The effects of sustained high hydrostatic pressure on membrane lipid composition and homeoviscous acclimation within shallow-water benthic marine invertebrates

Brief Report – MASTS Small Grant Award

Shallow-water fauna have repeatedly colonised the deep sea following numerous climate-driven deep-sea dysoxic extinction events. Consequently, extant deep-sea fauna comprises ancient and more recent shallow-water lineages. The combination of low temperature and high hydrostatic pressure, physical factors characteristic of deep-sea environments, is thought to limit range extensions by shallow-water fauna into the deep sea. Acute experimental works support this notion. Low temperature and high hydrostatic pressure reduce cellular membrane function by significantly decreasing membrane fluidity; this may contribute to hyperbaric limitation of bathymetric ranges. Invertebrates can increase the level of unsaturation of the fatty acids comprising their lipid bilayers in response to decreased temperature, a process termed homeoviscous acclimation. There is also limited evidence to suggest that homeoviscous acclimation may occur in response to increased pressure, though most evidence is based on microbes.

Recently, shallow-water decapods have been maintained for several weeks at high hydrostatic pressure and low temperature; the longest continuous maintenance of shallow-water fauna under deep-sea conditions. The monitoring of behaviour during these experiments indicated acclimation to these deep-sea conditions. Subsequent experiments demonstrated increased tolerance to hydrostatic pressure following prolonged exposure to deep-sea conditions, indicating acclimation to these conditions. The aim of this project was to test whether increased tolerance of high hydrostatic pressure following prolonged exposure to high hydrostatic pressure and low temperature was a consequence of homeoviscous acclimation.

Figure 1. Photograph of the shallow-water shrimp, *Palaemonetes varians*, which was subjected to high hydrostatic pressure across a range of temperatures. The membrane lipid composition was subsequently analysed in this project to investigate homeoviscous acclimation. Photograph from http://en.wikipedia.org/wiki/File:PalaemonetesVariansCommonDitchShrimp3.JPG

The major objective of this project was, therefore, to analyse the membrane lipid composition of decapod crustaceans collected from costal environments (Figure 1) and subjected to prolonged exposure to deep-sea conditions to investigate whether changes in lipid composition (homeoviscous acclimation) occur in response to high hydrostatic pressure and low temperature. This would be the first time homoeviscous acclimation has been demonstrated within a shallow-water animal.

MASTS Small Grant Award supported Andrew Oliphant for a four weeks period whilst he visited Dr David Pond at the Scottish Association for Marine Science, Dunbeg. Here, David instructed Andrew in laboratory techniques and procedures for the analysis of membrane lipids. David provided access to laboratories, equipment, and consumables. During the visit, samples were processed to extract lipids, separate membrane lipids, and then run these membrane lipids through a Gas Chromatograph to give data on the composition of membrane lipids. This was done for 79 samples. The result of Andrew's time at SAMS was a data set on the membrane lipid compositions of decapods, which had been experimentally exposed to deep-sea conditions. Further, Andrew learned techniques for lipid analysis. The data set produced is now being statistically analysed and is in the process of being writing-up as a manuscript with the aim to publish in a peer-reviewed, international journal in 2014. Data on the relative composition of saturated and mono- and poly-unsaturated fatty acids within *Palaemonetes varians* after seven days at atmospheric pressure (0.1 MPa) and hydrostatic pressure equivalent of 1000m (10 MPa) and across three temperatures are shown in Figure 2.

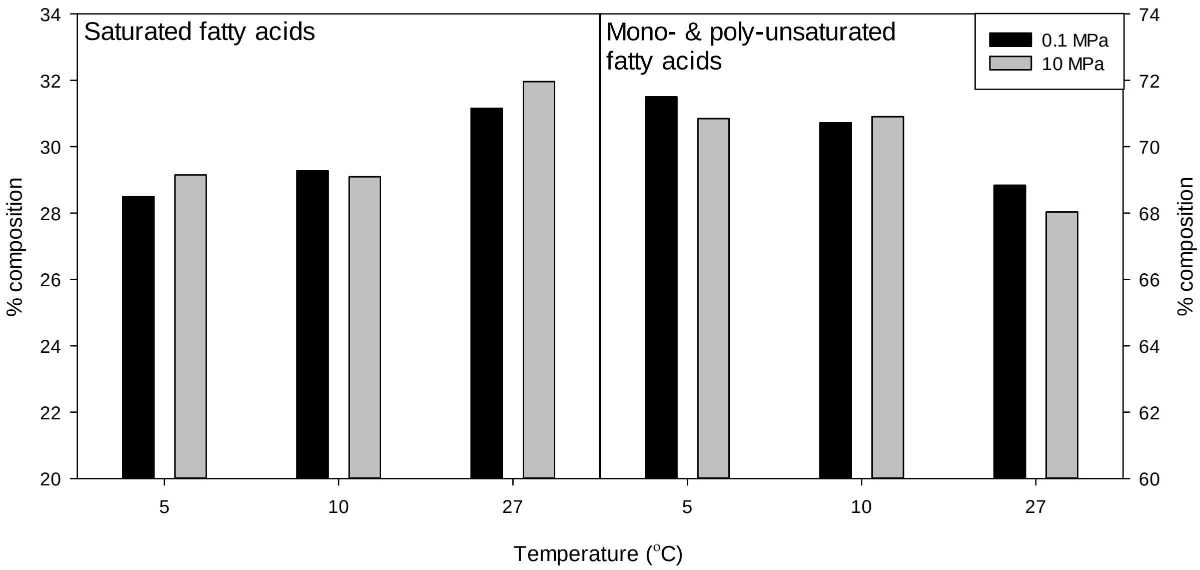

Figure 2. The relative composition of saturated and mono- and poly-unsaturated fatty acids within *Palaemonetes varians* cell membrane after 7 days under 0.1 MPa or 10 MPa at three different temperatures (5, 10, 27 °C)