Received: 4 October 2022 | Accepted: 7 October 2022
DOI: 10.1111/1462-2920.16247

CRYSTAL BALL

environmental microbiology | Applied Microbiology International

# Picturing natural microbiomes: Matrix-assisted laser desorption/ionization mass spectrometry imaging for unravelling the architecture of environmental microbial communities

**Carmen Michán**

Departamento de Bioquímica y Biología Molecular, Campus de Excelencia Internacional Agroalimentario CeiA3, Universidad de Córdoba, Campus de Rabanales, Córdoba, Spain

**Correspondence**
Carmen Michán, Departamento de Bioquímica y Biología Molecular, Campus de Excelencia Internacional Agroalimentario CeiA3, Universidad de Córdoba, Campus de Rabanales, Edificio Severo Ochoa, E-14071 Córdoba, Spain.
Email: bb2midoc@uco.es

**Funding information**
Spanish Ministry of Science and Innovation (MICINN), Grant/Award Number: PID2019-110049RB-I00

Ecosystems are complex matrixes with a multitude of physical, chemical, and biological interactions that are continuously changing both along space and time. In the current scenario of climate warming, their physicochemical properties are quickly altered, and thus, the living beings could be heavily affected (Malhi et al., 2020). Microorganisms can be found in almost every environment in the biosphere, where they provide most of their metabolic potential. Unravelling the structure of the microbial communities is crucial to identify hotspots of vulnerability and resilience and to develop possible interventions that can decelerate or even stop undesirable changes.

The surge of OMIC techniques has undoubtedly proven useful in deciphering the complex nature of the microbial communities in natural ecosystems. Metagenomics have certainly contributed, and metaproteomics and metabolomics are quickly being developed although they still have severe limitations, since isolation of proteins and metabolites from complex matrixes can be challenging (Herruzo-Ruiz et al., 2021). Nevertheless, all these techniques do not give information of the spatial structure of the microbial communities. On the other hand, transmission and scanning electron microscopic techniques can provide with beautiful 2D and 3D images, but the different microorganisms can be rarely identified, and little information of the biomolecules and metabolites is obtained.

Among omics, those coupled with label-free matrix-assisted laser desorption/ionization (MALDI) mass spectrometry (MS) allows the identification and quantification of complex molecules as peptides, lipids, and metabolites from biological samples. Both microscopic and MALDI-MS are combined in MALDI imaging spectrometry (MSI) producing snapshots with semi-quantitative identification of biomolecules, which can be associated with 2D microscopic images. Integration of different MSI analyses, serial slicing of the samples and successive sampling along time can provide information about different biomolecules (e.g. proteins, peptides, lipids, metabolites), and their changes along space and time in a certain biological sample. Although MSI was initially used in basic research of plant and animal tissues, it is currently mainly used in more applied biomedical and pathological studies, for example, to analyse the distribution and effects of therapeutical drugs in a certain whole-body or organ (Chicano-Gálvez et al., 2022). To date, environmental applications have been very limited, most of them covering the disposition and the effect of toxic chemicals into animals or plants to elucidate their mechanisms of action (Poter & Sánchez, 2022) and most microbiological studies are in man-made plated/cultured environments. Current efforts include the study of natural ecosystems

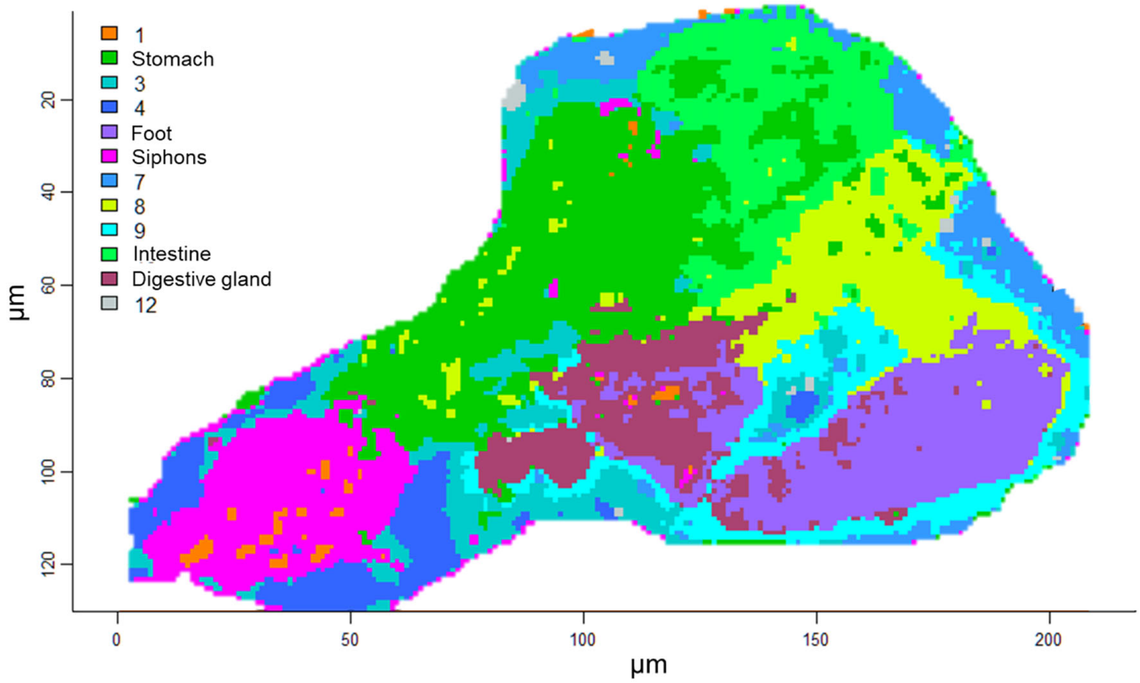


FIGURE 1 MSI peptidome analysis of *Scrobicularia plana* clam

that are already embedded in a matrix, that is, biofilms, or the microbiome inside more complex organisms as bivalves for aquatic environments monitoring (Figure 1, manuscript in preparation) (Michán et al., 2021).

Nevertheless, MSI has important challenges that need to be improved, including wider identification of molecules, higher sensitivity particularly of low abundant analytes, and selection of non-interfering matrixes. Bottlenecks for its use in natural environmental studies are also even more difficult and include fixation of inconsistent environments and management of an extremely high amount of data.

Sooner rather than later all the technical problems will be overcome, and we will be able to see beautiful 3D kaleidoscopic images of the microbiomes from natural ecosystems, plus the molecular interactions among them, both intra- and inter-species. Let us hope that this understanding will help in tackling the degradation of natural ecosystems and to improve our resilience to increasing situations of extreme climate episodes and pollution.

## ACKNOWLEDGEMENTS

The authors would like to thank Dr. Eduardo Chicano-Gálvez and Dr. Ana M. Herruzo-Ruiz for introducing MSI to us and showing its potential, and to Eduardo and Dr. José Alhama Carmona for critical reading of the manuscript. MSI studies undertaken by the author are currently supported by the Spanish Ministry of Science and Innovation (MICINN, PID2019-110 049RB-I00).

## CONFLICT OF INTEREST

The author has no conflict of interest to declare.

## ORCID

*Carmen Michán* https://orcid.org/0000-0003-2921-0987

## REFERENCES

Chicano-Gálvez, E., Salzet, M. & Fournier, I. (2022) MALDI mass spectrometry imaging and spatially-resolved proteomics. In: Siegel, T.P. (Ed.) *MALDI mass spectrometry imaging: from fundamentals to spatial omics, new developments in mass spectrometry series*. London: The Royal Society of Chemistry.

Herruzo-Ruiz, A.M., Fuentes-Almagro, C.A., Jiménez-Pastor, J.M., Pérez-Rosa, V.M., Blasco, J., Michán, C. et al. (2021) Meta-omic evaluation of bacterial microbial community structure and activity for the environmental assessment of soils: overcoming protein extraction pitfalls. *Environmental Microbiology*, 23, 4706–4725. https://doi.org/10.1111/1462-2920.15673

Malhi, Y., Franklin, J., Seddon, N., Solan, M., Turner, M.G., Field, C. B. et al. (2020) Climate change and ecosystems: threats, opportunities and solutions. *Philosophical Transactions of the Royal Society*, 375, 20190104.

Michán, C., Blasco, J. & Alhama, J. (2021) High-throughput molecular analyses of microbiomes as a tool to monitor the wellbeing of aquatic environments. *Microbial Biotechnology*, 14, 870–885.

Poter, J.E. & Sánchez, L.M. (2022) MALDI mass spectrometry imaging for microbiology. In: Siegel, T.P. (Ed.) *MALDI mass spectrometry imaging: from fundamentals to spatial omics, new developments in mass spectrometry series*. London: The Royal Society of Chemistry.

**How to cite this article:** Michán, C. (2023) Picturing natural microbiomes: Matrix-assisted laser desorption/ionization mass spectrometry imaging for unravelling the architecture of environmental microbial communities. *Environmental Microbiology*, 25(1), 135–136. Available from: https://doi.org/10.1111/1462-2920.16247