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Swimming in the sea hare *Aplysia brasiliana*: Cost of transport, parapodial morphometry, and swimming behavior

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Abstract

The energetics and behavior of the parapodial-swimming *Aplysia brasiliana* were investigated in order to compare net cost of transport (COT_{net}) between swimming and crawling, and to compare transport costs with other swimmers. Oxygen consumption (V_{O_2}) increased with increasing animal mass for resting, crawling, and swimming animals. Slopes of the regressions of log V_{O_2} on log mass were 0.90, 0.91, and 0.89 for resting, crawling, and swimming, respectively. The regression for resting V_{O_2} on mass was significantly lower than regressions of crawling and swimming on mass, which fell into a statistically homogenous subgroup. During 4-h swimming bouts, parapodial beat frequency dropped by less than 10% of starting values after 2 h and then stabilized for the remainder of the trial, whereas velocity steadily decreased to about 70% of starting values over the 4-h period. Initial beat frequency (at the start of a swimming bout) was negatively related to body mass, varying from 1.1 beat s^{-1} for a 34 g individual to 0.7 beats s^{-1} for a 500 g individual. Final beat frequency (at the end of a swimming bout) was also negatively related to body mass, but had a significantly lower intercept than initial beat frequency. Neither initial swimming velocity nor final swimming velocity was related to mass, but final velocity was significantly lower than initial velocity. A 250 g *A. brasiliana* swam at 345 m h^{-1} and crawled at 7 m h^{-1}. Swimming COT_{net} (0.1 ml O_2 kg^{-1} m^{-1}) for a 250 g *A. brasiliana* was 50 times less than crawling COT_{net} (5.3 ml O_2 kg^{-1} m^{-1}). While the crawling COT_{net} for *A. brasiliana* fell within the range of other marine gastropods, swimming COT_{net} was less than that of swimming crustaceans, and much less than another gastropod, *Melibe leonina*, that uses lateral bending to swim.

Keywords: *Aplysia*; Cost of transport; Gastropod; Sea hare; Swimming

1. Introduction

The energetics of running, swimming, and flying have been studied in a wide variety of taxa, both vertebrate (Schmidt-Nielsen, 1972) and invertebrate

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(Full, 1997). Gastropod crawling has also received attention and is considered the most costly form of locomotion, in part owing to the expense of producing mucus (Denny, 1980). Some gastropods, however, swim as well as crawl. The costs and benefits of swimming vs. crawling in these organisms are poorly understood because to our knowledge only one study has investigated the costs of both in a single species (Caldwell and Donovan, 2003). Three general modes of swimming in gastropods have been reported: (1) parapodial or mantle flapping (*Gastropteron*, *Hexabranchus*, and *Aplysia*), (2) dorsoventral undulation (*Tritonia* and *Pleurobranchaea*), and (3) lateral bending (*Melibe* and *Dendronotus*) (see Farmer, 1970 for complete review). Of these, only the cost of lateral-bend swimming in the nudibranch *Melibe leonina* has been studied. Swimming using lateral bending is relatively costly due to high energy expenditure, and a non-directional and slow swimming speed (Caldwell and Donovan, 2003). In comparison, parapodial flapping is characterized by directional, relatively fast swimming, and it is likely that the transport costs would be much lower. In this paper we examine the costs of swimming by parapodial flapping in *Aplysia brasiliana*.

While the neurobiology of *Aplysia* has been well studied, less is known about its behavior and ecology (Carefoot, 1987). In particular, the physiology, ecology, and physics of swimming are poorly understood. Of over 30 species of *Aplysia*, less than six are known to swim, and this number may be reduced as further species synonymies are disclosed (Medina et al., 2001). Sea hares swim by flapping large parapodia (bilateral extensions of the mantle) across their backs. Hypotheses on the mechanism of propulsion during swimming in sea hares include sculling (Farmer, 1970; Bebbington and Hughes, 1973), jetting (Neu, 1932), and hydrodynamic lift (von der Porten et al., 1982), but definitive tests of these hypotheses have not been done.

Most of our knowledge of swimming in sea hares comes from two species, *Aplysia fasciata* and *A. brasiliana*, which are closely related (Medina et al., 2001). *A. fasciata* occurs in the eastern Mediterranean Sea where it inhabits sand and rock areas, eats mainly green-algal foods, and is nocturnally active (Susswein et al., 1983, 1984). Its swimming has been studied primarily from an ethological perspective (Susswein et al., 1983, 1984; Ziv et al., 1991a,b, 1994). *A. brasiliana* occurs in the northern Gulf of Mexico where it inhabits seagrass beds, favors red-algal foods, and is active in the late afternoon and early evening. Early studies on swimming in *A. brasiliana* focused on mechanisms of propulsion and orientation (Hamilton and Russell, 1982; von der Porten et al., 1982; Hamilton, 1986), but more recent work has examined ecological aspects of swimming (Carefoot and Pennings, 2003). Swimming in both *A. brasiliana* and *A. fasciata* appears to be highly dependent upon food-related cues (Aspey et al., 1977; Susswein et al., 1984; Ziv et al., 1991a,b; Carefoot and Pennings, 2003), but other factors such as social interactions and avoidance of noxious stimuli are also involved (Ziv et al., 1991a,b, 1994; Levy and Susswein, 1999; Carefoot and Pennings, 2003). Greater understanding of the behavioral and evolutionary significance of swimming in sea hares has been hindered by a lack of data on the energetic costs of swimming. Knowledge of the cost of swimming in *Aplysia* is also a prerequisite for comparing the physiology and ecology of locomotion between sea hares and other swimming gastropods.

A full understanding of swimming in *Aplysia* will require attention to scaling issues. Within a single species, the range in mass of swimming individuals may span over an order of magnitude (authors' personal observations). Thus, understanding how swimming behavior and energetics scale with mass may be critical to understanding the function and ecology of swimming. Similarly, because *Aplysia* may swim for extended periods (hours), it is important to begin to understand how swimming behavior changes over time during bouts of swimming.

Here, we focus on the energetics and scaling of swimming in *A. brasiliana*. We ask three major questions. First, what is the energetic cost of swimming, and how does this compare with the cost of crawling, and with the cost of swimming in other invertebrates? Second, how does the energetic cost and other features (velocity, parapodial beats s^{-1}) of swimming scale with animal size? Third, as animals tire, how do they modify their swimming behavior?

2. Materials and methods

2.1. Experimental animals

A. brasiliana ranging from 34 to 506 g live mass were collected from harbors at south Padre Island, Texas in May 2003, and maintained indoors in 60-l plastic tubs with fresh running seawater (24–26 °C, 30 ppt) at the Marine Science Institute, University of Texas. Only vigorous, healthy animals were collected. Animals were provided with a mixture of green (*Ulva fasciata*) and red (*Hypnea musciformis*, *Gracilaria dibilis*) algae ad libitum. No individual was used more than once in any experiment. Once in the laboratory, animals became active throughout the day, presumably due to the absence of the particular environmental cues used to entrain their normal circadian rhythms. To standardize behavior, all measurements were made during 1000–1400 h.

2.2. Aerobic energy expenditure

To determine aerobic energy expenditure during swimming, crawling, and resting, oxygen consumption was measured during each of these states in a closed-system respirometer. For each oxygen-consumption trial, an individual sea hare ($N=25$; 34–506 g live body mass) was placed in a plastic container (1500, 2500, 4000, or 7500 ml depending on animal size) with a tight-fitting lid. Respirometer size was chosen to permit each sea hare to swim freely without contacting the side with its parapodia while being small enough to accurately measure declining oxygen levels. Thus, the size of the respirometer constrained swimming such that the sea hares could not swim in one direction for an extended length of time. Seawater (24–26 °C) was run freely into the container for a few moments, allowing all bubbles to be displaced. An oxygen electrode (Cameron Instrument Company; Pt. Aransas, TX) was inserted into a hole drilled into the lid and connected to an oxygen monitor (DI 2000, Cameron Instrument Company), which in turn was connected to a data-acquisition system (DataQ; Ackron, OH). This allowed the oxygen tension in the respirometer to be monitored continuously. The respirometer assembly sat on a magnetic stirrer that turned a stir-rod within the container turning at a speed to ensure complete mixing of the seawater during all activity states but low enough so as not to influence swimming.

Oxygen-consumption rates were determined during three states of activity for each animal: resting (sea hare stationary and displaying little obvious movement), crawling (foot attached to the sides or bottom of the container and sea hare exhibiting forward movement), and swimming (foot detached from the substratum and parapodia flapping) with the seawater being refreshed within the container between activity bouts. Most animals spontaneously exhibited all three states, although only 14 crawled in the respirometer for a period long enough to obtain a reliable V_{O_2} (see below). An activity had to continue for at least 10 min for the P_{O_2} data to be used. We did not allow the P_{O_2} in the respirometer to drop to less than 70% of the theoretical saturation value.

2.3. Swimming and crawling behavior

To determine how velocity and parapodial beat frequency scaled with animal mass, and how these variables changed as a function of animal fatigue, we conducted swimming trials with the same 25 animals used in the respirometry experiments. Swimming trials were conducted in an indoor, circular, fiberglass tank (0.5 m depth; 1.5 m diameter) containing non-agitated seawater at 24–26 °C. After completion of the respirometry measurements, animals were rested for 24 h with abundant food and then used in swimming trials. Animals were allowed to swim continuously for 4 h, and they did this typically at the water surface. At the beginning of the trial, and every 30 min thereafter for 4 h, we recorded parapodial beat frequency and swimming velocity. Swimming velocity was measured at the surface, in the center of the tank (i.e., animals were not touching tank walls), by placing a meter stick alongside a swimming animal and recording the time taken to swim 100 cm. Parapodial beat frequency was measured immediately after by counting the number of beats in 60 s. Velocity measurements were made in triplicate and averaged for each animal at each time period. To reduce variation in initial values, we averaged results from 0 and 30 min for each animal (it was customary for animals to take several minutes to adopt the streamlined posture of the foot, rhinophores, and oral tentacles that is characteristic of a long-duration swimming bout; Carefoot and Pennings, 2003).

After an additional 24-h rest for the 25 test animals, with ad libitum food provided, crawling velocity was determined by allowing individuals to crawl in a shallow tank (0.2 m depth; 1.5 m diameter) and recording distance covered during timed bouts (1-min duration, averaged over two discrete bouts separated by a 1–2 min interval). Because five animals refused to crawl, crawled for less than 1 min, or failed to crawl for two discrete 1-min bouts (in general, *A. brasiliana* is a poor crawler, see Discussion), we obtained data for only 18 animals (34–443 g live mass, representing a mean of 250 g).

2.4. Swimming and crawling costs of transport

Total cost of transport is used as a measure of aerobic energy expenditure per unit distance traveled (COT_{tot} ml O_2 kg^{-1} m^{-1}; Schmidt-Nielsen, 1972). Net cost of transport (COT_{net}) is calculated by subtracting standard oxygen consumption (resting) from active oxygen consumption (swimming or crawling) before dividing the result by the speed of locomotion.

Because swimming was constrained by the close-system respirometer, the net cost of transport during swimming was estimated as follows. Mass-specific resting oxygen consumption for each animal was subtracted from its mass-specific swimming oxygen consumption to give the amount of oxygen required for swimming per unit mass of animal. This was then divided by the initial speed (time averaged over 0–30 min) of the same animal as measured in the swimming behavior experiments.

Cost of transport for crawling was estimated in a slightly different manner because of the inconsistent crawling behavior already noted. Mass-specific resting oxygen consumption for each animal was subtracted from its mass-specific crawling oxygen consumption to give the amount of oxygen required for crawling per unit mass of animal. This was then divided by a speed determined for each animal from the regression of log speed vs. log mass generated from a larger sample of sea hares (see Results).

2.5. Parapodial morphology

To determine how parapodial area scaled with mass, individual *A. brasiliana* ($N=42$) were weighed and the area of their right parapodium measured. Parapodial area was determined by taking a digital photograph of each animal while it was resting in a relaxed position on its left side in a small amount of seawater. A 10-cm ruler was included in the photograph for reference. Image analysis software (NIH Image) was used to determine parapodial area.

2.6. Statistical analysis

Although Model II regression is most appropriate for data in which the independent variable is not fixed, Model I regression was used in this study since measurement error of the independent variable (mass) was less than that of the dependent variables. Since the correlation coefficient for the Model I regression was large (see Results), the difference between Model I and Model II regression is small (Laws and Archie, 1981). When the Model I regression equation had been determined, the resulting slope was compared to the expected slope for isometry using the comparison-of-slopes methods described in Zar (1996).

3. Results

3.1. Aerobic energy expenditure

During swimming in the respirometer, parapodial beat frequency ranged from 0.8 to 1.1 beats s^{-1} for all of the animals. This beat frequency and the observed swimming behavior (parapodia completely overlapping in the closed position of the swimming cycle) were similar to that of rested animals in large tanks (see below).

Oxygen consumption increased significantly with increasing animal mass for all three activity states (Fig. 1; $t>7.0$, $P<0.001$ for all slopes). The slopes of the regressions (b) were not significantly different ($F_{2,58}=0.02$, $P>0.5$; ANCOVA) and ranged between 0.89 and 0.91. The intercepts, however, were significantly different ($F_{2,60}=47.02$, $P<0.001$; ANCOVA), with the resting treatment falling into its own statistical subgroup, and the crawling and swimming treatments forming another subgroup ($P<0.05$; Tukey's multiple comparison tests).

Rates of oxygen consumption for a "standard" 250 g *A. brasiliana* were calculated from these regression equations. Oxygen consumption during swimming

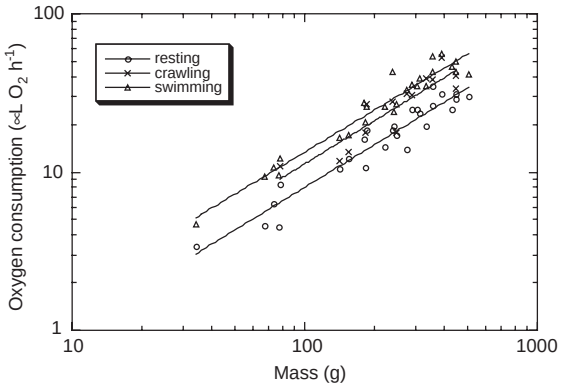

Fig. 1. Oxygen uptake of swimming ($N=25$), crawling ($N=14$), and resting ($N=25$) *A. brasiliana* as a function of mass. The regression equations for each state of activity are: log resting $V_{O_2}=-0.90+0.90\log$ mass, $r^2=0.91$, $P<0.001$; log crawling $V_{O_2}=-0.76+0.91\log$ mass, $r^2=0.81$, $P<0.001$; and log swimming $V_{O_2}=-0.65+0.89\log$ mass, $r^2=0.94$, $P<0.001$.

was the highest (30 ml O_2 h^{-1}) followed by oxygen consumption during crawling (26 ml O_2 h^{-1}) and oxygen consumption during resting (18 ml O_2 h^{-1}).

3.2. Swimming and crawling behavior

Swimming *A. brasiliana* adopted a characteristic streamlined body posture (Carefoot and Pennings, 2003), with oral tentacles rolled over, edges of the foot curled inward such that none of the foot is exposed, and rhinophores laid back along the head. Swimming is accomplished by opening and closing the parapodia in a sinusoidal flapping motion, but whether propulsion is derived from sculling, jetting, or hydrodynamic lift, or some combination of the three, is not known. In freshly rested animals, the parapodia overlapped (left over right) in the closed position at the end of a beat cycle. As the animals tired, however, the parapodia overlapped less, or even failed to meet at the end of a beat cycle.

Initial (time 0–30 min) parapodial beat frequency was negatively related to body mass (log–log slope, b, equal to -0.18), varying from 1.1 beat s^{-1} for a 34 g individual to 0.7 beats s^{-1} for a 500 g individual (Fig. 2A). After 4 h of swimming, the relationship between final beat frequency and body mass differed from the initial relationship as follows. The slopes of these two regressions were not statistically different ($b=-0.13$ for final beat frequency vs. mass; $F_{1,46}=1.94$, $P>0.2$; ANCOVA) but the intercepts were ($F_{1,48}=7.10$, $P=0.02$; ANCOVA). However, beat frequency of a standard 250 g *A. brasiliana* fell to only 95% of initial frequency after 4 h of swimming. Both initial swimming velocity and final swimming velocity were unrelated to mass (Fig. 2B). Mean initial velocity (345 ± 50 m h^{-1}; all values are mean ± S.D. unless otherwise noted) was significantly greater than mean final velocity (241 ± 51 m h^{-1}; $t=9.63$, $P<0.001$; paired t-test). Thus, after 4-h continuous swimming, velocities dropped to 70% of initial rates.

Changes in beat frequency and velocity were gradual over the 4-h duration of the swimming experiment (Fig. 3). Note that the reduction in velocity, which was continuous over the entire 4-h trial, did

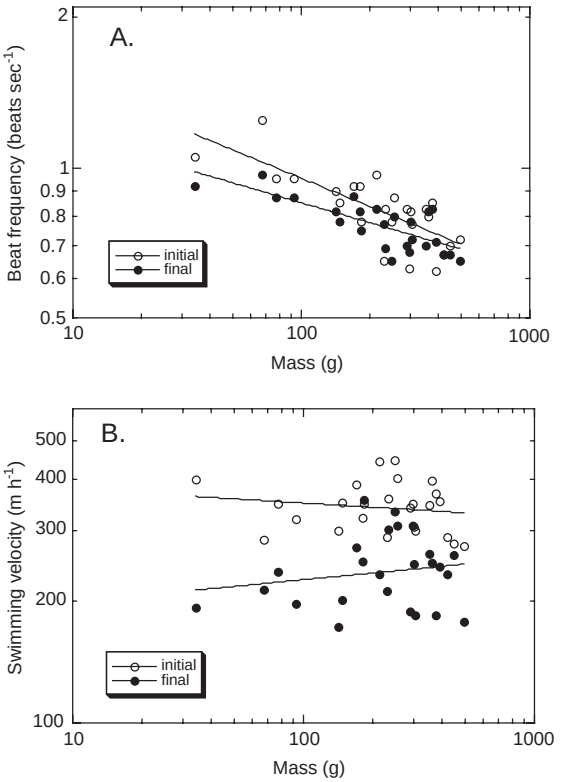

Fig. 2. Parapodial beat frequency (A) and swimming velocity (B) as a function of mass in *A. brasiliana*. Initial (time 0–30 min) values are indicated by open symbols and final (after 4 h of swimming) values by closed symbols. The regression equations were: log initial beat frequency $=0.33-0.18\log$ mass, $r^2=0.63$, $P<0.0001$; log final beat frequency $=0.19-0.13\log$ mass, $r^2=0.57$, $P<0.0001$; log initial velocity $=2.61-0.03\log$ mass, $r^2=0.03$, $P=0.45$; log final velocity $=2.24+0.05\log$ mass, $r^2=0.03$, $P=0.40$.

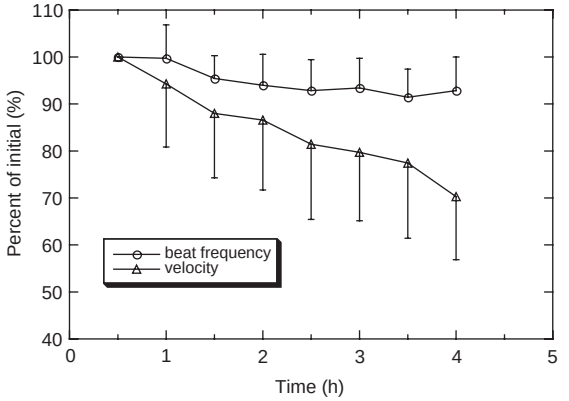

Fig. 3. Changes in parapodial beat frequency and velocity in *A. brasiliana* at 30 min intervals over 4 h of continuous swimming. Data are expressed as a percent of initial (times 0 and 30 min averaged) values. Each point represents the mean and S.D. of $N=25$ animals.

not match the pattern of reduction in beat frequency, which dropped slightly between 1 and 2 h and appeared to remain stable thereafter.

Crawling speed was quite variable but increased significantly with mass (Fig. 4). The speed of crawling for a standard 250 g *A. brasiliana* was 7 m h^{-1}.

3.3. Cost of transport

Neither swimming nor crawling COT_{net} was affected by body mass ($t=0.76$, $P=0.46$ and $t=0.45$, $P=0.51$, respectively) so a mean COT_{net} was determined regardless of size. Thus, COT_{net} was 0.1 ± 0.05 ml O_2 kg^{-1} m^{-1} during swimming for *A. brasiliana* and 5.3 ± 2.8 ml O_2 kg^{-1} m^{-1} during crawling.

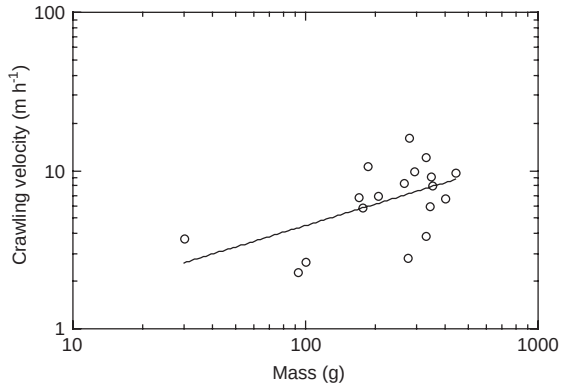

Fig. 4. Crawling velocity of *A. brasiliana* as a function of mass. The regression equation is log crawling velocity $= -0.25 + 0.45$ log mass, $r^2=0.29$, $P=0.02$.

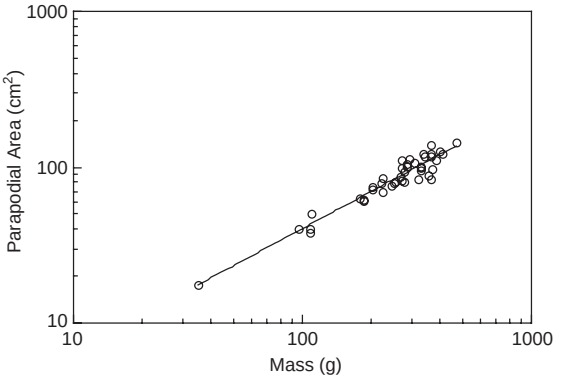

Fig. 5. Relationship between parapodial area and body mass in *A. brasiliana*. The relationship is described by the equation log parapodial area $= 0.03 + 0.79$ log mass, $r^2=0.92$. The slope is significantly greater than the predicted slope of 0.67 for isometry ($t=37.5$, $P<0.001$).

3.4. Parapodial morphology

Parapodial area scaled allometrically to mass (Fig. 5). Thus, the observed log–log slope of 0.79 was significantly greater ($t=37.5$, $P<0.001$) than the expected value of 0.67 (based on the isometric relationship between area and mass) indicating that larger *A. brasiliana* had disproportionately large parapodia.

4. Discussion

COT_{net} for *A. brasiliana* when swimming in still water (0.1 ml O_2 kg^{-1} m^{-1}) was 50 times less expensive than when crawling (5.3 ml O_2 kg^{-1} m^{-1}). Since energy expenditure during these two modes of locomotion was similar (30 and 26 ml O_2 h^{-1} for swimming and crawling, respectively; Fig. 1), this large difference was due to the relatively fast swimming pace (345 m h^{-1}, Fig. 2B) of *A. brasiliana* compared with a slow crawling pace (7 m h^{-1}, Fig. 4). The high COT of crawling is likely a combination of the inherent inefficiencies of crawling (mucus production, friction; Denny, 1980) and the fact that *A. brasiliana* is a particularly bad crawler, with a small (narrow) foot and weak foot musculature compared to other aplysiid gastropods (authors' personal observa-

tions). For example, *Aplysia dactylomela*, a non-swimmer, can crawl at 30–90 m h^{-1} (Carefoot, 1987), with a COT_{net} of 2.8 ml O_2 kg^{-1} m^{-1} (V_{O_2} data from Carefoot, 1989). For this "good crawler", the COT of crawling is about half that of crawling *A. brasiliana*, but still 28 times greater than that of swimming *A. brasiliana*.

Swimming COT likely varies in the field depending upon the degree of waves and currents, since speed and effort may also vary. In the field, *A. brasiliana* swims at average speeds of 230 to 440 m h^{-1} (bracketing the initial and final speeds recorded in this study; 345 and 241 m h^{-1}, respectively), regardless of current magnitude, water depth, or sun aspect (Hamilton and Ambrose, 1975). However, animals swimming down-current or experiencing high waves have slower water speeds than animals swimming up-current or experiencing small waves (Hamilton and Ambrose, 1975; Hamilton, 1984, 1986). Whether swimming effort varies depending on conditions in the field is not known; however, the effect of waves and current on water speed, coupled with displacement due to currents (which may increase or decrease net displacement depending on swimming direction relative to current direction and speed) will increase variation in COT. While the effects of environmental conditions on COT in the field deserve further study, the general result that swimming is far more efficient than crawling will likely hold under all but the most unusual conditions.

Compared with *M. leonina*, the only other swimming gastropod for which COT_{net} has been measured (Caldwell and Donovan, 2003), *A. brasiliana* displays a much more efficient swimming mode (Fig. 6). *M. leonina* swims using lateral bending which results in the oral-hood rhinophores hitting the tail as the body whips from side-to-side. While effective in lifting the body off of the substratum, this type of swimming is not effective for directional movement in still water. Note that while COT_{net} for crawling is similar for *A. brasiliana* and *M. leonina* (about 5 ml O_2 kg^{-1} m^{-1}; Fig. 6), COT_{net} for swimming is almost two orders of magnitude less in *A. brasiliana* than in *M. leonina*. This clearly illustrates the effectiveness of parapodial flapping as compared with lateral bending. It is probable that the other main

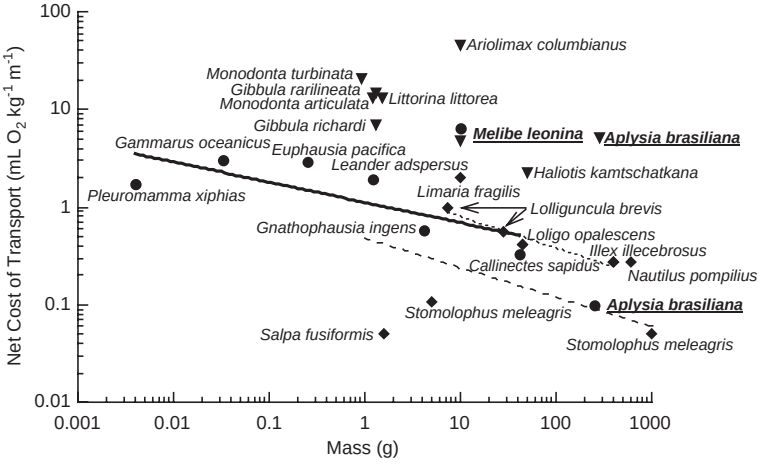

Fig. 6. Cost of transport of crawling (triangles), swimming (circles), and jetting (diamonds) invertebrates. The solid line represents the best fit line for swimming COT_{net} vs. mass for six species of swimming crustaceans (*Pleuromamma xiphias*, Morris et al., 1985; *Gammarus oceanicus*, Halcrow and Boyd, 1967; *Euphausia pacifica*, Torres and Childress, 1983; *Leander adspersus*, Ivlev, 1963; *Gnathophausia ingens*, Cowles and Childress, 1988; *Callinectes sapidus*, Houlihan et al., 1985). Jetting swimmers are squid (*Loligo opalescens*, O'Dor, 1982; *Illex illecebrosus*, Webber and O'Dor, 1986), a salp (*Salpa fusiformis*, Trueman et al., 1984), a nautilus (*Nautilus pompilius*, O'Dor et al., 1990), a jellyfish (*Stomolophus meleagris*, Larson, 1987), and a file shell (*Limaria fragilis*, Donovan and Baldwin, 1999). Data points for crawling and swimming *M. leonina* are from Caldwell and Donovan (2003). Crawling gastropods include tropical marine snails (*Gibbula rarilineata*, *Gibbula richardi*, *Monodonta articulata*, and *Monodonta turbinate*, Houlihan and Innes, 1982), temperate marine snails (*Littorina littorea*, Innes and Houlihan, 1985; *Haliotis kamtschatkana*, Donovan and Carefoot, 1997), and a terrestrial slug (*Ariolimax columbianus*, Denny, 1980). The dashed line represents a regression for 11 species of fish (Beamish, 1978) for comparison.

form of gastropod swimming, dorsoventral undulation, has high transport costs similar to those of lateral bending, owing to the non-directional nature of this type of swimming.

Parapodial flapping also appears to be an efficient mode of swimming compared with other swimming invertebrates. While crawling COT_{net} for *A. brasiliana* falls within the range of other marine gastropods, its swimming COT_{net} is less than that of swimming crustaceans and falls more in line with swimming fish and some jetting invertebrates (Fig. 6).

It should be noted that only aerobic energy expenditure was measured in this study, and if *A. brasiliana* uses substantial amounts of anaerobic energy during swimming or crawling, then COT_{net} would be higher. However, anaerobic contribution to locomotion in aplysiids may in fact be low. Pyruvate reductase enzymes, those that convert pyruvate into lactate or into opines to maintain cellular NAD^+ levels and thus maintain glycolysis during hypoxia or anoxia, have been measured in a variety of opisthobranchs (Livingstone et al., 1983, 1990; Sato et al., 1993), and their concentrations are generally low. Moderate levels of lactate dehydrogenase (LDH) have been found in *Aplysia kurodai* and *A. juliana* (Sato et al., 1993), but none in *A. dactylomela* (Livingstone et al., 1990). Although LDH levels have not been measured in *A. brasiliana*, it is known to exhibit elevated levels of hemolymph lactate during experimental exposure to air, although whole animal levels remain constant (Bedford and Lutz, 1992). Thus it is unclear whether *A. brasiliana* could have high enough levels of anaerobic enzymes to contribute significantly to energy expenditure during exercise.

Scaling principles dictate that swimming animals will have an increasingly harder time to produce enough propulsion/lift to get them off the substratum as they grow. Many fish and invertebrates solve this problem by being neutrally buoyant, but *A. brasiliana* is negatively buoyant. However, large *A. brasiliana* have disproportionately larger parapodia than smaller individuals (Fig. 5) and this may allow them to generate more thrust to offset their increasing mass. Regardless of the exact mechanism by which *Aplysia* swim (sculling, jetting, hydrodynamic lift, or some combination of the three), an allometry in parapodial growth would be energetically favorable.

As expected from data for wing-beat frequency in flyers, and cycle frequency in invertebrate swimmers and jetters (Full, 1997), parapodial beat frequency was negatively related to mass in *A. brasiliana* (see also Carefoot and Pennings, 2003). As animals tired, swimming velocity decreased disproportionately more than did parapodial beat frequency (Fig. 3). Because tired animals closed the parapodia at the end of a beat much less tightly than did fresh animals, the data suggest that fatigue slows the animal by affecting muscular contraction more than by affecting beat frequency, which is regulated by neuronal oscillators in the pedal ganglia (von der Porten et al., 1980, 1982; McPherson and Blankenship, 1991) and modulated by neurons in the cerebral ganglion (Gamkrelidze et al., 1995).

A comparison of swimming gastropods suggests that a gradient in swimming efficiency and duration is coupled with the context in which swimming occurs. In *Tritonia diomedea*, a poor swimmer, swimming occurs only as an escape response to predators or noxious stimuli (Willows et al., 1973; Getting, 1975). Swim duration is brief (<1 min), and movement over the substratum occurs primarily through water motion (surge and currents) rather than directional swimming (Donovan, unpublished data). Another swimming opisthobranch, *M. leonina*, swims spontaneously for moderate durations (about 1 h), as well as in response to aversive stimuli (Watson et al., 2001; Lawrence and Watson, 2002; Caldwell and Donovan, 2003). *M. leonina* may rely on swimming to move short distances between kelp stipes (Ajeska and Nybakken, 1976) although longer distances are probably accomplished with the help of currents (Mills, 1994). In comparison, *A. brasiliana* swims spontaneously in the context of food-finding, for extended durations (up to several hours), in a highly directional manner (Hamilton and Russell, 1982; Carefoot and Pennings, 2003). Not surprisingly, this increasing importance of swimming as a part of routine daily activity is coupled with a sharp reduction in the cost of transport across these three taxa.

Given the low COT of swimming in *A. brasiliana*, why do all *Aplysia* species not swim? We consider two hypotheses. First, it may be that swimming carries risks that make it unfavorable in certain habitats. *A. brasiliana* occurs in shallow, expansive seagrass habitats, making it unlikely that

it would be carried into unfavorable habitats by long swims. In contrast, many other sea hares occur in rocky intertidal and subtidal habitats on steeply-sloping coasts. In these habitats, swimming might be disrupted by heavy surf, or might rapidly carry the animals into deep water away from their food sources. A rigorous evaluation of this hypothesis will require more detailed information on the habitats of swimming and non-swimming sea hares than is currently available. Second, it may be that the evolution of swimming would be favorable for all species but that it is an evolutionary bottleneck that has only occurred once (but see Medina and Walsh, 2000; Medina et al., 2001). Rigorously evaluating this hypothesis is difficult due to a lack of a complete molecular phylogeny of the genus *Aplysia*, and common misidentifications in the literature with respect to the ability of individual species to swim. Given these caveats, our current understanding of this issue is that swimming is an ancestral condition for *Aplysia* that has been lost and regained repeatedly (Medina and Walsh, 2000; Medina et al., 2001). Because all species of *Aplysia* retain parapodia, and most can open and close them in a flapping motion to flush water across the gills (a behavior that probably gives rise to the belief that more species swim than actually do), it is likely that the transition from a non-swimming to a swimming state in *Aplysia* would require only an increase in parapodial size and musculature. Similarly, Whiting et al. (2003) have argued that evolutionary transitions from a flightless to a flying state in insects may have been common, as has been the occurrence of flightlessness in birds.

In sum, we have shown that the cost of transport for *A. brasiliana* is over an order of magnitude less for swimming than crawling. The directional swimming method used by *A. brasiliana* is far more efficient than the non-directional swimming employed by some other gastropods, and is similar in efficiency to swimming by fish. More study is needed to understand exactly how *Aplysia* generates propulsion, and why all species of *Aplysia* do not swim. It is already clear, however, that swimming serves different purposes in different gastropod taxa. For some, like *T. diomedea*, swimming is an inefficient, rarely-used, escape response that is an interruption to, rather than a part of, regular behavior. In contrast, for *A. brasiliana*, swimming is a highly-efficient mode of locomotion that is fundamentally integrated with all other aspects of its behavior and ecology.

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