Visualization of the Ionome in Planktonic Symbioses

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In the ocean, planktonic microorganisms display a wide range of trophic modes, such as symbiotic associations [1]. The association between the single-celled radiolarians (host) and their symbiotic microalgae is one of the most prevalent eukaryotic symbioses in surface waters where nutrients are poorly available. However, despite their key ecological role, the functioning and physiology of the partnership remain poorly understood. The study of microbial symbioses where partners cannot be separated calls for dedicated high-resolution imaging methods that maintain the physical and chemical integrity of the host-symbiont association. More particularly, chemical imaging can provide information about the homeostasis of the nutrients (e.g. ionome) at the subcellular level, therefore highlighting the metabolic capacity and needs of the host and the symbiont [2]. The ionome can also provide information about the quality of the energy that is transferred up the food web and exported to the deep ocean.

Here, we visualized the ionome (composition and distribution of nutrients) in intact planktonic symbioses at the subcellular level, using a combination of chemical imaging instruments, such as secondary ion mass spectrometry (NanoSIMS) and synchrotron X-ray fluorescence (S-XRF). Live cells (host and the symbiotic microalga Brandtidinium nutricula) were collected in the Mediterranean Sea and cryo-fixed with high-pressure freezing. Vitreous cells were then freeze-substituted and embedded in resin. Consecutive semi-thin sections were analyzed in parallel on nanoSIMS and S-XRF to unveil the macronutrients (nitrogen and phosphorous) and trace metals (iron and manganese), respectively, in organelles of each partner. Additional consecutive ultra-thin sections were also used for electron microscopy to obtain the ultrastructure of the regions of interest.

The subcellular distribution of nitrogen (nanoSIMS: \(^{12}\text{C}^{14}\text{N}\)), which is limiting in the ocean and mainly contained in proteins and amino acids, shows that symbiotic microalgae are N-rich, particularly their photosynthetic machinery (Figure 1). Sulfur (\(^{32}\text{S}\)) is also highly concentrated in algal cells especially in vacuoles (Figure 1). By contrast, phosphorous content unveiled by nanoSIMS (\(^{32}\text{P}^{16}\text{O}_{2}\)) and S-XRF is lower in microalgae than in the host (Figures 1 and 2). Iron (Fe) exhibits a high concentration in specific vacuoles of the host, presumably as a source for the symbiotic microalgae, while manganese is homogeneously distributed in both partners (Figure 2). Our nanoscale elemental mapping paves the way for complementary studies to understand the metabolic interactions in planktonic symbioses.
References:
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Figure 1. From left to right, mapping of Nitrogen ($^{12}$C$^{14}$N), Phosphorous ($^{32}$P$^{16}$O$_2$) and Sulfur ($^{32}$S) in the symbiotic microalga and host cell unveiled by nanoSIMS. Graph below: The content of these three secondary ions were compared across the dashed line in the organelles of the symbiont.

Figure 2. Quantitative mapping of Phosphorous (red) and the trace metals Iron (green, left image) and Manganese (green, right image) in the host and symbiotic microalgal cell unveiled by S-XRF.