

Photosensitivity in the Sixth Abdominal Ganglion of Decapod Crustaceans: A Comparative Study

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Received September 29, 1975

Summary. The existence of photosensitive neurons in the 6th abdominal ganglion of the crayfish has been known for over forty years. These structures have been considered unique to freshwater crayfish, largely due to their apparent absence in a closely related species, the lobster *Homarus*. We have shown that abdominal photoreceptor neurons are widespread in occurrence among other marine decapod crustaceans. In addition to the electrical responses which result from illumination of the 6th ganglion, similarities in the distribution of afferent synaptic input and of the axon location in the nerve cord suggest that these cells are homologous central interneurons.

Introduction

The paired caudal photoreceptor (CPR) interneurons, located in the sixth abdominal ganglion of crayfish, are primitive light sensitive cells lacking both peripheral light conditioning structures and specialized phototransductive segments. These cells are of interest not only because of their role in a negatively photokinetic locomotory reflex (Welsh, 1934; Harris and Stark, 1974), but due to the combined function of the CPR as a sensory interneuron receiving tactile information from receptors on the telson and uropods (Kennedy, 1963; Wilkens and Larimer, 1972; Galeano and Beliveau, 1973). These unique features are relatively simple to assay by electrophysiological techniques.

Caudal photoreceptors have been reported in at least nine crayfish species, including blind cave-dwelling forms (Larimer, 1960). Since their discovery by Prosser 41 years ago (Prosser, 1934) these cells have been considered a unique characteristic of crayfish due to the apparent absence of similar light sensitive interneurons in closely related species, the lobster *Homarus* in particular (Prosser, personal communication; Wilkens and Larimer, 1974). Despite the lack of direct

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electrophysiological evidence in species other than crayfish, behavioral studies (Welsh, 1934; Hess, 1940) have indicated the possibility of abdominal photosensitivity in two other species of decapod crustaceans, the spiny lobster *Panulirus argus*, and the snapping (pistol) shrimp *Alpheus*.

In this report we will demonstrate electrophysiologically that abdominal photosensitive cells occur widely among marine decapods. Basic similarities, including the specificity of the light-sensitive region, the bimodal function as a somato-sensory interneuron, and axon location suggest that, in addition to the giant fibers, the caudal photoreceptor interneurons evolved as homologous structures.

Methods and Materials

A qualitative comparison of both the photic responses and the directional somatosensory input of the CPR, as found in the crayfish, was the basis for our tests on other species. However, in most of the smaller caridean shrimps, single-fiber recordings were difficult to maintain long enough to test each of the ganglionic roots for afferent input. Thus, these data are available only for the larger decapod species.

The abdomen of each animal was isolated from the cephalothorax, and the ventral nerve cord was exposed and desheathed. Electrical activity was recorded extracellularly by drawing into suction electrodes small fiber bundles that had been dissected from 5–6 abdominal connectives. Larger suction electrodes were used to stimulate the roots of the 6th ganglion. Our light source was a microscope lamp, fitted with a heat-absorbing filter and a shutter. Although constant supra-threshold intensities were used throughout these experiments, the amount of light was not measured quantitatively. Saline solutions were used for those species where formulae have been reported; crayfish saline (van Harreveld, 1936), lobster saline (*Panulirus*; Mulloney and Selverston, 1972), and shrimp saline (Kusano and LaVail, 1971). Otherwise, artificial sea water was the bathing medium. All preparations were tested at temperatures of 15–17 °C.

Results

A survey for photoreceptor interneurons in marine decapod crustaceans was begun following the discovery in the blue mud shrimp, *Upogebia pugettensis*, of a cell physiologically very similar to the crayfish CPR. Excluding crayfish, a total of eighteen decapod species and one stomatopod have been tested for

Fig. 1. Response patterns of light-sensitive interneurons, in five species of decapod crustaceans, to light shined on the caudal region of the isolated abdomens (upward pointing and downward pointing arrows signal the duration of the light stimulus). Records of electrical activity in three species have been shortened by 3, 2.7 and 1.6 sec respectively. Note that since light intensities were not measured quantitatively in these experiments, relative latencies to onset of response cannot be accurately compared. Cross-sections of the 5–6 connectives in each species are shown at the left. An arrow indicates the approximate position of the photoreceptor axon in each connective. Calibration bar: 500 μ . A schematic drawing of the 6th abdominal ganglion in each species, shown on the right, illustrates the number and relative morphology of ganglionic roots. Positive and negative signs indicate the synaptic effect of direct electrical stimulation of the roots while recording from the CPR teased from the upper connective

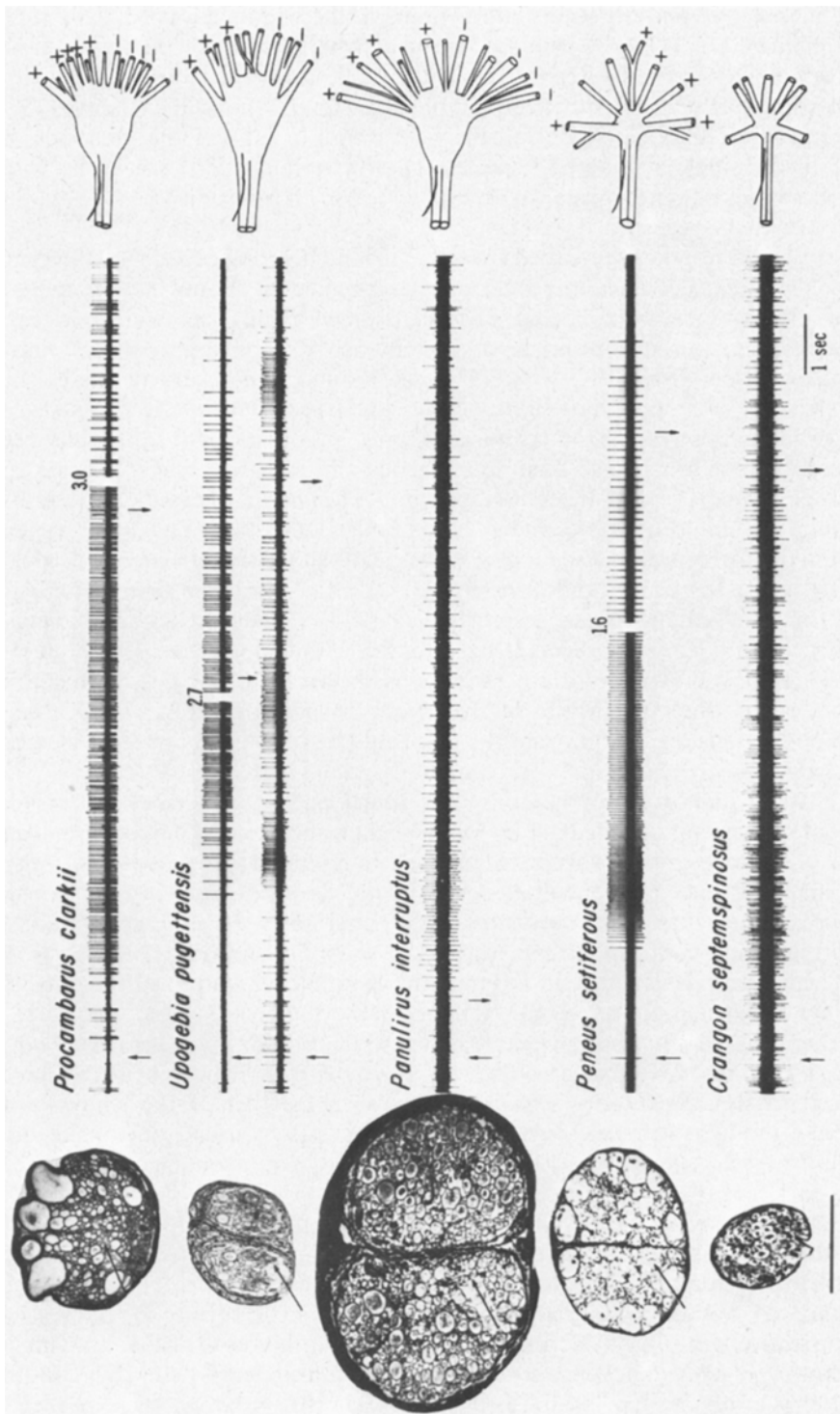


Fig. 1

the presence of photoreceptor interneurons in the abdominal cord. This survey (summarized in Table 1), includes representatives of most of the major groups of decapods except the deep water and pelagic forms. The animals are listed according to their presently accepted phylogenetic relationships (Barnes, 1974). As indicated, the light-sensitive cells were found in nine of the decapods, but not in the single stomatopod species. The distribution of the neurons within the decapods does not appear to accurately reflect the presumed genetic relationships of each species.

The light responses recorded from the blue mud shrimp *Upogebia*, the Pacific coast spiny lobster *Panulirus interruptus*, a commercial shrimp *Peneus setiferus*, and the common sand shrimp *Crangon septemspinus*, are shown in Fig. 1 compared to the light response of the crayfish *Procambarus clarkii* (Girard). Unlike the crayfish CPR, those of *Upogebia* and *Peneus* are normally silent in the dark-adapted state. In *Upogebia*, high-frequency bursts of spikes are superimposed on the light response in most preparations (Fig. 1), but occasionally, these bursts also arise spontaneously in the dark. The *Upogebia* cells also dark-adapt rapidly by comparison to *Procambarus* where the process may require a minute or more under comparable conditions. The light response of the photoreceptor in *Peneus* characteristically has a shorter latency, a greater initial burst frequency, and a less prolonged after-discharge than the crayfish neuron. The caudal photoreceptor of *Panulirus interruptus* is spontaneously active in the dark and shows a light response similar to that of the crayfish although only a short test flash response is shown here. A light-sensitive interneuron was observed in one of five *Crangon* specimens (Fig. 1), but due to the poor recording conditions, little else could be concluded about its electrical activity.

Caudal photoreceptor neurons were found and characterized in five additional species (not included in Fig. 1). The light responses of the prawn, *Pandalus*, and of the broken-back shrimp, *Spirontocaris*, resemble the response in *Crangon* in that both are spontaneously active in the dark and both appear to have approximately the same burst frequencies and rates of dark adaptation. In the remaining caridean shrimp tested, the snapping shrimp, *Alpheus armatus*, the photoreceptor showed an intermittent spontaneous output in the dark and a high-frequency light response similar to the CPR of *Peneus*. The level of spontaneous activity and the light response in the Florida spiny lobster *Panulirus argus*, both resemble that described for *P. interruptus*. However, in the shovel-nosed lobster, *Scyllarides aequinoctialis*, also a member of the spiny lobster family, the light response once again shows bursts of spikes not unlike those found in *Upogebia*. The CPR of this species is also spontaneously active in the dark.

After discovery of the photoreceptor in *Upogebia*, the possible homology of this cell in other decapods to that of the crayfish CPR was investigated further by determining the distribution of afferent tactile receptors. Input distribution, as well as gross ganglionic morphology in four species, is presented comparatively in Fig. 1. Tactile input entering via the roots of the 6th ganglion in both *Upogebia* and *Panulirus* is similar to the ipsilateral-excitatory, contralateral-inhibitory pattern established in the crayfish, although there are a few

differences in the number of roots and input specificity in each instance. In *Peneus*, however, the distribution of afferent input is significantly different. No inhibition was observed ipsilaterally or contralaterally while excitatory inputs appear primarily among the contralateral roots. Three paired roots were consistently observed in all of the natantians represented in this survey, whereas 5 or 6 paired roots are present in all reptantian decapods.

Additional evidence for the homology of the various CPR interneurons was obtained from the localization of the axons within the nerve cords. When referenced to a cross-sectional map of the 5-6 connectives, the neurons appear to be positioned very similarly in all the species tested. The approximate location of the axon in four species is indicated by the arrows in Fig. 1.

Table 1. Decapod crustaceans surveyed for abdominal photoreceptor interneurons. The phylogenetic relations are adapted from Barnes (1974). The column at the right indicates the number of animals in which photoreceptors were found, followed by the total number of experiments on that species. An asterisk indicates positive demonstration of CPR. References for the crayfish (parentheses) are: (1) Kennedy, 1963; (2) Larimer, Trevino and Ashby, 1966; (3) Larimer, 1960; (4) Prosser, 1934; (5) Galeano and Beliveau, 1973

Phylogeny	Species	No. tested
Order Decapoda		
Suborder Natantia		
Section Penaeidea	* <i>Peneus setiferus</i>	4-4
Section Caridea		
Superfamily Palaemonida	<i>Palaemonetes vulgaris</i>	0-4
	<i>Macrobrachium carcinus</i>	0-2
	<i>M. acanthurus</i>	0-2
Superfamily Alpheoidea	* <i>Alpheus armatus</i>	3-3
Superfamily Pandaloida	* <i>Pandalus</i> sp.	1-2
Superfamily Hippolytidae	* <i>Spirontocaris</i> sp.	1-1
Superfamily Crangonoida	* <i>Crangon septemspinosus</i>	1-5
Section Stenopodidea	<i>Stenopus</i> sp.	0-2
Suborder Reptantia		
Section Macrura		
Superfamily Scyllaridea	* <i>Panulirus interruptus</i>	4-4
	* <i>P. argus</i>	1-7
	* <i>P. guttatus</i>	0-1
	* <i>Scyllarides aequinoctialis</i>	1-1
Superfamily Nephropsidea	<i>Homarus americanus</i>	0-6
	* <i>Procambarus clarkii</i> (1)	
	* <i>P. simulans</i> (2)	
	* <i>Cambarus setosus</i> (3)	
	* <i>C. sciotensis</i> (2)	
	* <i>C. virilis</i> (4)	
	* <i>C. limosus</i> (4)	
	* <i>Orconectes virilis</i> (2)	
	* <i>O. rusticus rusticus</i> (5)	
	* <i>O. pellucidus australis</i> (2)	
Superfamily Thalassinidea	* <i>Upogebia pugettensis</i>	7-7
	<i>Callinassa gigantea</i>	0-6
Section Anomura	<i>Pagurus pollicarus</i>	0-2
	<i>Clibanarius vittatus</i>	0-2
Order Stomatopoda	<i>Squilla empusa</i>	0-3

Discussion

The concept of cellular homology has been applied to the evolution of nervous systems based both on structure and function, e.g., giant fibers including the Mauthner neurons (Stefanelli, 1951), the Retzius' cells of leeches (Lent, 1973) and the giant fibers of annelids and crustaceans. Although a great deal of variation exists within the Annelida, giant axons show patterns of similarity in certain polychaete families and, in a number of oligochaete families, the giant fibers also are likely to have had a common ancestral origin (Nicol, 1948). In decapod crustaceans, the median giant fibers appear comparable in a variety of species, including *Homarus* (Allen, 1894), *Palaemonetes* and *Cambarus* (Johnson, 1924), and *Callinassa* (Turner, 1950). Further, Sakharov (1970) cites histochemical and pharmacological evidence for tracing cellular homologies, as illustrated by the monoaminergic cells found in segmental ganglia of both leeches and earthworms.

Our evidence for the presence or absence of CPR interneurons in crustaceans was based on their light sensitivity. Once located by this means, not only could the light response be characterized for comparison but other data including spontaneous activity, synaptic input, and cord position were readily available. Based upon the fundamental similarities of all these criteria, it seems reasonable to assume that the CPR cells that have been located thus far are probably homologous. The alternate interpretation, i.e., that the caudal photoreceptors represent analogous structures, would imply that the ability to perceive illumination of the abdominal segments is of great importance and, as a result, was independently evolved by each individual species. No evidence is available to support this contention.

Unfortunately, in those instances where the search for CPR's was negative, we do not know that the homologous neurons are absent. For example, an organism could evolve a loss of light sensitivity in the cell but retain other homologous characteristics such as basic morphology, position, and synaptic input. If the light sensitivity were not present, in fact, the physiological function of the CPR's could not be distinguished from a large class of directionally-sensitive and purely mechano-receptive interneurons found in the abdominal cords of many crustaceans. In addition, our failure to identify photoreceptors in several species may have been due to experimental technique, a possibility suggested by the fact that only one out of five specimens of *Crangon* exhibited a light response. Nevertheless, we can be fairly certain that some species, including *Homarus* and the fresh water shrimp, do not possess caudal photosensitivity on the basis of these experiments.

One explanation for the uneven distribution of CPR interneurons among decapods, then, is that there has been an evolved loss of light sensitivity in some species. Assuming this, one must also suggest that, of the various functions performed by the interneuron, light sensitivity is perhaps less important to the survival of the species. This conclusion is not totally satisfactory, however, since the basic role of the interneuron in the control of coordination of behavior is still largely unknown. No direct evidence is available to suggest that the CPR functions as a command interneuron. Nevertheless, we feel that the present

evidence for an apparently homologous neural system in several decapod crustacean species will offer a valuable comparative resource for studies on the photoreceptor behavioral reflex, its neural basis, and on the evolution of identified neurons.

We thank Dr. R. Fernald of the Friday Harbor Laboratories and Dr. B. Ache, Florida Atlantic Univ., Boca Raton, Fla., for the generous supply of laboratory facilities and Dr. D. Kennedy for reviewing the manuscript. This research was supported in part by NIH research grants NS 11751-01 (to L.A.W.) and NS 05423-08 (to J.L.L.) from the National Institute of Neurological Diseases and Stroke, NIH Postdoctoral Fellowship NS 43039-02 (to L.A.W.) and by a Grass Fellowship in Neurophysiology (to L.A.W.). This paper is a publication of the Tallahassee, Soppchopy, and Gulf Coast Marine Biological Association (No. 40).

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