Reference: *Biol. Bull.* **203:** 144–151. (October 2002)

Swimming Behavior of the Nudibranch *Melibe leonina*

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Abstract. Swimming in the nudibranch *Melibe leonina* consists of five types of movements that occur in the following sequence: (1) withdrawal, (2) lateral flattening, (3) a series of lateral flexions, (4) unrolling and swinging, and (5) termination. *Melibe* swims spontaneously, as well as in response to different types of aversive stimuli. In this study, swimming was elicited by contact with the tube feet of the predatory sea star *Pycnopodia helianthoides,* pinching with forceps, or application of a 1 *M* KCl solution. During an episode of swimming, the duration of swim cycles (2.7 ± 0.2 s [mean ± SEM], $n = 29$) and the amplitude of lateral flexions remained relatively constant. However, the latency between the application of a stimulus and initiation of swimming was more variable, as was the duration of an episode of swimming. For example, when touched with a single tube foot from a sea star ($n = 32$), the latency to swim was 7.0 ± 2.4 s, and swimming continued for 53.7 ± 9.4 s, whereas application of KCl resulted in a longer latency to swim (22.3 ± 4.5 s) and more prolonged swimming episodes (174.9 ± 32.1 s). Swimming individuals tended to move in a direction perpendicular to the long axis of the foot, which propelled them laterally when they were oriented with the oral hood toward the surface of the water. The results of this study indicate that swimming in *Melibe,* like that in several other molluscs, is a stereotyped fixed action pattern that can be reliably elicited in the laboratory. These characteristics, along with the large identifiable neurons typical of many molluscs, make swimming in this nudibranch amenable to neuroethological analyses.

Received 7 November 2001; accepted 28 June 2002.

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Introduction

Swimming is a common form of locomotion in marine gastropods. It has been described for at least 47 species (reviewed by Farmer, 1970), and the neural mechanisms underlying swimming have been investigated in detail in four of these: *Pleurobranchaea californica* (Jing and Gillette, 1995, 1999), *Tritonia diomedea* (Willows *et al.,* 1973; Hume *et al.,* 1982; Getting, 1983), *Clione limacina* (Arshavsky *et al.,* 1985; Satterlie, 1985, 1991; Satterlie and Spencer, 1985; Satterlie *et al.,* 1985; Satterlie and Norekian, 1996), and *Aplysia brasiliana* (von der Porten *et al.,* 1982; McPherson and Blankenship, 1991a,b). In general, both rhythmic swimming and fictive swimming in molluscs are highly stereotyped and reliably expressed in intact and semi-intact preparations, as well as in isolated ganglia. These characteristics of molluscan swimming, combined with the suitability of their nervous systems for neurophysiological experimentation (Willows, 1965), have allowed scientists to elucidate some of the fundamental neural mechanisms responsible for producing swimming and other rhythmic behaviors (reviewed in Audesirk and Audesirk, 1985; Getting, 1989).

In opisthobranchs, five general types of swimming have been described, but only three types are common (Farmer, 1970): (1) parapodial or mantle flapping (as in *Gastropteron, Hexabranchus,* and *Aplysia*); (2) dorsoventral undulation (as in *Tritonia* and *Pleurobranchaea*); and (3) lateral bending (as in *Dendronotus*). Of the 47 swimming species listed by Farmer (1970), 21 swim by flapping either the mantle or some part of the foot, 5 swim using dorsoventral undulation, and 18 swim using lateral flexions. The latter is the most common type used by aeolidaceans and dendronotaceans. Lateral-bending swimming in these animals does not seem to propel them in a particular direction; rather, it appears as if swimming moves these animals into the water

column where the current may carry them away from potential predators.

Thompson (1976) hypothesized that swimming in opisthobranchs evolved as a means of escape. For most of the species studied, this seems to be a feasible explanation, since swimming can be elicited by noxious stimuli such as strong tactile stimulation or contact with a potential predator (Mauzey *et al.,* 1968; Edmunds, 1968; Farmer, 1970; Willows *et al.,* 1973; Page, 1993). Some animals, such as *T. diomedea,* apparently swim solely as a means of escape (Willows *et al.,* 1973). In others, such as *A. brasiliana,* which has no known predators, swimming is fairly directional and may serve a migratory role (Hamilton and Ambrose, 1975).

Studies of the opisthobranch *Melibe leonina* offer conflicting hypotheses as to the function and general characteristics of swimming behavior in this species. In one of the earliest papers on the subject, Agersborg (1921) states that the position of animals during a swimming episode may vary from dorsal aspect up to ventral aspect up. He further notes that swimming seems to be correlated with copulating masses of animals, suggesting that it may be a voluntary method for finding mates. In the same paper, Agersborg also refers to a method of "falling" through the water column, by completely relaxing the body musculature, which looks like "a feigned death." Hurst (1968) briefly describes the swimming behavior of this species as occurring only dorsal aspect up, and does not mention the ecological significance of the behavior. Farmer (1970) concluded that *Melibe* uses swimming to move from one kelp blade to the next. Most recently, Bickell-Page (1991; Page, 1993) suggested that swimming is an escape response. However, it is unclear which organisms in the natural habitat of *Melibe* might elicit escape swimming. Mauzey *et al.* (1968) and Bickell-Page (1991) have observed the sea star *Crossaster paposus* eating *Melibe,* and Ajeska and Nybakken (1976), Mauzey *et al.* (1968), and Bickell-Page (1991) have reported that several crab species, including *Pugettia producta,* will capture and eat *Melibe.* In contrast, it has also been reported that several species of sea stars avoid *Melibe,* presumably because they find the secretions of its repugnatorial gland repulsive (Ajeska and Nybakken, 1976; Bickell-Page, 1991). One goal of this study was to determine if *Melibe* would swim in response to these potential predators, which would indicate that one function of swimming in this species is escape.

In this paper we present results from three types of experiments concerned with swimming behavior in *M. leonina.* First, we analyzed swimming in 29 animals to enhance our understanding of the behavior and to quantify the various components of the swim. Second, we sought to determine the types of stimuli and potential predators that elicit swimming. Third, we assessed the movement of animals through the water column to determine whether swimming propels animals in random directions or predictable ones. We found that swimming in *Melibe* is a stereotyped rhythmic behavior that is most readily elicited in the laboratory by touching animals with the tube feet of the predatory sea star *Pycnopodia helianthoides.* Furthermore, what appears to be random motion during swimming has a fairly predictable directional component, with an animal typically moving in a path perpendicular to the long axis of its foot. These studies clarify some controversial issues concerning the swimming behavior of *M. leonina* and lay the framework for the neurophysiological studies presented in the subsequent paper (Watson *et al.,* 2002).

Materials and Methods

Animals

Specimens of the nudibranch *Melibe leonina* (Gould, 1852) were collected during May and June of 1994 and January through June of 1995 in sheltered bays throughout the San Juan Archipelago, Washington. Collections were made by scuba divers and the animals were maintained in flow-through seawater tables, typically between 10 and 12° C, at the University of Washington's Friday Harbor Laboratories. Animals were provided with blades of eelgrass (*Zostera marina*) to crawl upon, and they fed on planktonic crustaceans from the unfiltered water supply, supplemented with *Artemia* nauplii twice weekly.

Analysis of normal swimming

We analyzed the progression of a complete bout of swimming (from initiation to termination) in 29 animals. Each animal was placed in a 50-l aquarium with a small amount of eelgrass. Swimming was initiated by using a 3-ml syringe without a needle to apply 1 ml of 1 *M* KCl to the skin of either the oral hood or body wall. Not every animal responded to the salt stimulus. Our analyses are based on the 29 animals that swam. The following parameters were then measured: (1) latency between application of the stimulus and initiation of the swim; (2) swim duration; (3) number of complete swim cycles in each swimming episode; (4) average swim cycle duration (duration of an episode divided by the number of swim cycles); and (5) direction (right or left) of the first and last flexions. Finally, for one animal, we measured the duration of each individual swim cycle from a videotape of a complete swim episode. All averages are presented as the mean ± SEM.

To assess the magnitude of lateral flexions throughout the course of a swim, three animals were videotaped while they swam in place and the tapes were digitized for measurement of flexion angles. Two loops of 4-0 surgical silk were attached to the middle of the body wall of each animal, one on either side, at the point where their body pivots during swimming. After one day of recovery, animals were individually suspended by these loops, ventral aspect up, in an

acrylic plastic chamber. The chamber was supplied with a continuous flow of natural seawater so that it remained at 10–12° C. A video camera was mounted above the chamber, and the output of the camera was recorded onto videotape. Swimming was induced by dislodging the foot of the animal from its attachment to the surface tension of the water in the chamber. The video recordings were digitized, one frame/second, and version 1.55 of the public domain NIH Image software program (developed at the U.S. National Institutes of Health and available on the Internet at ⟨http://rsb.info.nih.gov/nih-image/⟩) was used to measure changes in the angle of the portion of the body anterior to the pivot point, relative to the region of the body posterior to that point.

Stimuli that elicit swimming

Different stimuli were applied to animals to determine their effectiveness in eliciting swimming behavior. The stimuli included (1) pinching of the cerata with self-closing forceps; (2) prodding of the foot with a glass rod; (3) application of a 1 *M* KCl solution to the oral hood or body wall; and (4) presence of, or contact with, potential natural predators (sea stars—*Pycnopodia helianthoides, Henricia leviuscula, Pisaster* spp; crabs—*Cancer magister, Scyra acutifrons, Oregonia gracilis, Cancer productus;* and an anemone found on eelgrass—*Epiactus prolifera*). In each experiment, the stimulus was given at time zero, and the latency to swim and the duration of any ensuing swim episodes were recorded. In the sea star contact experiments, a single tube foot was excised from a live sea star, held in self-closing forceps, and brought into contact with the back of the oral hood of a specimen of *Melibe.*

To determine if certain animals commonly found in the natural habitat (eelgrass beds) of *Melibe* were potential predators, we performed a series of predation experiments. Individual *Pycnopodia, Epiactus,* or crabs were placed in a 50-l aquarium with flow-through seawater. Then one specimen of *Melibe* was placed in the tank, which also contained a small amount of eelgrass, and left there for 24 h. Every 6–8 h the nudibranch was examined for evidence of an attack. Three trials were carried out with each potential predator.

Direction of swimming

The movement of *Melibe* through the water column during a swimming episode was determined for seven animals. The goal of this study was to test the hypothesis that this species moves ventrally, in a direction roughly perpendicular to the long axis of the foot. Individual nudibranchs were induced to swim in a 50-l aquarium that had *x*-*y* coordinates drawn on a clear plastic cover placed over the top and on one side. Each animal was placed on a blade of eelgrass that was located in the center of the tank and secured to the bottom, then induced to swim using a brief touch on the back of the oral hood with a single sea star tube foot. The tube foot had been excised from a live *Pycnopodia* with fine scissors and was held with a pair of self-closing forceps. A line representing the orientation of the foot was marked on the *x*-*y* grids at 5-s intervals. These lines were then plotted in two dimensions and used to calculate the predicted position of the midpoint of the foot at successive time intervals. The "variance angle" (the angle between the actual and predicted position of the foot at the next time point) was then calculated for each 5-s time interval, averaged, plotted using polar coordinates, and compared to the predicted path.

Results

Analysis of normal swimming

Although *Melibe* is occasionally observed swimming near the ocean surface (Mills, 1994; pers. obs.), typically it is found attached to eelgrass or kelp blades. *Melibe* swims spontaneously, as well as when forced off the substrate or exposed to a noxious stimulus. Its swimming behavior is characterized by slow, rhythmic, lateral flexions that last for 1–2 s (2–4 s for a full swim cycle). During each flexion, the body bends laterally into a shape resembling the letter C, with the oral hood approaching the tip of the tail (Fig. 1). A digitized video of swimming can be viewed at the following web site: http://zoology.unh.edu/faculty/win/Melibe/melibeswimming.htm.

The overall organization of a swim episode can be broken down into five basic components, some of which are illustrated in Figure 1. When not swimming, *Melibe* is often found attached, by means of its flattened foot, to a vertical substrate, such as a blade of eelgrass, with oral hood open and cerata extended (Fig. 1, top). A spontaneous swimming episode begins with closure of the oral hood and release of the foot, usually starting with the anterior portion, from the substrate (component 1—withdrawal). Once in the water column, the bottom of the foot is rolled medially, the body is compressed laterally, and the cerata are extended dorsally (2—lateral flattening). Swimming movements consist of an alternating lateral-bending, or flexion, of the body that brings the closed oral hood in close proximity to the tail (3—flexions). These rhythmic flexions may continue for a period of a few seconds to over an hour. The conclusion of a swim episode is preceded by the anterior tip of the foot unrolling and "probing" for an appropriate substrate on which to settle (4—unrolling and swinging). A swim episode is terminated (5—termination) when the anterior foot comes in contact with a suitable substrate and the animal uses its flattened foot to attach to it. Termination does not necessarily occur during the first encounter of the foot with a substrate; often the animal will make multiple contacts before ceasing to swim. Moreover, animals will occasion-

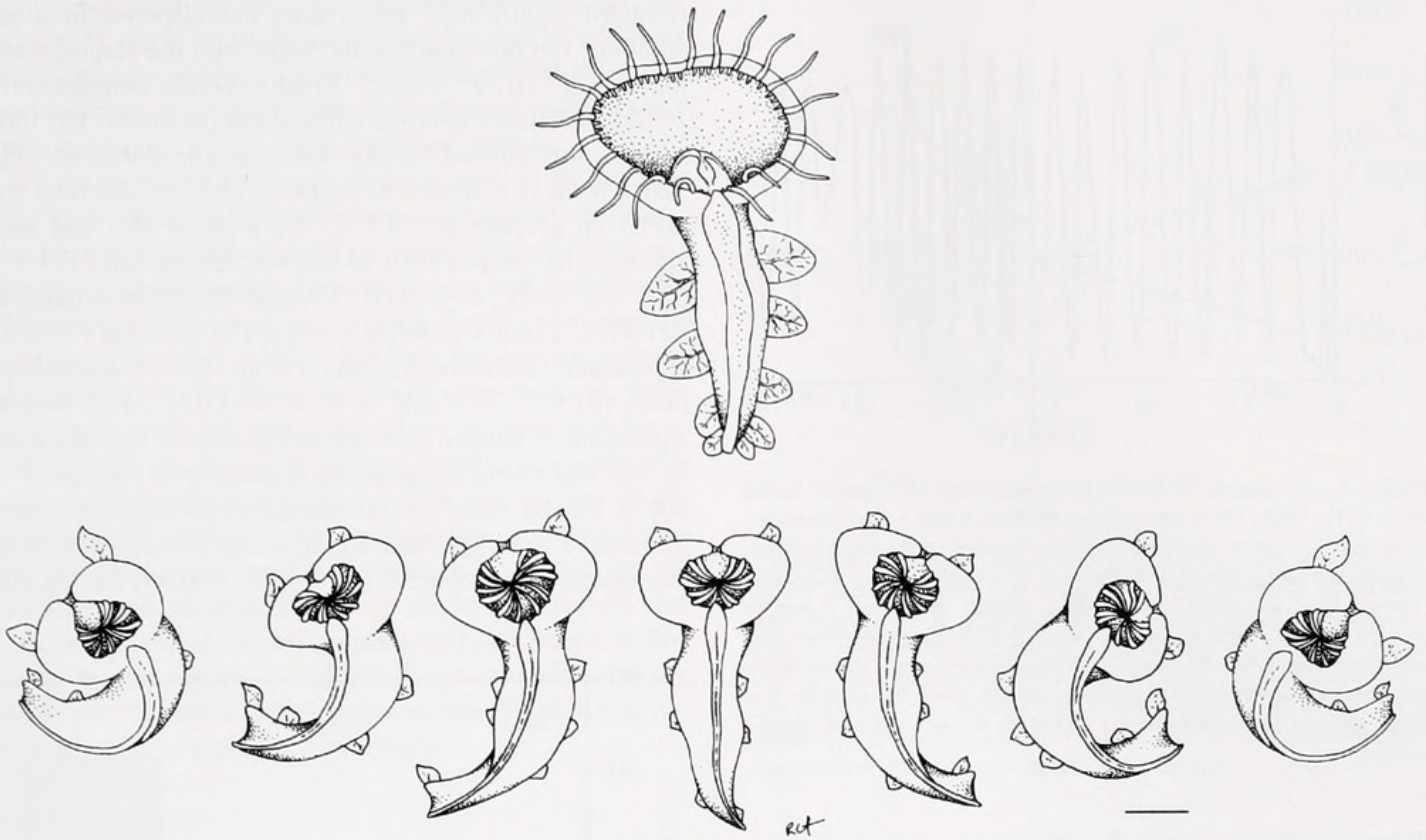

Figure 1. Illustrations of *Melibe leonina* at rest (top) and swimming (all ventral views). The sequence of drawings on the bottom show 50% of a swim cycle, as the nudibranch goes from being fully flexed to the right, to fully flexed to the left. Note the slight twisting of the posterior portion of the foot, which creates a sculling motion that propels animals in the ventral direction. Scale bar = 1 cm.

ally stop swimming in the water column, without contact of the foot with a substrate.

The flexions involved in the swim are not equivalent along the entire length of the body. In addition to the lateral flexions, there is also a concurrent twisting of the posterior part of the body, so that the foot becomes the leading edge during each lateral flexion (Fig. 1). This "sculling" motion provides a propulsive force that pushes water dorsally and moves the animal in a ventral direction, much as the sculling movements of the wings of the pteropod *Clione* cause it to move in the anterior direction (Satterlie *et al.,* 1985). This sculling movement was originally described by Hurst (1968), but no further mention of it has appeared in the literature on *Melibe.* The combination of lateral bending of the entire body and dorsal twisting of the foot typically propels the animal in the ventral direction. If the animal is oriented with its oral hood toward the surface, swimming will propel it in a lateral direction.

Although some aspects of swimming are quite variable in *Melibe,* the duration of each swim cycle, the magnitude of rhythmic lateral flexions, and the instantaneous swimming velocity are all very consistent during a swim episode. For example, in a single swim episode lasting 58 s, the average duration of each swim cycle was 2.03 ± 0.03 s, with no appreciable variation throughout the course of the swim. In 29 different animals, the average duration of a swim cycle was 2.7 ± 0.2 s. The magnitude of the lateral flexions was also quite consistent throughout a swim episode (Fig. 2). Other than the first and last few flexions in the swim episode, the contractions of the body in both directions were similar in amplitude for most of the episode.

In contrast to the stereotyped swimming escape response of *Tritonia,* where the duration of the swim and the direction of the first and last swimming flexion can be reasonably predicted (Willows *et al.,* 1973; Hume *et al.,* 1982), the swims of *Melibe* show little consistency in those parameters. Fifty-eight percent of the animals tested began with a left flexion (n = 29), and 53% of the animals finished a swimming episode with a flexion to left. The variation in the duration of swim episodes was also quite large. The mean swim duration in response to a salt stimulus was 174.9 ± 32.1 s (n = 11; data were taken from the 11 animals that swam, out of the 49 tested with a salt stimulus); some animals swam for only 33 s, but others continued for as long as 1546 s (25.7 min).

One of the unique features of swimming in *Melibe* is the motionless floating behavior which Agersborg (1921) referred to as "feigned death." During these floating events the animals lie in one place, dorsal aspect up, with the cerata inflated and spread parallel to the surface of the water; they

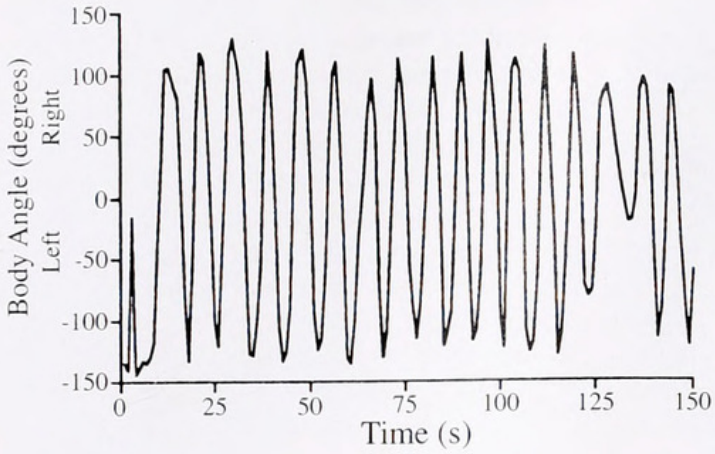

Figure 2. Swimming in *Melibe* is characterized by vigorous lateral flexions of the body. These motions are repeated without significant variation in timing or amplitude throughout the duration of the swim episode. This graph is a plot of the angle of the body of a single tethered individual, as viewed from the ventral side, during slightly more than 2 min of swimming. Animals were induced to swim by dislodging them from their attachment to the surface tension of the water. In the example shown in this figure, the animal was slightly flexed to the left when the stimulus was applied, and also remained slightly flexed to the left when it stopped swimming.

will occasionally remain in this position for several minutes. During a separate set of long-term swimming experiments (60–90 min), during which the animals were not allowed to attach to any substrate, these pauses occurred every 10–20 min.

Stimuli that elicit swimming

To determine what external factor probably causes *Melibe* to swim in its natural habitat, and how to reliably stimulate swimming in the laboratory, we screened a number of possible noxious stimuli, including pinches with forceps, salt (KCl), and contact with several different putative predators. There was a significant effect of these treatments on the tendency of *Melibe* to swim, with some treatments being more effective than others ($P < 0.001$, G-test for independence). Of the three stimuli, the touch of the predatory sea star *Pycnopodia* yielded the most reliable response (Fig. 3A; 62% of the 32 animals that were touched swam, $P < 0.0001$ Fisher's exact test, comparing sea star responses to pooled KCl and pinch responses). In fact, a very brief (<1 s) touch with an individual *Pycnopodia* tube foot was usually sufficient to elicit a swim. This finding contrasts with an earlier report that "*M. leonina* rarely swim following sea star contact" (Page, 1993). Single pinches to a cerata, as well as trains of pinches, caused rapid escape crawling but rarely swimming (5% swam, $n = 20$). A salt solution (1 ml of 1 *M* KCl) applied to the skin of the head elicited swimming in 22% of the trials ($n = 49$).

Of the three stimuli that were found to be most effective, contact with sea stars elicited a rapid escape response that occurred significantly faster than the response to a salt stimulus, but not significantly faster than the response to a pinch (Fig. 3B; $P < 0.05$ Kruskal-Wallis nonparametric ANOVA, Dunn's multiple comparison post-test). The three stimuli also elicited swims with variable durations (Fig. 3B). Swims in response to contact with a sea star tube foot lasted an average of 53.7 ± 9.4 s ($n = 20$) and were significantly shorter than swims elicited by salt (174.9 ± 32.1 s, $n = 11$, $P < 0.01$, Kruskal-Wallis nonparametric ANOVA, Dunn's multiple comparison post-test), but not significantly different in duration from swims triggered by a pinch (91.0 ± 76.9 s, $n = 6$, $P > 0.05$). There was no significant difference between the duration of swims elicited by salt and swims triggered by a pinch with forceps ($P > 0.05$). Finally, there was no correlation between the latency to respond to a particular stimulus and the duration of the subsequent swim episode ($r^2 = 0.05$). However, animals

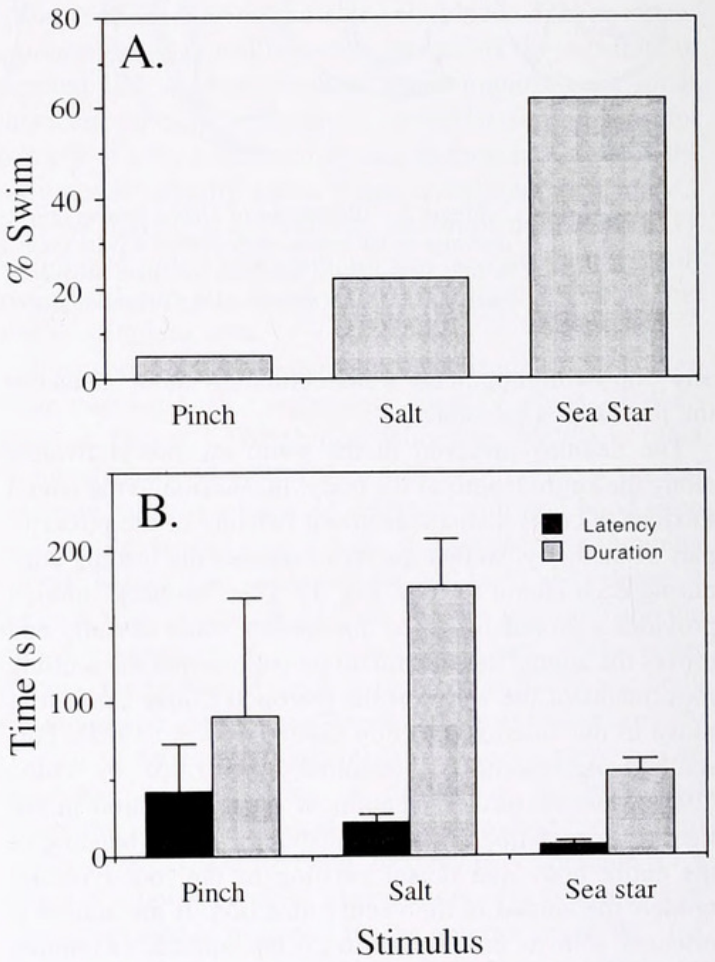

Figure 3. The influence of various stimuli on swimming behavior in *Melibe*. Animals were exposed to three different stimuli (pinch, $n = 20$; salt, $n = 49$; tube foot of the sea star *Pycnopodia*, $n = 32$) and their tendency to swim (panel A, % that swam), latency to swim (Panel B, time from stimulus to initiation of swim), and duration of the swim episode (Panel B) were measured. The sea star stimulus was significantly more effective than the other stimuli. Animals touched with sea star tube feet responded very rapidly and consistently in comparison to those given the other stimuli.

often swam multiple times in response to a single sea star contact. If these multiple swims are viewed as one long swim episode, then in general, "stronger" stimuli (sea star>salt>pinch) caused animals to respond more quickly and swim longer compared to "weaker" stimuli.

All the crabs and anemones tested, as well as the sea stars other than *Pycnopodia,* elicited no responses at all in *Melibe.* Neither animal seemed to take any notice of the other's presence. When contact between crabs and nudibranchs occurred, the nudibranch would often simply crawl over the carapace of the crab without incident. No contact between the anemone and the nudibranch was ever observed. Some nudibranchs were left with crab and sea star predators for up to 48 h, with no signs of predation. Finally, in a number of cases, nudibranchs were placed on the oral surface of potential sea star predators, and no ingestion occurred. However, we did not control for the state of hunger of the test predators, and on other occasions we have observed both *Pycnopodia* and anemones eating small specimens of *Melibe* in the laboratory. In addition, Ajeska and Nybakken (1976) have reported that *Pugettia,* a crab found in California kelp beds, is a predator of *Melibe.*

Direction of swim

Preliminary observations indicated that, when swimming, *Melibe* moved in a ventral direction, perpendicular to the long axis of the foot. To test this hypothesis, we analyzed the instantaneous swimming direction of seven animals, in 5-s intervals, as described in the Materials and Methods. Five of these animals moved, on average, in a direction that varied less than one standard deviation (14°) from the predicted direction (90° from the long axis of the foot). The variance angles of the other two animals were only slightly different than predicted (Fig. 4). These data support the hypothesis that the general direction of movement, from one swimming flexion of *Melibe* to the next, can be predicted if the orientation of the foot is known. This prediction is most accurate after the first two swimming flexions, which tend to propel the animal upward. Subsequently, most movement generated by an individual flexion is in a plane that is perpendicular to the long axis of the foot. Therefore, if an animal is positioned vertically in the water column, with its oral hood toward the surface, as it is often found on blades of eelgrass (pers. obs.), swimming would most likely move it in a lateral direction.

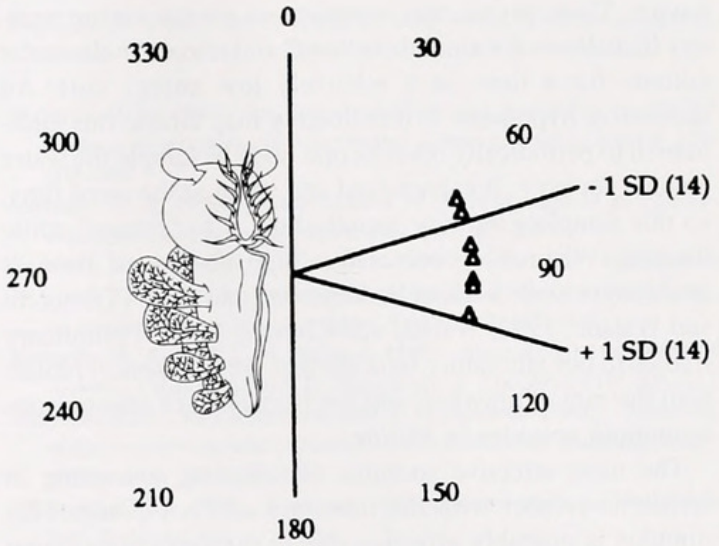

Figure 4. A comparison of the swimming direction of a specimen of *Melibe* with a predicted path (perpendicular to the foot). Seven animals were observed for the duration of an induced swim, and the angle of travel, relative to the long-axis of the foot, was measured at 5-s intervals. An angle of 90 degrees represents movement in a direction perpendicular to the long axis of the foot. The average angles were calculated and plotted for each animal. The average angle traveled by five of seven animals fell within one standard deviation of the prediction (=14 degrees).

Discussion

In this study we examined the swimming behavior of *Melibe leonina,* from initiation, elicited by a variety of stimuli, to termination, marked by reattachment to a suitable substrate. As in other lateral-bending swimmers (Agersborg, 1922), and many other swimming molluscs (Thompson, 1976), the behavior is elicited most reliably by noxious stimuli, such as contact with the tube feet of the predatory sea star *Pycnopodia.* It is also very stereotyped in terms of the consistent rhythmic flexions used to propel the animals through the water.

Melibe is rarely observed swimming in its natural eelgrass and kelp habitats, even though potential predators such as *Pycnopodia,* anemones, and crabs are present (Ajeska and Nybakken, 1976; unpubl. obs.). One explanation for this apparent low frequency of swimming might be that *Melibe* rarely encounters predators. This nudibranch tends to situate itself near the distal portions of eelgrass and kelp blades, where large sea stars and crabs are rarely found. *Melibe* may also have less of a tendency to swim where currents are strong, because swimming animals have a high probability of being carried away from their preferred habitat, and potential mates, by these currents. The high density of swimming and floating individuals observed intermittently at considerable distances from local eelgrass beds suggests that other factors, besides predators, may also trigger swimming in *Melibe* (Mills, 1994). Farmer (1970) suggested that these animals might use voluntary swimming episodes to move from one kelp blade to another, and we have observed spontaneous bouts of swimming on many occasions in the laboratory. Voluntary swimming may also be a means of dispersal, which would allow mixing of the gene pools from spatially isolated populations inhabiting eelgrass beds located several kilometers from each other (Mills, 1994).

A unique feature of swimming in *Melibe,* in comparison with other swimming molluscs, is motionless floating be-

havior. These pauses may represent an energy-saving strategy that allows the animals to "rest" and remain in the water column for a time, at a relatively low energy cost. An alternative hypothesis is that floating may enable this nudibranch to periodically open its oral hood to sample the water column for prey. It cannot feed and swim at the same time, so this sampling activity would allow it to "forage" while floating. When it encountered a high density of food, it could stop, seek a suitable substrate, and feed (Trimarchi and Watson, 1992; Watson and Chester, 1993). Preliminary studies in our laboratory indicate that prey (*Artemia*) reduce both the rate of crawling and the frequency of spontaneous swimming episodes in *Melibe*.

The most effective stimulus for eliciting swimming in *Melibe* is contact with the tube feet of *Pycnopodia*. This stimulus is probably effective due to the surfactants found on the tube feet of certain predatory sea stars (Mauzey *et al.*, 1968; Mackie, 1970). Oddly enough, we rarely observe sea stars attacking an adult *Melibe*. Page (1993) suggests that sea stars avoid *Melibe* because its repugnatorial glands, located throughout the epidermis, release a chemical that renders it repulsive to predators. These glands do not mature until the animals are 4–7 weeks old, and sea stars do attack and consume younger individuals. It is interesting that even though their repugnatorial glands help deter potential predators, mature specimens retain their tendency to escape when they sense the presence of certain sea stars.

The direction that *Melibe* travels during a swim appears to be random, upon casual observation. However, certain features of the path taken during a swim episode are fairly predictable. When this nudibranch starts to swim, it first releases the anterior part of its foot from the substrate. Then, no matter what the initial orientation of the animal is, its head moves toward the surface and the first few lateral flexions tend to move its body in the anterior direction. Once an individual has "pushed off" and is in the water column, the combination of lateral flexions and twisting of the posterior portions of the foot and tail region creates a "sculling" motion that reliably propels it along a plane perpendicular to the long axis of the foot (Fig. 4). Thus, although its swimming behavior has less of a directional component than seen in some molluscs that use parapodial flapping, such as *Aplysia brasiliana* and *Clione, Melibe* appears to have more control than the animals that use dorsal-ventral flexions, like *Tritonia* and *Pleurobranchaea*. This raises the question of whether *Melibe* has the ability to seek out its preferred habitats or potential mates, or whether it attempts to move laterally from one eelgrass or kelp blade to another. Certainly, in the laboratory, it swims spontaneously, especially during the night (Watson and Newcomb, unpubl. obs.), and our working hypothesis is that it uses swimming both as a response to predators and as a means of intermittent locomotion.

According to Audesirk and Audesirk (1985), three criteria must be fulfilled for a behavior to be useful in neuroethological studies: reliability, robustness, and stereotypy. All of these criteria are characteristic of the swimming behavior of *Melibe leonina*. It can be reliably initiated in the laboratory with natural stimuli or a salt solution. The robustness and stereotypy are illustrated in Figure 2, which shows that over the time course of a swim, the flexion amplitude and frequency do not change significantly. Furthermore, *Melibe* is amenable to electrophysiological investigations, as are many other opisthobranch species, because it has large, identifiable neurons, and impulses from these neurons can be recorded both in swimming, semi-intact animals and in isolated brains (see companion paper, Watson *et al.*, 2002). Finally, in *Melibe*, relatively few higher order interneurons constitute the swim central pattern generator, so a very thorough neuroethological understanding of the behavior is possible (Watson *et al.*, 2001).

Acknowledgments

We thank A. O. Dennis Willows and Glen Brown for their advice during the experimental phases of this study and for providing critical comments on this paper, Eric Abrahamson and Jen Wishinski for their help with several behavioral experiments, Jim Newcomb and Stuart Thompson for constructive comments on the manuscript, and the entire staff of FHL for their continuous support and assistance with all phases of the work. This study was supported by Center for Marine Biology grants and Summer Teaching fellowships to K.A.L. through the University of New Hampshire and an NIH grant to W.H.W. It is contribution number 386 of the Center for Marine Biology/Jackson Estuarine Laboratory series.

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