

Internal pH distribution and post-metamorphic biomineralization in the tubeworm, *Hydroides elegans*

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The serpulid tubeworms produce a diverse tube structure through controlled calcification. Cellular environment associated with actively calcifying serpulid tubeworms at metamorphosis were studied using pH and calcium sensitive indicators. With a notable degree of compartmentation, the thoracic region between the collars showed a high pH value above 8.5 and elevated calcium ion levels. As suggested by SEM-EDX results, such region also demonstrated a higher Ca signal. To analyze the presence of crystalline CaCO₃, the unpolished sample was characterized using SEM-EBSD at 20kV, this low voltage and non-destructive approach showed the direct formation of aragonite. Applying in situ lift-out technique at the calcified region, TEM specimen was prepared for structural analysis using selected area diffraction pattern. This study documents the cellular environment during the first calcification event in the serpulid tubeworm at the transition of metamorphosis and the subsequent aragonite formation.

Keywords: imaging, serpulid tubeworms, visualization, calcifier, biomineralization