Anemonia viridis primary cell culture: a new tool for cnidarian studies

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Member of the cnidarian phylum, the temperate symbiotic sea anemone Anemonia viridis is a relevant experimental model to investigate, in a post-genomics approach, the molecular and cellular events involved in the preservation or in the rupture of the symbiosis between the animal cells and their symbiotic microalgae, named zooxanthellae (Sabourault et al., 2009; Ganot et al., 2011; Moya et al., 2012).

In this aim, we developed a primary culture from A. viridis epidermal and gastrodermal cells. By adapting and optimizing previous published methods, i.e. spontaneous or chemical dissociations (Frank et al., 1994; Domart-Coulon et al., 2004), we extracted cells from whole tentacle or from a separated epithelial cell layers corresponding to the epiderm or the gastroderm. Each plating resulted in a heterogeneous primary culture of different cell types as discharged cnidocytes, free zooxanthella cells (A. viridis symbiotes) and many regular, small rounded and adherent cells (of 3-5 µm diameter). The different culture observations showed that this last cell group contains A. viridis epithelial undifferentiated cells. Moreover, PCR analyses conducted on primary cultures, maintained for 2 weeks, confirmed a specific signature of A. viridis. In parallel, we evaluated the cell viability of these cultures by vital staining. Serial dilutions, led during 4 weeks, of re-suspended small rounded cells isolated using chemical dissociation allowed us to obtain a homogenous primary culture of A. viridis epithelial undifferentiated cells.

The maintenance and the propagation of this homogenous primary cell culture for several weeks provide suitable model for in vitro cnidarian studies and preliminary step for further investigations on symbiosis mechanisms.