Application of Synchrotron Radiation-Based Nano-Transmission X-ray Microscopy (SR-nTXM) on Marine Animal-Plant Symbiosis

Shao-En Peng (彭紹恩)[1,2], Huai-Ting Huang (黃懷葶)[1], Jian-Hua Chen (陳建樺)[3], Yen-Fang Song (宋豔芳)[3] , and Chii-Shiarng Chen (陳啟祥)[1,2]

[1]Institute of Marine Biotechnology, National Dong Hwa University, Hualien, Taiwan
[2]National Museum of Marine Biology and Aquarium, Pingtung, Taiwan
[3]National Synchrotron Radiation Research Center, Hsinchu, Taiwan

Endosymbiosis in cnidaria-dinoflagellate association plays critical role in regulating productivity of corals and related marine ecosystems. Cellular endosymbiosis with dinoflagellate *Symbiodinium spp*. (i.e. the symbiont or "zooxanthellae" in generic name) in corals (i.e. the host) is initiated by the internalization of symbionts via phagocytic process into host endoderm cells (fig 1). However, the mechanism by which the symbiont is able to reside inside the host cell and establish an obligatory and mutualistic association remain unclear after four decades of investigation. Due to the large quantity of calcium carbonate skeleton, it is difficult to analyze how endosymbiosis is regulated and coordinated at organismal level. Fortunately, the non-destructive synchrotron-radiation-based microscopy in energy range of the infrared and hard X-ray provides a solution for investigating 2D images for chemical composition and 3D tomography images within coral tissue.

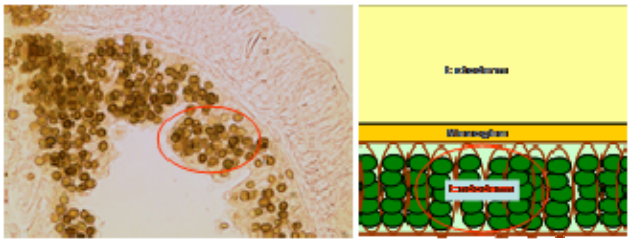

Figure 1. Photograph of the cross sectioned tentacles of stony coral (*Euphyllia glabrescens*).

With the comparative analysis of coral proteome, it revealed that the actin cytoskeleton protein of coral may play the important roles on endosymbiosis as the base for architectural support, molecule translocation or signal transduction (unpublished data). In this study, we applied the synchrotron radiation-based nano-transmission X-ray microscopy (SR-nTXM) to investigate the cellular localization of cytoskeleton protein in coral tissue.

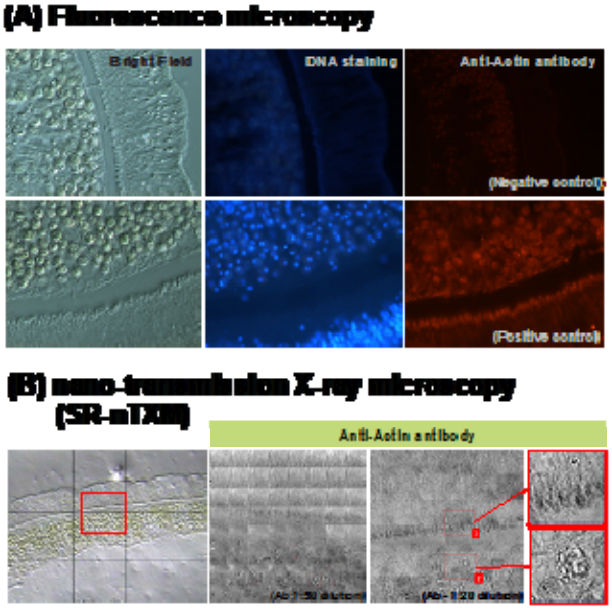

Figure 2. Analysis of the cellular location of coral actin cytoskeleton proteins by SR-nTXM. The tentacles of coral (*Euphyllia glabrescens*) were removed, paraffin-embedded, sectioned and subjected to immunostaining, then observed by fluorescence microscope of SR-nTXM. The fluorescence microscopy demonstrated the cellular location of actin cytoskeleton protein that recognized by anti-actin antibody (A). The SR-nTXM also revealed the same cellular location of actin cytoskeleton protein in coral tissue (B).

The preliminary data (Fig 2) demonstrated that the synchrotron radiation-based nano-transmission X-ray microscopy (SR-nTXM) should be applied to investigate the endosymbiosis in cnidarian-dinoflagellate successfully.