

# Science and Technology Group Annual Report FY2025

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## **1 Introduction**

Cnidarians represent a large group of marine animals that includes sea anemones, jellyfish, corals and others. A unique feature of this phylum is the presence of nematocytes, stinging cells involved in defense and prey capture. The venom composition of cnidarian nematocytes as well as their discharge mechanism is still rather poorly understood. The main goal of the current project is to gain insights into the payload composition of these cells and establish a natural source protein purification workflow for nematocyst toxin isolation compatible with structure determination by single particle cryo-electron microscopy.

The sea anemone *Aiptasia* is a cnidarian phylum representative that is well known as a model organism in photosymbiosis research. *Aiptasia* anemones possess defensive filaments termed acontia that are released upon physical stimulation (Lam, Cheng et al. 2017). These contain a highly abundant nematocyte population, which makes them an ideal target for this project.

## **2 Activities and Findings**

Last year, I initiated extensive sample characterization and conducted preliminary experiments that enabled us to establish a robust approach for nematocyte protein payload sample preparation. In FY2025 we established a sample preparation pipeline, explored various plunge-freezing and data collection conditions, and performed analysis of the acquired single-particle cryo-EM data. These efforts led to the reconstruction of an initial density map for one of our targets (data not shown). Further analysis is currently ongoing.

Furthermore, to provide a broader biological context for these results, we also performed room-temperature volume EM on anemone samples, establishing a suitable sample preparation protocol and collecting a large dataset, now under analysis.

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In addition, as one of the major FY2025 activities, we collected, isolated, and preserved another cnidarian specimen by vitrification and room-temperature fixation protocols. This sample was also characterized using confocal microscopy and analyzed by micro-CT to elucidate structural features of interest. Further downstream experiments are ongoing, including acquisition of cryo-volume EM data. Cryo-volume EM pipelines were established and datasets were acquired for a third sample, a unicellular marine organism. These datasets are currently being processed and segmented. As these projects are conducted as sensitive collaborations, details on the nature of the last two samples will be disclosed upon acceptance of the relevant manuscripts or future agreement from my collaborators.

## **3 Collaborations**

All unit members, Sitsel Unit, OIST

Yuki Yoshioka and Eiichi Shoguchi, Satoh Unit, OIST

Malgorzata Hall, Imaging section, OIST

Prof. Cheryl Ames, Graduate School of Agriculture, Tohoku University

Prof. Shunichi Takahashi, Tropical Biosphere Research Center, University of the Ryukyus

Prof. Atsuko Tanaka, Tohoku University

Prof. Alexander Belyy, University of Groningen

## **4 References**

Lam, J., Y. W. Cheng, W. U. Chen, H. H. Li, C. S. Chen and S. E. Peng (2017). "A detailed observation of the ejection and retraction of defense tissue acontia in sea anemone (*Exaiptasia pallida*)."