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Temperature-controlled experiments for the shell microstructural formation of S. broughtonii (Mollusca: Bivalvia)

Kozue Nishida[1*], Atsushi Suzuki[2], Ryosuke Isono[3], Yusuke Watanabe[3], Yukihiro Nojiri[4], Chiharu Mori[1], Mizuho Sato[2], Kei Sato[1], Takenori Sasaki[1]

[1]The University of Tokyo, [2]AIST, [3]MERI, [4]National Institute for Environmental Studies

A temperature experiment was performed to corroborate the thermal control of microstructural formation, and the cultured specimens were found to exhibit differences in shell formation by temperature. It has been suggested that the cyclical changes in the shell microstructures of *S. broughtonii* might be affected by temperature seasonality based on a study of field specimens (Nishida et al., 2012). This is the first report of temperature experiments in relation to the microstructural formation of shells. Additionally, this experiment contributes to the reconstruction of the paleoenvironments using shell microstructures and to our understanding of the mechanisms of shell microstructural formation.

We cultured specimens of *S. broughtonii* under five different temperature conditions at the Demonstration Laboratory, Marine Ecology Research Institute (MERI) in Kashiwazaki City, Niigata Prefecture, Japan. The investigated temperatures were 13 C, 17 C, 21 C, 25 C, and 29 C, and the specimens were cultured for approximately 58 days. We placed 5 aquariums (12 liters) in the laboratory with 5-7 specimens placed in each aquarium. We removed part of the marginal periostracum to determine shell growth during the experiment.

The shell sizes and increment of the shell deposition during the experiment show that the most rapid growth occurs at 17 C. Based on the d18O data, the specimens at 17 C, 21 C, 25 C, and 29 C formed shell material at each temperature condition. The thickness of the composite prismatic structure increases at higher water temperatures, and this trend is same as that of the field specimens. The specimen at 17 C showed the sharpest edge in the marginal part of the outer layer in comparison to the specimens cultivated at 21 C, 25 C, and 29 C. Accounting for the outer layer, the area of the composite prismatic structure increases as the water temperature is reduced. The growth increment of the crossed lamellar structure was relatively constant, whereas that of the composite prismatic structure increased rapidly as the thickness of the composite prismatic structure increased at cooler temperatures. This finding suggests that the optimum temperature for *S. broughtonii* growth as determined experimentally is consistent with the shell growth in the temperate area and that the formation of the composite prismatic structure increases the shell growth, especially the expansion of the growth increments in the outermost part of the outer layer.

Keywords: Bivalve, shell microstructure, Stable oxygen isotopes, temperature experiment, shell growth