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A Systematic Study of Neuroendocrine system of millipede *Anoplodusmus tanjoricus* (Pocock) (Myriapoda : Diplopoda)

KEYWORDS: Millipede, neurosecretory cells, distribution, Anoplodesmus.

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ABSTRACT *In the millipede Anoplodesmus tanjoricus dense neurosecretory material was observed in the brain .the structure of the neurosecretory system was studied. In the brain of adult millipede Anoplodesmus Tanjoricus three types of neurosecretory cells, designated as A,B,C were distinguished in the brain. The A cells were differentiated into A1, A2, A3, A4 and C cells into C1 and C2 types. The types differed from each other in their cytomorphology, stainability, distribution, neurosecretory activity and mode of discharge of secretary material. The neurosecretory cells became more evident, scattered among the neuroblasts along the periphery of the protocerebrum. The neurosecretory cells were distinguished from other neurons because of their large size and presence of neurosecretory material.*

INTRODUCTION

The neurosecretory system of myripodes is through be limited to a group of neurosecretory cells situated on either side in the protocerebrum closed to the globuli cells, the product of which in the end organ, the cerebral gland(Gabe1952,1954a,b,;1956a; Plam1956). It also believed that the cerebral glands , besides being a storage gland and presumably release centres also elaborate another secretion Of their own , which is distinguishable from that produced in the neurosecretory cells of the brain due to difference in its stainability. The system is more extensive . at least in Jonespeltis, where additional neurosecretory cells and a new storage centre (connective body) have been located (Prabhu 1959, 1961), though the secretary role of the cerebral glands in the animal has been found to be improbable. However, recent work has shown that the system is extensive in Anoplodesmus tanjoricus also.

MATERIALS AND METHOD

Adult millipede Anoplodesmus tanjoricus of body length 35-40mm were collected from fields at Amravati region .They were kept in laboratory and feed cucumber, apple, vegetables and rotten leaves with plenty of humus. For the histological procedure the brain was removed from the healthy and fully matured millipede by careful dissection. The animals were dissection fixative or in normal saline(0.7% Nacl).In either case the nerve ring, with the cerebral gland and connecting bodies intact, was transferred to Bouin's fluid in which acetic acid was replaced by 0.5% trichloroacetic acid. Paraffin section if 3-10µ were cut and stained in Gomori's chrome alum haematoxylene - phloxine and mounted in DPX.

OBSERVATIONS AND RESULTS

The neurosecretory cells became evident, scattered among the neuroblast, along the periphery of the protocerebrum. The neurosecretory cells could be identified from other neurons because of their large size and the presence of material, stainble with Gomori's chrome alum haematoxylene -phloxine. The cells were found to be either oval, spherical or pear shaped. They were distributed in the medium, lateral and ventral aspect of the brain. The spherical cells, four to five in number had an average diameter of 14µm.The oval shaped cells five to six in number, were 10-15 µm (breadth and length respectively).The pear shaped cells two to three in number, were 9 to 12µm in diameter. Present studied on a local diplopod, Anoplodesmus has shown that the neurosecretory system in this animal consist of various, distant type neurosecretory cells distributed in the brain, the ventral nerve cord and the visceral ganglion together with the cerebral gland and connective bodies, which from the storage the neurosecretory material produced in these cells. There are 3 distinct types of neurosecretory cells, namely as A , B, C. A and C cells are divided into subgroup based on their morphological characteristics.

A Cells pattern:

A cells :- A cells are pyriform with abundant golgi bodies, no mitochondria and are visible with the light microscope and show reduction in the amount of the nissal substance as secretary activity progresses. They have distinct axons. The secretary granules of these cells are differentiated by Gomori's chrome alum haematoxylene- phloxine stain. The A cells are of four different pattern such as A_1,A_2,A_3 and A_4.

i) A_1 cell

A_1 cells are the largest of all the cells and there measure about 25-26µm in diameter. They are oval or pyriform in shape with axon. Nucleus is 17µm to 18µm in diameter oval in shape and especially nucleolus shows an increase in size during neurosecretion. The secretary granules are stainable with Gomori stain. They are blue black in color. They discharge their secretary products peripherally as well axonally in the form of small granules.

ii) A_2 cell:

They are smaller than the A_1 cells. They are oval or pryiform in shape measure about 22-23µm in diameter. Their nucleus is oval in shape and is 14µm to 15µm in diameter. They discharge intra-nuclear secretary products axonally and have no peripheral discharge. The granules Observed are stainable with chrome alum haematoxylene- phloxine.

iii) A_3 cell:

They are smaller than the A_1 and A_2 cells . They are pryiform in shape measuring about 20 to 21µm in diameter. Their nucleus is 14µm to 15µm in diameter. They discharge intranuclear secretary product in peripheral and have no axonal discharge.

iv) A_4 cell:

In size these are comparable smaller than first three, that is A_1,A_2 and A_3 cells. They are oval in shape and their secretary products from large discrete irregular clumps in the cytoplasm which stain deep blue with chrome alum haema-

Volume : 3 | Issue : 11 | Nov 2013 | ISSN - 2249-555X

toxylene- phloxine. The nuclei are oval in shape. These are sometimes very much elongated or even lobed to the extent suggesting amitotic division. Each lobe is having a nucleolus. However, the nuclei actually do not appear to divide. The four patterns of the A cells have secretary granules which are clearly differentiated blue in color by Gomori's chrome alum haematoxylene-phloxine stain. Their discharge is probably axonal in the from of submicroscopic particles. It may be noticed that the differences among the subtypes of A cells are less stricking than those of C cells.

II) B Cells:
B cells are smaller than A cells. These are pyriform and have never been observed to exceed 16µm -17µm in diameter. The cell membrane is not especially stainable. B cell have small nucleus with deeply staining chromatin network of red color. Their oval nucleus measures about 7 to 8µm in diameter.

A distinct nucleolus could not be marked in these cells. The nuclei are very often seen pushed to one side of the cell, closely opposed to the cell membrane, usually at the axonal end of the cell. Sometimes the nuclei are in distingusiable due to the cell being practically filled with dense colloid mass intensely axocarminophilic and phloxinophilic with chrome alum haematoxylene- phloxine. Intra-nuclear secretion is absent, as the nuclei are clearly distinguishable without any droplets, even when the whole cytoplasm is practically covered by a mass of colloid or during the growth of smaller colloid droplets into larger masses.

III) C Cells:
The 'C' cells are one of the three types of neurosecretory cells of millipede, Anoplodesmus tanjoricus found distributed in the brain. In general they are small.15 µm to 17 µm in diameter, pyriform or spherical in shape differing from other neurosecretory cells by their totally different chromatin rich nuclei. The nucleus, measuring about 5µm to 6µm diameter, is either spherical or oval usually dense chromatin material. The secretary granules of the cytoplasm are stainble, deep blue with chrome alum haematoxylene- phloxine. These granules thus represent cystrine/ cysteine material, according to the presence or absence of axonal transport of neurosecretory material. The C cells could be distinguished into two subtypes C_1 and C_2 cells.

i) C_1 Cells: The C_1 cells are distributed along the ventral and mesial side of the brain. They are small, 15µm to16µm in diameter. They are situated along the middle line and either sides of the brain. Thus an arch of highly stainable tracts could be seen along the inner margin of the brain encircling the dorsal boundary of the oesophagus. The axons of the majority of the C1 neurosecretory cells are directed towards the lateral oesophageal connectives and they end in a pair of neurohaemal organs called connective bodies.

ii) C_2 cell:
This cell is distributed at random in different regions of the brain. They have no distinct cellular processes to name as exons. C_2 cell are scattered uniformly in the brain. They measure about16µm to17µm in diameter. They discharge in peripheral, evidently they have no end organ. They occur scattered irregularly among other neurons and neurogalia of the brain and ventral nerve cord. They also occur though infrequently, In the neurophile region and liberate secretary products probably from the cell periphery.

Axonal path of neurosecretory cells:
Axons of the A_1 type of cells on each side travel in the from of a bundle towards the median line and take a curve to the periphery, backward and lateral. Much of the path therefore lies in the horizontal plane. Later these tracts curve downward and posterior -lateral and emerge out of the brain as the anterior nerve of the cerebral gland. fibres from the A pattern cells are situated in the "Visceral ganglion", that enter the brain the travel along the periphery , emerge again close together, as the posterior nerve of the cerebral gland. The "cerebral gland" is not a gland but is made up of mainly swollen nerve fibre termination of the anterior and posterior nerve to the "cerebral gland".

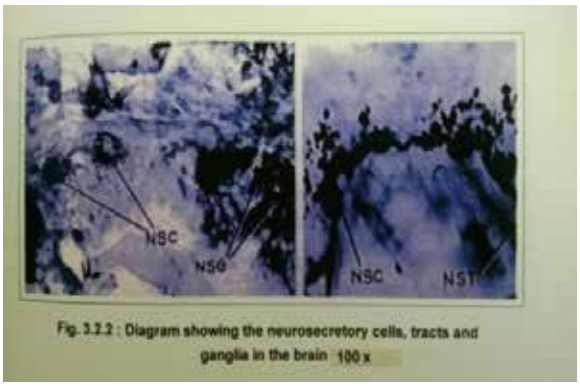

Fig. 3.2.2 : Diagram showing the neurosecretory cells, tracts and ganglia in the brain 100 x

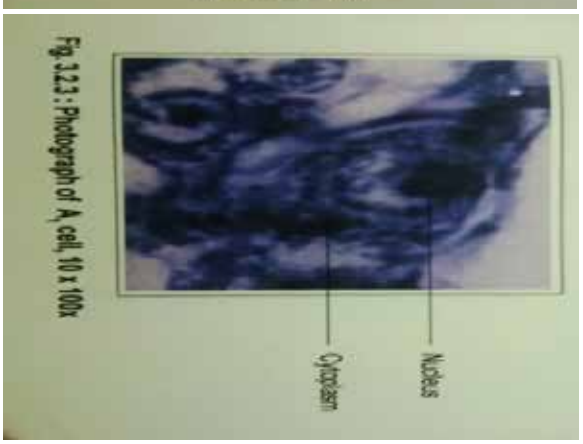

Fig. 3.2.4 : Photograph of A_2 cell, 10 x 100x

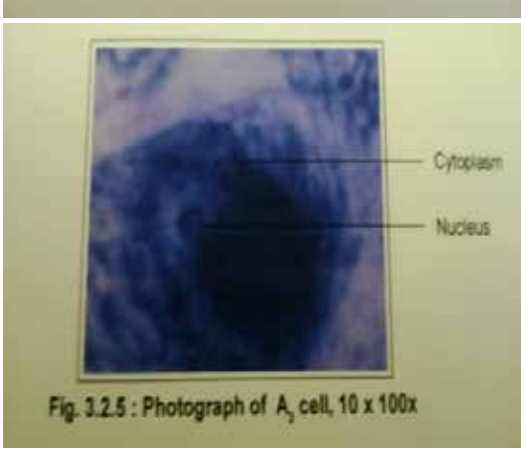

Fig. 3.2.5 : Photograph of A_3 cell, 10 x 100x

Volume : 3 | Issue : 11 | Nov 2013 | ISSN - 2249-555X

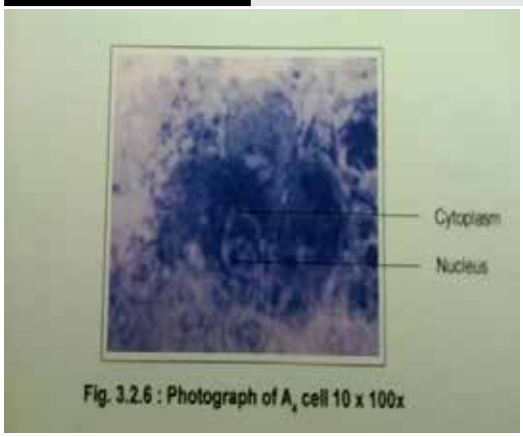

Fig. 3.2.6 : Photograph of A_4 cell 10 x 100x

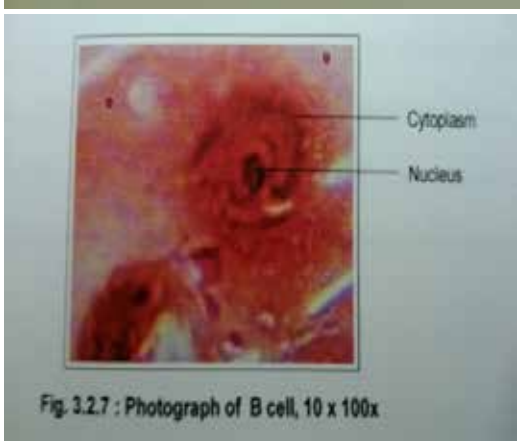

Fig. 3.2.7 : Photograph of B cell, 10 x 100x

Fig. 3.2.8 : Photograph of C_1 cell, 10 x 100x

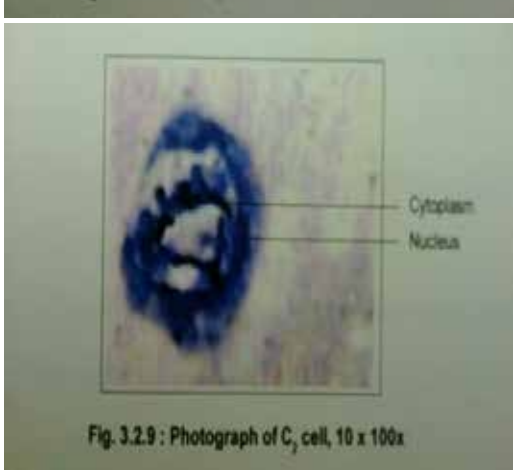

Fig. 3.2.9 : Photograph of C_2 cell, 10 x 100x

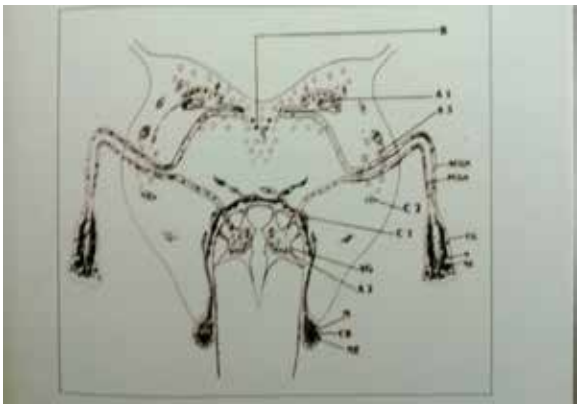

Diagram showing the neurosecretory cell types in the brain and visceral ganglion. A1,A2 and A3 :three patterns of Acells; B : B cell; C1 and C2subtypes of C cell; VG : Visceral ganglion; CG : Cerebral gland; N : Nucleous; NE : Nerve ending swollen with accumulated neurosecretory material ; CB : Connective body; ACGN and PCGN : Anterior and Posterior nerve of the cerebral gland.

DISCUSSION

The histomorphological structure of cerebral ganglia, visceral ganglia, suboesophageal ganglion, ventral ganglia and neurosecretory organ in anoplodesmus tanjoricus are similar to that of Jonespeltis splendidus described by Prabhu 1959, 1961, 1964, 1962a) the distribution of neurosecretory tract in species under study correspondence to that of other millipede species in the idea that neurosecretory tract arising from the brain and pass towards suboesophageal ganglion via the lateral oesophageal connective an identical neurosecretory tract arising from the brain and passing into perioesophageal collar going to the ventral nerve chain has been described in diplopod, polydesmus testaceuls, Gersch(1958) however in the symphylid scutigerella pagesi, Juberthie-Jupean (1961)described two tracts one originating from protocerebral neurosecretory cells and other form deutocerebral neurosecretory cells in the millipede , Polyxenus have only one neurosecretory path way in the brain arising from the deutocerebral neurosecretory cells has been disrcibed, Nguyen- Duy-Jaquemin(1971c).

Pattern of distribution of neurosecretory tracts and neurosecretory cells in millipedes species understudy Anoplodesmus tanjoricus, Jonespeltis splendidus , Prabhu(1962a) and in millipede Orthomorpho gracilis Sahli(1966) is more or less similar. The neurosecretory system of Anoplodesmes tanjoricus consist of neurosecretory cells in brain, suboesophageal ganglion and ventral ganglia. It is associated with the neurohaemal organs. Three types of neurosecretory cells in the brain and cerebral glands, connective bodies and visceral ganglia are A,B,C cells.The A cell type is A1,A2,A3,A4 present and distributed in the protocerebral lobe, cerebral ganglia, suboesophageal ganglia and visceral ganglia.B cell present in the dorsal part of the brain.C1and C2 cells are present on the ventral side, mesial side, different region of the brain and ventral nerve cord is more similar to that of Jonesplitus splendidus Prabhu (1961, 1962a) and Lithobius forficatus palm (1956) and Chez les chilopodes Gabe(1956). Another report of similar studies in the Chilopod Geophilus longicornis by Ernst (1971). The histomorphological structure of cerebral ganglia, visceral ganglia, suboesophageal ganglion, ventral ganglia and neurosecretory organ in Anoplodusmus tanjoricus are similar to that of Jonespletis splendidus described by Prabhu (1959,1961, 1964, 1962a). The distribution of neurosecretory tract in species under study corresponds to that of other millipede species in the idea that neurosecretory tract arising from the brain and pass towards suboesophageal ganglion via the lateral oesophageal connective and identical neurosecretory tract from the brain and passing

Volume : 3 | Issue : 11 | Nov 2013 | ISSN - 2249-555X

into perioesophageal coller going to the ventral nerve chain has been described in diplopod, polydismus testaceculs, Gersch(1958) however in the symphylid scutigerella pagesi, Juberthie- Jupeau(1961) described two tract, one originating from protocerebral neurosecretory cells and other from deutocerebral neurosecretory cell in the millipede only one neurosecretory path way in the brain arising from the deutocerebral neurosecretory cell has been described Nguyen-Duy-Jaquemin(1971c).

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