## Microplastics as cargo for pathogenic bacteria and hotspot of antimicrobial resistance genes transfer? Insights into microbial communities and the emergence of antibiotic resistance

## Méril MASSOT, Stéphanie BEDHOMME

## CEFE, CNRS, 1919 route de Mende, 34293 Montpellier

Microplastics (MP) have emerged as a significant source of pollution in marine ecosystems<sup>1</sup>. Microbial biofilms rapidly establish on the surface of these MPs and studying these microcosms is crucial to characterize the role of MP as vector for pathogens, and their role as reservoir of antimicrobial resistance (AMR) genes.

To tackle these questions, we used the fish-pathogen *Vibrio anguillarum*, as model pathogenic bacteria and evaluated by flow cytometry and microscopy the intraspecific variability in the efficiency of colonization of different types of MP: polystyrene spheres (30  $\mu$ m), and polypropylene and polyethylene terephthalate fragments (38-50  $\mu$ m). The set of sixteen strains tested revealed a broad spectrum of attachment phenotypes. We therefore looked for an association between colonization phenotype and gene content and phylogenetic relatedness.

Furthermore, we investigated the potential role of MP in structuring microbial community composition in marine environments. MP samples were incubated at three distinct locations in the Mediterranean Sea, representing varying levels of human activities: an aquaculture area (étang de Thau), a harbor (Carnon city), and a less polluted area (national reserve of Cerbères-Banyuls). Variation in composition of the MP-associated biofilms among locations and MP polymers were analyzed by shotgun metagenomics, using MetaSpades<sup>2</sup>. Obtained sequences were screened against the CARD database<sup>3</sup> for AMR gene identification.

To understand the influence of marine MP-associated biofilms on the emergence of antibiotic-resistant pathogenic bacteria, we investigated the impact of natural marine microbial communities on the attachment of three Vibrio strains covering the range of attachment phenotype. Attachment efficiency was measured by comparing attachment to sterile MP versus colonized MP. Additionally, we are planning to study whether the spatial structure provided by MP in the water column promotes horizontal transfer of resistance genes detected in these communities.

It is crucial to decode the potential consequences of MP-associated biofilms on the emergence of antibiotic-resistant pathogenic bacteria. The colonization of microplastics by bacteria in marine environment enables their long-distance transport, facilitating the spread to susceptible new host populations.

## References

- Bowley, J., Baker-Austin, C., Porter, A., Hartnell, R. & Lewis, C. Oceanic Hitchhikers Assessing Pathogen Risks from Marine Microplastic. *Trends Microbiol.* 29, 107–116 (2021).
- Nurk, S., Meleshko, D., Korobeynikov, A. & Pevzner, P. A. metaSPAdes: a new versatile metagenomic assembler. *Genome Res.* 27, 824–834 (2017).
- 3. Alcock, B. P. *et al.* CARD 2020: antibiotic resistome surveillance with the comprehensive antibiotic resistance database. *Nucleic Acids Res.* **48**, D517–D525 (2020).