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Sex specific anatomical variation in the organization of Jacobson's organ (VNO) with respect to its olfactory function in common worm snake *Typhlina bramina*

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Abstract : Jacobson's organ also known as vomeronasal organ (VNO) has an important function in prey detection feeding and reproduction in the biological activities of worm snakes. In this article the anatomical and histological layout of this sensory organ in a worm snake *Typhlina bramina* has been reported. For this study, 5-7 micrometric histological sections were cut from their heads where the Jacobson's organ remain located in the mid dorsal plain of the skull having the vomar and nasal plate flooring. The micrometric measurement of the organ lodged in the vomeronasal cranial capsule also laterally flanked by baso-medial side of nasal cavity. In this species of the worm snake (*Typhlina bramina*) the Jacobson's organ has been found to possess an average linear length of 550 μ m in male and 510 μ m in female which is slightly smaller than other reported species. It has also a prominent connection to the nasal cavity along with a communicating tunnel with the oral cavity through 2 small apertures in the anterior palate. On close observation of the micro sections, the lachrymal duct descending from the eye (internal pocket) is also found to join the organ. Surprisingly the organs lumen since to be also lying by 2 different cellular layer likely to be sensory in function. Cellular differentiation in the sensory epithelial layer which is relatively very thick has also been noticed and discussed in the paper. The central mushroom body covered by special epithelial layer has ciliated cells. Histochemically the epithelial cells have been found to be PAS positive due to heavy deposition of glycogen.

Key words: Typhlina bramina, Sensory function, Jacobson's organ/ VNO, sensory epithelium, PAS positive.

INTRODUCTION

In comparison to general vertebrates and in contrast to the mammals, great variation is found in the organization of sense organ particularly that of snakes with reference to the blind worm snake, *Typhlina bramina*. Snake mostly use the power of smell and touch for detection of any object present in their vicinity as because they do not posses movable eye lids and rudimentary eyes remain sunken and

*Corresponding author : Phone : 08789865993 E-mail : ranjanamdp@gmail.com fixed which arrest their movement. Moreover they also lack external parts of ears, middle ear and simpanic membrane excepting the ear ossicles and as a result they can only perceive the sound wave vibrations.

To supplement these lapses in the optic and auditory organ, the nature has equipped snakes with well developed organ of smell and touch in the form of accessory sensory system (ACS) known as vemero nasal organ.¹⁻⁴

The lineage of VNOs in tetrapods and many groups of mammals including rodents has very prominent evolutionary organization but in squamates the VN

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chemosensory system is anatomically and functionally quite distinct from the main olfactory system. Several researches have been done on the VNO of snakes⁵⁻⁷ but very little has been done for the worm snakes.

MATERIALS & METHODS

6 adults (3 males & 3 females) were kept in laboratory experimental jar after their collection from local habitats of Madhepura and Saharsa. The specimens were killed with 5% chloroform and their heads were decapitated sex wise and kept separately in the smaller jars, washed with normal saline and fixed in 4% formalin for at least one week. The preserved and fixed decapitated head were later decalcified using formic acid sodium citrate buffer solution for 3-4 days as per the protocol of Smith *et al.* (1997)⁸. The tissues were dehydrated in graded series of ethanol, cleared in xyline and paraffin wax blocks were made from the decapitated head tissues. Microtomic sections were procured in transverse plain at 5-7 μm thickness.

The sections were mounted on glass slides and stained with Haematoxylin-Eosin or Alcian blue-Periodic Acid Schif (PAS). Then stained sections were scrutinized in details by the aid of a light microscope. Ocular graded graticule was used to measure specific histological parameters in the organs including the lumen's width and distance between the two capsules enclosed the organs.

RESULTS

As per the experimental work plan, the microtomic section of VNO complex have been procured from both male and female subject in order to record the sex specific variations in its structural organization. In the Typhlops bramina, the VNO consists of a paired bilateral structure located at the baso-medial side of the nasal cavity, with direct relationship with the vomer bone and palatine process of maxillary bone. In general, each organ appeared as a hollow spindle with an average length of 550μ m male and $510\ \mu$ m in female. Each organ begins at rostral one-third of the nasal cavity. Anteriorly, the organ is narrow but gradually widens. This increase in diameter continues to two-third of the organ while in its caudal one-third it undergoes a reverse process so that it again becomes tapered. Along with this, a marked variation in the dimension as obtained from the micrometric measurement has been found in both male and female individuals. The sex specific measurement data showing the micro level variation has been furnished in tables 1 & 2 and displayed in graphs A & B

In transversal sections the organ is observed as a dome-shaped structure which is enclosed by an osseous capsule composed of medial palatine and lateral vomer bones. The capsule is incomplete as in some regions the bone is replaced by connective or cartilage tissues. The sites and locations of these replacing tissues varies along the organ' length but mostly is seen in dorsolateral and ventromedial regions. The lumen of organ has become crescent-shaped as the result of Mushroom Body (MB) which is a bulge-like evagination protrudes from ventral or ventrolateral side into the lumen.

Along the length of the organ the lumen is lined with three types of epithelia. The dorsal or concave side of the VNO lumen is lined with a sensory epithelium whose main role is to detect chemical and elicit impulses to be conducted into the central nervous system. This is a pseudo stratified epithelium in which three types of cells can be recognized. Just beside the lumen there is a layer of supportive cells which have elongated nuclei. At the top of supportive cells a several layers of sensory cells (on average 10 layers) with rounded nuclei are observed. The receptor cells are indeed bipolar neurons whose dendrites terminate as microvilli at the luminal surface.

Table 1- Micrometric measurements of the nasal cavity and VNO complex dimensions in µm with VLD/ NCD ratio in male *T.bramina*

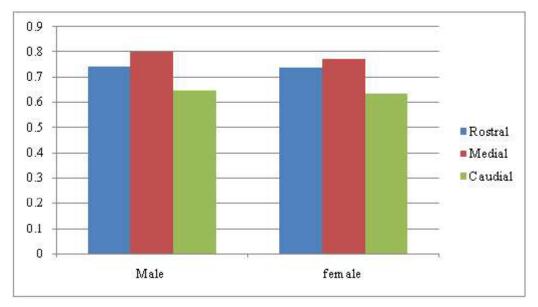
Intercapsular space of nasal cavity	VNO dimensions	VNO lumen Diameter (VLD)	Nasal Cavity Diameter (NCD)	VLD/NCD ratio
18	Rostral	162	218.2	0.742
22	Medial	215	269.0	0.799
20	Caudial	187	289.0	0.647

Ranjana- Sex specific anatomical variation in the organization of Jacobson's organ (VNO) with respect to its olfactory function in common worm snake *Typhlina bramina*

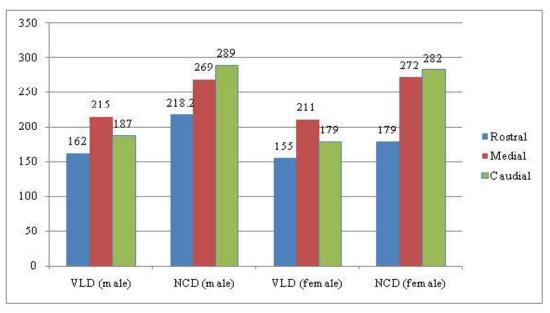
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Intercapsular space of nasal cavity	VNO dimensions	VNO lumen Diameter (VLD)	Nasal Cavity Diameter (NCD)	VLD/NCD ratio		
18	Rostral	155	210.0	0.738		
22	Medial	211	272.0	0.775		
20	Caudial	179	282.0	0.634		

Table 2- Micrometric measurements of the nasal cavity and VNO complex dimensions in µm with VLD/NCD ratio in female *T.bramina*

Graph A.- Sex specific variation in the three-dimentional VLD & NCD measurements of Jacobso's organ complex (VNO) in *T.bramina*



Graph B.- Sex specific variation in the three-dimentional VLD/NCD ratio of Jacobso's organ complex (VNO) in *T.bramina*



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