



Fig. 4. High power view of part of the germinal vesicle (upper left hand corner) and of the perinuclear basophilic organelles of the same oocyte as in Figure 3. The cross-sectional area of most of these organelles is stirrup-shaped in this tangential section.

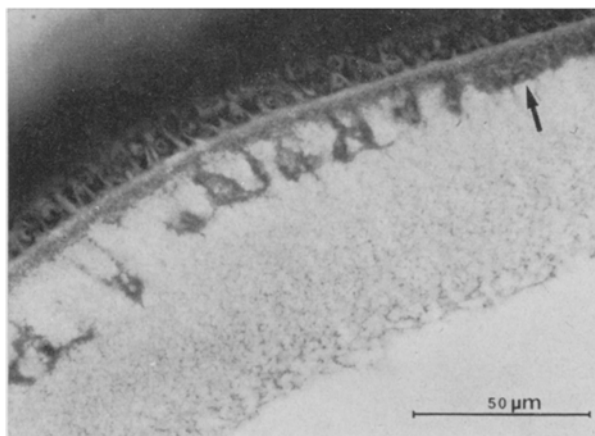


Fig. 5. High power view of perpendicular germinal disc section of an oocyte, 4.5 mm diameter after staining with toluidine blue. The perigerminal cortex is indicated by an arrow.

organelles are usually the last to disappear. In oocytes with a diameter greater than 8 mm (in a laying quail there are usually only some 3 oocytes of this size) these subcortical masses are usually no longer visible.

**Conclusion.** In Japanese quail oocytes of 4.5 to 7 mm diameter subcortical cytoplasmic organelles, situated about the germinal vesicle, show the singular property of being arranged in concentric circles (often also a radial disposition can be seen) about the dorsoventral axis of the future quail embryo (the dorsal side being situated at the surface of the cicatricular region, the ventral side towards the deeper part). This indicates an evident RNA- and probably also DNA-bound cytoplasmic polarity. As no other gradient is to be seen by this technique, it seems as though the quail oocyte in the stage studied exhibits symmetry around this axis only.

**Résumé.** Chez la caille japonaise pondeuse la région cicatriculaire des oocytes, au début de leur période de

grand accroissement, présente une symétrie radiale cytoplasmique uniaxiale. Cette symétrie radiale est caractérisée par l'emplacement d'organites cytoplasmiques subcorticaux sur des cercles concentriques autour de la vésicule germinative.

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## Occurrence of Synapses in Olfactory Epithelium of Fish

The olfactory epithelium of vertebrates is known to comprise<sup>1-5</sup> olfactory or sensory cells intermingled with supporting cells. Other cellular elements present are the mucin-secreting goblet cells and the basal cells, the latter being arranged in a layer at the base of the epithelium. The structure of the olfactory epithelium in fishes has been studied by several workers<sup>6-9</sup> and it has been concluded that it shows no fundamental variation from the general vertebrate pattern.

The commonly accepted picture of a sensory cell of the olfactory epithelium is that it is a bipolar primary neurone with its peripheral dendritic end swollen into an olfactory vesicle bearing sensory hairs or cilia. The greater part of the body of the sensory cell is occupied by its spherical nucleus, there being only scant cytoplasm present around it. The thin, centrally directed axons of sensory cells reach the olfactory bulb directly, there being no interconnecting neurones. This being so, the olfactory organ of vertebrates, as pointed out by KLEEREKOPER<sup>10</sup>, is 'the most primitive among the receptors which has remained un-

affected by evolutionary changes in its peripheral organization'. On reaching the olfactory bulb, the axons of sensory cells interlace with dendrites of mitral cells in structures called glomeruli. The axons of mitral cells gradually join to constitute, together with axons of spindle

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<sup>4</sup> A. J. D. DE LORENZO, *J. Biophys. biochem. Cytol.* 3, 839 (1957).

<sup>5</sup> D. FRISCH, *Am. J. Anat.* 127, 87 (1967).

<sup>6</sup> O. TRUJILLO-CENÓZ, *Z. Zellforsch.* 54, 654 (1961).

<sup>7</sup> L. H. BANNISTER, *Q. J. microsc. Sci.* 106, 333 (1965).

<sup>8</sup> J. A. F. WILSON and R. A. WESTERMAN, *Z. Zellforsch.* 83, 196 (1967).

<sup>9</sup> G. GEMNE and K. B. DOVING, *Am. J. Anat.* 126, 457 (1969).

<sup>10</sup> H. KLEEREKOPER, *Olfaction in Fishes* (Indiana University Press 1969).

cells, the secondary olfactory fibres which extend directly to the telencephalon, without synapses<sup>11-15</sup>.

We have been studying the structure of olfactory epithelium in the teleost *Channa* (= *Ophicephalus*) *punctatus*. Besides the sensory, supporting, basal and goblet cells, we have been able to identify yet another type of cell, the spindle-shaped cell or the secondary neurone. These secondary neurones lie at a level lower than that of the primary neurones or olfactory cells. The axon of a primary neurone in *C. punctatus* does not extend as an independent element towards the basement membrane. Instead, the axonal end of this cell synapses with the dendritic end of a secondary neurone and it is the axon of the secondary neurone which actually passes out of the basement membrane. Figure 1 is a photograph of a synapse between the primary and secondary neurones as seen in stained preparation of a paraffin section of the olfactory epithelium, while Figure 2 shows the same taken after isolating individual cellular elements through centrifugation of fresh and unfixed olfactory lamellae stained with methylene blue. An examination of frozen sections processed according to Baker's acid haematein method for determining the localization of phospholipids in the olfactory epithelium confirmed the above observations, since, apart from mucin granules of goblet cells, a positive reaction was obtained exclusively in the primary and secondary neurones and in their synaptic connections (Figure 3).

In fishes and all other vertebrates studied so far, the axon of the primary neurone is stated to synapse with the dendrite of a mitral cell after it has reached the olfactory bulb. The synopsis of the axonal end of the primary neurone with the dendritic end of a secondary neurone within the olfactory epithelium, therefore, presents a unique situation in *C. punctatus*. Is it conceivable that the secondary neurone in this fish represents a mitral cell and its axon, therefore, corresponds to a secondary olfactory fibre which contributes to the formation of the olfactory tract? There is a possibility that this may be so. In *C. punctatus* the olfactory lobes are distinguishable while the bulbs are absent and two lateral and one medial bundles of fibres arise directly from the olfactory organ and constitute together the olfactory tract. We are extending our studies to some other species of fish in order to determine whether or not synapses similar to those observed in *C. punctatus* occur in their olfactory epithelia and, after a preliminary examination, we are convinced that there is need for a careful reexamination of this aspect in fishes, and probably in other vertebrates as well.

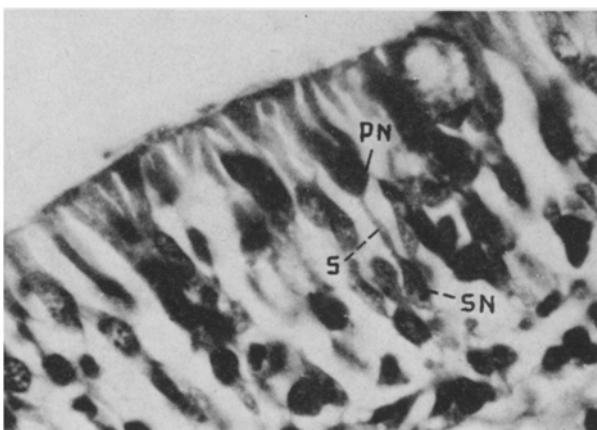


Fig. 1. Olfactory epithelium of *C. punctatus* to show the synapse (S) between primary (PN) and secondary (SN) neurones.  $\times 1,000$ .

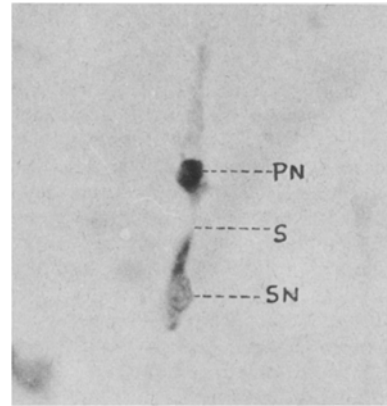


Fig. 2. Synapse (S) between primary (PN) and secondary (SN) neurones as seen after isolation of cellular elements through centrifugation of fresh and unfixed olfactory lamellae of *C. punctatus* stained in methylene blue.  $\times 400$ .

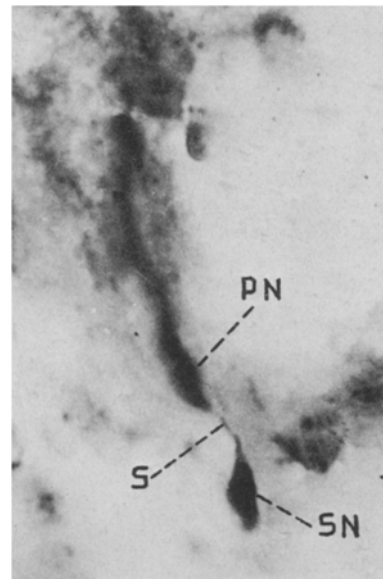


Fig. 3. Synapse (S) between primary (PN) and secondary (SN) neurones as seen after processing frozen sections according to Baker's acid haematein method for demonstrating phospholipids in olfactory epithelium of *C. punctatus*.  $\times 1,000$ .

**Zusammenfassung.** Nachweis von Synapsen in der Riechschleimhaut des Fisches *Channa punctatus*, die als Verbindung zwischen primärer und sekundärer Riechzelle aufgefasst werden.

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<sup>15</sup> R. NIEUWENHUY, in *Progress in Brain Research* (Ed. Y. ZOTTERMAN; Elsevier, Amsterdam 1967); vol. 23.

<sup>16</sup> We wish to thank Dr. RAMESHWAR SINGH, our colleague in this department, for his helpful suggestions.