Out of the deep-sea to a land-based aquarium setting: investigating acclimatization processes in the hydrothermal vent mussel *Bathymodiolus azoricus*

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Marine molluscs represent suitable models for the study of the effect of environmental factors on physiological reactions. Deep-sea hydrothermal vents are considered as some of the most extreme environments in the world, yet the animals dwelling around the vent sites exhibit high productivity and therefore must deal with unusual levels of heavy metals, pH, temperature, CO2 and sulfide, in addition to environmental microbes. In an attempt to understand the physiological reactions of animals normally adapted to endure extreme conditions we set out to investigate acclimatization parameters during the mussel Bathymodiolus azoricus processes of adaptation to laboratory conditions. The DeadEnd colorimetric TUNEL system was used to detect apoptotic cells in gill tissues of mussels brought to surface and subsequently maintained in aquaria for periods up to 3 months. With this method no visible cells undergoing apoptosis were observed during our studies. In addition, the signal transduction protein kinases MAPK, p38 and JNK were detected by Western blotting in gill and hemocyte extracts, from same experimental animals. Their detection was variable during the course of acclimatization experiments, suggesting that signal transduction reactions are likely to occur as part of vent mussel's strategy to adapt to land-based aquarium conditions. We also monitored the gradual loss of endosymbiont bacteria in gill bacteriocytes through fluorescence insitu hybridization (FISH). In addition, mucopolysaccharide granules (proteoglycans) at the distal apex of gill filaments were also monitored over the course of acclimatization utilizing the Alcian blue-Periodic Acid Schiff staining method and by auto-fluorescence visualization. Taken together, the above parameters were proven successful in assessing vent mussel's adaptation to laboratory settings at atmospheric pressure and therefore, set the stage for the study of immune reactions in Bathymodiolus azoricus.

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