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The S-Layer of *Nitrosopumilus maritimus*

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In 2005, the first ever isolated and cultivated **A**mmonia **O**xidizing **A**rchaeum (AOA), the mesophilic, marine and extremely small Archaeon *Nitrosopumilus maritimus* [1] was successfully enriched from a tropical marine tank. Only three years later, the use of ribosomal proteins as phylogenetic markers led to the proposal of a new archeal phylum, the Thaumarchaeota [2], with *N. maritimus* as one of the first members (see fig. 1 [3]). Due to its extraordinary metabolic [4] and phylogenetic features, *N. maritimus* rapidly gained the interest of researchers around the world. Since then, it has been recognized that AOA make a significant contribution to the first and rate limiting step in nitrification [5] and are highly abundant in their habitats. The discovery of a novel type of ammonia monooxygenases and putative new metabolic pathways [4] further emphasize their role in the global nitrogen circle.

Nevertheless, ultrastructural information on *N. maritimus* is still scarce; the S-Layer as its most obvious surface structure has not yet been investigated (see fig. 2). The quasi-periplasmic space between the cellular membrane and the S-Layer lattice comprises about one third of the total volume of a single cell. This astonishing figure points to the importance of the S-Layer for *N. maritimus* cells and indicates that is has to fulfill a certain, yet unknown, function. The question whether it is also providing a functional compartmentalization to the cell remains to be answered. To address these topics, our current experiments focus on the structural and biochemical investigation of the S-Layer and its composing protein(s). This work gives a first insight into compositional, organizational and molecular details of the *N. maritimus* S-Layer. The investigative methods that were used for this study include TEM preparations [6] like freeze etching, (ultra-)thin sectioning, tomography and cryo tomography as well as biochemical approaches. The center to center-distance was determined to be ~20 nm. A two-dimensional reconstruction of the S-Layer lattice was done to identify its symmetry (p6 or p3), while a three-dimensional reconstruction provides information about putative pores and links between the protein subcomplexes. By this multi-technical approach we examined our target structure from different points of view, in 2D and 3D, regarding compositional and biochemical aspects. The resulting, overall image shows that the S-Layer has not only structural, but also a putative functional importance for *N. maritimus* cells. A comparison with other archaeal relatives will be given as a conclusion.

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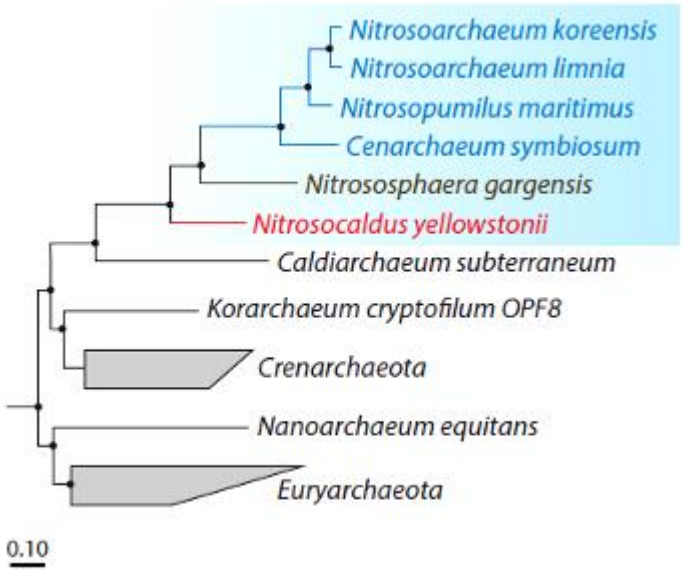

Figure 1:
Phylogenetic tree of the Archaea

Figure 1. Recently published phylogenetic tree of AOA, based on ribosomal proteins [3]; Thaumarchaeota are marked blue.

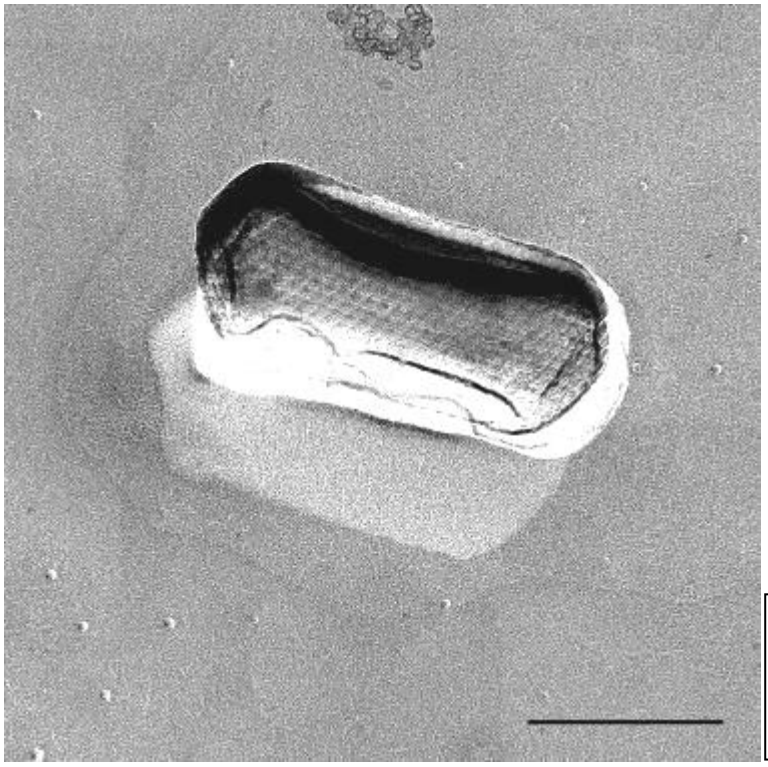

Figure 2:
Freeze etching preparation of *N. maritimus*;
bar = 200 nm

Figure 2. Freeze etched *N. maritimus* cell, revealing the S-Layer (unpublished data). The regularly arranged protein oligomers are clearly visualized; center to center distance ~ 20 nm, symmetry p6 or p3.