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## Ecological functions and environmental fate of exopolymers of *Acidobacteria*

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# Summary

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*Acidobacteria* is a widely distributed phylum but their functional roles in ecosystem processes are still largely elusive. The high abundance and ubiquity of *Acidobacteria* in different environments, especially soils, raises intriguing questions about the physiological traits underlying their marked abundance. This lack of fundamental knowledge is mostly due to difficulties to isolate *Acidobacteria* and their slow growth in vitro. Increased knowledge of the traits of different *Acidobacteria* subdivisions is critical for understanding their persistence in soil as well as their interactions with other soil microorganisms.

The *Granulicella* genus belongs to the class *Acidobacteriia* and is known to produce copious amount of Extracellular Polymeric Substances (EPS) in culture medium. EPS are fundamental for microbial life and provide an ideal substrate for chemical reactions, nutrient entrapment and protection against environmental stresses such as salinity and drought. Microbial EPS can enhance the aggregation of soil particles and benefit plants by maintaining the moisture of the environment and trapping nutrients. The EPS of the two strains *Granulicella* sp. WH15 and 5B5 were chemically and physically characterized previously, showing interesting emulsification properties. Therefore, the major goal of my thesis was to investigate and understand the metabolism of these two *Granulicella* sp. strains, and the functions and environmental fate of EPS of *Granulicella* sp. WH15 (WH15EPS). We optimized growth conditions of *Granulicella* sp. WH15 and 5B5 in the laboratory to obtain higher biomass and enhance EPS production while using multi-omics approaches to investigate their metabolic adaptations. Furthermore, we used high-throughput sequencing to detect other soil microorganisms and their functions associated with the degradation of WH15EPS.

In **chapter 3**, we showed that the transcriptional and proteomic responses of *Granulicella* sp. WH15 grown at different concentrations of cellobiose resulted in higher expression of genes encoding excretory functions and reallocation of resources to maintain basic cell metabolism instead of generating new cell biomass. In **chapter 4**, we observed that the addition of a mix of trace elements to culture medium improved significantly the growth yield of *Granulicella* sp. WH15 and 5B5. After evaluating the effects of each of the trace elements separately, our results showed that manganese (Mn) had a significant positive effect on the growth of both strains. Further proteomic and genomic analyses showed that the strains had different proteomic profiles and several uncharacterized metal ion transporters that could be involved in metal ion homeostasis and could contribute to survival under high manganese concentrations.

Optimization of carbon concentration and manganese in culture medium allowed our strains, especially WH15, to grow faster in laboratory conditions, producing extractable amounts of EPS.

In **chapter 5**, we labeled WH15EPS with  $^{13}\text{C}$  and investigated its effect on the assembly and co-occurrence of the active bacterial and fungal communities in topsoil by stable isotope probing (SIP). Our results demonstrated that WH15EPS was primarily assimilated by *Planctomycetes*, *Verrucomicrobia*, *Ascomycota* and *Basidiomycota* and co-inertia analysis

suggested overall relationships between these bacterial and fungal kingdoms. We observed the incorporation of WH15EPS by *Singulisphaera* and its connections to other *Planctomycetes* and *Acidobacteria*, which were not reported before.

In **chapter 6**, we applied WH15EPS as an enrichment factor to target microorganisms and functions associated with EPS degradation through culture-independent and culture-dependent techniques. Our results showed a large diversity of glycoside hydrolase families with biotechnological potential and a high number of unclassified microorganisms that could be targeted for future studies.

In **chapter 7**, I integrated the overall findings of my thesis and discussed the most important observations concerning the impact of carbon sources and trace elements on the physiology of *Granulicella* and, more general, the ecological functions and environmental fate of EPS of *Acidobacteria*. The data presented in this thesis establish a solid fundamental basis for more mechanistic studies of *Acidobacteria* and other uncultivated microbes.