ENERGETICS OF THE EXERCISING WHARF CRAB SESARMA CINEREUM[1]

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Crabs (1.9 g) were exercised for up to 60 min on a miniature treadmill respirometer. O_2 consumption ($\dot{V}O_2$) at all velocities was low, a maximum of only 1.5 times the resting rate. $\dot{V}O_2$ was not related to the velocity of running. Five weeks of treadmill training produced a minor rise in $\dot{V}O_2$ at the maximum sustainable velocity, but the highest $\dot{V}O_2$ was still only two times the resting $\dot{V}O_2$. Crabs exercising in 100% O_2 significantly increased their endurance capacity, indicating that this crab's O_2 conductance system is severely limited. Anaerobic metabolism with lactate as an end product supplemented aerobic energy production to a minor extent. Whole-body lactate (WBL) increased at a constant but small rate (0.10 $mg \cdot g^{-1} \cdot min^{-1}$) when the crab ran at the highest velocity (0.14 $km \cdot h^{-1}$). Wharf crabs fatigued rapidly when run in a 100% N_2 atmosphere, indicating they do not have unusual anaerobic capacities or unusually high levels of O_2 stores or phosphagen reserves to compensate for the very low $\dot{V}O_2$. Hence, all data suggest that *Sesarma cinereum* has a uniquely reduced metabolic rate during exercise.

INTRODUCTION

The wharf crab *Sesarma cinereum* is a semiterrestrial decapod belonging to the family Grapsidae. *Sesarma cinereum* occupies the upper fringe of the littoral zone. During the daylight *S. cinereum* hides near wharves under stones and debris and in crevices. At night, individuals have been observed wandering the beach and congregating in moist areas to feed (Seiple 1981). *Sesarma* species are herbivorous. The more terrestrial *Sesarma* species, such as *S. cinereum,* do not construct burrows (Seiple 1979). As a result, these crabs are mobile, not residing in any one location. They defend only temporary refuges, and it appears that reproductive success depends more on the time spent searching for a mate than holding a resource such as a burrow (Seiple and Salmon 1982). In spite of the fact that locomotion is important in *S. cinereum*'s behavioral repertoire, no evaluation of its energetic cost has been attempted. Actually, very little information exists on the energetics of locomotion in crabs (Herreid 1981). Two different energetic responses have been found in the species examined thus far. Swift-running ghost crabs, *Ocypode gaudichaudii* and *O. quadrata,* have shown a remarkable aerobic capacity (7–12-fold elevations over resting $\dot{V}O_2$) and a considerable ability to sustain treadmill locomotion (Full and Herreid 1983; Full 1984). In contrast, a number of other land crabs have been shown to consume oxygen at relatively low rates in response to exercise; oxygen uptake increased sluggishly three to six times resting levels (Herreid, Lee, and Shah 1979; Wood and Randall 1981*a,* 1981*b;* Herreid, O'Mahoney, and Full 1983; Full and Herreid 1984). These species require significant amounts of energy derived from anaerobic fermentation to sustain even slow rates of locomotion.

The present study examines both the endurance capacity and the energetic response during treadmill locomotion in the wharf crab *S. cinereum.* The effects of exercise intensity, duration, and training were determined so that the crab's energetic capacity could be rigorously defined. In each case, oxygen consumption ($\dot{V}O_2$) was determined and compared with resting $\dot{V}O_2$. Whole-body L-lactate levels (WBL)

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served as an indicator of anaerobic metabolism. The results suggest that the response of *S. cinereum* to exercise represents a third type of energetic pattern. Both $\dot{V}O_2$ and lactate production showed only modest elevations during exercise when compared with other crab species tested at similar rates of locomotion.

MATERIAL AND METHODS

ANIMALS

Sesarma cinereum (Bosc) were collected on beaches near the Duke University Marine Laboratory in June of 1982. In our laboratory at Buffalo, New York, an environmental chamber, set on a 12L:12D cycle, was used to house the animals. Crabs were kept in large aquaria at 24 C with sand, sea grass, and access to 75% seawater. Tetramin fish food was added to the aquaria twice a week. Experiments began within 1 wk of the crabs' arrival, and all experiments were completed within 4 wk. Both intermolt males and females were used. Females carrying eggs were avoided whenever possible. The mean mass of the animals used in the oxygen consumption experiments was 1.92 ± 0.11 g (SE) and for the whole-body lactate studies was 1.79 ± 0.09 g (SE).

ENDURANCE CAPACITY

Crabs were run to fatigue in a miniature, motor-driven, treadmill respirometer after a 10-min rest period. When fatigue occurred, the endurance time was recorded. The animals were considered fatigued when they did not maintain pace with the treadmill, dragged their abdomens, or did not respond to three successive prodding attempts. Running velocities ranged from 0.05 to 0.40 $km \cdot h^{-1}$. At these velocities, all animals adjusted their gait immediately to match the speed of the treadmill belt. Any experiment in which an animal struggled or ran erratically was discarded.

In a separate series of trials, another group of crabs was tested for endurance capacity during exposure to different oxygen concentrations, 100% and 0% O_2. The maintenance of the high and low O_2 concentrations required that the Plexiglas cover be placed on the treadmill respirometer. Consequently, a control group (21% O_2, room air) was tested because the animals could not be prodded as effectively as in the experiments described previously, where no cover was necessary. All crabs were given a 10-min rest period before exercise at 0.10, 0.15, 0.20, or 0.25 $km \cdot h^{-1}$. Crabs belonging to one experimental group were exposed to 100% O_2 for the 10-min rest period. This was accomplished by adding 100% O_2 to the chamber initially and for every 3 min thereafter. Pure O_2 was then added every 2 min during exercise until fatigue. In the other experimental group, N_2 was added rapidly to flush the chamber after the conclusion of a 10-min rest period in room air. After this initial addition, N_2 was added continuously during running until the crab showed fatigue. No animal was tested more than once on a single day.

AEROBIC METABOLISM

Oxygen consumption measurements.—*Sesarma* were exercised on a treadmill enclosed in a Lucite chamber, while $\dot{V}O_2$ was determined by open-flow respirometry. All measurements were made at 24 C and 50% relative humidity unless otherwise specified. O_2 concentration was monitored by an oxygen analyzer (Applied Electrochemistry) that was interfaced with an integrating chart recorder (Linear Instruments model 282). Mass-specific instantaneous $\dot{V}O_2$ was derived by a method used by Bartholomew, Vleck, and Vleck (1981), given the following parameters: (1) the fractional O_2 concentration entering and leaving the respiratory chamber, (2) the flow rate of air (35 $cm^3 \cdot min^{-1}$) passing through the respirometer, (3) the "washout" characteristics of the respiratory chamber, and (4) the animal's mass (see Herreid, Prawel, and Full 1981*b;* Full and Herreid 1983, 1984). The mean respiratory exchange ratio (R) was taken to be 1.0 in the above calculation. The fractional error in $\dot{V}O_2$ is ±9% if R was taken to be 1.0 but is actually 0.50 or 1.50 (Withers 1977). For each trial, instantaneous $\dot{V}O_2$ values were calculated at 1-min intervals. A curve fitting technique, interpolation using the cubic spline method, was executed on an Apple II+ computer (Warme 1981). This method fits a cubic polynomial through

each of four successive points. All $\dot{V}O_2$ values were corrected to STPD conditions.

Sustained exercise protocol.—All crabs were given a 60-min rest period on the treadmill prior to exercise. After the rest period the animals were exercised for 20 or 60 min at a given velocity. Three velocities were selected for the 20-min exercise bouts; 0.06, 0.10, and 0.13 $km \cdot h^{-1}$. The runs lasting 60 min were conducted at 0.06 $km \cdot h^{-1}$. A 60-min recovery period followed each bout of exercise. $\dot{V}O_2$ was determined continuously during rest, run, and recovery. A similar protocol, with respect to time, was used in a 140-min control experiment. Following 60 min of rest, control animals remained in the chamber, unexercised, for 80 additional min. No animal was tested more than once on any given day.

In a separate set of trials, five crabs (1.22 ± 0.14 g SE) were run after a 60-min rest period at 0.13 $km \cdot h^{-1}$ for 20 min with the relative humidity (RH) adjusted to 99%–100%. The RH was increased by drawing air into the treadmill from a chamber partially filled with water and containing damp paper towels. The percent saturation of the air leaving the treadmill was determined by measuring the increase in weight of a water absorbent (Drierite). This amount of trapped water was then compared with the expected amount contained in saturated air at that temperature.

Maximal $\dot{V}O_2$ protocol.—Crabs were forced to run progressively faster while $\dot{V}O_2$ was monitored. Experiments were preceded by a 60-min rest period. Exercise began at 0.14 $km \cdot h^{-1}$ and was sustained for 5 min. The treadmill velocity was then increased by increments of 0.014 $km \cdot h^{-1}$ at 3-min intervals over the next 9 min.

ANAEROBIC METABOLISM

Whole-body lactate preparation.—Whole-body lactate (WBL) content of *Sesarma* was determined at specified time intervals during sustained exercise. WBL content was determined by the procedure described by Full and Herreid (1984). At the conclusion of an exercise bout or control experiment, the animal was removed from the treadmill and frozen in liquid nitrogen. The time required for this process was less than 2 s. Immediately on freezing the animal was pulverized in a mortar precooled with liquid nitrogen. Subsequently, the tissue powder was placed in a 0.6 N perchloric acid solution whose volume equaled five times the animal's mass. The mixture was then homogenized and kept on ice for 30 min. After this period, the homogenate was centrifuged and the supernatant was filtered and stored in a refrigerator for less than 2 days before analysis.

Lactate analysis.—The quantitative determination of L-lactate in the supernatant was accomplished by a specific spectrophotometric assay (Sigma diagnostic kit no. 826-UV). The glycine-hydrazine buffer (stock no. 826-3; pH = 9.2) was selected for use. The buffer was modified by adding EDTA to produce a 12-mM concentration and by introducing concentrated HCL dropwise until a pH of 9.0 was reached. The modification in buffer was necessary to remove the observed drift in spectrophotometric readings produced by heavy metal ions (Engel and Jones 1978; Graham et al. 1983). Assay performance characteristics have been described in detail elsewhere (Full and Herreid 1984).

Protocol.—Crabs were allowed a 60-min rest period on the treadmill before exercise. For the sustained exercise experiments, animals were run at one of three velocities (0.06, 0.10, and 0.14 $km \cdot h^{-1}$) for a given duration ranging from 2 to 20 min. Crabs run at 0.14 $km \cdot h^{-1}$ were removed for analysis at 2-min intervals, while those run at the medium and low velocities were analyzed at 10 and 20 min. After the exercise bout, the crab was rapidly removed from the treadmill and prepared for WBL analysis. During the recovery WBL content was measured at specified intervals (5, 15, and 45 min) after 20 min of exercise. Crabs were exercised at the same velocities used in the experiments focusing on the exercise period.

WBL content was determined in control animals (1) immediately after removal from holding tank, (2) after a 60-min rest period on the treadmill, and (3) after 20 min of rest in addition to the initial 60-min rest period. Any experiment in which a crab struggled was aborted.

TRAINING

Forty crabs were trained for endurance running over a 5-wk period. During the entire period, this group of crabs was kept under the same laboratory conditions as the untrained animals. All animals were trained simultaneously on a large treadmill (Quinton Instruments 1871D). Each crab was exercised in one of 40 separate cubicles (3.8 × 7.6 cm) formed by a custom-made Lucite frame which was fastened a few millimeters above the belt. The observer could check the progress of the crabs by a large mirror that was suspended over the training apparatus. Periodically crabs required prodding. This was accomplished through the frame's open top by the use of a long wire. Most animals ran well with only brief lapses of inactivity or struggling. Animals that either lost a leg or were injured during training were immediately removed from the experiment.

Each crab was trained 5 days a week for the 5-wk period. During the first 3 wk of training, animals were exercised at 0.06 $km \cdot h^{-1}$. The duration of each training bout was 30 min for the first week and was increased to 60 min for the next 2 wk. Crabs were run for 30 min at 0.10 $km \cdot h^{-1}$ for the last 2 wk of training (weeks 4 and 5).

At the conclusion of the training period, the crabs were randomly assigned to two groups so that endurance capacity and $\dot{V}O_2$ response could be evaluated. Endurance was tested as described previously. $\dot{V}O_2$ measurements were conducted during a 60-min rest, a 20-min run at the high velocity (0.13 $km \cdot h^{-1}$), and a 60-min recovery period. All crabs were subsequently used in WBL experiments. WBL was determined at intervals of 5, 10, 15, and 20 min during a run at 0.14 $km \cdot h^{-1}$.

RESULTS

ENDURANCE CAPACITY

The grapsid crab *Sesarma cinereum* ran well on the treadmill system but at times used different running styles. It abandoned the normal sideways locomotion and ran facing forward. The pattern of forward locomotion was primarily used at medium and low velocities (0.05–0.13 $km \cdot h^{-1}$). During sideways locomotion crabs have been shown to switch or reverse the set of legs that lead (Full and Herreid 1984). Forward running was followed by an eventual switching of the leading set of legs and may be linked to preventing fatigue of particular muscles involved in locomotion. Sideways running was adopted almost exclusively at the higher exercise velocities (0.14–0.40 $km \cdot h^{-1}$).

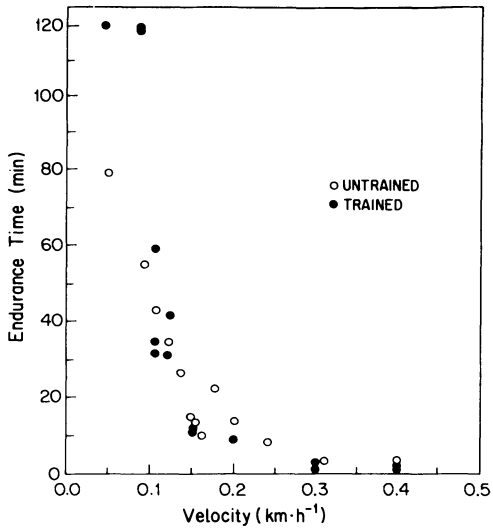

FIG. 1.—Endurance time of crabs running on a treadmill as a function of velocity. Each data point is a separate trial. Open circles represent untrained crabs, while closed circles represent trained animals. Three trained animals ran for 120 min without fatiguing and are plotted at the top of the graph at the appropriate velocity.

Figure 1 shows the relationship between endurance time and velocity. Crabs running at velocities below 0.15 $km \cdot h^{-1}$ could sustain exercise well over 20 min, while above this velocity, fatigue was observed in 3–20 min. At velocities slower than 0.06 $km \cdot h^{-1}$, crabs tended to wander about the chamber. Crabs trained for 5 wk showed a similar pattern. However, at very low velocities (less than 0.10 $km \cdot h^{-1}$), trained crabs appeared to exhibit greater endurance; three animals ran for 2 h without fatiguing.

Table 1 shows the changes in endurance capacity when *S. cinereum* was exposed to pure oxygen and nitrogen while running at a range of speeds. Both factors, velocity and O_2 concentration, were judged to have a significant effect on endurance when a

TABLE 1

ENDURANCE CAPACITY OF *Sesarma cinereum* IN ROOM AIR, 0% OXYGEN, AND 100% OXYGEN AS A FUNCTION OF THE VELOCITY OF LOCOMOTION

VELOCITY ($km \cdot h^{-1}$)	ENDURANCE TIME (min)		
	Room Air 21% O_2	Nitrogen 0% O_2	Oxygen 100% O_2
.10	14.6 ± 2.3	5.2 ± .5	91.4 ± 14.6
.15	6.4 ± .6	3.2 ± .3	9.5 ± 1.1
.20	3.9 ± .2	2.6 ± .2	7.3 ± .6
.25	3.4 ± .7	2.0 ± .3	4.3 ± .3

NOTE.—Each value represents the mean ± 1 SE for seven animals.

two-way ANOVA was conducted. As the velocity of running increased, endurance time decreased for all conditions ($F = 42.5$, $P < .001$). When the O_2 concentration in air was increased, endurance time showed a significant increase ($F = 38.7$, $P < .001$). A significant interaction between velocity and O_2 concentration was also present ($F = 27.6$, $P < .001$).

OXYGEN CONSUMPTION

The $\dot{V}O_2$ of 14 *Sesarma* was determined for the last 15 min of the 60-min resting period. During the rest period the animal's movement was carefully recorded at 15-min intervals. Crabs initially explored the chamber, but no activity was observed after 45 min of rest. The average resting $\dot{V}O_2$ was 0.043 ml $O_2 \cdot g^{-1} \cdot h^{-1} \pm 0.002$ (SE) for 28 experiments.

Sustained exercise.—$\dot{V}O_2$ of five animals running at each of the three velocities showed only a modest increase during exercise (fig. 2). An obvious steady-state O_2 consumption ($\dot{V}O_{2ss}$) was not attained during the 20-min exercise bout. The time to attain 50% of the peak $\dot{V}O_2$ ($t_{1/2\ peak}$) was approximately 5–6 min at each velocity. $\dot{V}O_2$ reached a mean value of 1.5 times the resting $\dot{V}O_2$ at the high velocity (fig. 2). $\dot{V}O_2$ in the control experiment ($n = 4$) decreased to the resting $\dot{V}O_2$ level in 45 min and showed no change during the subsequent 95 min.

The mean $\dot{V}O_2$ early in recovery showed a slight increase over the exercise rates. After approximately 10 min of recovery, $\dot{V}O_2$ declined to an asymptote somewhat

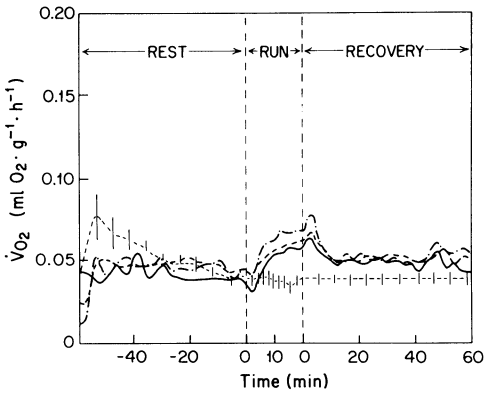

FIG. 2.—Oxygen consumption ($\dot{V}O_2$) of *Sesarma cinereum* on a treadmill during rest, exercise, and recovery periods. The three curves showing an increase in $\dot{V}O_2$ during the run represent three running velocities: 0.13 (– · –), 0.10 (——), and 0.06 (– – –) $km \cdot h^{-1}$. The curve with vertical bars (±1 SE) is a control experiment where animals were not exercised. Each curve is the mean of four to five animals.

above the resting levels (fig. 2). The elevated recovery $\dot{V}O_2$ remained relatively constant until the experiment was terminated 60 min after the run.

No significant increase in $\dot{V}O_2$ with velocity was found. The net volume of O_2 consumed (area under $\dot{V}O_2$ curve minus resting $\dot{V}O_2$) was not different from that at medium velocity ($P > .05$). However, three of five animals did show their greatest increase in $\dot{V}O_2$ at the high velocity.

In figure 3 the mean $\dot{V}O_2$ of five crabs

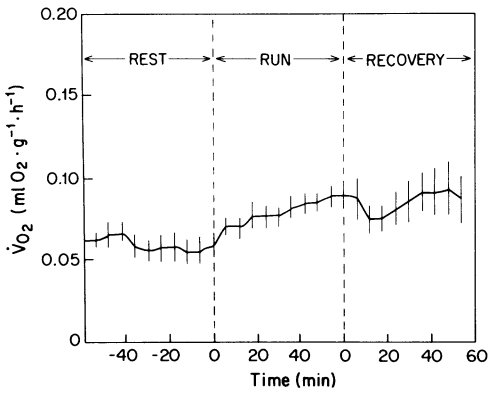

FIG. 3.—Oxygen consumption of *Sesarma cinereum* on a treadmill during 60-min rest, run (0.06 $km \cdot h^{-1}$), and recovery periods. The curve is a mean of five animals and vertical bars represent ±SE of mean.

is shown for the long-duration (60-min) exercise experiments at 0.06 km · h^{-1}. $\dot{V}O_2$ increased continuously throughout the exercise period. Again, the rate of O_2 uptake rose only to 1.6 times the resting levels. During the first 10 min of the recovery period, $\dot{V}O_2$ began to decline. After this time $\dot{V}O_2$ increased to near exercise levels. This elevated recovery $\dot{V}O_2$ occurred when the animals became very active as they attempted to escape from the respiratory chamber.

The $\dot{V}O_2$ kinetics of five crabs exercising in air saturated with water vapor was similar to those running at the same velocity at 50% RH. The aerobic factorial scope for crabs exercising at 100% RH (1.7) was not significantly different from those tested at 50% RH (1.5; t = 0.24, P = .59). Also, crabs exercising at 100% RH showed no weight loss.

$\dot{V}O_{2max}$.—Five crabs were subjected to a progressive $\dot{V}O_{2max}$ test after a 60-min rest period (fig. 4). Maximal O_2 consumption is generally defined as the $\dot{V}O_2$ attained when consecutive increases in velocity result in no further increase in $\dot{V}O_2$ (Seeherman et al. 1981). A $\dot{V}O_{2ss}$ was not clearly attained during the present test; $\dot{V}O_2$ continued to show a small rise during exercise until the crab fatigued (therefore, $\dot{V}O_{2max}$ may not have been reached, or, more likely, the animal could have been exercising near its maximum aerobic capacity

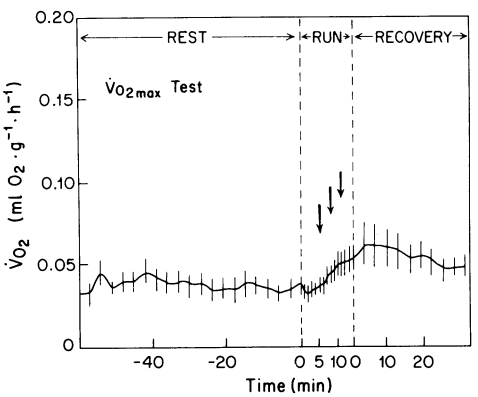

FIG. 4.—Progressive maximal oxygen consumption test after a 60-min rest period. Arrows represent 0.014 km · h^{-1} increases in velocity after 5 min of initial exercise at 0.14 km · h^{-1}. The curve is a mean of five animals and vertical bars show ±SE of mean.

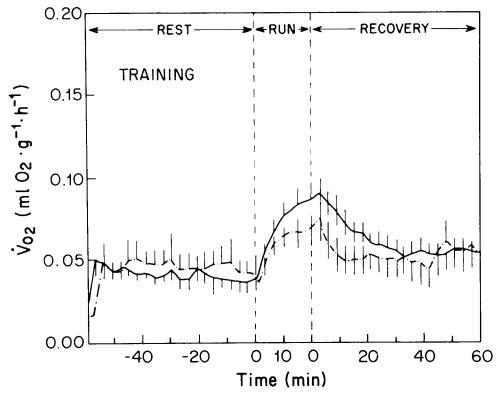

FIG. 5.—Oxygen consumption of *Sesarma cinereum* on a treadmill during rest, run (0.13 km · h^{-1}), and recovery periods. The two curves represent the mean of four untrained animals (– · –) and eight crabs trained for 5 wk (——). Vertical bars represent ±1 SE of the mean.

even at the initial velocity tested). Later, in the 30-min recovery, $\dot{V}O_2$ showed a decline toward resting levels.

Training.—In figure 5 the $\dot{V}O_2$ of trained vs. untrained crabs is compared. The resting $\dot{V}O_2$ during the last 15 min of a 60-min rest period was not different between the groups ($P > .05$). The volume of O_2 consumed over resting levels (net $\dot{V}O_2$) by the trained crabs (11.2 µl $O_2 \cdot g^{-1}$) for 20 min of exercise at 0.13 km · h^{-1} was significantly greater than for the untrained crabs (5.0 µl $O_2 \cdot g^{-1}$; t = 1.94; $P < .05$).

LACTATE PRODUCTION AND REMOVAL

Net WBL content was determined after a 60-min rest period; the level found was 0.30 mg · g $animal^{-1}$ ± 0.025 SE (n = 4). This amount was not different from the pooled, nonexercised control values (0.31 mg · g^{-1} ± 0.017 SE; n = 7; see fig. 6).

Sustained exercise.—Net WBL increased during the entire exercise period at the highest velocity tested (fig. 6). A stepwise polynomial regression analysis (Zar 1974) showed that a linear function was the best fit ($P < .05$), while a t-test revealed that the slope of a least-squares regression line was significantly different from zero (t = 2.34; $P < .05$). The regression equation is WBL = 0.010 T + 0.339 (n = 34; r = .55), where T represents time

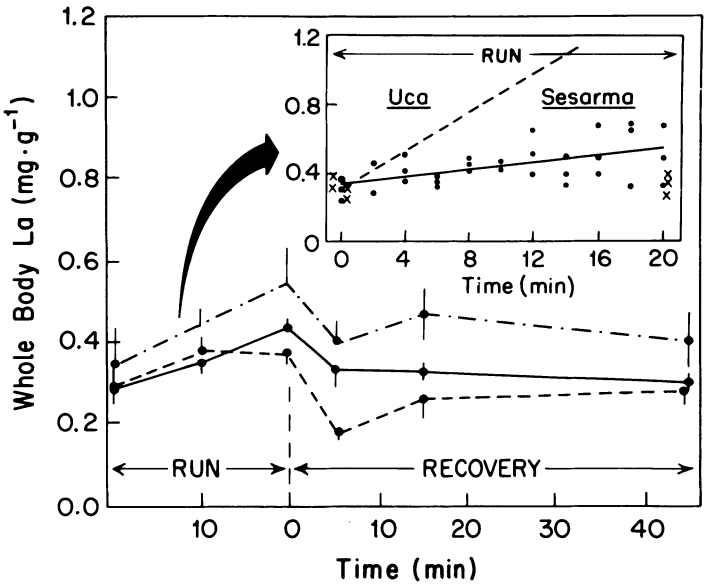

FIG. 6.—Time course of net whole-body lactate content during rest (t = 0), exercise, and recovery at three velocities: 0.14 (–•–), 0.10 (——), and 0.06 (– – –) km·h^{-1}. Vertical bars are ±1 SE except for the regression line at 0.14 km·h^{-1} where bars represent 95% confidence intervals. Inset: Expanded time scale showing individual data points and regression line for net whole-body lactate during exercise at 0.14 km·h^{-1}. Control data are designated by the letter x; x's to the left of t = 0 represent values of animals immediately after removal from holding tank. The regression for *Uca pugilator* running at 0.16 km·h^{-1} is shown for comparison (Full and Herreid 1984).

in minutes. No significant difference at the low and medium velocities was found when 10- and 20-min WBL values were compared with resting levels, although there was some suggestion of an increase in the mean levels with time (fig. 6).

During the first 5 min of recovery, WBL declined. At the low velocity the decline (0.18 mg·g^{-1}) was below the resting WBL level (t = 3.30; $P < .05$). WBL content at 45 min of recovery was not significantly different from the resting WBL levels.

Training.—Fifteen crabs were analyzed for WBL content after 5 wk of training. Net WBL was lower in trained crabs (0.20 mg·g^{-1}) than untrained animals (0.30 mg·g^{-1}) after a 60-min rest period on the treadmill (t = 3.00; $P < .05$). Net WBL in the trained crabs was not significantly different from the untrained animals after 5, 10, 15, or 20 min of exercise at 0.14 km·h^{-1} ($P > .05$). At the end of the 20-min exercise bout, trained crabs did show a 45% decrease in mean net WBL, but because of the large variation for the untrained animals, the decrease was not statistically significant.

DISCUSSION

ENDURANCE CAPACITY

The endurance versus velocity relationship found in the present study is typical of other animals. As the crabs were exercised at faster velocities the time to fatigue declined (fig. 1). The major decrease in endurance occurred at 0.10–0.15 km·h^{-1}. This range of maximum sustainable velocities is somewhat below that of *Uca pugilator* and is only half that of the ghost crab, *Ocypode gaudichaudii* (Full and Herreid 1983, 1984). Five weeks of training does not result in a faster maximum sustained velocity, in contrast to our observations for the trained fiddler crab, *U. pugilator* (unpublished results). *Sesarma cinereum* does not appear exceptional with respect to its endurance capacity. If any statement can be made in light of the

small number of other crab species tested, it is that this crab can sustain only very slow rates of locomotion over extended periods.

AEROBIC METABOLISM

Sesarma cinereum displays a limited aerobic response to exercise; its $\dot{V}O_2$ differs from that previously reported for exercising crabs with respect to (1) the pattern of the $\dot{V}O_2$ increase and (2) the relationship of $\dot{V}O_2$ versus running velocity. In addition, the wharf crab demonstrates little change in its aerobic response after training.

Aerobic response.—Figure 7 provides a direct comparison of the aerobic responses which have been observed in exercising crabs. Running crabs such as *O. gaudichaudii* show a rapid elevation in $\dot{V}O_2$ leading to a steady state up to 10 times the resting $\dot{V}O_2$ (Full and Herreid 1983). A rapid aerobic adjustment of this type is characteristic of running mammals which rely primarily on aerobic metabolism to power sustained locomotion and can elevate $\dot{V}O_2$ to 10 times their resting consumption (Taylor et al. 1980). Other exercising pedestrian crabs seem to be limited in their ability to increase the rate of O_2 transport. A significant lag in the $\dot{V}O_2$ increase at the onset of exercise has been reported for *Uca pugilator* (Full and Herreid 1984). In addition to *U. pugilator, Cardisoma guanhumi* and *Gecarcinus lateralis* do not attain a $\dot{V}O_{2ss}$ over a 10- to 20-min exercise bout (Herreid et al. 1979, 1983). These species are capable of increasing O_2 transport by only a factor of three to five times.

Exercising *S. cinereum* increased $\dot{V}O_2$ during locomotion by only 50% during 20 min of exercise. No greater $\dot{V}O_2$ elevation was found even after 60 min of exercise (fig. 3). *Ocypode gaudichaudii* and *U. pugilator,* running at comparable velocities, elevated $\dot{V}O_2$ by three- and sevenfold, respectively (fig. 7). It is clear that the $\dot{V}O_2$ of *S. cinereum* represents an extraordinarily reduced aerobic response, more so than that described for any other exercising crab.

$\dot{V}O_2$ *vs. velocity.*—A wide variety of birds, mammals, and reptiles show a linear increase in $\dot{V}O_{2ss}$ with an increase in speed

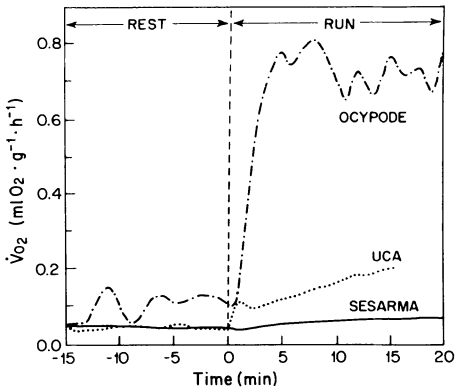

FIG. 7.—Oxygen consumption of *Sesarma cinereum* (——) at 0.13 km · h^{-1} compared with *Ocypode gaudichaudii* (– · –) and *Uca pugilator* (· · ·) during rest and exercise. *Ocypode gaudichaudii* (2.69 g) was run at 0.13 km · h^{-1}; *U. pugilator* (mean mass = 2.26 g) was run at 0.11 km · h^{-1}.

(Taylor 1977; Taylor, Heglund, and Maloiy 1982). Previous research, using techniques identical with those employed in the present study, has revealed a similar relationship for five species of cockroaches and a ghost crab (Herreid, Full, and Prawel 1981*a;* Full and Herreid 1983; Herreid and Full 1984). Crabs walking underwater appear to follow the same trends (Houlihan and Innes 1984; Houlihan, Mathers, and El Haj 1984). Even exercising crabs which do not attain a $\dot{V}O_{2ss}$ consume greater amounts of O_2 as they run at faster velocities (Herreid et al. 1979, 1983; Wood and Randall 1981*a;* Full and Herreid 1984). For some of these crab species the increase in energy demand is predominantly met by anaerobic ATP production (Full and Herreid 1984).

The amount of oxygen used by *S. cinereum* did not increase significantly as the crab doubled its running velocity (0.06–0.13 km · h^{-1}; fig. 2). Moreover, the rate of O_2 uptake was not different from that measured at the fastest sustained velocity, even when the velocity was progressively increased ($\dot{V}O_{2max}$ test). A comparable relationship has been described for mammals exercising near or beyond the rate at which $\dot{V}O_{2max}$ is attained (Seeherman et al. 1981). At these velocities no increase in $\dot{V}O_2$ was observed, while the rate of anaerobic metabolism showed a

proportional elevation with velocity. Perhaps *S. cinereum* is exercising near its $\dot{V}O_{2max}$ even at slow rates of locomotion.

Training.—Studies on humans show that the aerobic response to exercise depends on the condition of the individual. Sedentary humans have sluggish $\dot{V}O_2$ kinetics and a lower $\dot{V}O_{2max}$ compared with athletes (Hickson, Bomze, and Holloszy 1978; Cerretelli et al. 1979). Training decreases the lag time required to reach $\dot{V}O_{2ss}$ and $\dot{V}O_{2max}$ increases. Training in *S. cinereum* leads to a near doubling in the amount of O_2 consumed during exercise (fig. 5), but the reasons for this improvement are unknown.

The data from sustained exercise, high-velocity, and training experiments all support the conclusion that the oxygen transport capacity of *S. cinereum* is limited. Since little is known about the respiratory physiology of this crab, the site of limitation cannot be determined. Nevertheless, the improvement of exercise endurance with 100% oxygen (table 1) indicates that the limitation exists not with the muscles' ability to use oxygen but with the transport from gill to the mitochondria. Individuals of *Sesarma* have been reported to pump water from their gill chamber out of their mouth, across the branchiostegite, to the base of the walking legs (Horn 1968; Felgenhauer and Abele 1983). Water traveling this route is apparently aerated and returned to the branchial chamber. The use of this respiratory mechanism could result in significant desiccation during prolonged exposure to air and thereby affect oxygen diffusion at the gill. However, two lines of evidence suggest that the gill may not be the site limiting oxygen conductance. First, no change was observed in oxygen consumption when crabs were exercised in 100% RH where no desiccation was possible. Second, *Sesarma* has a more than adequate gill area/O_2 uptake ratio (14.8 mm^2 per μl O_2 consumed) when compared with other terrestrial and semiterrestrial crabs such as *Ocypode quadrata* (1.9 $mm^2/\mu l$ O_2) and *Uca pugilator* (12.7 $mm^2/\mu l$ O_2; Gray 1957). If this reasoning is sound, then the circulatory system becomes the most likely candidate for the area of limitation. Whatever the site of O_2 limitation, O_2 conductance is certainly low in comparison to other crab species.

LACTATE PRODUCTION

In vertebrates, anaerobic metabolism provides energy for activity primarily under two conditions: (1) at the onset of exercise, where the lag in $\dot{V}O_2$ is observed before a $\dot{V}O_{2ss}$ is attained (O_2 deficit period); and (2) at work levels exceeding $\dot{V}O_{2max}$. A similar pattern has been suggested for exercising crustaceans (Full and Herreid 1984).

For *S. cinereum* no clear $\dot{V}O_{2ss}$ is present, and $\dot{V}O_2$ appears to be always near the maximum possible rate. Both conditions suggest a dependence on anaerobic metabolism. However, we noted a significant increase in net WBL only at the high velocity, 0.14 $km \cdot h^{-1}$ (fig. 6). Because the lactate increase was constant over the 20-min exercise bout, a regression slope can be used to estimate the net rate of WBL production. This rate, 0.010 $mg \cdot g^{-1} \cdot min^{-1}$, is unexpectedly low when compared with a crab such as *U. pugilator* that relies on anaerobic fermentation at a comparable velocity (fig. 6, inset). In the fiddler crab, *U. pugilator,* L-lactate fermentation accounted for approximately 70% of the energy requirement for exercise at the maximum sustained velocity (0.16 $km \cdot h^{-1}$). The fiddler crab not only consumes nearly six times more O_2; it also produces six times more lactate (Full and Herreid 1984). These data suggest that lactate fermentation in *S. cinereum* does not provide sufficient energetic compensation for the low rate of aerobic metabolism. Are there other sources for energy to explain the low $\dot{V}O_2$ patterns of *S. cinereum?*

There is little evidence that unusual anaerobic end products, such as those found in molluscs (e.g., deZwaan 1977), are involved. We (in collaboration with W. R. Ellington) have analyzed *S. cinereum* after exercise and found no increase in D-lactate, alanine, strombine, octopine, succinate, or malate. Moreover, L-lactate appears as the primary indicator of anaerobic ATP production in the only crustacean thoroughly examined during hypoxia

(Zebe 1982). Also, L-lactate is produced in abundant quantities during exercise in other crustacean species (e.g., McMahon 1981; Taylor 1982). Only the provocative results of Smatresk and Cameron (1981) and Booth (1982) reporting an unknown metabolic acid suggest an unusual anaerobic end product may be produced in some crabs.

Oxygen stores and high-energy phosphate stores such as arginine phosphate are also unlikely to account for major amounts of energy during prolonged exercise. According to the analysis of Herreid (1981), a 2-g crab with 30% blood and an O_2-carrying capacity of 2% would have only 0.025 ml O_2 stored in blood and tissues. Assuming (1) 25% of the muscle mass is used for locomotion, (2) crustacean muscle contains 6.95 μmol ATP/g and 33.36 μmol arginine phosphate/g (Beis and Newsholme 1975), and (3) 1.0 μl O_2 = 0.283 μmol ATP, we estimate that there might be 20 μmol of ATP and phosphagen in the locomotory muscles of these crabs. This is the equivalent of 0.07 ml O_2. Together, O_2 and high-energy phosphate stores would amount to only 0.1 ml O_2; ghost crabs of the same size used five times this amount of O_2 to travel at the same speed. As further evidence of the inadequacy of these energy sources, we found the wharf crab had no extraordinary endurance capacity when it was exercised in an N_2 atmosphere where it had to rely solely on stored O_2 or phosphates and on anaerobically produced energy.

Since we cannot readily account for *S. cinereum*'s low $\dot{V}O_2$, we must consider the possibility that it may require less energy to run than other crabs previously tested. How this might be accomplished is not clear, although greater elastic storage in the legs, greater transfer of potential and kinetic energy during walking, and the use of a more efficient muscle fiber type are possibilities (see Cavagna, Heglund, and Taylor 1977).

Whatever the solution to the problem may be, *S. cinereum* appears unique among running animals. It requires very small amounts of O_2 during exercise compared with other crab species, with no obvious compensation by other energetic mechanisms. Its pattern of energetic utilization represents a third type of response to exercise, distinct from the aerobic pattern of ghost crabs and the mixed aerobic and anaerobic (lactate) mechanisms of fiddler crabs.

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