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Saroj Kumar Ghosh
Fisheries Laboratory, Department of Zoology, The University of Burdwan, Golapbag, Burdwan-713 104, West Bengal, India

Padmanabha Chakrabarti
Fisheries Laboratory, Department of Zoology, The University of Burdwan, Golapbag, Burdwan-713 104, West Bengal, India

Correspondence:
Padmanabha Chakrabarti
Department of Zoology,
The University of Burdwan, Golapbag, Burdwan-713 104, West Bengal, India
Email: dr.pchakrabarti@yahoo.in
Tel: +91-342-2634798

Histophysiological Studies on the Olfactory Epithelium of Banded Pearl Spot *Etroplus suratensis* (Bloch, 1790)

Saroj Kumar Ghosh, Padmanabha Chakrabarti

ABSTRACT

The histoarchitecture and function of various cells lining the olfactory epithelium of *Etroplus Suratensis* (Bloch, 1790) was investigated with light microscope. Histologically, the olfactory lamella consisted of two layers of epithelium separated by central core. On the basis of cellular organization and their distribution in the surface of each lamella is characterized by sensory and non-sensory regions. Four distinct type of receptor cells (primary receptor cells, secondary receptor cells, microvillous cells and rod cells) as well as mast cells and basal cells were identified in the sensory epithelium. The non-sensory epithelium was provided with mainly stratified epithelial cells and large number of mucous cells. The different cells on the olfactory epithelium were correlated in relation to their role in olfaction.

Keywords: Cellular architecture, Function, Olfactory organ, *Etroplus suratensis*.

1. Introduction

The study of the olfactory organ in fish is of great significance because it is essentially a chemoreceptor and plays a vital role not only locating food but also in detecting the presence of odoriferous substances in the aquatic ecosystem. Fish ascertain chemical stimuli in consequence of two major channels of chemoreception, olfaction and gestation [1]. Olfaction is accomplished principally by stimulation of the receptor cells on the neuroepithelial surface of the olfactory organ, which is innervated by the olfactory nerve [2]. Olfactory signals and cues serve a crucial role for several life functions in fish such as migration, communication, feeding, schooling, defense and reproduction [3]. Comprehensive research has been performed on the cellular organization of the olfactory epithelium of different teleosts through electron microscope [4, 5, 6, 7, 8, 9, 10, 11] but there is little information by the use of light microscopy [12, 13, 14, 15, 16]. Apparently it seems that no worth mentioning work has been done on correlation and cellular variation of the olfactory epithelium and their role in sensory reception in teleosts.

The aim of the present investigation was to examine the structural characterization and functional aspects of different cell types on the olfactory epithelium of brackish water pearl spot *Etroplus suratensis* (Cichlidae, Perciformes) by histological analysis.

2. Materials and Methods

Adult specimens of *E. suratensis* (13 to 15 cm in total length) were procured from Junput brackish water fish farm, Purba Midnapore, West Bengal. Fishes were sacrificed within a few second of capture by decapitation. The olfactory rosettes were then carefully dissected out from the olfactory chamber on the dorsal side of the head under a stereoscopic binocular microscope and were fixed in aqueous Bouin's fluid for 16-18 h. After fixation the tissues were washed repeatedly in 70% ethanol and dehydrated properly through ascending series of ethanol. Then the tissues were cleaned with xylene and embedded in paraffin wax of 56-58˚C under a thermostat vacuum paraffin-embedding bath for a period of 1 h and 30 min. Sections were cut at 4 µm thick using a rotary microtome (Weswox). After routine histological procedure deparaffinized sections were stained with Delafield's Haematoxylin-Eosin and Mallory's [17] triple stain. Staining slides were examined and photographed under Olympus-Tokyo PM-6 compound microscope.

3. Results
Olfactory epithelium of *E. suratensis* is a continuous thick sheet of stratified epithelial cells, which is folded to form lamellae. Histologically, the olfactory lamellae are based on raphe and composed of two layers of olfactory epithelium separated by wide central lamellar space, the central core (Fig. 1). Secondary foldings in the olfactory lamella are found also. The epithelium is dissevered into sensory and non-sensory regions which are made up of the following cell types:

3.1. Primary neurons or receptor cells
Receptor cells are the sensory elements of the olfactory epithelium, mainly present beneath or adjacent to the stratified epithelial cells. They are provided with a cell body and a long dendrite. The cell body is characterized by rounded or oval structure and deeply stained nuclei (Figs. 2, 5 and 6). The dendrite of the receptor cells are stretched as a slender process up to the surface of the epithelium. Synaptic contact is discernible in between primary and secondary neurons (Fig. 5).

3.2. Secondary neurons or spindle-shaped cells
The secondary neurons lie either beneath or side of the primary neurons (Figs. 2 and 3). The secondary neurons with synapse are differentiated by their elongated-oval nuclei. These receptor cells are highly basophilic with darkly stained nuclei in the distal part of the cell (Figs. 5 and 6). The axons of the secondary neurons extend up to the deeper region of the olfactory epithelium and pass into the central core of the lamella.

3.3. Microvillous cells
Microvillous cells are situated in more superficial layer of the epithelium, provided with light stained inconsiderable nuclei. They are small in size, rounded and possess minute dendrites on the apical rim of the cell (Fig. 5).

3.4. Rod cells
Rod cell containing an elongated nucleus is situated on the side of the receptor cells in the epithelium. They are ovoid in appearance, protrude as a spike like structure and characterized with highly basophilic nuclei (Figs. 2 and 6).

3.5. Mast cells
These cells are small in extent, more rounded with relatively gentle amount of cytoplasm and have a polymorphous nucleus (Figs. 2 and 6).

3.6. Basal cells
These cells are distributed in the deeper part of the olfactory epithelium, adjacent to the central core. They are cuboidal, oval and rounded in outline with distinct nuclei and chromophobic cytoplasm (Figs. 3, 4 and 6). Basal cells verily conceive the reservoir for the creation of supporting and receptor cells as they migrate about the upper part of the olfactory epithelium.

3.7. Stratified epithelial cells or Supporting or sustentacular cells
These cells are elliptical to columnar in shape with prominent central basophilic nucleus situated superficially in the epithelium (Fig. 4). Cytoplasm is less granular and faintly stained. Supporting cells give the basic structure of the olfactory lamella.

3.8. Mucous cells
Mucous cells are oval glandular cells, dispensed in considerable abundance at the free margin of the olfactory epithelium mainly in the non-sensory region (Figs. 1 and 4). The mature mucous cell exhibits a globular shape (Fig. 4) due to homogenous mucin and locates in the superficial layer.

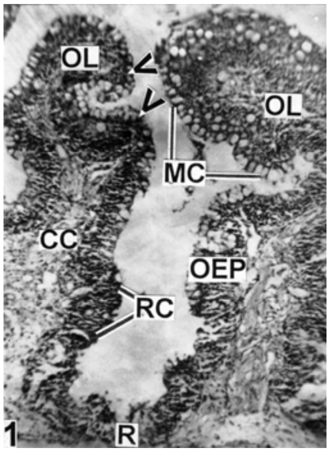

Fig 1: Olfactory lamellae (OL) attached with raphe (R) showing compactly arranged receptor cells (RC) in the olfactory epithelium (OEP) which are separated by broad central core (CC). Arrow heads indicate secondary folding of lamellae. Note the presence of mucous cells (MC) at the apical border of OL (HE) × 100 X.

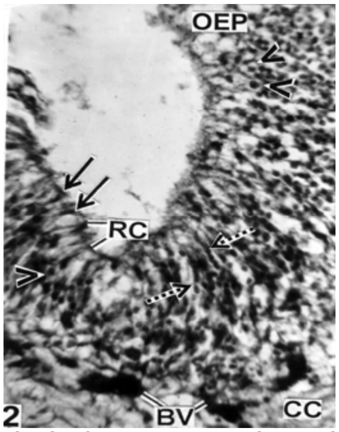

Fig 2: Sensory OEP lined with primary RC, secondary RC (broken arrows), rod cells (RD) (solid arrows) and mast cells (M) (arrow heads). Note blood vessels (BV) present in CC (MT) × 400 X.

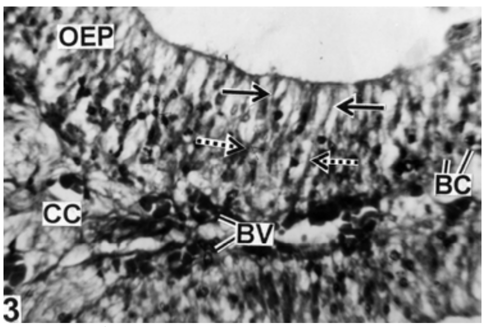

Fig 3: OEP comprised of primary RC (solid arrows), secondary RC (broken arrows) and basal cells (BC). Note CC provided with BV (MT) × 400 X.

Central core of the lamella

The olfactory lamellae of *E. suratensis* are composed of two layers of olfactory epithelium enclosing a broad central space called central core. It consists of loosely arranged connective tissue stroma. Nerve fibres and blood vessels cross through the central core (Figs. 2, 3, 4 and 6).

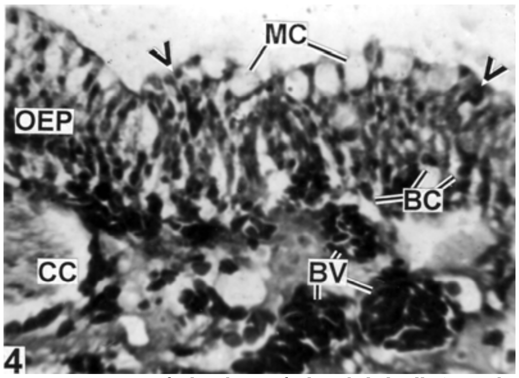

Fig 4: Non-sensory OEP typified with stratified epithelial cells (arrow heads), MC and BC. Note compact BV in CC (HE) × 600 X.

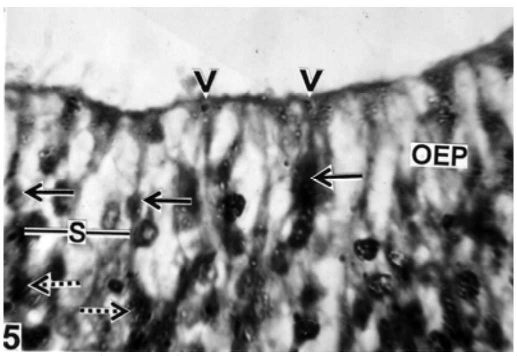

Fig 5: Higher magnification of sensory OEP showing the arrangement of primary RC (solid arrows), secondary RC (broken arrows) and their synaptic contact (S). Note the presence of microvillous cells (arrow heads) on the surface epithelium (MT) × 1000 X.

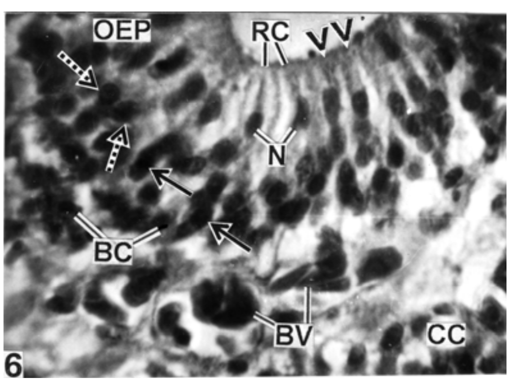

Fig 6: OEP exhibiting primary RC with dendrite processes and conspicuous nuclei (N), secondary RC (solid arrows), RD (arrow heads), M (broken arrows) and BC. Note the presence of BV in CC (HE) × 1000 X.

4. Discussion

The olfactory organ of fish is placed at the ethmoid region of the skull and composed of many folds to form lamellae [18]. It has an acute sense of smell in various aspects of life history and exhibits considerable diversity which reflects the degree of development and ecological habitats [19]. The ecological niche inhabited by a given species has an immense impact on structural characterization of the olfactory epithelium [20]. The folding on the lamellae of the olfactory epithelium increases the surface area of the epithelium as well as the sensitivities and efficacy of the olfactory organ [21]. The distribution of the sensory and non-sensory epithelia on the surface of the lamellae shows a great variety in different fish species for adaptation to a specific environment [22].

The olfactory epithelium of *E. suratensis* consists of receptor, mast, basal, stratified epithelial and mucous cells. The receptor cells are blended with stratified epithelial cells in the surface epithelium. The dendrite of receptor cells runs to the surface of the lamella as a slender process are of unique attention as they form a part of the olfactory transduction mechanism and are stimulated by odour-bearing substances. It is constituted that the receptor cells bestow in the olfactory epithelium are accomplished to ascertain chemical changes in the surrounding environment [23]. The most interesting characteristic of the present investigation is the histological identification of secondary neurons in addition to primary neurons and the presence of synaptic connections between these two types of neurons in the olfactory epithelium. The synaptic connection between the primary and secondary receptor cells may extend from the epithelial surface to the central core. Ojha and kapoor [12] also reported secondary neurons in the olfactory epithelium of *Labeo rohita*. Graziadei and Metcalf [24] assumed that new neurons rehabilitate the old and degenerating ones and establish the synaptic contact in the olfactory bulb.

In the present investigation in contrast to the primary and secondary neurons, the microvillous receptor cells with slightly sunken apex consist of minute dendrites. This also conforms to the observation of Camacho *et al*. [25] in the olfactory epithelium of sturgeon. The microvillous receptor cells might form a different olfactory transduction mechanism for pheromones or amino acids. Zippel *et al.* [26] suggested microvillous cells in the olfactory epithelium of gold fish mediate responses to pheromones. Bhute and Baile [6] also advocated that the microvillous cells perceive and process signals of pheromone, which is a significant proceeding of reproduction in *Labeo rohita*. On contrary, Bakhtin [27] and Bannister [28] reported that microvillous cells in the olfactory epithelial surface of *Squalus acanthias* and teleostean fishes are forerunners of ciliated receptor cells.

The existence of rod cells in *E. suratensis* is a peculiar feature. The olfactory epithelial cells with rod shaped processes have also been described as usual receptors by Yamamoto [22]. The sensitivity of the rod receptor cells may change in the euryhaline *E. suratensis* when they migrate into sea water from brackish water or vice versa. Hernadi [29] proposed that the occurrence of the rod-shaped olfactory neuron has been observed in the presence of a new physiological condition.

Mast cells in the olfactory mucosa are believed to play an important role in reproduction of Rhesus monkey [30], Baltic trout [31] and Indian major carp, *Labeo rohita* [7]. Mast cells also can change metabolic activity of receptors and thereby the sensitivity of olfactory epithelium. In the present study the mast cells in the olfactory epithelium of *E. suratensis* are thought to cause fluctuations in the production of mucus over the olfactory epithelium. As the terminal mucus film is believed to be an important factor in the olfactory process this may also influence the variations in the olfactory sensitivity [32].

The basal cells are small and lying adjacent to the central core and having no cytoplasmic processes reaching to the free surface. These cells can function as stem cells for regeneration of lost or damaged receptor cells, supporting cells and mucous cells [19, 33]. Using tritiated thymidine followed by autoradiography, Thornhill

[34] and Graziadei and Metcalf [24] have shown that the basal cells, apart from differentiating into supporting cells, also give rise to the olfactory receptor cells, which are continually replaced during life. Andres [35, 36] also suggested that the basal cells are the precursors of regenerating receptor cells.

The stratified epithelial cells abet the olfactory epithelium particularly in the non-sensory areas. The supporting cells may produce a serous secretion which affirms the continued directional flow along the surface of the epithelium. This flow removes the remains of the stimulating substances and keeps the receptors ready for new stimuli. Moulton and Beidler [32] reported that the supporting cell has complex secretory and nutritional functions.

Mucous cells of varying magnitude with secretory activity are profusely present in the olfactory epithelium of *E. suratensis*. The secreted mucin from the mucous cells maybe help in the smooth flow of water circulation in the olfactory chamber by binding microscopic debris which is ejected out through the posterior nostril to keep free the receptor surface. This is special adaptive feature of this bottom dwelling teleost. This is in compliance with the findings of Rahaman and Khan [37] in the olfactory epithelium of *Anabas testudineus* and Bandyopadhyay and Data [13] in the olfactory organ of *Heteropneustes fossilis*.

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6. References

1. Hara TJ. Mechanism of olfaction. Fish chemoreception, Chapman & Hall, London, 1992, 151-170.
2. Hansen A, Zeiske E. The peripheral olfactory organ of the zebrafish, *Danio rerio*: an ultrastructural study. Chem Senses 1998; 23:39-48.
3. Wilson DA. Fish smell. Focus on "odorant specificity of single olfactory bulb neurons to amino acids in the channel catfish". J Neurophysiol 2004; 92: 38-39.
4. Ruzhinskaya NN, Gdovskii PA, Devitsina GV. Chloride cells, a constituent of the fish olfactory epithelium. J Evol Biochem Physiol 2001; 37:89-94.
5. Mana RR, Kawamura G. Olfactory organs of two pelagic teleost fish-Opah (*Lampris guttatus*) and Dolphin fish (*Coryphaena hippurus*). South Pacific Study 2002; 22:53-64.
6. Bhute YV, Baile VV. Organization of the olfactory system of the Indian major carp *Labeo rohita* (Ham.): a scanning and transmission electron microscopy study. J Evol Biochem Physiol 2007; 43:342-349.
7. Chakrabarti P, Ghosh SK. Ultrastructural organisation and functional aspects of the olfactory epithelium of *Wallago attu* (Bleeker). Folia Morphol 2009; 68: 40-44.
8. Ma AJ, Wang XA. Functional morphology of the olfactory organ of the tongue sole, *Cynoglossus semilaevis*. Chin J Oceanol Limnol 2010; 28:209-217.
9. Ghosh SK, Chakrabarti P. Cellular organization and functions of the olfactory epithelium of pearl spot *Etroplus suratensis* (Bloch): a light and scanning electron microscopic study. Folia Morphol 2010; 69:154-159.
10. Ghosh SK, Chakrabarti P. Histological organization and microarchitecture of various cells lining the olfactory epithelium of *Rita rita* (Hamilton, 1822) (Siluriformes: Bagridae). Biological Lett 2012; 49:89-96.
11. Ghosh SK, Chakrabarti P. Topological organization and functional aspects of the olfactory epithelium of whipfin silver biddy *Gerres filamentosus* (Cuvier 1829). Pakistan Journal of Biological Sciences 2013; 16:245-250.
12. Ojha PP, Kapoor AS. Histology of the olfactory epithelium of the fish, Labeo rohita (Ham. Buch.). Arch Biol Bruxelles 1973a; 44: 425-441.
13. Bandyopadhyay SK, Datta NC. Morphoanatomy and histology of the olfactory organs of air-breathing catfish, *Heteropneustes fossilis* (Bloch). J Anim Morphol Physiol 1996; 43:85-96.
14. Chakrabarti P. Histological and histochemical studies on the olfactory rosette of *Mugil parsia* (Hamilton). Folia Morphol 2005; 64:41-46.
15. Ghosh SK, Chakrabarti P. Cellular architecture and functional aspects of the olfactory rosette of *Wallago attu* (Bleeker). Turk J Fish Aquat Sci 2009; 9:187-190.
16. Chakrabarti P, Ghosh SK. Histological and histochemical studies of the olfactory organ in bagrid catfish *Rita rita* (Hamilton, 1822). Journal of Biological Sciences 2013; 13:242-249.
17. Mallory FB. The aniline blue collagen stain. Stain technol 1936; 11:101.
18. Hara TJ. Olfaction in fish. Prog Neurobiol 1975; 5:271-335.
19. Zeiske E, Theisen B, Breucker H. Structure, development and evolutionary aspects of the peripheral olfactory system. In. *Fish chemoreception* (ed. Hara TJ). Chapmann & Hall, London 1992, 13-39.
20. Hara TJ. The diversity of chemical stimulation in fish olfaction and gestation. Rev Fish Biol Fish 1994; 4:1-35.
21. Zeiske E, Melinkat R, Breucker H, Kux J. Ultrastructural studies on the epithelia of the olfactory organ of Cyprinodonts (Teleostei, Cyprinodontoidae). Cell Tiss Res 1976; 172:245-267.
22. Yamamoto M. Comparative morphology of the peripheral olfactory organ in teleosts. In. *Chemoreception in fishes* (ed. Hara TJ). Elsevier, Amsterdam, 1982, 35-59.
23. Datta NC, Das A. Anatomy of the olfactory apparatus of some Indian Gobioids (Pisces: Perciformes). Zool Anz Jen 1980; 3:241-252.
24. Graziadei PPC, Metcalf JF. Autoradiographic and ultrastructural observations on the frog's olfactory mucosa. Z Zellforsch 1971; 116:305-318.
25. Camacho S, Ostos-Garrido MV, Domezain A, Carmona R. Study of the olfactory epithelium in the developing sturgeon characterization of the crypt cells. Chem Senses 2010; 35:147-156.
26. Zippel HP, Sorensen PW, Hansen A. High correlation between microvillous olfactory receptor cell abundance and sensitivity to pheromones in olfactory nerve-sectioned goldfish. J Comp Physiol 1997; 180:39-52.
27. Bakhtin EK. Peculiarities of the fine structure of the olfactory organ of *Squalus acanthias*. Tsitol 1977; 19:725-731.
28. Bannister LH. The fine structure of the olfactory surface of teleostean fishes. Q J Micr Sci 1965; 106:333-342.
29. Hernadi L. Fine structural characterization of the olfactory epithelium and its response to divalent cations Cd^{2+} in the fish *Alburnus alburnus* (Teleostei, Cyprinidae): a scanning and transmission electron microscopic study. Neurobiol 1993; 1:11-31.

30. Saini KD, Breiphol W. Frequency of mast cells in the olfactory mucosa of Rhesus monkeys. Cell Tissue Res 1977; 178:671.
31. Bertmer G. Structure and function of the olfactory mucosa of migrating Baltic trout under environmental stresses, with special reference to water pollution. In. *Fish chemoreception* (ed. Hara TJ). Elsevier, Amsterdam 1982, 395-422.
32. Moulton DG, Beidler LM. Structure and function in the peripheral olfactory system. Physiol Rev 1967; 47:1-52.
33. Frabman AI. Developmental biology of olfactory sensory neurons. Semin Cell Biol 1994; 5:3-10.
34. Thornhill RA. Cell division in the olfactory epithelium of the lamprey, *Lampetra fluviatilis*. Z Zellforsch 1970; 109:147-157.
35. Andres KH. Der Feinbau der Regio olfactoria von Makrosmatikern. Z Zellforsch 1966; 69:140-154.
36. Andres KH. Der olfaktorische saum der Katze. Z Zellforsch 1969; 96:250-274.
37. Rahmani AR, Khan SM. Histology of the olfactory epithelium and the accessory nasal sacs of an anabantoid fish, *Anabas testudineus* (Bloch). Arch Biol 1980; 91:397-411.