

Neurosecretion in the Marine Crab, *Charybdis lucifera* (Fabricius)

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Five different types of neurosecretory cells (NSC), designated as A, B, C, D, and E cells, were identified. A, B, and D cells occurred in the brain, thoracic ganglion, circumoesophageal connective ganglion and optic ganglia. C cells were confined to the anterior region of the thoracic ganglion and E cells in the brain. A single X - organ was present in the medulla interna and 3 in medulla terminalis. Axonal fibres from these X - organs terminated in the sinus gland forming the sinus gland X - organ tract. Presence of NS material throughout the tract suggested the transport of NS product to the sinus gland via these axons.

Studies have been carried out on a number of crustaceans to elucidate the source and nature of neurohormones and their significance in the maintenance of different homeostatic mechanisms¹⁻⁴. Considering the importance of neurohormones in the control of various physiological processes of decapods⁵⁻⁷ and the discrepancy in correlation of the different types of neurosecretory cell (NSC) types so far described in different species⁸ the central nervous system of the marine crab, *Charybdis lucifera* (Fabricius), has been examined histologically to study different types of NSC and their distribution.

Materials and Methods

Crabs were collected from Bombay waters. The eyestalks were excised from live specimens and chitinous exoskeleton was removed without damaging the soft tissue. The inner tissue was fixed in aqueous Bouin's fixative. Cerebral ganglion, thoracic ganglion and circumoesophageal connective ganglion were dissected and fixed in the same fixative. Tissues were dehydrated in ethanol and embedded in paraffin wax (56 - 58°C). Sections (8 - 10 μ m thick) were cut and stained with Gomori's chromealumhaematoxylenephloxine.

Results

Neurosecretory cell types—A cells : These are the largest among the 5 types of cells identified and are present in the eyestalks, brain circumoesophageal connective ganglion and thoracic ganglion either singly or in combination with other types of NSC. They are oval or spherical in shape with 29 - 90 μ m diam. The nucleus is spherical or oval (12 - 35 μ m diam.). A single large nucleolus with unevenly distributed chromatin material is present, generally in

association with the nuclear membrane. During resting stage the cells are large in size and cytoplasm is densely packed with uniformly distributed coarse granules (Fig. 1). They exhibit secretory phases as judged by changes in the cytoplasm and nucleus. Secretory cycle starts with an increase in vacuolation, the cytoplasmic granules also diminish in number (Figs 2 and 3). In the final stage of activity the cytoplasm is occupied by a large vacuole and the nucleus is surrounded by a thin film of cytoplasm (Fig. 4). This is considered as the vigorous stage of secretion⁹. The gradual reduction of cytoplasmic granules and the increase in vacuolation suggest the possibility that some of the material that goes into secretion may be these granules.

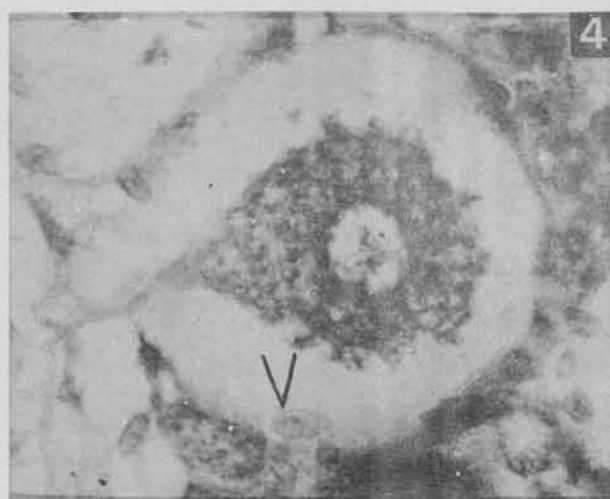
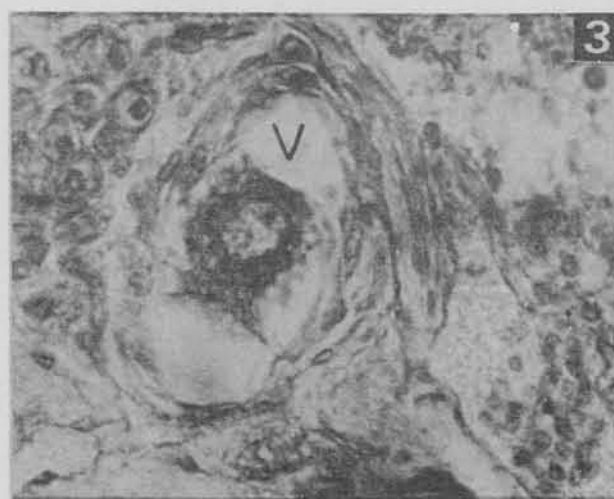
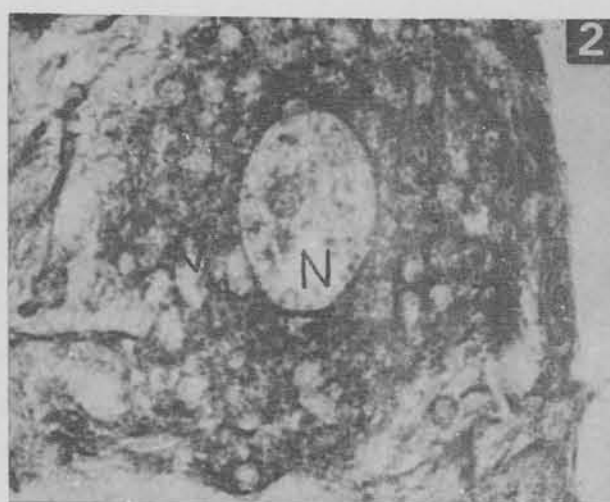
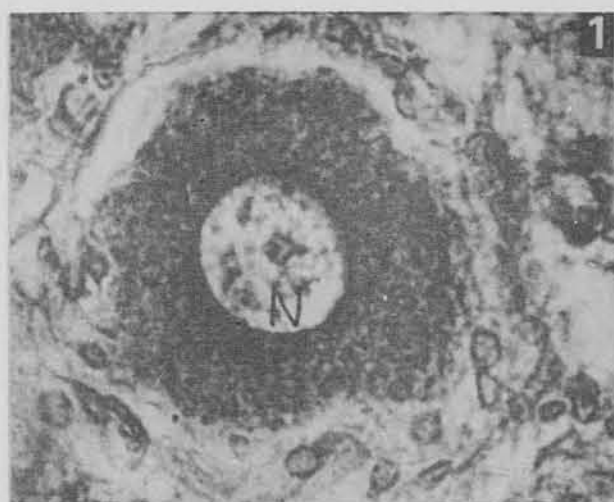
B cells : These are much smaller (13 - 45 μ m diam.) in comparison with A cells. Nucleus (7 - 18 μ m diam.) is prominent with a nucleolus. These cells are spherical or oval, mostly with a long axon, present singly or in groups in brain, eyestalks, circumoesophageal connective ganglion and thoracic ganglion (Fig. 5). B cells also display secretory activities similar to those of A cells. Vacuoles are usually restricted to the peripheral region (Fig. 6).

C cells : The cells are restricted to the anterodorsal region of thoracic ganglion where they are arranged in 2 groups. These cells (17 - 33 μ m diam.) with characteristic clumpy cytoplasm are devoid of vacuoles (Fig. 7). The nucleus measures 6 - 11 μ m diam. Secretory activities of the cells are not detectable.

D cells : The cells (10 - 20 μ m diam.) are scattered throughout the central nervous system. Nucleus is prominent and large (5 - 10 μ m diam.) compared to the size of the cell. Single prominent nucleolus is present in each cell. These cells do not exhibit any detectable secretory activity (Fig. 8).

E cells : These are the smallest of all the 5 types of cells observed, with scanty cytoplasm and relatively large nucleus. The cell boundaries are indistinct. They

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Figs 1 to 4—A type cells showing different stage of secretory activity ($\times 500$). Figs 5 and 6 - B type cells exhibiting secretory activity ($\times 500$) [N - nucleus, V - vacuole, AX - axon]

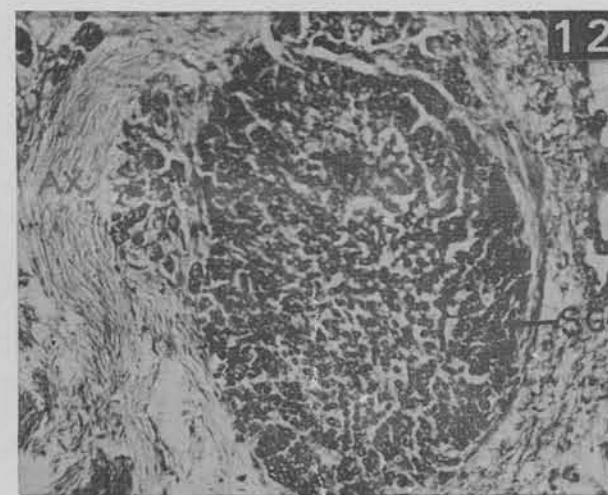
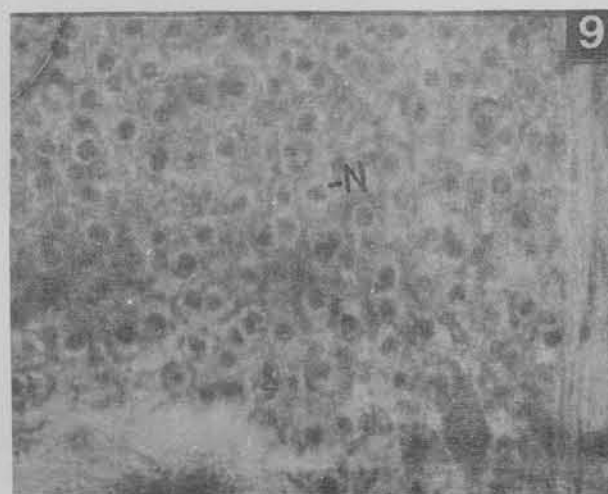
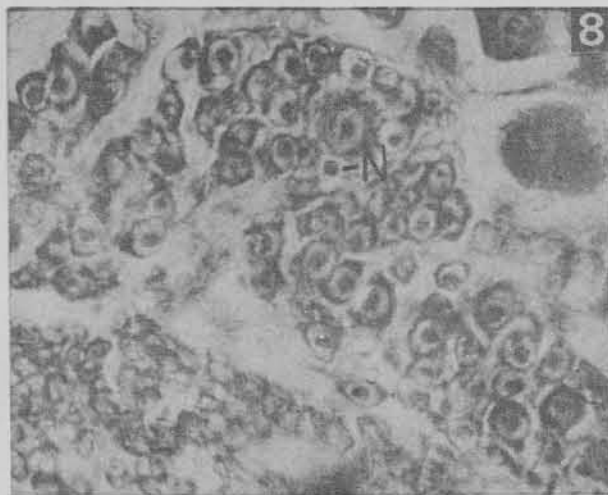
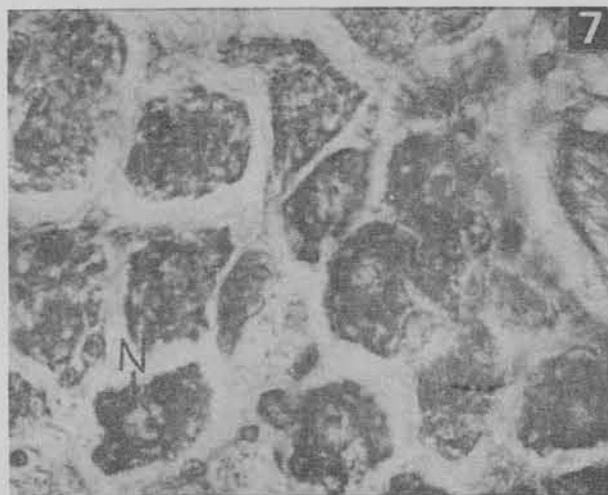


Fig. 7—C type cells with clumpy cytoplasm ($\times 500$). Fig. 8 - D type cells with large nucleus ($\times 500$). Fig. 9 - E type cells showing scanty cytoplasm and large nucleus ($\times 850$). Fig. 10 - Medulla interna ganglionic X - organ (MIGX) and axonal tract (AX) ($\times 200$). Fig. 11 - Medulla terminalis ganglionic X - organ (MTGX) I, II, III ($\times 200$). Fig. 12 - Sinus gland (SG) and axonal tract (AX) ($\times 150$) [N - nucleus]

are found in 2 large groups on the outer side of lobous olfactorius (Fig. 9).

Sinus gland X - organ complex—Eyestalk comprises of 4 optic ganglia, viz. lamina ganglionaries, medulla externa, medulla interna, and medulla terminalis. NSC are distributed throughout the optic ganglia. In medulla interna and medulla terminalis NSC are clustered in groups forming X - organs. In medulla interna there is one group of NSC on the abaxial aspect of the posterior region which sends axon to the sinus gland. Three types of cells, A, B, and D are present in this group. A cells outnumber the other 2 types. Axons from this cell group contribute bulbous endings to the sinus gland. This group of cells is identified as medulla interna ganglionic X - organ (MIGX) (Fig. 10). Besides the scattered NSC, there are 3 distinct groups of cells present in medulla terminalis. The first group, medulla terminalis ganglionic X - organ - I (MTGX - I) is located in the dorsolateral abaxial margin. A, B, and D cells are present in this group. D cells are more in number (Fig. 11). MTGX - II, the second NSC group, is situated at the dorsoabaxial margin. Of the 3 types of cells (A, B and D) present, D cells outnumber others (Fig. 11). Towards the posterior region of the ganglion, a 3rd group of NSC identified as MTGX III is situated. Besides numerous D cells, very few A and B cells are present in this group (Fig. 11).

Sinus gland, the principal neurohaemal organ in the present crab is located at the abaxial side of the anterior region of the medulla terminalis. The gland is composed of bulbous endings of axon fibres from NSC a blood sinus is found in association with the collection of nerve cell terminals. The sinus gland receives groups of axons from MIGX, and MTGX organs I, II, III. The axonal fibres form distinct axonal tract termed as the sinus gland X - organ tract. Careful examination of the tract revealed the presence of phloxinophilic secretory droplets throughout the tract (Fig. 12).

Discussion

All the 5 types of cells present in *C. lucifera* resemble the types described in the crabs *Parathelphusa jacquemonti*, *Scylla serrata*, *Portunus pelagicus* and in the lobster *Panulirus polyphagus*¹⁰⁻¹³. The pattern of distribution of NSC is similar to that found in *S. serrata* but differs from that in *P. jacquemonti* and *P. pelagicus* where E cells are present in thoracic ganglion also¹⁰⁻¹².

Three X - organs are located in the medulla terminalis of the crab *C. lucifera*. In addition to this, one X - organ is found to be present in medulla interna.

Carlisle and Knowles¹⁴ have reported the presence of an X - organ in the medulla interna of *G. lateralis*, and a similar group of cells in the medulla externa of *L. serratus*. The 3 MTGX - organs in the present crab are similar to those in *P. polyphagus*¹³ but differ from those in *Sesarma*¹⁵ and *S. serrata*¹¹. A single X - organ is identified in these 2 species, while in *P. pelagicus* 2 X - organs are present in the medulla terminalis¹².

A number of axonal fibres are terminating in the sinus gland from the NSC present in the eyestalks. The terminal end of these axons form bulbous structures. The sinus gland is the collection of these axonal terminals. In *C. lucifera* phloxine stained material is observed throughout the axonal tract that connect the X - organs to the sinus gland as observed earlier^{8,13,16}. It is evident from the present study and earlier observations that NS material is transported to the sinus gland through the axonal connection. It is reported that at least some of the hormones which influence the metabolic activities and chromatophore control are transported to the sinus gland¹⁷. It has been suggested that the NS materials undergo chemical alterations as they pass along the axons from the pericarya towards the sinus gland^{8,18}. Nuclei belonging to the neuroglial cells are noticed within the sinus gland of *C. lucifera*. Similar observations are also made on different decapods^{8,16}, but their role if any in the chemical transformation of the secretory materials remains doubtful.

References

- 1 Kleinholz L H, *Gen Comp Endocrinol*, **19** (1972) 473.
- 2 Silverthron S U, *Cell Tissue Res*, **115** (1973) 125.
- 3 Mumbach M W & Scheer B T, *Comp Biochem Physiol*, **56B** (1977) 47.
- 4 Nagabushanam R & Kulkarni G K, *Hydrobiologia*, **67** (1979) 113.
- 5 Keller R, *Z Verg Physiol*, **51** (1965) 49.
- 6 Rangneker P V & Phatak S S, *Ann Zool (Agra)*, **14** (1978) 27.
- 7 Chandy J P & Kolwalkar D G, *Comp Physiol Ecol*, (1985) (in press).
- 8 Lake P S, *Pap Proc R Soc Tasmania*, **105** (1970) 87.
- 9 Matsumoto K, *Biol J Okayama Univ*, **4** (1958) 103.
- 10 Nirmal H B, *J Biol Sci*, **7** (1964) 15.
- 11 Deshmukh R D & Rangneker P V, *J Biol Sci*, **16** (1965) 23.
- 12 Kolwalkar D G, *Studies on some physiological aspects of the marine crab, Portunus pelagicus (Rathbun)*, Ph D thesis, University of Bombay, 1974.
- 13 Phatak S S & Rangneker P V, *Rivista di Biologia*, **29** (1976) 59.
- 14 Carlisle D B & Knowles F G W, *Endocrine control in crustaceans*, (Univ Press, Cambridge) 1959, pp 150.
- 15 Enami M, *Biol Bull*, **101** (1951) 241.
- 16 Smith G & Naylor E, *J Zool*, **116** (1972) 313.
- 17 Knowles F G W & Carlisle D B, *Biol Rev*, **31** (1956) 396.
- 18 Bliss D E, Durand J R & Welsh T H, *Z Zellforsch*, **39** (1954) 520.