

## Screening and Quantification of Tetracycline and Sulfonamide Resistance Genes in Natural Environmental Samples

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Screening of antibiotic resistance is important factor for assessing risk of antibiotic resistance in aquaculture environment. In this study, tetracycline (*tet A* and *tet M*) and sulfanamide (*sul 1* and *sul 2*) resistance genes were screened in 24 sampling locations including marine water samples at different levels (100, 500 and 1000 m), lagoons (Chilow, Puttalam), rivers (Kelani, Kalu, Walawe, Mahaweli) and from Horton plains. The Tetracycline (TET and OTC) and Sulfanamide (SUF) contamination levels in the sampling locations were analyzed by using High Performance Liquid Chromatography (HPLC). DNA was extracted and Polymerase Chain Reaction (PCR) method was followed to screen the presence of Antibiotic Resistance Genes (ARGs) in the samples. Real time PCR assays were employed to quantify ARGs. None of the samples were shown the contaminations of tetracycline and sulfonamides. The *tet (M)* gene was detected in 7 sampling sites. From the tested ARGs, both *tet A* and *tet M* were found at 3 marine samples (100m), 2 lagoon samples, whereas both *sul 1* and *sul 2* were recorded at only in one lagoon sample. No resistance genes were found in >100 m depth marine samples and in any river samples. The average concentrations of the *Sul 1* and *Sul 2* was ranged between  $0.12 \times 10 - 1.56 \times 10$  copies/ml,  $0.05 \times 10 - 0.56 \times 10$  copies/ml in lagoon samples while *tet M* and *tet A* were ranged  $0.11 \times 10 - 1.23 \times 10^2$  copies/ml,  $0.13 \times 10 - 4.56 \times 10$  copies/ml in marine samples. The presence of tetracycline and sulfonamide resistance genes may limit the effectiveness of antibiotics in treating fish illnesses, thereby causing a potential risk to the aquaculture industry.

**Keywords:** Antibiotic resistance genes, Tetracycline, Sulphonamide