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Comparative anatomy of the vomeronasal organ complex in bats†

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INTRODUCTION

The first description of the vomeronasal organ (of Jacobson) was by Ruysch in 1703. Since Jacobson's (1811) report there have been many studies of this organ in a wide spectrum of vertebrates (cf. Negus, 1958). Investigations in the order Chiroptera, however, have been relatively scarce and brief. Schwink (1888) reported the absence of the organ in embryos of *Vespertilio murinus*. Herzfeld (1889), reporting on *Pteropus edwarsi* (*sic*) and an unidentified bat, found no organ. Duval & Garnault (1895) reported the presence of the organ in an embryo of *Vesperugo pipistrellus* but reported its absence in *Rhinolophus ferrum-equinum* and *Vespertilio murinus*. Broom (1895, 1897) described a remarkably well-developed organ in the vespertilionid *Miniopterus schreibersi*, but in *Pteropus poliocephalus* and *Nyctophilus* (sp.?) he found none. Grosser (1900) reported the organ in *Rhinolophus ferrum-equinum* and *R. hipposideros* but found none in several vespertilionid bats. Simonetta & Magnoni (1939) found it in an embryo of *R. ferrum-equinum*. Mann (1961) gave a gross description of the organ in *Phyllostomus hastatus*, *Glossophaga soricina*, *Desmodus rotundus*, and *Artibeus jamaicensis*. Schmidt & Greenhall (1971) commented on the well-developed organ in *D. rotundus* and indicated its position in the nasal septum. Histological descriptions were provided by Broom on *Miniopterus* and by Bhatnagar & Kallen (1974*b*) on *A. jamaicensis*. The latter authors reported no organ in *Myotis lucifugus*. Suthers (1970) provided a review of the vomeronasal organ in bats. Preliminary observations on the vomeronasal organ complex in ten species of bats have been reported by Cooper & Bhatnagar (1975).

Considering the suggested role of the vomeronasal organ in feeding (Negus, 1956), the diverse interspecific dietary preferences of bats make the order Chiroptera a significant group in which to conduct comparative investigations on this organ. We report here our anatomical findings on fourteen species of bats, representing six of the sixteen Recent chiropteran families (Koopman & Jones, 1970; Smith, 1972) and belonging to five of six known dietary groups (Bhatnagar & Kallen, 1974*a*). This paper describes the organ complex in eight species of bats and includes the first reports on *Rhinopoma microphyllum*, *Megaderma lyra* and *Hipposideros lankadiva*. Also reported for the first time is an accessory olfactory bulb in *Miniopterus schreibersi*.

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MATERIALS AND METHODS

The following species were investigated:

Suborder Megachiroptera	
Family Pteropodidae	
Rousettus leschenaulti	1♀
Pteropus giganteus	1♂
Cynopterus sphinx	1♀
Suborder Microchiroptera	
Family Rhinopomatidae	
Rhinopoma microphyllum	1♀
Family Megadermatidae	
Megaderma lyra	1♀
Family Rhinolophidae	
Rhinolophus lepidus	1♀
Hipposideros lankadiva	1♀
Family Phyllostomatidae	
Glossophaga soricina	1?
Artibeus jamaicensis	2♂♂, 2♀♀
Desmodus rotundus	2♀♀
Family Vespertilionidae	
Myotis lucifugus	3♂♂
Pipistrellus subflavus	1♂
Eptesicus fuscus	2♂♂
Miniopterus schreibersi	1♂

Rousettus and *Cynopterus* were collected in Jhabua, India and preserved in 10% formalin. *Pteropus*, *Rhinopoma*, *Megaderma*, *Rhinolophus* and *Hipposideros* were also collected in India (Gwalior and Guna, M.P.) and were perfused with Bouin's fluid as described by Bhatnagar & Kallen (1974*b*, 1975). *Artibeus*, collected in Mexico, and *Myotis*, captured in Buffalo, New York, were similarly preserved with Bouin's fluid. Two specimens of *Desmodus* (one glutaraldehyde-perfused) and one specimen of *Glossophaga* (preserved in 10% formalin) became available through the courtesy of Professor William A. Wimsatt. For one alcohol-fixed specimen of *Miniopterus* we are grateful to Dr Robert L. Martin. *Eptesicus* and *Pipistrellus* were locally collected and perfused with Bouin's fluid. Many voucher specimens have been deposited with the American Museum of Natural History, New York, and the Museum, Texas Tech University. We are thankful to Dr Karl F. Koopman for help in identifying some specimens.

Frontal serial sections of 10 μm were prepared from the decalcified cranium (Evans & Krajian, 1930) of each bat as described by Bhatnagar & Kallen (1974*b*, 1975). Each series was stained with Gomori's (1950) one-step trichrome. Selected

sections were treated with the periodic acid-Schiff (PAS) technique to demonstrate carbohydrate-containing structures. Others were stained using the protargol silver method of Bodian (1936, 1937) for the examination of neural structures.

RESULTS

General observations

There is no prototype of the vomeronasal organ in the order Chiroptera. Certain general characteristics, however, may be cited at this point to provide an outline for specific morphological descriptions.

The term 'vomeronasal organ complex' is used in reference to the vomeronasal organ, the vomeronasal glands, and all associated structures contained within, and including, the vomeronasal cartilage (Fig. 1). The chiropteran vomeronasal organ complexes are bilaterally located in the anterior ventral nasal septum. Each organ is contained in a vomeronasal cartilage which in cross section is either C-shaped, hook-shaped, or some modification thereof. Anteriorly, the cartilage may completely encircle the organ. Processes from these cartilages accompany the nasopalatine ducts in their course between the oral and nasal cavities. Other nasal cartilages may also contribute to the support of the nasopalatine ducts (e.g. palatine cartilages).

The anterior nasal cartilages are strikingly similar in all species examined, regardless of the presence or absence of the vomeronasal organ (Fig. 2). The bilateral vomeronasal cartilage is ventrolateral to the septal cartilage. In those bats which have no vomeronasal organ, the homologue of this cartilage, the paraseptal cartilage, occupies a similar position in the septum. This paraseptal cartilage is a simple elongated structure in contrast to the curved vomeronasal cartilage. Both cartilages have anterior processes which accompany the nasopalatine ducts. The palatine cartilages may also contribute to the support of the ducts and the inferior nasal cartilages may also show similar anterior connections.

The nasopalatine ducts open anteromedially into the roof of the oral cavity. The ostium (Fig. 2G) of the vomeronasal organ is located near where the nasopalatine duct joins the nasal cavity. The epithelium of this region continues into the ostium, but soon differentiates to become characteristic of the organ. In better developed organs the medial epithelium is pseudostratified ciliated columnar and resembles olfactory epithelium; the lateral epithelium is ciliated columnar. Characteristic receptor and supporting cell nuclei are readily observable in the sensory epithelium of several species. The basal cell nuclei are, however, difficult to identify in our 10 μm series. Vomeronasal glands located within the septum open into the vomeronasal organ. The epithelium of the gland ducts becomes continuous with that of the organ (Fig. 1). These glands are predominantly 'serous' but 'mucous' elements may also be present. Secretory material is seen in the lumen of the organ. Venous sinuses are present and are typically oriented dorsally or laterally relative to the organ and are most prominent in the better developed organ complexes. The unmyelinated vomeronasal nerve corresponds in size to the apparent sensory differentiation of the vomeronasal epithelium.

The paravomeronasal ganglion is frequently seen in the chiropteran vomeronasal

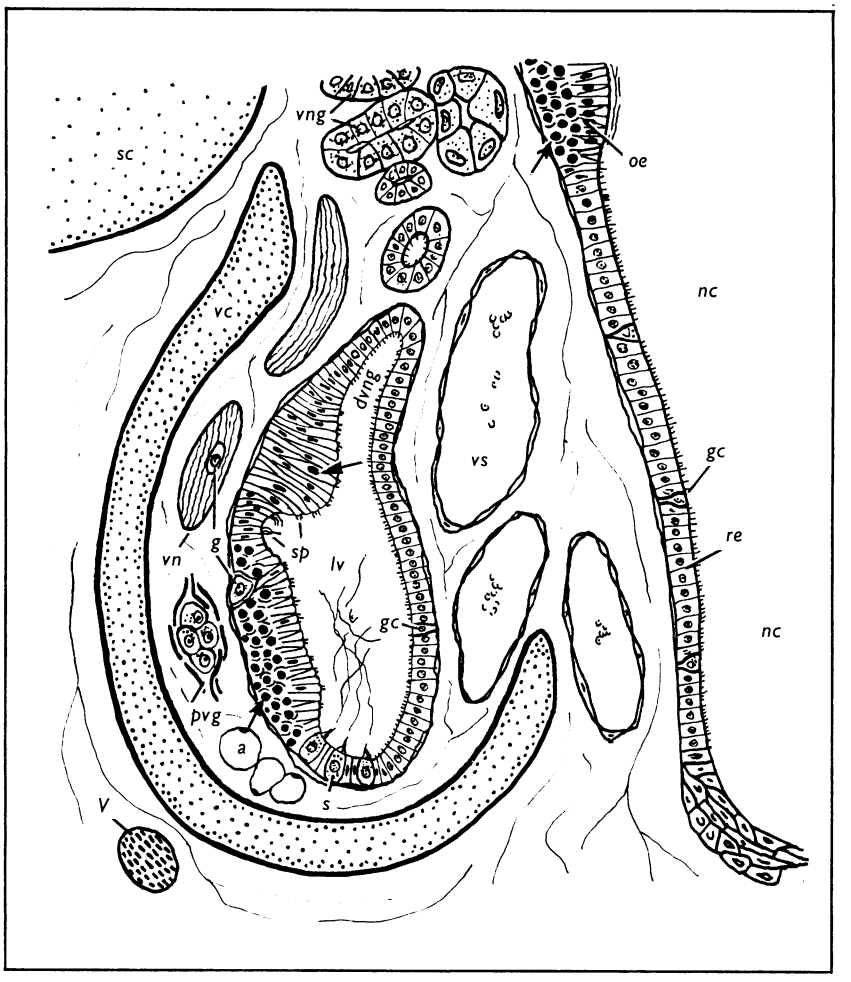


Fig. 1. Composite drawing of 'chiropteran' vomeronasal organ complex, incorporating structures observed in species investigated in this study. It is emphasized that this is idealized and not a depiction of the organ complex of any one species. Abbreviations: *a*, adipose cell; *dvng*, duct of vomeronasal gland; *g*, ganglion cell body; *gc*, goblet cell; *lv*, lumen of the vomeronasal organ; *nc*, nasal cavity; *oe*, olfactory epithelium; *pvg*, paravomeronasal ganglion; *re*, respiratory epithelium; *s*, secretory cell; *sc*, septal cartilage; *sp*, surface projections; *vc*, vomeronasal cartilage; *vn*, vomeronasal nerve; *vng*, vomeronasal gland; *vs*, vascular sinus; *V*, branch of trigeminal nerve. Arrows indicate receptor cell nuclei.

organ complex. It also corresponds to the degree of development of the organ, and is in close association with the vomeronasal nerve (Fig. 1, *pvg*). It is sometimes seen as diffuse ganglion cell bodies. In at least one species intraepithelial ganglion cell bodies have been observed. Accessory olfactory bulbs are present in those bats which have well-developed vomeronasal organs. Presence of the organ does not necessarily indicate presence of the accessory olfactory bulb. The vomeronasal nerve passes medially to project to the accessory olfactory bulb (Fig. 3).

Anatomical features of the vomeronasal organ complex in each investigated species are described below. Measurements are provided in Tables 1 and 2. For this comparative study, consideration of as many species as possible has been much more informative than the investigation of many specimens of individual species. In consideration of differential tissue shrinkage, and since observations were made on single serially sectioned specimens, all measurements should be regarded as approximations.

Suborder Megachiroptera

Family Pteropodidae

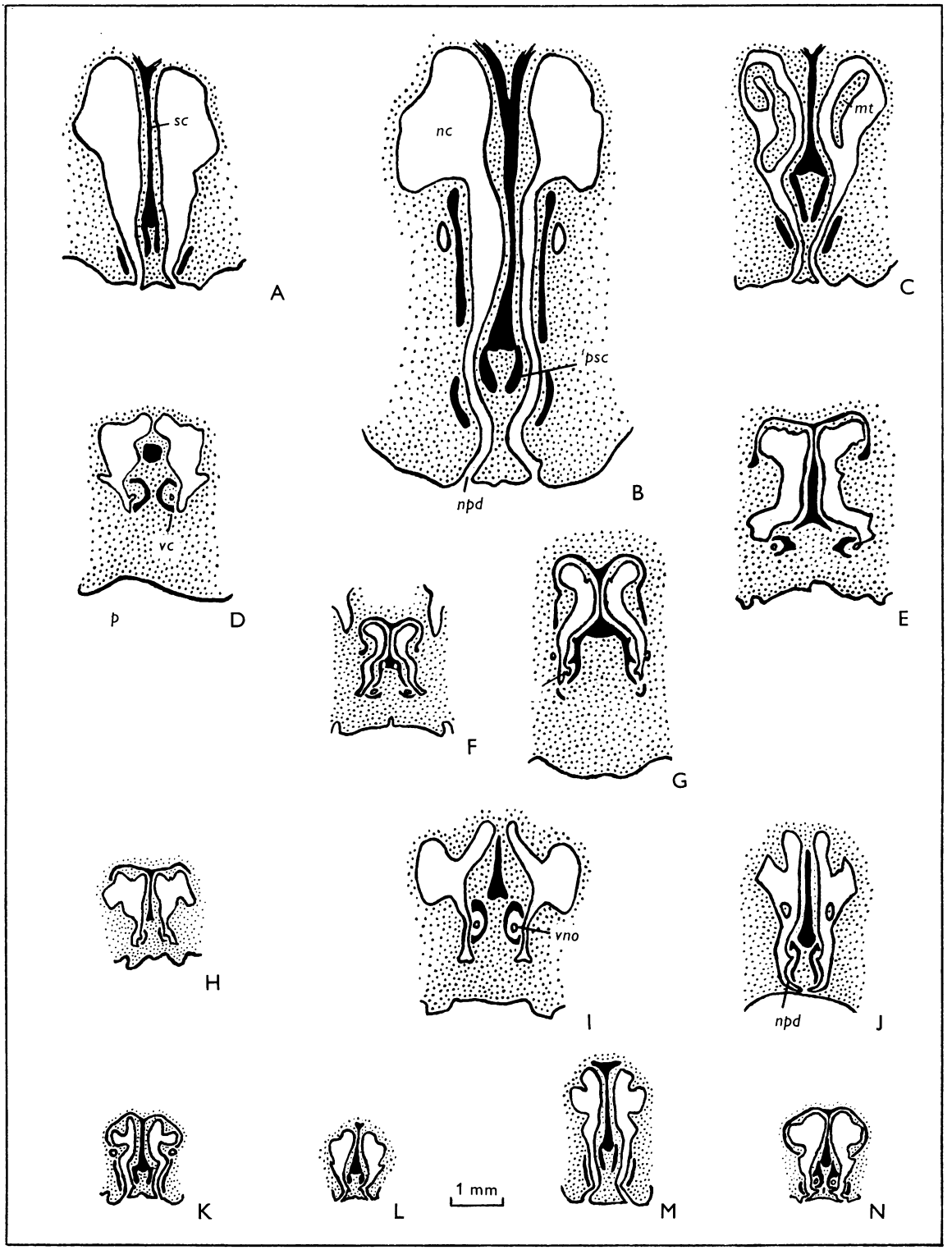
Rousettus leschenaulti, *Pteropus giganteus*, *Cynopterus sphinx* (Figs. 2A–C). The vomeronasal organ has been reported to be absent from this family (Herzfeld, 1889; Broom, 1897; Matthes, 1934). The three megachiropteran species we investigated support this conclusion. Despite the absence of the vomeronasal organ, patent nasopalatine ducts (Figs. 2A–C) lined with stratified squamous epithelium are present in all three pteropids. The nasal cavities of these bats are dorsoventrally elongated and this is reflected in the structure of the anterior nasal cartilages. The structural correspondence with the cartilages of the species which have vomeronasal organs is, however, readily apparent (also see Broom, 1897). The olfactory bulbs of these species were not investigated for the presence of accessory olfactory bulbs.

Suborder Microchiroptera

Family Rhinopomatidae

Rhinopoma microphyllum (Figs. 2D, 4A–B and 12). The nasal septum is exceptionally broad except where it joins the roof of the nasal cavities (Fig. 4A). For most of its extent the tiny vomeronasal organ is completely encircled by the large vomeronasal cartilage, and is suspended in the centre of the cartilage by loose connective tissue (Fig. 4B). The ostium of the organ opens 4·6 mm posteriorly in the nasal cavity. Lined with stratified squamous epithelium, the nasopalatine ducts, through most of their course, are about 300 μm in diameter, and open into the oral cavity through the extremely thick palate (Fig. 4A) medial to the upper incisors. The ducts are 1·28 mm long and are accompanied by processes from the vomeronasal cartilages.

The organ opens into an anterior–inferior extension of the nasal cavity which is lined with pseudostratified ciliated columnar epithelium. The ostium of the organ is similarly lined. Simple ciliated columnar epithelium, however, predominates in the remainder of the organ. A basement membrane is clearly observed. Long surface projections are prominent throughout the lumen, but are longer posteriorly. These are reminiscent of stereocilia, but structures resembling basal bodies are present. The epithelium does not appear to be characteristically olfactory. Vomeronasal glands are not present. No glandular tissue is contained within the cartilage and no ducts open into the organ. The mucous septal glands, localized in the prominent bulges on either side of the septum (Fig. 4A), seem not to be associated with the organ. PAS staining failed to reveal any mucins in the lumen. The loose connective tissue contains adipose cells and vascular sinuses. The paravomeronasal ganglion (Fig. 4B) is not as striking as in other species. The minute vomeronasal nerve is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identifiable, but not traceable on the septum. Careful examination of the olfactory bulbs revealed no distinct areas which could be described as accessory olfactory bulbs.

Family Megadermatidae

Megaderma lyra (Figs. 2E, 5A–B and 13). The vomeronasal organ is located low with respect to the nasal cavities and the septum (Figs. 2E and 5A). The two organs are widely separated and the most ventral portion of the septum broadens to accomodate the superior aspects of the vomeronasal cartilages. The lumen of the organ is located near the level of the floor of the nasal cavity. The ostium of the vomeronasal organ is situated at the juncture of the nasopalatine duct and nasal cavity. Anteriorly, for a short distance, the vomeronasal cartilage completely encircles the organ, but the C shape remains the dominant configuration (Figs. 2E and 5A). The cartilage lies superior to the palatine processes of the maxilla. The nasopalatine ducts open into the oral cavity 2·1 mm from the tip of the snout, medial to the canines and 2·5 mm rostral to the beginning of the nasal cavity. The ducts are lined with stratified squamous epithelium and are accompanied by processes from the vomeronasal cartilage, into which muscle fibres are inserted. Passing posteriorly, the ducts (4 mm long) separate and run adjacent to the maxilla and superior to its palatine processes.

The tubular vomeronasal organ is accompanied in its cartilage sheath by loose connective tissue. Vascular sinuses surround the organ randomly. Anteriorly, 'serous' vomeronasal glands are superior to the vomeronasal cartilage. Gland ducts can be seen along the length of the organ entering through its lateral wall. Posteriorly, where the vomeronasal cartilage tapers, the glands are closer to the organ. Laterally, the epithelium of the organ is low ciliated columnar, whereas medially, it is pseudostratified ciliated columnar and appears sensory (Fig. 13). Large, round nuclei characteristic of olfactory receptors are observed. Though no paravomeronasal ganglion is observed, the small vomeronasal nerve can be seen accompanying the organ. No accessory olfactory bulbs were observed.

Family Rhinolophidae

Rhinolophus lepidus (Figs. 2F, 6A–B and 14A–C). The broad ventral portion of the wedge-shaped nasal septum contains the vomeronasal organ complex. The vomeronasal and septal cartilages are in continuity with each other. Anteriorly, the organ is completely encircled by the cartilage (Figs. 6A–B). The nasopalatine ducts open into the oral cavity 1·5 mm posterior to the tip of the snout, and 1·8 mm

Fig. 2. Scheme of the anterior nasal cartilages in bats. The cartilages of this region are strikingly similar in all species examined regardless of the presence or absence of the vomeronasal organ. In all species the representative sections are from the region where the nasopalatine ducts join the nasal cavities. A, *R. leschenaulti*; B, *P. giganteus*; C, *C. sphinx*; D, *R. microphyllum**; E, *M. lyra**; F, *R. lepidus**; G, *H. lankadiva**; H, *G. soricina**; I, *A. jamaicensis**; J, *D. rotundus**; K, *M. lucifugus*; L, *P. subflavus*; M, *E. fuscus*; N, *M. schreibersi**. (*Species where vomeronasal organ is present.) Abbreviations: *mt*, maxilloturbinal; *nc*, nasal cavity; *npd*, nasopalatine duct; *o*, ostium; *p*, palate; *psc*, paraseptal cartilage; *sc*, septal cartilage; *vc*, vomeronasal cartilage; *vno*, vomeronasal organ.

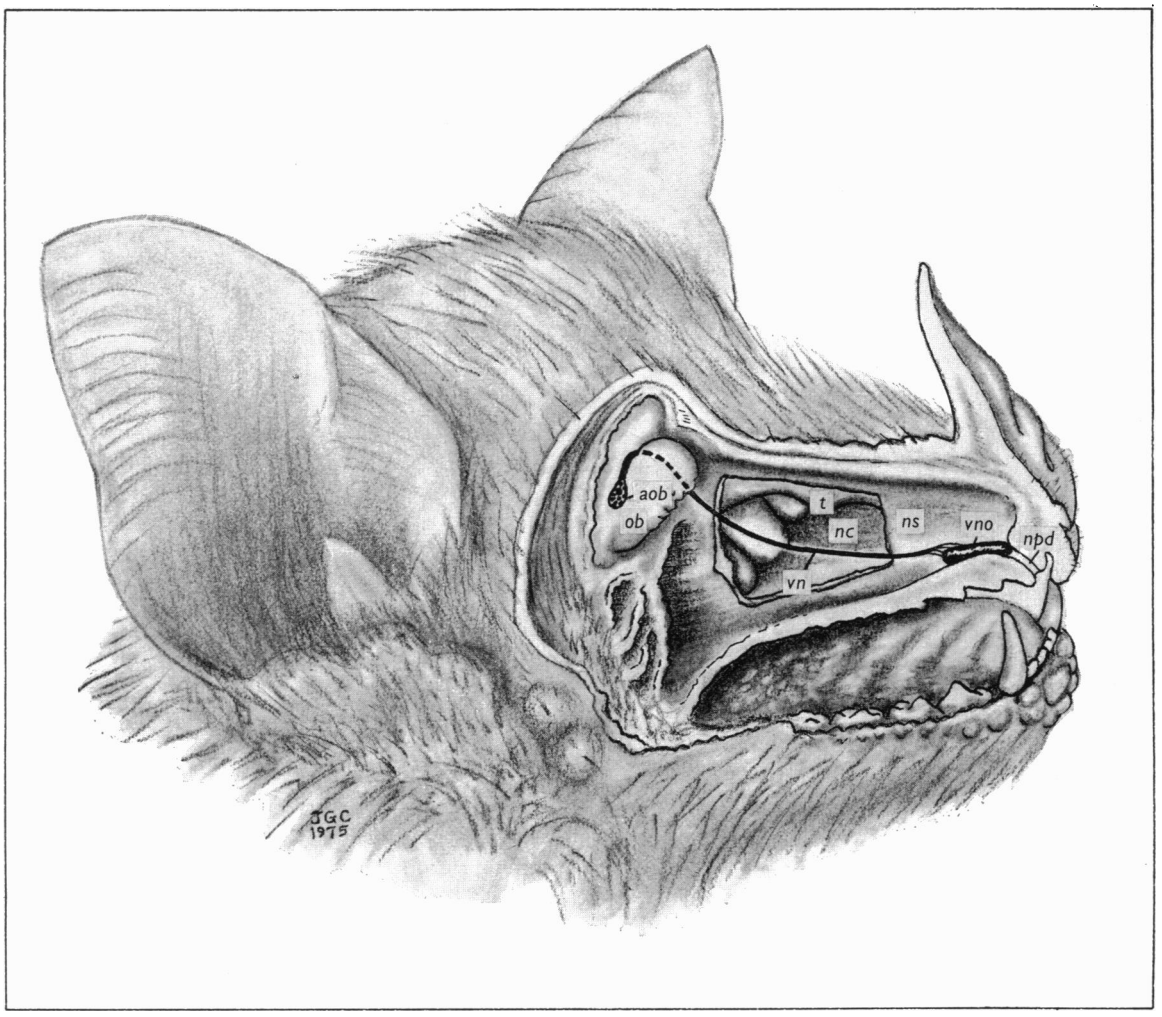


Fig. 3. Stylized illustration of dissection of *Artibeus jamaicensis* ♀. The lateral wall of the right nasal cavity and the turbinates have been removed to show the position of the right vomeronasal organ with respect to the nasal cavity and nasopalatine duct. Note the ascending course of the vomeronasal nerve (here diagrammatically depicted as a single nerve bundle) projecting to the ipsilateral accessory olfactory bulb located postero-dorsolaterally with respect to the olfactory bulb. Part of the nasal septum has been removed to show turbinates within the left nasal cavity. Abbreviations: *aob*, accessory olfactory bulb; *nc*, left nasal cavity; *npd*, nasopalatine duct; *ns*, nasal septum; *ob*, olfactory bulb. *t*, turbinates; *vn*, vomeronasal nerve; *vno*, vomeronasal organ; (ca. × 3½).

before the beginning of the nasal cavity. In this region the ducts lined with stratified squamous epithelium measure approximately 300 μm. Anterior processes of the vomeronasal cartilages accompany the ducts.

The ostium of the vomeronasal organ opens into the medial wall of the nasopalatine duct 3·2 mm posteriorly in the snout. The nasal cavities also begin at this point. The organ is initially lined with stratified squamous epithelium, but 300 μm

Table 1. *Selected measurements relating to the vomeronasal organs (left side) of the investigated species*

The vomeronasal organ was found to be absent in *Rousettus leschenaulti*, *Pteropus giganteus*, *Cynopterus sphinx*, *Myotis lucifugus*, *Pipistrellus subflavus* and *Eptesicus fuscus*. Dietary preferences (C, carnivorous; F, frugivorous; I, insectivorous; N, nectivorous; S, sanguivorous) are from Walker (1975). *Measurements taken in areas of sensory epithelium, where present.

	Vomeronasal organ								
Species	Greatest length (mm)	Greatest diameter (lumen) (μm)	Epithelial thickness* (μm)	Distance from beginning of nasal cavity (mm)	Distance from roof of oral cavity: Anteriorly (mm)	Distance from roof of oral cavity: Posteriorly (mm)	Paravomero-nasal ganglion	Accessory olfactory bulb	Diet
Rhinopoma microphyllum	0·65	90	22	4·60	1·750	1·310	Present	?	I
Megaderma lyra	1·05	132	13	1·56	0·910	0·998	?	?	C
Rhinolophus lepidus	1·17	135	15	0·00	0·345	0·611	Present (diffuse)	?	I
Hipposideros lankadiva	0·64	135	32	1·00	1·766	1·862	Present (diffuse)	?	I
Glossophaga soricina	3·90	250	60 (variable)	0·87	0·312	0·164	Present (diffuse)	Present	F, N, I
Artibeus jamaicensis	1·60	345	50	1·60	1·344	1·344	Present	Present	F
Desmodus rotundus	2·30	650	63	2·10	0·520	0·293	Present	Present	S
Miniopterus schreibersi	1·70	296	23	1·20	0·221	0·481	?	Present	I

Table 2. *Dimensions of the olfactory bulb and the accessory olfactory bulb where present (pteropid olfactory bulbs were not investigated; for their measurements see Bhatnagar & Kallen, 1974a)*

Species	Olfactory bulb		Accessory olfactory bulb	
	Length (mm)	Greatest diameter (mm)	Length (μm)	Greatest diameter (μm)
Rhinopoma microphyllum	1·40	1·77	—	—
Megaderma lyra	2·20	2·25	—	—
Rhinolophus lepidus	1·19	1·61	—	—
Hipposideros lankadiva	2·90	1·59	—	—
Glossophaga soricina	1·65	1·44	240	455
Artibeus jamaicensis	3·10	3·26	530	470
Desmodus rotundus	2·22	2·55	280	1270
Myotis lucifugus	1·07	1·53	—	—
Pipistrellus subflavus	1·05	0·92	—	—
Eptesicus fuscus	1·88	1·82	—	—
Miniopterus schreibersi	1·40	1·82	160	360

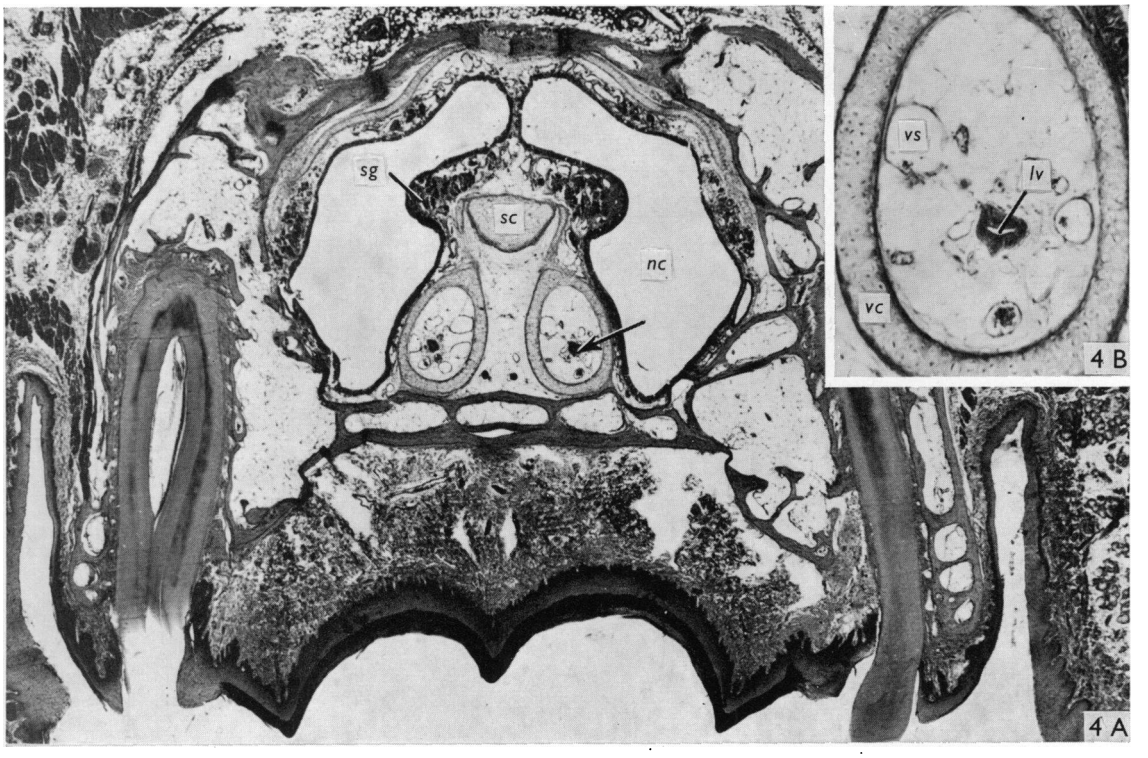
posteriorly this epithelium differentiates into ciliated columnar epithelium. The organ is circular in cross section and shows no medial–lateral differentiation of the epithelium, which does not resemble olfactory epithelium; no sensory cells could be positively identified. The vomeronasal glands appear to be mixed but predominantly 'serous'. Ducts open into the organ posteriorly. The lumen is filled with mucus and particulate matter (Fig. 14B). Large, ciliated, mucus-secreting cells are seen in the epithelium (Figs. 14B–C). Prominent vascular sinuses are located around the organ. The vomeronasal nerve is not identifiable. A branch of the trigeminal nerve can be seen in close association with the organ posteriorly. A few ganglionic cell bodies are seen located adjacent to the epithelium. Accessory olfactory bulbs are apparently absent.

Hipposideros lankadiva (Figs. 2G, 7A–B and 16). The vomeronasal organ is rudimentary and is located in an impressive nasal septum. The broad ventral septum contains massive septal and vomeronasal cartilages. The latter are continuous superiorly with the septal cartilage. Ventrally, the cartilages form troughs to contain the small vomeronasal organs which are situated low in the nasal septum, level with the floor of the nasal cavity.

Figs. 4–11. Representative photomicrographs from 10 μm frontal sections showing position and (insets) detailed structure of vomeronasal organ complexes in eight species of bats. Arrows indicate the lumen of the organ. All main photomicrographs, ×22; all insets, ×80. Gomori trichrome. Abbreviations as in Figure 1.

Fig. 4. *Rhinopoma microphyllum.* A and B, small vomeronasal organ is contained with a large amount of connective tissue within a large vomeronasal cartilage. Note the extremely thick palate and the absence of glandular tissue within the cartilaginous capsule. Septal glands (*sg*) are contained in prominent bulges of the nasal septum.

Fig. 5. *Megaderma lyra.* A, the organ complex is oriented low with respect to the nasal cavities; B, the vomeronasal glands are superior to the cartilage.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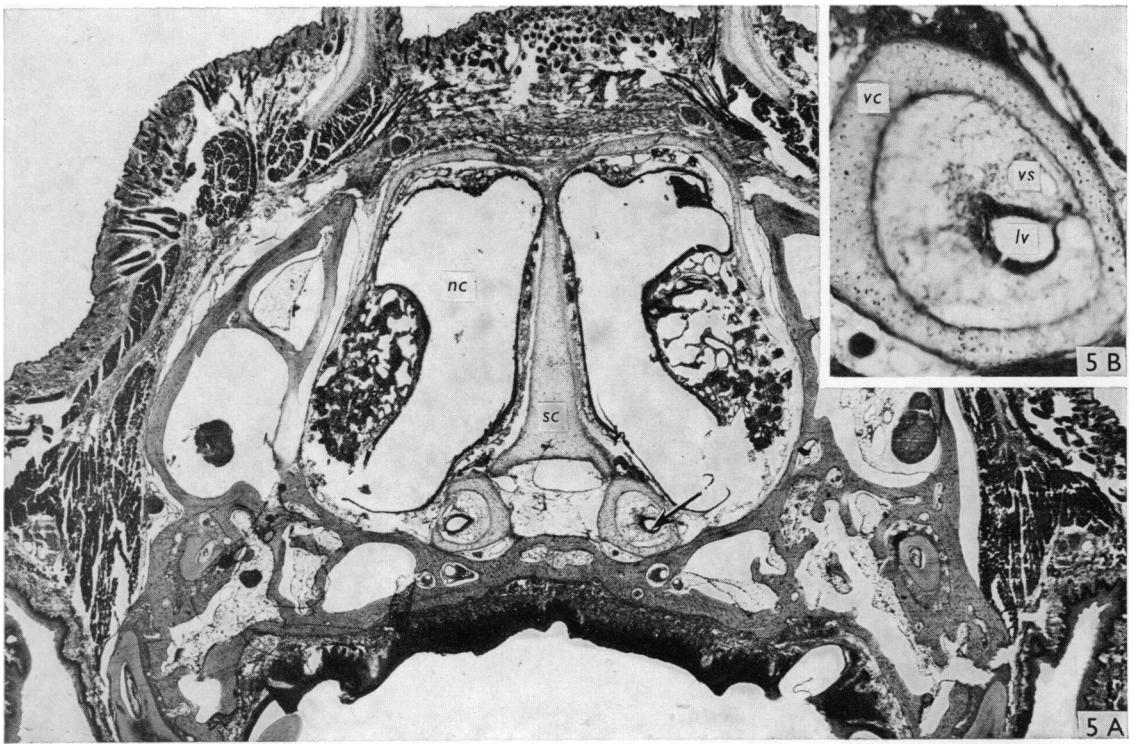
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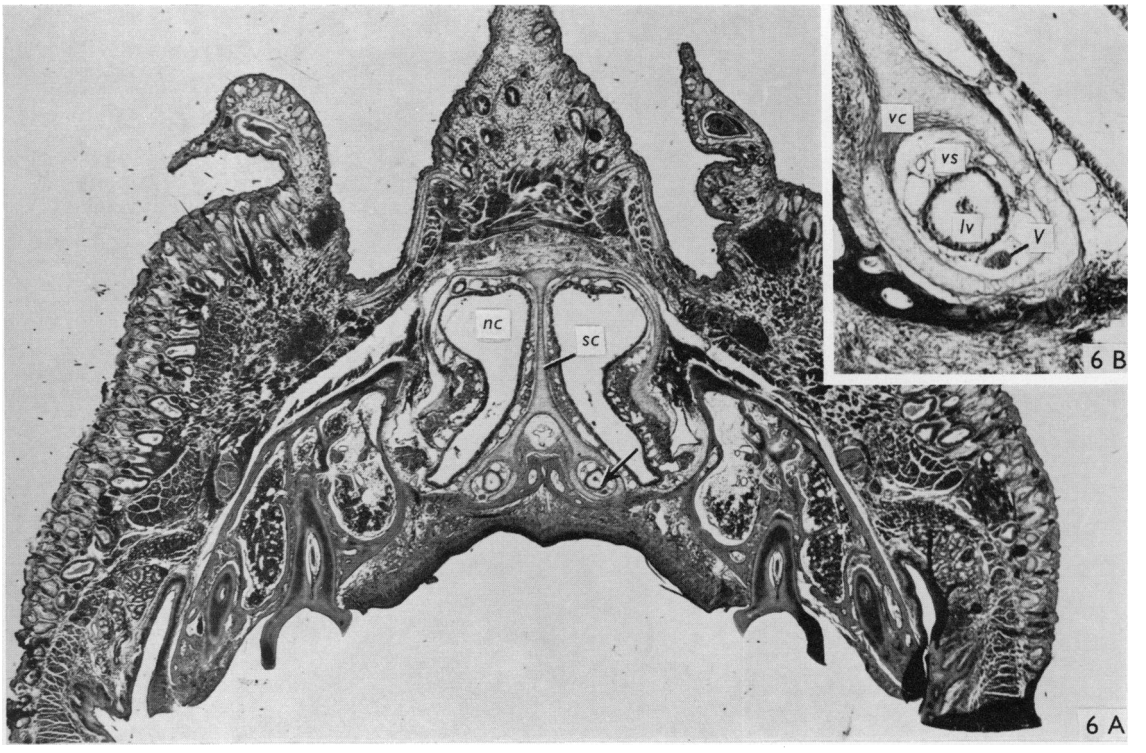
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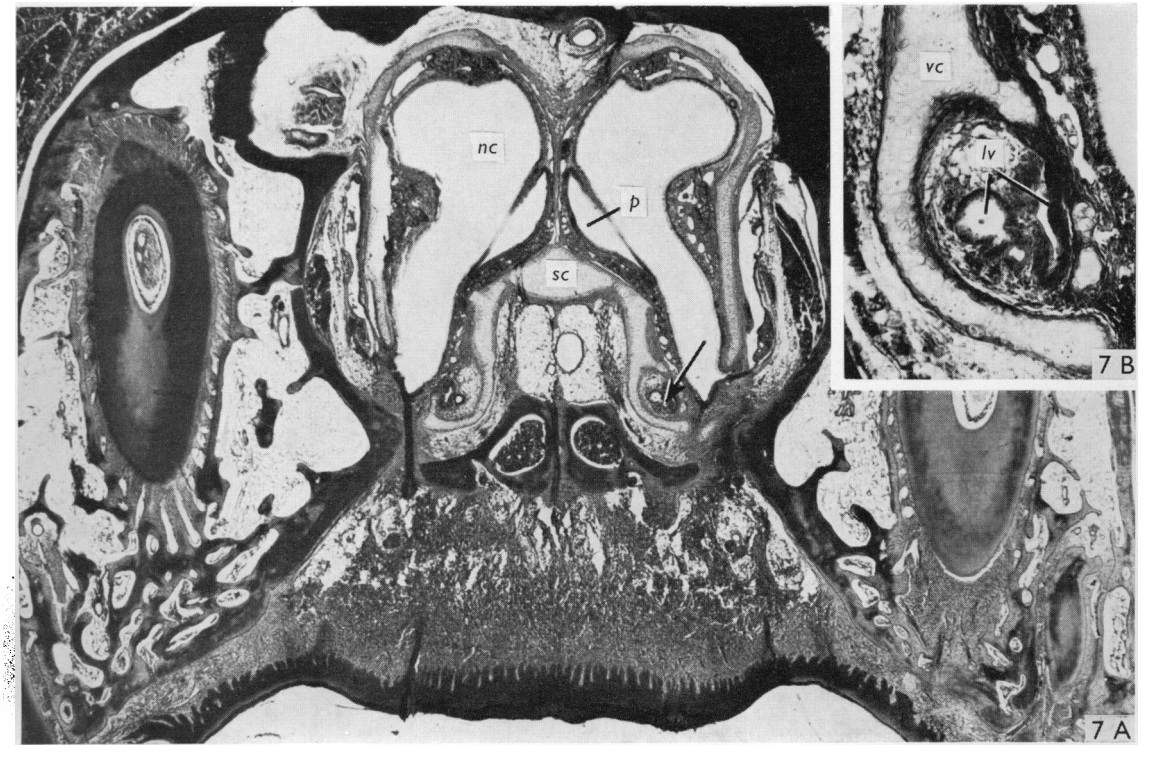
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Another noteworthy feature of the nasal septum is the presence of bilateral 'pockets' (Fig. 7A, *p*) located superolateral to the base of the septal cartilage. These septal pockets are 700 μm long. The wide mouths of the pockets open anteriorly in the nasal cavities. The superior edges of these pockets take origin anteriorly from the roof of the nasal cavities. Anteriorly, they are lined with pseudostratified columnar epithelium but posteriorly, the pseudostratified epithelium shows at least some superficial resemblance to olfactory epithelium. Glandular tissue is seen associated with the walls of these pockets.

The nasopalatine ducts are 4·2 mm long, begin in the nasal cavity 6·2 mm posteriorly, and open into the roof of the oral cavity 1·9 mm from the tip of the snout. The ducts measure 0·50 mm in diameter. These ducts, as well as the ostium of the organ (100 μm in diameter), are lined with stratified squamous epithelium.

The vomeronasal organ is not a distinct tubular structure. Anteriorly, it has several lumina (Fig. 7B). Posteriorly, the organ is better integrated and the epithelium is pseudostratified ciliated columnar. In some regions low columnar ciliated cells line the lateral wall of the organ. However, medial–lateral differentiation of epithelium cannot be considered a prominent characteristic of this organ. Epithelium, distinctly olfactory in nature, is present in the organ posteriorly. Large round nuclei characteristic of olfactory receptors are seen. Surface projections are very prominent. Terminal bars are observed at the epithelial surface. No surface investment of secretory material is observed. Secretory material is present in the lumen, especially posteriorly, but is not PAS-positive. The vomeronasal glands appear 'serous'. The organ complex is not well vascularized. Most of the venous sinuses are located posteriorly. The vomeronasal nerve is small but readily detected. Though no distinct paravomeronasal ganglion is present, ganglion cell bodies are observed. Inspection of the olfactory bulbs revealed no accessory olfactory bulbs.

Family Phyllostomatidae

Glossophaga soricina (Figs. 2H, 8A–B, 17 and 21). Though our specimen was poorly preserved, satisfactory observations were possible. The long, well-developed vomeronasal organ is compactly situated in the ventral nasal septum. The vomeronasal cartilage encircles the organ anteriorly for a short span, but the C shape predominates. Though the cartilage ends 3·9 mm posteriorly, the organ continues for 1·7 mm without any investment of the vomeronasal cartilage. However, in this region the organ receives its support medially and ventrally from the adjacent bony structures. The nasopalatine duct opens medially into the oral cavity. As in other species, an anterior process from the vomeronasal cartilage accompanies the duct. The duct joins the nasal cavity 170 μm posteriorly in the snout and 0·5 mm posterior

Fig. 6. *Rhinolophus lepidus*. A, the septal and paraseptal cartilages are integrated as a single cartilage unit, the latter enclosing the vomeronasal organ; B, note material in lumen of the vomeronasal organ and fascicle of trigeminal nerve (*V*) adjacent to the organ.

Fig. 7. *Hipposideros lankadiva*. A, in the massive ventral portion of the septum, the septal and paraseptal cartilages are continuous. Also note the thick palate and the *septal pockets* (*p*) on either side of the nasal septum and superior to the septal cartilage (*sc*). Further explanation in text; B, the multiple lumina (*lv*) of the vomeronasal organ are readily seen.

to the beginning of the nasal cavity. Slightly posterior to where the duct joins the nasal cavity is the ostium of the vomeronasal organ. The organ is initially lined with stratified squamous epithelium, but for most of its extent the organ is lined laterally with low ciliated columnar epithelium and medially with pseudostratified ciliated columnar epithelium. In some areas the cells are extremely tall while in other areas they barely rise above the basement membrane. Surface projections are visible. The basement membrane is closely applied to the vomeronasal cartilage with very little intervening connective tissue. The vomeronasal nerve is, however, present in this area, and its large fascicles are readily seen in the nasal septum. Scattered nerve cell bodies, apparently belonging to the paravomeronasal ganglion, are present. The nerve courses postero-dorsomedially on the olfactory bulb and projects to the well-developed accessory olfactory bulb (Table 2) which lies *within the substance* of the olfactory bulb. The glomeruli of the oval-shaped accessory bulb are much smaller than those of the olfactory bulb and are scattered throughout the accessory bulb. The characteristic laminar organization of the olfactory bulb is absent in the accessory bulb. These observations apply to the accessory bulbs of other species as well (Table 2) and therefore will not be repeated.

Artibeus jamaicensis (Figs. 2I, 3, 9A–B, 18 and 22). The impressively well-developed vomeronasal organ complex fills the thick anterior nasal septum. The crescent-shaped organ and its cartilage lie ventrally in the septum and, superiorly, the entire septum is occupied with the vomeronasal gland. Anteriorly, the vomeronasal cartilage completely encircles the organ for a short distance. Posteriorly, it becomes U-shaped allowing the gland ducts to enter the organ through the deficient roof. The nasopalatine ducts open as mere slits into the oral cavity immediately posterior to the medial incisors, but are of the order of 30 μm in diameter through most of their length. Though the ducts are accompanied by anterior cartilaginous processes they are not in intimate association as in some other species. This may be due to the protection afforded by the surrounding dentition and the premaxilla. The ducts are approximately 1·5 mm long and are lined with stratified squamous epithelium. The vomeronasal duct opens into the nasopalatine duct as it joins the nasal cavity 1·8 mm posterior to the beginning of the latter.

The organ is initially lined with stratified squamous epithelium but, 100 μm posterior to the ostium, pseudostratified ciliated columnar epithelium appears on the medial wall. Olfactory-like sensory cells are readily detected by their characteristic nuclei which measure 6–7 μm in diameter. Our series proved to be of such quality that even the terminal bars are clearly seen. Sensory epithelium is present on both medial and lateral walls but that of the medial wall is consistently taller. Medial–lateral differentiation, however, is present posteriorly. In the posterior half of the organ glandular tissue is present within the cartilage sheath, with the ducts entering the inferior aspect of the organ. Secretory material is seen in the lumen of the organ but is not PAS-positive, though the vomeronasal glands are predominantly 'mucous'. Vascular sinuses are mainly dorsal and lateral with respect to the organ.

The paravomeronasal ganglion is a prominent structure. The nuclei of its neurons measure 7·5–9·0 μm in diameter. It is in close association with the large vomeronasal nerve, which courses on the septum in at least two separate strands. Several intra-

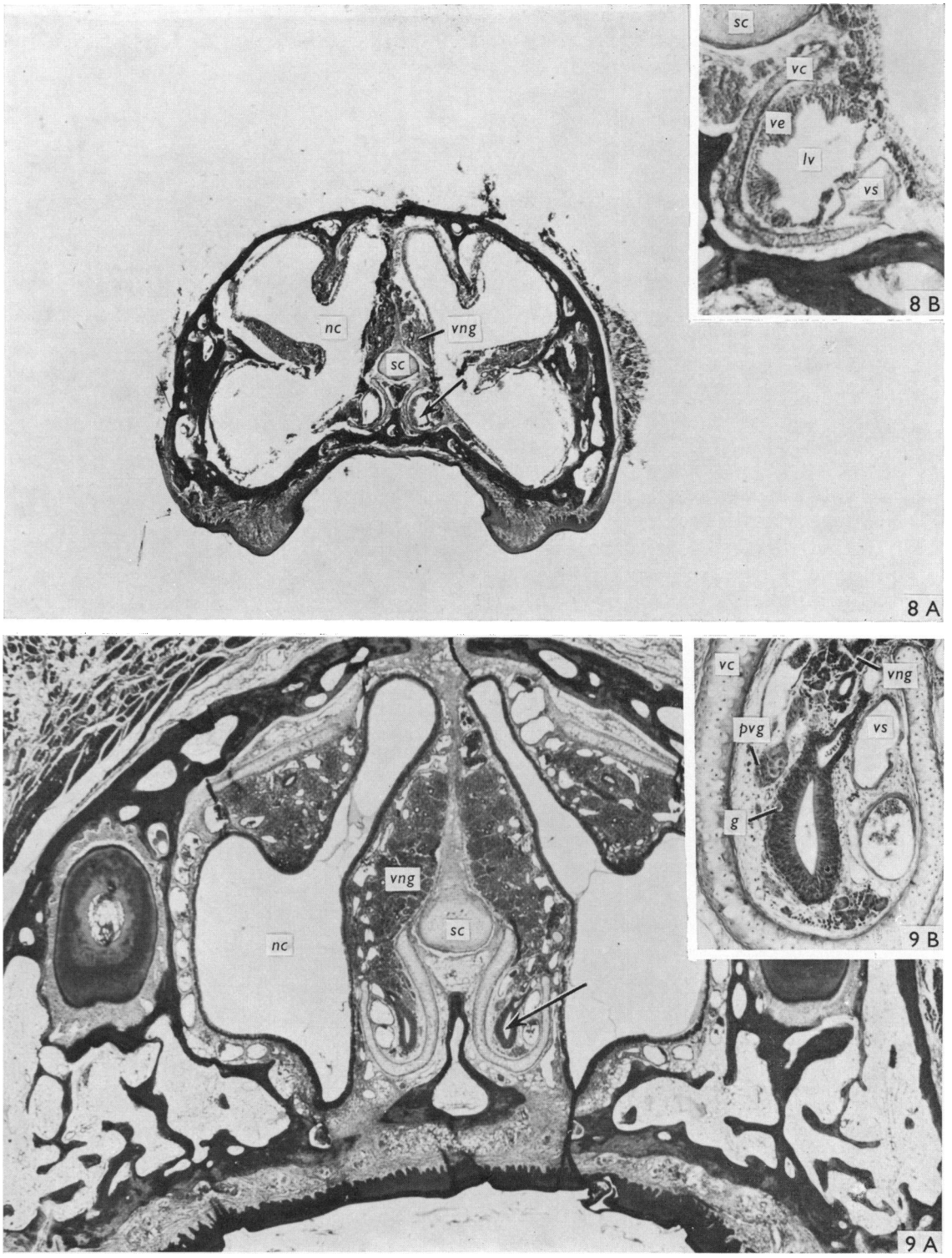

Fig. 8. *Glossophaga soricina.* A, the integument has been removed from the poorly preserved specimen; B, note the irregular contour of the vomeronasal epithelium unique to this species.

Fig. 9. *Artibeus jamaicensis.* A, prominent vomeronasal glands (*vng*) fill the septum; B, large paravomeronasal ganglion (*pvg*) and intraepithelial ganglion cell body (*g*) are clearly seen.

epithelial ganglion cell bodies are clearly seen at various levels in the epithelium The vomeronasal nerve courses dorsomedially on the olfactory bulb and enters the tear-drop-shaped accessory bulb which is located dorsolaterally on the posterior aspect of the main bulb (Figs. 3 and 22).

Desmodus rotundus (Figs. 2J, 10A–B, 15 and 20). The vampire bat possesses the most spectacular organ of all the species examined. Here, the organ complex closely fits the classical description of the mammalian vomeronasal organ (cf. Negus, 1958). The large well-developed organ is contained in a bulge in the ventral nasal septum and corresponds in shape to the dorsoventrally elongated nasal cavities. Anteriorly, the vomeronasal cartilage is a modified C shape, but for a short span (250 μm) it completely encircles the organ. Posteriorly, a hook shape predominates and the organ complex is close to the roof of the oral cavity (Figs. 10A–B). The ventral aspect of the vomeronasal cartilage is part of the structural support of the palate in this area. The crescent-shaped vomeronasal organ nearly fills its cartilage sheath. The nasopalatine ducts (40–60 μm in diameter) are nearly vertical and open anteromedially in the oral cavity posterior to the incisors. Processes from the vomeronasal cartilages accompany the ducts medially. Due to the depth of the nasal cavities the maxillae also lend lateral support to the ducts.

Unlike other species, the ostia of the organs and the nasopalatine ducts open separately in the nasal cavities. The ostium of the organ (80 μm india meter), and the bulge which the organ occupies in the anteroventral septum, can be readily observed by gross dissection, since the organ opens into the nasal cavity proper. Anteriorly, the organ is lined with respiratory epithelium. 200 μm posteriorly, tall pseudostratified ciliated columnar sensory epithelium, similar to olfactory epithelium, is seen on the medial wall of the vomeronasal organ, and predominates on this wall for the remainder of the organ. Artefactual separation of the cellular components in this glutaraldehyde-perfused specimen made further detailed observations difficult. Anteriorly, large vascular sinuses surround the organ within the cartilage, posteriorly they are lateral. Little glandular tissue is seen within the cartilage. The predominantly 'mucous' vomeronasal glands are prominent and open into the superior aspect of the organ. As might be expected, the vomeronasal nerve is large and in its course contains the well-defined paravomeronasal ganglion. The nerve enters the large well-developed accessory olfactory bulb which nearly covers the entire posterodorsal aspect of the olfactory bulb (Fig. 20).

Family Vespertilionidae. (Figs. 2K–N, 11A–B, 19 and 23)

The vomeronasal organ is absent in three of the four species investigated: *M. lucifugus*, *P. subflavus*, and *E. fuscus*. A 17·0 mm crown–rump length embryo of *M. lucifugus* also lacked the organ. Broom (1895, 1897) described the organ in

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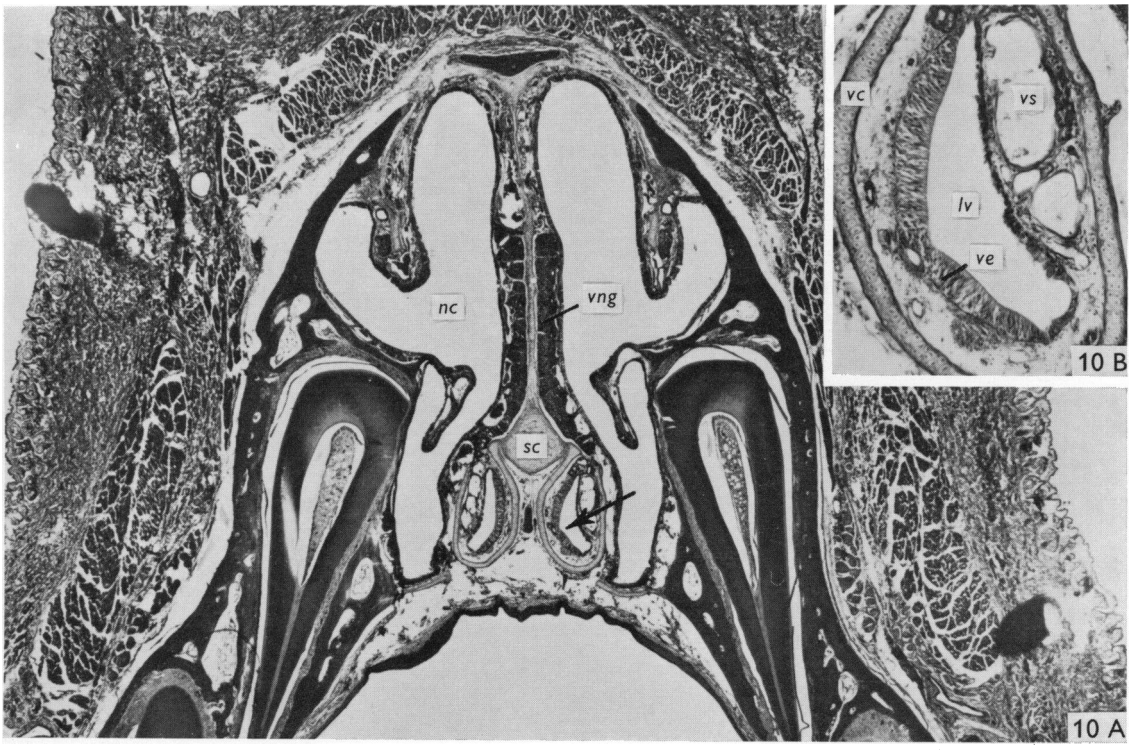
Fig. 10. *Desmodus rotundus*. A, the dorsoventrally elongated nasal septum and vomeronasal organ correspond to the similarly structured nasal cavities. Glutaraldehyde-perfused specimen; B, crescent-shaped vomeronasal organ shows medial–lateral differentiation of the epithelium.

Fig. 11. *Miniopterus schreibersi*. A, note large vomeronasal organ and low orientation of the septal cartilage; B, the vomeronasal epithelium here is closely applied to the cartilage.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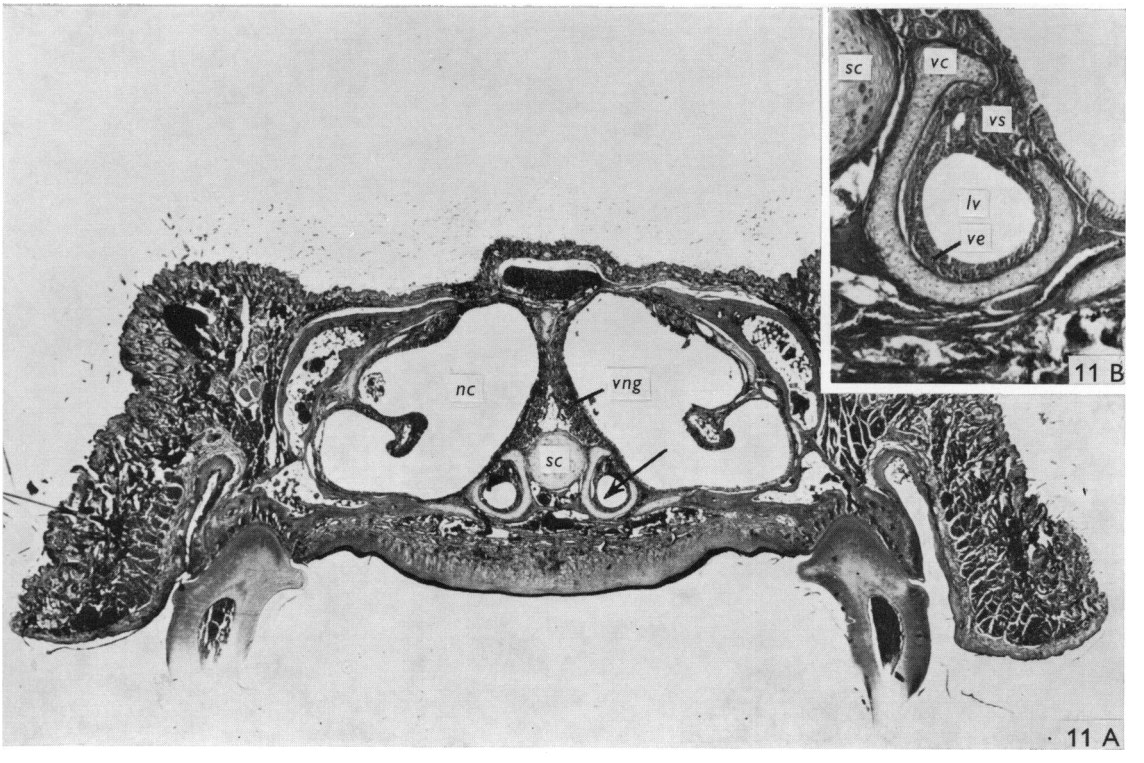
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Miniopterus schreibersi. Our investigation confirms his report and provides additional information.

Myotis lucifugus, *Eptesicus fuscus*, *Pipistrellus subflavus* (Figs. 2K–M). Patent nasopalatine ducts are present in each species and are so nearly vertical that the entire course from the nasal to the oral cavities is visible on several sections (Figs. 2K–M). All nasopalatine ducts are lined by stratified squamous epithelium and are associated with cartilage structures homologous to the cartilages of bats possessing the vomeronasal organ, though variations do exist. No accessory olfactory bulbs are present in these species, including the *Myotis* embryo.

Miniopterus schreibersi (Figs. 2N, 11A–B, 19 and 23). The nasal cavities of *Miniopterus* look distinctly similar to the other vespertilionids. Homology of the cartilage structures is evident (Figs. 2K–N), but the anterior ventral nasal septum is modified to contain the large well-developed organ (Figs. 11A–B). The round, bar-like, septal cartilage is lodged between the paired vomeronasal cartilages, and appears to have lost its dorsal extension in the nasal septum. The patent nasopalatine ducts, lined with stratified squamous epithelium, are nearly vertical. Processes from the vomeronasal cartilages and the palatine cartilages accompany the ducts. The organ begins 1·2 mm posterior to the beginning of the nasal cavities and is initially lined with stratified squamous epithelium. The organ fits so snugly into its cartilage that little room is left for any connective tissue. It has a large round lumen.

Like *Glossophaga*, this specimen was also in a poor state of preservation, making histological observations difficult. Consistent with other species, posteriorly the medial wall is lined with sensory epithelium; laterally the organ is lined with simple ciliated columnar epithelium. Some surface projections are evident. 'Serous' vomeronasal glands open into the lumen from the superior aspect of the organ. Venous sinuses are present superior to the organ. Large fascicles of the vomeronasal nerve are seen travelling between the organ epithelium and its investing cartilage. A distinct paravomeronasal ganglion was not observed. The vomeronasal nerve projects medially to the accessory bulb, which is situated posterodorsally on the olfactory bulb (Fig. 23).

DISCUSSION

Structural aspects of the chiropteran vomeronasal organ complex

Position

The vomeronasal organ has been described as located in the anteroventral portion of the nasal septum (Negus, 1958). In general, the orientation of the organ in the order Chiroptera is consistent with this description. However, in *Megaderma* and *Hipposideros* the organs are placed so low with respect to the nasal septum that they appear to be located more within the floor of the nasal cavity. In these two species the organs are separated more widely than usual. The bases of the nasal septa broaden to allow for this arrangement (Figs. 5A and 7A). The lumina of the organs in these cases are at or near the level of the floor of the nasal cavity. This design may very well serve to keep the organ as near as possible to the oral cavity. This could be of considerable advantage to *Hipposideros*, where the palate is extremely thick. Prominent vomeronasal ducts are not seen.

The ostium may open into the medial wall of the nasopalatine duct as in *Artibeus*,

directly into the nasal cavity proper as in *Desmodus*, or into an anterior extension of the nasal cavity as seen in *Rhinopoma*, but in all cases the anterior extremity of the organ is near where the nasopalatine duct joins the nasal cavity. The functional significance of this anterior position in various species has been speculated on by Negus (1956, 1958), who emphasizes the possible role of the organ in detection of food odour and flavour, and by Estes (1972), who emphasized a sexual function in detection of pheromones. Such reports epitomize a basic 'food–sex dichotomy' which seems to have arisen concerning the function of the organ. This will be discussed later. Here we should like to cite, specifically, Estes' description of the '*flehmen*' response which particularly relates to the position of the organ in the anterior snout. Citing Schneider (1930) for the use of the term, Estes defines the '*flehmen*' reflex as a distinctive grimace in response to the urine of conspecific females, exhibited by a wide spectrum of mammals including ungulates: "In a typical performance by a hoofed mammal, the animal stands open-mouthed, with head extended and elevated, while the upper lip is retracted, wrinkling the nose and baring the gum... Rapid licking movements and mouthing may accompany or follow *flehmen* in some species"... Estes further speculated that this response could be involved in the functioning of the vomeronasal organ since ... "inhalation through the incisive ducts could well serve to empty and fill the organs". Mann (1961) has observed phyllostomatid bats retracting their upper lip as though exhibiting the '*flehmen*' response. Our observations indicate that the functions suggested by Negus and Estes seem anatomically feasible in the investigated bats.

Nasopalatine ducts and nasal cartilages

The functional implications of the patency of the nasopalatine ducts have been alluded to above. We emphasize here that the nasopalatine ducts were patent, and that cartilage support to maintain this patency was apparent in each species (Fig. 2). Of necessity, our treatment of the nasal cartilages has been very brief. It is important to note, however, that homologous cartilage structures were seen in all species investigated, irrespective of the presence or absence of the organ. This homology was also noted by Broom (1897). The cartilages are basic components of the organ complex, and may play a role in the functioning of the organ (Hamlin, 1930). According to Mann (1961), contraction of the levator labii muscle in *Phyllostomus hastatus* "forced the antero-dorsal horn of Jacobson's cartilage upwards so that its antero–ventral prong opened the canalis incisivus to a maximum". He speculated that a retraction of this muscle increased the patency of the nasopalatine duct, thereby facilitating the entrance of odorous substances into the organ. Such muscular attachments on the cartilage accompanying the nasopalatine ducts were readily seen in our specimen of *Megaderma*.

Concerning the significance of the homologous cartilage structures seen in bats lacking the organ, Duval & Garnault (1895) concluded that the presence of the cartilage in the absence of the organ supported the theory that the cartilages of this region were basically supportive in nature and had only a secondary relation to the vomeronasal organ. Grosser (1900) emphatically rejected this theory. We tend to agree with Grosser that the presence of the cartilage in bats lacking the organ reflects the regression of this primitive sense organ.

The course of the nasopalatine ducts with respect to the nasal and oral cavities has been previously mentioned. The path of the ducts may be nearly vertical, as in the pteropids, the vespertilionids, and *Desmodus*; may travel over a great distance horizontally as in *Megaderma* (4 mm); or, more commonly, may travel at a dorso-ventral angle to join the nasal cavities. In our opinion, the long nasopalatine ducts of *Megaderma* and *Hipposideros* compensate for the receded nasal cavities. It is sometimes difficult to determine where the nasopalatine ducts end and the nasal cavities begin. Such is the case in *Rhinopoma*, in which the ducts open into anterior–inferior extensions of the nasal cavity.

The nasopalatine ducts in *Desmodus* are noteworthy. They are nearly vertical, but curve anteromedially in their paths (Fig. 2J) to open in close proximity to each other in a slight recess in the roof of the oral cavity posteromedial to the incisor teeth. This position emphasizes a possibility – which may apply to other species as well – that movements of the tongue may play a role in moving air through the nasopalatine ducts. This may be especially significant for the blood-lapping vampire. The close association of the vomeronasal cartilages with the roof of the oral cavity in *Desmodus* has been previously noted. This proximity could possibly have further functional implications, since tongue movements could manipulate the hook-shaped cartilages resulting in pressure on the organ itself, thus aiding in the expulsion of its contents.

Vomeronasal organ epithelium

Of the species investigated, the organ is present in eight and is best developed in the phyllostomatids *Desmodus*, *Artibeus*, and *Glossophaga*. It is intermediate in development in *Megaderma* and rudimentary in *Hipposideros*, *Rhinolophus*, and *Rhinopoma*. A prominent organ is also present in the vespertilionid *Miniopterus*, as described by Broom (1895, 1897). The best-developed organs are consistent with the generalized description of the mammalian organ (cf. Negus, 1958), even regarding the epithelium, which is differentiated into pseudostratified columnar type medially and ciliated columnar laterally. *Artibeus*, however, is the only species to have sensory epithelium all around the organ (Bhatnagar & Kallen, 1974*b*). Elsewhere the epithelium on the medial aspect is sensory, resembling olfactory epithelium. Large round

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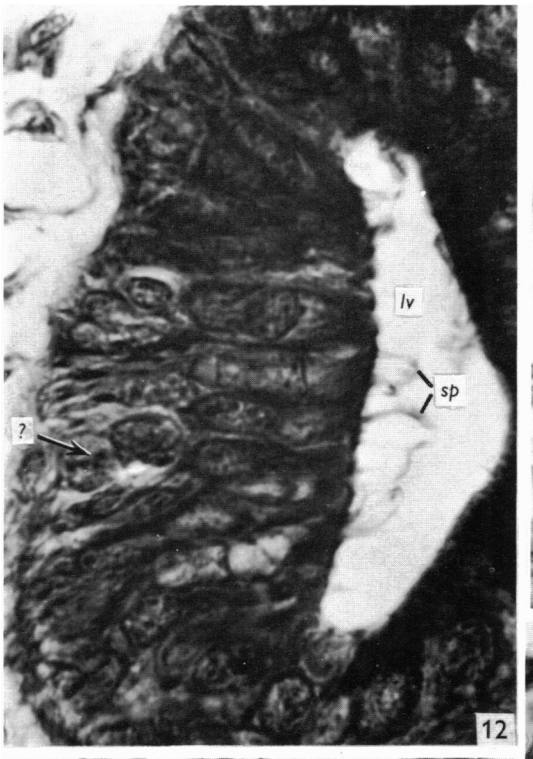
Figs. 12–15. Photomicrographs showing the vomeronasal organ epithelium. Abbreviations as in Figure 1. Arrows indicate receptor cell nuclei. Gomori trichrome. 10 μm.

Fig. 12. *Rhinopoma microphyllum*. The epithelium is tall and dense with long surface projections. × 1350.

Fig. 13. *Megaderma lyra*. Densely packed ciliated cells and a fascicle of the vomeronasal nerve are shown. × 1350.

Fig. 14. Vomeronasal organ of *Rhinolophus lepidus*. A, note glandular nature of the epithelium and secretory material in the lumen. × 550; B, lower half of section adjacent to Figure 14A stained with the periodic acid–Schiff (PAS) reaction for carbohydrates demonstrating the secretion of 'mucus' (*m*) into the lumen of the organ. × 550; C, secretory cells from an epithelial area corresponding to the area outlined in Figure 14B. Note tall surface projections. × 1350.

Fig. 15. *Desmodus rotundus*. Note surface projections and ganglion cell body. Spaces seen in the epithelium are fixation artefacts. Glutaraldehyde-perfused specimen. × 550.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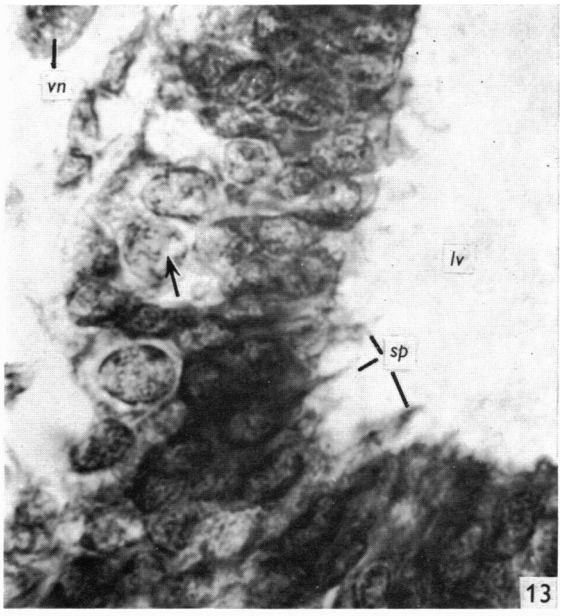
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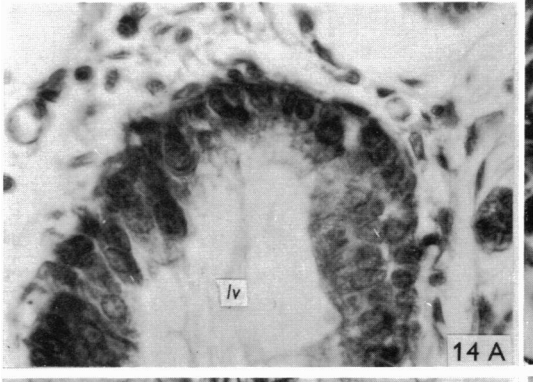
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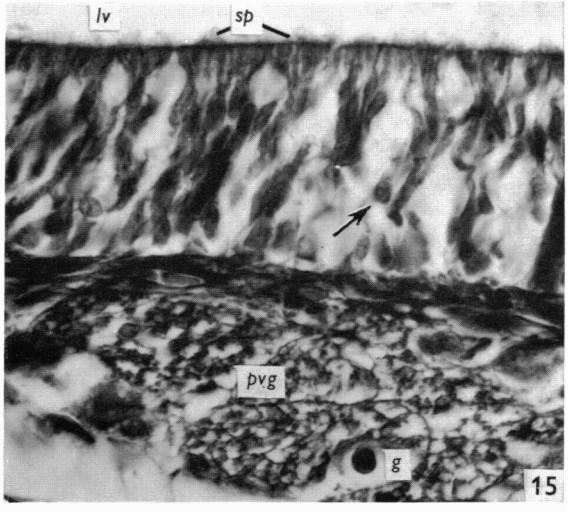
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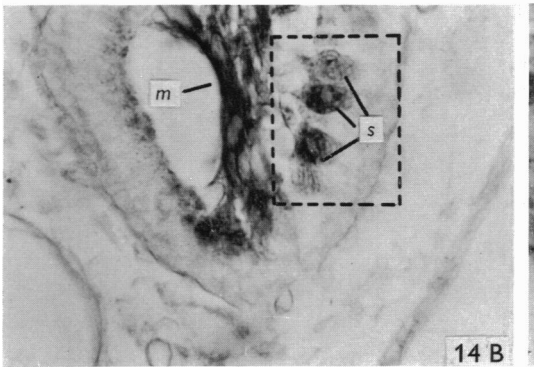
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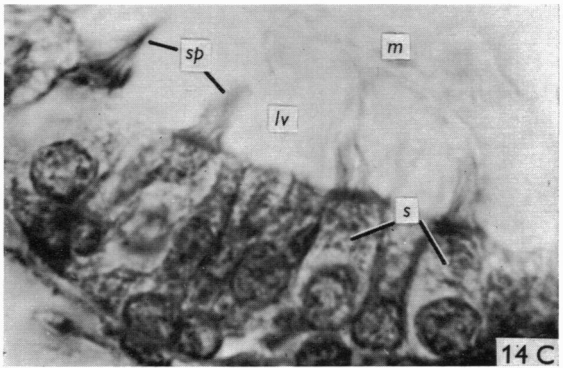
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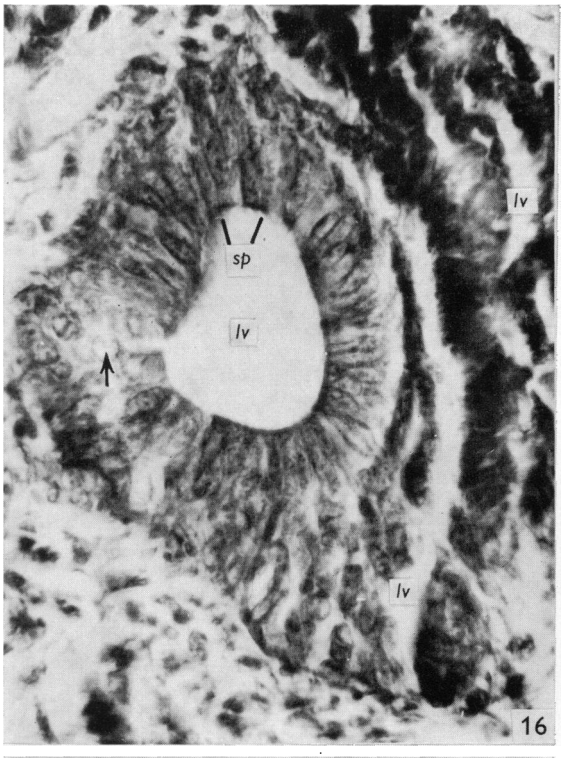
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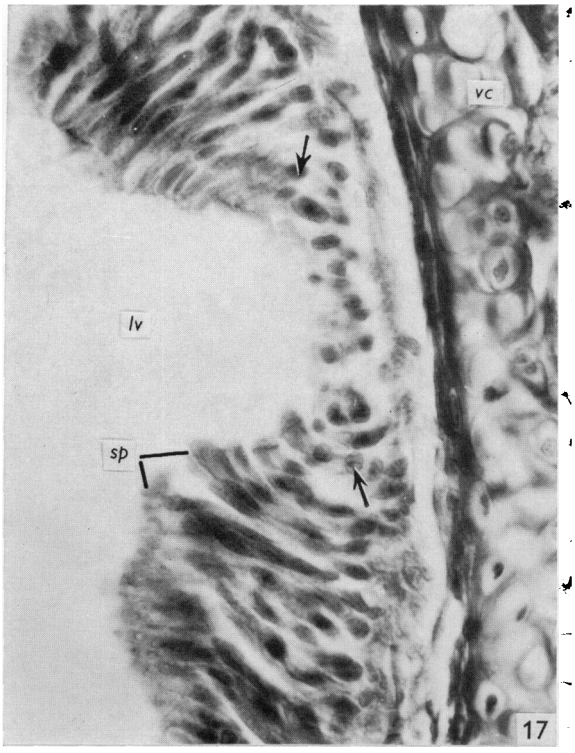
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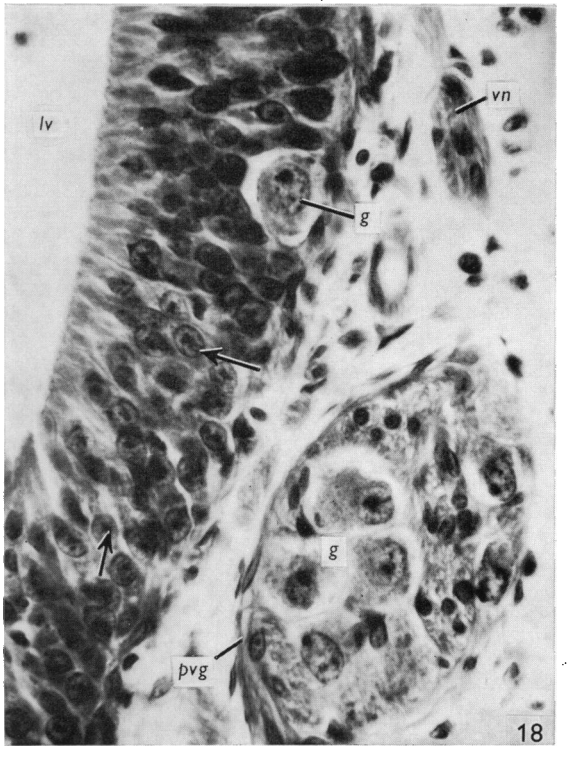
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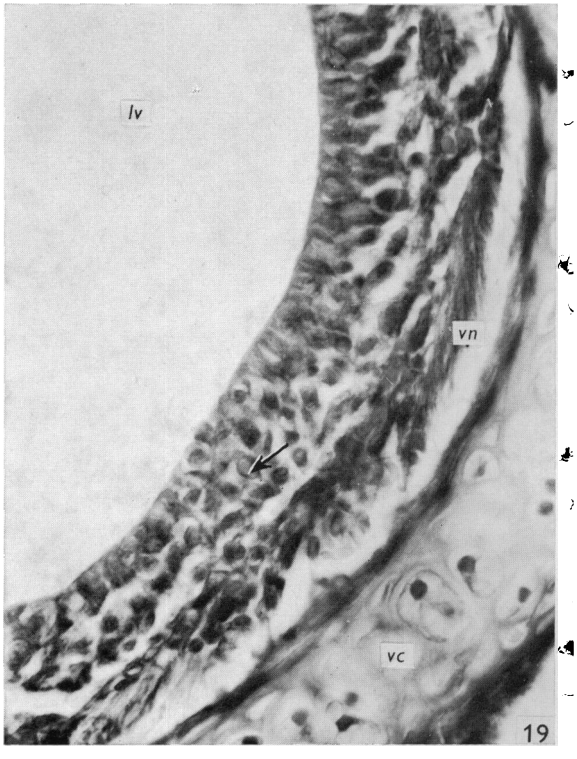
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nuclei characteristic of receptor cells, and oval nuclei of supporting cells, highly variable in diameter, are readily identified. The epithelium is tallest in phyllostomatids. In *Glossophaga* it showed such variation in height that the surface presented a highly contoured appearance in cross section. This would, perhaps, serve to increase the surface area (i.e. receptor area) of the epithelium. A multiple-lumen organ in *Hipposideros* may serve similarly.

Compared to the well-developed organs, the rudimentary organs are highly variable in epithelial characteristics. Here the question of sensory receptors is difficult to resolve. Grosser (1900) remarked that the rudimentary organs of *Rhinolophus ferrum-equinum* and *R. hipposideros* were not innervated. Our present observations do not contradict him, but the subject deserves further study in the light of the apparently innervated rudimentary organs observed in this study. The question of the presence and absence of surface projections in the vomeronasal epithelium has been debated for different mammals (see discussion by Bhatnagar & Kallen, 1974*b*). In our chiropteran organs we have consistently noted tall surface projections. Presence of secretory material in the organ lumen is a characteristic feature.

This study has revealed previously undescribed *septal pockets* on each side of the nasal septum superior to the vomeronasal organ. In *Artibeus*, Bhatnagar & Kallen (1974*b*) have described septal flaps in a similar location. They speculated that these flaps may protect the glands from back-up of secretions due to pressures created by nasophonation. Along these lines, the septal pockets of nasophonating *Hipposideros* (Novick, 1958) would supply even greater protection to the glands, which are prominent beneath their medial walls and empty into the vomeronasal organ. These pockets, located directly in the path of air currents, would seem unavoidably to have some effect on the phonation itself. These cavities and their tightly stretched lateral walls could supply a variable degree of resonance to the nasal sounds emitted by this bat. Could the septal flaps function similarly in *Artibeus*? Further studies may resolve such questions.

An olfactory patch, or septal olfactory organ, lying like an island in the respiratory region bilaterally on the nasal septum at the entrance to the nasopharyngeal meatus, has been described in several rodents (cf. Adams & McFarland, 1971; Bojsen-Møller, 1975). Even though we do not now know whether olfactory receptors line these septal pockets, we have noticed many nuclei characteristic of olfactory receptors in its epithelium. Should our observations be confirmed later on, these pockets could be considered analogous to the septal olfactory organ. It may not be

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Figs. 16–19. Photomicrographs showing the vomeronasal organ epithelium. Abbreviations as in Figure 1. Arrows indicate receptor cell nuclei. Gomori trichrome. 10 μm.

Fig. 16. *Hipposideros lankadiva*. Epithelium shows densely packed cells with surface projections. Note multiple lumina (*lv*). × 600.

Fig. 17. *Glossophaga soricina*. Irregular thickness of the epithelium and presence of surface projections are apparent. The epithelium is closely associated with the cartilage. × 600.

Fig. 18. *Artibeus jamaicensis*. Note the vomeronasal nerve, the paravomeronasal ganglion, and an intraepithelial ganglion cell body. Surface projections, although present, are not seen here. × 600.

Fig. 19. *Miniopterus schreibersi*. The epithelium is in close association with the vomeronasal cartilage. Note the vomeronasal nerve. × 600.

surprising to find scattered olfactory receptors in this area, since olfactory epithelium begins on the nasal septum in this region only 400 μm posterior to the pockets.

Glands

The predominantly 'serous' vomeronasal glands are located in the nasal septum superior to the vomeronasal organ. They are present in all species except *Rhinopoma*, where the organ is quite rudimentary. Since secretions from the olfactory glands have been implicated in the mechanism of olfaction (cf. Moulton & Beidler, 1967), lack of vomeronasal glands in *Rhinopoma* is consistent with the rudimentary nature of its vomeronasal organ. Significance of the glands is emphasized by the observation that well-developed organs possess well-developed glands. In *Artibeus* and *Glossophaga* the glands fill the septal region. The wide lumina of the ducts do not open through the epithelium but seem to be continuous with the epithelium of the organ (Fig. 1, *dvng*). Secretions are evident in the lumina of some organs, most notably in *Rhinolophus*. Here the organ is filled with mucus from the epithelium which includes goblet cells and 'mucus'-producing secretory cells with prominent surface projections (Fig. 14C). Goblet cells have also been reported in the anterior organ epithelium of *Artibeus* (Bhatnagar & Kallen, 1974*b*). The functional implications of the epithelium of the rudimentary organ in *Rhinolophus* remain to be determined. It would be useful to know whether the vomeronasal glands more closely resemble olfactory glands (of Bowman) or the salivary gland 'pseudomucous' and 'pseudoserous' cells of Wimsatt (1956).

Vascular sinuses

Vascular sinuses were seen accompanying the organ in all species. However, their degree of development directly paralleled that of the organ. In the well-developed organs the sinuses were dorsolaterally oriented (e.g. *Glossophaga* and *Desmodus*). This orientation would allow engorgement of these vessels to compress the organ against the medial wall of the cartilage and facilitate emptying of its contents. The crescent-shaped lumen in *Desmodus* (Fig. 10B) appears consistent with this thought. Broman (1920) and Hamlin (1930) proposed that varying blood pressure in the region might result in a pumping action of the organ within its cartilaginous capsule. While this may well be true for the species having well-developed organs, it is questionable for the remaining species where the organ is not prominent. Hamlin further suggested that the elastic tissue associated with the organ helps suck in substances when sinuses are emptied.

Neural structures

The degree of development of the vomeronasal nerve, the paravomeronasal ganglion, and the accessory olfactory bulb directly parallels that of the organ in general. Thus, in the rudimentary organs, examination of these structures may help in assessing the degree of sensory importance of the organ.

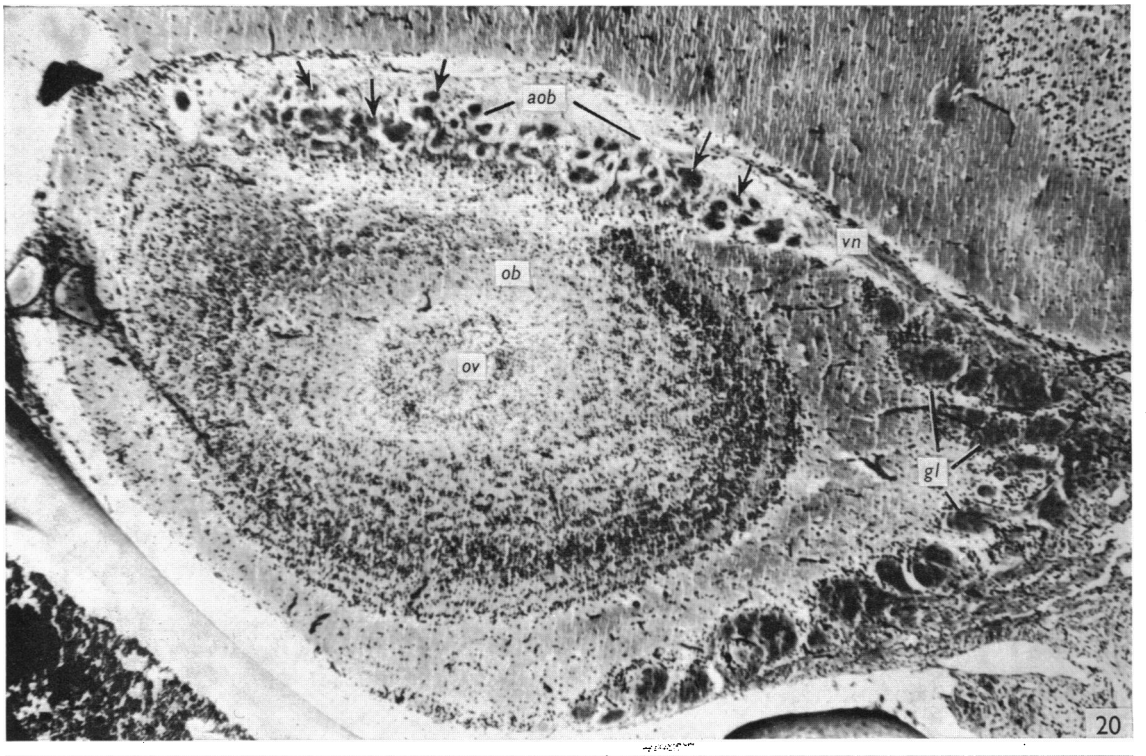
In all species, with the exception of *Rhinolophus*, it was possible to identify the vomeronasal nerve. In well-defined bundles it followed a gradually ascending course towards the accessory bulb (as in *Desmodus*, *Artibeus*, *Glossophaga*, and *Miniopterus*). Mann (1961) described a myelinated vomeronasal nerve in *Phyllostomus*

hastatus, *Artibeus jamaicensis*, *Glossophaga soricina*, and *Desmodus rotundus*. In all the species examined by us we have found this nerve to be unmyelinated. This is consistent with the unmyelinated nature of the vomeronasal nerve in other mammals (cf. Suthers, 1970; Adrian, 1955).

Ganglion cell bodies associated with the vomeronasal nerve are well known in other mammals (Kolmer, 1927). An impressive and discrete paravomeronasal ganglion was described in *Artibeus* by Bhatnagar & Kallen (1974*b*). Similar well-defined ganglia were also observed in *Desmodus* and *Rhinopoma*. In *Hipposideros* and *Glossophaga* scattered neurons, apparently belonging to the paravomeronasal ganglia, were observed in association with the vomeronasal nerve. A few neurons were also observed in *Rhinolophus* in association with the organ. According to Johnston (1913), this ganglion consists of bipolar ganglion cells resembling cerebrospinal ganglion cells giving rise to the nervus terminalis (also see discussion by Grüneberg, 1973). Intraepithelial ganglion cell bodies such as those reported by Bhatnagar & Kallen (1974*b*) were observed by us only in *Artibeus*. These intraepithelial ganglion cell bodies presumably belong to the nervus terminalis system supplying free nerve endings to the vomeronasal organ epithelium (Bojsen-Møller, 1975). This is analogous to the similar distribution of free nerve endings to the olfactory epithelium from the trigeminal nerve. Our inability to observe ganglionic structures in the otherwise very well-developed organ complex of *Miniopterus* could be due to poor preservation of the specimen.

Although by 1961 it was common knowledge that the mammalian vomeronasal organ projects to the accessory olfactory bulb (McCotter, 1912; Humphrey, 1936; Schneider, 1957), apparently the first chiropteran accessory bulbs were not described until Mann's (1961) report on *Glossophaga soricina*, *Artibeus jamaicensis*, *Desmodus rotundus*, and *Phyllostomus hastatus*. Bhatnagar & Kallen (1974*b*) also described the accessory bulb in *A. jamaicensis*. Schneider (1957, 1966) investigated all these bats and others, including *Rousettus aegyptiacus*, yet did not observe the accessory olfactory bulbs. Humphrey (1936) denied the presence of this structure in *Tadarida mexicana*. Broom (1895) described the vomeronasal organ in *Miniopterus schreibersi*, but did not investigate its projections. Even though Schneider reported no accessory bulb in *Miniopterus*, we have discovered a well-developed accessory formation. Of our four species *Desmodus* possessed the largest accessory bulb, almost occupying the entire dorsal aspect of the large olfactory bulb. Mann (1963) noticed no variation as to the caudal and lateral position of the accessory olfactory bulb on the main bulb in phyllostomatids. We found the accessory bulb in *Artibeus* consistent with his description, but in the other species variations of a posterodorsal position were noted. Furthermore, in striking contrast to his description of the histological elements "arranged from the peripheria towards the center", we found no orderly laminar organization in any of the accessory bulbs, presumably due to their small size (also see Stephan, 1965).

Though accessory olfactory bulbs were not identified in bats possessing rudimentary organs, other indications of sensory function of the organ such as the presence of vomeronasal nerves, paravomeronasal ganglia, and receptor-like cells were seen. Further investigations to trace the projections from these rudimentary structures would be of interest.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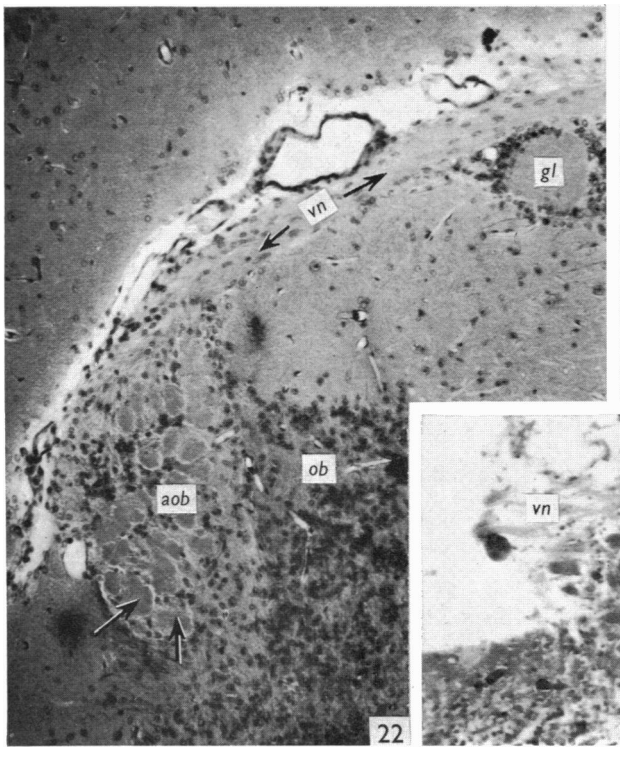
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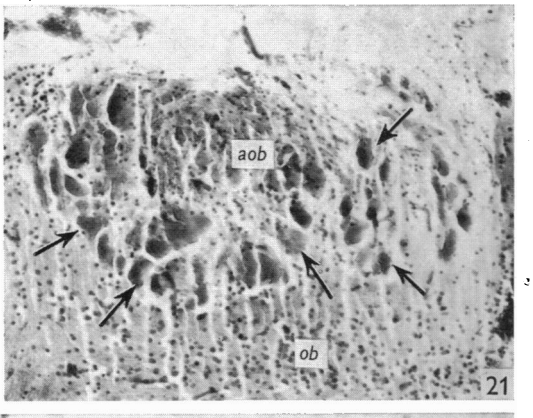
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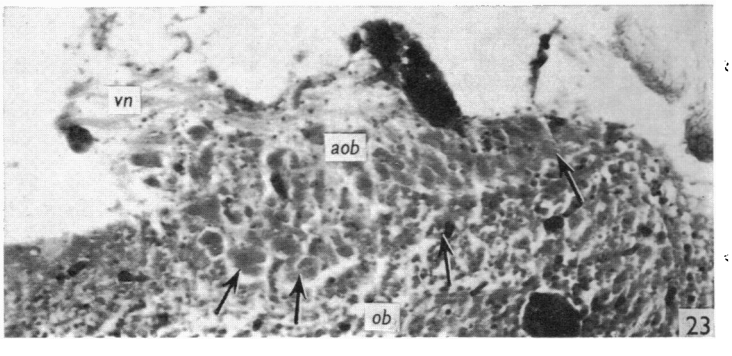
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Functional considerations

Earlier observations (cf. Negus, 1956) on the vomeronasal organ led to speculation that it is involved in detection of odour and flavour of food. More recently, investigations of neural connections of the organ (Winans & Scalia, 1970; Raisman, 1972; Barber & Raisman, 1974) and considerations of the effects of pheromones on reproduction (Estes, 1972; Whitten & Champlin, 1973), have led to increased emphasis on a sexual role for the vomeronasal organ. Feeding behaviour and sexual behaviour are most frequently involved in discussions of the organ's function. It seems, however, that a dichotomy of views has evolved from such considerations, to the extent that the two functions are often regarded as mutually exclusive. The vomeronasal organ is, in fact, basically understood to be a component of the accessory system of olfaction (see review by Scalia & Winans, 1975). If the function of the organ is considered in this broader context of a dual olfactory system, the dichotomy should be resolved, for, just as different functions of the main olfactory system are emphasized in different species (Whitten & Champlin, 1973), the accessory olfactory system could also function in sexual behaviour in one species while functioning in feeding in another, and in yet others both aspects may be involved. Since the accessory olfactory bulb projects to neural areas that are involved in both reproductive and feeding behaviour (Winans & Scalia, 1970; Raisman, 1972), it is not unlikely that these neural areas may differ in relative significance in different animals. The first definitive report on the function of the vomeronasal organ (Powers & Winans, 1975) has established a role for it in the mating behaviour of the male hamster. To what degree this role applies in other mammalian species remains to be seen.

Since knowledge of sexual behaviour in bats is limited, it is not possible to speculate adequately on the role of the vomeronasal organ in this regard. Its role in food-seeking behaviour is, perhaps, more apparent. The bats with the best developed vomeronasal organs are those most dependent on a well-developed olfactory sense, i.e. the frugivorous, nectivorous, and sanguivorous phyllostomatids. In the insectivorous bats, which rely more heavily on their sense of echolocation (Griffin, 1974), the organ is either rudimentary or absent. The insectivorous *Miniopterus*, which has a prominent vomeronasal organ, seems to be an exception. It should be noted, however, that it has proportionately larger olfactory bulbs than its close relatives, such as *Pipistrellus*, *Eptesicus*, and *Myotis* (Table 2). These observations point to

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Figs. 20–23. Photomicrographs of frontal sections through the posterior aspect of the olfactory bulbs showing the accessory olfactory bulbs. Bulbs of the right side are shown except in Figure 23 where the left one is used. 10 μm.

Fig. 20. *Desmodus rotundus*. The accessory bulb occupies nearly the entire dorsal aspect of the olfactory bulb. Protargol silver. × 75.

Fig. 21. *Glossophaga soricina*. Gomori trichrome. × 130.

Fig. 22. *Artibeus jamaicensis*. Gomori trichrome. × 130.

Fig. 23. *Miniopterus schreibersi*. Gomori trichrome. × 180.

Abbreviations: *aob*, accessory olfactory bulb; *gl*, glomerulus; *ob*, olfactory bulb; *ov*, olfactory ventricle; *vn*, vomeronasal nerve. Arrows indicate accessory olfactory glomeruli.

some yet unknown behaviour patterns of *Miniopterus* where use of this prominent dual olfactory sense could be made. It is no coincidence that the vomeronasal organ in *Megaderma* shows an intermediate state of development. This appears consistent with the feeding behaviour of the carnivorous *Megaderma* (Walker, 1975), in which this species could benefit from an olfactory sensitivity intermediate between that of insectivorous and frugivorous species.

In considering the feeding behaviour of the vampire bat perhaps we can construct a model which may help resolve the 'food–sex dichotomy'. Vampire bats are known to void urine profusely immediately after feeding (Wimsatt & Guerriere, 1962; Greenhall, Schmidt & Lopez-Forment, 1971). They have been observed to urinate on their prey and to return to the same animal repeatedly for feeding (Turner, 1975). Since the mammalian vomeronasal organ has been implicated in response to pheromones in urine (Estes, 1972; Whitten & Champlin, 1973), the accessory olfactory system could be the means through which the vampire bat relocates its marked prey. Thus the typically sexual response to urine would be modified to facilitate food-seeking. It is further interesting to speculate that the '*flehmen*' response might be exhibited by vampires during their feeding process. Schmidt & Greenhall (1971, Fig. 5) describe and include a photograph of *Desmodus* exhibiting a typical sniffing posture during olfactory orientation which has apparent likeness to '*flehmen*'. Mann (1961) has also observed retraction of the upper lip in a characteristic manner in sexually motivated phyllostomatids. One of us (K.P.B.) has observed *A. jamaicensis* sniffing on many occasions while locating food (unpublished observations).

Bruner (1914) proposed 'monosmesis' (a condition in which the olfactory organ is used to test only the external medium) and 'diosmesis' (where both the external medium and the contents of the oral cavity are tested) as terms to distinguish two separate types of respiration in Amphibia. He further explained that 'monosmatic' animals had simple olfactory systems with the vomeronasal organ lacking, whereas, 'diosmatic' animals had a complex olfactory system in addition to a vomeronasal organ. Since the vomeronasal organ is generally accepted as an accessory olfactory organ, we would like to propose that the terms 'diosmatic' and 'monosmatic' be brought into general use to characterize animals with or without the organ respectively. We feel that these concise terms should greatly facilitate reports dealing with the vomeronasal organ.

Of the six families investigated, all except the family Pteropodidae were found to have at least one diosmatic member. Absence of the organ in the megachiropterans is remarkable in the light of the otherwise well-developed olfactory organ in these nectivorous and frugivorous bats (Stephan, Pirlot & Schneider, 1974). It should be noted, however, that only a few of the 150 Recent species of this suborder (Koopman & Jones, 1970) have been investigated. Even though *Rousettus*, which is said to be the most primitive member of this group (Jones & Genoways, 1970), lacks the organ (this study), and the accessory olfactory bulb (Schneider, 1966), it is possible that the organ may yet be found in a megachiropteran species. In the final analysis, should megachiropterans prove to be exclusively monosmatic, then this would be yet another significant disparity between the two suborders.

The presence of a vomeronasal organ, though rudimentary, in the primitive

microchiropteran *Rhinopoma*, seems logical in light of the primitive nature of the organ itself. *Rhinopoma*, *Megaderma*, *Hipposideros*, and *Rhinolophus* are insectivorous bats, and are heavily dependent on echolocation (Novick, 1958). These facts, in addition to the insectivorous *Miniopterus* being diosmatic, tempt us to speculate that the organ may be more widespread within, and characteristic of, the suborder Microchiroptera than has previously been suspected. Investigations of many other species are needed to resolve this question.

It seems ironic that the first investigations on the chiropteran vomeronasal organ were conducted on pteropids and vespertilionids in which the vomeronasal organ is characteristically absent, and that this condition was taken to be representative of the entire order. Previous investigations, and the present report, strongly indicate that the vomeronasal organ is a characteristic structure in several chiropteran families, and that it is most developed in the family Phyllostomatidae. Further studies along these lines should help to establish the functional morphology of the vomeronasal organ complex in the order Chiroptera.

SUMMARY

The morphology of the vomeronasal organ complex was histologically described in eight out of fourteen chiropteran species investigated. Of the six families examined, all except the family Pteropodidae (suborder Megachiroptera) were found to have at least one member possessing the organ. The organ is best developed in phyllostomatids. It is absent in vespertilionids (including a *Myotis* embryo) except in *Miniopterus*. An accessory olfactory bulb is reported for the first time in the latter. The organ is described for the first time in *Rhinopoma*, *Megaderma*, and *Hipposideros*. The organ in *Rhinolophus* is also described. Homologous anterior nasal cartilages and patent nasopalatine ducts are present in all species. The organ occupies the anterior ventral nasal septum region. In *Megaderma* and *Hipposideros* it is level with the nasal cavity floor. Areas of epithelium similar to olfactory epithelium have been observed in some organs. Epithelia, vascular sinuses, vomeronasal nerves, paravomeronasal ganglia, accessory olfactory bulbs, and vomeronasal glands have been investigated. In bats with regressed or rudimentary organs (*Megaderma*, *Rhinopoma*, *Rhinolophus*, *Hipposideros*) accessory olfactory bulbs could not be identified. Thus, presence of the organ does not necessarily indicate presence of the accessory olfactory bulb. Septal pockets located superior to the organ complex and lined with pseudostratified columnar epithelium are described in *Hipposideros* and may play a part in nasophonation. A unique role is proposed for the organ in the feeding behaviour of *Desmodus*. The desirability of extending the useful terms 'diosmatic' and 'monosmatic' to all vertebrates in reference to their respective possession or lack of the vomeronasal organ is suggested.

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