Ultrastructural analysis of chilled and cryopreserved coral larvae

Luca Cirino[1,2], Zhi-Hong Wen[1], Su-June Tsai[3] and Chiahsin Lin*[2,4]
[1]Department of Marine Biotechnology and Resources, National Sun Yat-sen University.
[2]Department of Planning and Research, National Museum of Marine Biology & Aquarium.
[3]Department of Post Modern Agriculture, Mingdao University.
[4]Graduate Institute of Marine Biology, National Dong Hwa University.

Abstract

The cryobanking of marine invertebrates in early developmental stages has been extensively explored, but few ultrastructural studies have investigated the effects of cryopreservation on biomaterials. Transmission electron microscopy (TEM) provides useful information on sites and extent of cryoinjuries. Ultrastructural observations were conducted on larvae of corals *Seriatopora caliendrum* and *Pocillopora verrucosa* after subjecting samples to vitrification and nano-laser warming, with lipid-free vitrification solutions (VSs) or VSs supplemented with erucic acid (EA), phosphatidylethanolamine, or linoleic acid. Samples then underwent TEM, and several ultrastructural parameters were examined. Cryoinjury in larval epithelia manifested as a lack of tissue coherency and affected the chloroplast structure of Symbiodiniaceae. Vitrified and laser-warmed coral larvae exhibited a patchy well-preserved ultrastructure. On average, liposome-supplemented VSs allowed for better preservation of thickness and area occupied by microvilli and flagella. Ultrastructural analysis of vitrified and nanoparticle laser–warmed coral larvae revealed the essential role of nanoparticle dispersion stability in the homogeneous laser thawing of the sample, as well as the relevance of cellular projection preservation to cryopreservation outcomes. The ultrastructural effects of vitrified, and nanoparticle laser–warmed coral larvae were examined for the first time. The findings will aid the development of efficient coral cryopreservation protocols.

Keywords: coral larvae, vitrification, laser-warming, ultrastructure

***Corresponding author. Email: chiahsin@nmmba.gov.tw**