ESRF	**Experiment title:** Mesostructure evolution the of nacreous layers in marine shells	**Experiment number**: SC- 4155
Beamline:	**Date of experiment**: from: 24.03.2016 to: 29.03.2016	**Date of report**: 05.09.2017
Shifts:	**Local contact(s)**: Alexandra Pacureanu	*Received at ESRF:*
Names and affiliations of applicants (* indicates experimentalists): Igor Zlotnikov, Paul Zaslansky, Bernd Bayerlein		

Report:

Background

The objetive of this beamtime was to obtain high quality 3D information on the spatial arrangement of the nacreous layer of several molluscan shells. The nacreous layer is typically one of two or more layers that comprise the shell of many molluscan species. All layers are hierarchically structured biocomposites consisting of calcium carbonate and a very small percentage of organic material, such as proteins, polysaccharides and lipids, though the nacreous layer is known to be the mechanically most advanced. Its outstanding properties are based on its architecture, which exhibits a “brick and mortar” texture. The stiff mineral units, which consist of the calcium carbonate polymorph aragonite, form polygonal shaped platelets (~ 10 µm in diameter) that are stacked on top of each other in direction perpendicular to the surface of the shell, which corresponds to the direction of growth. The platelets are separated/surrounded by a softer organic phase – the interlamellar membranes, which are approximtely 40 nm thick. To understand how this complex structure is formed, the investigation of the spatial morphology is crucial. Previously performed experiments on ID19 allowed us to resolve the organic phase of another molluscan shell ultrastructure – the prismatic layer - where the organic phase is much thicker (1 µm). Using these data we could show that this mesostructure can be predicted using classical theories for normal grain growth and coarsening[1]. Here, we planned to perform similar morphological analyses on the nacreous layer, where the very thin interlamellar membranes require a tomographic technique that provides higher resolution.

Experiments and Setup at ID16a

The nacreous layers of species from three different molluscan genera were scanned at the nano-imaging beamline ID16A-NI. Cylindrically shaped samples, with a diameter of aproximately 300 µm, were prepared from the nacreous layer of the bivalves *Unio pictorum* and *Pincata nigra*, the gastropods *Strombus decorus* and *Haliotis coccoradiata*, and the cephalopod *Nautilus pompilius*. The samples were scanned using a beam energy of 33.6 keV and 1800 angular projections were taken over 180 degrees with exposure times of 1 s. Scans with pixel resolutions ranging from 60 nm to 25 nm were performed, whereas always 4 scans with varying sample to detector distances were taken to enable the holotomographic reconstructions.

[1] Bayerlein, B., Zaslansky, P., Dauphin, Y., Rack, A., Fratzl, P., & Zlotnikov, I. (2014). Self-similar mesostructure evolution of the growing mollusc shell reminiscent of thermodynamically driven grain growth. Nature Materials, 13(October), 1102–1107. http://doi.org/10.1038/NMAT4110

Analysis and Results

Due to the high-resolution setup at ID16A-NI we were able to resolve the thin organic interlamallar membranes (thicknesses are in the order of 40 nm) which separate the mineral nacreous platelets in *U. pictorum* and *P. nigra* (displayed for *U. pictorum* in Figure 1). Processing of the data enabled us to visualize the spatial arrangement and the morphology of nacre platelets in three dimensions for the first time. Analysis of these data will provide us with important information on the biomineralization prozess and the morphological evolvement of nacre ultrastructure. Furthermore, a comprehensive characterization of the spatial morphology of this material can contribute to the characterization of the advanced material properties of nacre, which are based on its hierachically structured architecture.

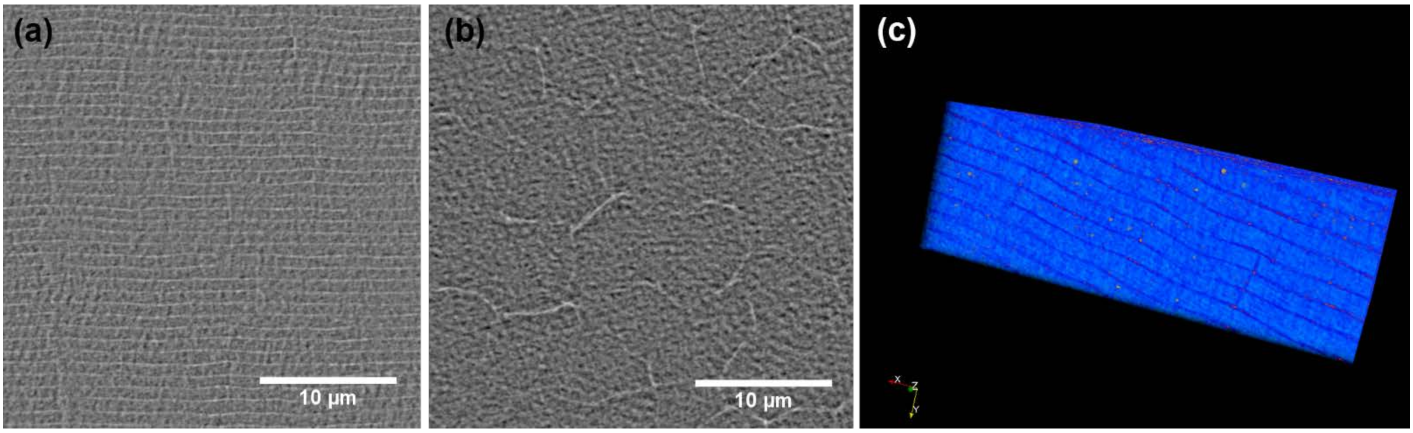

Figure 1: Reconstructions of the nacreous layer of *U. pictorum*. (a) cross-section of nacre in direction perpendicular to the shell surface. (b) cross-section of the platelets parallel to shell surface. Bright areas correspond to the organic phase, and dark areas to mineral component. (c) 3D visualization of a cropped region of the tomography data.