRANCO, E. F.^{1,2}

INSTITUTION: 1. Instituto Tecnológico de Santo Domingo (INTEC); 2. Universidade Federal do Pará-UFPA

ABSTRACT

Although antibiotic resistance genes (ARGs) are naturally prevalent in lotic ecosystems such as rivers, their outstanding ability to disseminate in untreated anthropogenic effluents could lead to the rise of clinically important multi-drug resistant bacteria as shown in previous studies done in Europe and Asia. This issue is even more compelling in developing countries, where access to clean water and sanitary pipelines is restricted to the affluent districts, allowing illegal river bank householders to consume it without proper treatment as in the Dominican Republic. For this study, we sampled three regions of Isabela river with contrasting characteristics (pristine, medium anthropogenic impact and high anthropogenic impact). The main defining characteristics for these regions to be selected were the relative population density surrounding the river and the presence of illegal untreated effluents. We analyzed water from each region to compare the prevalence of multi-drug resistant bacteria to understand the diversity and complexity of antibiotic resistance genes and genetic mobile elements in lotic ecosystems impacted by untreated effluents. For this purpose, water sampled from each region were vacuumed-filtered (Millipore) with a 0.22- μ m-pore-size nylon membrane and two membranes from each replicate were obtained and cultured in two MacConkey agars, one supplemented with cefotaxime and other supplemented with imipenem. Individual colonies were then purified in the same medium and stored. The antibiotic susceptibility test was performed on a BD Phoenix platform following the CLSI guidelines. Phylogeny affiliation of the colonies was determined through MALDI-TOF and DNA from each individual colony was extracted with Qiagen Dneasy Blood and Tissue kit. ARGs were amplified using PCR for each colony to determine the resistance genotype. One isolate was selected for whole genome sequencing. Nine samples from three sites in March 2019 we found to have water quality parameters were within limits, also nineteen bacterial multi-drug resistant isolates were obtained. From the isolates obtained, 17 of 19 were resistant to most beta-lactamases and were resistant to at least 3 different families of antibiotics. Most antibiotics tested resulted in complete resistance from all isolates except trimethoprim, only 63.2% were resistant; amoxicillin/clavulanate with 74% resistance; and imipenem, with 89.5% resistance. One of these isolates will be sequenced.

Keywords: antibiotic resistance, extended spectrum β -lactamases, environmental resistance

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