

Characterization of cellular and molecular responses of *Actinia equina* (Linnaeus, 1758) MG Parisi, MR Trapani, A Damiano, G Benenati, N Parrinello, M Cammarata

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Sea anemones (Actinaria, Cnidaria) are benthic sessile species that depend of their toxic venom for survival, providing to defense and predation.

Pore-forming cytolytic hemolysins, sodium channel toxins and potassium channel toxins have been isolated and characterized from a great number of sea anemones. However little is known about toxins compositions and their cellular origin.

Aim of this research is to describe the cell types of internal cavity of *Actinia equina* and their biological activity and characterize, after purification, cytolytic and antimicrobial molecules from different tissues.

Extracts from mucus and tissues showed cytolytic activity toward rabbit and sheep erythrocytes and human chronic myelogenous leukemia cells K562.

A new lytic peptide of about 6 kDa from percoled body fluid and tentacles extract has been isolated by SEC-HPLC followed by mass spectrometry and identified as a neurotoxin specific for sodium channels (AeI) existing in the database.

Then, we showed clearly that the presence of cytolytins is not exclusive of nematocysts. A plaque-forming assay carried out with cell populations extracted from the percoled body fluid showed for the first time that anthozoan granulocytes are able to form plaque of lysis. We have separated the total population of free cells into three distinct discrete bands by discontinuous Percoll gradient, and we have identified different cell types. Cell lysate of each cellular band showed cytolytic activity towards different erythrocytes types and was inhibited by sphingomyelin.

Tentacles acid extracts were subjected to purification on SEP PAK C8 Vac column followed by several HPLC runs on C18. Among the several peaks, two showed biological activity towards *E. coli*, *S. aureus* and *C. albicans* and were pooled and lyophilized.

These first results on *A. equina* immunobiology lead us to explore the evolution of cnidarian immune response.