Teixeira, A. M., Matos, D., Halwatura, L., Aga, D., & Manaia, C. (2023). *Monitoring of antibiotic resistance and contaminants of emerging concern in small-scale wetland-based municipal treatment systems*. Poster session presented at MICROBIOTEC'23, Portugal.

Human sewage is one of the major sources of antibiotic resistant bacteria (ARB), antibiotic resistance genes (ARGs) and chemical contaminants of emerging concern (CECs). Wastewater treatment is a crucial barrier to prevent environmental contamination. This study aimed to assess the efficacy of three constructed wetlands (CWs) (<200 p.e.) located in small villages, in Northern Portugal dedicated to the treatment of domestic effluents. Twenty-four hour composite samples of influent and effluent were collected over four campaigns in the winter (March), spring (May), summer (July) and autumn (October) during the year of 2023. Triplicate total DNA extracts from 50-250 ml of sample were used to measure the abundance of biomarkers associated with anthropogenic contamination (intl1; uidA; sul1; crAssphage; ermB, ermF, qacE Δ 1, tetX, mefC and aph($3^{''}$)-ib)1 and the bacterial load through 16S rRNA gene quantification by qPCR. Cultivable Escherichia coli and total coliforms were quantified on Chromogenic Coliform Agar (CCA). CECs extracted by solid-phase extraction (SPE) were quantified by liquid chromatography-mass spectrometry (LC-MS). Total coliforms ranged from 4.5 – 6.1 log UFC/mL in influent samples and 1.7 – 3.8 log UFC/mL in effluent samples. Total bacterial abundance, assessed based on the 16S rRNA gene, ranged between 8.0 – 8.9 log-units gene copy/mL in influent and 6.3 – 7.6 log-units in effluent. The biomarkers tested showed removal values of up to 3 log-units gene copy/mL. The chemical analysis of 119 compounds showed that pain killers as acetaminophen, illicit drugs as cocaine, antihyperlipidemic as fenofibric-acid, antihypertensives as irbesartan or psychoactive drugs as oxazepam were present in all samples (1st and 2nd campaigns), persisting after treatment. The results obtained so far suggest that the three CWs have good treatment capacity, with an important role of macrophytes, although dependent on the growth stage along the year, and with limited capacity to remove CECs.