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Gap Junctions Suggest Epithelial Conduction Within the Comb Plates of the Ctenophore *Pleurobrachia bachei***

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Summary. Intercellular gap junctions occur between the ciliated cells that make up the comb plates of the ctenophore *Pleurobrachia*. Similar junctions are found within the ciliated grooves which run from the apical organ to the first plate of each comb row, as well as throughout the endoderm of the meridional canals. Gap junctions were not found in the ectodermal tissue between the comb rows. The distribution of junctions suggests that excitation conduction within the ciliated grooves, comb plates and meridional canal endoderm may be epithelial.

Key words: Gap junction – Epithelial conduction – Ctenophora – Cilia.

Conduction of excitation between the ciliary plates of ctenophores is suspected to be non-nervous (Horridge, 1965, 1966, 1974; Mackie, 1970; Spencer, 1974). Evidence for neurotid, or epithelial, conduction has come from behavioral experiments utilizing excess Mg^{++} and a variety of surgical manipulations (Parker, 1905; Horridge, 1965; Sleigh, 1968). Tamm (1973), on the basis of the latter, indicates that conduction between comb plates of cydippids and beroids depends on mechanical coupling and not on conduction of electrical events. In the lobates, however, electrical conduction along the comb rows is still suggested (Horridge, 1965; Tamm, 1973).

Morphological analysis of conduction systems of ctenophores has been primarily concerned with ectodermal and mesogleal nerve elements, which form a network maintaining an inhibitory influence over the comb plates (Horridge, 1965; Hernandez-Nicaise, 1973a, b, c). Synapses have been described between neurites

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and the comb plate cells of several ctenophores (Horridge and Mackay, 1964; Horridge, 1965; Hernandez-Nicaise, 1973a, c). However, junctional specializations suggestive of intercellular electrical coupling have not previously been reported in areas where they are behaviorally indicated.

We find intercellular junctions similar to the gap junctions found in coelenterates (Hand and Gobel, 1972) and higher animals (Staehelin, 1974) in the comb plates of the cydippid ctenophore *Pleurobrachia bachei*. This report represents a portion of an ongoing morpho-physiological investigation of conduction systems of various ctenophores.

Materials and Methods

Specimens of *Pleurobrachia bachei* Agassiz were collected in the Santa Barbara Channel by surface tows, or individually by divers. Live material was fixed in 2% glutaraldehyde in cacodylate buffer followed by post-fixation in 1% OsO_4 in a similar buffer. The solutions were adjusted to 900–930 MOsM (calculated). Following ethanol-propylene oxide dehydration, the tissue was embedded in araldite. Thin sections were cut on a Porter-Blum MT-2 ultramicrotome with glass or diamond knives. The sections were stained with uranyl acetate and lead citrate prior to examination in a Siemens Elmiskop IA electron microscope.

Results

The ciliated cells forming the comb plates of *Pleurobrachia* are tall and columnar (65 μm long, 2.3 μ wide). Each cell bears 10–50 cilia which are packed together to produce a plate of hundreds of cilia (Figs. 1, 2). Each comb plate moves as a single unit. The cells of the comb plate extend from the apical surface to the basement membrane bordering the mesoglea. They generally possess a medium-dense cytoplasm with many longitudinally oriented microtubules and several extremely large mitochondria (up to 2 μ wide and 4 μm long). Neurons are frequently found running between the comb plates (Fig. 7) onto which they occasionally synapse (Fig. 4).

Intercellular membrane junctions are found at all levels between adjacent comb plate cells. The junctions are typically 0.5–0.8 μm long and exhibit close apposition of the adjacent outer leaflets of the two plasma membranes (Figs. 3, 5–7). The junctions result in the narrowing of the intercellular cleft from 20–30 nm in a non-junctional area to 3–4 nm (Fig. 4). The junctional complexes have a seven-layered appearance typical of gap junctions prepared with glutaraldehyde, osmium and uranyl acetate (Staehelin, 1974). They are most numerous in the apical and basal regions of the comb plate but were also encountered in the central areas of the plate. Gap junctions were observed between the ciliated cells of the ciliated grooves leading from the apical organ to the comb rows, but not in the ectodermal tissue between comb plates.

Gap junctions similar to those described above were also found in the endoderm of the meridional canals which underlie the comb rows. They have been identified between at least three undescribed cell types. The gap junctions tend to be

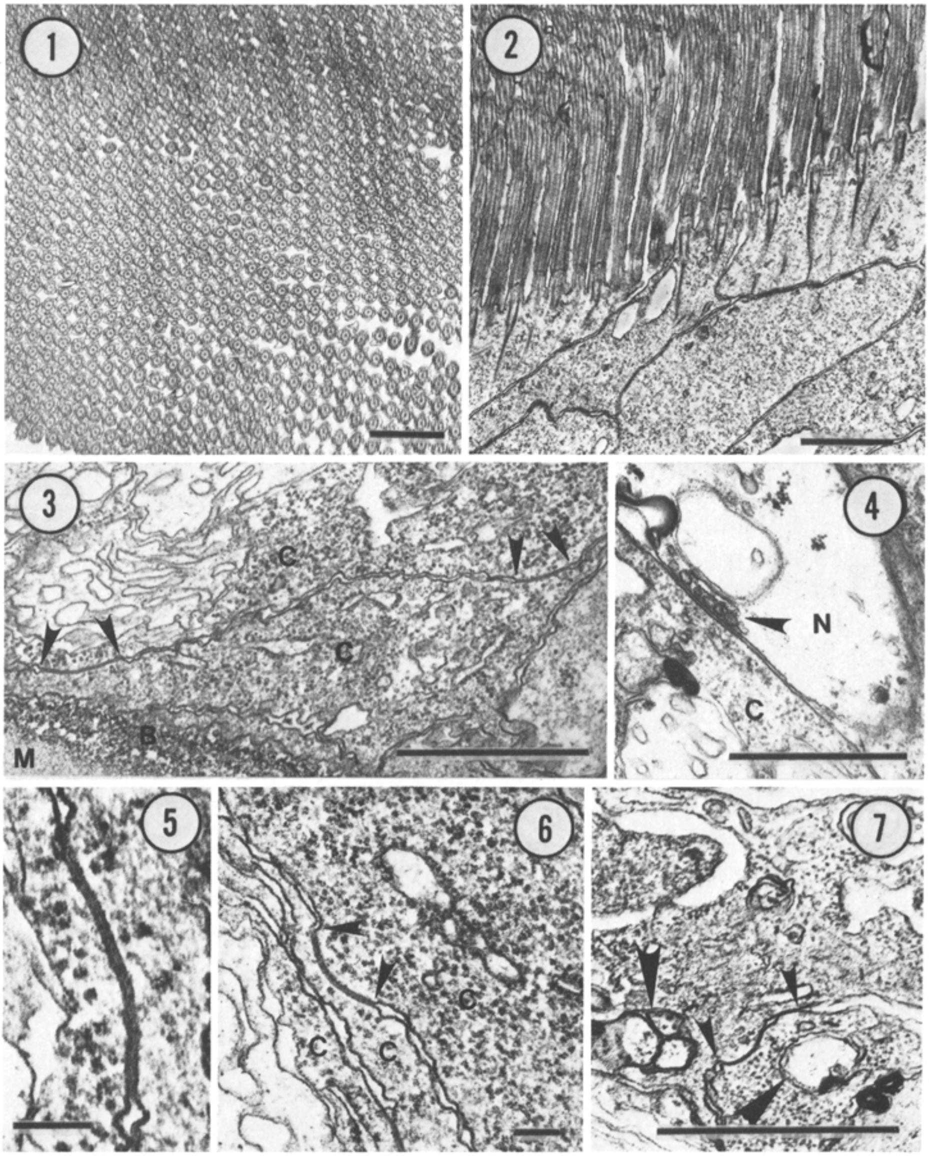

Fig. 1. Cross-section through the tip of a comb plate. Note the tight packing of the numerous cilia, each of which is contained in an individual sheath. Scale = 1 μ

Fig. 2. Tangential section through the apical portions of comb plate cells. Each cell contributes several cilia to the plate. Scale = 1 μ

Fig. 3. Membrane apposition of two comb plate cells (*C*) near the basement membrane (*B*) and mesoglea (*M*). Two gap junctions are delineated by arrows. Scale = 1 μ

Fig. 4. Neuro-comb plate cell synapse (arrow) from the apical region of a comb plate. *N* neuron; *C* comb plate cell. Scale = 1 μ

Fig. 5. Enlarged view of the left gap junction of Fig. 3. Note the close apposition and parallel relationship of the two membranes. Scale = 0.1 μ

Fig. 6. Gap junctions (arrows) within the central region of the comb plate. *C* comb plate cells. Scale = 0.1 μ

Fig. 7. Gap junctions (small arrows) in the apical portion of the comb plate. The large arrows indicate neurons which penetrate the comb plate cells (*C*). Scale = 1 μ

distributed throughout the entire meridional canal endoderm rather than being localized under the comb plates, suggesting that a non-nervous conduction system may be present in the endodermal lining of the canals.

Discussion

Anatomical evidence suggests the existence of non-nervous conduction of electrical activity within the comb plates of the cydippid ctenophore *Pleurobrachia*. Gap junctions are present throughout the comb plates but not in the tissue between them. This discontinuity is consistent with the behavioral observations of Tamm (1973), and supports his supposition that coordination between plates is not electrical but mechanical. The function of the gap junctions on *Pleurobrachia,* therefore, does not appear to be conduction of excitation from plate to plate, but rather in coordination within a plate. Electrical coupling of comb plate cells presumably contributes to a unification of the beating of all cilia making up the plate, some of which can be separated by up to a millimeter.

However, the gap junctions of the ciliated grooves appear to be involved in longitudinal conduction of excitation from the apical organ to the first comb plate of *Pleurobrachia.* This observation takes on added significance when comparing it to the lobate ctenophores. In these forms, in which non-nervous plate-to-plate coordination is still suggested (Tamm, 1973), the ciliated grooves extend throughout the length of the comb rows.

Presence of an endodermal conduction system within the meridional canals has no known behavioral or physiological correlate. The identification of gap junctions within these tissues of *Pleurobrachia* suggests that they might be concerned in the control of bioluminescent responses, since the luminescent effector cells are located in the meridional canal endoderm (Freeman and Reynolds, 1973).

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