

FUNCTIONAL ANATOMY OF OLFACTORY SENSE ORGANS

ALFRED CUSCHIERI

Department of Anatomy, University of Malta

The olfactory organ is the most primitive of the organs of special sense and the one with the simplest structural organization, and yet it has defied a clear exposition of the way in which it perceives and distinguishes between different odours. Undoubtedly one of the main reasons for this has been the greater interest which scientists have taken in other aspects of sensory physiology, particularly sight and hearing, impairment of which produces severe handicap, and is positively detrimental to the independent survival of an affected individual. The sense of smell is not so indispensable for Man; its loss is a more tolerable burden depriving him only of the emotional experiences aroused by odours.

Yet the olfactory organ in Man is endowed with a remarkable degree of sensitivity and a finesse of its discriminatory powers. The sensitivity of the human olfactory mucosa is more than one hundred times better than that of the best gas chromatograph, and its discriminatory powers enable Man to distinguish between an immense variety of odours and to identify specifically those with which he is familiar. For example almost anyone can distinguish between water, gin, eau-de-cologne, kerosene, acetone and other substances, all of which look very much alike, by their distinctive odours. An experienced perfumer can do very much better and is frequently capable not only of distinguishing between a large variety of lavender oils but also of naming their country of origin.

The significance of odours to man is mainly psychological and emotional. In most other animals, including mammals odours have a more profound significance in terms of survival. In fact most animals rely on their sense of smell for the detec-

tion and recognition of prey for feeding, for escaping from predators and for selecting their mates for reproduction. However in animals it is much more difficult than in man to estimate the sensitivity and the range of discriminatory capacity of their olfactory organs. Certainly many animals are much more keenly scented than Man as can be appreciated for example from the ability of dogs to follow a trail and to distinguish the odour complex of a particular individual from all other extraneous odours.

Not all animals share the same degree of olfactory acuity and it is customary to categorize them roughly as macrosmatic and microsmatic. Attempts to identify an anatomical basis for this difference have shown that there is no simple relationship between the degree of development of the sense of smell and the size of the olfactory area or its density of receptors.

Olfaction is the most primitive of the special senses, the first to be developed in the evolutionary scale and the first sense which enabled perception of objects from a distance without the necessity of actual physical contact. The olfactory organ also has a simple structural organization consisting of receptor cells surrounded by supporting cells and overlying a layer of basal cells. Besides, the receptor cells are themselves primary sensory neurons the cell bodies of which lie close to the sensory surface, a common feature in invertebrates but unique in the vertebrate series. Their axons proceed directly to the cerebral cortex without any complicating synapses; the olfactory bulb in which they terminate is developmentally a forward extension of the cerebral hemisphere and histologically has the structure of a cortex. It is pertinent to point out that this direct connection of the receptors with the

cerebral cortex is considered to be an expression of the fact that, from an evolutionary point of view, the cerebral hemispheres were initially developed as correlation centres for the olfactory sense.

In spite of this apparent simplicity in structural organization, the mechanisms underlying olfactory perception and discrimination pose serious and far reaching problems; and when we attempt to interpret the electrical responses of the olfactory epithelium to odour stimulation it becomes clear that we are dealing with a far more complex system than was originally supposed. Apart from the receptors there are other factors in the olfactory epithelium and its environment which must be considered, such as the structure and function of the supporting and basal cells, and the chemical composition and properties of the surface fluid which bathes the receptor endings and which is secreted by Bowman's glands underlying the epithelium.

The nature of the olfactory stimulus itself present a number of problems. Unlike visual and auditory stimuli, namely light and sound waves whose physical properties and variables are accurately understood, the physical and chemical properties of odoriferous molecules which constitute the olfactory stimulus are still the subject of considerable controversy and much speculation.

The Fine Structure of the Olfactory Mucosa

It is remarkable that the olfactory epithelium is very similar in all vertebrates from cyclostomes to mammals. It is a pseudostratified epithelium, much thicker than the surrounding non-sensory respiratory epithelium of the nasal cavity. The classification of the component cells into three distinct types namely receptor cells, supporting cells and basal cells was first established by Schultze in 1856 and is still valid today. Histologically the three cell types are recognizable from the position of their nuclei (Fig. 1 and 2). Deep to the epithelium are situated the olfactory fasciculi of unmyelinated axons and the distinctive Bowman's glands whose ducts

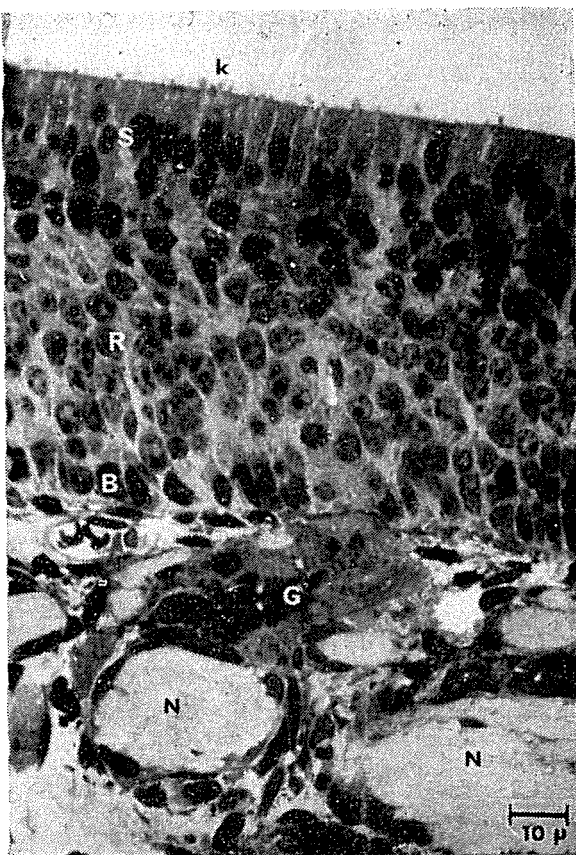


Fig. 1: Vertical section through the olfactory mucosa of an adult mouse. The supporting cell nuclei (S), the receptor nuclei (R) and the basal cell nuclei (B) form three distinct zones. Olfactory knobs (K) can be seen at the surface of the epithelium. Olfactory nerve bundles (N) and a Bowman's gland (G) are present deep to the epithelium 1 μ m epoxyresin section stained with toluidine blue.

extend vertically to the surface of the epithelium. Here the secretions of Bowman's glands form an adherent film of fluid. Further details which have been added on to this basic picture have been obtained from various lines of research. A considerable amount of knowledge regarding the ultrastructure of the component cells has been gained from numerous electron microscope studies of the olfactory mucosa amongst which are those in

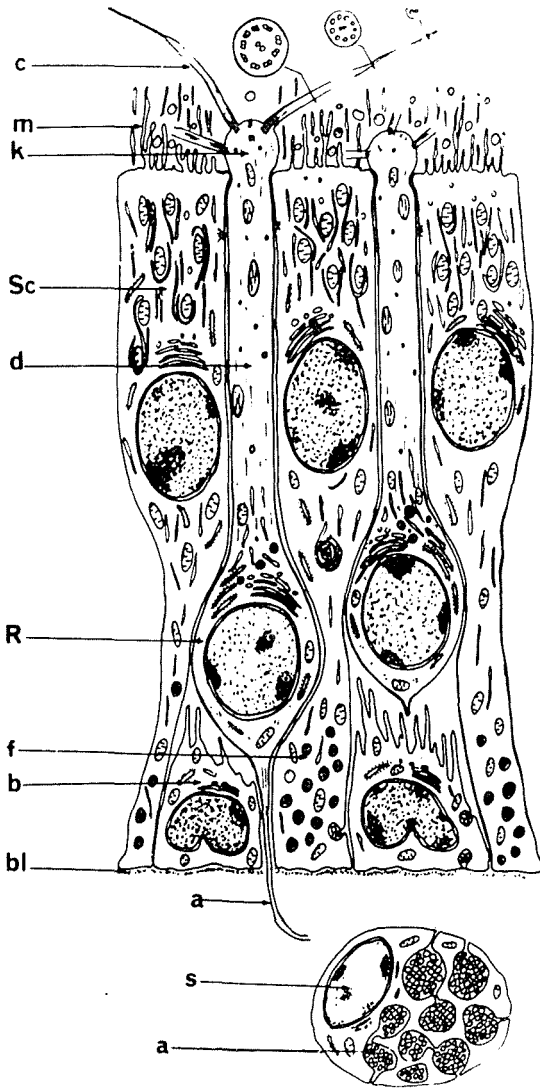


Fig. 2: Diagram illustrating the structure of the olfactory epithelium. C — cilia; m — microvilli; K — olfactory knobs; — Sc — supporting cells; d — distal process of receptor cell bodies; f — foot process of supporting cell; b — basal cell; bl — basal lamina; a — olfactory axon; s — schwann cell.

the frog (Reese, 1965), rabbit (de Lorenzo 1957), mouse (Frish, 1967), primates and Man (de Lorenzo, 1970). More recently attention has also been turned to the fine structural development of the olfactory mucosa (Briephol et al 1973; Cuschieri

and Bannister 1975) contributing to a better understanding of certain ultrastructural features and their functional significance.

A few histochemical studies (Baradi and Bourne 1953; Brncnshtein 1965; Shantha and Nakajima, 1970; Shapiro, 1970; Cuschieri and Bannister, 1974) have described the location and distribution of various enzymes and other substances in the olfactory mucosa. Such histochemical studies are potentially useful in elucidating some aspects of the metabolism and functions of the various cells but the interpretation of the precise metabolic role of the enzymes demonstrated has been complicated by the existence of variations amongst the different animals studied and by the inherent limitations of the histochemical techniques employed.

Of direct relevance to the study of olfaction is the vomeronasal organ, a paired structure present in the nasal cavity of many animals, being particularly well developed in reptiles and some mammals but vestigial or absent in primates and birds. It usually takes the form of a diverticulum lined in its greater part by a sensory epithelium similar in basic structure to the olfactory epithelium, and also sensitive to odours. Although it shows some minor differences from the olfactory epithelium both in structure and in its electrophysiological responses to odours it has provided useful information on the sense organs of smell.

The following account of the detailed structure of the component cells of the olfactory mucosa will attempt to correlate the results obtained from these various lines of research. In this way a better evaluation of the current status of knowledge of the anatomy of the olfactory organ will be possible.

The Olfactory Receptors

The olfactory receptors are bipolar neurons. A thick "distal process" is directed towards the surface of the epithelium where it is expanded into a terminal knob projecting for about $2\mu\text{m}$ beyond the surface. The distal process, sometimes inaccurately named the olfactory dendrite,

contains longitudinally oriented microtubules, vesicles and mitochondria. The terminal knob bears a variable number of cilia (usually not exceeding 20) which arise from basal bodies within its cytoplasm. They also contain free contriesoles and an abundance of mitochondria indicating a high level of metabolic activity as is to be expected in sensory nerve endings.

"The cilia" are the parts of the olfactory receptors which are most readily accessible to odoriferous molecules. It has therefore been generally assumed that the odour receptor sites, where the initial events in olfactory stimulation occur, lie on their surface membrane but there is no conclusive evidence for this. It should be noted that modified cilia are also found in other sensory organs, notably the rods and cones in the retina, and appear to be the initial transducing elements (Stiegh, 1962). 1962).

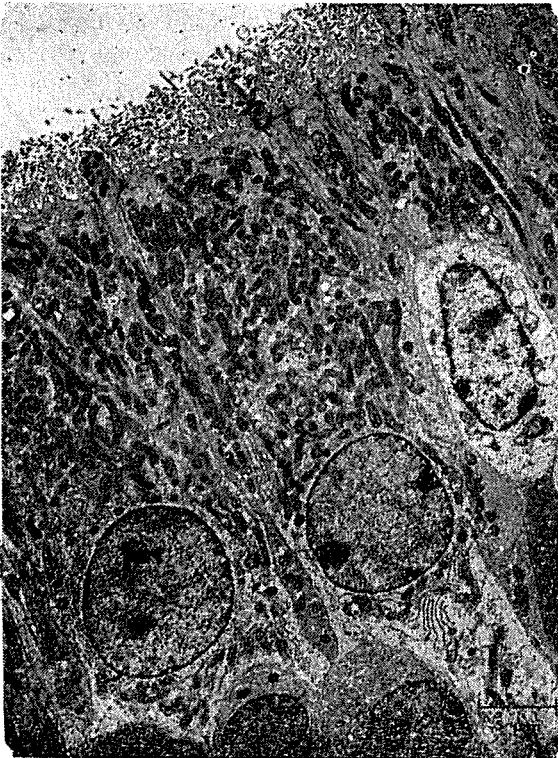


Fig. 3: Electron micrograph of superficial part of the olfactory epithelium. Note the abundance of mitochondria in the supra nuclear parts of the supporting cells.

The olfactory cilia possess in their proximal parts an array of nine pairs of microtubules surrounding one central pair, an arrangement typical of motile cilia. The modified cilia of other sense organs typically lack the central pair of microtubules. The olfactory cilia show a modified structure only in their distal part which tapers into a long thin segment containing decreasing numbers of microtubules (Reese, 1965) (Fig 2). The ciliary structure does not, however, appear to be indispensable for olfactory function since receptor endings having microvilli, instead of cilia are found in certain species of fish (Bannister, 1965) and in the vomeronasal organs of reptiles and mammals (Altner and Muller, 1968; Bannister 1968).

The length and motility of cilia have been subjects of much disagreement. The ciliary movements which have been observed were slow, irregular and asynchronous and it has been suggested that such movement may have been stimulated by breakage of their delicate, thin distal parts (Reese, 1965). Even if ciliary movement does normally occur it is unlikely to have any mechanical significance in wafting odoriferous particles to and from the receptor sites or in the movement of the olfactory surface fluid. It would be more reasonable to suppose that the olfactory cilia simply provide an increased receptive area for the cell.

The technical difficulties involved in measuring the lengths of cilia have cast doubts on the validity of the reported measurements which have varied from 1 to 200 μm . The question of ciliary length is significant in so far as it determines whether the cilia are completely covered by the olfactory surface fluid which odour molecules would have to penetrate before reaching the receptor sites; or whether they are long enough to lie at the air-fluid interface directly accessible to air borne odours. In most electron micrographs the former appears to be the case.

The "cell body" of the receptor neuron is small, being only about 8 μm in diameter and is largely occupied by its vesicular nucleus. The scanty cytoplasm it contains is occupied by a lamellar system of

smooth-surfaced and granular endoplasmic reticulum and a well developed Golgi apparatus, (both indicative of synthetic activity), as well as large numbers of lysosomes some of which contain membranous remnants indicative of autophagic activity. Histochemical studies have shown that acid phosphatase is present in high concentration and is located mainly within the lysosomes and partly within the Golgi apparatus. The internal organization of the receptor cells suggests the occurrence of a continuous process of synthetic and degradative activity possibly providing a mechanism for cellular maintenance. Presumably a significant part of this process is renewal of the delicate exposed surface membrane of the receptor endings, which is prone to physical damage and to exhaustion by repeated stimulation.

The "proximal processes" of the receptors cells form the unmyelinated olfactory axons. These are amongst the smallest of axons having a diameter of $0.2 \mu\text{m}$. Within the epithelium they are enclosed in small invaginations of the supporting and basal cells. In the sub-epithelial tissues large numbers of axons are clustered together and collectively are enclosed within single invaginations of the surrounding Schwann cells. This arrangement is unique for olfactory nerves since unmyelinated axons elsewhere, such as those in the sympathetic grey rami, are enclosed singly in Schwann cell invaginations. It may also have functional implications possibly allowing for axonal interaction to occur although synaptic contacts between the axons have not been observed.

Are There Morphologically Distinct Types Of Receptors?

In an attempt to find some anatomical basis for olfactory discrimination several workers have claimed that they could distinguish different types of receptors and classified them according to such criteria as differences in shape, size and number of cilia. None of these classifications appears to be justified since differences in shapes and size of the receptors are to be expected from the close packing of the

cells in the epithelium. Electron microscope studies have also failed to detect any differences which would not be expected as a result of random variation and the idea that differences in receptor function may be reflected in their gross morphology is now tending to be discredited.

The Supporting Cells

The supporting cells surround the individual receptor cells and isolate them from one another. Each supporting cell extends from the surface to the basal lamina. From the free surface project long branched microvilli which extend beyond and enmesh most of the olfactory cilia. The part of the cell above the oval nucleus contains most of the cell cytoplasm, the most characteristic feature of which is the abundance of mitochondria surrounded by an elaborate system of smooth surfaced endoplasmic reticulum. This region of cytoplasm also contains an abundance of enzymes including dehydrogenases, cytochrome oxidase, adenosine triphosphatase and esterase all of which are present in concentrations far greater than in any other part of the olfactory epithelium. The supporting cells, therefore, far from being passive supporting elements are highly active metabolically and may have important functions.

In amphibians and reptiles secretion granules containing mucosubstances are present within the supporting cells indicating that these have a secretory function contributing to the formation of the olfactory surface fluid. In mammals and birds, however, the supporting cells show no evidence of secretory activity.

The way in which the supporting cells ensheath the receptors suggests that there may be a relationship between these two cells in a manner analogous to that between neuroglia and neurons. The supporting cells may be responsible for maintaining the chemical composition and, in particular, the ionic balance in the intercellular fluid which forms the immediate environment of the receptor cells. Molecular and ionic transport may also occur between the supporting and receptor cells.

It has been shown (Cuschieri 1972) that at the junctional complexes between these two cells alkaline phosphatase activity is present, an enzyme which would favour the occurrence of such transport processes at these sites. There could even be, as a result of such ionic interchange, some degree of electrical coupling between the receptor and supporting cells.

In the olfactory epithelium of mammals almost all the receptors are completely isolated from one another by the supporting cells. However, adjacent receptors are common in the vomeronasal organ and are also found in the olfactory epithelium of lower vertebrates. The degree of receptor cell isolation may be of functional importance since interaction might occur between adjacent receptors. Studies in the olfactory epithelium during development have shown that when the receptors are first formed they are grouped together in clusters and that they become separated from one another by the supporting cells during later stages of development (Cuschieri and Bannister, 1975). It would therefore appear that any close proximity of receptors in adult olfactory epithelia is the result of incomplete separation rather than of any specific functional association of receptors.

The Basal Cells

Like the supporting cells, the basal cells also show evidence of high metabolic activity and contain a variety of enzymes including succinic dehydrogenase, cytochrome oxidase and adenosine triphosphatase. Alkaline phosphatase is present in particularly high concentration and is located on the plasma membrane which is thrown into finger like processes extending between the receptor cells and also enclosing olfactory axons. This enzyme indicates that the basal cells are active in molecular transport across the base of the epithelium. This may be important for meeting the nutritional requirements of the olfactory epithelium which is much thicker than any other epithelium and yet is supplied only by a sub-epithelial vascular plexus. It is perhaps significant that

during embryonic development, before the basal cells have been differentiated, the olfactory epithelium is supplied by capillary loops invaginating deeply into the epithelium. A similar situation occurs in the adult vomeronasal organ which also lacks basal cells but is supplied by intra-epithelial capillary loops.

A further function which has been attributed to the basal cells is that of providing a blastema for the continuous regeneration of receptor and supporting cells. Autoradiographic studies (Moulton et al 1970) have shown that continuous cell proliferation occurs within the olfactory epithelium and that initial uptake of tritiated thymidine occurs mainly in clusters of nuclei close to the base of the epithelium. It is, however, unlikely that this is the only function of the basal cells, since they do not resemble undifferentiated cells in their structure or enzyme complement.

Bowman's glands and the Olfactory Surface Fluid

The surface of the olfactory epithelium is continually bathed by a highly tenacious film of fluid. In mammals it is derived from Bowman's glands only but in amphibians and reptiles the supporting cells also contribute their secretions (Graziadei, 1971).

The chemical composition of the olfactory surface fluid is only poorly known. A few enzymes such as acid phosphatase and succinic dehydrogenase have been demonstrated histochemically in the surface fluid and in the Bowman's glands of mammals (Baradi and Bourne, 1953; Cuschieri, 1974b).

The secretion of Bowman's glands have been reported to contain acid mucosubstances in some animals and neutral mucosubstances in others. The supporting cells of amphibians and reptiles also contain neutral or acid mucosubstances. The histochemical composition of the mucosubstances secreted by Bowman's glands have been analysed in some detail in mice. It has been shown that they differ from all the other secretions in the nasal cavity in that they contain sulphated mucosub-

stances, (Cuschieri 1974 a), a finding which has been confirmed by autoradiographic studies with the isotope sulphur-35 (Dodson et al, 1976). Similar findings have also been obtained in other mammals (unpublished results).

Functions of the Surface Fluid

Undoubtedly the olfactory surface fluid is important for protection against drying, osmotic damage, infection and mechanical abrasion, functions common to secretions covering most other mucous membranes. In addition it is to be expected that this surface fluid will influence the access of odour molecules to the receptor sites and their subsequent removal. This may be affected by such factors as solubility of the odour molecules in the surface fluid, the air/fluid partition coefficient and physical interactions between polar groups on the odour molecule and in the surface fluid. Chemical interactions may also occur and it is possible that certain odour molecules may be altered by enzymic action.

It is also likely that the surface fluid forms a reservoir for inorganic ions, which are necessary for the electrical events associated with sensory transduction, and a conduction pathway for the flow of ions at the olfactory surface. It is interesting that sulphated mucosubstances occur in the olfactory surface fluid of mammals since similar mucosubstances have also been demonstrated in the central nervous system (Saigo and Egami, 1970) and in the extracellular fluid at the nodes of Ranvier (Langley and Landon, 1967). These polyanions are known to bind inorganic cations strongly.

The Olfactory Pigment

It had long been noted that in many animals the olfactory mucosa had a yellowish or brownish colour. It had also been assumed that the pigment involved was important in olfaction. This belief was supported by the observation that certain albino animals, which were presumed to lack pigment in their olfactory mucosa, died from eating poisonous plants (e.g. St. John's wort, 'Hypericum crispum') because it was thought that they had a poor

sense of smell and were unable to recognise them. Experimental evidence has not supported these assumptions. In fact it has been shown that albino animals do not lack olfactory pigment (Moulton, 1962) and that albino rats had lower thresholds for certain odours than pigmented rats (Moulton, 1960). The death of albino animals from eating St. John's wort is almost certainly the result of photodynamic sensitisation from exposure of unpigmented skin (Horsley, 1934). Pigmented animals are unharmed by eating the plant.

The olfactory pigment has been found to be located in the supporting cells and in Bowman's glands; and has never been demonstrated in receptors. It is thought that the pigment which is a complex of carotenoids and non-carotenoid phospholipids is a metabolic by product which is stored in the basal parts of the supporting cells. There is no definite evidence which might suggest that the pigment has any functional significance.

Electrophysiology of the Olfactory Mucosa

Recordings of the electrical responses of the olfactory mucosa to odour stimuli provide quantitative data regarding its sensitivity and differential responses to various odours. Most electrophysiological studies have been carried out on frogs for technical convenience but studies on other animals have shown that there is no essential difference in response in the different species. Most recordings have been made from an electrode placed on the sur-

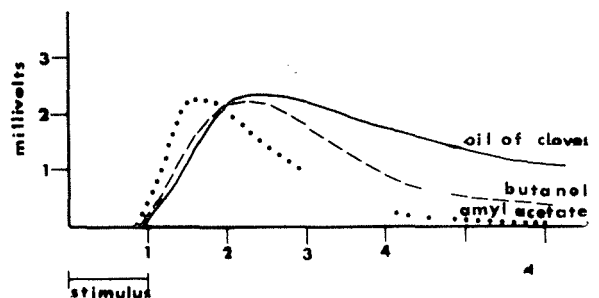


Fig. 4: Electro-olfactogram (E.O.G.) responses to three different odours. The duration of the stimulus was 1 second in each case (after Ottoson).

face of the olfactory mucosa. When a puff of odourised air is directed towards the region being studied a slow, negative, purely monophasic potential develops. This is the electro-olfactogram (E.O.G.). The E.O.G. response to various odours shows differences in the time course of the potential change. Some odours show a faster rising phase than others; and some show a longer falling phase than others (Fig. 4). Compared with corresponding potentials from other sense organs, the E.O.G. response is very slow. A puff of air containing a low concentration of an odour and lasting one second produces a response lasting about 5 seconds or longer. The slow response would not be surprising if the odour molecules have to diffuse through a layer of fluid before reaching the receptors. The time course of the response depends on the number of molecules reaching the receptors per unit time and its duration on the time it takes for odour molecules to be removed or inactivated.

Increasing odour concentration results in a logarithmic increase in amplitude of the E.O.G. potential. Continuous stimulation results in a sustained potential which remains throughout the duration of the stimulus, indicating that the olfactory re-

ceptors adapt extremely slowly. This finding conflicts with the common observation that the sensation of a smell rapidly weakens and soon becomes imperceptible.

The E.O.G. is only obtained by recording from the surface and it declines progressively as the electrode is inserted into the epithelium. It reflects the summated electrical activities at the olfactory surface but it is still uncertain how it is related to the 'generator potential' elicited in the receptors. It could also be a composite potential caused not only by potentials arising in the receptors but also by potentials arising in other cells.

Recordings from single receptors was made possible by the use of special glass microelectrodes, which were pushed into the mucosa until the spike activity from the receptor axons could be recorded (Gestland, 1965).

The spikes arising from different axons could be distinguished from one another by their amplitude — the spikes arising from one axon are constant in amplitude. The response of one receptor could therefore be distinguished from that of other receptors (Fig 5). Using this method of single unit recording three important facts emerged: (1) a single receptor can be excited, inhibited or remain unaffected by an odour; (2) one unit responds differently to different odours; and (3) no two receptors respond in the same way to a variety of odours. The responses could also be measured quantitatively. These results, which have been considerably amplified by further research, have provided a basis for understanding the differential sensitivity of olfactory receptors to odours, which is of considerable importance in olfactory discrimination.

Theories of Olfaction

Perhaps the greatest paradox in olfactory research is that we still do not know the nature of the essential stimulus that constitutes a smell. The original idea that the chemical configuration of an odoriferous molecule determined its smell was soon discarded since no correlation between the two could be found. Numerous



Fig. 5: Spike activity of olfactory axons. Spikes of three different amplitudes arising from 3 separate receptors can be distinguished (a) resting state; (b) response to stimulation with tetraethyl tin (after Gestland).

alternative theories were therefore advanced attempting to explain which physical or chemical characteristics of molecules determined their particular odour, how these characteristics could account for the specificity of odour sensations, and in what way the stimulating molecules altered the receptor cell so as to generate an impulse.

The molecular characteristics which have been implicated in formulating these theories have included such features as the polarity of molecules, their over-all shape or profile, the chemical nature of their end groups and intra-molecular vibrations, or various combinations of these. Some theories have even suggested that odorant substances emit waves which stimulate the receptors.

It is not intended to review the numerous and varied theories which have been proposed but merely to point out that they testify to the lack of adequate experimental evidence which has given way to considerable imagination. The theories of olfaction have therefore been based mainly on theoretical consideration. However, theories are important for the researcher in designing critical experiments which may throw light on the fundamental problems of olfaction, but the difficulties encountered in doing this have so far been quite formidable.

REFERENCES

1. ALTNER, H., and MULLER, W. 1968 Elektro-physiologische und Elektronmikroskopische Untersuchungen an der Vielschleimhaut des Jacobson'schen Organs von Eidechsen. *Z. vergl. Physiol.*, 60: 151-155.
2. BANNISTER, L.H. 1965 The fine structure of the olfactory surface of teleostean fishes. *Q. J. microsc. Sci.*, 106, 333-342.
3. BANNISTER, L.H. 1968 Fine structure of the sensory endings in the vomeronasal organ of the slow worm *Anguis fragilis*. *Nature, Lond.*, 217: 275-276.
4. ARADI, A.F., and BOURNE, G.H. 1953 Gustatory and olfactory epithelia. *Int. Rev. Cytol.*, Vol. II, 289-322.
5. BRIEPHOL, W., MESTRES, P., and MELLER 1973 Licht und Elektronenmikroskopische Befunde zur Differenzierung des Riechepithels der Weissen Maus. *Verh. Anat. Gesell.*, 67: 443-449.
6. BRONSHTEIN, A.A. 1965 Histochemistry of the olfactory organ. *Arkh. Anat. Gistol., Embriol.*, 48:106-116.
7. CUSCHIERI A. 1972 The structure and histo-chemistry of the olfactory and vomeronasal organs in the mouse. Ph.D. Thesis, University of London.
8. CUSCHIERI A. and BANNISTER L.H. 1974a Some histochemical observations on the mucosubstances of the nasal glands of the mouse. *Histochem. J.*, 6:543-558.
9. CUSCHIERI A. and BANNISTER L.H. 1974b. Enzyme histochemistry of the olfactory mucosa and vomeronasal organ in the mouse. *J. Anat.*, 118:447-487.
10. CUSCHIERI A. and BANNISTER L.H. 1975 The development of the olfactory mucosa in the mouse: electron microscopy. *J. Anat.*, 119: 471-498.
11. DE LORENZO A., A.J. 1957 Electron microscopic observations of the olfactory mucosa and olfactory nerve. *J. biophys. biochem. Cytol.*, 3:839-850.
12. DE LORENZO A.J. 1970 The olfactory neuron and the blood-brain barrier. In *Taste and smell in vertebrates*. G.E.W. Wolstenholme and J. Knight (Ed.). Churchill, London, p.151-173.
- 12a DODSON, H.C., BANNISTER L.H. and CUSCHIERI A. 1976 The secretions of the nasal cavity in mice. *Proc. Royal Micro. Soc.*, 6.
13. FRISCH D. 1967 Ultrastructure of mouse olfactory mucosa. *Am. J. Anat.*, 121:87-120.
14. GESTLAND R.C., LETTIN J.Y. and PITTS W.H. 1965 Chemical transmission in the nose of the frog. *J. Physical, Lon.* 181:525-559.
15. GRAZIADEI P.P.C. 1971 The olfactory mucosa of vertebrates. In *Handbook of sensory physiology, Vol. IV pt. 1*. Ed. L.H. Beidler, Springer, Berlin.
16. HORSLEY C.H. 1934. Investigations of the actions of St. John's wort. *J. Pharmacol. Epth. Therap.*, 50:310-322.
17. LANGLEY O.K. and LANDON D.N. 1967 A light and electron histochemical approach to the node of Ranvier and myelin of peripheral nerve fibres. *J. Histochem. Cytochem.* 15:722-731.
18. MOULTON D.G. 1960 Studies in olfactory acuity. 5. Comparative olfactory sensitivity of pigmented and albino rats. *Animal Behaviour*, 8:129-133.
19. MOULTON D.G. 1962 Pigment and the olfactory mechanism. *Nature*, 195:1312-1313.
20. MOULTON D.G., CELEBRI, G., and FINK R.P. 1970 Olfaction in mammals — two aspects: proliferation of cells in the olfactory epithelium and sensitivity to odours. In *Taste and smell in vertebrates*. Ed. G.E.W. Wolstenholme and J. Knight p. 227-245
21. REESE T.S. 1965 Olfactory cilia in the frog. *J. Cell. Biol.*, 25:209-230.
22. SAJGO K. and EGAMI F. 1970 *J. Neurochem.*, 17:633-647.
23. SCHULTZE M. 1856 *Über die Endigungsweise des Geruchsnerven und der Epithelialgebilde der Nasenschleimhaut*. Monatsber. ent. Akad. Wiss. Berlin., 21:504-515.
24. SHANTHA T.R. and NAKAJIMA 1970 Histological and histochemical studies on the rhesus monkey (*Macaca mulatta*) olfactory mucosa. *Z. Zellforsch, mikv. Anat.*, 103:291-399.
25. SHAPIRO B.L. 1970 Enzyme histochemistry of the embryonic nasal mucosa. *Anat. Rec.*, 166: 87-98.
26. SLEIGH M.A. 1962 *The biology of cilia and flagelle*. Macmillan, New York.