

Introduction

During its lifetime an organism is subject to a range of environmental factors. The organism's ability to survive and adapt to change is an important one. Of all the factors in the marine environment temperature has one of the most striking effects on the functions and distribution of organisms (Schmidt-Nielson, 2002). Adaptation to temperature change is therefore very important. Adjustment to changes in temperature is termed 'temperature acclimation'. 'Temperature acclimation confers upon an organism the ability to stabilise its energy requirements in the face of alterations in habitat temperature' (Halcrow, 1963). Temperature differentials of as much as 10°C above or below a thermocline are common. Many planktonic organisms migrate over a considerable vertical range (Bainbridge, 1961; Vinogradov, 1970). Annual or seasonal range in temperature due to climatic change although gradual can also vary substantially especially in shallow or restricted waters (Schmidt-Nielson, 2002). The Menai Strait can vary by as much as 17°C (Jones *et al.*, 1982).

There are two ways that organisms regulate temperature. Homeotherms are organisms that regulate body temperature to constant level; poikilotherms are organisms whose body temperature conforms to that of the ambient environment. All sub-tidal marine invertebrates and most fish are poikilotherms. In poikilotherms an increase in temperature usually increases metabolic and behavioural activity (Levinton, 2001). Poikilotherms are able to compensate for changes in habitat conditions (Halcrow, 1963).

An organism adapted to suit winter freezing conditions may die if placed in seawater at say 20°C whereas no mortality occurs in organisms collected in summer transferred into water of the same temperature. This organism has adjusted physiologically between seasonal variations in water temperature. It has learnt to acclimatise. Changing environmental factors and studying responses can aid in studying acclimation (Levinton, 2001).

In this experiment we have captured *Temora longicornis* using medium plankton net from the pier in Menai Bridge. *T. longicornis* is a neritic copepod of the Phylum

Crustacea and is one of the most common zooplankton in the North Sea and adjacent waters (Harris and Pfaffenhöfer, 1976). *T. longicornis* is also common across the east and west side of the North Atlantic Ocean (Gill and Crisp, 1984) and the Indian Ocean. The wide distribution of *T. longicornis* is an indication of its wide thermal tolerance. The seawater temperature at the time of collection was 15°C. The sample captured was split into three groups. One third was kept at 5°C for 15 days, another third at 10°C for 15 days and the final third at 20°C also for 15 days. Over the succeeding three days limb beat rates of each group were measured over a range of temperatures from 5-25°C. We are testing the hypothesis that acclimation temperature has no effect on beat rate at various temperatures. We want to see if *T. longicornis* exhibits a degree of physiological adaptation to temperature.

Results

Figure 1 shows the increase in temperature for the three groups of *T. longicornis* kept at different temperatures for 15 days. The correlation coefficients range from 0.963 to 0.983 all with associated probabilities of <0.001 . This indicates a close linear fit and a good probability of always obtaining a good linear relationship.

The plot of residuals versus fits did not show any curve relationship. The data proved to be of approximately normal distribution with an Anderson Darling value of 0.616 and associated probability of 0.103 on the standardised residuals. The test for heterogeneity of variance showed no significant difference in variability between treatments with a test statistic of 0.57 and an associated probability of 0.753. Bartlett's test statistic was used because the data is approximately normal. The data fits the assumptions of the ANOVA test and therefore can be properly analysed using regression analyses.

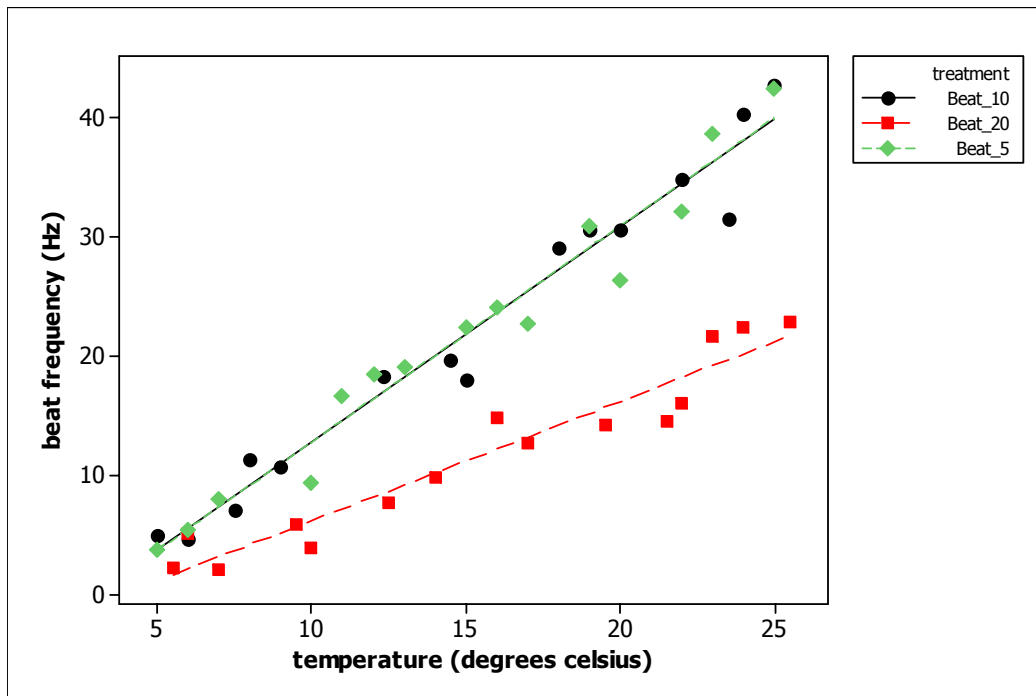


Figure 1 Beat rate of three groups of *T. longicornis* captured in seawater (temperature 15°C) recorded over a range of temperatures from 5-25°C over 3 days. Each group was initially kept at constant temperature for 15 days before the start of the experiment. One third of *T. longicornis* was kept at 5°C (Beat_5), another third at 10°C (Beat_10) and the final third at 20°C (Beat_20).

Table 1. The analysis of three groups of *T. longicornis* beat rate recorded over 3 days when exposed to a range of temperatures from 5-25°C after being initially kept at constant temperature of 5°C, 10°C or 20°C over preceding 15 days.

Table 1 (a) Slopes, intercepts (\pm their SE) from regression analysis of the increase in beat rate with increased temperature.

| Temperature | Slope (Hz °C ⁻¹) | Intercept (°C) |
|-------------|------------------------------|------------------|
| 5°C | 1.82 \pm 0.10 | -5.41 \pm 1.21 |
| 10°C | 1.81 \pm 0.09 | -5.36 \pm 1.20 |
| 20°C | 1.00 \pm 0.09 | -3.90 \pm 1.21 |

Table 1 (b) Analysis of variance table with temperature in degrees Celsius as covariate, Seq = sequential, Adj = adjusted for entry order into the model, DF = Degrees of freedom, SS = Sum of Squares. MS = mean of squares.

| Source | DF | Seq SS | Adj SS | Adj MS | F | P |
|-------------|----|--------|--------|--------|--------|--------|
| Treatment | 2 | 1031.4 | 3.4 | 1.7 | 0.33 | 0.719 |
| Temperature | 1 | 4459.9 | 4501.3 | 4501.3 | 876.30 | <0.001 |
| Interaction | 2 | 284.1 | 284.1 | 142.0 | 27.65 | <0.001 |
| Error | 39 | 200.3 | 200.3 | 5.1 | | |
| Total | 44 | 5975.7 | | | | |

Table 1 (c) Comparisons between beat rate regression slopes and intercepts and their respective group averages. Slope difference in Hz °C⁻¹, Standard Error of difference = 0.072 Hz °C⁻¹, intercept differences in °C, Standard Error of differences = 1.2 °C.

| Treatment | Average | Difference | T-value | Probability |
|-------------------|---------|------------|---------|-------------|
| Slopes | 1.54 | | | |
| 5°C | | 0.27 | 3.76 | <0.001 |
| 10°C | | 0.26 | 3.70 | 0.001 |
| 20°C | | -0.54 | -7.43 | <0.001 |
| Intercepts | -4.89 | | | |
| 5°C | | -0.52 | -0.43 | 0.670 |
| 10°C | | -0.47 | -0.39 | 0.697 |
| 20°C | | 0.99 | 0.82 | 0.420 |

Table 1 (a) shows that the temperature curves for *T. longicornis* kept at 5°C and 10°C remains the same. The two slopes for 5°C and 10°C differ by only 0.01 Hz per °C. The intercept value differs by only 0.05°C. The temperature curve for *T. longicornis* kept at 20°C is quite different from the curves for 5 and 10°C with a slope of 1.00 Hz per °C compared to 1.81 and 1.82 Hz per °C, an average difference of 0.8 Hz per °C. The intercept for *T. longicornis* kept at 20°C is -3.90 an average of 1.49°C different from the two intercepts for *T. longicornis* kept at 5°C and 10°C, which are -5.41 and -5.36 respectively.

The negative values for the intercepts are however extrapolated from the straight-line data. In reality you cannot have negative beat rates so the beat rate at 0°C would be zero. As the lowest temperature at which beat rate was measured in this experiment was 5°C it is not possible to make any valid conclusions of what happens to the frequencies below this temperature but the line probably curves to zero.

Table 1 (b) shows that both the temperature increase and interaction between the acclimation temperature and temperature increase have a significant effect in changing beat rate with probability values less than 0.05 (p-value = <0.001). The effect of temperature acclimation alone shows no significant effect on beat rate (probability value >0.05)

Table 1 (c) shows that the slopes of the beat rate / temperature curves for all three acclimation temperatures are significantly different from the average (probability value of <0.05). The difference in slopes for the beat rate temperature curve of *T. longicornis* kept at 5°C and 10°C are the same but are both significantly higher than the average. The slope for *T. longicornis* kept at 20°C is significantly lower than average. This slope is less steep than the average slope showing rotation of the slope indicating a significantly lower increase in limb beat rate as temperature increases than the *T. longicornis* kept at either 5°C or 10°C.

The lower portion of table 1 (c) shows that the intercepts for *T. longicornis* acclimatised at 5°C and 10°C are higher than average and the intercept for *T. longicornis* kept at 20°C is lower than average. There is however no significant difference between the intercepts and the average (probability value of >0.05).

Overall the results indicate that *T. longicornis* acclimatised at 5°C and 10°C respond very similarly to increase in temperature. *T. longicornis* acclimatised at 20°C suggests a gentler slope. Due to the significant changes in slopes between the beat rate / temperature curves at the different acclimation temperatures it can be said that the temperature curve for *T. longicornis* acclimatised at 20°C has rotated.

Discussion

Some changes in temp tolerance occur in response to natural climatic change. The change between summer & winter are an example of natural acclimatisation. Prolonged exposure of organisms to high or low temperatures within their thermal tolerance can result in compensatory changes. Adaptation is not restricted to change in lethal limits, other compensating mechanisms also exist (Schmidt-Nielson, 2002). Within limits a temperature increase accelerates most processes. Within the temperature range an animal can tolerate the rate of oxygen consumption often increases in a fairly regular manner with increasing temperature (Levinton, 2001).

Acclimatization in poikilotherms such as *T. longicornis* often includes adjustments at the cellular level. Cells may increase the production of certain enzymes, helping to compensate for the lowered activity of each enzyme molecule at temperatures that are not optimal. In other cases cells produce variants of enzymes that have the same function but different temperature optima. Membranes may also change the proportions of saturated lipids they contain, which helps keep membranes fluid at different temperatures. (Campbell and Reece, 2002).

Copepod activity is dependant upon the environmental temperature. In our data *T. longicornis* has a range of limb beat rates between 2 and 42 Hz. It is known that temperature affects both the physiology and ecology of zooplankton (Heinle *et al.*, 1969). Like all poikilotherms *T. longicornis* metabolic rate and behavioural activity tends to increase exponentially with temperature (Levinton, 2001). The shape of the beat rate / temperature curve can show a translocation, a rotation or a combination of these factors following thermal acclimation (Newell, 1973). If the beat rate / temperature curve has a significantly different intercept then a translocation has

occurred and the copepod has acclimated to the temperature change by raising or lowering their heat tolerance in response to acclimation. This is normally achieved by using the same or variants of the same enzymes adapted to a different optimum temperature (Newell, 1973) increased enzyme concentration, altered haemolymph pH or altered tissue osmolarity. If the beat rate/ temperature curve has rotated; shown by a significant change in the slope, then different enzymes have been produced to cope with activity at change in temperature. Rotation and translocation indicate both processes are occurring.

The results from table 1 show that the *T. longicornis* previously kept at 5 and 10°C showed the same acclimation rates and processes whereas the temperature curve slope rotation for *T. longicornis* kept at 20°C implies possible use of different enzyme systems as a result of acclimation to higher temperature.

T. longicornis acclimated to 5°C and 10°C the results in table 1 (c) indicate that no cellular level changes have been made; enzyme types and concentrations have probably remained the same. Results in table 1(c) for *T. longicornis* acclimated at 20°C suggests an increase in production of certain enzymes to compensate for lowered activity of other enzymes that are not within their optimal temperature range. This type of activity is often an indication of an organism being subject to short-term temperature changes such as diurnal fluctuations of tides rather than the more long term climatic changes in temperature experienced with seasonal changes which you would expect to result in translocation of the beat rate temperature curve (Newell, 1973).

Most biochemical and physiological processes are very sensitive to changes in body temperature. The rates of most enzyme-mediated reactions increase by a factor of 2-3 for every 10°C temperature increase. This is known as the Q_{10} effect (Schmidt-Nielson, 2001). The Q_{10} value is therefore represented as the slope in the results. The group of warm acclimated *T. longicornis* (20°C) has a lower slope therefore a lower Q_{10} than the *T. longicornis* acclimated at 5°C and 10°C. The rotation at 20°C is clockwise suggesting a decrease in Q_{10} value (Crisp and Gill, 1985). The Q_{10} at 5°C acclimation temperature is 3.36 compared to the Q_{10} value at 20°C which is 2.95 confirming the decrease in Q_{10} when the acclimation temperature increases.

Crisp and Gill (1985) showed that as acclimation temperature increased from 5°C to 15°C there was a marked translocation in the limb beat rate / temperature curve to the right along the temperature axis. However when the acclimated curves of winter and summer populations were considered together the winter curves showed a lower Q_{10} (clockwise rotation of the curve) as well as a translocation to the left along the temperature axis. A clockwise rotation and a shift of the beat rate / temperature curve to the left appear to be most common in cold acclimation (Halcrow, 1963). Whereas the acclimated curve of summer animals acclimated to warmer water shifts to the right (Halcrow, 1963).

T. longicornis have the ability to acclimatise to temperature changes. Survival of organisms especially the more passive forms depends on their tolerance to daily and seasonal fluctuations in temperature (Bradley, 1975)

References

Scmidt-Nielson, K. 2002. Animal Physiology. Adaptation and environment. Pages 218 – 297.

Vinogradov, M.E. 1970. Vertical distribution of the oceanic zooplankton. Acad. Sci. U.S.S.R Inst. Oceanography. Israel programme for Scientific translations, Israel., 339.

Gill, C.W. Crisp, D.J. 1985. The effect of size and temperature on the frequency of limb beat of *Temora Longicornis*. J. Exp. Mar. Biol. Ecol., 86: 185-196.

Campbell, N.A. Reece, J.B. 2002. Biology. Benjamin Cummings, San Francisco. 926-935.