METABOLIC ADAPTATIONS (AND THE LACK THEREOF) TO OXYGEN LIMITATION IN A BURROW-DWELLING CRUSTACEAN

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ABSTRACT

The mantis shrimp Hemisquilla californiensis routinely encounters hypoxia or anoxia in its burrows, which it often enters and caps for hours at a time. Given this environment the species should exhibit adaptations for living in low oxygen conditions that are not generally observed in crustaceans. In this study I compared the aerobic metabolism of H. californiensis with that of the shrimp Pandalus platyceros, which lives on the aerobic surface of the sediment rather than in hypoxic burrows. As expected, P. platyceros was a poor oxyregulator with a moderate anaerobic survival time of a few hours. Aerobic metabolism at 20-30 mm Hg O_2 was depressed by a factor of 2.5 from metabolism at 110-130 mm Hg. Unexpectedly, H. californiensis is also a poor oxyregulator. Aerobic metabolism of H. californiensis at 20-30 mm Hg O_2 was depressed 2.3-fold from that at 130-140 mm Hg. H. californiensis, therefore, which routinely encounters hypoxic conditions, is little better at oxyregulating than is P. platyceros, which inhabits a nearly normoxic environment. However, H. californiensis has dramatically greater anaerobic survival of 24-48 hours or more.

Figure 1: Hemisquilla californiensis. Photo by Ruwan Randeniya

INTRODUCTION

Fine, organic-rich marine sediments are typically anoxic below the surface. Animals that live in such sediments typically have one of several strategies for surviving the low oxygen conditions. Many have alternate metabolic pathways for anaerobic metabolism which increase the amount of energy available anaerobically and/or reduce the buildup of toxic by-products (Hochachka and Somero, 2002). Others build and ventilate burrows, which serves to bring oxygenated water down to them within the sediment.

Crustaceans in general have not been found to utilize alternate anaerobic metabolic pathways to any substantial extent (Zebe, 1991). Anaerobiosis in crustaceans nearly always results in the buildup of lactate as the primary by-product. As a result, with the possible exception of some barnacles, anaerobic capacity in crustaceans is quite limited (Zebe, 1991). Few crustacean species live within anaerobic sediment. Those which do, such as some amphipods (Gamble, 1970; Hervant et al., 1998) and thalassinideans (Forster and Graf, 1995; Rowden and Jones, 1995; Stamhuis et al., 1997), tend to have a similar suite of adaptations which serve to enhance their ability to extract oxygen from the water. Most live in burrows, some of which are lined with mucus which serves to isolate the burrow water from the oxygen demand of microorganisms in the sediment. Burrows tend to have multiple openings, which facilitates circulation of water through the burrow (Fig 2A). The residents often station themselves at or near the burrow entrance and beat their large pleopods for ventilation. Burrow-living crustaceans are usually strong oxyregulators, and can maintain nearly normal rates of aerobic metabolism even in hypoxic conditions (Figure 2B).

Hemisquilla californiensis (Figure 1) is a large burrow-dwelling stomatopod which does not seem to fit these generalizations well. This species lives in blind-ended burrows in silty sand (Basch and Engle, 1989). It often caps the burrow entrance for many hours at a time. Preliminary observations suggest that the shrimp maintains a continuous high level of activity in the burrow, even while remaining deep within the burrow and distant from the entrance for hours at a time.

In this experiment I compared the aerobic metabolism of Hemisquilla californiensis with that of the spot shrimp Pandalus platyceros (Figure 3B), which is of a similar size and lives on soft bottoms at a similar range of depths, but lives epibenthically instead of within burrows and thus is not likely to routinely encounter low oxygen conditions. I hypothesized that Hemisquilla would much better oxyregulating capacity than did Pandalus and that neither species would be able to survive any extended periods of anoxia.

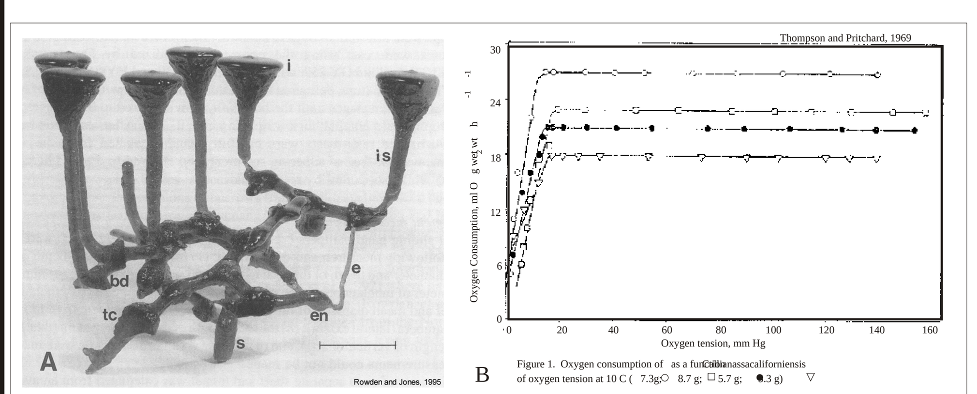

Figure 2: Typical adaptations seen in burrow-dwelling crustaceans. A. Multiple burrow entrances provide flow-through ventilation of the burrow. B. Burrow-dwelling species are typically strong oxyregulators with very low critical oxygen pressure (P_c). This facilitates maintenance of aerobic metabolism while in the hypoxic conditions of the burrow. (A from Rowden and Jones, 1995; B from Thompson and Pritchard, 1969)

MATERIALS AND METHODS

10 Pandalus platyceros ranging from 21 to 94 g, and 7 Hemisquilla californiensis ranging from 56 to 149 g were placed individually in a temperature-controlled respirometer (Figure 3). Temperature in the respirometers was held at the temperature the animals were acclimated to (11C for Pandalus and 14C for Hemisquilla), and reflected the ambient temperatures where the animals were captured. After a short acclimation period the respirometer was sealed and the subject's aerobic metabolism was monitored by oxygen electrode. Water in the respirometers was stirred by magnetic stirbar or by peristaltic pump. Aerobic metabolism was measured continuously as oxygen level decreased from near air saturation to zero, which normally took about 4-8 hours. Some experiments were ended when the animals had reached severe hypoxia, defined for this experiment as 10 mm Hg O_2 (or 6% saturation, air saturation is 157 mm Hg). Other experiments were allowed to extend for up to 48h beyond this point in complete anoxia, while the animals were monitored for behavior and activity. The rate of the subject's aerobic metabolism in each interval of 10 mm Hg O_2, and in each interval of 2 mm Hg in the range of 0-30 mm Hg, was calculated.

The extent to which each subject oxyregulated was assessed after plotting the rate of aerobic metabolism (MO_2, micromoles O_2 g wet wt-1 h-1) as a function of the partial pressure of oxygen in the water (pO_2, mm Hg) (Figure 4). The subjects were determined to be oxyregulating if the plot of MO_2 as a function of PO_2 showed an inflection, with MO_2 remaining steady or dropping slowly at first until a critical oxygen pressure (P_c) was reached, after which MO_2 dropped rapidly toward zero. Pure oxyregulation occurred if MO_2 remained steady or even increased over a broad range of pO_2 above Pc. Partial oxyregulation occurred if MO_2 declined slowly with decreasing pO_2, but not as rapidly as the decline in pO_2. Either of these conditions could be detected by the fact that the full data set could be described by two distinct linear regressions covering the range above and below Pc with lower residual error than from a single regression of the entire data set (Yeager and Ultsch, 1989). The subjects were determined to be oxyconforming if MO_2 decreased monotonically toward zero as pO_2 decreased, and a single regression described the relationship with less residual error than two regressions could.

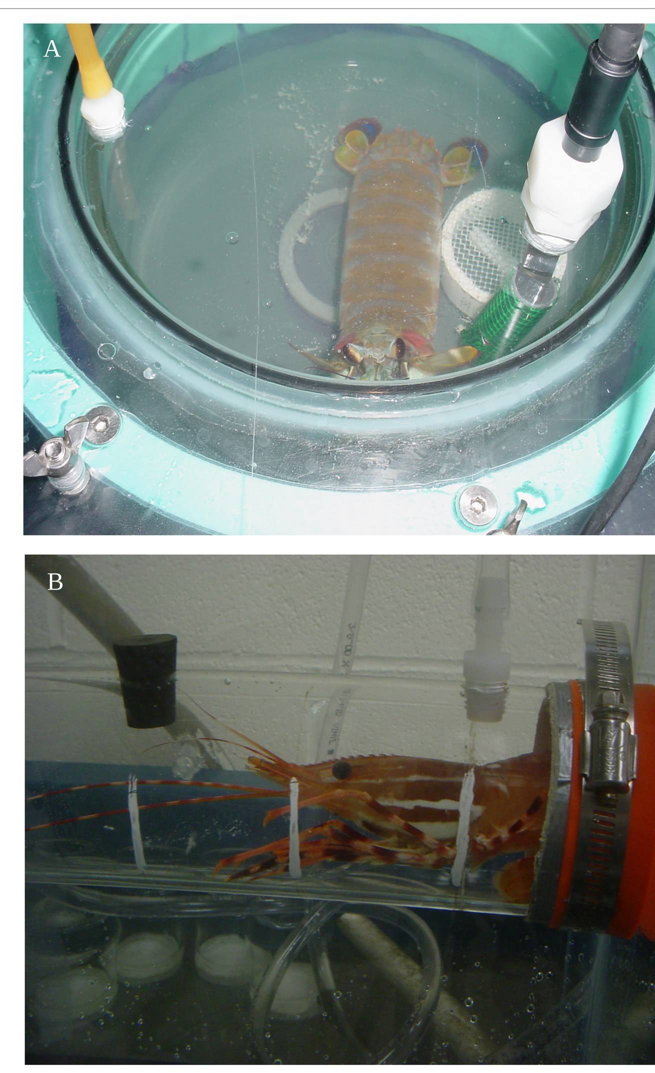

Figure 3. The sealed, temperature-controlled respirometers used for this experiment. A. Freestanding water-jacketed type (Hemisquilla inside), mixed by a magnetic stirbar. B. Tube type immersed in a temperature-controlled water bath (Pandalus inside). Water is mixed by a peristaltic pump, which also pumps water past the oxygen electrode which is outside the chamber.

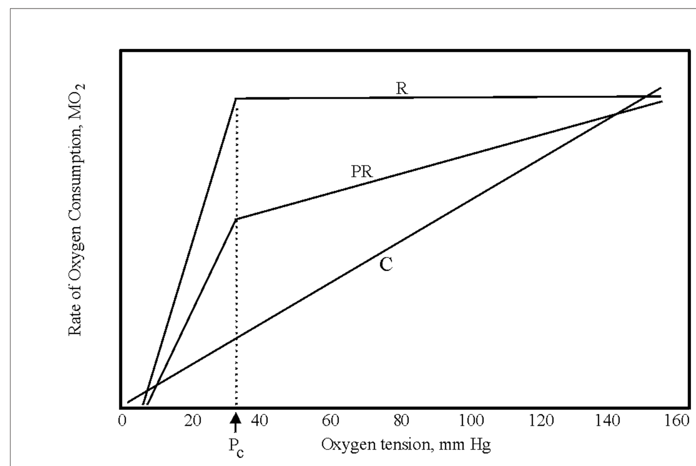

Figure 4: Possible patterns of aerobic metabolism. R. Pure oxyregulation. Rate of aerobic metabolism MO_2 is independent of oxygen partial pressure pO_2 over a range of pO_2 above the critical oxygen pressure P_c, then rapidly drops toward zero. PR. Partial or weak oxyregulation. MO_2 declines gradually with pO_2 above P_c then drops more sharply below P_c. C. Oxyconformity. MO_2 varies directly with oxygen partial pressure over the entire range of oxygen pressures encountered.

RESULTS

As expected, Pandalus platyceros is an oxyconformer or only partial oxyregulator. Most individuals only partially regulated within the range above their P_c (Figure 5A). Others had a pattern indistinguishable from oxyconformity (Figure 5B), while one individual was able to regulate down to 50 mm Hg with only a slight decrease in MO_2 (Figure 5C). Even in the partial oxyregulators, however, P_c was high–averaging around 50 mm Hg (Table 1). Mean rate of aerobic metabolism in moderate hypoxia of 20-30 mm Hg was only 1/3 the rate observed at pO_2 > 100 mm Hg. No Pandalus survived long periods of anoxia, with 1.4h being the longest period which an animal, which was near death on removal, survived.

Unexpectedly, Hemisquilla californiensis is also an oxyconformer or only a partial oxyregulator. Several Hemisquilla showed at least weak oxyregulation (Figure 6A). The regulation pattern in others was so weak that it was difficult to distinguish it from oxyconformity (Figure 6B). One individual was able to regulate down to around 20 mm Hg O_2 with only a slight decrease in MO_2 (Figure 6C). The oxyregulating Hemisquilla also had a high P_c, averaging 46 mm Hg (Table 1). Mean rate of aerobic metabolism at 20-30 mm Hg O_2 was less than half the rate observed at pO_2 > 100 mm Hg. However, Hemisquilla survival of anoxia was dramatically better than that of Pandalus. Several individuals survived over 24 and even up to 48 h of anoxia. The largest Hemisquilla individual survived anoxia the longest, but survival of up to 47 hours was observed in even the smallest individual. One individual died after less than 24h of anoxia.

Table 1: Metabolic characteristics of Pandalus platyceros (Caridea) and Hemisquilla californiensis (Stomatopoda). R = oxyregulator. PR = partial oxyregulator. C = oxyconformer. MO_2 = rate of aerobic metabolism, micromoles O_2 g wet wt h^{-1} $^{-1}$

Species	number tested	Percent of group			Mean MO_2			P_c mmHg	Maximum anaerobic survival
		R	PR	C	>100 mm Hg	20-30 mm Hg	0-10 mm Hg		
Pandalus	10	10	60	20	4.2	1.4	0.20	50	1.4h
Hemisquilla	7	14	71	14	2.5	1.2	0.41	46	>48h

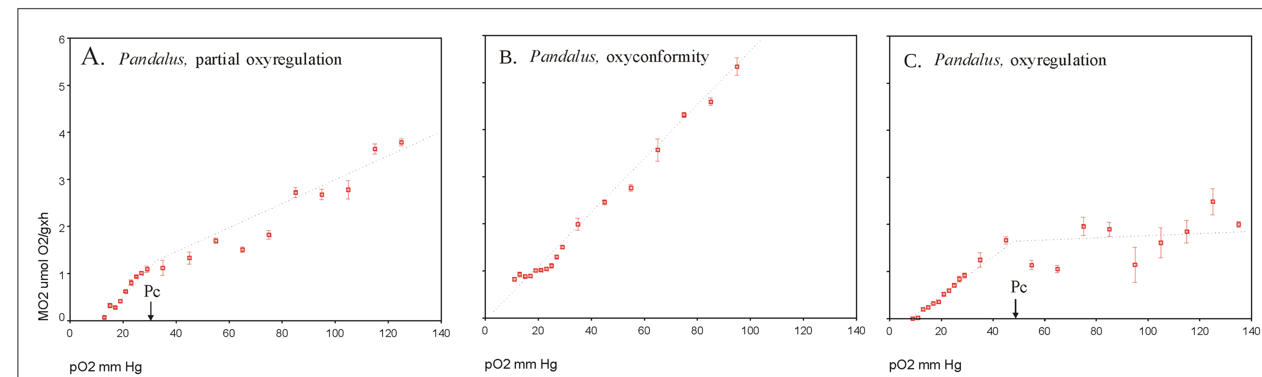

Figure 5: Patterns of aerobic metabolism observed in Pandalus platyceros. A. Partial oxyregulation (observed in most individuals). B. Near complete oxyconformity (observed in several individuals). C. Possible oxyregulation down to about 50 mm Hg O_2 (observed in one individual).

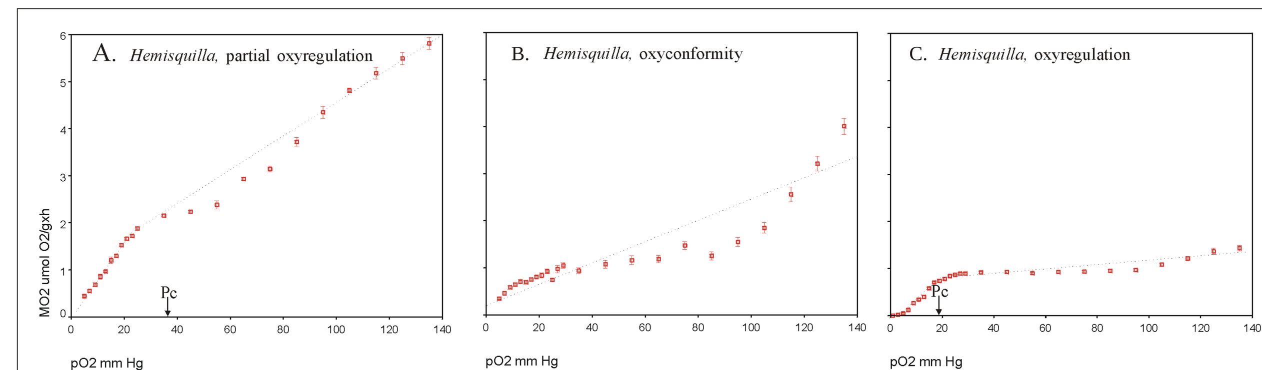

Figure 6: Patterns of aerobic metabolism observed in Hemisquilla californiensis. A. Partial oxyregulation (observed in most individuals). B. Very weak regulation or oxyconformity (observed in several individuals). C. Possible oxyregulation down to about 20 mm Hg O_2 (observed in one individual).

DISCUSSION

Unexpectedly, Hemisquilla does not follow the metabolic pattern which has been reported for other species of burrow-dwelling crustaceans. Its mean P_c of 46 mm Hg is surprisingly high given the low oxygen levels it must encounter in the burrow. This species must spend a large portion of its time under oxygen conditions that sharply limit its aerobic metabolism. Even at very high oxygen levels the species does only a partial job of oxyregulation, suggesting that oxygen level may restrict this species' aerobic scope for activity under all but completely air saturated conditions. Indeed, the pattern of aerobic metabolism of this species is much more similar to that of epibenthic crustacean species such as Pandalus than to other burrow-dwelling crustaceans.

Differences in anatomy may partially explain why this species is not a strong oxyregulator as are the other burrow dwellers such as the thalassinidean shrimp. The gills of burrow-dwelling shrimp such as Callianassa and Upogebia are enclosed within a thoracic gill chamber and ventilated by a scaphognathite. The gills of mantis shrimp such as Hemisquilla, on the other hand, are fully exposed threadlike structures between the pleopods (Figure 7). The beating of the pleopods no doubt circulates water past the gills, but the lack of a gill chamber may limit Hemisquilla's ability to manipulate water flow and the O_2 extraction efficiency of the respiratory system.

The very long anaerobic survival time of Hemisquilla, on the other hand, puts it squarely among the best anaerobic survival abilities of crustaceans and clearly distinguish it from epibenthic species such as Pandalus. Indeed, 48h survival under truly anaerobic conditions is among the most extreme anaerobic capacities reported for crustaceans. We are presently investigating the biochemical mechanisms by which this species achieves this metabolic feat (McFadden and Cowles, 2004).

SUMMARY

1. The burrow-dwelling stomatopod Hemisquilla californiensis is a poor oxyregulator and when regulating has a high critical oxygen pressure. In this respect it is little different from Pandalus platyceros, an epibenthic shrimp which does not routinely encounter hypoxia. Hemisquilla is likely to be limited in aerobic metabolic scope under many of the hypoxic conditions which it routinely encounters.

2. In contrast to Pandalus' low tolerance, Hemisquilla californiensis has very extensive anaerobic capacity, ranking among the highest measured for any crustacean. This is likely to be adaptive for a burrow-dwelling species.

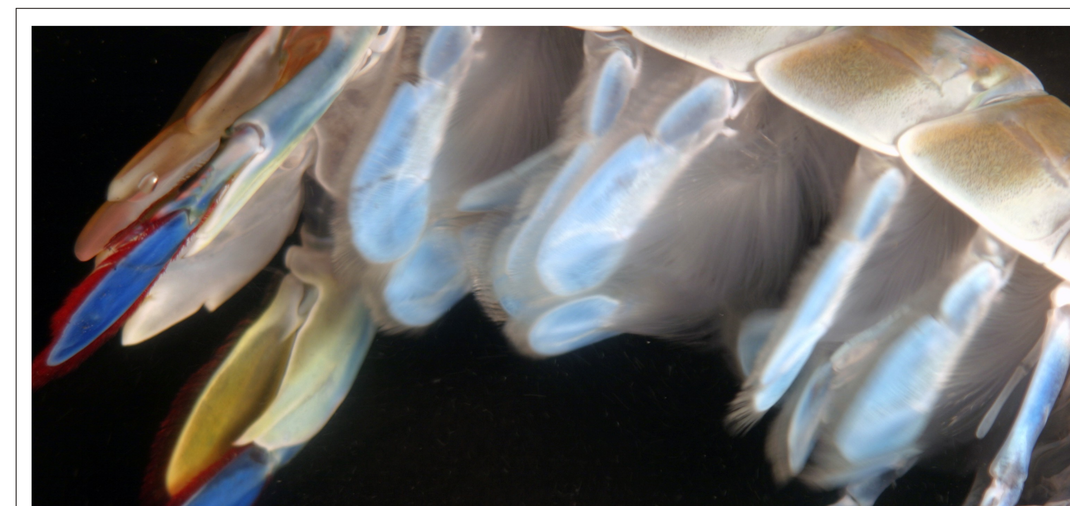

Figure 7: The threadlike abdominal gills of Hemisquilla californiensis, seen between the blue-tinted pleopods. The pleopods beat rhythmically, keeping water circulating past the gills. Note that the gills are fully exposed rather than enclosed within a chamber. Photo by Ruwan Randeniya

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