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Topological Organization and Functional Aspects of the Olfactory Epithelium of Whipfin Silver Biddy *Gerres filamentosus* (Cuvier 1829)

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Abstract: The structural organization of different cells lining the olfactory epithelium of *Gerres filamentosus* (Cuvier 1829) were studied by light and scanning electron microscopy respectively to correlate their role in olfaction. The fan shaped olfactory rosette of *G. filamentosus* was more or less oval in outline and composed of 13 lamellae of different sizes in both sides. The olfactory epithelium was partitioned into sensory and non-sensory regions. The sensory epithelium was restricted on the flat apical end of the lamellae and embossed with two types of receptor cells bearing either cilia or microvilli. The non-sensory epithelium, covering the middle and basal region of lamellae was comprised of stratified epithelial cells and mucous cells. The orientation of various cells in the surface contour of olfactory epithelium was discussed in light of their functional significance.

Key words: Cytoarchitecture, surface ultrastructure, function, olfactory epithelium, *Gerres filamentosus*

INTRODUCTION

Olfactory organ in fish is of great biological importance because it is essentially a chemoreceptor and plays an indispensable role to detect water soluble compounds in the aquatic ecosystem. Chemoreception is involved in the searching of food, recognition of sex, discrimination between individuals of the same or different species, in defenses against predators, in parental behaviour, in orientation and in many other activities (Uva, 1985). Olfaction results from stimulation of the sensory receptor cells in the olfactory organ, which is innervated by the olfactory nerve (Ichikawa and Ueda, 1977). The morphohistology and the fine structural organization of different cells lining the olfactory epithelium in various fishes have been described by several workers (Ojha and Kapoor, 1973; Bandyopadhyay and Datta, 1996; Ruzhinskaya *et al.*, 2001; Ma and Wang, 2010; Chakrabarti and Ghosh, 2010a, 2011; Ghosh and Chakrabarti, 2012). These studies have revealed the considerable morphological variability regarding the shape and location of the olfactory organ, number and arrangement of the olfactory lamellae, the distribution of sensory and non-sensory epithelium as well as an abundance of various receptor cells, which correlate with the enormous diversity of life-styles among fish species. The ecological niche inhabited by a given species

probably has a great impact on the structure of olfactory lining and level of specialization (Hara, 1994; Kuciel *et al.*, 2011).

However, lacunae still remain in the structural and functional entities of the different sensory and non-sensory cells pertaining to the olfactory epithelium of brackish water teleosts. An attempt has therefore, been made in the present study to portray the distribution and characterization of the olfactory epithelial components and the functional significance of various cells in the olfactory epithelium of *Gerres filamentosus* (Cuvier 1829) with its mode of life and living. *G. filamentosus* (Perciformes: Gerreidae) is an omnivorous bottom feeder (Golikatte and Bhat, 2011) which subsists on small crustaceans, tunicates, polychaetes, filamentous algae, diatoms, molluscan shell, fish scales and miscellaneous items (Aziz *et al.*, 2012). In this study structural organization of different cells lining the olfactory epithelium of *Gerres filamentosus* (Cuvier 1829) were studied by light and scanning electron microscopy respectively to correlate their role in olfaction.

MATERIALS AND METHODS

Twelve healthy adult fishes of *G. filamentosus* (8-10 cm in length) were procured from the Junput brackish water fish farm in Purba Midnapore, West

Bengal, India. Fish were anaesthetized with tricaine methone-sulphonate (MS 222; Sigma Chemical Co.) solution (100 mg L^{-1}) and sacrificed following the guidelines given by the Institutional Ethical Committee. Olfactory rosettes were dissected out from the dorsal side of the olfactory chamber under a stereo microscope and immediately processed for the histological and Scanning Electron Microscopic (SEM) preparation.

For the purpose of SEM study, the olfactory rosettes were perfused *in vivo* with 2.5% glutaraldehyde solution in 0.1 M phosphate buffer (pH 7.4) for 10 min. The entire olfactory rosettes were carefully dissected out and the adhering mucus on the epithelial surface was removed by repeated rinsing with 1% Tween 40 solution. After being rinsed in 0.1 M phosphate buffer (pH 7.4), the tissues were infiltrated with 2.5% glutaraldehyde buffered with 0.1 M phosphate buffer (pH 7.4) for 24 h at 4°C . After proper fixation, the tissues were rinsed in the same buffer for 10 min and post fixed in 1% osmium tetroxide (OsO_4) in 0.1 M phosphate buffer (pH 7.4) for 2 h. After secondary fixation the tissues were washed thoroughly in buffer, dehydrated through ascending series of acetone, followed by isoamyl acetate and then subjected to critical point drying method with liquid carbon-dioxide. After being dried the olfactory rosettes were mounted on metal stubs, coated with gold palladium with a thickness of approximately 20 nm and scanned in a Hitachi, S-530 SEM.

For histological study, olfactory tissues were fixed in aqueous Bouin's fluid for 16-18 h and were dehydrated properly through ascending series of ethanol, cleared with xylene and embedded in paraffin wax of $56\text{-}58^{\circ}\text{C}$. Tissues were sectioned serially at $4 \mu\text{m}$ thickness. After routine histological procedure the sections were stained with Delafield's Haematoxylin-Eosin and Mallory's triple stain.

RESULTS

Under SEM observations, the olfactory rosette of *G. filamentosus* beholds as fan like, more or less oval in outline and consists of 13 lamellae in each left and right sides. The outer margins of the lamellae are free, while the inner margins are affixed to the broad median raphe (Fig. 1). Lamellae are closely set and originated from raphe with a convex ventral and concave dorsal surface. The length of lamellae varies according to their position. The apical parts of the lamellae are flat and almost tongue shaped but the middle and bases are slender (Fig. 1, 2). Sensory and non-sensory are distinguished on each lamella. The sensory epithelium is restricted on the flat apical end whereas the non-sensory epithelium organizes mainly in the lateral surface of middle and basal portion of the lamellae (Fig. 2). Histologically, the olfactory lamellae

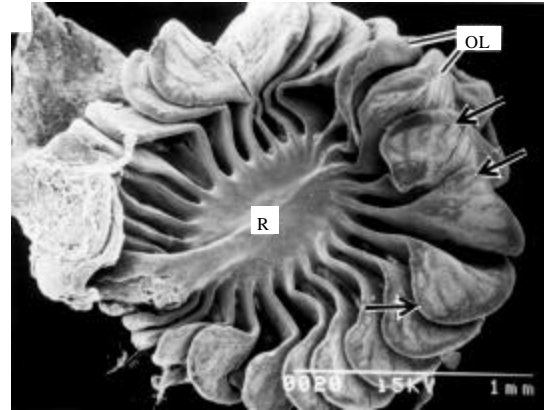


Fig. 1: Fan like oval shaped olfactory rosette showing Olfactory Lamellae (OL) attached with broad median Raphe (R). Arrows indicate tongue shaped apical part of the OL (SEM)50X

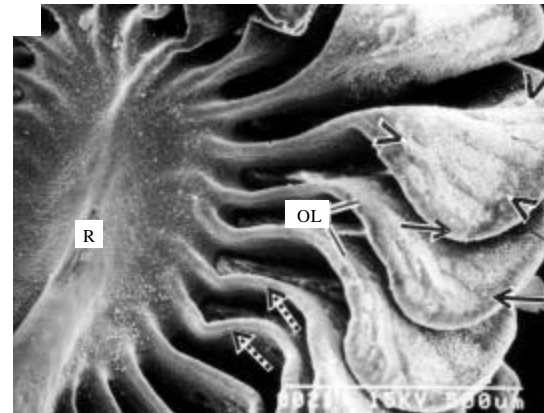


Fig. 2: Higher magnification of (OL) Olfactory Lamellae based on (R) Raphe showing sensory areas (arrow heads) on the flat and tongue shaped apical ends (solid arrows). Note slender part of OL having non-sensory area (broken arrows) (SEM)100X

are comprised of two layers of epithelium enclosing a thin stromal layer, the central core which constitutes loose connective tissues, nerve fibers and blood vessels (Fig. 3). The surface contour of sensory epithelium is chiefly lined with receptor cells in one side of the lamellae and non-sensory epithelium on other side of lamellae arranged with mucous cells (Fig. 3). The dendron of each receptor cell expands as a narrow cylindrical process (Fig. 4, 5) and provided with knob like vesicle on the free epithelial surface (Fig. 5). In some areas of the sensory epithelium few microvillous receptor

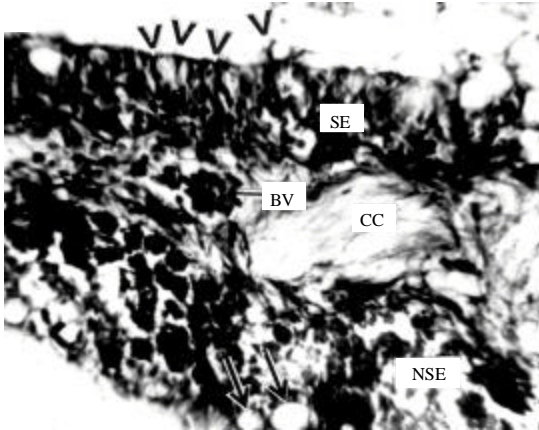


Fig. 3: Section of Olfactory Epithelium (OEP) showing Sensory Epithelium (SE) lined with Receptor Cells (RC) (arrow heads). Non-sensory Epithelium (NSE) on another side provided with Mucous Cells (MC) (solid arrows). Central Core (CC) provided with Blood Vessels (BV) (MT) 400X

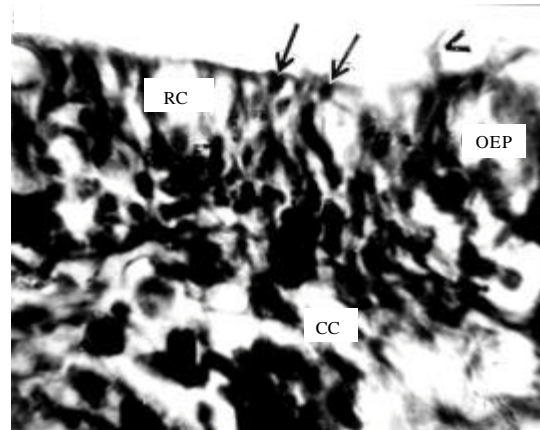


Fig. 5: Magnified view of sensory OEP showing dendrite process of cylindrical RC and MV (solid arrows). Arrow head indicates RC with knob like vesicle. CC marks central core (MT) 1000X

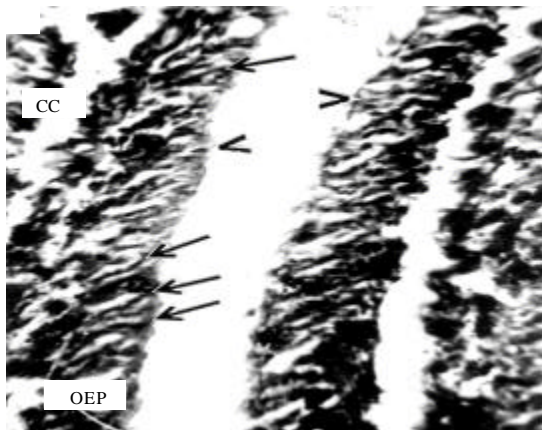


Fig. 4: Section of sensory OEP provided with a series of RC (solid arrows) and scattered microvillous cells (MV) (arrow heads). CC indicates central core (HE) 400X

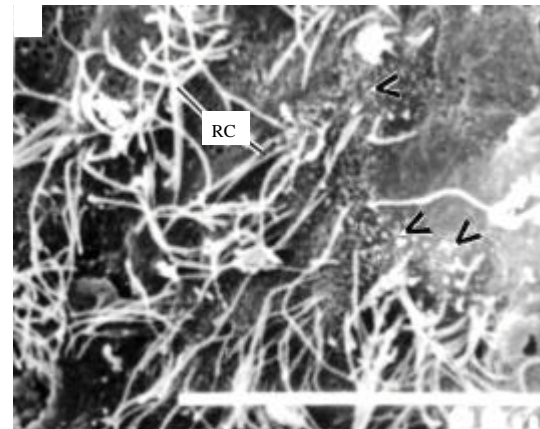


Fig. 6: Sensory epithelium exhibiting microvillous cells (arrow heads) and patches of ciliated RC (SEM) 4500X

cells are present in between cluster of ciliated receptor cells. The microvillous receptor cells have a slightly sunken apex and consist of minute dendrites (Fig. 5). Under SEM studies the sensory epithelium is also embossed with numerous microvillous cells which are discernible at the base of the ciliated receptor cells and give a sculptured manifestation to the surface architecture (Fig. 6). The transitional

zone of sensory and non-sensory epithelium is made up of patchy distribution of ciliated receptor cells, among which oval or polygonal stratified epithelial cells are located (Fig. 7).

Histologically, the surface zone of non-sensory epithelium is mainly comprised of stratified epithelial cells having prominent nuclei and mucous cells (Fig. 8). According to SEM examination the surface of the non-sensory epithelium is provided with few scattered receptor cells in between the stratified epithelial cells which are afforded with concentric microridges alternating

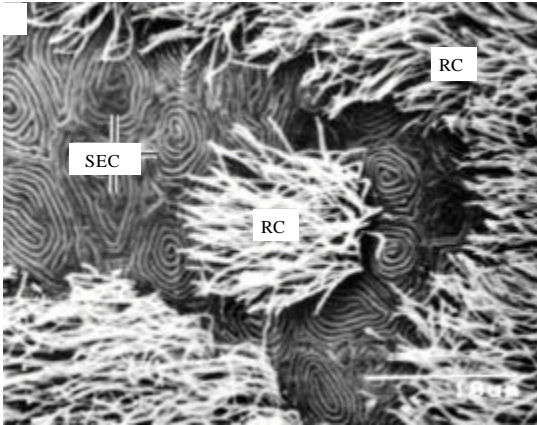


Fig. 7: Transitional zone of sensory and non-sensory olfactory epithelium showing dendrite patches of RC in between prominent stratified epithelial cells (SEM) 3500X

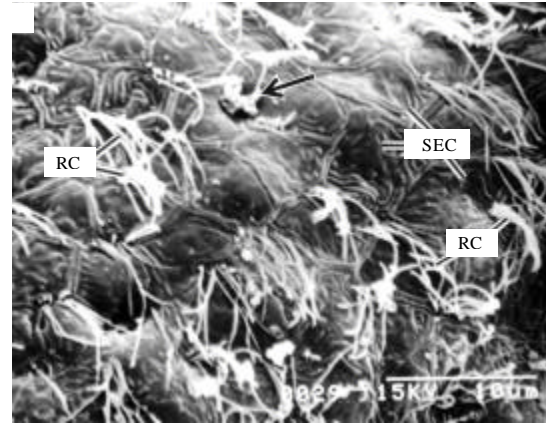


Fig. 9: Showing dendrite process of scattered RC in between SEC. Note the presence of MC (solid arrows) in between SEC (SEM) 3000X

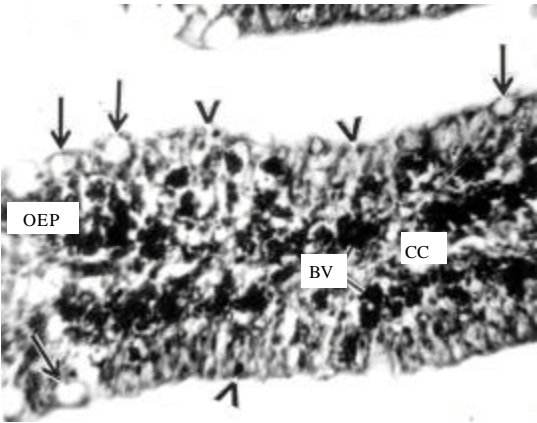


Fig. 8: Section of non-sensory OEP comprised of Stratified Epithelial Cells (SEC) (arrow heads) and mucous cells (solid arrows). CC points out central core with BV (HE) 400X

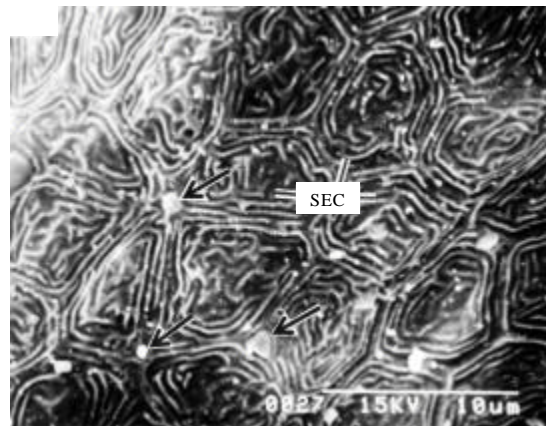


Fig. 10: Surface feature of raphe represented by compactly arranged SEC provided with labyrinth pattern microridges. Note the presence of mucous cells with mucin mass (solid arrows) in between SEC (SEM) 4000X

with furrow (Fig. 9). The raphe is delineated by compactly arranged stratified epithelial cells intercalated with the opening of mucous cells (Fig. 10). The apical surface of the stratified epithelial cells is characterized with labyrinth pattern microridges leaving shallow channels in between. Secreted mucin droplets are laid in the opening of mucous cells (Fig. 10).

DISCUSSION

The olfactory organ is lined with olfactory epithelia which is raised from the floor of the olfactory chamber and

often folded to form olfactory lamellae (Hara, 1975). The olfactory epithelium shows considerable diversity, reflecting the degree of development and ecological habitats (Zeiske *et al.*, 1992). The present study reveals that the oval shaped olfactory rosette of *G. filamentosus* consists of 13 olfactory lamellae arranged on both side of the median raphe and can be classified under Bateson, 1890 rosette type-3 or Burne, 1909 rosette column I. According to Teichmann (1954), the oval type of olfactory organ falls under the category of eye-nose fish, which means that this category of fish possesses similarly developed optic and olfactory sensitivity. In

terms of the arrangement of the lamellae against the raphe, the olfactory rosette of *G. filamentosus* have been recognized as Type G, fan-shaped (Yamamoto, 1982). The olfactory epithelium of *G. filamentosus* comprises a complex system having sensory and non-sensory regions. The sensory receptor epithelium is restricted in the flat apical tongue like portions of the lamellae while the middle and basal part of the lamellae is provided with non-sensory epithelium. This arrangement may be due to the fact that the tongue shaped area of sensory epithelium faces the flow of incoming water current and the receptor cells mobilizing different olfactory cues. Similar tongue like projections of the olfactory lamellae has been observed by Chakrabarti and Ghosh (2010b) in the olfactory epithelium of *Catla catla*.

In the present study, in *G. filamentosus* the sensory epithelium mainly consists of two morphologically distinct types of receptor cells: ciliated and microvillous cells. They occur together but in different proportions Zeiske *et al.* (2003) also observed that the ciliated and microvillous receptor cells also occur together in the olfactory organ of genus *Acipenser* but in different proportions in different species. The present study reveals that the ciliated receptor cells dominate over the microvillous receptor cells. The ciliated receptor cells are of special interest because they form a part of the olfactory transduction mechanism, are stimulated by odour bearing substances and also enable the fish to detect food. In the present observation, the ciliated receptor cells correspond to the type I cells of Yamamoto and Ueda (1978). In contrast to the ciliated receptor cells, the microvillous receptor cells have a slightly sunken apex and consist of minute dendrites. This also conforms to the findings of Camacho *et al.* (2010) in the olfactory epithelium of sturgeon. The microvillous receptor cells might form a different olfactory transduction mechanism for pheromones or amino acids. Bhute and Baile (2007) also advocated that the microvillous receptor neurons perceive and process signals of pheromone, which is an important step of breeding in *Labeo rohita*. On the other hand Bakhtin (1977) and Bannister (1965) reported that microvillous cells in the olfactory surface of *Squalus acanthias* and teleostean fishes are predecessors of ciliated receptor cells.

In the transitional zone of sensory and non-sensory epithelium aggregation of ciliated receptor cells are responsible for better monitoring of the water quality even up to this zone. Furthermore, the non-sensory epithelium consists of stratified epithelial cells provided with concentric microridges on their apical surface that help in holding mucus film over the epithelium and in protecting the sensory receptor cells

from mechanical injury or from different hazardous substances. The mucous cells are distributed in between the stratified epithelial cells of the non-sensory epithelium. The mucus covering the olfactory lamellae constitutes an important medium in which odorants are diffused. On the other hand the mucin probably helps in binding microscopic debris and keeps the sensory cells ready for new stimuli. This is in conformity with the findings of Rahmani and Khan (1980) in the olfactory mechanism of *Anabas testudineus*.

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REFERENCES

- Aziz M., V. Ambily and S. Bijoy Nandan, 2012. Food and feeding habits of *Gerres filamentosus*. *Fishery Technol.*, 49: 10-13.
- Bakhtin, E.K., 1977. Peculiarities of the fine structure of the olfactory organ of *Squalus acanthias*. *Tsitologiya*, 19: 725-731.
- Bandyopadhyay, S.K. and N.C. Datta, 1996. Morphoanatomy and histology of the olfactory organ of an air-breathing catfish *Heteropneustes fossilis* (Bloch). *J. Anim. Morphol. Physiol.*, 43: 85-96.
- Bannister, L.H., 1965. The fine structure of the olfactory surface of teleostean fishes. *Q. J. Microsc. Sci.*, 106: 333-342.
- Bateson, W., 1890. The sense organs and perceptions of fishes, with remarks on the supply of bait. *J. Mar. Biol. Assoc. UK.*, 1: 225-256.
- Bhute, Y.V. and V.V. Baile, 2007. Organization of the olfactory system of the Indian major carp *Labeo rohita* (Ham): A scanning and transmission electron microscopic study. *J. Evol. Biochem. Physiol.*, 43: 342-349.
- Burne, R.H., 1909. The anatomy of the olfactory organ of teleostean fishes. *Proc. Zool. Soc. London*, 2: 610-663.
- Camacho, S., M.V. Ostos-Garrido, A. Domezain and R. Carmona, 2010. Study of the olfactory epithelium in the developing sturgeon. Characterization of the crypt cells. *Chem. Senses*, 35: 147-156.

- Chakrabarti, P. and S.K. Ghosh, 2010a. Histoarchitecture and scanning electron microscopic studies of the olfactory epithelium in the exotic fish *Puntius javanicus* (Bleeker). Arch. Polish Fish., 18: 173-177.
- Chakrabarti, P. and S.K. Ghosh, 2010b. Histological and scanning electron microscopical study of the olfactory epithelium of the Indian major carp, *Catla catla* (Hamilton). Folia Morphol., 69: 24-29.
- Chakrabarti, P. and S.K. Ghosh, 2011. The structural organization and functional aspects of the olfactory epithelium of tigerperch, *Terapon jarbua* (Forsskal, 1775) (Perciformes: Terapontidae). Turk. J. Zool., 35: 793-799.
- Ghosh, S.K. and P. Chakrabarti, 2012. Histological organization and microarchitecture of various cells lining the olfactory epithelium of *Rita rita* (Hamilton, 1822) (Siluriformes: Bagridae). Biological Lett., 49: 35-42.
- Golikatte, R.G. and U.G. Bhat, 2011. Food and feeding habits of the whipfin silver biddy *Gerres filamentosus* from Sharavati estuary, central west coast of India. World J. Sci. Technol., 1: 29-33.
- Hara, T.J., 1975. Olfaction in fish. Prog. Neurobiol., 5: 271-335.
- Hara, T.J., 1994. The diversity of chemical stimulation in fish olfaction and gestation. Rev. Fish. Biol. Fish., 4: 1-35.
- Ichikawa, M. and K. Ueda, 1977. Fine structure of the olfactory epithelium in the goldfish, *Carassius auratus* a study of retrograde degeneration. Cell Tiss. Res., 183: 445-455.
- Kuciel, M., K. Zuwała and M. Jakubowski, 2011. A new type of fish olfactory organ structure in *Periophthalmus barbarous* (Oxudercinae). Acta Zool., 92: 276-280.
- Ma, A. and X. Wang, 2010. Functional morphology of the olfactory organ of the tongue sole, *Cynoglossus semilaevis*. Chinese J. Oceanol. Limnol., 28: 209-217.
- Ojha, P.P. and A.S. Kapoor, 1973. Structure and function of the olfactory apparatus in the freshwater carp, *Labeo rohita* (Ham. Buch.). J. Morphol., 140: 77-85.
- Rahmani, A.R. and S.M. Khan, 1980. Histology of the olfactory epithelium and the accessory nasal sacs of an anabantoid fish, *Anabas testudineus* (Bloch). Arch. Biol., 91: 397-411.
- Ruzhinskaya, N.N., P.A. Gdovskii and G.V. Devitsina, 2001. Chloride cell, A constituent of the fish olfactory epithelium. J. Evol. Biochem. Physiol., 37: 89-94.
- Teichmann, H., 1954. Vergleichende untersuchungen an der nase der fische. Zoomorphology, 43: 171-212.
- Uva, B., 1985. Olfaction, related structures and functional adaptations. Boll. Zool., 52: 331-337.
- Yamamoto, M. and K. Ueda, 1978. Comparative morphology of fish olfactory epithelium-III. Cypriniformes. Bull. Japan Soc. Sci. Fish., 44: 1201-1206.
- Yamamoto, M., 1982. Comparative Morphology of the Peripheral Olfactory Organ in Teleosts. In: Chemoreception in Fishes, Hara, T.J. (Ed.). Elsevier, Amsterdam, pp: 39-59.
- Zeiske, E., B. Theisen and H. Breucker, 1992. Structure, Development and Evolutionary Aspects of the Peripheral Olfactory System. In: Fish Chemoreception, Hara, T.J. (Ed.). Chapman and Hall, London, pp: 13-39.
- Zeiske, E., A. Kasumyan, P. Bartsch and A. Hansen, 2003. Early development of the olfactory organ in sturgeons of the genus *Acipenser*: A comparative and electron microscopic study. Anat. Embryol., 206: 357-372.