



Live confocal imaging of intrashell cytoplasm and calcification process in a symbiotic large benthic foraminifer *Amphisorus kudakajimensis*

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Algal symbiont-bearing large benthic foraminifers are primary and carbonate producers as well as paleoenvironmental indicators in tropical and subtropical reef environments. Despite their importance, their cellular physiology is not well known. In this study, we have developed methods to observe *in vivo* images of a living symbiotic porcelaneous large foraminifer, *Amphisorus kudakajimensis*. Nikon A1 confocal laser scanning microscope with Calcein-AM as a fluorescent indicator was used to visualize morphology and streaming of cytoplasm in living *A. kudakajimensis*. The observation showed that in the marginal part of intrashell cytoplasm, cytoplasm gradually became highly branched and vacuolated. We also succeeded in observing elevated pH (pH 9.0) and lowered pH (pH 6.0) vesicles in reticulopodia-like cytoplasm, using a cell membrane-impermeable fluorescent pH indicator with low toxicity (HPTS). This study demonstrates the use of confocal microscopy in studying cytoplasmic dynamics and initial calcification process such as seawater endocytosis and alkalization of seawater vacuoles.