Mass spectrometry in wastewater-based epidemiology (WBE) for the determination of small and large molecules as biomarkers of exposure. Needs for COVID-19 testing with environmental proteomics (EP-WBE).

Damià Barceló[1,2], Yolanda Picó[3], Carlos Perez-Lopez[1], Antoni Ginebreda[1], Romà Tauler[1], Montserrat Carrascal[4], Joaquin Abian[4],

[1] Water and Soil Quality Research Group, Department of Environmental Chemistry, IDAEA-CSIC, C/Jordi Girona 18–26, 08034 Barcelona, Spain

[2] Catalan Institute for Water Research (ICRA), C/Emili Grahit 101, 17003 Girona, Spain

[3] Environmental and Food Safety Research Group-University of Valencia (SAMA-UV), Desertification Research Centre (CIDE), Joint Center CSIC-University of Valencia-Generalitat Valenciana, Moncada Naquera Road km 4.3, 46113 Moncada, Valencia (Spain).

[4] Biological and Environmental Proteomics, Institute of Biomedical Research of Barcelona, Spanish National Research Council (IIBB-CSIC/IDIBAPS), Rosellón 161, E- 08036 Barcelona, Spain

Wastewater-based epidemiology (WBE) estimates collective consumption or exposure to chemicals or pathogens by tracking the substances excreted in the population's wastewater. WBE is generally applied using mass spectrometry (MS) and provides a fingerprint of health hazards, habits, and lifestyle of the population served by a wastewater treatment plant (WWTP). This presentation reports three applications of WBE using MS: 1) Detection for small molecules as biomarkers, 2) Detection for large molecules as biomarkers, and 3) Detection of viral proteins of SARS-CoV-2 by High Resolution MS (HR-MS) and MALDI-TOF-MS.

The identification of small molecules used as biomarkers is linked to the search of human biomonitoring (HBM) studies[1]. Few examples of non-target analysis are reported here. By doing this, the identification of unknown compounds is made feasible thanks to the possibility to share the most probable empirical formula and fragmentation pattern with several internet available databases. In short, with the combined application of HRMS, the improvement of raw data processing, identification systems, and bioinformatics, a new window for the discovery of new useful biomarkers in wastewater can be achieved.

Proteins can be a good indicator of prevalent diseases in the population and, together with biomarkers composition in sewage, can reflect the surrounding human activities (e.g., industry, agriculture, and pest control) as well as the health status of populations [2]. While biomarkers might be found in the soluble part of the effluent, proteins are likely expected to be also in the particulate matter because they travel in the water media attached to the surface of particles and bacteria. MS approaches based on HR-MS and MALDI–TOF-MS could be feasible for rapid detection and routine monitoring of proteins. By applying HRMS, we demonstrated the presence in urban sewage waters of proteins from diverse origins, including human proteins such as uromodulin, α-amylase, and S100A8, which have been proposed as health

markers associated with renal function or stress-related changes. Due to the large number of data recorded, the bioinformatics treatment to recognize the complex pattern of the omics already proposed for small molecules becomes crucial here. In this work, a recent metabolomic mass spectrometry non-target analytical methodology named the Regions of Interest-Multivariate Curve Resolution (ROIMCR)[3] was applied to our proteomics data. This methodology combines the searching of the Regions of Interest (ROI) and the Multivariate Curve Resolution-Alternating Least Squares (MCR-ALS) methods for the resolution of the constituents of the analyzed samples. In short, Environmental Proteomics (EP) applied to WBE provides additional information on human protein biomarkers together with characteristic peptides of other species such as rat, mouse, cattle, chicken as well as distinctive peptides of a large number of bacteria.

We believe that the EP-WBE approach can be applied to SARS-CoV-2 proteins and peptides in the coming years, in a similar way it has already been used for clinical samples. EP-WBE is a complementary tool to PCR and follows the concept of "thinking outside the box" to detect Covid-19 outbreak.

REFERENCES

[1] Y.Picó, D. Barceló. Identification of biomarkers in wastewater-based epidemiology: main approaches and analytical methods TrAC - Trends in Analytical Chemistry (2022) In press.

[2] M. Carrascal, J. Abian, A. Ginebreda, D. Barceló. Discovery of large molecules as new biomarkers in wastewater using environmental proteomics and suitable polymer probes. Science of the Total Environment 747 (2020) 141145.

[3] C. Perez-Lopez, A. Ginebreda, M. Carrascal, D. Barceló, J. Abian, R.Tauler.
 Non-target protein analysis of samples from wastewater treatment plants using the regions of interest-multivariate curve resolution (ROIMCR) chemometrics method. Journal of Environmental Chemical Engineering 9 (2021) 105752.