Selected Emerging Pollutants and Microbial Diversity Monitoring In Wastewater Treatment Plant Maturation Ponds

Lehlohonolo B. Qhanya^{1*}, Olga de Smidt¹, Abidemi O. Ojo¹, Errol Cason², Gabre Kemp³

¹ Centre for Applied Food Sustainability and -Biotechnology (CAFSaB), Faculty of Health and Environmental Sciences, Central University of Technology, Bloemfontein, South Africa.

* lqhanya@cut.ac.za, odesmidt@cut.ac.za, aojo@cut.ac.za

² Department of Animal Sciences, University of the Free State, Bloemfontein, South Africa CasonED@ufs.ac.za

³ Department of microbiology and biochemistry, University of the Free State, Bloemfontein, South Africa <u>kempg@ufs.ac.za</u>

Abstract

The presence of emerging pollutants (EPs) in the water matrices has received considerable attention in the last decade and has been reported in literature worldwide. The current conventional wastewater treatment processes utilized cannot completely remove EPs and these untreated pollutants are eventually released into the downstream water systems. Many wastewater treatment plants (WWTPs) in South Africa utilize wastewater stabilization ponds (WPS), particularly maturation ponds, as the final stage in treating municipal sewage water [1]. EPs such as plasticizers, flame retardants, pesticides, personal care products and pharmaceuticals are known to be endocrine disrupting chemicals (EDCs) which could pose a risk to human and ecological health [2], [3]. South Africa does not have a regulation to monitor the EPs water matrices. This study aimed to monitor the physico-chemical properties, which included selected EPs, and microbial diversity in WWTPs maturation ponds. Wastewater samples were collected from the inflow and outflow maturation ponds of a WWTP located in Bloemfontein, South Africa over 6 months. Physico-chemical parameters like pH, DO, ORP, EC, Temperature and TDS were measured onsite using multi-parameter probes. Atrazine, 17a-ethinylestradiol (EE2), Triclosan and Bisphenol A were measured using LC-MS/MS and targeted metagenomics (16S rRNA and ITS) were assessed using the Illumina MiSeq platform. pH and dissolved oxygen (DO) significantly influenced the microbial diversity found in both ponds. EDC concentrations fluctuated with the following ranges: atrazine $(0.025 - 0.15 \ \mu g \ L^{-1})$, EE2 $(0.0057 - 125 \ \mu g \ L^{-1})$, triclosan $(0.147 - 1.87 \ \mu g \ L^{-1})$ and bisphenol A $(0.003 - 0.145 \ \mu g \ L^{-1})$. When comparing the ponds, it was evident that the maturation ponds did not significantly influence the removal of most EDCs entering the inlet and outlet ponds. Microbial composition assessment results showed a total of 10121 high-quality 16S rDNA and ITS. All the DNA sequences obtained belonged to 43 phyla, 1978 families and 1774 genera. These ASVs were clustered in QIIME 2 at 99% similarity, with 7680 belonging to the bacterial taxa and 2441 belonging to the fungal taxa. Proteobacteria was the most abundant bacterial phylum at 30,3% and Ascomycota (70,2%) for the fungal taxa. Genus *Pirellula* and *Mycobacterium* dominated the outflow pond samples. Further assessment of the core microbiome, with a specific focus on the bacterial diversity of *Flavobacterium*. Functional content prediction revealed the presence of several xenobiotic metabolism pathways associated with the degradation of EDCs (Bisphenol A and Atrazine) were present. This research presents a baseline for selected EP concentrations and an understanding of the core microbiome of the maturation ponds on a genetic and functional level. This data could be useful in future research for EPs degrading microbial consortia.

References

 N. A. Khan, S. U. Khan, S. Ahmed, I. H. Farooqi, M. Yousefi, A. A. Mohammadi, and F. Changani, "Recent trends in disposal and treatment technologies of emerging-pollutants- A critical review," Jan. 01, 2020, Elsevier B.V. doi: 10.1016/j.trac.2019.115744.

- [2] I. C. Vasilachi, D. M. Asiminicesei, D. I. Fertu, and M. Gavrilescu, "Occurrence and fate of emerging pollutants in water environment and options for their removal," Water (Switzerland), vol. 13, no. 2, pp. 1–34, 2021, doi: 10.3390/w13020181.
- [3] R. Kumar and P. Kumar, "Wastewater stabilisation ponds: Removal of emerging contaminants," Journal of Sustainable Development of Energy, Water and Environment Systems, vol. 8, no. 2, pp. 344–359, 2020, doi: 10.13044/j.sdewes.d7.0291.