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Photoreceptor-like Cells in the Prostomium of a Scaleworm, *Lepidonotus helotypus*

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ABSTRACT Pear-shaped photoreceptor-like cells are widely scattered in the prostomial epithelium of the scaleworm, *Lepidonotus helotypus*. The cell is provided with cilia, the basal structure of which is situated beneath the cuticle. Having run inward through a narrow neck, ciliary sheaths soon branch out and form a bulbous mass of microvilli (phaosome) above the nucleus. No positive evidence was obtained as regards the continuity between the phaosome and the cell membrane but membranous clefts extending from either side are frequently observed. The cell is surrounded by supportive epithelial cells and also by presumably chemo- or mechanoreceptoral cells of low electron density. One of the two types of electrical responses to light which were recorded over the prostomial surface is considered to originate from extraocular receptors. The possibility of involvement of the photoreceptor-like cells in such responses is discussed. (*Zool. Mag. 85: 265-269, 1976*)

Though dermal photosensitivity can be seen in a number of animals, only a few cells have been claimed to be responsible for it, such as ciliated cells found on the siphonal tip of *Nassarius reticulatus* (Crisp, 1971), phaosome-containing cells in the epithelium of *Lumbricus terrestris* (Röhlich *et al.*, 1970), etc. In some polychaetes, cells called photoreceptor-like cells (Hermans and Cloney, 1966), sensory sacs (Zahid and Golding, 1974), or simple photoreceptors (Whittle and Golding, 1974), have been reported. The lack of electrophysiological evidence has restrained these authors from using the term photoreceptors for such cells. During the course of ultrastructural studies of the prostomial photoreceptive systems of the scaleworm, *Lepidonotus helotypus*, I have encountered a new type of cell scattered in the epithelium of the prostomium. They appear to come under the same category as those mentioned above and I shall follow the terminology of Hermans and Cloney (1966) for the time being. Morpho-physiological elucidation of those dermal photoreceptive systems is the principal aim of the present work.

Material and Methods

Scaleworms, *Lepidonotus helotypus*, were collected from the Seto Inland Sea. Prostomial regions were dissected out from excised whole heads in 3% glutaraldehyde in 0.1 M cacodylate buffer (pH 7.8) with the addition of 8.5% sucrose. The pre-fixation that lasted for 3hr was followed by post-fixation in 1% osmium tetroxide in the same buffer overnight on ice. Fixed tissues were routinely treated for thin sectioning on a Porter-Blum MT-2B ultratome. The sections stained with uranyl acetate and lead citrate were examined in a Hitachi HS-8 electron microscope operated at 50 kV.

For electrophysiological experiments, a head piece isolated under dim red light was mounted on a sheet of moistened filter paper with the cuticular side up. A glass capillary microelectrode filled with 2 M KCl (2.5 MΩ in the tip resistance) was applied vertically onto the preparation and the indifferent electrode of a silver plate was placed beneath the filter paper. Electrical responses were amplified and displayed on a dual beam oscilloscope (Nihon Kohden, VC-7A) with a time constant of 0.2 sec. The positivity of

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the recording electrode was shown as an upward deflection. The stimulating light source was a 35 W tungsten filament lamp which was admitted by means of an electromagnetic shutter and led to the preparation through an infrared absorbing filter and a light guide (10,000 lux at the surface of preparation).

Results

Photoreceptor-like cells that lie perpendicular to the cuticular surface (Fig. 1) are scattered under the cuticle of the prostomium, about 30 μm deep, and their distribution is not limited to the ocellar region. The cell is pear-shaped and ciliated. The uniqueness of its structure is that the nucleus is situated at the bottom of the cell whereas the cilia have their basal structure beneath the cuticle and run through a narrow neck (2-3 μm in diameter; 15-20 μm in length) that connects the two extreme sides of the cell. At about the middle of the cell length, the ciliary sheath branch out and form a bulbous mass (7-10 μm in diameter) of microvilli above the nucleus. The microvilli, 0.15 μm thick and spaced with a gap of about 14 nm, are not arranged in a fixed pattern but appear to be a spirally coiled whorl. This may be called a phaosome. Microtubules of each cilium have a pattern of $2+9\times2$ at first but the central pair are soon lost. The fate of the peripheral doublets was not determined, but single tubules were occasionally observed within some of the villi.

The center of the phaosome is filled with an amorphous electron-opaque matrix (Fig. 1), which may be regarded as a primitive lens-like structure by analogy to a humor in a pulmonate snail, *Helix aspersa* (Eakin and Brandenburger, 1967).

The important question of whether or not the phaosome is structurally connected with the outer plasmalemma cannot be positively answered. However, the fact that clefts are seen frequently extending outward from the phaosome (Fig. 2) as well as inward from the cell surface (Fig. 3) may suggest the presence of membrane continuity.

The nucleus, 6 μm by 3 μm, is often irregularly shaped. The cell is provided with well-developed Golgi complexes and many vesicles (0.1 μm in diameter), rough-surfaced endoplasmic reticula and ribosomes (0.02 μm) but only few mitochondria and pigment granules (Fig. 1). A narrow process projecting from the innermost region of the cell was always observed (arrow). This may be an axon, but lacking evidence with serial sections, any conclusion must be reserved for future work.

The photoreceptor-like cell is surrounded by epithelial cells and electron light cells of unknown function. The epithelial cells may be supportive in function, for they not only deeply interdigitate with the receptor-like cells, but also desmosomes and septate junctions are frequently observed between the two types of cells. The light cell, on the other hand, may be characterized by the abundance of mitochondria and microtubules. Though not shown in the figure, it projects in addi-

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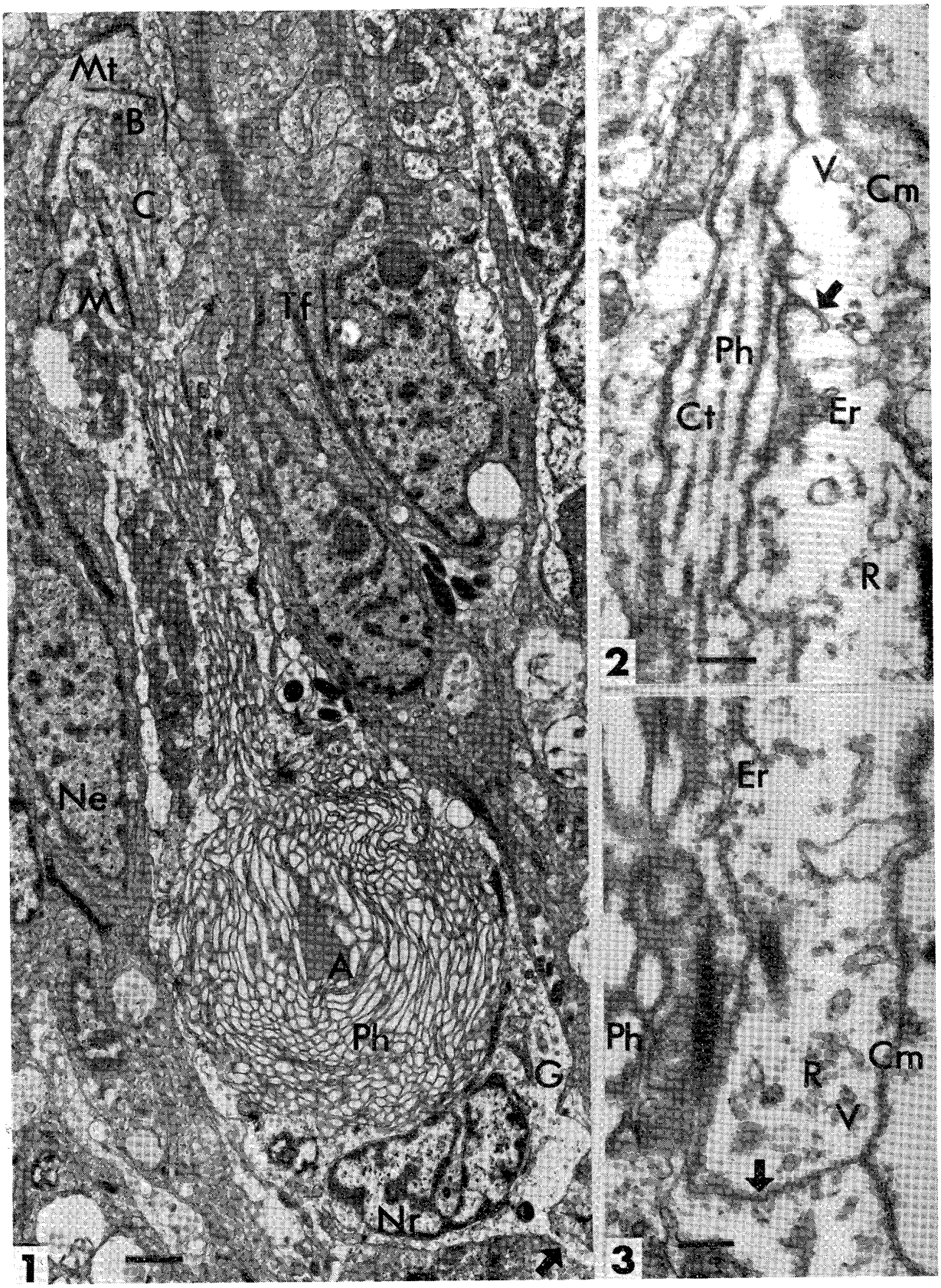
Fig. 1. The photoreceptor-like cell. A, amorphous matrix; B, basal body; C, cilium; G, Golgi complex; M, mitochondria of light cell; Mt, microtubules; Ne, nucleus of epithelial cell; Nr, nucleus of receptor-like cell; Ph, phaosome; Tf, tonofilament. An arrow indicates an axon-like process. Scale: 1 μm.

Fig. 2. The cleft (arrow) extending outward from the phaosome in the receptor-like cell. Cm, cell membrane; Ct, ciliary microtubules; Er, rough-surfaced endoplasmic reticulum; R, ribosomes; V, vesicle. Scale: 0.2 μm.

Fig. 3. The cleft (arrow) extending inward from the cell membrane in the receptor-like cell. Scale: 0.2 μm.



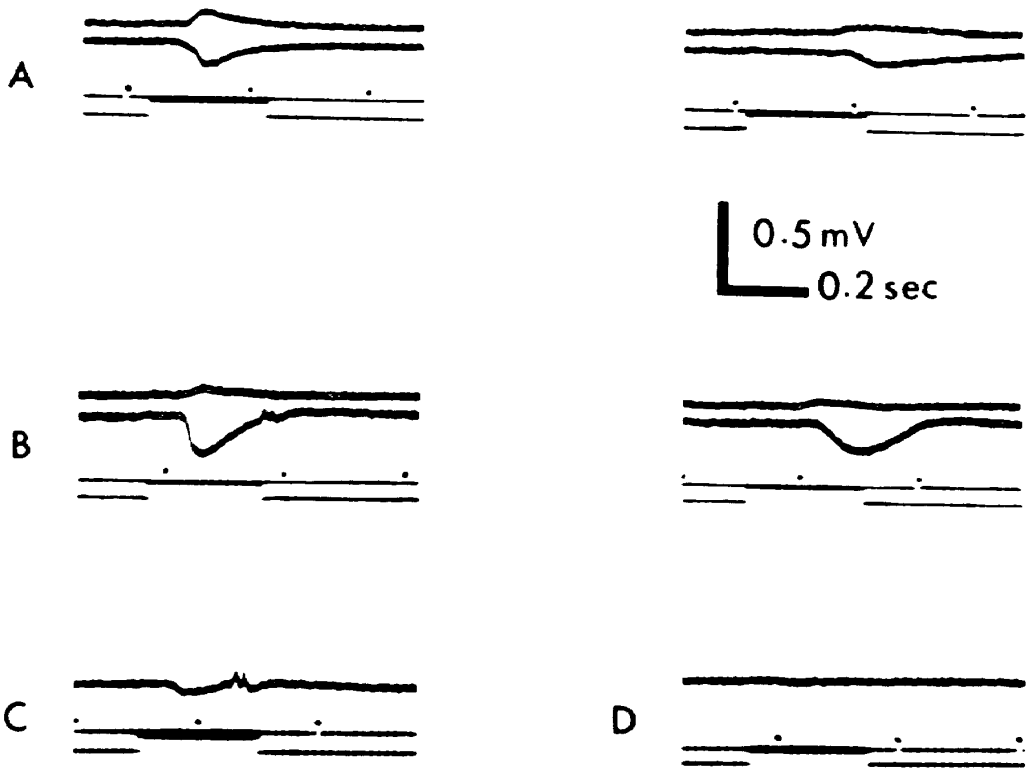


Fig. 4. Responses from the cuticular surface of the head region (pro- and peristomium), (18°C). Light stimuli were delivered at constant intervals (2 sec). Occasionally, spontaneous pulses from the brain were seen (B and C). Upper records in A and B; responses from the median part of the prostomium. Lower records in A; responses above the anterior ocellus: in B; above the posterior ocellus. Records on the left in A and B; responses after 3 min in darkness. Records on the right in A and B; responses to the 9th and 8th stimuli, respectively. C; a response after 3 min in darkness from a part between anterior and posterior ocelli. D; a response after 3 min in darkness from the peristomium.

tion a cilium into the cuticular layer. These features suggest that the cell may be chemo- or mechanoreceptoral.

Electrical responses recorded from the prostomial surface were either positive or negative in sign, depending on the position of the electrode (Fig. 4). Compared with the negative responses which were restricted to the ocellar region, the positive ones were wider in distribution, smaller in amplitude and longer in latency in dark-adapted preparations (Figs. 4 A and B, left). The sequence of occurrence of the two types of responses can be reversed by repetitive stimulation, however, so that the positive responses appear before the negative ones at the 8th or 9th stimuli (A and B, right). The difference in adaptability as well as the opposite polarity may be taken to indicate that the two are originated from different sources. The negative responses are possibly derived from the ocellus, because as the electrode was shifted away from the ocellar region, the amplitude became smaller (Figs. 4 C and D). It is plausible that the positive responses have been derived from some photoreceptoral elements other than ocelli in the prostomium.

Discussion

The phaosomal structure described above is similar in some respects to that of the earthworm, *Lumbricus terrestris* (Röhlich *et al.*,

1970) and the leech, *Helobdella stagnalis* (Clark, 1967). Though the presence of the ciliary structure resembles that of the earthworm cells, the separation of the basal structure and the nucleus at the two extremes of the pear-shaped cell is entirely new.

The analogy among the three cells lies in that the mass of microvilli is surrounded by the cell from which they originated. In the leech, the continuity between the phaosome and the cell membrane has been demonstrated morphologically (White and Walther, 1969) as well as electrophysiologically (Fioravanti and Fuortes, 1972), but not in the earthworm. Nor was it possible to reveal the continuity in this species, but we cannot dismiss at this stage the possibility of its presence through deep invaginations of the cell membrane. The presence of the dermal photoreceptors has been suggested in eyeless preparations of *Nereis* by behavioral responses (Evans, 1969) as well as by impulse generations (Gwilliam, 1969). The slow potentials recorded in the present study may be considered to have been originated from extraocular elements. It is thus highly probable, but is still to be determined that the photoreceptor-like cells are the source of such electrical activities.

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