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Morpho-anatomy of x-organ, sinus gland complex in eye stalk of *Sartoriana spinigera* (WOOD MASON 1871)

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Abstract : In the present investigation it was observed that eyestalk of *Sartoriana spinigera* contained optic ganglia, on the distal part of optic ganglia compound eye was present and from the proximal end optic nerve arose. Eyestalk contained four ganglia arranged in a consecutive manner *i.e.* just beneath the compound eye lamina ganglionalis was present, then medulla externa, medulla interna and medulla terminalis were observed. Sinus gland was located on the dorsal side of the medulla externa just beneath the compound eye and the X-organ on the ventral side of the medulla terminalis. Paraldehyde Fuchsin positive neurosecretory cells were observed in the X-organ sinus gland complex. Sinus gland was ellipsoid structure which was bent on itself at the anterior extremity of the eyestalk . The sinus gland was surrounded by a large outer sinus and a small inner sinus which appear to be in the form of blood vessel. Neurosecretory cells in X-organ were Paraldehyde Fuchsin positive which entered obliquely in the Sinus gland.

Keywords : *S. spinigera,* eyestalk, X-organ/sinus gland, Neurosecretory cells.

INTRODUCTION

Crustacean growth and development are characterized by the periodic shedding (molting) and fresh replacement of the exoskeleton. The cellular events that lead to molting are stimulated by the steroid hormones produced by paired endocrine glands the Y organs. The X-organ sinus gland complex is present in the eyestalks.

This X-organ sinus gland complex in the eye stalk is involved in growth and reproduction of crustaceans. Several physiological processes are controlled by neuro hormones from the eyestalk. The neuro hormones synthesized by a proximal cluster of neuroendocrine cells were called the X-organ. They are conducted distally via axons regrouped in tracts to the storage and release in neurohemal organ the sinus gland . Soyez(1997), Van Herp (1998), Lacoumbe (1999) reported a wide variety of 8-9.5 K.D peptides isolated from the XO/SG complex and exhibiting a sequence homology and overlapping biological effects. Eye stalk hormones from the X-organ sinus gland system are involved in pigment condensation and dispersion in chromatophores, movement of distal retinal pigment in compound eye, inhibition of molting, inhibition of ovarian development, control of blood sugar level and osmotic pressure regulation.

Neurosecretory cells in each ganglia are classified into several types according to stainability with Gomori chrome alum haematoxylene Fuchsin and paraldehyde Fuchsin method. In present investigation morphology and anatomy of eyestalk and its endocrine glands were studied using various histochemical stains.

MATERIALS AND METHODS

Live specimens of both the sexe were collected from the different sites of Ranchi. Some of them were preserved in 5% formalin at the spot. Now both, alive as well as preserved specimens were carried to the laboratory. Live specimens were kept in glass aquaria containing and water. The animals were regularly fed with fish food and sometimes with small pieces of fish.

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The water was also changed after 2-3 days. Dead animals and remaining food pieces werw also removed to prevent infections to rest of the animals.

For morphological study of *Sartoriana spinigera* both sexes were observed externally. For anatomical study, the live specimens of both sexes were sacrificed by immersing the animal into 5% formalin solution. After killing, the animals were washed under running tap water. It was dissected by obeying the standard method as suggested by Whitehouse and Grove(1956) to trace out the reproductive system of male and female specimens.

For the localization of neurosecretory cells in eyestalk the live crabs were sacrificed by removing the eyestalk with fine scissors. The chitin were removed carefully and fixed in aqueous Bouins' fluid for 24 hours. After 24 hours eyestalk were washed by 70% alcohol to remove the excess stain. Tissues were dehydrated . Now tissue were oxidized in freshly prepared acidified $KMnO_4$ solution. Bleached in 4% Sodium bisulfate solution until tissue were perfectly white. Now tissues were stained by Paraldehyde Fuchsin for 10-20 min depending upon the size of tissue. Tissue were washed with alcohol then differentiated with Acid alcohol for 1-2 min and cleared in Cedar Wood Oil for 10 min and passed through Xylene for 5 min and mounted in DPX. Photomicrograph were taken for study.

OBSERVATIONS AND RESULT

Fig 1 showed the optic ganglia present inside the exoskeleton, on the distal part of optic ganglia compound eye is present and from the proximal end optic nerve arise.

Fig 2 showed that eyestalk contained four ganglia arranged in a consecutive manner i.e just beneath the compound eye, lamina ganglionalis is present then medulla externa, medulla interna and medulla terminalis are observed.

Fig 3 showed the location of sinus gland on the dorsal side of the medulla externa just beneath the compound eye and the X organ on the ventral side of the medulla terminalis. Optic nerves arose from the medulla terminalis.

Fig 5 showed whole mount of optic ganglia using Paraldehyde Fuchsin stain. Paraldehyde Fuchsin positive neurosecretory cells were seen in purple colour. The neurosecretory cells were present in X organ. Sinus gland was also Paraldehyde Fuchsin positive which was bent on itself at the anterior extremity of the eyestalk. The gland is surrounded by a large outer sinus and a small inner sinus which appear to be in the form of blood vessel. It is ellipsoid structure. Positive reaction with Paraldehyde Fuchsin showed the presence of neurosecretory materials. Neurosecretory cells were Paraldehyde fuchsin positive, which were present in X organ entered, in the sinus gland. Longitudinal section of eyestalk contained four ganglia arranged in consecutive manner.

i. Lamina ganglionalis
ii. Medulla externa
iii. Medulla interna
iv. Medulla terminalis

Fig 5 showed the diagrammatic representation of the ganglia and X organ Sinus gland in eyestalk. The gland was surrounded by a large outer blood sinus and a smaller inner sinus which appeared to be in the form of a blood vessel.

DISCUSSION

The freshly dissected X-organ appeared as a pale mass. In *Sartoriana spinigera* it was located at the ventral base of the medulla terminalis. Sinus gland was typical neurohaemal organ and the site of storage and release of neurosecretory substances

through the axonal endings which originated from the X-organ. The axonal endings of the neurosecretory cells were also present in optic ganglia and brain. The axonal endings contained numerous neurosecretory granules and secretion occured into the blood sinus, haemocoel. Sinus gland appeared on the dorsal aspect of the medulla externa in *Sartoriana spinigera*. Its shape is flat slightly doom shaped.

Similar observations and results were almost observed by other scientist in the past. Hanstrom(1937,1947) studied *Squilla mantis* and pointed out that it contained inverse type of sinus gland found in a number of Decapods. Hanstorm(1947) described that the sinus gland was situated between the medulla externa and medulla interna but according to Carlisle and Passano(1955a) it occupied a position distal to lamina ganglionalis but in the present study in *Sartoriana spinigera* the sinus gland is situated at dorsal aspect of medulla externa (Fig -2&3)

Ruby Daniel(1958) from the examination of sections of the eyestalk *of Squilla holoschista* , it was reported that the sinus gland lied on the dorsal side of the optic ganglia between the lamina ganglionalis and medulla externa over which it extends proximally. As in Decapoda in *Sartoriana spinigera* also ganglia were found in vertical series one behind the other (Fig 2).As in *Squilla mantis* in *Sartoriana spinigera* also the gland is completely separated from the neurilemma covering the optic ganglia (Fig 5) and unlike in *Squilla demaresti* in which the gland is partly connected with the neurilemma. Ruby Daniel from the examination of sections of the eyestalk of *Squilla holoschista* , reported that the sinus is triangular in shape and measures about 0.5 mm in length and about 0.65 mm. across the broadest part. But in present investigation it is curved with the inner side in contact with the portions of the optic ganglia (fig-5) and it is doom shaped.

Observation reported by Ruby Daniel[(1958)] showed that at the anterior extremity of the gland it is bent on itself to form two well marked folds.

In *Sartoriana spinigera* also sinus gland is slightly bent doom shaped structure , but folds have not been observed. Such folds have not been described in any other crustacean also and appeared to be peculiar to this type. According to her these folds lie on each side of the median longitudinal plane of the gland. A blood vessel passed in between the folds. The extensions of the folds enclosed the blood sinus partially. Similarly to *Sartoriana spinigera* from present investigation the gland is surrounded by a large outer blood sinus, and a smaller inner sinus which appeared to be in the form of a blood vessel which occurred between and partially enclosed by the folds of the gland. The nerve supplying the gland entered on its inner side which is concave corroborating *Sartoriana spinigera.* The structure less membrane or the limiting membrane is found covering the gland on the convex side and is absent on the inner concave side. By possessing the structureles membrane on the outside and the nerves on the inside, it resembles the gland found in the *Anomura((Honstrom1939).*

In the present investigation the X-organ lies on the ventrolateral side in a depression formed by the medulla terminalis. In being situated in *Sartoriana spinigera* distal to the medulla terminlis it resembles the X-organ of *Homarus americanus(Welsh1951)* and *Palaemonetes vulgaris* and differs from that of *Squilla mantis* as well as those of *Sesarma dahaan(Enami1951 b), Cambarus bartoni* and many species of Brachyura. In *Sartoriana spinigera* the X-organ extends on the medulla terminalis from the ventral ide to the dorsal side in an oblique manner, and is undivided unlike in *Squilla mantis.* The only indication of a division, if any, is a light constriction in the distal region which gives it a lobed appearance. It has the characteristic shape of a cluster of grapes surrounding the thick nerve which descends through the medulla terminalis recalling the structure found in *Natanti*a(Hanstrom 1939).By comparing with it, the X-organ of *Sartoriana spinigera,* shows more advanced features and resembles in a number of respects that found in *Homarus* and *Squilla holoschista*.

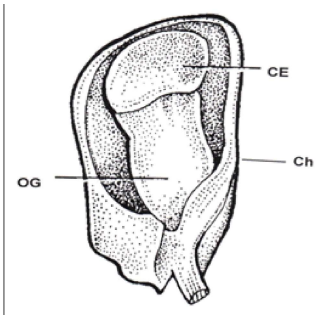

Fig. 1 Diagrammatic representation of the Optic ganglia in the Eyestalk

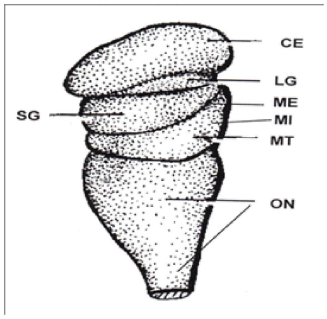

Fig. 2 Diagrammatic representation of the Optic ganglia in the Eyestalk and sinus Gland.

Abbreviations – Compound Eye (CE), Chitin (Ch), Lamina Ganglionalis (LG), Medulla terminalis (MT) Medulla Externa (ME), Medulla Interna (MI), Optic Nerve(ON), Optic Ganglia (OG) and Sinus Gland (SG)

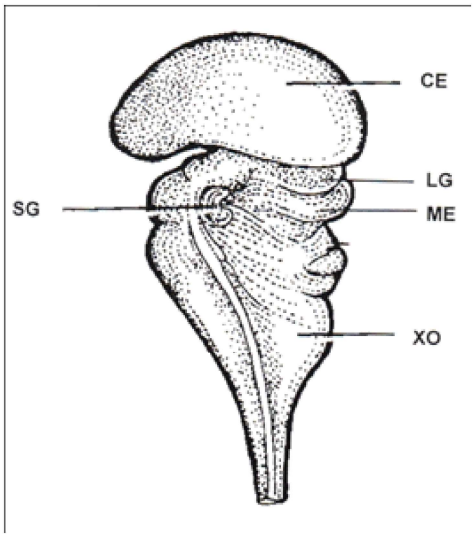

Fig. 3 Diagrammatic Representation of External Side view of the left Eyestalk showing locations Of SG, MT, CE and ON.

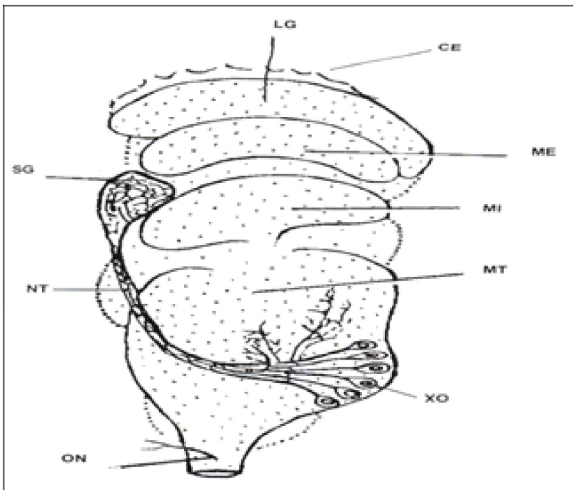

Fig. 4 Diagrammatic Representation of Ganglia and X-organ Sinus gland Complex of the *S. spinigera*.(Dorsal View).

Abbreviations – Compound Eye (CE), Chitin (Ch), Lamina Ganglionalis (LG), Medulla terminalis (MT) Medulla Externa (ME), Medulla Interna (MI), Optic Nerve(ON), Optic Ganglia (OG) and Sinus Gland (SG)

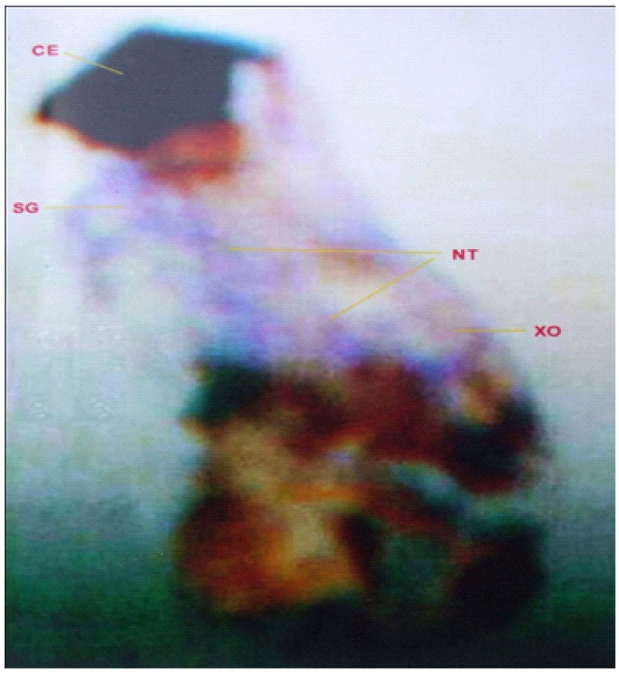

Fig 5. Photograph showing distribution of Paraldehyde Fuchsin positive Neurosecretory cells in Eyestalk of *S. spinigera.*

Abbreviations – Compound Eye (CE), Nerve Tract (NT), X-organ (XO), Sinus Gland (SG)

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