

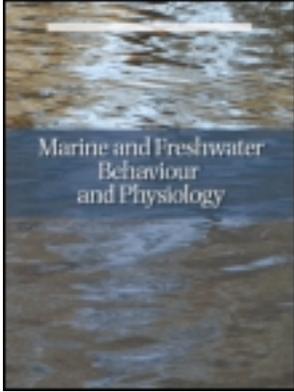
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PHYSIOLOGICAL AND BEHAVIORAL STUDIES OF ESCAPE RESPONSES IN CALANOID COPEPODS

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Electrophysiological techniques have been applied to monitoring sensory discharges from the first antennae of calanoid copepods. Extracellular nerve impulse traffic from both mechanoreceptors and putative chemoreceptors has been recorded. The first antennae of some, but not all, calanoid groups possess "giant" mechano receptive axons generating very large (mV) extracellular signals. There are two such giant antennal mechano receptors (GAMs) innervating setae of each distal tip. These are sensitive to small (< 10 nm) controlled hydrodynamic disturbances, including abrupt displacements and sinusoidal vibrations with frequencies up to and exceeding 2 kHz. Behavioral studies show that escape "jumps" can be triggered in *Labidocera madurae* by the same types of disturbances. Sensitivities as low as 4 nm were observed at frequencies of ca. 900 Hz. Behavioral sensitivities are similar to those measured physiologically and suggest that firing of the GAMs is capable of triggering escape behavior, perhaps even with a single nerve impulse.

KEY WORDS: Mechanoreception, hearing, crustaceans, neuroethology

INTRODUCTION

Predation pressure in the pelagic environment is high. Zooplankton, including pelagic copepods, are preyed upon by many fish species, pelagic invertebrate predators (cnidarians, ctenophores, chaetognaths, copepods), and benthic predators (corals, sea anemones, barnacles). Copepods have a characteristic escape jump, which can effectively lower predator capture efficiencies (e.g., Browman *et al.*, 1989; Trager *et al.*, 1994). Involvement of sensory systems in this behavior would be expected to include detection of a predator, discriminating it from irrelevant signals, and localizing it. Behavioral studies on copepods have shown that detection of approaching predators can occur at a distance (e.g., Strickler, 1975; Kerfoot *et al.*, 1980; Browman *et al.*, 1989). Predator avoidance behaviors, such as escape "jumps", can be elicited by mechanical stimulation (Haury *et al.*, 1980; Gill, 1985; Gill and Crisp, 1985; Costello *et al.*, 1990). Copepods can detect and react to both predatory lunges (e.g. Drenner *et al.*, 1978; Wright and O'Brien, 1984; Kils, 1992) and flow fields of predators (Tiselius and Jonsson, 1990; Yen and Fields, 1992). Understanding the mechanosensory substrate of this escape system would give substantial insight into the functioning of a key factor in copepod survival.

In deducing what information might be available to a copepod through its sensory systems, past work (e.g. Price *et al.*, 1988; Marine Zooplankton Colloquium I, 1989) has had to rely on assumptions derived from physiological studies on large, mostly benthic,

crustaceans from very different taxa (e.g. Laverack, 1963; Mellon, 1963; Tazaki and Ohnishi, 1974; Bush and Laverack, 1982; Ache, 1982; Sandeman, 1989). The issue of whether such assumptions could withstand direct scrutiny led to electrophysiological studies in our laboratory on sensory neurons in the first antennae of calanoids. Despite the small size of the animal, neural responses to controlled stimulation can be recorded successfully (Yen *et al.*, 1992; Lenz and Yen, 1993). This approach has opened the way to analyzing the physiological substrates of behavioral sensitivity of the animals to stimuli which are crucial to finding food and mates, and avoiding or escaping from predators. In focusing initially on the latter behavior, we find involvement of mechanosensors with properties differing markedly from those inferred from other crustaceans. We will review some of our past work in this area and present some recent results, especially relating to behavioral correlates.

METHODS

Physiological Experiments

In most of the physiological work to be described, past and present, electrical potential differences have been recorded between the body of a copepod drawn up into an insulating layer of oil, and the sea water bath into which one of the first antennae is left projecting. This leaves the distal half to two-thirds of the sensory setae, both mechanoreceptive and chemoreceptive, exposed to the sea water under fairly "natural" conditions. The recording point can be controlled by varying the position of the oil-water interface along the antenna. The experiments to be described have focused on mechano-sensitivity in these setae, although we have also succeeded in recording from presumptive chemosensory elements with the same configuration. Controlled mechanical stimulation was produced by somewhat different means in different experiments. The most recent of these used the methods of Gassie *et al.*, (1993), as diagrammed in Figure 1. A computer-controlled piezoelectrically-driven sphere generates near-field water disturbances the magnitude and direction of which can be calculated using classical dipole equations (Bergeijk, 1967; Kalmijn, 1988). In the particular cases we have employed so far, the sphere is located directly in front of the animal, with its axis of movement parallel to the shaft of the antenna. This produces an opposite movement of water at the antennal tip which can be calculated from the equation:

$$d = Da^3/2r^3 \quad (1)$$

where d is the displacement of the water at the antenna, D is the displacement of the sphere, a is the radius of the sphere, and r is the distance from the center of the sphere to the antenna. In the frequency ranges we have studied (100–2500 Hz) the effect of the boundary layer on the effective radius of the sphere is small and has been ignored.

In another electrophysiological approach, the antennal nerve of *Gaussia princeps* was exposed by cutting a small window in the ventral exoskeleton at the base of the antenna. A suction electrode was lowered through the opening and applied to the surface of the antennal nerve in this region. "En passant" recordings were thereby obtained of sensory nerve impulses ("spikes") travelling into the CNS. Electrical stimulation of motor axons in the same trunk was possible.

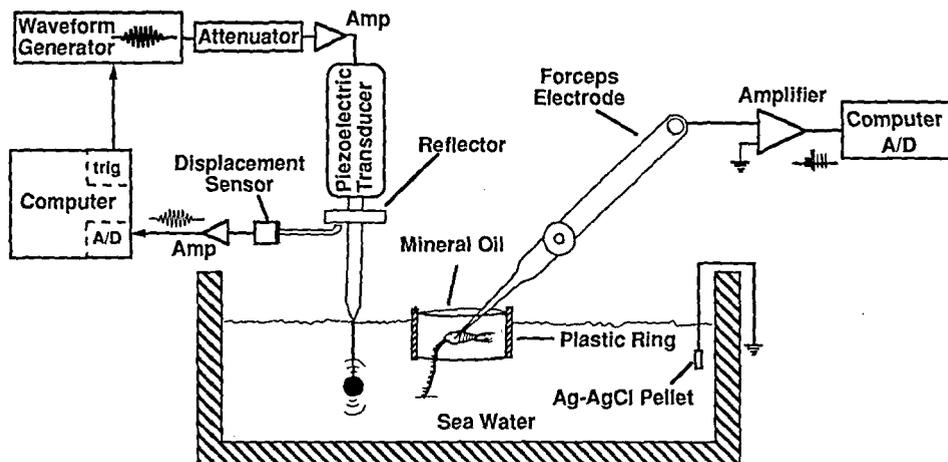


Figure 1 Stimulation and recording set-up. Pre-programmed stimulus waveforms are down-loaded into the generator. Output of the waveform is triggered by the computer, which then digitizes both the amplified electrical responses picked up with the forceps electrode and the output from the displacement sensor (modified from Gassie *et al.*, 1993).

Behavioral Experiments

L. maduræ were collected weekly using 2 to 5 min. subsurface horizontal tows (333 μm mesh plankton net) in shallow (< 10 m depth) nearshore areas off Oahu (Kaneohe Bay and Waikiki Beach). Animals were sorted into 2-l jars, maintained at room temperature (22–25°C), and fed *Artemia nauplii*. Only actively swimming adult *L. maduræ* females were used in the behavioural experiments. In order to maintain a fixed position in relation to the stimuli, copepods were tethered on their dorsal side to a stainless steel wire (diameter = 0.05 mm) with cyanoacrylate adhesive (Krazy Glue Pen). To avoid damage, copepods were transferred from the tethering dish to the experimental dish (10 × 10 × 5 cm) in a pool of seawater. There the wire was held in stainless steel forceps mounted on the micro-manipulator of the set-up described above. Copepod behavioral responses were observed with a dissecting microscope. The stimulus waveforms used in these experiments were a single abrupt water displacement (“trapezoid”) and a sine wave amplitude-modulated by a trapezoid function with a duration of 30 msec (“tone pip”; see Gassie *et al.*, 1993). Behavioral thresholds and best frequencies were determined by changing the amplitude and frequency of the stimulus. Successive stimuli were presented at 1 min intervals. Behavioral responses to stimuli were scored as either a “full escape”, an “antennal flick”, or as no response. A full escape behavior was defined as a response that caused vertical or horizontal motion of the copepod’s entire body while it was secured on the wire. Antennal flicks involved only the movement of usually one of the antennae away from the stimulus sphere and towards the cephalothorax. Other behaviors observed in response to stimulation, such as “trembling” (a flurry of feeding appendage movement), and “tail flicks”, were noted for future reference, but were scored as “no response” for the present experiments.

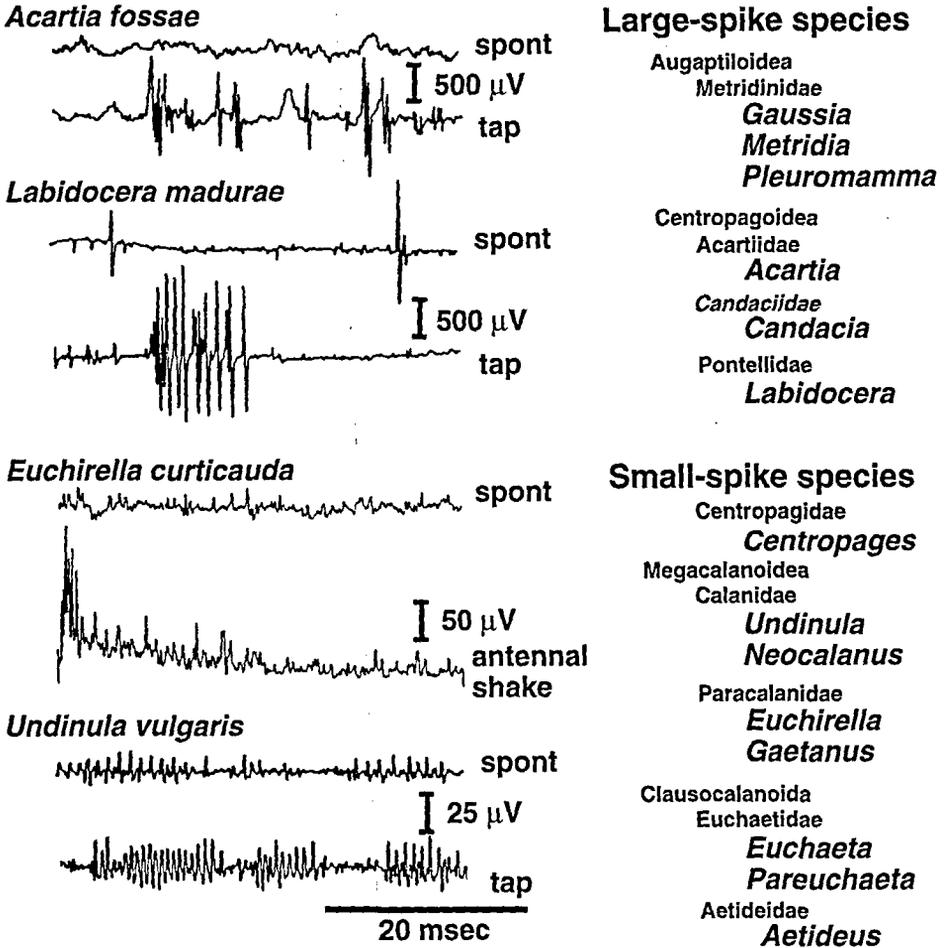


Figure 2 Nerve impulses found in the first antenna of various copepod species. Left: representative nerve impulse traffic from 4 calanoid species; Top trace of each pair: unstimulated (except by ambient noise levels); bottom trace: activity elicited by various forms of mechanical stimulation. Note the change in voltage scale between the top two and bottom two pairs. Right: current assessment of the presence or absence of GAM spikes in various species (modified from Yen *et al.*, 1992).

MECHANORECEPTIVE PHYSIOLOGY OF COPEPOD ANTENNAE: A BRIEF REVIEW

Giant Mechanoreceptor Spikes Are Present in Some Species

In the earliest experiments, we and our colleagues made a survey of neural activity in antennae of several species of calanoids (Yen *et al.*, 1992). Nerve impulse firing was highly sensitive to extraneous sources of mechanical stimulation, including tapping the

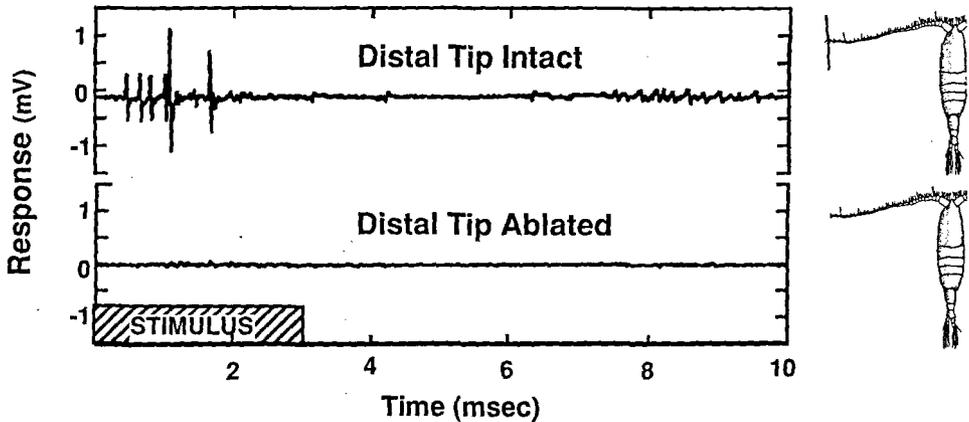


Figure 3 Giant spikes originate at the distal tip. Water movements (tone pip of 700 Hz, 7 nm amplitude) elicit GAM discharges when the distal tip of the antenna is intact (upper trace), but not after the terminal two segments are ablated (lower trace). *P. xiphias*. (modified from Lenz and Yen, 1993).

preparation table, touching the water surface or antennal setae, and even loud talking in the room. Examining the different species, a key characteristic of the activity was the presence of very large spikes (amplitudes up to 5 mV) in some animals (Figure 2A). Not all genera we have examined have exhibited these “giant” spikes (Yen *et al.*, 1992), so we have chosen to focus initial physiological work on those which do: *Labidocera*, *Acartia*, *Pleuromamma* and *Gaussia*. We have termed these “giant antennal mechanoreceptors” (GAMs). A key issue in the sensory ecology of zooplankton is how the sensory environment changes from habitat to habitat. This in turn should impact the “neuroecology” of the copepod: the adaptations of its nervous system to the ecological situation. Might the presence of GAMs represent such an adaptation? Figure 2 tabulates the known giant-spike and small-spike genera. Of those species we have found to possess the giant antennal receptors, several are open ocean forms, some inhabiting shallow and some quite deep regions, while others are bay species. Other species lacking GAMs occupy some of the same habitats. While clearly of interest, we have not succeeded in establishing a firm correlation between ecological conditions and type of mechanoreceptive units present. A better correlation appears to be made based on phylogenetic relationships. Giant spikes tend to be found in superfamilies assigned by Park (1986) an earlier evolutionary divergence (Augaptiloidea; Centropagoidea) and have not been found in superfamilies with later ones (Megacalanoidea, Clausocalanoida). The one possible exception shown in Figure 2 (*Centropages*) is based on a small sample (2) and needs to be reexamined.

Giant Spikes Originate at the Distal Tip

Several different types of experiments have shown directly the mechanosensitivity of copepod antennal setae. Figure 3, upper trace, shows an example of responses elicited by a sudden brief direct translation of the antenna (stationary water). Such responses disappear if the distal tip of the antenna is removed (Figure 3, lower trace: Lenz and

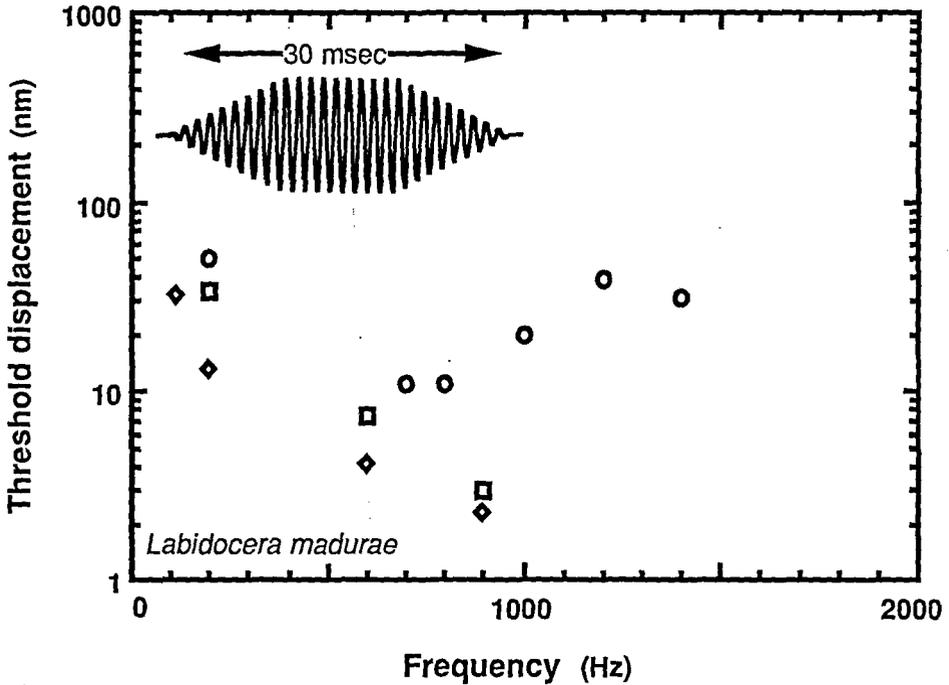


Figure 4 Threshold displacements for tone pips of various frequencies. Ordinate indicates (on a log scale) the peak-to-peak water displacement which elicits at least one spike in half of the trials of a series of 10 spaced 10 sec apart. Different symbols represent different animals. Data obtained in collaboration with Michele Solis.

Yen, 1993). Clipping (shortening) the long setae of the distal tips also eliminates or raises the threshold for firing of the most prominent units, which suggests a GAM origin in these setae (Lenz, unpublished observations).

Receptors Are Very Sensitive to Mechanical Stimulation

Perhaps the single most important issue for this type of mechanoreceptor is sensitivity. The giant units of the calanoids we have studied are exceptionally sensitive. For a variety of different stimulus forms, we find that displacements just sufficient to elicit an impulse can be in the nanometer range. Several studies using various methodologies all give similar results (Yen *et al.*, 1992; Lenz and Yen, 1993; Lenz, 1993). However, the magnitude of water movement needed to elicit a response depends significantly on the characteristics of the movement. One of the stimulus forms we use in exploring GAM sensitivity is the "tone-pip", a sinusoidal movement the amplitude of which is allowed to increase over 10 msec, is held constant for various lengths of time (typically 10 msec), then decreases to zero over the final 10 msec. This form is widely used in studies of auditory and hydrodynamic reception in other organisms, and permits performance comparisons with this work. In Figure 4, we show typical results of sensitivity measurements in *L. madurae*. In this experiment, a tone pip was adjusted in amplitude

until it elicited a single GAM spike in 50% of the trials. This magnitude was recorded as the threshold. The figure shows how threshold displacement varied as a function of frequency in the pip: sensitivity was relatively low at the lower (e.g. < 100 Hz) and higher (e.g. >1200 Hz) frequencies tested, and was greatest at intermediate values. Similar results have been found for *Pleuromamma xiphias* (Lenz and Yen, 1993). A "best" frequency around 800–1000 Hz is typical of the large-spike species we have examined so far.

NEUROETHOLOGICAL RESULTS

There Are Two Giant-spike Units in Each Antenna

One critical issue for the physiology is how many units with the highest sensitivity are present. Larger (decapod) crustaceans have typically thousands of exteroceptive mechanosensory neurons supplying the body and appendage surfaces, and the antennae and antennules (e.g. Laverack, 1968; Wiese, 1976; Tautz and Sandeman, 1980; Sandeman, 1989). Each first antenna of calanoid copepods, on the other hand, has around 111 (*Pleuromamma* females: Lenz *et al.*, this volume) and those confined to the distal tips number only a few (Weatherby *et al.*, 1994). While records such as those shown in Figures 2 and 3 appear to have several spikes of different amplitudes, careful analysis suggests that there are in fact only two units in most of the calanoids so far examined. We term the smaller amplitude unit "A" and the larger one "B." These two units had the lowest thresholds of any mechanoreceptive units observed under our experimental conditions. In most cases, the threshold for unit A was lower than that for unit B. Exceptionally large spikes, seen typically at the onset of a stimulus, can be accounted for as superpositions of firing in these two units. Figure 5A shows several records analyzed from this point of view. Insets above the middle trace show that superpositions can be resolved into A and B components: on an expanded time scale the two compound spikes labeled "A+B" in the trace are plotted with solid lines and computed superpositions of appropriately time-shifted A and B spikes (light dotted lines) are shown with dashed lines (barely visible in places). Previously published records from several species showing the "giant" spikes are consistent with two units (Yen *et al.*, 1992; Lenz and Yen, 1993). To date, these two units have proven to be the only giant spikes recorded at any point along the antennal shaft. They are present throughout the shaft, to near the distal tip, confirming the distal location of the primary sensors. These physiological conclusions are supported by work on giant-spike species for which electron microscopic cross sections of the antennal shaft are available. Typically two axon profiles are found with diameters distinctly larger than the rest in the same antennal cross section (Figure 4B; Weatherby, personal communication). While these "giant" axons have not yet been traced to specific sensory somata, the size distribution of axon diameters in the antennal innervation is consistent with the sensory physiological results.

Setal Position is Under Motor Control

Calanoid first antennae participate in escape reactions, being folded posteriorly to reduce drag and probably contribute to the forward propulsive force (Kerfoot *et al.*, 1980). One of the intriguing observations we have made with suction electrodes in

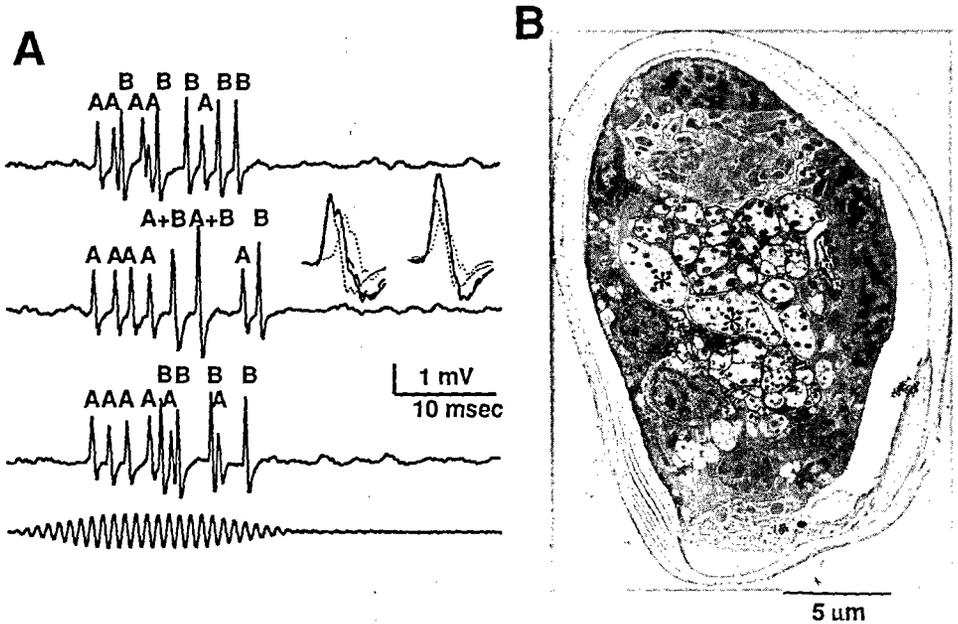


Figure 5 Two giant antennal mechanoreceptors are reidentifiable.

A. Three presentations of a tone-pip stimulus, showing the unit-analysis of the response in *P. xiphias*. Extra large spikes in the middle trace ("A+B") are consistent with a coincidence with superposition of A and B unit firing. Insets show these compound events on an expanded time scale (1.5 msec trace duration) with a dashed line showing the computed sum of individual A and B spikes (dotted lines). Spike A is delayed 0.27 and 0.08 msec, respectively, compared to spike B for the two compound events. Data obtained in collaboration with M. Solis.

B. Electron micrograph showing axon profiles in a cross section of the antennal nerve trunk, taken half way from base to tip in *L. madurae*. Note the two exceptionally large profiles, presumed to correspond to GAM action potentials (Courtesy of Tina Weatherby).

G. princeps (in addition to being able to record discharges to mechanical stimulation of sensory setae) is that single electrical shocks to the antennal nerve cause movements of antennal setae, and in particular, a dramatic rapid remotion of seta #8 (nomenclature of Weatherby *et al.*, 1994). The seta is brought to lie parallel to the antennal shaft, then rapidly returns to its rest orientation perpendicular to the shaft (Hartline and Weber, unpublished observations). In addition to moving the seta out of the way and contributing to streamlining, this particular movement might make a small contribution to forward propulsion.

Behavioral Properties Parallel Physiological Properties

Tethered *L. madurae* survived in good condition for the several hours of a typical

behavioral experiment, and in several cases were still in good condition and responding sensitively the following day. Some animals generated escape responses spontaneously, but in the better preparations these occurred rarely (several minutes between). At least 30 sec were permitted to elapse after a spontaneous escape response before testing thresholds.

Abrupt water displacements consistently and repeatedly elicited escape jumps. These displacements consisted of a short linear (constant velocity) ramp (< 1 msec duration) at the onset, then a constant value held for 25 msec. The return to zero was a ramp symmetrical to the onset. *L. madurae* responded to such abrupt water displacements of as little as 4 nm (Table 1). These responses showed no habituation to stimuli presented at 1 min. intervals. The probability of an escape response to a threshold level stimulus stayed constant even after 50 stimulus presentations.

L. madurae was more sensitive to oscillatory stimuli than to single abrupt displacements (Table 1). Threshold sensitivities to water displacements depended on sine wave frequency with a "best" frequency around 900 Hz (Figure 6). Escape responses were also elicited at all other frequencies tested (100–1800 Hz), but greater water displacements were required. Expressed in terms of peak water velocity, lowest thresholds ranged from 7 to 27 $\mu\text{m}/\text{sec}$ (16.8 ± 43 , mean \pm SEM). These threshold velocities correspond well to velocities required to trigger single sensory impulses in this species and in *P. xiphias* (Yen *et al.*, 1992; Lenz and Yen, 1993). Figure 6A and B plot behavioral thresholds vs frequency for tone pips eliciting escape jumps (Figure 6A: for peak displacements; Figure 6B: for peak water velocities). For comparison with 6B, Figure 6C replots the physiological data of Figure 4 in terms of velocity. Physiological and behavioral sensitivities are comparable. Thus, velocities of water movement of just a few microns per second are sufficient to trigger both GAM spikes and escape jumps. In both cases, the peak sensitivity to water displacement is near 1 kHz.

Table 1 Behavioral thresholds for two different stimulus waveforms
Magnitude of stimuli needed to trigger escape jumps in two *Labidocera Madurae* tested with both a 30 msec duration 900 Hz tone pip (Sine wave column) and an abrupt displacement of 25 msec duration and 0.1 msec rise and fall times (Trapezoid column).

	Sine wave		Trapezoid	
Animal	1	2	1	2
Displacement (nm)	8.6	2.2	4.3–6.4	15–22
Velocity ($\mu\text{m}/\text{sec}$)	24	6.3	43–64	150–220

DISCUSSION

The correspondence in the unusual movement sensitivity and frequency-dependence for stimuli triggering jumps with those triggering spikes in GAMs supports the hypothesis that disturbance detection by GAMs is one pathway leading to escape jumps. The employment of giant axons in the circuit is characteristic of an escape system for an invertebrate. It undoubtedly indicates an exceptionally high velocity of propagation (compared with other copepod neurons), and hence a minimal delay for the CNS to be informed of a triggering mechanical event. These are the conditions needed for a rapid escape response generated to a sudden and unexpected predatory

attack. The localization of the GAMs to the distal tips of the first antennae is consistent with behavioral studies using water jets and physical stroking of different parts of the body, which have found that stimulating the setae on the first antennae was most likely to elicit an escape response (Gill, 1985; Gill and Crisp, 1985). Perhaps most surprising in the comparison between physiology and behavior is the conclusion that the data are consistent with a single impulse in a GAM being sufficient to trigger an escape jump. In most escape systems, at least some sensory summation is required, involving multiple firings of a unit or multiple units firing, or both.

An important issue is identifying what natural stimuli lead to escape jumps via the GAM pathway. As noted by Yen *et al.* (1992), the special sensitivity to sudden stimuli exhibited by the GAMs is not especially appropriate for detecting the low-frequencies that must characterize flow fields around predators. On the other hand, predatory attack stimuli are sudden and brief in nature. When analyzed in terms of frequency components (Fourier decomposition), both sudden and brief stimuli are characterized by energy in the high frequencies. This situation would appear to fit qualitatively with our findings of high sensitivities (even expressed in terms of velocity) up to exceptionally high frequencies (1 kHz), as compared to benthic crustaceans (*Procambarus*, 0.1 μm , < 100 Hz; Wiese, 1976; *Cherax*, 0.2 μm , < 300 Hz; Tautz and Sandeman, 1980; *Euphausia*, 0.2 μm , < 500 Hz; Wiese and Marschall, 1990; Wiese, this volume). Lenz and Yen (1993) have discussed some of the uncertainties in this conclusion, however. Further, it is worth noting that Bleckmann *et al.*, (1991) studied frequency compositions of naturally occurring mechanical stimuli, including rapidly swimming fish, and found frequency spectra falling off well below the 1 kHz copepod cut-offs. The highest frequencies they found with significant energy (< 200 Hz) occurred in turbulent wakes, which would presumably be too delayed to serve as a useful warning signal. Frequency components in front of a lunging fish have not, to our knowledge, been measured quantitatively, but in the absence of such measurements, they might be expected to approximate those from rapid swims. Low frequencies might still trigger GAM-mediated escapes, but water displacements required would be well above those needed by a high-frequency stimulus. The lunge of a predatory copepod, which is potentially capable of very high velocities and accelerations (Strickler, 1985; Trager *et al.*, 1994), might generate the high frequency components to which the giant-spike species are especially sensitive. Certain other rapid attacks might generate high frequencies as well, but so far, no stimulus by a threatening situation with the high frequency components matching GAM sensitivity has been clearly identified. The morphological sophistication of the sensory apparatus (Weatherby *et al.*, 1994) and the exceptional characteristics of the physiology compared to benthic crustaceans argues against this being just a byproduct of building a mechanosensory seta, and implies a major adaptive advantage.

Escape reactions are found in most if not all calanoids. The GAM system, apparently, is not. Although we do not know from experimental results what the sensory architecture underlying escape reactions is for the "small spike" species (lacking GAMs), they presumably possess non-giant homologues of GAMs. Why do not all copepod species possess giant mechanoreceptors? Presumably the ecological/ethological needs for such receptors must be balanced against the significant energetic and structural costs of producing and maintaining such large axons. A much more extensive central projection might be one explanation for a large neuron, but there is no obvious behavioral difference that we have seen reported between giant-spike and small-spike species as might be expected from a major difference in wiring.

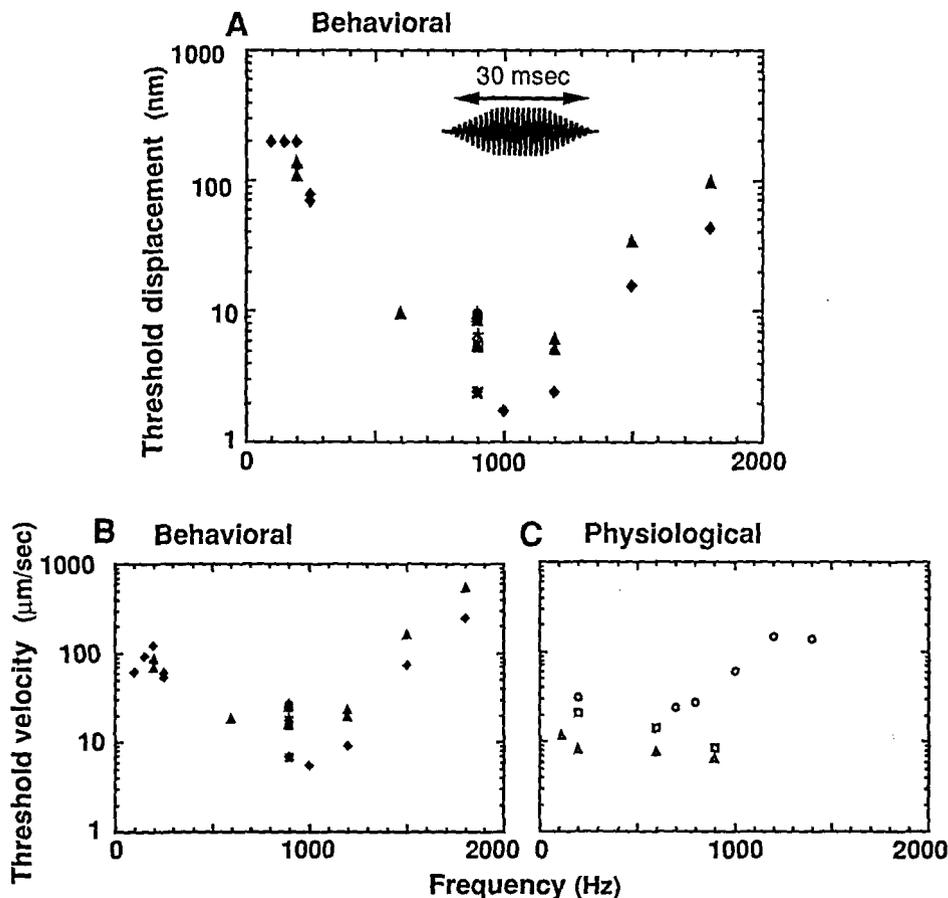


Figure 6 Comparison of behavioral and physiological thresholds.

A. Water displacement (ordinate; logarithmic scale) at different frequencies of a tone pip (abscissa) just sufficient to elicit an escape "jump" in 50% of the presentations.

B. Same data as A., plotted in terms of maximum water velocity.

C. Threshold velocities for tone pips which elicit a GAM impulse (data from Figure 5).

Minimization of errors in comparing timing of signals received by two antennae might be another advantage. However, in terms of time savings of a simple escape reaction, having axons that are *ca.* 50% larger in diameter than the rest (e.g. see Figure 4B) saves perhaps in the order of a millisecond (for an antenna 3 mm long and estimating conduction velocities for this diameter range of non-myelinated axons to be 1 m/s). For a copepod capable of escape velocities of 300 body lengths per second (Kerfoot *et al.*, 1980; Trager *et al.*, 1994), this would amount to less than a body length's advantage over a non-giant warning system. Such an advantage would be most critical in evading predators with capture volumes of dimensions comparable to the size of a copepod (e.g. another copepod), and less so for predators with large capture volumes.

A question is raised by the conclusion that there are only two giant receptor units: to which setae do these two belong? At this point, a clear answer cannot be given, but it is tempting to speculate that since clipping the long setae of the tip (in particular probably #8) appears to eliminate the giant units, and since each of the long setae (in particular #8) is dually innervated by mechanoreceptive cells, that it is seta #8 which is producing the high sensitivity large amplitude spikes (the next most likely candidate is seta #1). It is also of interest that this seta (#8) is the only one controlled by its own muscle, which is capable (as demonstrated in the suction electrode experiments on *G. princeps*) of rapidly remoting the seta. It seems likely that this motor capability is used in the escape behavior, although the significance of having the capability associated with the most sensitive receptors is not yet clear.

In our behavioral study, we observed responses to water velocities as low as $7 \mu\text{m}/\text{sec}$. This translates into a shear-rate sensitivity of around 0.002 s^{-1} for a 3 mm long antenna, much greater than that reported by Yen and Fields (1992) for *Acartia nauplii* escaping from *Temora longicornis* flow fields (0.8 s^{-1}). One contributor may be that distal setal mechanoreceptors do not develop until the last naupliar stage. It is also greater sensitivity than for adult *Pleuromamma* escaping suction-tube shears reported by Fields and Yen (this volume: 15 s^{-1}). Such flow fields, however, generate mostly low frequency stimuli to which GAMs tend to be insensitive.

Studies on copepod behavior in turbulent conditions have shown a decrease in escape responses over time (Costello *et al.*, 1990; Hwang *et al.*, 1994). This type of mechanical stimulation is likely to be very different from the discrete stimulus presentations given in Gill's (1985) and the present studies. Gill applied a supra-threshold stimulus to the antennal setae of *T. longicornis* at 1 min. intervals and observed no decrease in escape behavior over time. For a discrete fluid disturbance, our data indicate that, in addition to no decrease in the probability of a behavioral response, there is no change in mechanosensitivity if the stimulus occurs at 1 min. intervals.

Oceanic processes occur at many spatial and temporal scales, and the least studied ones are the micrometer and microsecond scales (*Marine Zooplankton Colloquium* 1989; Haury and Yamazaki, 1995). Our behavioral data indicate that calanoid sensory systems are detecting fluid disturbances at these scales. This further suggests that copepods are interacting with their environment and biological community on these micron scales. Processes occurring at these spatial and temporal scales need to be better understood in order to understand biological interactions in the planktonic community.

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References

- Ache, B.W. (1982) Chemoreception and thermoreception. In: *The Biology of Crustacea*, Vol. 3. (H. C. Atwood, D. C. Sandeman, D. H. Bliss, eds., Academic Press, New York. pp. 369–398.
- Bergeijk, W. A. van. (1967) Introductory comments on the lateral line function. In: *Lateral Line Detectors*, P. Cahn, ed., Indiana Univ. Press, Bloomington, IN. pp. 73–81.
- Bleckmann, H., Breithaupt, T., Blickhan, R. and Tautz, J. (1991) The time course and frequency content of hydrodynamic events caused by moving fish, frogs, and crustaceans. *J. Comp. Physiol.*, **168**, 749–757.
- Browman, H. I., Kruse, S. and O'Brien, W. J. (1989) Foraging behavior of the predaceous cladoceran *Leptodora kindtii*, and escape responses of their prey. *J. Plankton Res.*, **11**, 1075–1088.
- Bush, B. and Laverack, M. S. (1982) Mechanoreceptors. In: *The Biology of Crustacea*, Vol. 3. H. C. Atwood, D. C. Sandeman, D. H. Bliss, eds., Academic Press, New York. pp. 339–456.
- Costello, J. H., Strickler, J. R., Marrasé, C. Trager, G. Zeller, R. and Freise A. J. (1990) Grazing in a turbulent environment: Behavioral response of a calanoid copepod, *Centropages hamatus*. *Proc. Natl. Acad. Sci. USA.*, **87**, 1648–1652.
- Drenner, R. W., Strickler, J. R. and O'Brien, W. J. (1978) Capture probability: the role of zooplankton escape in the selective feeding of planktivorous fish. *J. Fish. Res. Bd. Can.*, **35**, 1370–1373.
- Gassie, D. V., Lenz, P. H., Yen, J., and Hartline, D. K. (1993) Mechanoreception in zooplankton first antennae: Electrophysiological techniques. *Bull. Marine Sci.*, **53**, 96–105.
- Gill, C.W. (1985) The response of a restrained copepod to tactile stimulation. *Mar. Ecol. Progr. Ser.*, **21**, 121–125.
- Gill, C. W. and Crisp, (1985) Sensitivity of intact and antennule amputated copepods to water disturbance. *Mar. Ecol. Progr. Ser.*, **21**, 221–227.
- Haurly, L. R., Kenyon, D. E. and Brooks, J. R. (1980) Experimental evaluation of the avoidance reaction of *Calanus finmarchicus*. *J. Plankton Res.*, **2**, 187–202.
- Haurly, L. R. and Yamazaki, H. (1995) The dichotomy of scales in the perception and aggregation behavior of zooplankton. *J. Plankton Res.*, **17**, 191–?
- Hwang, J.-S., Costello, J. H. and Strickler, J. R. (1994) Copepod grazing in turbulent flow: elevated foraging behavior and habituation of escape responses. *J. Plankton Res.*, **16**, 421–431.
- Kalmijn, Ad. J. (1988) Hydrodynamic and acoustic field detection. In: *Sensory Biology of Aquatic Animals*, J. Atema, R. R. Fay, A. N. Popper, W. N. Tavolga, eds., Springer-Verlag, New York, pp. 83–130.
- Kerfoot, W. C., Kellogg, D. C. and Strickler, J. R. (1980) Visual observations of live zooplankters: Evasion, escape and chemical defenses. In: *Evolution and Ecology of Zooplankton Communities*, W. C. Kerfoot, ed., New England University Press, Hanover, NH. pp. 10–27.
- Kils, U. (1992) The ecoSCOPE and dynIMAGE: Microscale tools for *in situ* studies of predator-prey interactions. *Arch. Hydrobiol.*, **36**, 83–96.
- Laverack, M. S. (1963) Responses of cuticular sense organs of the lobster, *Homarus vulgaris* (Crustacea). III. Activity invoked in sense organs of the carapace. *Comp. Biochem. Physiol.*, **10**, 261–272.
- Laverack, M. S. (1968) On the receptors of marine invertebrates. *Oceanogr. Mar. Biol. Ann. Rev.*, **6**, 249–324.
- Lenz, P. H. (1993) Vibration sensitivity in marine copepods In: *Sensory Systems of Arthropods*, K. Wiese, F. G. Gribakin, A. V. Popov and G. Renninger, eds., Birkhäuser Verlag, Berlin. pp. 423–430.
- Lenz, P. H. and Yen, J. (1993) Distal setal mechanoreceptors of the first antennae of marine copepods. *Bull. Marine Sci.*, **53**, 170–179.
- Marine Zooplankton Colloquium I. 1989. Future marine zooplankton research – a perspective. *Mar. Ecol. Progr. Ser.*, **55**, 197–206.
- Mellon, D. (1963) Electrical responses from dually innervated tactile receptors on the thorax of the crayfish. *J. Exp. Biol.*, **40**, 137–148.
- Park, T. (1986) Phylogeny of calanoid copepods. *Sylogus*, **58**, 191–196.
- Price, H. J., Paffenhöfer, G.-A., Boyd, C. M., Cowles, T. J., Donaghay, P. L., Hamner, W. M. Lampert, W., Quentin, L., Ross, R. M., Strickler, J. R. and Youngbluth, M. J. (1988) Future studies of zooplankton behavior: Questions and technological developments. *Bull. Mar. Sci.*, **43**, 853–872.
- Sandeman, D. C. (1989) Physical properties, sensory receptors and tactile reflexes of the antenna of the Australia freshwater crayfish *Cherax destructor*. *J. Exp. Biol.*, **141**, 197–217.
- Strickler, J. R. (1975) Swimming of planktonic *Cyclops* (Copepoda, Crustacea): Pattern, movements and their control. In: *Swimming and Flying in Nature* Vol. 2. T. Y.-T. Wu *et al.*, eds. pp. 599–616.

- Strickler, J. R. (1985) Feeding currents in calanoid copepods: two new hypotheses. In: *Physiological adaptations of marine animals*, M. S. Laverack, ed., Society for Experimental Biology, pp. 459–485.
- Tautz, J. and Sandeman, D. C. (1980) The detection of waterborne vibration by sensory hairs on the chelae of the crayfish. *J. Exp. Biol.*, **88**, 351–356.
- Tazaki, K. and Ohnishi, M. (1974) Responses from tactile receptors in the antenna of the spiny lobster *Panulirus japonicus*. *Comp. Biochem. Physiol.*, **47A**, 1323–1327.
- Tiselius, P. and Jonsson, P. R. (1990) Foraging behavior of six calanoid copepods: observations and hydrodynamic analysis. *Mar. Ecol. Prog. Ser.*, **66**, 23–33.
- Trager, G., Achituv, Y. and Genin, A. (1994) Effects of prey escape ability, flow speed, and predator feeding mode on zooplankton capture by barnacles. *Mar. Biol.*, **120**, 251–259.
- Weatherby, T. M., Wong, K. K. and Lenz, P. H. (1994) Fine structure of the distal sensory setae on the first antennae of *Pleuromamma xiphias* Giesbrecht (Copepoda). *J. Crust. Biol.*, **14**, 670–685.
- Wiese, K. (1976) Mechanoreceptors for near-field water displacements in crayfish. *J. Neurophysiol.*, **39**, 816–833.
- Wiese, K. and Marschall, H. P. (1990) Sensitivity to vibration and turbulence of water, in context with schooling in Antarctic krill, *Euphasia superba*. In: *Frontiers in Crustacean Neurobiology*, K. Wiese, W.-D. Krenz, J. Tautz, H. Reichert, B. Mulloney, eds., Birkhauser, Basel, Switzerland. pp. 121–139.
- Wright, D. I. and O'Brien, W. J. (1984) The development and field test of a tactical model of the planktivorous feeding of white crappie (*Pomoxis annularis*). *Ecol. Monogr.*, **54**, 65–98.
- Yen, J. and Fields, D. M. (1992) Escape responses of *Acartia hudsonica* (Copepoda) nauplii from the flow field of *Temora longicornis* (Copepoda). *Int. Revue ges. Hydrobiol.*, **36**, 124–134.
- Yen, J., Lenz, P. H., Gassie, D. V. and Hartline, D. K. (1992) Mechanoreception in marine copepods: electrophysiological studies on the first antennae. *J. Plankton Res.*, **14**, 495–512.