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The Occurrence of the Preoptic Neurosecretory Cells in the Diencephalic Ventricle of the Arctic Lamprey, *Lampetra japonica*

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Lampreys, a representative of the most primitive living vertebrates, have a definite preoptico-hypophysial neurosecretory system in their diencephalon comparable to that of higher vertebrates (Sterba, 1972). As was shown in previous papers (Öztan and Gorbman, 1960; Tsuneki and Gorbman, 1975a; Nakai et al., 1979; Yui and Honma, 1979), the preoptic nucleus of lampreys contains so-called cerebrospinal fluid (CSF)-contacting neurons which project their dendritic process into the ventricular cavity. During the course of our anatomical studies on the brain-ventricular system of the arctic lamprey, *Lampetra japonica* (Chiba et al., 1981; Chiba and Honma, 1986a), the preoptic neurosecretory cells, which should be better-called intraventricular perikarya according to Vigh-Teichmann and Vigh (1983), were encountered. In spite of extensive studies on the periventricular structures of the brain of the arctic lamprey (Honma, 1969; Honma and Honma, 1970; Ochi and Hosoya, 1974; Tsuneki, 1974; Tsuneki et al., 1975; Shioda et al., 1977; Nakai et al., 1979; Yui and Honma, 1979; Chiba et al., 1981; Yamamoto et al., 1983; Chiba and Honma, 1986a; Ushiki et al., 1986), no cytological details of such intraventricular perikarya have been described.

In this short communication, we report ultrastructural features of the intraventricular neurosecretory perikarya in the lamprey brain, along with a brief discussion on a possible role of the cells in neuroendocrine integration.

Material and methods

Twenty adult arctic lamprey, *Lampetra japonica* (Martens), of both sexes, ranging 45–52 cm in total length, were caught during their upstream migration in the lower reaches of the Agano River and the Shinano River flowing into the Sea of Japan through Niigata City. The period of collection was from February 1986 to March 1988. The animals were killed by decapitation under anesthesia in 0.5% MS 222 and their brains were quickly removed from the cranium, and immersed in various fixatives.

For light microscopy, the brains were fixed in Bouin's solution, dehydrated through an ethanol series, embedded in paraffin, cut serially at 8 μm thickness in saggital and transverse directions, and stained with aldehyde fuchsin (AF)-fast green-orange G.

For transmission electron microscopy, the brains were immersed overnight in Karnovsky's solution, postfixed in 1% OsO_4 for 2 hrs., dehydrated through an ethanol series, and embedded in Epon 812 or Spurr. Ultrathin sections cut with a LKB ultratome were double-stained with uranyl acetate and lead solutions, and examined with a JEOL 1200 EX electron microscope.

For scanning electron microscopy, tissue blocks prefixed with the same solution adopted for transmission electron microscopy were postosmicated, dehydrated, critical point-dried, sputter-coated with platinum-palladium, and observed with a Hitachi S-800 electron microscope.

Results and discussion

By light microscopic examination, round or oval cells measuring 7–10 μm in diameter were found lying solitarily or in a small cluster on the ventro-lateral wall of the preoptic recess (Fig. 1). These cells were identified as preoptic neurosecretory neurons by their staining affinity for AF and their location close to the preoptic nucleus. In one of the sections, an intraventricular cell projecting an AF-positive axon into the subependymal neuropil was also recognized (Fig. 1). Of the twenty animals examined, five specimens comprising both sexes showed such intraventricular neurosecretory perikarya.

At the ultrastructural level, the intraventricular cell contained a round nucleus with a very prominent nucleolus. The perikaryon was characterized by the presence of parallel arrays of granular endoplasmic reticulum, scattered granules ranging 120–220 nm in diameter, and a small number of mitochondria (Fig. 2). The content of the granules showed varied densities (Fig. 2, inset). Polysomes, microtubules, vesicles of various sizes, and Golgi apparatus were also recognized as well as coated vesicles. These ultrastructural features

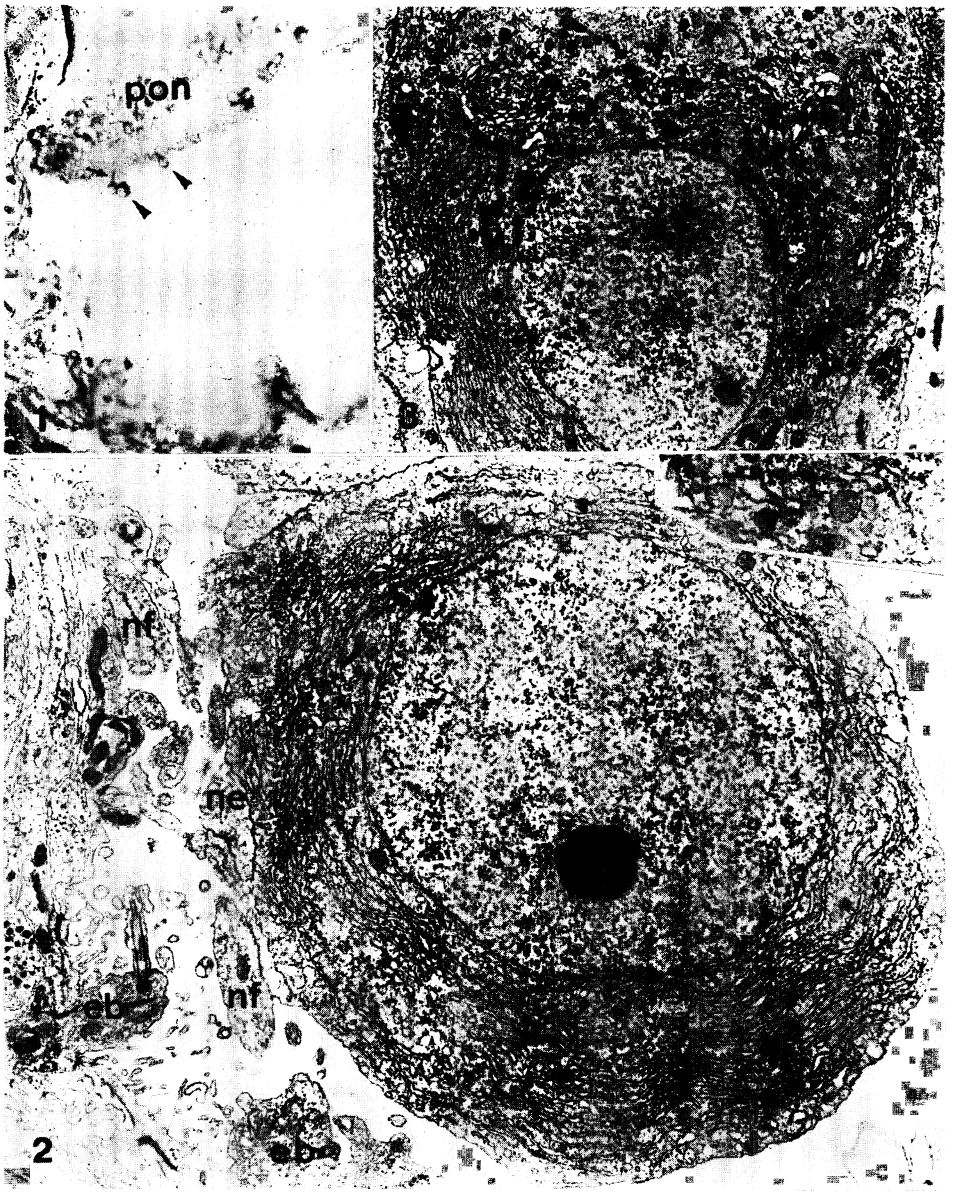

Fig 1. Section of the preoptic area of the arctic lamprey *Lampetra japonica* showing the intraventricular neurosecretory perikarya (arrowheads) protruding into the preoptic recess. Note the intraventricular cell sending AF-stainable axon (small arrow) toward the preoptic nucleus (pon). ×200.

Fig. 2. Detail of the intraventricular neurosecretory cells in the preoptic recess of the lamprey brain. The cells show secretory perikarya characterized by the presence of parallel arrays of granular endoplasmic reticulum, scattered elementary granules, and few mitochondria. Intraventricular nerve fibers (nf) and their ending (ne), and endbulbs of the CSF-contacting neurons (eb) are also noticed. ×8,800. Inset shows elementary granules of varied densities. ×23,000.

Fig. 3. Ultrastructural features of an ordinary neurosecretory cell of the preoptic nucleus showing stacks of granular endoplasmic reticulum, a number of elementary granules, and Golgi bodies in the perikaryon. ×7,500.

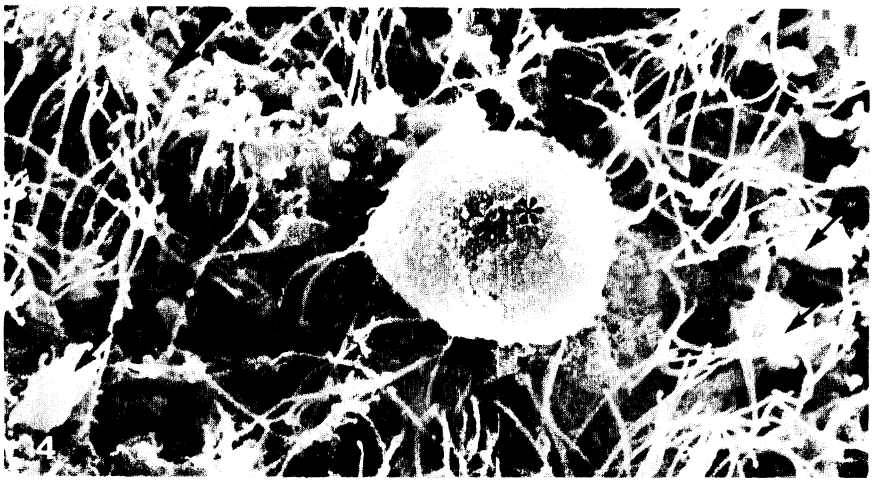

Fig. 4. Scanning electron microscopy of an intraventricular cell lying on the ependymal surface of the preoptic recess of the lamprey brain. Endbulbs of the CSF-contacting neurons (small arrows) and an intraventricular cell (asterisk) associated with intraventricular nerve fibers (large arrow) are shown. Note the rough surface of the intraventricular cell bearing protuberances. ×3,800.

were common to the ordinary preoptic cells found within or beneath the ependyma (Fig. 3). The intraventricular cells showed an intimate association with intraventricular nerve fibers and ciliated endbulbs of the CSF-contacting neurons with or without small-cored vesicles, thus forming supraependymal neuronal complexes. The nerve ending which comes in contact with the intraventricular perikaryon was also demonstrated (Fig. 2).

By scanning electron microscopy, a large supraependymal cellular structure corresponding to the aforementioned intraventricular cell was demonstrated (Fig. 4). This structure appeared to be plunging into the ventricular cavity through the ependyma and to be associated with the intraventricular nerve fibers. The surface of this object was uneven and provided with granular protuberances (Fig. 4). Minute caveolae were occasionally seen, but neither cilia nor pseudopod-like processes were demonstrated (Fig. 4).

In general, supraependymal or intraventricular cells found within the cerebral ventricles of vertebrates include different cell types: neurons, glial cells, and wandering cells typically represented by Kolmer cells (intraventricular macrophages). All these cell types have already been reported to occur in the cerebral ventricles of the cyclostomes (Tsuneki and Gorbman, 1975b; Shioda et al., 1977; Chiba et al., 1981; Chiba and Honma, 1986a, b). However, the intraventricular cells presented here differ from the supraependymal free cells previously reported to exist in the cyclostome brain (Tsuneki and Gorbman, 1975b; Chiba and Honma, 1986b). As evidenced by light and electron microscopic data, the cells in question are identifiable as neurosecretory neurons belonging to the preoptic nucleus. Even in the ventricular cavity, these cells appeared to have secretory capacity, although the site of release of the cytoplasmic granules was not seen. On the basis of these morphological findings, we consider that these cells are displaced into the ventricular lining to represent a specialized form of CSF-contacting neuron of the preoptic nucleus. A similar disposition of the neuronal perikarya was reported in the mesencephalic nucleus of the 5th cranial nerve of the shark brain (Kemali and Miralto, 1979).

Intraventricular neurons, synapses, and axons were found in almost all vertebrates from fishes to mammals (Vigh-Teichmann and Vigh, 1983). These neurons may have various capacities, i.e., sensory or secretory function as proposed for the CSF-contacting neurons (Smoller, 1965; Vigh-Teichmann and Vigh, 1983). The intraventricular neurosecretory perikarya presented here were associated with intraventricular nerve fibers and endbulbs of the CSF-contacting neurons to form intraventricular neuronal complexes. In such an anatomical disposition, chemical or

physical information within the cerebral ventricle may be directly perceived or received by these cells, or indirectly transferred via intraventricular nerve fibers. Consequently, these neuronal complexes may participate in hypothalamic neuroendocrine integration. Further physiological and phylogenetical studies are necessary to elucidate their exact function.

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カワヤツメの第三脳室内に認められた視束前核由来の神経分泌細胞

千葉　晃・本間義治

円口類の脳室および視床下部—下垂体系に関する比較解剖学的研究の過程で，邦産カワヤツメ成魚の第三脳室前視束陥凹内に，しばしば髄液内（上衣上）細胞の出現

を認めた．これらを光学顕微鏡ならびに電子顕微鏡によって精査した結果，視束前核由来の神経分泌細胞であることが判明した．この髄液内神経分泌細胞は，微細構造上分泌能を有しており，かつ髄液接触ニューロンの脳室内突起や髄液内神経線維と接触を保ち，いわゆる上衣上神経複合体を形成していることから，神経内分泌協関の一役を担っているものと予想された．

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