J. Steroid Biochem. Molec. Biol. Vol. 39, No. 4B, pp. 601–614, 1991
Printed in Great Britain.

0960-0760/91 $3.00 + 0.00

SENSORY PROCESSING IN THE MAIN AND ACCESSORY OLFACTORY SYSTEMS: COMPARISONS AND CONTRASTS

MICHAEL MEREDITH
Department of Biological Science, B-157, Florida State University, Tallahassee, FL 32306-3050, U.S.A.

Summary—The vomeronasal organ (VNO) and accessory olfactory system (AOS) are present in most terrestrial vertebrates except birds and higher primates. The receptor neurons of the AOS are sequestered inside the VNO, away from the main airflow to the main olfactory receptor neurons. Mechanisms of stimulus access to the sensory neurons vary across species but in most cases there is a system for delivering stimuli faster than would be possible by diffusion. Vomeronasal (VN) receptor neurons typically lack cilia, the site of most of the transduction apparatus in the main olfactory receptors. The VN receptor neurons have a restricted but privileged pathway to the areas of the brain concerned with reproduction and social behavior. In contrast, the main olfactory neurons have a broad pathway to wide areas of the brain, including the neocortex. Experiments where the VNOs or other parts of the accessory olfactory pathway were ablated indicate that the system is important in many behavioral and physiological responses to pheromones (chemical signals carrying information about gender or reproductive or dominance status), some of which may be proteins. VN sensory neurons respond to both volatile and non-volatile stimuli. There is no evidence in the vertebrate AOS for the extreme sensitivity or selectivity characteristic of insect pheromone detectors, but this has not been adequately tested. There is some evidence for learning, possibly by synaptic modification at the second-order neuron level. Social and reproductive cues stimulating the AOS often elicit an intracerebral release of LHRH—which may act at receptors different from those of the pituitary to facilitate behavior. Whether the LHRH release is necessary for AOS-mediated behavioral response is not yet clear.

FUNCTIONS OF THE TWO SYSTEMS

The accessory olfactory system (AOS) consists of chemoreceptor neurons in the vomeronasal organ (VNO) and their central neural pathway through the accessory olfactory bulb (AOB), amygdala and basal forebrain. It has been implicated in pheromone detection and chemical communication in a variety of species [1]. The system is not present in humans or other higher old-world primates. However, a residual recess, opening at the base of the nasal septum, is reported to be common in humans [2] but lacking bipolar vomeronasal (VN) sensory neurons [3]. The AOS is an important sensory system for most other terrestrial vertebrates except birds. In snakes, the AOS appears to be the more important chemoreceptor system and is used in feeding behavior (prey trailing and attack), sexual behavior and social behavior [4].

In mammals, the AOS appears to be specialized to detect species-specific chemical signals (pheromones) that carry specific information about gender, reproductive or dominance status [1, 5, 6]. The main olfactory system (MOS) is suggested to have a more generalized function as a "molecular analyzer" for environmental chemicals having no predetermined meaning. In this role, it would have an extensive capacity for making associations between odors and contexts. This ability would make the MOS valuable for triggering appropriate responses in a variety of situations where characteristic odors may be present. The MOS certainly is involved in some circumstances where chemical cues are learned. Many animals appear to learn tasks that are cued by odors better than tasks that are cued by visual signals. In such tasks, for example, a rat shows similar sophistication in learning to that of a monkey performing on visual tasks [7].

Although useful heuristically, the proposed segregation of function between the main system and the AOS is not absolute. Indeed, various pieces of evidence show that the MOS

Proceedings of the International Symposium on Recent Advances in Mammalian Pheromone Research, Paris, France, 6–9 October 1991. Sponsored by the EROX Corporation.

is also involved in chemical communication. For example, the MOS is responsible for individual recognition in animals in some circumstances [8], and it is also involved in the detection of maternal pheromones by newborn rabbits [9]. In addition, some behavioral and hormonal responses that depend on an intact VN system in naive animals, may be elicited by main olfactory cues in experienced animals, once they have learned to use olfactory input to identify a behavioral situation previously indicated by VN input [10, 11]. Learning of "odor signatures" is also not the exclusive province of the MOS. In the female mouse, pregnancy may be aborted on exposure to strange male odors (Bruce effect), but the stud male's odor is learned after mating, apparently via the AOS [12, 13] (see below).

Is the nervus terminalis (NT) involved?

Most evidence implicating the AOS in specific behavioral or hormonal responses comes from experiments where the VNOs were removed or the VN nerves were cut. Both treatments also damage the NT, a cranial nerve of unknown function that connects the nose to the ventral forebrain [14]. This nerve has been indirectly implicated in chemical communication in fish [15] and in hamsters [16] so some deficits attributed to VNO malfunction after lesions may in fact be due to NT damage. Experiments which specifically exclude NT involvement (e.g. Bruce effect) [17] demonstrate conclusively that NT could not be responsible for ALL the functions attributed to the AOS. The NT in rodents and other species contains LHRHir cells and could contribute to hormonal responses to chemosignals. It is associated with the VN nerve and may have endings within the VNO [18, 19]. The human NT ends in the nasal cavity [20] and may innervate the recess that is all that remains of the human VNO.

STIMULI

Non-volatile stimuli may be involved but not all VN stimuli must be non-volatile

In most behavioral and hormonal responses where the VNO/AOS is implicated, the chemical stimuli involved are unknown. Many pheromonal interactions in rodents and ungulates involve chemicals present in urine, special skin-gland secretions, or in vaginal fluid. In several cases non-volatile components of a crude stimulus are found to be capable of producing a nearly complete response [1, 5]. For example, the active component for male hamster copulatory behavior appears to be a protein which has been named aphrodisin [21].

Despite strong evidence that some VN stimuli are non-volatile, there is no evidence that all VN stimuli must be non-volatile. The VN system is sensitive to volatiles in experimental situations and the mechanisms of stimulus access, described below, could deliver volatiles to the sensory epithelium, especially if in solution in mucus. Furthermore, the MOS is not precluded from responding to non-volatiles [22].

STIMULUS ACCESS

Stimuli may be delivered by tongue or nose movements in some species

In most species, including all the mammalian species with the organ, the VN sensory epithelium on each side is sequestered in a separate chamber connected to the outside world only by a narrow duct. Effective stimulus access is therefore an important question. In snakes, the paired ducts open into the mouth and stimuli are delivered to the duct entrances via the tips of the forked tongue. In many mammalian species, the organ opens into the nasopalatine canal, connecting nose and mouth and can thus receive stimuli either from the nasal cavity or from the mouth. There is indirect evidence in several species that the tongue may be used to introduce materials into the nasopalatine canals [23–26]. A facial grimace called "Flehmen" appears to mobilize the tissue around the ducts and to assist in delivering stimuli to the VN sensory epithelium [27]. In the mouse lemur, which communicates via chemosignals in urine, Schilling *et al.* [26] could stimulate both AOS and MOS with liquid urine via the nasopalatine canal, but only the MOS with urine vapor (possibly at a lower concentration).

VN stimuli should probably be aqueous-soluble or bind to aqueous-soluble carriers

In rodents, the VN ducts open onto the floor of the nasal cavity just inside the nostrils (Fig. 1), far anterior to the nasal end of the nasopalatine canal, and stimuli probably enter via the nostrils. If the stimuli are non-volatile, they are presumably carried to the sensory epithelium in solution in the ventral mucus

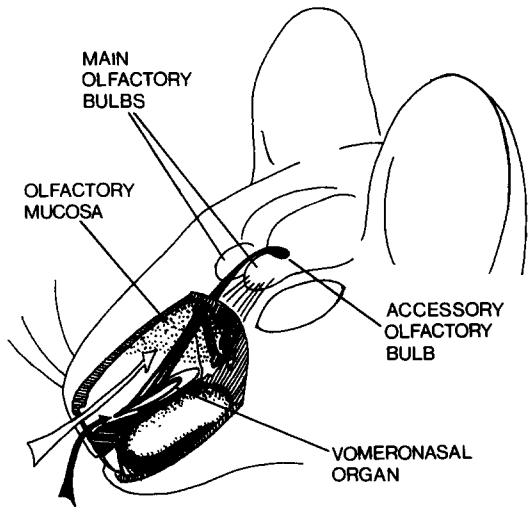

Fig. 1. The VNO opens antero-ventrally into the ventral groove of the nasal cavity where aqueous-soluble, (and sometimes non-volatile) stimuli (solid arrow) pass by in the mucus stream. The olfactory sensory mucosa is more postero-dorsal (stippled area) and receives volatile odors (open arrow). VNO has a separate centrally projecting neural pathway via the AOB to brain areas concerned with social and reproductive functions.

stream. This stream arises from glands opening into the nasal vestibule [28] at the anterior end of the nose and continually passes back along the floor of the nasal cavity, past the opening of the VN duct [22], to the nasopharynx where it is swallowed. Thus, a prime requirement for a VN stimulus may be that it be mucus-(i.e. aqueous)-soluble. One component of mucus of the rat is olfactory binding protein (OBP) produced by the lateral nasal gland [29]. OBP binds odors including some with little aqueous-solubility. It has been proposed as a carrier of odors to the main olfactory epithelium but appears to be uniquely suited to be a carrier of stimuli to the VNO [22].

VN pump delivers stimuli from the nasal cavity to the sensory neurons—is activated by novel situations

In the hamster there is a special mechanism for delivering aqueous-soluble stimuli into the lumen of the VNO. The organ is enclosed in a capsule which also contains large blood vessels (Fig. 2). Stimulation of the nasopalatine nerve, which carries autonomic fibers into the posterior end of the organ, results in vasoconstriction of the vascular tissue around the organ and a dramatic inflow of mucus into the duct [30]. Most mammalian species have large blood vessels adjacent to the VN lumen, so this mechanism may be common [25–27, 31, 32]. In the hamster, the pump was shown to be behaviorally important because lesions of the nasopalatine nerve result in deficits in mating behavior [33].

We originally thought that the sequestration of the VNO might be to prevent inadvertent stimulation from eliciting inappropriate behavior [5]. However, recordings of pump operation in behaving male hamsters, using electrodes implanted in the VN capsule, did not support that suggestion. The pump appeared to operate in any novel situation—with an estrous female, but also with an anestrous female or a male, or on simply opening the cage lid [34]. Thus, although not stimulated continuously, the organ is not stimulated very selectively. Possibly VN receptors are sequestered because they require a special mucus environment. The details of the electrophysiological response suggested an oscillation in vasomotor activity with a period of 1–2 s that could summate in arousing situations, indicating an overall increase in vasomotor tone. The electrophysiological signals were verified as arising from pump operation by comparing the activity in awake behaving animals with that recorded through the same electrodes in anesthetized animals, while activating the pump by nasopalatine nerve stimulation (Fig. 3).

RECEPTOR CELLS

VN receptor neurons lack cilia—but is the transduction mechanism different from that of the main olfactory receptor neurons?

VN receptor cells are bipolar neurons derived from the olfactory placode [6] like main olfactory receptor cells, but with microvilli

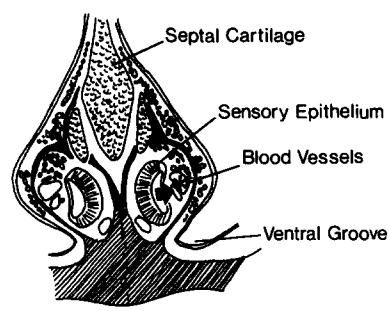

Fig. 2. Cross section of the base of the nasal septum in the hamster nose showing components of the VN pump. Sympathetic system activity constricts blood vessels (asterisk) within the VN capsule (hatched). The lumen of the VNO expands (solid arrow) and odor laden mucus is drawn into the VNO from the ventral groove of the nasal cavity (through a duct anterior to the section shown).

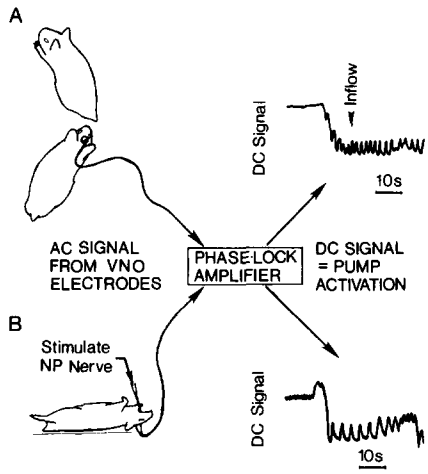

Fig. 3. (A) Activation of the VN pump in awake behaving animals was recorded by placing electrodes in VNO and passing a low voltage AC signal between them. The constriction and dilation of the blood vessels constituting the pump alters the size of the "in phase" signal (detected with a phase-lock amplifier). Downward movement (indicating stimulus inflow) of the DC signal trace from the phase-lock amplifier occurs when a female (or other novel stimulus) is introduced into the cage of a resting male (top right). Oscillations appear to be due to repetitive bursts of impulses in sympathetic vasomotor nerve. They can be reproduced in the anesthetized animal (B) by repetitive electrical stimulation of the nasopalatine (NP) nerve (bottom right). Initial upward movement may be due to inevitable stimulation of parasympathetic fibers in the nerve.

rather than cilia on their apical membrane [35, 36]. A few cilia have been reported in the dog [37], and in the hamster the microvilli also contain microtubules, but without the characteristic ciliary arrangement [38].

Main olfactory receptor neurons have cilia on their apical surface that appear to contain most of the transduction apparatus [39, 40]. Odors induce increases in intracellular cAMP or IP_3 via G-protein linked receptor molecules [41, 42]. Depolarization via cAMP-activated non-specific cation channels [43] and/or IP_3-activated Ca^{2+} channels [44] generates action potentials. So far, there is no reason to suppose that the transduction process in the VNO would be different: just located on microvillar rather than ciliary membranes.

Recordings from individual main olfactory receptor cells show them to be relatively non-specific. Each appears to be sensitive to a different, often overlapping range of individual chemicals [45], and to be less specific as intensity (concentration) is increased. Thus, the neurons with the lowest threshold are activated first and the less sensitive ones for that odor (which may be more sensitive for another) are recruited at higher intensity. Activity (firing rate) increases steeply with intensity, reaching a maximum (saturation?) at about 1–2 log units above that cell's threshold. It is not yet clear whether this means that high intensity information is carried only by the less sensitive neurons, as may be the case in other systems [46].

The distribution of sensitivity across different individual neurons has been interpreted as evidence for multiple molecular receptors, distributed in different proportions to different neurons. Recent molecular biological work [47] (see elsewhere in this issue) suggests multiple receptor proteins (of β-adrenoceptor type). The results from single receptor neuron recordings could be explained if many of these genes are expressed in each receptor neuron but with various densities of product. One advantage of that arrangement, over one where one receptor molecule type is expressed in each cell, could be the ability to code a wide range of intensities—despite the saturation of the most sensitive cells that have many activated receptors.

In the AOS, despite the lack of cilia, the organization may be similar. However, if VN stimuli can be proteins as suggested, some novel interactions between receptor and ligand may be possible. It is not clear how many different VN stimuli might have to be discriminated. There are probably more than just one male and one female pheromone [48] even though one pheromone might elicit many context-specific responses. If the system is set up to detect only a few pheromones it may not need the large number of receptors proposed for the MOS. On the other hand, discriminating even a few closely related molecules might require the simultaneous analysis by many receptors of different specificity—as in the laboratory where several chemical tests are needed to identify a chemical. No successful recordings have yet been made from individual VN receptor neurons so their specificity is unknown. In the tortoise (where the VN epithelium is exposed to the nasal airflow) multi-unit nerve-bundle recordings show a similar responsiveness in the VN and main olfactory nerves to a range of chemicals [36]. The VN nerves appeared somewhat more sensitive to the smaller, more water-soluble members of homologous series of acetates or alcohols—but possibly only because these dissolve easily in the mucus overlying the VN epithelium [5]. There is no evidence as yet for extreme specificity as in insect pheromone receptors.

CENTRAL CONNECTIONS: STRUCTURAL AND FUNCTIONAL

Main olfactory bulb (MOB): wide topographic input; chemotopic odor map

The sheet of receptor neurons making up the main olfactory epithelium is distributed over the surface of a complex array of turbinates in most species (although relatively simple in humans). Odors may distribute unevenly across it, but the epithelium also varies in sensitivity from place to place [49]. The majority of receptor neuron axons appear to project to the olfactory bulb in a loose topographic pattern but some proportion of axons from each epithelial region diverge widely to a more extensive area of the bulb [50, 51]. Evidence from 2-deoxyglucose (2DG) metabolic mapping suggests a "chemotopic" organization across the bulbar surface with different areas activated by different odors [52–54]. Of course, the divergence from a strict topographic projection could actually increase the chemotopic specificity of a small region of the bulb, if receptor cell axons with particular specificity converged onto that region. Although the most active areas on the map do not appear to be essential for odor discrimination [55], there is evidence for behavioral significance of spatial activity in the bulb [56].

Accessory olfactory bulb (AOB): restricted input; no topography (or chemotopy?)

In the AOS, the VN receptor neurons are arranged in a relatively narrow strip along one side of the VN lumen. The opposite non-sensory epithelium may contain trigeminal mechanosensory endings [37] and possibly NT endings. The axons of VN receptor neurons gather into bundles, pass under the septal epithelium and between the MOBs to the AOBs. There is a clear topographic relation in the hamster between rostro-caudal regions of the epithelial sheet and the three main nerve bundles, with the dorsal, middle and ventral nerves containing mainly axons from the anterior, middle and posterior parts (Fig. 4). However, when the axons arrive at the AOB, there is no obvious topography between the nerve bundles and the AOB and thus no topographic projection from the epithelium to the AOB [57]. VN receptor cell axons reaching the medial and the lateral side of the AOB do differ in their binding of certain monoclonal antibodies, but these axons are scattered in the VN nerves and appear to come

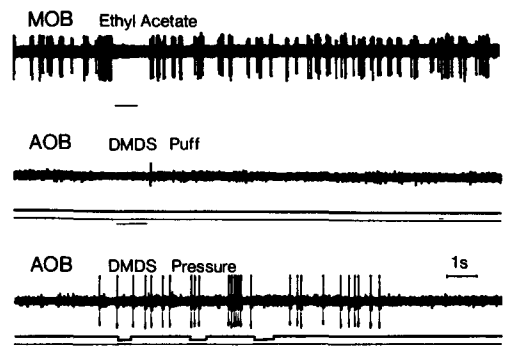

Fig. 4. *Top.* MOB output neuron (probably mitral cell) responds briskly with a burst of spikes then a pause to ethyl acetate puffed into a constant airflow through the nose (airflow 115 ml/min, barbiturate anesthesia). This response was reproducible with repetition of the stimulus. *Middle.* AOB second-order neuron (probably mitral cell) does not respond to a puff of DMDS vapor directed at the vomeronasal duct entrance (nasal cavity opened). *Bottom.* The same AOB cell fires 27 spikes in 10 s when the sidewall of the VNO capsule is pressed in and released—sucking odor-laden mucus into the lumen. This neuron fired significantly fewer spikes (7) when pressure was applied without a preceding odor puff. This response was reproducible.

from non-topographically arranged cells scattered throughout the VN epithelium [58]. No function has been proposed for the antigenic differences. The lack of topography does not mean that there is no *Chemo*topic organization of the AOB, since dispersed neurons with similar functions might still converge on one spot in the AOB, but it does not provide any support for the notion either.

MOB activity is characteristically complex: patterns may code odor information

Output neurons in the MOB are of several types. The most superficial cells, with the least extensive lateral inhibitory connections, are the most excitable to electrical stimulation of the olfactory nerve. The deepest cells (mitral cells) have the most extensive lateral inhibitory connections and are the least excitable to electrical stimulation [59]. They appear to have the same hierarchy of excitability to odor stimulation, but their responses to odors are much more complicated, often involving temporal patterns of excitation and suppression within the response to a single odor pulse. The patterns vary with type of odor and intensity so that these cells also show non-monotonic intensity response functions [60]. A computer simulation of the known anatomy and connectivity of olfactory bulbar circuits [61, 62] suggests an explanation for this strange behavior: (1) the saturation of the most sensitive receptor neurons and the recruitment of less sensitive ones as intensity increases, leads

to a spreading of activity at the bulb; (2) the strong lateral inhibitory circuits create waves of inhibition in front of, and perhaps behind, the spreading excitation. How this process could help to code odor quality and intensity is not clear as yet, but it does provide one coherent explanation for several aspects of bulbar odor response. Odor coding is difficult to understand anyway since there is no clear topographic projection from the bulb to higher centers that could preserve the chemotopic spatial pattern of activity in the bulb. One possibility is that local spatial patterns are converted to temporal patterns during the rising phase of an odor stimulus, as a result of the changing spatial patterns described above.

AOB appears similar to MOB. Lateral inhibitory circuits suggest non-specific activation but specificity has not been measured

If there is no topography or chemotopy in the AOB, this arrangement would contrast with the MOB where a chemotopic organization may underlie odor coding. Spatial analysis of VN stimuli (chemotopy) might NOT be expected if the organ were analyzing relatively few stimuli with relatively specific receptors. The unique activation of particular cells by single stimuli (as in an ideal labeled line system) would require no comparison between parallel inputs (except in the case of mixtures) and thus, no spatial analysis in the AOB. However, the organization of the AOB seems to be set up for spatial analysis because, as in the MOB, mitral cells

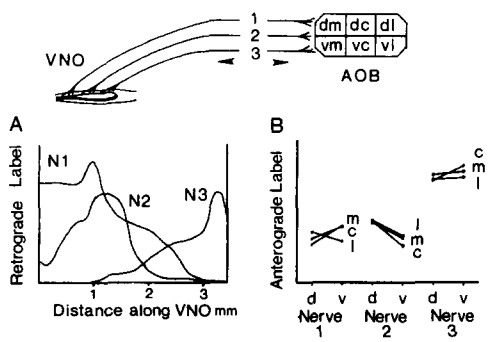

Fig. 5. (A) HRP-label injected in nerve 1, 2 or 3 travels back to VNO (retrograde label density); nerve 1 (N1) receives axons from the anterior VNO, nerve 2 (N2) from the central VNO and nerve 3 (N3) from the posterior VNO. (b) HRP-label also travels to AOB (anterograde label density). Nerve 3 is larger and carries more label but the label is distributed evenly to all AOB segments from each nerve with no significant gradients either dorso-ventrally or medio-laterally—i.e. there is no topographic projection from VNO to AOB. d = dorsal; v = ventral; m = medial; c = central; l = lateral (see diagram of AOB segments at top right).

interact with lateral inhibitory interneurons. The circuits underlying lateral and feedback inhibition in the AOB appear to function similarly to those of the MOB in physiological experiments using electrical stimulation of the AOB input and output [63, 64].

In mammals, no AOS response has been observed when odors were simply blown over the entrance of the VN duct but without activation of the VN pump [26, 30, 65] (Fig. 5). Responses to odors were recorded from single second-order neurons in the hamster AOB, to amyl acetate vapor delivered directly to the surgically exposed sensory epithelium [66] and to dimethyl disulfide (DMDS) and other odors drawn into the lumen of the organ [30, 67]. DMDS is thought to be a hamster attractant-pheromone [68] but its behavioral effect is dependent on the main system not the AOS [66]. The other odors were arbitrarily selected organic esters and alcohols widely used in olfactory research but not suspected of being biologically significant chemosignals. In both cases, then, the responses appear to be relatively *non*-specific. Firing rates of individual AOB mitral cells in the hamster were slow and responses to odor stimuli drawn into the VNO were relatively small and sluggish compared to MOB responses (although rapid compared to the expected rate of stimulus delivery by diffusion). A relatively slow response under these circumstances might reflect the 1–3 s latency to peak pump operation [30]. Both excitatory and suppressive responses could be recorded in the hamster AOB. Hatanaka and Shibuya [69] also recorded both excitatory and suppressive responses, in the turtle AOB, and noted temporal patterns of response remarkably similar to those of the MOB recorded in the hamster [60]. However, very high stimulus concentrations were used, and stimuli included some non-odorous ionic solutions, so it is not clear if these results reflect a similarlity between main system and AOS processing at more normal intensities. Attempts to stimulate the VNO with solutions of hamster vaginal fluid—known to contain a VN stimulant [70]—were unsuccessful [M. Meredith, unpublished], possibly because the continuous saline perfusion washed the VN gland secretions away from the sensory epithelium. Thus, in mammals, no responses have been recorded in the AOS to known pure VN stimuli, so conclusions that the AOS is non-specific may be premature. In snakes, AOB responses have been recorded to a

glycoprotein purified from prey extracts and known to activate behavior through the VNO [71]. The specificity of AOB neurons for this substance was not reported.

Some insights into MOB organization have come from mapping activity with 2-deoxyglucose (2DG). However, all attempts to demonstrate AOB function in the hamster via 2DG uptake were unsuccessful. No uptake was seen in awake or anesthetized animals, in response to natural or artificial odor stimulation, including a mating test where the AOS should be involved. No 2DG uptake occurred in the AOB even when its entire input was stimulated electrically, although field potentials recorded in the amygdala confirmed that the AOB had been activated [M. Meredith, J. Kauer, R. O'Connell and G. Shepherd (1980), unpublished]. Other 2DG studies, although not directed at demonstrating AOB uptake, also show blank spots for 2DG uptake in the AOB of the rat [54] and tree shrew [72]. Nevertheless, recent 2DG studies show uptake in the AOB of the lemur, in response to urine stimulation [26] so our conclusion that the system was not suitable for the 2DG method may only apply to some species.

AOB output is restricted mainly to the amygdala. MOB output is widespread but the two systems may converge onto individual neurons in the VN amygdala

The MOS and AOS in mammals are separate, at least up to the tertiary neuron level in the amygdala [73, 74]. The VN nerves project only to the AOB, the olfactory nerves project only to the MOB and there is no functional cross connection between the two even though MOB output axons pass through the granule layer of the AOB [75]. Both systems project to the amygdala but their endings are clearly segregated to different nuclei. The AOB mitral cells send axons to the medial nucleus (MN) and posteromedial cortical nucleus (PMCN) of the amygdala (the VN amygdala [76])—and in some species there is a small projection directly to the bed nucleus of the stria terminals (BNST) [73, 74]. There is no connection from any component of the AOS to the neocortex either directly or via the thalamus. More highly convoluted pathways can be imagined but there is no reason to suspect that VN sensory input would be handled by the nervous system at a cognitive—or conscious—level [5], *cf* Ref. [77]. What the AOS does have is a relatively direct connection to the limbic areas of the brain concerned with social and reproductive functions.

In contrast to this restricted projection, the main olfactory output cells send axons widely to the accessory olfactory nucleus (AON), the olfactory tubercle (OT), the amygdala and to the pyriform and entorhinal cortices. From these regions, main olfactory information passes to many parts of the brain including the neocortex both via the mediodorsal thalamus and directly [78]. The amygdala projections from the MOB end in the anterior and posterolateral cortical nuclei (ACN and PLCN; the olfactory amygdala). Olfactory and VN cortical amygdalae are adjacent and there are short connections between them. There is some evidence that connections may be important and that they do occur at this level. In behavioral experiments, the removal of the VNO in sexually inexperienced hamsters produces severe deficits in mating behavior, but removal of the organs in experienced animals has virtually no effect [10]. The conclusion from these experiments was that the animals could learn to use main olfactory cues from females, once the olfactory input had been associated with VN input *during* experience and before VNOs were removed. The amygdala may be where convergence of the two systems occurs. In separate experiments, single neurons were identified in PMCN that could be driven by electrical stimulation of both the accessory pathway (electrodes in the VNO) and of the main pathway (electrodes in the antero–ventral lateral olfactory bulb). These two electrode positions were chosen to avoid cross stimulation between systems and resulted in different response latencies that were characteristic of the system stimulated [79].

AOB learning proposed in a specific chemosensory response—the Bruce effect

Indirect evidence for function in the AOB has come from a series of ingenious experiments from Keverne's laboratory [13]. Pregnancy is blocked in female mice if they are exposed to "strange" males (i.e. of a strain differing from the impregnating male), and this block is prevented if the female's VNOs are ablated [77, 80]. The stud male does not block his own mate's pregnancy because she forms a memory of his odor during the 2–3 h after mating (if he is allowed to remain). Keverne's group implicated the AOB as the site of memory because depletion of NE in the posterior bulb or local

anesthetization there prevented memory formation, whereas anesthetization of the next relay, the amygdala, did not [13, 81]. They suggest a selective enhancement of inhibitory feedback synapses from activated granule cells under the influence of NE [82]. Subsequent activation by stud male odor would then be selectively attenuated. This mechanism ignores the powerful lateral inhibition present in the AOB. However, in a simple computer model of olfactory bulb circuits that includes the lateral inhibition [62], selective attenuation can occur when odors are represented spatially. Responses to an "odor" are eliminated when inhibitory synapses are strengthened according to the spatial response pattern for a previous response to that odor. Other overlapping patterns are altered but not eliminated [62] (Fig. 6). A much more elaborate model, based on Freeman's model for the main bulb [83] suggests that modifying the granule-mitral synapses would raise the frequency of oscillations in the circuit—possibly preventing activation of the more central neural or hormonal responses that cause pregnancy block. The oscillatory model, while mathematically more sophisticated, appears to have no more external validation than the simple attenuation model, however.

In the MOB, Freeman's elaborate model [84] also suggests that "learning" may occur by synaptic modification. An alternative mechanism for "learning" in the main bulb, implicates a modification of structure. Rat pups can learn new odors when these are paired with mechanical

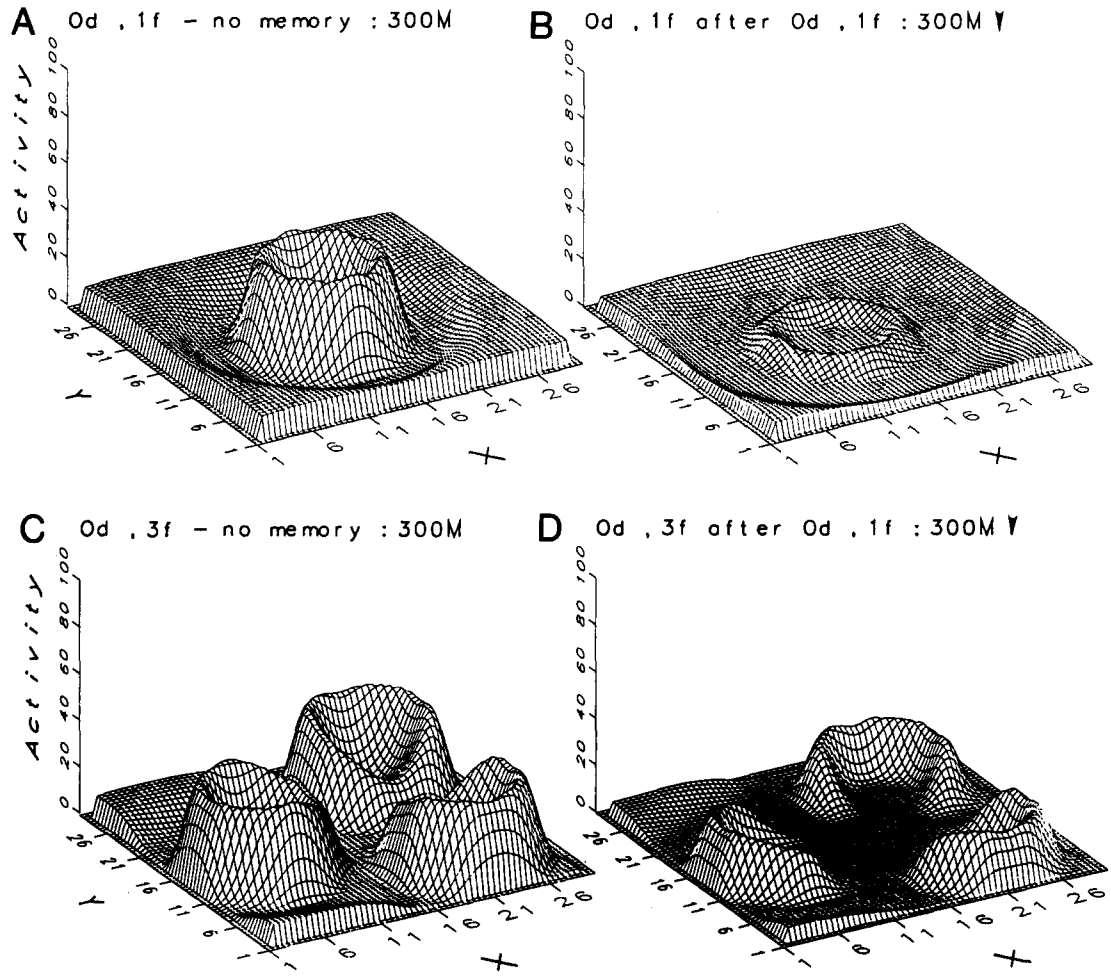

Fig. 6. Computer simulation of a 30 × 30 array (X,Y axes) of output neurons activated (Z axis) by an odor. Neurons near the center are excited. There is a central depression [62] and a laterally inhibited surround. (A) This spatial pattern is (in the simulation) the pattern to be learned and ignored (see the text). Learning consists of strengthening inhibitory interneuron synapses according to the degree of excitation produced by the original odor. (B) After learning, this odor produces minimal response and a wide area of surround inhibition. (C) A second odor produces a different (3-focus) pattern of activity in the same array of neurons. (D) The second response is preserved (although altered) after learning the first pattern.

stroking (possibly mimicking maternal grooming) [85]. Associated with the learning is an increase in metabolic activity [86] and a hypertrophy of the region of the bulb associated with the maximum activation by the odor (as indicated by 2DG uptake) [87]. No similar mechanism has yet been suggested for the accessory bulb. All three suggested mechanisms for "learning" implicate NE as a facilitator.

HORMONAL CONSEQUENCES OF CHEMOSENSORY INPUT

VN input may affect reproductive events by altering LHRH release

The release of LH in response to social stimulation, such as encounters between conspecific animals of opposite sex, has long been recognized [5, 88] and presumably implies a preceding release of LHRH. This response to social stimulation was later shown to depend in part on the VNO. Coquelin *et al.* [89] found an increase in serum LH within 5 min in male mice exposed to females or to female urine. The response to urine disappeared in animals with VNOs removed. Wysocki *et al.* [90] showed that an increase of testosterone (presumably resulting from increased gonadotropin secretion), in males exposed to anesthetized females disappeared in mice lacking VNOs.

The VN system thus appears to be involved in the release of LHRH in the brain. The VN system is also important for initiating mating behavior in male hamsters, especially in inexperienced animals [10] and in mice [11]. At the same time, LHRH is important in facilitating mating behavior, especially in animals that are otherwise in a suboptimal state for mating—as, for example, males with low testosterone levels or impaired genital sensory input [91, 92] or ovariectomized females with reduced steroid priming doses [93]. There are many LHRH containing neurons and fibers associated with the AOBs (and with the MOBs and the NT), as well as in forebrain areas with connections to chemosensory input. Thus, it is a small step to hypothesize that LHRH release may be an important step in the pathway by which VN sensory input facilitates mating behavior. The fact that many LHRH neurons do not communicate with the median eminence (approx. 50% [94]), suggests that the LHRH release associated with the behavioral facilitation need not act through the pituitary. A facilitation of mating behavior in female rats by extrapituitary action of LHRH has been demonstrated in hypophysectomized, ovariectomized animals [95].

Recent evidence suggests that exogenous LHRH delivered intra-cerebro-ventricularly (icv) can substantially restore mating behavior in male hamsters with VNOs removed [96] (Fig. 7), supporting the hypothesis that the VN acts through LHRH release. These data do not prove the hypothesis because VN sensory input and LHRH could independently facilitate behavior. The AOS and LHRH release are also associated in female rats [97, 98], hamsters [99],

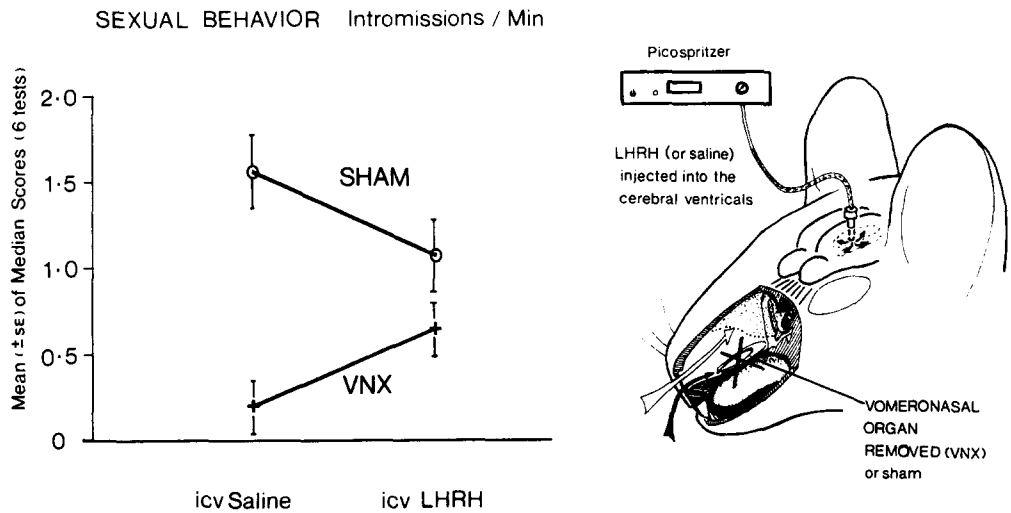

Fig. 7. LHRH injected into the ventricles (icv) partially restores mating behavior lost after VNO removal. Male hamsters without vomeronasal organs (VNX) continue to show severe deficits in mating behavior when saline is injected icv—compared to sham operated animals (SHAM). When LHRH (50 ng in 2 μl) is injected icv, performance of VNX animals is significantly improved and no longer significantly different from SHAM animals (but the performance of SHAM animals declines).

voles and mice [100, 101]. Rajendren *et al.* [97] demonstrated a reduction in mating behavior and a decrease in LH release in female rats with VNOs removed (suggesting a reduction in LHRH release). Moss and Dudley [93, 102] had previously shown by an elegant series of experiments that LHRH and various analogs facilitate mating behavior in estrogen treated ovariectomized female rats. In these experiments mating can be facilitated by analogs that are ineffective for LH release, such as the pentapeptide fragment Ac^{5-10}, or even by antagonists. Moss and Dudley [93] conclude that mating behavior is facilitated by LHRH action at a receptor different from those controlling pituitary LH release. Preliminary experiments [103] suggest that Ac^{5-10} and the LH-release antagonist Nal-Glu can also significantly facilitate mating behavior in male hamsters, extending the findings to a different species and opposite sex.

LHRH released by AOB input may act through MPOA. Prolactin may also be involved

Dudley and Moss [104] determined that the lowest dose of Ac^{5-10} effective for female mating behavior was found when the peptide was injected directly into the medial preoptic area (MPOA), suggesting that the MPOA may be the site of LHRH action. The MPOA is sexually dimorphic and known to be important for male mating behavior. Despite the suggestion that LHRH may act through the MPOA, receptor binding studies using recognized LHRH agonists [105, 106] actually demonstrate few LHRH receptors in the MPOA fewer, for example, than in the hippocampal formation. However, the agonists used may not bind to the hypothetical receptor involved in mating behavior enhancement. This hypothetical receptor might bind the C-terminal end of the peptide, given the effectiveness of the C-terminal fragment Ac^{5-10}, rather than the N-terminal end which appears to be important for LH release [93]. Jennes *et al.* [107] recently reported that there were some differences between LHRH receptor binding in pituitary membranes and hippocampal membranes but Ac^{5-10} did not displace the standard LHRH agonist buserelin from either. There was also no detectable binding of labeled Ac^{5-10} to pituitary or to hippocampal membranes. MPOA membranes were not tested. It is not certain, of course, that the identified mammalian LHRH molecule or a fragment of it is the molecule that normally facilitates mating behavior. Other LHRH-related peptides may be available [108, 109].

Regardless of the nature of the receptors, LHRH can certainly modulate the activity of neurons in MPOA tissue slices in the rat [110]. The MPOA is a region with extensive LHRH input including connections from the diffuse array of LHRH cells and fibers extending back to it from the MOB and AOB—and the NT [111]. It also receives input via the stria terminalis [112] from the medial amygdaloid nucleus, a specific target of the AOB. Thus, VN inputs could cause LHRH release in MPOA, either via specific LHRH projections or via non-LHRH activation of local release.

The preovulatory LH release can be enhanced by AOB or medial amygdala stimulation and inhibited by MOB or lateral (olfactory) amygdala stimulation. Beltramino and Taleisnik [113] conclude that this surge system operates via the ventral premammillary nucleus rather than the MPOA so this evidence does not rule out MOB participation in socially-stimulated LHRH release. Similarly, most of the known steroid and neurotransmitter control systems for LHRH release [111, 114, 115] refer to the surge mechanism. Control of socially-stimulated LHRH release, which could be small and localized, may be different. Steroid receptors or steroid accumulating cells have been localized to several stations on putative accessory olfactory pathways, including the corticomedial amygdala, BNST and the MPOA itself [116–118]—although not involving the LHRH cells themselves [119]. Thus, there may be activational steroid influences on the LHRH/behavioral pathway. There are certainly organizational steroid influences. The AOB, medial amygdala and MPOA are all sexually dimorphic in some species, due to the normal influences of androgen [120–122].

A second suggestion for a hormonally-mediated VN effect is that VN input lowers prolactin (PRL) levels. Keverne's group were able to mimic several effects of VN removal in female mice by injecting bromocryptine, a DA agonist. These effects included: (1) the estrus suppression that occurs in group-housed females [123]; (2) the acceleration of puberty in immature females exposed to male odors [124]; and (3) the blockage of pregnancy in females exposed to chemosignals from strange males (see above). Because DA normally inhibits PRL release from the pituitary, they concluded that the effect of VN input was to lower PRL. The DA agonist

must affect many systems in the brain, however, and also has a suppressive effect on LHRH and LH release. Electrical stimulation in the AOB does drive arcuate nucleus cells that have projections to the median eminence [125], but whether these are dopaminergic is not known. Blockade of GABA action in the AOB enhanced the driving of these cells, consistent with the proposed mechanism for AOB "memory".

REFERENCES

1. Halpern M.: The organization and function of the vomeronasal system. *A. Rev. Neurosci.* **10** (1987) 325–362.
2. Johnson A., Josephson R. and Hawke M.: Clinical and histological evidence for the presence of the vomeronasal (Jacobson's) organ in adult humans. *J. Otolar.* **14** (1985) 71–79.
3. Moran D. T., Jafek B. W. and Rowley J. C.: Ultrastructure of the vomeronasal organ in man: a pilot study. *Chem. Senses* **10** (1985) 420–421.
4. Halpern M. and Kubie J. L.: The role of the ophidian vomeronasal system in species-typical behavior. *TINS* **7** (1984) 1–6.
5. Meredith M.: Sensory physiology of pheromone communication. In *Pheromones and Reproduction in Mammals* (Edited by J. G. Vandenbergh). Academic Press, New York (1983) pp. 199–252.
6. Wysocki C. J.: Similarities and differences between the vomeronasal and olfactory systems. Presented at the International Symposium on Recent Advances in Mammalian Pheromone Research. Oct. (1991).
7. Slotnick B. M.: Olfactory stimulus control in the rat. *Chem. Senses* **9** (1984) 157–165.
8. Johnston R. E. and Rasmussen K.: Individual recognition of female hamsters by males: role of chemical cues and of the olfactory and vomeronasal systems. *Physiol. Behav.* **33** (1984) 95–99.
9. Hudson R. and Distel H.: Pheromonal release of suckling in rabbits does not depend on the vomeronasal organ. *Physiol. Behav.* **37** (1986) 123–128.
10. Meredith M.: Vomeronasal organ removal before sexual experience impairs male hamster mating behavior. *Physiol. Behav.* **36** (1986) 737–743.
11. Wysocki C. J.: Vomeronasal chemoreception—Its role in reproductive fitness and physiology. *Neurol. Neurobiol.* **50** (1989) 545–566.
12. Bellringer J. F., Pratt H. P. M. and Keverne E. B.: Involvement of the vomeronasal organ and prolactin in pheromonal induction of delayed implantation in mice. *J. Reprod. Fert.* **59** (1980) 223–228.
13. Brennan P., Kaba H. and Keverne E. B.: Olfactory recognition: a simple memory system. *Science* **250** (1990) 1223–1226.
14. Schwanzel-Fukuda M. and Pfaff D. N.: Migration of LHRH-immunoreactive neurons from the olfactory placode rationalizes olfacto–hormonal relationships. *J. Steroid Biochem. Molec. Biol.* **39**(4B) (1991) 565–572.
15. Dulka J. G., Stacey N. E., Sorenson P. W., Van Der Kraak G. S. and Marchant T. A.: A sex pheromone system in goldfish: is the nervus terminalis involved. *Ann. N.Y. Acad. Sci.* **519** (1987) 411–420.
16. Wirsig C. R. and Leonard C. M.: Terminal nerve damage impairs the mating behavior of the male hamster. *Brain Res.* **419** (1987) 293–303.
17. Rajendren G. and Dominic C. J.: Effect of bilateral transection of the lateral olfactory tract on the male-induced implantation failure (the Bruce effect) in mice. *Physiol. Behav.* **36** (1986) 587–590.
18. Wirsig C. R. and Leonard C. M.: Acetylcholinesterase and luteinizing hormone-releasing hormone distinguish separate populations of terminal nerve neurons. *Neuroscience* **19** (1986) 719–740.
19. Witkin J. W. and Silverman A. J.: Luteinizing hormone-releasing hormone (LHRH) in rat olfactory systems. *J. Comp. Neurol.* **218** (1983) 426–432.
20. Pearson A. A.: The development of the nervus terminalis in man. *J. Comp. Neurol.* **75** (1941) 39–66.
21. Singer A. G.: A chemistry of mammalian pheromones. *J. Steroid Biochem. Molec. Biol.* **39**(4B) (1991) 627–632.
22. Meredith M. and O'Connell R. J.: HRP uptake by olfactory and vomeronasal receptor neurons: use as an indicator of incomplete lesions and relevance for non-volatile chemoreception. *Chem. Senses* **13** (1988) 487–515.
23. Jacobs V. L., Sis R. F., Chenoweth P. J., Klemm W. R. and Sherry C. J.: Structures of the bovine vomeronasal complex and its relationships to the palate: tongue manipulation. *Acta Anat.* **110** (1981) 48–58.
24. Jacobs V. L., Sis R. F., Chenoweth P. J., Klemm W. R., Sherry C. J. and Coppock C. E.: Tongue manipulation of the palate assists estrous detection in the bovine. *Theriogenology* **13** (1980) 353–356.
25. Schilling A.: The possible role of urine in territoriality of some nocturnal prosimians. In *Symp. Zool. Soc. Lond.* (Edited by D. H. Stoddart). (1980) pp. 165–193.
26. Schilling A., Serviere J., Gendrot G. and Perret M.: Vomeronasal activation by urine in the primate *Microcebus murinus*: a 2 DG study. *Expl Brain Res.* **81** (1990) 609–618.
27. Melese-d'Hospital P. Y. and Hart B. L.: Vomeronasal organ cannulation in male goats: evidence for transport of fluid from the oral cavity to the vomeronasal organ during flehmen. *Physiol. Behav.* **35** (1985) 941–944.
28. Bojsen-Moller F.: Topography of the nasal glands in rats and some other mammals. *Anat. Rec.* **150** (1964) 11–24.
29. Pevsner J., Hwang P. M., Sklar P. B., Venable J. C. and Snyder S. H.: Odorant binding protein and its mRNA are localized to lateral nasal gland implying a carrier function. *Proc. Natn. Acad. Sci. U.S.A.* **85** (1988) 2383–2387.
30. Meredith M. and O'Connell R. J.: Efferent control of stimulus access to the hamster vomeronasal organ. *J. Physiol.* **286** (1979) 301–316.
31. Eccles R.: Autonomic innervation of the vomeronasal organ of the cat. *Physiol. Behav.* **28** (1982) 1011–1015.
32. Hamlin H. E.: Working mechanism for the liquid and gaseous intake and output of Jacobson's organ. *Am. J. Physiol.* **91** (1929) 201–205.
33. Meredith M., Marques D. M., O'Connell R. J. and Stern F. L.: Vomeronasal pump: significance for male hamster sexual behavior. *Science* **207** (1980) 1224–1226.
34. Meredith M.: Chronic electrophysiological recordings of vomeronasal pump activation in awake animals. *Chem. Senses* **12** (1987) 683.
35. Graziadei P. P. C.: Functional anatomy of the mammalian chemosensory system. In *Chemical Signals in Vertebrates* (Edited by M. Mozell and D. Muller-Schwarze). Plenum Press, New York (1977) pp. 435–454.
36. Tucker D.: Non-olfactory responses from the nasal cavity: Jacobson's organ and the trigeminal system. In *Handbook of Sensory Physiology* (Edited by L. M. Beidler). Springer, Berlin, Vol. 4 (1971) pp. 151–181.
37. Adams D. R. and Wiekamp M. D.: The canine vomeronasal organ. *J. Anat.* **138** (1984) 771–787.

38. Taniguchi K. and Mochizuki K.: Morphological studies on the vomeronasal organ in the golden hamster. *Jap. J. Vet. Sci.* **44** (1982) 419–426.
39. Lowe G. and Gold G. H.: Localization of transduction to olfactory receptor cilia. *Chem. Senses* (1991). In press.
40. Menco B. Ph. M.: Ultrastructural localization of the transduction apparatus in the rat's olfactory epithelium. *Chem. Senses* (1991). In press.
41. Boekhoff I. and Breer H.: Differential stimulation of second messenger pathways by distinct classes of odorants. *Neurochem. Int.* **17** (1990) 553–557.
42. Breer H., Boekhoff I. and Tareilus E.: Rapid kinetics of second messenger formation in olfactory transduction. *Nature* **344** (1990) 65–68.
43. Gold G. H. and Nakamura T.: Cyclic nucleotide-gated conductances: a new class of ion channels mediates visual and olfactory transduction. *TINS* **8** (1987) 312–316.
44. Kalinoski L., Aldinger S., Boyle A., Huque T. and Restrepo D.: Characterization of an inositol-1,4,5-trisphosphate (IP_3) receptor in isolated olfactory cilia. *Chem. Senses* (1991). In press.
45. Duchamp A., Revial M. F., Holley A. and MacLeod P.: Odor discrimination by frog olfactory receptors. *Chem. Senses Flavour* **1** (1974) 213–233.
46. Sachs M. B., Voigt H. F. and Young E. D.: Auditory nerve representation of vowels in background noise. *J. Neurophysiol.* **50** (1983) 27–65.
47. Buck L. and Axel R.: A novel multigene family may encode odorant receptors: a molecular basis for odor recognition. *Cell* **65** (1991) 175–187.
48. Keverne E. B., Murphy C. L., Silver W. L., Wysocki C. J. and Meredith M.: Non-olfactory chemoreceptors of the nose: recent advances in understanding of the vomeronasal and trigeminal systems. *Chem. Senses* **11** (1986) 119–133.
49. Mackay-Sim A. and Shaman P.: Topographic coding of odorant quality is maintained at different concentrations in the salamander olfactory epithelium. *Brain Res.* **297** (1984) 207–216.
50. Astic L., Saucier D. and Holley A.: Topographical relationships between olfactory receptor cells and glomerular foci in the rat olfactory bulb. *Brain Res.* **424** (1987) 144–152.
51. Kauer J. S.: Olfactory receptor staining using horseradish peroxidase. *Anat. Rec.* **200** (1981) 331–336.
52. Royet J. P., Sicard G., Souchier C. and Jourdan F.: Specificity of spatial patterns of glomerular activation in the mouse olfactory bulb: computer-assisted image analysis of 2-deoxyglucose autoradiograms. *Brain Res.* **417** (1987) 1–11.
53. Slotnick B. M., Panhuber H., Bell G. A. and Laing D. G.: Odor-induced metabolic activity in the olfactory bulb of rats trained to detect propionic acid vapor. *Brain Res.* **500** (1989) 161–168.
54. Stewart W. B., Kauer J. S. and Shepherd G. M.: Functional organization of rat olfactory bulb analyzed by the 2-deoxyglucose method. *J. Comp. Neurol.* **185** (1979) 715–733.
55. Slotnick B. M., Graham S., Laing D. G. and Bell G. A.: Detection of propionic acid vapor by rats with lesions of olfactory bulb areas associated with high 2-DG uptake. *Brain Res.* **417** (1987) 343–346.
56. Mouly A. M. and Holley A.: Perceptive properties of the multisite micro stimulation of the olfactory bulb in the rat. *Behav. Brain Res.* **21** (1986) 1–12.
57. Wisgirda M. and Meredith M.: Projection from vomeronasal organ to accessory olfactory bulb: no topography. (1991) Submitted.
58. Imamura K., Mori K., Fujita S. C. and Obata K.: Immunochemical identification of subgroups of vomeronasal nerve fibers and their segregated terminations in the accessory olfactory bulb. *Brain Res.* **328** (1985) 362–366.
59. Scott J. W. and Harrison T. A.: The olfactory bulb: anatomy and physiology. In *Neurobiology of Taste and Smell* (Edited by T. E. Finger and W. L. Silver). Wiley-Interscience, New York (1987) pp. 151–178.
60. Meredith M.: Patterned response to odor in mammalian olfactory bulb: I. Influence of intensity. *J. Neurophysiol.* **56** (1986) 572–597.
61. Meredith M.: Temporal and spatial patterns of response to odor in hamster olfactory bulb: single unit recordings and computer simulation. *Chem. Senses* **11** (1986) 638.
62. Meredith M.: Spatial distribution of inhibition during olfactory bulb response to odor: computer simulation. *Chem. Senses* (1991). In press.
63. MacLeod N. K. and Reinhardt T. W.: An electrophysiological study of the accessory olfactory bulb in the rabbit—I. Analysis of electrically evoked potential fields. *Neuroscience* **10** (1983) 119–129.
64. Reinhardt W., MacLeod N. K., Ladewig J. and Ellendorf F.: An electrophysiological study of the accessory olfactory bulb in the rabbit—II. Input–output relations as assessed from analysis of intra and extracellular unit recordings. *Neuroscience* **10** (1983) 131–139.
65. Adrian E. D.: Synchronised activity in the vomeronasal nerves with a note on the function of the organ of Jacobsen. *Pflügers Arch. Ges. Physiol.* **260** (1955) 188–192.
66. Meredith M.: The vomeronasal organ and accessory olfactory system in the hamster. In *Chemical Signals in Vertebrates and Aquatic Animals* (Edited by D. Muller-Schwarze and R. M. Silverstein). Plenum Press, New York (1980) pp. 303–326.
67. Meredith M.: Stimulus access and other processes involved in nasal chemosensory function: potential substrates for neural and hormonal influence. In *Olfaction and Endocrine Regulation* (Edited by W. Breipohl). IRL Press, London (1982) pp. 223–236.
68. Singer A. G., Agosta W. C., O'Connell R. J., Pfaffman C., Bowen D. V. and Field F. H.: Dimethyl Disulfide: an attractant pheromone in hamster vaginal secretion. *Science* **191** (1976) 948–950.
69. Hatanaka T. and Shibuya T.: Odor response patterns of single units in the accessory olfactory bulb of the turtle *Geoclemys reevesii*. *Comp. Biochem. Physiol.* **92A** (1989) 505–512.
70. Clancy A. N., Macrides F., Singer A. G. and Agosta W. C.: Male hamster copulatory responses to a high molecular weight fraction of vaginal discharge: effects of vomeronasal organ removal. *Physiol. Behav.* **33** (1984) 653–660.
71. Inouchi J., Jiang X. C., Wang D., Kubie J. and Halpern M.: Garter snake accessory olfactory bulb neurons respond to a chemoattractive protein purified from earthworm secretions. *Chem. Senses* **15** (1990) 594.
72. Skeen L. C.: Odor-induced patterns of deoxyglucose consumption in the olfactory bulb of the tree shrew. *Brain Res.* **124** (1977) 147–153.
73. Davis B. J., Macrides F., Young W. M., Schneider S. P. and Rosene D. L.: Efferents and centrifugal afferents of the main olfactory bulb and accessory olfactory bulb in the hamster. *Brain Res. Bull.* **3** (1978) 59–72.
74. Winans S. S. and Scalia F.: Amygdaloid nucleus: new afferent input from the vomeronasal organ. *Science* **170** (1970) 330–332.
75. Johnson J. I., Switzer R. C. and Kirsch J. A. W.: The course of the dorsal lateral olfactory tract as an indicator of dichotomy in the phylogeny of placental mammals. *Neuroscience* **5** (1979) 142.

76. Kevetter G. A. and Winans S. S.: Connections of the corticomedial amygdala in the golden hamster. I. Efferents of the vomeronasal amygdala. *J. Comp. Neurol.* **197** (1981) 99–111.
77. Lloyd-Thomas A. and Keverne E. B.: Role of the brain and accessory olfactory system in the block to pregnancy in mice. *Neuroscience* **7** (1982) 907–912.
78. Price J. L.: The central olfactory and accessory olfactory systems. In *Neurobiology of Taste and Smell* (Edited by T. E. Finger and W. L. Silver). Wiley-Interscience, New York (1987) pp. 179–203.
79. Licht G. and Meredith M.: Convergence of main and accessory olfactory pathways onto single neurons in the hamster amygdala. *Expl Brain Res.* **69** 1987) 7–18.
80. Keverne E. B. and de la Riva C.: Pheromones in mice: reciprocal interaction between the nose and brain. *Nature* **296** (1982) 148–150.
81. Kaba H., Rosser A. and Keverne B.: Neural basis of olfactory memory in the context of pregnancy block. *Neuroscience* **32** (1989) 657–662.
82. Jahr C. E. and Nicoll R. A.: Noradrenergic modulation of dendrodendritic inhibition in the olfactory bulb. *Nature* **297** (1982) 227–229.
83. Taylor J. G. and Keverne E. B.: Accessory olfactory learning. *Biol. Cybern.* **64** (1991) 301–305.
84. Freeman W. J. and Skarda C. A.: Spatial EEG patterns, non-linear dynamics and perception: the neo-Sherringtonian view. *Brain Res. Rev.* **10** (1985) 147–175.
85. Sullivan R. M. and Hall W. G.: Reinforcers in infancy: classical conditioning using stroking or intra-oral infusions of milk as UCS. *Devl Psychobiol.* **21** (1988) 215–223.
86. Coopersmith R., Henderson S. R. and Leon M.: Odor specificity of the enhanced neural response following early odor experience in rats. *Devl Brain Res.* **27** (1986) 191–197.
87. Leon M.: Plasticity of olfactory output circuits related to early olfactory learning. *TINS* **10** (1987) 434–438.
88. Bronson F.: The reproductive ecology of the house mouse. *Q. Rev. Biol.* **54** (1979) 265–299.
89. Coquelin A., Clancy A. N., Macrides F., Noble E. P. and Gorski R. A.: Pheromonally induced release of luteinizing hormone in male mice: involvement of the vomeronasal system. *J. Neurosci.* **4** (1984) 2230–2236.
90. Wysocki C. J., Katz Y. and Bernhard R.: Male vomeronasal organ mediates female induced testosterone surges in mice. *Biol. Reprod.* **28** (1983) 917–922.
91. Dorsa D. M. and Smith E. R.: Facilitation of mounting behavior in male rats by intracranial injections of luteinizing hormone-releasing hormone. *Regul. Peptides* **1** (1980) 147–155.
92. Moss R. L., Dudley C. A., Foreman M. M. and McCann S. M.: Synthetic LRF: a potentiator of sexual behavior in the rat. In *Hypothalamic Hormones* (Edited by M. Motta, P. G. Crosignani and L. Martini). Academic Press, London (1975) pp. 269–278.
93. Moss R. L. and Dudley C. A.: Neuropeptides and the social aspects of female reproductive behavior in the rat. In *Advances in Comparative and Environmental Physiology* (Edited by J. Balthazart). Springer, Berlin (1989) pp. 209–237.
94. Silverman A. J., Jhamandas J. and Renaud L. P.: Localization of luteinizing hormone-releasing hormone (LHRH) neurons that project to the median eminence. *J. Neurosci.* **7** (1987) 2312–2319.
95. Pfaff D. W.: Luteinizing hormone-releasing factor potentiates lordosis behavior in hypophysectomized ovariectomized female rats. *Science* **182** (1973) 1148–1149.
96. Meredith M., Howard G. and Wisgirda M.: LHRH injected intracerebrally relieves some behavioral deficits in male hamsters after vomeronasal organ lesions. *Chem. Senses* **15** (1990) 619.
97. Rajendren G., Dudley C. A. and Moss R. L.: Role of the vomeronasal organ in the male-induced enhancement of sexual receptivity in female rats. *Neuroendocrinology* **52** (1990) 368–372.
98. Saito T. R. and Moltz H.: Sexual behavior in the female rat following removal of the vomeronasal organ. *Physiol. Behav.* **38** (1986) 81–87.
99. Mackay-Sim A. and Rose J. P.: Removal of the vomeronasal organ impairs lordosis in female hamsters: effect is reversed by luteinizing hormone-releasing hormone. *Neuroendocrinology* **42** (1986) 489–493.
100. Dluzen D. E. and Ramirez V. D.: Localized and discrete changes in neuropeptide (LHRH and TRH) and neurotransmitter (NE and DA) concentrations within the olfactory bulbs of male mice as a function of social interaction. *Horm. Behav.* **17** (1983) 139–145.
101. Dluzen D. E., Ramirez V. D., Carter C. S. and Getz L. L.: Male vole urine changes luteinizing hormone-releasing hormone and norepinephrine in female olfactory bulb. *Science* **212** (1981) 573–575.
102. Moss R. L. and Dudley C. A.: Differential effects of an luteinizing-hormone-releasing hormone (LHRH) antagonist analogue on lordosis behavior induced by LHRH and the LHRH fragment Ac-LHRH$^{5-10}$. *Neuroendocrinology* **52** (1990) 138–142.
103. Fernandez G., Howard G. and Meredith M.: Early vomeronasal lesions cause severe deficits in male hamster mating behavior; relieved in part by intracerebral LHRH peptides. *Chem. Senses* (1991). In press.
104. Dudley C. A. and Moss R. L.: Facilitation of lordosis in female rats by CNS-site specific infusions of an LH-RH fragment, Ac-LH-RH-(5-10). *Brain Res.* **441** (1988) 161–167.
105. Badr M. and Pelletier G.: Characterization and autoradiographic localization of LHRH receptors in the rat brain. *Synapse* **1** (1987) 567–571.
106. Jennes L., Dalati B. and Conn P. M.: Distribution of gonadotropin releasing hormone agonist binding sites in the rat central nervous system. *Brain Res.* **452** (1988) 156–164.
107. Jennes L., Janovick J., Braden T. and Conn T. M.: Gonadotropin releasing hormone binding sites in rat hippocampus: different structure/binding relationships compared to the anterior pituitary. *Molec. Cell Neurosci.* **1** (1990) 121–127.
108. Millar R. P., Wormald P. J. and De L. Milton R. C.: Stimulation of gonadotropin release by a non-GnRH peptide sequence of the GnRH precursor. *Science* **232** (1986) 68–70.
109. Nowak F. V.: Cloning of two hypothalamic cDNAs encoding tissue-specific transcripts in the preoptic area and testis. *Molec. Endocr.* **4** (1990) 1205–1210.
110. Pan J. T., Kow L. M. and Pfaff D. W.: Modulatory actions of luteinizing hormone-releasing hormone on electrical activity of preoptic neurons in brain slices. *Neuroscience* **27** (1988) 623–628.
111. Silverman A. J.: The gonadotropin-releasing hormone (GnRH) neuronal systems: immunocytochemistry. In *The Physiology of Reproduction* (Edited by E. Knobil and J. Neill). Raven Press, New York (1988) pp. 1283–1303.
112. Maragos W. F., Winans-Newman S., Lehman M. and Powers J. B.: Neurons of origin and fiber trajectory of amygdalofugal projections to the medial preoptic area in Syrian hamsters. *J. Comp. Neurol.* **280** (1989) 59–71.
113. Beltramino C. and Taleisnik S.: Ventral premammillary nuclei mediate pheromonal-induced LH release stimuli in the rat. *Neuroendocrinology* **41** (1985) 119–124.

114. Fink G.: Gonadotropin secretion and its control. In *The Physiology of Reproduction* (Edited by E. Knobil and J. Neill). Raven Press, New York (1988) pp. 1349–1377.
115. Weiner R. I., Findell P. R. and Kordon C.: Role of classic and peptide neuromediators in the neuroendocrine regulation of LH and prolactin. *The Physiology of Reproduction* (Edited by E. Knobil and J. Neill). Raven Press, New York (1988) pp. 1235–1281.
116. Pfaff D. W.: *Estrogens and Brain Function (Neural Analysis of a Hormone Controlled Mammalian Reproductive Behavior)*. Springer, New York (1980).
117. Rainbow T. C., Parsons B., MacLusky N. J. and McEwen B. S.: Estradiol receptor levels in rat hypothalamic and limbic nuclei. *J. Neurosci.* **2** (1982) 1439–1445.
118. Sheridan P. J.: The nucleus interstitialis striae terminalis and the nucleus amygdaloideus medialis: prime targets for androgen in the rat forebrain. *Endocrinology* **104** (1979) 130–136.
119. Shivers B. D., Harlan R. E., Morell J. I. and Pfaff D. W.: Absence of estradiol concentration in cell nuclei of LHRH immunoreactive neurons. *Nature* **304** (1983) 345–347.
120. Nishizuka M. and Arai Y.: Organizational action of estrogen on synaptic pattern in the amygdala: implications for sexual differentiation. *Brain Res.* **231** (1981) 422–426.
121. Page R. B.: The anatomy of the hypothalamus–hypophyseal complex. In *The Physiology of Reproduction* (Edited by E. Knobil and J. Neil). Raven Press, New York (1988) p. 1161.
122. Roos J., Roos M., Schaeffer C. and Aron C.: Sexual differences in the development of accessory olfactory bulbs in the rat. *J. Comp. Neurol.* **270** (1988) 121–131.
123. Reynolds J. and Keverne E. B.: The accessory olfactory system and its role in the pheromonally mediated suppression of oestrus in grouped mice. *J. Reprod. Fert.* **57** (1979) 31–35.
124. Lomas D. E. and Keverne E. B.: Role of the vomeronasal organ and prolactin in the acceleration of puberty in female mice. *J. Reprod. Fert.* **66** (1982) 101–107.
125. Li C.-S., Kaba H., Saito H. and Seto K.: GABAergic mechanisms are involved in the control of tubero-infundibular arcuate neurons by the accessory olfactory bulb. *Neurosci. Lett.* **120** (1990) 231–233.