Indian Journal of Biological Sciences, 20 : 36 – 39, 2014

FUNCTIONAL ANATOMY OF CELLULAR JUNCTIONS IN OLFACTORY NEUROEPITHELIUM OF *PSEUDAPOCRYPTES LANCEOLATUS* (BLOCH AND SCHNEIDER)

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ABSTRACT ■ The olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801) is a pseudostratified tissue structure that includes different types of cells *i.e.,* sensory receptor cells, supporting cells, basal cells, *etc*. The various types of cell junctions (*viz.,* tight junctions, desmosomes, gap junctions and hemidesmosomes) between the neuroepithelial cellular components has been described and characterized under transmission electron microscope (TEM: Morgagni 268D). The presence of these cellular junctions or complexes is significantly involved in various types of vesicle mediated cellular communications during olfaction.

Key words: *Pseudapocryptes lanceolatus*, olfactory, desmosomes, hemidesmosomes, vesicle, *etc.*

INTRODUCTION

Olfactory neuroepithelium is a unique part of peripheral nervous system of vertebrates (Farbman, 1992). It shows multicellular appearance and forms pseudostratified structure (Firestein, 2001). On the basis of distinct morphology, the neuroepithelial cellular components are categorized as sensory receptor cells, supporting cells and basal cells (Hansen *et al.,* 2003). These cells are commonly observed in across the vertebrate phyla and in particular the fishes (Eisthen, 2002). The olfaction in vertebrates is mediated through a type of bipolar neuron *viz.,* olfactory sensory receptor cell that is involved in detection and discrimination of various odorants from the external environment (Hildebrand and Shepherd, 1997). The perceived chemical cues are generally conveyed to the brain through neural networks (Hamdani and Døving, 2007). Cell junctions are the most significant part of neuroepithelial system which is hardly characterized in vertebrate peripheral olfactory structure. This study considered the olfactory neuroepithelium of *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801) [IUCN Red List Category: Least Concerned] as a likely model to explore the ultrastructural features of cellular junctions or complexes between sensory and non-sensory neuroepithelial cellular components as well as their functional significance in olfaction.

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MATERIALS AND METHOD

The adult, sex-independent specimens of *P. lanceolatus* were collected from the local markets of South 24 Parganas and brought to the laboratory for acclimatization [temperature: 20° to 25°C, humidity: >40%, *etc.*]. The specimens were anaesthetized by using MS – 222 (dose: 100mg/liter – 200mg/liter). Olfactory apparatus of *P. lanceolatus* were dissected out from the antero-dorsal side of the head and separately fixed in 2.5% glutaraldehyde in 0.1 M phosphate buffer (pH. 7.2-7.4) at 4°C for 2 hours. After primary fixation, the olfactory tissues were rinsed in the same buffer and then fixed in 1% osmium tetroxide in 0.1 M phosphate buffer (pH. 7.2-7.4) for 1 hour at 30°C. The olfactory tissues were then rinsed in the same buffer and dehydrated in chilled acetone. The tissues were embedded in Araldite CY212 (TAAB, UK) and resin polymerized for 48 hours at 60°C. The transverse ultrathin sections (70nm – 80nm) of the olfactory lamella were cut by using ultramicrotome (Leica Ultracut – UCT), collected on copper grids and stained with uranyl acetate and lead citrate respectively. The sections were observed under Morgagni 268D transmission electron microscope (Fei Electron Optics, Eindhoven, The Netherlands) operated at 80kV. Digital images were analysed at by using iTEM software (soft imaging system, Münster, Germany) attached to the microscope.

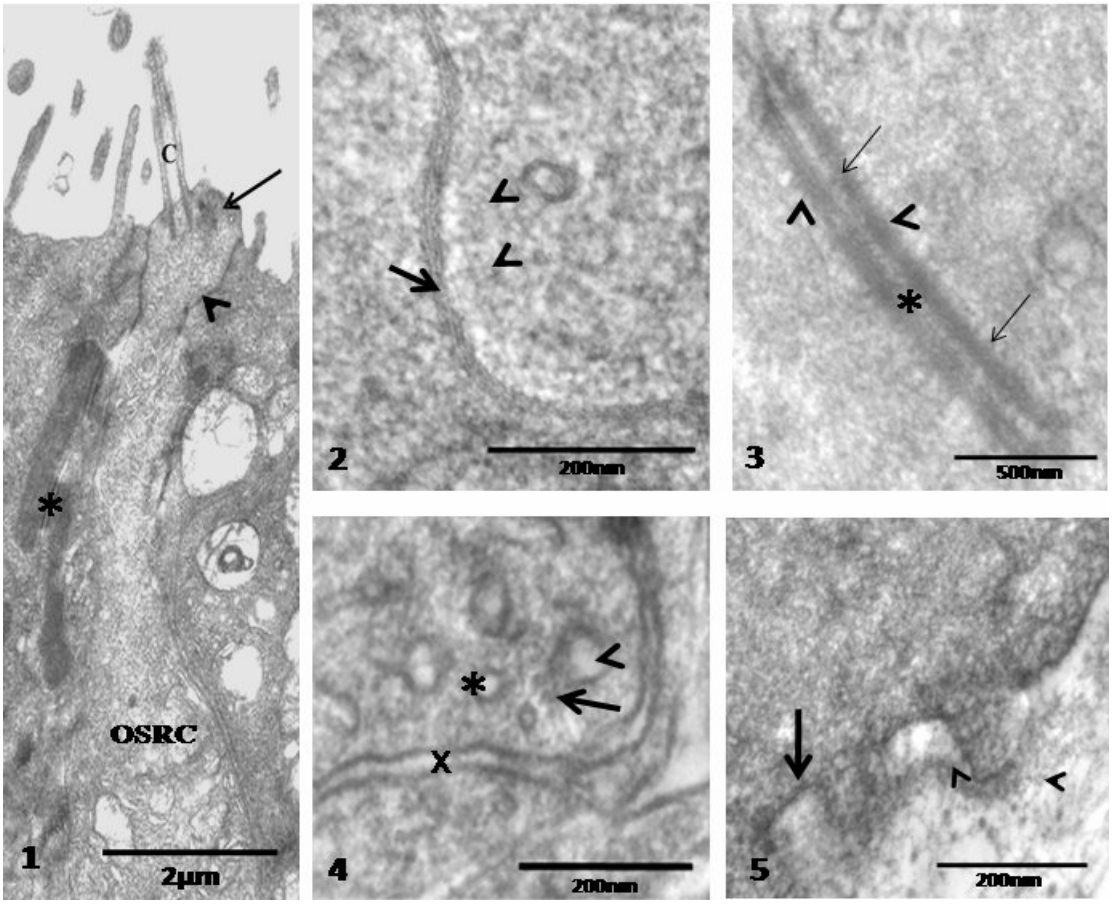

Fig. 1 – The electron micrograph indicates olfactory sensory receptor cell (OSRC) with prominent cilia (C) at the apical olfactory knob (→). The tight junction (>) and desmosomes (*) are also noted at the different depth of olfactory neuroepithelium.

Fig. 2 – Tight junction (→) is marked at the apical part of olfactory neuroepithelium of *P. lanceolatus.* Several vesicles (diameter: 5nm – 10nm) are also present near the tight junction.

Fig. 3 – Photomicrograph shows desmosome (*) along with intermediate filaments (>) at the middle part of olfactory neuroepithelium of *P. lanceolatus*. Vesicles (diameter: 10nm – 15nm) are also noted along with intermediate filaments of desmosome (→).

Fig. 4 – The terminal part of axon in olfactory sensory receptor cell is showing synaptic cleft (X). Different types of vesicles *i.e.,* small vesicles (→), coated vesicle (*) and synaptic vesicles (>) are also identified at this region.

Fig. 5 – Hemidesmosome (→) is marked at the basal region of olfactory neuroepithelium of *P. lanceolatus*. Small vesicles (>) are prominently accumulated just beneath the basal lamina.

RESULTS

The pseudostratified olfactory neuroepithelium of *P. lanceolatus* shows different types of cellular junctions between sensory receptor cells and supporting cells at various depths (Fig. 1). The apical part of the bipolar sensory receptor cell shows a prominent swelling to form olfactory knob towards nasal cavity. The olfactory knob possesses 4 to 6 number of kinocilia that are supported by axial microtubules and basal particles (Fig. 1). The lateral part of plasmamembrane in olfactory knob and supporting cells shows distinct tight junctions (Figs. 1 and 2). The thickness of tight junctions or zonula occludens is measured about 20nm. Numerous small vesicles having diameter of 5nm to 10nm are crowded at both the side of tight junctions (Fig. 2). Desmosomes between sensory receptor cell and supporting cell, appear at the middle part within olfactory neuroepithelium of *P. lanceolatus* (Figs. 1 and 3). The thickness of desmosomes varies from 60nm – 80nm. The intermediate filament like structures (diameter: 5nm to 7nm) are also noted in association with desmosomes (Fig. 3). Vesicles (diameter: 10nm to 15nm) are noted along with the intermediate filaments of desmosomes (Fig. 3). The gap junctions are found only in between olfactory sensory receptor cells within the olfactory neuroepithelium. The terminal knob of olfactory sensory receptor cell is forming gap junction or synaptic cleft (width: 20nm to 40nm) with the other sensory receptor cell (Fig. 4). Different types of vesicles *i.e.,* small vesicles (diameter: 20nm – 30nm), coated vesicle (diameter: 60nm – 70nm) and synaptic vesicles (diameter: 70nm - 90nm) are prominently marked at the synaptic knob of olfactory sensory receptor cell (Fig. 4). The lower part of olfactory neuroepithelium of *P. lanceolatus* shows hemidesmosomes on basal lamina (thickness: 70nm approx.) (Fig. 5). Small vesicles (diameter: 5nm – 10nm) are frequently marked just beneath the basal lamina near to the hemidesmosomes (Fig. 5).

DISCUSSION

Intercellular junctions in epithelial system are regarded as highly specialized structure for cell-cell interaction (Alberts *et al.,* 2008). The tight junctions are thought to act as a barrier for some biomolecule at the apical and basolateral domain of plasma membrane (Alberts *et al.,* 2008). In olfactory neuroepithelium of *P. lanceolatus*, we have observed that the tight junctions are only restricted at the apical part. We believe that

the tight junction may provide an adhesive contact between sensory receptor cell and neighbouring supporting cell. This structure is also helpful for defense against invading pathogens which may enter into the nasal cavity along with water during olfaction in fishes. Desmosomes are important for serving as anchoring sites between the epithelial cells (Alberts *et al.,* 2008). The intermediate filaments of desmosomes may form a structural framework for mechanical support where as gap junctions are important for neural communication between the olfactory sensory receptor cells. At the cell substratum interface of epithelial cell, the adhesion to matrix is maintained by hemidesmosomes (Green and Jones, 1996). Therefore all the cellular junctions *i.e.,* tight junctions, desmosomes, gap junctions and hemidesmosomes in olfactory neuroepithelium of *P. lanceolatus* are playing mutifunctional roles for providing mechanical support as well as in cellular communication. Apart from that, the crowding of morphoanatomically different types of vesicles is identified in association with various intercellular junctions. The morphoanatomical variation of vesicles may be indicative of functional diversity of the concerned vesicles (Alberts *et al.,* 2008). This study reveals that these vesicles are present at the site specific subcellular compartments within the olfactory sensory receptor neuron. The diversity and putative docking of various types of vesicles at the terminal part of axon (*i.e.,* synaptic knob) is representing the polarized nature of concerned vesicles as well as site specific docking for olfactory signal transduction (De and Sarkar, 2014). We assume that, these vesicles are significant for vesicle-mediated cellular communication during olfaction.

ACKNOWLEDGEMENTS

Authors are thankful to Prof. T. C. Nag, Department of Anatomy, AIIMS, New Delhi for EM facilities and UGC [F. No. 41-161/2012 (SR) of dated 13.07.2012] for necessary funding.

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