Neuroanatomy of a Dactylogyrid monogenean, from gold fish *Carassius auratus*, Nilsson, from Meerut (U. P.), India

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Abstract

Chemical named 5-bromo indoxyl acetate has been used to describe the nervous system of anoviparous Dactylogyrid monogenean *Pellucidhaptor Price and Mizelle* (1964), a gill parasite of *Carassius auratus*. Central nervous system consists of paired cerebral ganglia from which anterior and posterior neuronal pathways arise. These neuronal pathways are interlinked by cross connectives and commissures. Paired dorsal, ventral and lateral nerve cords emanate from the cerebral ganglia, connected at intervals by transverse connectives. Huge arrangement of dorsal, ventral and lateral nerve cords and their innervations have been examined. Peripheral nervous system (PNS) includes innervations of the alimentary tract, reproductive organs and attachment organs (anterior adhesive areas and haptor). Both the CNS and PNS are bilaterally symmetrical, and better developed ventrally than laterally and dorsally.
Introduction

Monogeneans are tiny, non-segmental and largest group of parasitic trematodes found primarily on skin or gills of fish. Monogeneans attach to their hosts using hooks, clamps and a range of other alternative structures. They are usually capable of dramatically elongating and shortening their body while moving. General nervous system of trematodes is composed of cerebral nerve cell clusters called ganglia in anterior and posterior nerve trunks. Nervous system of monogeneans is basic and ladder like. It consists of a pair of cerebral ganglia or brain and one pair of longitudinal nerve cords with transverse connections located at regular intervals. The central nervous system with its anterior ganglion (brain), sensory eye spots and some other structures indicate head specialization or cephalization. The central nervous system with longitudinal cords and ganglia (ladder type) is more highly developed than the nerve net of coelenterates.

Various authors have used the activity of cholinesterase (ChE) as indirect histochemical evidence of the presence of acetylcholine (ACh) in the nervous system of monogeneans (Halton and Jennings 1964; Halton and Morris 1969; Cable et al. 1996; Zurawski et al. 2001) using an indoxyl acetate as substrate for carboxylic esterases. Other studies have employed acetylthiocholine iodide (AcThI) as substrate (Rahemo and Gorgees 1987; Buchmann and Mellergaard 1988; Buchmann and Prentø 1989; Reda and Arafa 2002) to provide a more specific demonstration of ChE itself.

Like all Dactylogyrids, monogeneans of the genus *Pellucidhaptor* (Singh et al., 2003) are oviparous. It is a gill parasite of the aquarium gold fish, *Carassius auratus*. Present work involved a similar approach to explore the nervous system of *Pellucidhaptorkritskiya*. The central nervous system (CNS) and peripheral nervous system (PNS) were found to be highly reactive for cholinesterase and stained extensively in a dark blue colour.

It is hoped that such a study will help researchers to find an anthelmintic drug that may specifically target neural elements of the parasite without substantial side effects on the host.

Materials and Methods

Fishes for the present investigation were collected from aquarium fish vendors of Meerut. Worms were collected by method suggested by Mizelle (1936). In all 40 specimens were studied. Worms collected in live condition were washed thoroughly with cold distilled water. Study of nervous system was made with the help of histochemical localization of esterases, one of the very common neuro pharmacological elements, commonly found in the nervous system of all animals including monogeneans as suggested by Halton and Jennings (1964). Microphotographs were taken with the help of Motic image plus Software.

Results

The CNS consists of a thick, curved mass of paired cerebral ganglia (cg) located...
ventrally just anterior to the pharynx. Cerebral ganglia formed a circular path around the pharynx. Several thick projections extend from the anterior median region of the cerebral ganglia, imparting to this organ a butterfly-like appearance. Each projection gives rise to cerebral nerves (cn), which extend anteriorly to enter the head lobes where they innervate the anterior adhesive areas. Inside the pharynx few pharyngeal nerves (phn) were also detected.

Two thick ventral nerve cords (vnc) arise one from each ventro-lateralside of the cerebral ganglia and run posteriorly one on each side of the body where each join a prehaptoral ganglion (phg1). Another pair of prehaptoral ganglia (phg2) is located either side of the body a short distance posterior to the prehaptoral ganglia (phg1) in the peduncular region. Both phg1 and phg2 on each side of the body are connected via two ganglionic connectives. In the seminal vesicle region, two considerably thin branches arise, one from each ventral nerve cord, and reconnect to it. Another thin branch originates from the posterior region of each ventral nerve cord in the post-ganglionic region and reconnects to it. Five ventral transverse connectives (vc1–vc5) were detected, at the level of cirrus, egg, anterior to the ovary and between Phg1 and Phg2 respectively.

Staining for cholinergic elements revealed the presence of thirteen pairs of large neurons distributed bilaterally down the main body of the worm. Five pairs of these cells are located on the ventral side of the body (v1–v5) and eight pair on the dorsal side (d1–d8). Most of these cells are bipolar except fifth ventral pair and second dorsal pair which are multipolar. The first, second and third pair of ventral cell body (v1–v3) is positioned posterior to the egg, the fourth pair (v4) at the level of ovary, fifth multipolar pair (v5) in the peduncle region. The first dorsal pair (d1) of cell body is located at the level of egg, second pair (d2) is just behind it, third pair (d3) at the level of ovary and fourth to eighth dorsal pair (d4–d8) in the peduncular region. One lateral transverse connective (11) was also detected in the peduncle region anterior to the phg1. At regular intervals, the ventral nerve cords communicate with the lateral nerve cords by means of seven pairs of ventro-lateral connectives (vlc1–vlc7). Two thin dorsal nerve cords (dnc) arise from the postero-median region of the cerebral ganglia; extend posteriorly between the ventral nerve cords where they connect with the anterior pre-haptoral ganglia (phg1). The dorsal nerve cords are cross-linked by seven dorsal-transverse connectives (dc1–dc7).

Two considerably thin lateral nerve cords (lnc) arise from the postero-lateral region of the cerebral ganglia and run posteriorly, one adjacent to each lateral margin of the body where they join the phg2.
The haptor is extensively innervated by two outer (ohn) and two inner (ihn) haptoral nerves. The outer haptoral nerves originate from phg2 & inner haptoral nerves from phg1. The outer and inner haptoral nerves run ventrally in a posterior direction before branching into a plexus of numerous fine nerves in the posterior region of the haptor and innervates to marginal hooklets. Two dorso-lateral haptoral nerves (dhn), arising one from each lateral nerve cords, run posteriorly and attached to numerous fine nerves of outer and inner haptoral nerves.

The central nervous system (CNS) of *Pellucidhaptor* as revealed by cholinergic staining conforms to the basic orthogonal pattern described for other monogeneans. Typically, the PNS innervates the alimentary system, reproductive organs, attachment organs and sub-tegumental muscles.

**Discussion**

The present study has revealed that central nervous system of *P. kritskyia* consists mainly of a mass of cerebral ganglia and three pairs of ventral, lateral and dorsal longitudinal nerve cords connected by transverse connectives and commissures; it is better developed ventrally than dorsally and laterally. The main structure of the nervous system comprising cerebral ganglia, posterior and anterior nerve trunks and connecting commissures is analogous with its composition in other monogeneans (e.g. Halton & Morris, 1969, Venkatanarsaiah, 1981, Reuter, 1987). Typically, the peripheral nervous system innervates the alimentary system, reproductive organs, attachment organs and sub-tegumental muscles. Halton and Gustafsson (1996) reported that the peptidergic pathways in platyhelminths more follow closely those of the cholinergic system. The present study has revealed that cholinergic components of the nervous system of *P. kritskyia* are more extensive than the aminergic components.

The nerves supplying the head lobe nerves of *P. kritskyia* may play a role in temporary attachment of the adhesive sacs during locomotion and feeding and coordinating movements involved in the release of secretory bodies from the gland cells of the anterior adhesive apparatus. A ring commissure was detected around the mouth of *Chauhanellus indicus* (Rastogi, et.al. 2007). In the present study, a similar commissure is not observed in *P. kritskyia*. However, in all monogeneans studied hitherto, and oral ring commissure was found. Rohde (1968) suggested that the oral commissure is a character, which distinguishes the nervous system of the Monogenea from that of Digenea.

In the monogenean parasite *P. kritskyia*, the cholinergic nerves supplying the anterior region of the gonopore help coordinate events involved in copulation and ovulation. Cholinergic innervations of the reproductive organs of *P. kritskyia* may perform a similar role. The haptor of *P. kritskyia* is richly innervated with cholinergic element, suggesting a major role in motor function for the haptoral nerves.
Explanation of Plates

Plate 01: Diagrammatic representation of nervous system of Pellucidhaptor kritskyia Singh et al., 2003

Plate 02: Photomicrographs of nervous system of Pellucidhaptor kritskyia Singh et al., 2003.

Nerves in: (a) prohaptor region; (b) region of cirrus; (c) region of female gonopore; (d) region of gonads; (e) prepeduncular region; (f) peduncular region and (g) haptor region.

References


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PLATE 01
PLATE 02