J. Anat. (1965), **99**, 4. pp. 791–813
With 9 figures
Printed in Great Britain

The central olfactory connexions

BY T. P. S. POWELL, W. M. COWAN AND G. RAISMAN
Department of Human Anatomy, University of Oxford

INTRODUCTION

Following the studies of Adrian (1942), Rose & Woolsey (1943) and Fox, McKinley & Magoun (1944), all of whom used the evoked potential method, the central connexions of the olfactory bulb were investigated neurohistologically by several workers using the Glees method (Le Gros Clark & Meyer, 1947; Meyer & Allison, 1949; Allison, 1953*a*; Adey, 1953). Perhaps the most important contribution of these studies was to limit the projection of the olfactory bulb to the anterior olfactory nucleus, part of the olfactory tubercle, the pyriform cortex and certain of the amygdaloid nuclei, and to confirm that the entorhinal area and hippocampus are not in receipt of direct afferents from the olfactory bulb. More recent work, using the Nauta technique, has in general, substantiated these findings, but at the same time has raised several questions regarding the relationship of the anterior limb of the anterior commissure to this projection. For example, Lohman (1963) has presented considerable evidence to indicate that this bundle arises in the anterior olfactory nucleus rather than in the main or accessory olfactory bulbs. Other workers, including Cragg (1961*a*), Sanders-Woudstra (1961) and Powell, Cowan & Raisman (1963) have similarly questioned the existence of a projection, through the anterior limb of the anterior commissure, to the central nucleus of the amygdala and its supposed continuation, the bed nucleus of the stria terminalis.

In view of the general agreement that the pyriform cortex represents the major site of termination of the lateral olfactory tract it is perhaps surprising that so little is known of the efferent connexions of this area. Apart from the work of Lundberg (1960) who included this area in his study of the projection of the cerebral cortex upon the hypothalamus and of Cragg (1961*a*) who showed a projection from the pyriform cortex to the entorhinal area, there does not appear to have been any systematic study of the efferent connexions of the pyriform cortex. Such a study is clearly called for in view of the widespread interest in the influence of olfaction upon reproductive mechanisms (cf. Parkes & Bruce, 1961) and affective behaviour (Green, Clemente & De Groot, 1957). Whether these effects are mediated through the projection to the hypothalamus of the stria terminalis, which Cajal (1911) considered to be an important central olfactory projection pathway and to arise from the pyriform cortex, is not known as this opinion appears to have been largely overlooked in recent discussions of olfactory connexions.

Recent electrophysiological studies of the olfactory bulb following stimulation of the anterior commissure and the lateral olfactory tract (Kerr & Hagbarth, 1955; Green, Mancia & Von Baumgarten, 1962; Yamamoto, Yamamoto & Iwama, 1963; Phillips, Powell & Shepherd, 1963) have raised many problems of anatomical interest; for example, whether the axons of mitral and tufted cells have a differential

projection into the anterior commissure and lateral olfactory tract, and whether there are centrifugal fibres in the latter as described by Cajal (1911). With the exception of Cragg (1962) and Powell & Cowan (1963) the existence of such ipsilateral centrifugal fibres to the olfactory bulb has either been denied or overlooked.

In the present study the Nauta technique has been used to determine the central connexions of the olfactory bulb, the efferent projection of the pyriform cortex and the course and termination of centrifugal fibres to the bulb. The investigations of these three fibre systems have been included in one paper because of the obvious interrelationships between them. For convenience, however, the paper is divided into three sections in each of which the experimental results are described and discussed.

MATERIAL AND METHODS

For the first part of this study the olfactory bulb was removed in each of seven rats and in a further eighteen animals partial lesions of the bulb were made. For the investigation of the efferent connexions of the pyriform cortex, lesions were placed in and around this area in eighteen rats. For all operations the rats were anaesthetized with open ether. The olfactory bulb lesions were produced either by suction or with a fine needle through a small trephine hole in the overlying bone, while the pyriform cortex was approached through the infratemporal fossa in the manner described below. After a survival period of 5–7 ds, the animals were again anaesthetized and were perfused with saline and 10 % formalin. The brains were prepared according to either the conventional Nauta (1957) technique or to the modification of this method for paraffin sections (Nauta & Gygax, 1951). The majority of the brains were cut in the coronal plane, but some brains with each type of lesion were cut in the sagittal plane. The frozen sections were cut at 25 μm and the paraffin sections at 15 μm. For all brains a regular 1 in 5 series was stained.

As it was necessary to develop a subtemporal approach to the ventral surface of the brain so as to allow lesions to be placed in the pyriform cortex without concomitant damage to other parts of the cerebral hemisphere, and as this approach gives adequate access to other structures on the inferior surface of the brain, such as the median eminence of the hypothalamus, the operative details are given at some length.

With the head shaved and the animal lying on its side, a curved incision is made from a point on the dorsal surface of the head midway between the ears to a point just below and behind the prominent genal vibrissa lying approximately midway between the lateral angle of the palpebral fissure and the external auditory meatus (Fig. 1). The whole thickness of the scalp is incised so that the frontalis and platysma muscles are retracted with the skin. This exposes the temporal fascia overlying the temporalis muscle, together with the temporal and zygomatic branches of the VIIth cranial nerve, the lacrimal gland and the superficial temporal vessels. The attachment of the temporal fascia to the zygomatic arch is incised and the inferior and posterior borders of the temporal muscle mobilized. The muscle can now be elevated from the temporal bone as far dorsally as the temporal crest, the insertion of the muscle into the coronoid process of the mandible divided and the anterior border separated from the orbital fascia, care being taken to prevent bleeding from the

orbital venous sinuses. The periosteum over the zygomatic arch is stripped, and the origin of the masseter from its lower border is detached. Using fine pointed bone forceps most of the zygomatic arch can be removed leaving the temporomandibular joint clearly visible. The most anterior part of the joint capsule is incised to permit gentle dislocation of the head of the mandible in a posterior and inferior direction;

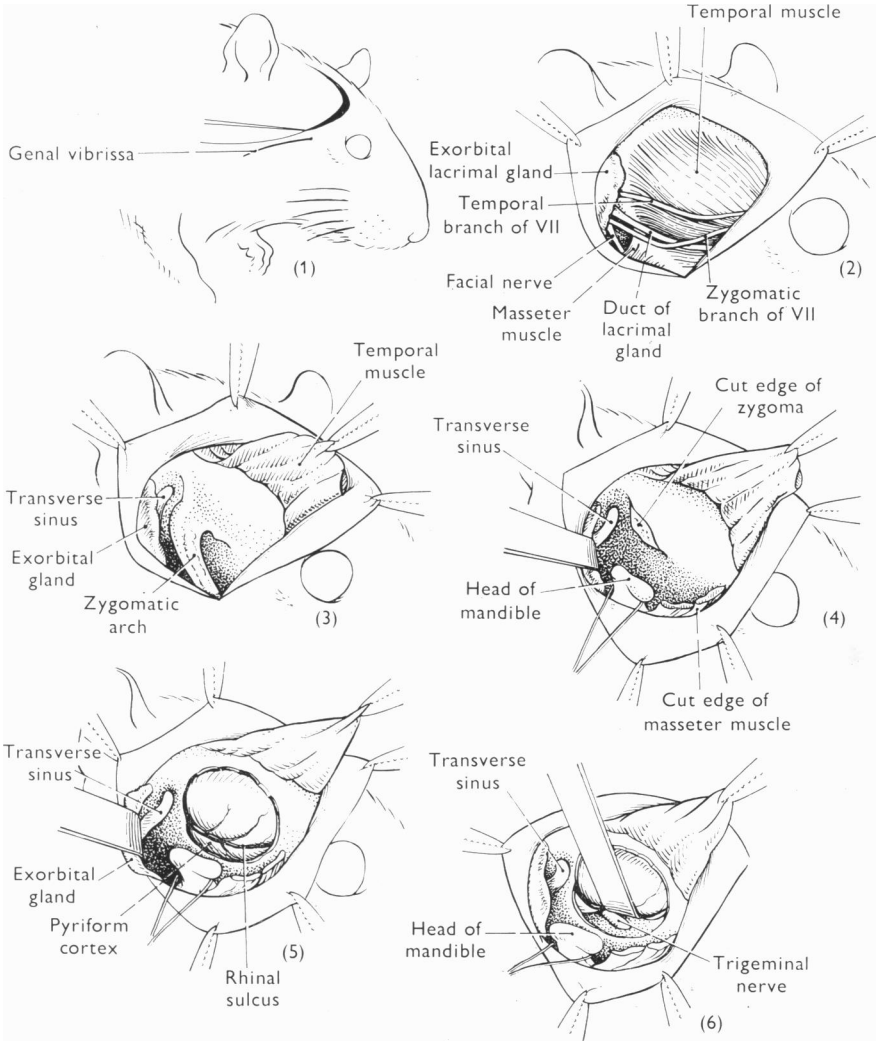

Fig. 1. Diagram to show the successive stages in the exposure of the pyriform cortex by the subtemporal approach described in the text.

great care is required at this stage to prevent tearing of the posterior part of the capsule of the joint in which the important articular tributary of the transverse venous sinus lies embedded. The retraction of the mandible lays bare a large area of skull below the cut edge of the zygomatic process. It has been found that the line of attachment of the zygomatic process coincides with the rhinal sulcus on the brain,

and if the bone ventral to this line only is removed, the pyriform cortex can be approached without rendering the neocortex liable to damage by exposure. The area of bone outlined in Fig. 1 (4) (which overlies the greater part of the pyriform cortex) is best removed by means of a dental drill and steel burrs of varying size. Little or no bleeding occurs if the bone over the whole area is reduced to almost paper thickness, and then removed piecemeal with a fine hook and forceps. The dura is opened with a curved needle and ground watchmakers forceps, revealing the entire mediolateral extent of the pyriform cortex, from the small vein in the rhinal sulcus laterally to the conspicuous lateral olfactory tract medially.

In order to reach more medial regions of the lower surface of the brain it is necessary to widen the bone removal dorsally to the level of the temporal crest and medially to the edge of the trigeminal nerve. After opening the skull and dura, as described above, the brain is gently elevated with a fine curved retractor (Fig. 1 (6)) until the divisions of the trigeminal nerve can be seen emerging from the ganglion. At this stage the head of the animal is rotated, and, with the aid of a dissecting microscope the vascular median eminence can be recognized postero-medial to the trigeminal ganglion. It is advisable that the retraction of the hemisphere be intermittent to minimize ischaemic damage to the neocortex.

At the end of the operation the head of the mandible slips back into the articular fossa, and the exposed area of brain and skull are covered by suturing the temporal muscle to the masseter muscle. Finally, the skin is closed by a continuous suture. Postoperative recovery is usually uneventful.

The essential advantage of this subtemporal approach is that without any special animal or head-holding apparatus the inferior surface of the brain can be exposed from below rather than laterally. Experience has shown that this technique permits a more ready exposure of the hypothalamus and probably results in less incidental damage to the rest of the cerebral hemisphere than other techniques at present available. For experiments in which selective lesions are to be placed in the pyriform cortex this approach has the added advantage that the region medial and inferior to the rhinal sulcus can be seen without exposing the overlying neocortex. The preservation of the temporalis muscle is also an advantage.

I. THE EFFERENT CONNEXIONS OF THE OLFACTORY BULB

Results

Of eight brains with partial lesions restricted to the olfactory bulb, Expt. P 63 will be described. In this case the rostrodorsal part of the main olfactory bulb has been removed without injury to the accessory bulb or any part of the anterior olfactory nucleus (Fig. 2).

In the sagittal sections of this brain there is a good deal of fibre fragmentation throughout the remaining parts of the main olfactory bulb and in the fibrous lamina beneath the accessory bulb. At the caudal end of the bulb coarse fibre degeneration can be traced into the lateral olfactory tract and through this bundle to the anterior olfactory nucleus, the olfactory tubercle, the pyriform cortex and amygdala. The anterior limb of the anterior commissure, which in sagittal sections can be seen over a considerable part of its antero-posterior extent, is free of degeneration. The

degeneration in the lateral olfactory tract is very superficial in its distribution, being virtually confined to the molecular layer of the affected structures.

Of the various subdivisions of the anterior olfactory nucleus degeneration is only

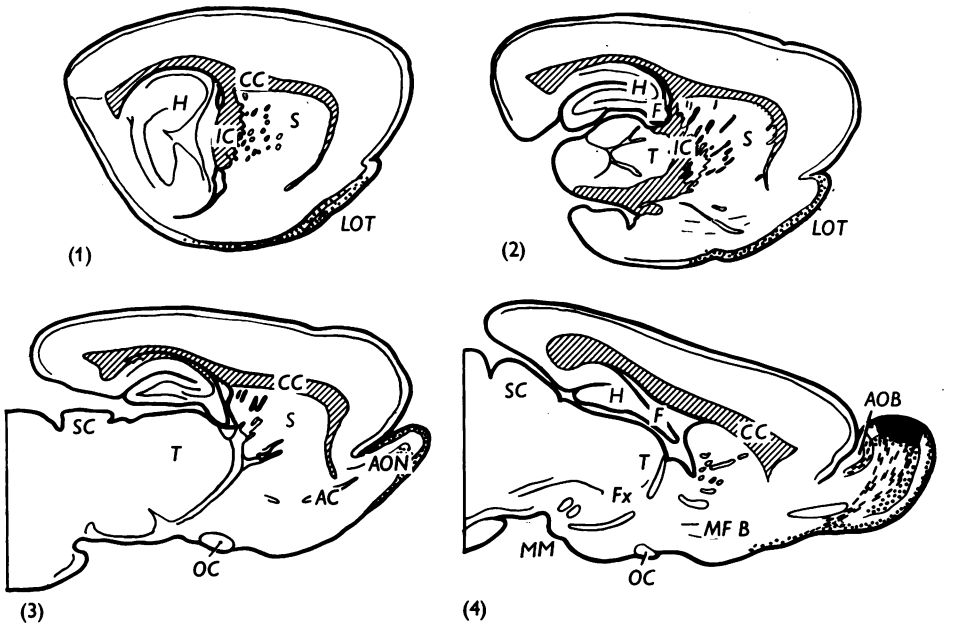

Fig. 2. Outlines of sagittal sections of the hemisphere to show the lesion and degeneration in Expt. P 63. In this and the subsequent figures the lesion is shown in solid black and the degeneration by broken lines and dots.

Abbreviations

A	anteromedial nucleus of thalamus	*LT*	lateral nucleus of thalamus
AC	anterior commissure	*MA*	medial nucleus of amygdala
AD	anterodorsal nucleus of thalamus	*MD*	mediodorsal nucleus of thalamus
AOB	accessory olfactory bulb	*MFB*	medial forebrain bundle
AON	anterior olfactory nucleus	*MG*	medial geniculate nucleus
AV	anteroventral nucleus of thalamus	*MM*	medial mamillary nucleus
BlA	basolateral nucleus of amygdala	*MSN*	medial septal nucleus
CC	corpus callosum	*MT*	mamillo-thalamic tract
ClA	central nucleus of amygdala	*NG*	nuclei gemini
CO	centrum ovale	*OC*	optic chiasma
CoA	cortical nucleus of amygdala	*OT*	olfactory tubercle
F	fimbria	*PC*	pyriform cortex
Fx	fornix	*PTA*	pretectal area
GP	globus pallidus	*R*	reticular nucleus of thalamus
H	hippocampus	*RF*	rhinal fissure
IC	internal capsule	*S*	striatum
LA	lateral nucleus of amygdala	*SC*	superior colliculus
LG	lateral geniculate nucleus	*SI*	substantia innominata
LH	lateral habenular nucleus	*ST*	stria terminalis
LHA	lateral hypothalamic area	*T*	thalamus
LM	lateral mamillary nucleus	*TO*	optic tract
LOT	lateral olfactory tract	*V*	ventral nucleus of thalamus
LPA	lateral preoptic area	*VM*	ventromedial nucleus of hypothalamus
LSN	lateral septal nucleus	*ZI*	zona incerta

found in the pars externa and the pars lateralis, but in the latter the fibre fragmentation extends on to the dorsal surface of the nucleus. As the distinction between this area of the pars lateralis and the pars dorsalis is difficult to determine, even in Nissl material, it is possible that a little degeneration may extend into the lateral parts of the pars dorsalis. The degeneration in the pars lateralis of the anterior olfactory nucleus is directly continuous caudally with that in the molecular layer of the pyriform cortex, the whole of which, from the rhinal sulcus laterally to the olfactory tubercle and amygdala medially, and from the anterior olfactory nucleus rostrally to the entorhinal area caudally is affected. As only an occasional fibre is seen amongst the pyramidal cells of this area it appears that the fibres of the tract terminate principally in relation to the apical dendrites of these cells. In the olfactory tubercle the degeneration is restricted to the molecular layer of its anterior and lateral parts. From the caudal part of the lateral olfactory tract degeneration extends medially into the nucleus of the tract and beyond it into the superficial parts of the cortical and medial amygdaloid nuclei, the density of the degeneration becoming less both medially and caudally. The other amygdaloid nuclei, and in particular the central nucleus and the bed nucleus of the stria terminalis, show no evidence of fibre degeneration.

In ten brains, lesions of the olfactory bulb, which have involved approximately the same area of the surface of the bulb, have suprisingly given considerably more degeneration in the brain and have caused severe degeneration in the anterior limb of the anterior commissure. The explanation for this difference is that these lesions have produced a wedge-shaped area of necrosis which extends well back, through the periventricular layer of the bulb, into the white matter in the junctional region of the bulb and anterior olfactory nucleus, and so directly involves the fibres of the anterior limb of the commissure as they emerge from this nucleus. In no case has degeneration been found in the anterior commissure or in the deep projection pathway associated with it when the lesion remained superficial to the periventricular layer.

The pattern of degeneration in these cases with involvement of the anterior limb of the anterior commissure is the same as after complete removal of the olfactory bulb. A typical example of such an experiment is found in R 81 where both the main and accessory olfactory bulbs have been completely destroyed (Fig. 3). The degeneration in the lateral olfactory tract in this case is very much heavier than in P 63 (described above as typical of a partial superficial lesion of the bulb), but its distribution in the molecular layer of the anterior olfactory nucleus, the olfactory tubercle, the pyriform cortex and amygdala is precisely the same. The principal difference between the two brains is the presence here of severe degeneration in the anterior limb of the anterior commissure which is distributed to both the ipsi- and contralateral sides. On the side of the lesion many degenerating fascicles can be seen spreading out diffusely towards the deep aspects of the cells of the external and lateral parts of the anterior olfactory nucleus, and behind this to the deep layers of the olfactory tubercle and pyriform cortex. Caudal to the level of crossing of the anterior commissure this projection to the pyriform cortex becomes progressively attenuated so that only an occasional degenerating fibre reaches the deep aspect of the pyriform cortex at the level of the amygdaloid nuclei. In view of previous accounts of a projection through the anterior commissure to the central amygdaloid

nucleus it may be emphasized that this nucleus is quite free of degeneration. The degeneration on the contralateral side is restricted to the main and accessory olfactory bulbs, in both of which there is an appreciable amount of fibre degeneration in the periventricular and granule cell layers with a few fibres extending superficially into the external plexiform layer. In this experiment no degeneration can be seen in the contralateral anterior olfactory nucleus, but in other experiments of this group unequivocal preterminal degeneration has been found in the external and lateral parts of this nucleus.

DISCUSSION

The present account of the central connexions of the olfactory bulb differs from the widely accepted description (cf. Allison, 1953*b*) in limiting its projection to the lateral olfactory tract; with small lesions strictly limited to the bulb degeneration is never seen in any part of the anterior commissure. This conclusion is in agreement with the recent findings of Lohman (1963), but as we do not have partial lesions in all parts of the bulb (and in particular limited to the accessory bulb), we cannot state with certainty that there are no fibres of bulbar origin in the anterior limb of the anterior commissure or in the ipsilateral deep projection pathway. The finding that even after comparatively small partial lesions a wedge of necrosis may extend deeply through the periventricular layer to involve the anterior commissure directly in the region of the anterior olfactory nucleus, may account for earlier descriptions of degeneration in the commissure after lesions apparently restricted to the olfactory bulb. It is clear from examination of sagittal sections of several brains that it is impossible in the rat to remove the whole, or even the greater part of, the olfactory bulb without involving the commissure behind the level of the bulb itself. The fact that there is no degeneration in the anterior commissure after partial lesions of the bulb must be taken as evidence against the older view that the fibres of the commissure are derived from the tufted cells of the bulb (Allison, 1953*a*). This conclusion is in accord with the recent electrophysiological observation (von Baumgarten, Green & Mancia, 1962; Yamamoto *et al.* 1963) that stimulation of the anterior commissure failed to activate tufted cells antidromically within the bulb.

In regard to the origin of the anterior limb of the anterior commissure, our evidence indicates that it does not arise from the olfactory bulb and so confirms the observation of Lohman (1963) who has reviewed the literature on this point. The contralateral projection of the commissure is to both the anterior olfactory nucleus and to the main and accessory olfactory bulbs. The fact that in experiments in which the anterior commissure is involved degeneration may be found either in the contralateral bulb alone, or in the bulb and anterior olfactory nucleus, indicates that there are at least two components in the contralateral projection and this is in accord with Lohman (1963); he found the origins of the components to the bulb and the anterior olfactory nucleus to be from the pars dorsalis and the pars rostralis respectively. The fibres to the contralateral olfactory bulb are therefore not strictly commissural but rather represent a crossed projection pathway.

The ipsilateral deep projection pathway associated with the anterior commissure which has also been described by Cragg (1961*a*) and Lohman (1963) is of interest for

two reasons. First, with the possible exception of the cortical and medial amygdaloid nuclei, it appears to duplicate the projection of the lateral olfactory tract, and secondly, because it projects to the deep aspect of the relevant cellular layers, in contrast to the lateral olfactory tract fibres which end predominantly upon the apical dendrites, it may well have a completely different effect upon the cells. It is not

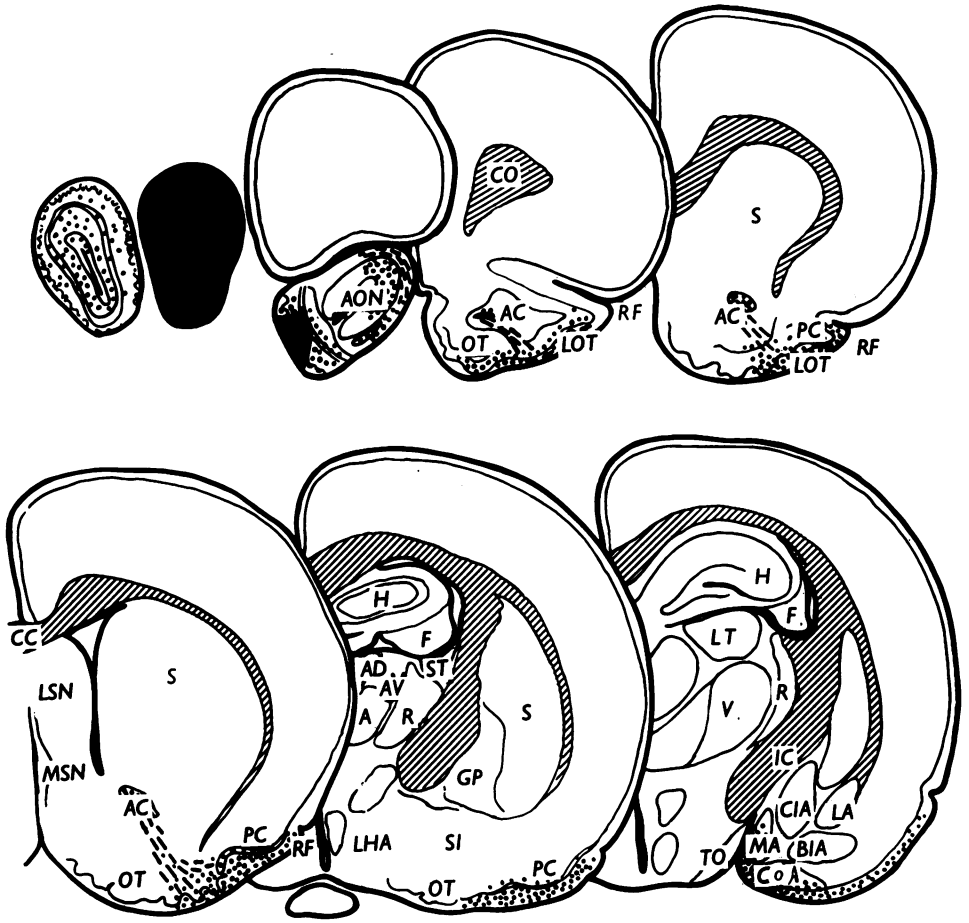

Fig. 3. Outlines of coronal sections at different rostro-caudal levels through the hemisphere to show the site and extent of the lesion and the distribution of the degeneration in Expt. R81.

known whether the fibres of this deep pathway arise in common with the decussating fibres of the anterior commissure.

The finding of a limited projection to the amygdaloid nuclei is in agreement with most recent studies with the Nauta technique (Cragg, 1961*a*; Sanders-Woudstra, 1961; Lohman, 1963). In none of these investigations has degeneration been found in the central amygdaloid nucleus of either side, or in the bed nucleus of the stria terminalis as was described in earlier studies with the Glees method (Le Gros Clark

& Meyer, 1947; Meyer & Allison, 1949; Allison, 1953*a*). The probable explanation for this discrepancy is that these nuclei commonly show pseudo-degeneration with the Glees method (Cowan & Powell, 1956).

II. THE EFFERENT CONNEXIONS OF THE PYRIFORM CORTEX

Results

In view of the difficulty involved in placing a lesion which destroys all, or even most, of the pyriform cortex without concomitant damage to either adjoining cortical areas or subcortical structures, brains with small lesions in the anterior and posterior parts of the pyriform cortex will be described. In Expt. P32 the anterior

Fig. 4. The lesion and the resulting degeneration in Expt. P32.

part of the pyriform cortex and the lateral olfactory tract have been damaged from just behind the level of the anterior olfactory nucleus back to the level of the genu of the corpus callosum. The pyriform cortex has been involved throughout its mediolateral extent, but the damage does not extend beyond the rhinal sulcus laterally or encroach upon the olfactory tubercle medially. The lesion is superficial throughout and does not involve the underlying white matter (Fig. 4). In addition to the rostrally directed degeneration in the lateral olfactory tract passing to the anterior olfactory nucleus and olfactory bulb, which will be described in detail later (section (3)), there is heavy fibre fragmentation extending medially and caudally from the lesion. The caudally directed degeneration can be followed through two pathways, superficial and deep. The superficial pathway in the molecular layer, contains considerably more degeneration than after lesions of the

olfactory bulb and much more appears to terminate in the caudal part of the pyriform cortex; in addition, many degenerating fibres can be traced beyond the pyriform cortex into the lateral part of the entorhinal area where they terminate. There is no degeneration in the subiculum or hippocampus. The deep pathway passes caudally beneath the pyriform cortex and distributes a heavy projection to the cellular layers of this area and beyond it to the entorhinal region. Associated with this deep pathway is a projection to the basal and lateral amygdaloid nuclei.

The medially directed degeneration passes into the nucleus of the diagonal band, the olfactory tubercle (especially its anterior and lateral parts), to the cortex below and in front of the genu of the corpus callosum, and into the lateral preoptic area. From the lateral preoptic area degeneration can be clearly traced caudally in the medial forebrain bundle as far as the mid-tuberal level. Much of this degeneration appears to terminate in the lateral hypothalamic area, but some is directed towards the suprachiasmatic region. There is no evidence, however, of degeneration in the supraoptic, paraventricular or suprachiasmatic nuclei. A second, heavier, projection extends caudally from the lateral preoptic area in a dorsal and medial direction. The majority of the degenerating fibres enter the inferior thalamic radiation and pass directly to the medioventral and mediodorsal nuclei through the subjacent thalamic nuclei. A smaller number of fibres take an even more caudal course, some reaching the most posterior part of the mediodorsal nucleus by passing dorsally along the medial margin of the habenulo-peduncular tract. Terminal degeneration is found throughout the anteroposterior extent of the central part of the mediodorsal nucleus, but is considerably heavier in its caudal third. From the region of this degeneration in the mediodorsal nucleus a smaller number of degenerating fibres extend dorsally into the lateral habenular nucleus where they again break up into a dense pericellular plexus (Fig. 5). In addition to this degeneration on the side of the lesion, there is a little fibre fragmentation in the comparable area of the mediodorsal and lateral habenular nuclei of the opposite side; this degeneration can be traced in continuity with that in the inferior thalamic radiation through the intermediodorsal commissure.

Only one experiment with a lesion in the posterior part of the pyriform cortex will be described. In this case (Expt. P7) there is a small area of destruction in the pyriform cortex from just behind the level at which the stria terminalis passes vertically from its origin in the amygdala. Throughout this extent only the lateral third of the pyriform cortex is involved and over a short distance the lesion extends beyond the rhinal fissure so that the neocortex is marginally involved. In the central part of the lesion the cortex has been damaged throughout its entire thickness, but the fibres of the external capsule have not been interrupted. It may be emphasized that no part of the amygdala has been encroached upon (Fig. 6). From the depths of the lesion heavy fibre degeneration can be traced into the external capsule and hence, in a medial and downwards direction, into the medial part of the pyriform cortex and into the lateral and basal amygdaloid nuclei. In these nuclei there is definite evidence of terminal degeneration, but the majority of the degenerating fibres appear to continue forwards through the anterior amygdaloid area to reach the lateral preoptic region. The further course and distribution of the degeneration is the same as that previously described for lesions of the rostral part of the pyriform

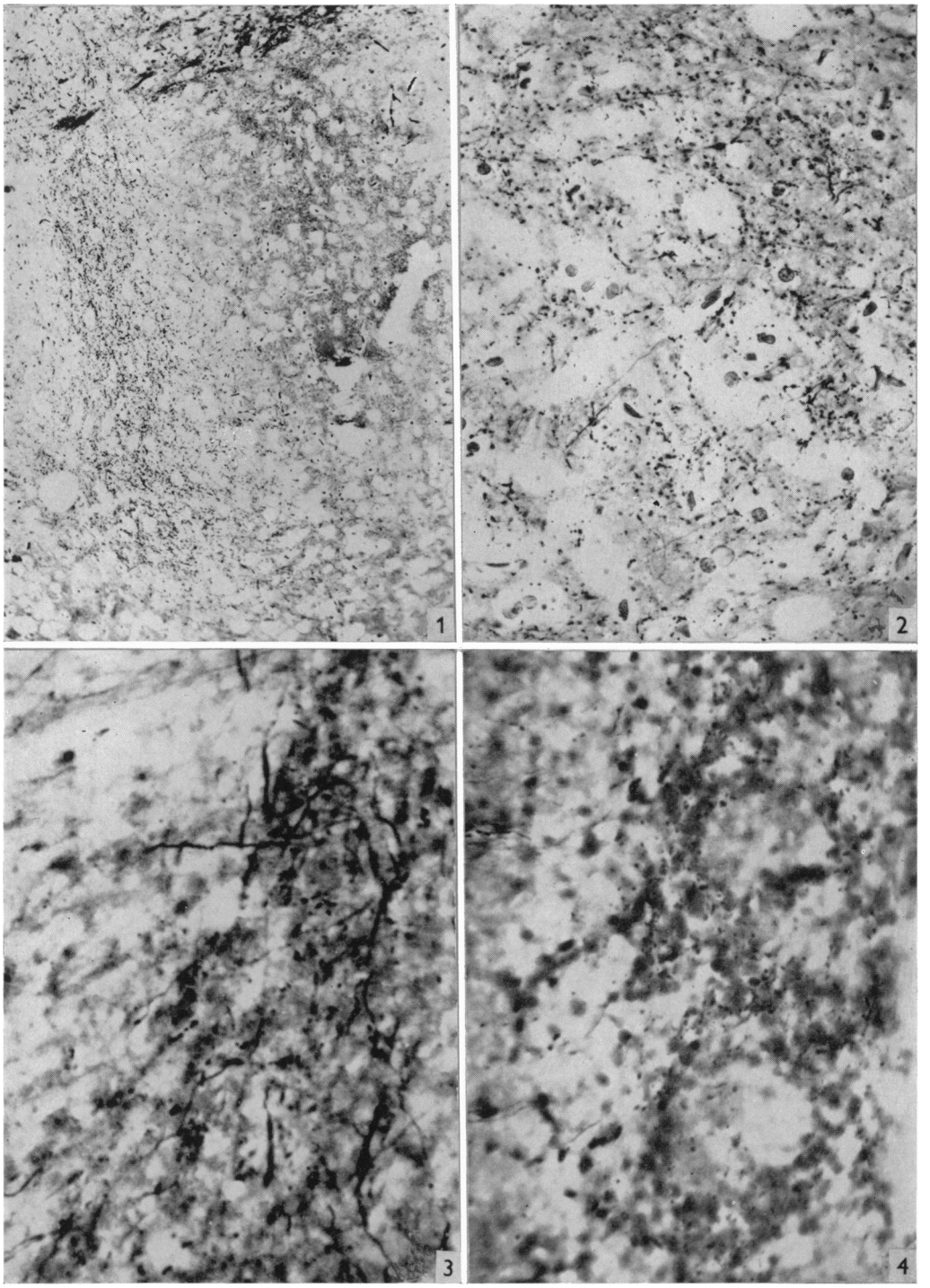

Fig. 5. (1) The fibre degeneration in the central part of the mediodorsal nucleus and in the lateral habenular nucleus of the ipsilateral side after a lesion of the pyriform cortex in Expt. P32. Method of Nauta & Gygax (1951). ×168. (2) Terminal degeneration in the mediodorsal nucleus. ×460. (3) Terminal degeneration in nuclei gemini in Expt. PL47. Method of Nauta (1957). ×680. (4) Terminal degeneration around the glomeruli of the ipsilateral olfactory bulb following section of the lateral olfactory tract in Expt. PL32. Treatment as (3). ×480.

cortex and need not be detailed further. However, it should be noted that there is no evidence for a centrifugal projection to the olfactory bulb from this region of the pyriform cortex (see later), nor is there any suggestion of degeneration within the stria terminalis.

In the rabbit, Lundberg (1962) has described a projection from the pyriform cortex, through the medial forebrain bundle, as far caudally in the hypothalamus as the premamillary region where the fibres terminate in two discrete cell masses, which he has designated the nuclei gemini. As these nuclei are only clearly

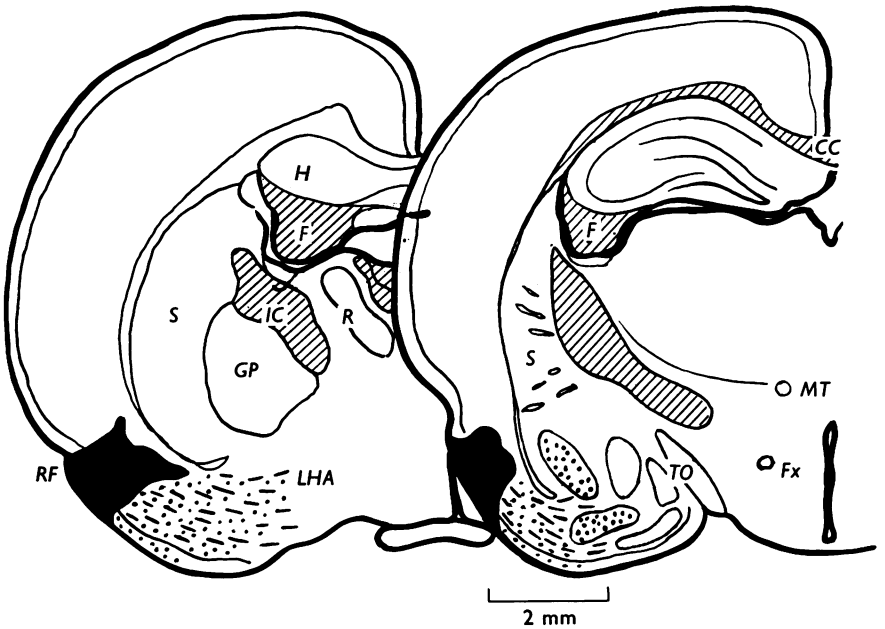

Fig. 6. Diagram to show the lesion and the distribution of the degeneration in the amygdala in Expt. P7.

recognized in sagittal section, four brains with lesions confined to the pyriform cortex were sectioned in this plane. In these sections (e.g. Expt. PL47) the degenerating fibres in the medial forebrain bundle can be seen to run throughout the entire rostro-caudal extent of the lateral hypothalamus and to enter the dorsal aspect of the nuclei gemini (Fig. 7) before breaking up into a fine preterminal plexus (Fig. 5). The morphology of these nuclei in the rat is exactly comparable to that described by Lundberg, as is the mode of entry and termination of the pyriform cortex fibres. The course and termination of the remaining degeneration in these brains is the same as that described earlier. Although nothing is known of the other connexions or functional significance of the nuclei gemini, the fact that they receive a distinct contribution from the pyriform cortex indicates that there is a direct olfactory influence extending as far as the level of the caudal hypothalamus.

In view of the possibility that the subcortical degeneration seen in the above experiments may be due to interruption of fibres arising in the neocortex above the

rhinal sulcus, several control experiments were done. Expt. PL 44 is typical, in that there is a lesion of moderate size in the neocortex but with no involvement of the pyriform area. The damage extends as a longitudinal strip from the level of the septum rostrally back almost to the caudal end of the hemisphere; centrally it extends down to the rhinal sulcus, and at its maximum width it involves almost

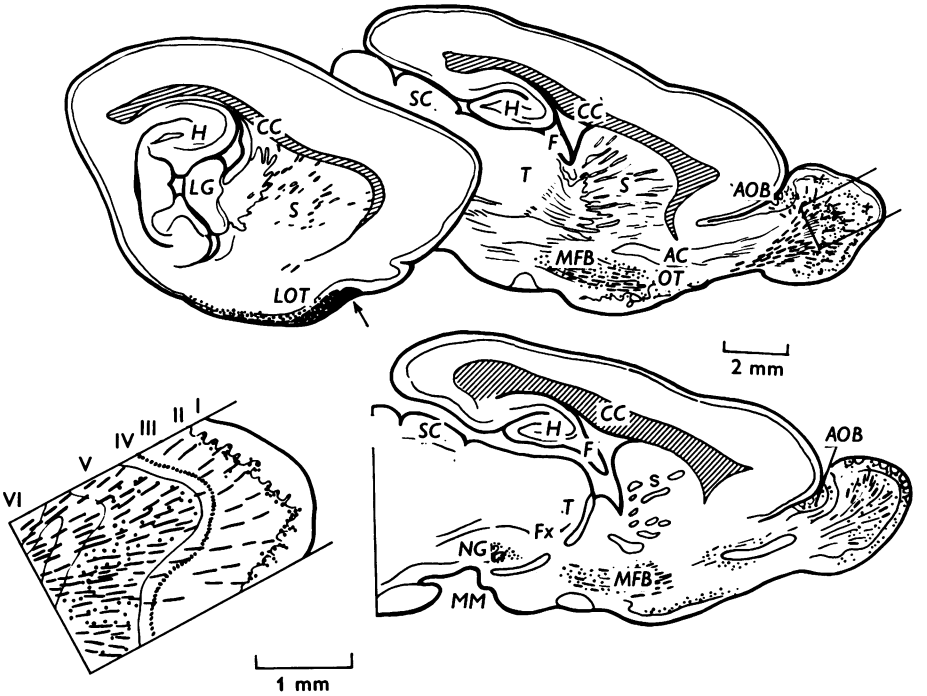

Fig. 7. Outlines of sagittal sections to show the lesion and the distribution of the degeneration in Expt. PL 47.

half the cortex on the lateral surface of the hemisphere. Although over the greater part of its extent the lesion is confined to the cortex, in its central part it has penetrated deeply to interrupt the fibres in the external capsule (Fig. 8). As it is not relevant to the present study, the distribution of the degeneration in this case will not be described in detail, but there is heavy fibre fragmentation in adjoining areas of the neocortex, the striatum and the thalamus. The significant feature of the present experiment is that there is no evidence of degeneration in the pyriform cortex, the amygdaloid nuclei, the ventral pathway, the hypothalamus or the mediodorsal nucleus of the thalamus, and as it is obvious that there can be no question that these negative observations are due to inadequate impregnation it may be concluded that the pattern of degeneration described in the earlier experiments is in fact due to the involvement of the pyriform cortex. Other similar experiments with additional involvement of the entorhinal area do not result in degeneration in the ventral pathway, the hypothalamus and the mediodorsal nucleus of the thalamus but do, of course, show degeneration in the perforant and alvear pathways to the hippocampus.

Discussion

In view of the similarity between the present findings on the projection of the pyriform cortex and earlier accounts of the connexions of the amygdala and temporal neocortex (Whitlock & Nauta, 1956; Nauta, 1961) it is necessary to emphasize that the pattern of degeneration described in our experiments is due to injury of the pyriform cortex alone and not to involvement of the underlying amygdaloid nuclei. The evidence for this rests primarily on the fact that the same subcortical distribution of degeneration has occurred in all the experiments with pyriform cortex lesions, but quite a different distribution of degeneration has been found in the control experiments with lesions in the adjacent neocortex. The

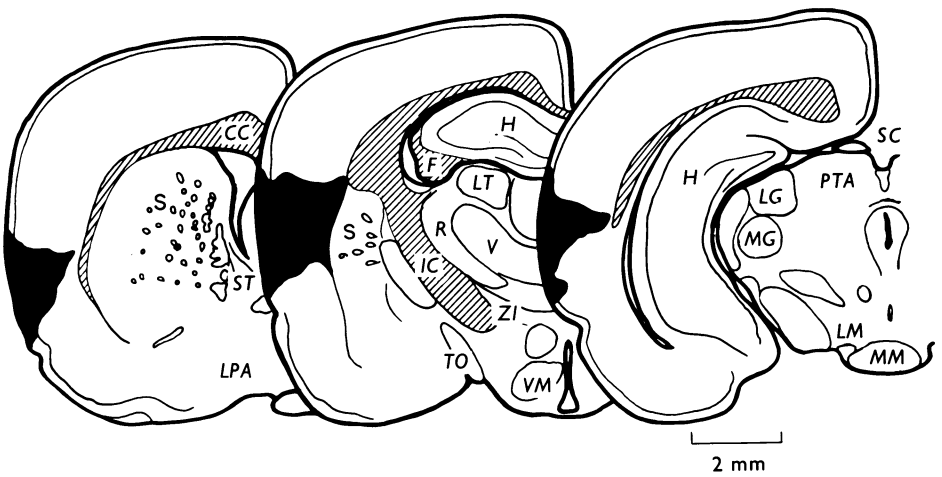

Fig. 8. The lesion in Expt. PL44.

superficial nature of the pyriform lesions rules out the possibility that the degeneration has been due to interruption of fibres from the amygdala, but as the pathway from the pyriform cortex to the preoptic area passes through the basal and lateral amygdaloid nuclei, lesions of these nuclei (even if placed stereotaxically) must inevitably damage the outflow from the pyriform area. The only difference between the present account of the subcortical projection of the pyriform cortex and published descriptions of the efferent connexions of the amygdala is that in the present experiments no degeneration has been seen in the stria terminalis. While the absence of degeneration in the stria terminalis after pyriform cortex lesions is of interest in contradicting the older view of Cajal (1911), its main relevance in the present context is that it serves as a useful control in showing that the pyriform cortex lesions have not involved the basolateral nuclear group of the amygdala in our experiments. Whether or not these nuclei also contribute fibres to the ventral pathway to the diencephalon cannot be excluded with current techniques as has been more fully discussed elsewhere (Cowan, Raisman & Powell, 1965). That the *bilateral* degeneration in the medio-dorsal and lateral habenular nuclei is not artefactual is apparent from several observations; it has not been seen in experi-

ments with lesions in the neocortex just dorsal to the rhinal sulcus; it has been described by several workers in similar studies (Guillery, 1959; Droogleever-Fortuyn, Hiddema & Sanders-Woudstra, 1960; Nauta, 1961; Cragg, 1961*b*; Sanders-Woudstra, 1961), and the degeneration can be traced in continuity from the inferior thalamic peduncle across the mid-line in the intermediodorsal commissure.

In addition to the amygdala and temporal neocortex, afferent fibres to the mediodorsal nucleus have been described as arising in several sites which could not possibly have been involved in the present experiments. Foremost amongst these are the septum, the nucleus of the diagonal band and the region of the olfactory tubercle (Guillery, 1959; Droogleever-Fortuyn, *et al.* 1960; Cragg, 1961*b*). In each case the fibres from these areas have been described as following essentially the same course as that taken by the efferent fibres from the pyriform cortex. Since, with the exception of the septum, it is impossible to place lesions in these structures without concomitant involvement of the fibres from the pyriform cortex, any degeneration found within this system must take into account such involvement. It is, of course, impossible to exclude that these areas are also contributing fibres to the mediodorsal nucleus of the thalamus, but all the evidence from the distribution and density of the degeneration in the nucleus points to the pyriform cortex as being the major, if not exclusive site of origin for this system. The finding of degenerating terminals as well as fibres of passage in the basolateral amygdaloid nuclei after pyriform cortex lesions makes it clear that the pyriform area has a dual projection to the hypothalamus: a direct projection through the ventral pathway, and a relayed projection either through the stria terminalis or through the ventral amygdalofugal pathway after synapsing in the basal and lateral amygdaloid nuclei. It may be relevant to note that the other major source of telencephalic afferents to the hypothalamus, viz. the hippocampus, similarly sends fibres both directly and indirectly by way of the septum.

Despite the well-known influence of exteroceptive sensory mechanisms upon the hypothalamus (and through it, upon the hypophysis) there has as yet been little or no convincing neuro-anatomical evidence for the fibre systems linking the primary sensory pathways and the hypothalamus. Interest in this problem has again been aroused in recent years by the demonstration of the dominant role played by olfaction in the hypothalamic regulation of reproduction (Lee & Boot, 1955; Whitten, 1958; Bruce & Parrott, 1960). The present findings provide evidence for an unexpectedly close relationship between the tertiary olfactory areas and the hypothalamus; indeed the whole rostro-caudal extent of the hypothalamus is to be regarded as one of the principal sites of influence of the olfactory system, being separated from the olfactory receptors by only two synaptic relays. As such, these observations provide an anatomical basis for several physiological observations on the influence of olfactory mechanisms upon the hypothalamus of which only two will be discussed here. As Whitten (1958) has shown, the presence of a male animal will bring about a dramatic synchronization of oestrous cycles in a randomly 'cycling' population of female mice, and since this effect is abolished by removal of the olfactory bulbs it has been interpreted as being due to the release of FSH in response to olfactory stimuli from the male. More recently, Parkes & Bruce (1962) have

found that pregnancy can be blocked during the first 4 days after mating by exposure of the pregnant mice to odours from strange males. Like the Whitten effect, this response is again dependent on the integrity of the olfactory bulbs; it is probably due to an inhibition of LH secretion by olfactory stimuli from the strange male. In addition to these behavioural studies, Barraclough & Cross (1963) have now adduced electrophysiological evidence of an olfactory input to the hypothalamus by the demonstration of changes in the firing rate of single units in the lateral hypothalamus in response to a variety of odours. To what extent the 'olfactory projection' to the hypothalamus may be involved in other aspects of reproductive functioning, such as determining the onset of puberty, is not known (cf. Donovan & van der Werff ten Bosch, 1959). That it is important for the maintenance of normal gonadal activity, however, is clear from the observation that removal of the olfactory bulb in the mouse (even in adult animals) results in a very significant reduction in ovarian weight (Whitten, 1956). The possibility also arises that the olfactory system may exert a degree of control upon the other endocrines especially in those animals in which olfaction is still the major exteroceptive sensory mechanism.

The pathways by which other sensory systems, such as the visual, influence the hypothalamus are not known, but it is now clear that they do not involve direct neocortico-hypothalamic fibres. Although earlier work with the Glees technique had indicated connexions between the frontal cortex and hypothalamus (Meyer, 1949; Le Gros Clark & Meyer, 1950), Lundberg's (1960) recent study has shown there is no direct projection to the hypothalamus from any area of the neocortex, and a number of our own unpublished observations are in agreement with this conclusion.

From this it is clear that the only cortical influences upon the hypothalamus are those due to connexions from the phylogenetically older cortical areas such as the hippocampus and pyriform cortex, and indeed the hypothalamus may be regarded as the principal subcortical projection site of these areas.

The other main projection of the pyriform cortex is that to the medial group of dorsal thalamic nuclei, the whole area projecting bilaterally to the mediodorsal and to the ipsilateral medioventral nucleus. This demonstration of an olfactory input to the dorsal thalamus is of interest from several points of view. First, because to some extent it brings olfaction into line with other sensory systems, all of which have long been known to have connexions with one or other of the thalamic nuclei, and in this context it may be helpful to consider the cortical projection areas of the mediodorsal and medioventral nuclei (viz. the orbitofrontal area and the adjacent cortex on the inferolateral aspect of the frontal lobe respectively) as being, in part at least, sensory projection areas for olfaction. There is, of course, a basic difference in organization of the thalamic relationship to olfaction and the other sensory systems in that the olfactory projection to the thalamus has already passed through a cortical region. This organizational difference must imply a functional dissimilarity between olfaction and the other sensory systems in which case the projection of the pyriform cortex to the mediodorsal and medioventral nuclei might be regarded as being essentially homologous to the projection of the main sensory areas of the cortex to the association nuclei of the thalamus (Mettler, 1935; Peele, 1942). A second

point of interest is the close topographical relationship between the 'thalamic projection areas' of olfaction and taste and between the corresponding cortical 'projection areas', that of the medioventral nucleus being directly contiguous with the region recently defined by Benjamin & Akert (1959) as being the cortical region for taste. The third interesting point raised by the finding of a direct projection from the pyriform cortex to the mediodorsal nucleus concerns the phylogenetic relations of these two structures. The mediodorsal nucleus has always attracted interest because of its close connexion with the frontal cortex, the progressive development of which is one of the more outstanding features of the evolution of the Primate brain (Le Gros Clark, 1932; Walker, 1938). At first sight it may seem anomalous that this nucleus should receive afferents from the phylogenetically old pyriform cortex which undergoes a significant regression in ascending phylogeny. However, it should be pointed out that even in the rat the projection of the pyriform cortex is to a relatively restricted part of the mediodorsal nucleus, which may well be homologous with the relatively poorly developed medial, magnocellular part of the nucleus, which in the Primate projects to the orbito-frontal cortex. That this is so, is suggested by the finding that what are probably the corresponding afferents to the mediodorsal nucleus in the monkey have in fact been described as terminating within this subdivision of the nucleus (Nauta, 1962).

The finding of a projection from the pyriform cortex to the entorhinal area is in agreement with Cragg (1961*a*), and since the entorhinal area is the main source of afferents to the hippocampus these observations may be taken as support for the older view that the hippocampus is closely related to the sense of olfaction. On the other hand, despite careful examination of several brains with lesions restricted to the pyriform cortex, a number of which were cut sagittally specifically for this purpose, no degeneration could be seen in the subiculum or the hippocampus. Cragg (1961*a*) has advanced evidence in support of a direct hippocampal projection from the pyriform cortex, but as few of his lesions were restricted to the basal aspect of the hemisphere one possible explanation for the degeneration which he has described is that some hippocampal afferent pathway such as the cingulum was involved in his experiments. In an attempt to clarify this point we have placed lesions similar to Cragg's, in all of which other structures were involved as well as the pyriform cortex; in these cases, although degenerating fibres can be seen passing caudally from the pyriform cortex into the lateral entorhinal area (in which the degeneration becomes confluent with that in the medial entorhinal area and subiculum from the cingulum), no degeneration has been seen in the hippocampus itself.

The relationship between the pyriform cortex and the hippocampus is of considerable interest since these areas provide the two major telencephalic projections into the hypothalamus. The similarity in the organization of the two cortical regions may be even closer: (1) both have a substantial direct projection to the dorsal thalamus (the hippocampus projecting directly to the anterior nuclei and the pyriform cortex to the medial), and (2) through their projections into the lateral preoptic area, both cortical areas may be indirectly related with the epithalamus (as the lateral preoptic area is now known to be a major source of afferents to the lateral habenular nucleus through the stria habenularis). One important respect

in which the two differ, however, is that the pyriform cortex is directly connected with the epithalamus, making a small but definite contribution to the lateral habenular nuclei of both sides. This finding is again in agreement with older views which stressed the close relationship between the habenular nuclei and the olfactory system, a relationship which is even more intimate in the reptilian brain (Kappers, Huber & Crosby, 1936; Gamble, 1952). Taken together with the projection into the hypothalamus, these findings indicate that there are at least two pathways for olfactory influences to the mid-brain, viz. by way of the medial forebrain bundle and the habenulo-peduncular tract.

III. CENTRIFUGAL FIBRES TO THE OLFACTORY BULB

Results

The centrifugal fibres to the olfactory bulb have been studied in two different types of experiment: first, in brains in which the lateral olfactory tract was divided at the rostral end of the pyriform cortex, and secondly, in brains in which both the lateral olfactory tract and the anterior limb of the anterior commissure were sectioned. In the previous section an experiment of the first type was described, and here only one further example will be given. In this case, the brain has been sectioned sagittally and has given a particularly good demonstration of the rostrally directed fibres in the lateral olfactory tract (Fig. 7). The lesion in this brain (PL47) is limited to the lateral olfactory tract and the adjacent part of the pyriform cortex at the level of the anterior part of the olfactory tubercle (which has not been encroached upon). The degeneration found behind the level of the lesion will not be described as this is essentially the same as in the earlier experiments with lesions of the pyriform cortex, but it may be pointed out that there is a marked difference in the appearance of the lateral olfactory tract on the two sides of the lesion. Behind the lesion, the tract is completely degenerated throughout its cross-sectional area, whereas rostral to the lesion there are comparatively few degenerating fragments both within the lateral olfactory tract itself and in the white matter deep to it. It is clear that the centrifugal fibres form only a very small proportion of the total number of fibres in the tract and indeed, as the bulb is approached the majority of the degenerating fibres lie in the white matter just deep to the tract and in this way come to reach the external and lateral parts of the anterior olfactory nucleus and all parts of the olfactory bulb. In the latter, severe terminal degeneration is found throughout the periventricular and granule cell layers, and many fibres extend superficial to the mitral cells to reach the external plexiform and glomerular layers. No degeneration is found within the glomeruli in relation to the synapses between the olfactory nerve fibre and the dendrites of the mitral cell, but the glomeruli are distinctly outlined by degenerating terminals apparently ending upon the periglomerular cells (Fig. 5). Many of the degenerating fibres from the dorsal part of the centrifugal pathway pass directly into the accessory bulb where again there are many fibres in the granule cell layers, and a few extend out into the plexiform and glomerular layers. As far as it is possible to judge, the severity and distribution of the degeneration is uniform throughout the whole extent of the bulb. On the opposite side, no degeneration is seen within the anterior commissure, and the

lateral olfactory tract, the anterior olfactory nucleus and the main and accessory bulbs are quite free of fibre fragmentation.

In the second type of experiment both the lateral olfactory tract and the anterior limb of the anterior commissure have been divided. Experiment R82 will be described as representative of this group, as the lesion is in the form of a 'leucotomy' cut through the hemisphere just in front of the level of the genu of the corpus callosum (Fig. 9). The anterior limb of the anterior commissure has been interrupted where it passes through the nucleus accumbens and the lateral olfactory tract just behind the anterior olfactory nucleus. The only important differences in the findings between this and the previous experiment are the presence of degenerating fibres in the anterior limb of the anterior commissure and the olfactory bulbs of both sides, and the substantially heavier degeneration in the ipsilateral bulb in the present experiment as compared with PL47. The fibre fragments in the anterior commissure are distinctly finer than those in the lateral olfactory tract, and these are the only degenerating fibres reaching the opposite anterior olfactory nucleus and olfactory bulb. In the anterior olfactory nucleus of the contralateral side the terminal degeneration is found in its external and lateral parts, and in the bulb there is moderately severe degeneration in the periventricular and granule cell layers, but only an occasional degenerating fibre can be seen passing beyond the mitral cells, and no fragmentation is present in relation to the periglomerular cells. In the accessory bulb also there is degeneration in the layer deep to the mitral cells but few, if any, degenerating fibres pass into the external plexiform layer. On the ipsilateral side, in both the main and accessory bulbs, the intensity of the degeneration is very much greater than on the contralateral side; the proximity of the lesion makes it difficult to interpret the degeneration in the ipsilateral anterior olfactory nucleus. The evidence for regarding the anterior olfactory nucleus rather than the olfactory bulb, as the site of origin of these fibres has already been discussed (section (1)).

It is clear from a comparison of these two types of experiments, not only that there are two groups of rostrally directed fibres to the olfactory bulb, namely those fibres which arise on the same side and run in the lateral olfactory tract and those which come through the anterior commissure from the anterior olfactory nucleus of the contralateral side, but also that these can be clearly distinguished on the basis of their origin, size, pathway and site of termination. It is possible that some fibres going to the bulb may join the anterior limb of the commissure from other parts of the ipsilateral hemisphere, but there is no evidence in support of this possibility.

Discussion

The major finding of the present study is that, in agreement with the recent observation in the rabbit by Cragg (1962), it provides direct experimental evidence for Cajal's (1911) description in Golgi material of centrifugal fibres in the lateral olfactory tract. No systematic attempt has been made to determine the origin of these fibres, but some evidence bearing on this problem has been obtained from an examination of several brains prepared for other purposes. As has been described in the previous section, a lesion in the posterior part of the pyriform cortex does not result in degeneration passing forwards in the lateral olfactory tract (Expt. P7), and

similarly, several brains with lesions of the amygdala (Cowan, *et al.* 1965) show that these nuclei do not contribute to this system. Lesions involving the medial forebrain bundle in the hypothalamus result in fibre degeneration which can be traced into the olfactory tubercle and anterior olfactory nucleus (principally into the pars medialis), but not into the lateral olfactory tract or the olfactory bulb. In a lesion of the rostral hypothalamus and preoptic areas with encroachment upon the caudal part of the olfactory tubercle (MI15), however, degenerating fibres can be seen passing laterally along the olfactory tubercle to join the lateral olfactory tract and

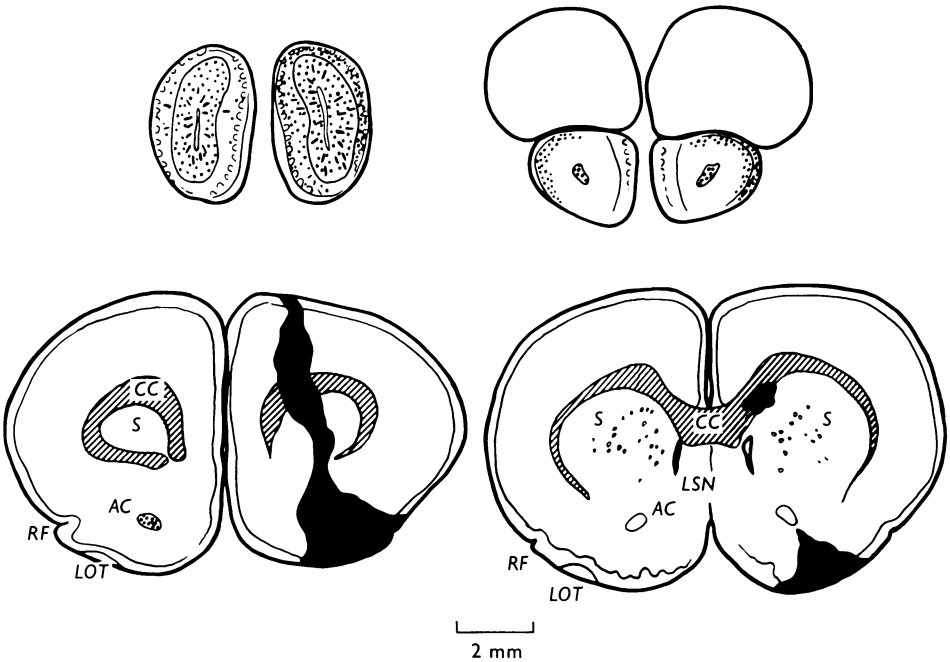

Fig. 9. The lesion and degeneration in Expt. R82.

run forwards in close association with it to the olfactory bulb. That the involvement of the olfactory tubercle in this experiment is critical is confirmed by another lesion in which the greater damage is in the hemisphere of the right side, but from a small area of destruction in the medial part of the olfactory tubercle of the left hemisphere degenerating fibres have been traced into the lateral olfactory tract and thence to the olfactory bulb. It seems reasonable to conclude, therefore, that many of the centrifugal fibres arise from the region of the olfactory tubercle (cf. Powell & Cowan, 1963). It is not possible to determine whether the rostral part of the pyriform cortex is also contributing fibres to the bulb since in all our experiments with lesions in this area there has also been some involvement of the lateral olfactory tract itself.

In regard to the termination of these fibres, they differ from those in the anterior commissure in that a substantial number reach the external plexiform and glomerular layers and from this it may be seen that the influence of the ipsilateral

hemisphere may be exerted in the region of the first synaptic relay of the olfactory pathway. That stimulation of the lateral olfactory tract can indeed influence periglomerular cells has been shown by Shepherd (1963), although it was not possible in that study to attribute this to the selective activation of either axon collaterals or centrifugal fibres. Until the site of origin of the centrifugal fibres is definitely established, and it becomes possible to excite them selectively, the precise role the centrifugal fibres play in olfaction will remain undetermined. In view of the close relationship of the periglomerular cells and the site of synapse between the incoming olfactory nerve fibres and the dendrites of the mitral and tufted cells it may be interesting to speculate that the centrifugal fibre (acting through these cells) may exert a presynaptic inhibitory influence on the olfactory nerve termination in a manner comparable to that found in the gracile and cuneate nuclei by Anderson, Eccles, Schmidt & Yokota (1964).

To substantiate this it would be necessary to show that the axons of the periglomerular cells end upon the terminal portion of the olfactory nerve fibres rather than upon the dendrites of the mitral cells as suggested by Cajal (1911). Whether the centrifugal fibres also terminate upon deeper granule cells, and thereby allow interaction with the commissural fibres and axon collaterals, cannot be determined as the degeneration seen in this layer after lesions of the lateral olfactory tract may be due simply to the degeneration of fibres passing through the layer to a more superficial termination.

SUMMARY

The central projection of the olfactory bulb, the efferent connexions of the pyriform cortex and the centrifugal fibres to the olfactory bulb have been studied in the rat with the Nauta method.

The olfactory bulb projects only through the lateral olfactory tract to the dorsal and external parts of the anterior olfactory nucleus, the antero-lateral part of the olfactory tubercle, the entire pyriform cortex, the nucleus of the lateral olfactory tract and the cortical and medial amygdaloid nuclei. The anterior limb of the anterior commissure receives no fibres from the olfactory bulb and projects only to the contralateral anterior olfactory nucleus and olfactory bulb.

The pyriform cortex sends efferent fibres (i) to the lateral entorhinal area; (ii) to the basal and lateral amygdaloid nuclei; (iii) to the lateral preoptic area from which fibres pass to the olfactory tubercle, the diagonal band nucleus and the cortex below the corpus callosum; (iv) through the medial forebrain bundle to the entire rostro-caudal extent of the hypothalamus (including the nuclei gemini); and (v) to the ipsilateral medioventral nucleus of the thalamus and bilaterally to the central parts of the mediodorsal nuclei and the overlying lateral habenular nuclei.

Centrifugal fibres pass forwards in the lateral olfactory tract and terminate in the olfactory bulb, at all levels except the outermost layer of the olfactory nerve fibres; they probably arise in the olfactory tubercle.

This work was supported by a grant from the Medical Research Council.

REFERENCES

ADRIAN, E. D. (1942). Olfactory reactions in the brain of the hedgehog. *J. Physiol., Lond.* **100**, 459–473.

ADEY, W. R. (1953). An experimental study of the central olfactory connexions in a Marsupial (*Trichosurus vulpecula*). *Brain*, **76**, 311–336.

ALLISON, A. C. (1953*a*). The structure of the olfactory bulb and its relationship to the olfactory pathways in the rabbit and the rat. *J. comp. Neurol.* **98**, 309–350.

ALLISON, A. C. (1953*b*). The morphology of the olfactory system in the vertebrates. *Biol. Rev.* **28**, 195–244.

ANDERSON, P., ECCLES, J. C., SCHMIDT, R. F. and YOKOTA, T. (1964). Depolarization of presynaptic fibres in the cuneate nucleus. *J. Neurophysiol.* **27**, 92–106.

BARRACLOUGH, C. A. & CROSS, B. A. (1963). Unit activity in the hypothalamus of the cyclic female rat: effect of genital stimuli and progesterone. *J. Endocrin.* **26**, 339–359.

BAUMGARTEN, R. VON, GREEN, J. D., and MANCIA, M. (1962). Recurrent inhibition in the olfactory bulb. II. Effects of antidromic stimulation of commissural fibres. *J. Neurophysiol.* **25**, 489–500.

BENJAMIN, R. M. & AKERT, K. (1959). Cortical and thalamic areas involved in taste discrimination in the albino rat. *J. comp. Neurol.* **111**, 231–260.

BRUCE, H. M. & PARROTT, D. M. V. (1960). Role of olfactory sense in pregnancy block by strange males. *Science*, **131**, 1526.

CAJAL, S. RAMON Y. (1911). Histologie du système nerveux de l'homme et des vertébrés. Paris: Maloine.

COWAN, W. M., RAISMAN, G. & POWELL, T. P. S. (1965). The connexions of the amygdala. *J. Neurol. Neurosurg. Psychiat.* (in the Press).

COWAN, W. M. & POWELL, T. P. S. (1956). A note on terminal degeneration in the hypothalamus. *J. Anat.* **90**, 188–192.

CRAGG, B. G. (1961*a*). Olfactory and other afferent connexions of the hippocampus in the rabbit, rat and cat. *Expl Neurol.* **3**, 588–600.

CRAGG, B. G. (1961*b*). The connections of the habenula in the rabbit. *Expl Neurol.* **3**, 388–409.

CRAGG, B. G. (1962). Centrifugal fibres to the retina and olfactory bulb, and composition of the supraoptic commissures in the rabbit. *Expl Neurol.* **5**, 406–427.

DONOVAN, B. T. & VAN DER WERFF TEN BOSCH, J. J. (1959). The hypothalamus and sexual maturation in the rat. *J. Physiol., Lond.* **147**, 78–92.

DROOGLEEVER-FORTUYN, J., HIDDEMA, F. & SANDERS-WOUDSTRA, J. A. R. (1960). A note on rhinencephalic components of the dorsal thalamus. The parataenial and dorso-medial nuclei. In A. Biemond *et al.* (eds.), *Recent Neurological Research*. Amsterdam: Elsevier.

FOX, C. A., MCKINLEY, W. A. & MAGOUN, H. W. (1944). An oscillographic study of olfactory system of cats. *J. Neurophysiol.* **7**, 1–16.

GAMBLE, H. J. (1952). An experimental study of the secondary olfactory connexions in *Lacerta viridis*. *J. Anat.* **86**, 180–196.

GREEN, J. D., CLEMENTE, C. D. & DE GROOT, J. (1957). Rhinencephalic lesions and behaviour in cats. An analysis of the Klüver-Bucy syndrome with particular reference to normal and abnormal sexual behaviour. *J. comp. Neurol.* **108**, 505–536.

GREEN, J. D., MANCIA, M. & VON BAUMGARTEN, R. (1962). Recurrent inhibition in the olfactory bulb. I. Effects of antidromic stimulation of the lateral olfactory tract. *J. Neurophysiol.* **25**, 467–488.

GUILLERY, R. W. (1959). Afferent fibres to the dorso-medial thalamic nucleus in the cat. *J. Anat.* **93**, 403–418.

KAPPERS, C. U. A., HUBER, G. C. & CROSBY, E. C. (1936). *The Comparative Anatomy of the Nervous System of Vertebrates, including Man*, vol. II, 1408–1411. New York: Macmillan.

KERR, D. I. B. & HAGBARTH, K. E. (1955). An investigation of olfactory centrifugal fibre system. *J. Neurophysiol.* **18**, 362–374.

LEE, S. VAN DER, & BOOT, L. M. (1955). Spontaneous pseudopregnancy in mice. *Acta physiol. pharm. neerl.* **4**, 442.

LE GROS CLARK, W. E. (1932). The structure and connections of the thalamus. *Brain*, **55**, 406–470.

LE GROS CLARK, W. E. & MEYER, M. (1947). The terminal connections of the olfactory tract in the rabbit. *Brain*, **70**, 304–328.

LE GROS CLARK, W. E. & MEYER, M. (1950). Anatomical relationships between the cerebral cortex and the hypothalamus. *Br. med. Bull.* **6**, 341–345.

LOHMAN, A. H. M. (1963). The anterior olfactory lobe of the guinea pig. *Acta anat.* (Suppl. 49), **53**.

LUNDBERG, P. O. (1960). Cortico-hypothalamic connexions in the rabbit. *Acta physiol. scand.* (Suppl. 171), **49**, 1–80.

LUNDBERG, P. O. (1962). The nuclei gemini. Two hitherto undescribed nerve cell collections in the hypothalamus of the rabbit. *J. comp. Neurol.* **119**, 311–316.

METTLER, F. A. (1935). Corticifugal fiber connections of the cortex of *Macaca mulatta*. The parietal region. *J. comp. Neurol.* **62**, 263–291.

MEYER, M. (1949). Study of efferent connexions of the frontal lobe in the human brain after leucotomy. *Brain*, **72**, 265–296.

MEYER, M. & ALLISON, A. C. (1949). An experimental investigation of the connexions of the olfactory tracts in the monkey. *J. Neurol. Psychiat., Lond.* **12**, 274–286.

NAUTA, W. J. H. (1957). Silver impregnation of degenerating axons. In *New research techniques of neuroanatomy*, pp. 17–26. Ed. W. F. Windle. Springfield, Illinois: C. C. Thomas.

NAUTA, W. J. H. (1961). Fibre degeneration following lesions of the amygdaloid complex in the monkey. *J. Anat.* **95**, 515–532.

NAUTA, W. J. H. (1962). Neural associations of the amygdaloid complex in the monkey. *Brain*, **85**, 505–520.

NAUTA, W. J. H. & GYGAX, P. A. (1951). Silver impregnation of degenerating axon terminals in the central nervous system. *Stain Tech.* **26**, 5–11.

PARKES, A. S. & BRUCE, H. M. (1961). Olfactory stimuli in mammalian reproduction. *Science*, **134**, 1049–1054.

PARKES, A. S. & BRUCE, H. M. (1962). Pregnancy-block in female mice placed in boxes soiled by males. *J. Reprod. Fert.* **4**, 303–308.

PEELE, T. L. (1942). Cytoarchitecture of individual parietal areas in the monkey (*Macaca mulatta*) and the distribution of the efferent fibres. *J. comp. Neurol.* **77**, 693–738.

PHILLIPS, C. G., POWELL, T. P. S. & SHEPHERD, G. M. (1963). Responses of mitral cells to stimulation of the lateral olfactory tract in the rabbit. *J. Physiol., Lond.* **168**, 65–88.

POWELL, T. P. S. & COWAN, W. M. (1963). Centrifugal fibres in the lateral olfactory tract. *Nature, Lond.*, **199**, 1296–1297.

POWELL, T. P. S., COWAN, W. M. & RAISMAN, G. (1963). Olfactory relationships of the diencephalon. *Nature, Lond.*, **199**, 710–712.

ROSE, J. E. & WOOLSEY, C. N. (1943). Potential changes in the olfactory brain produced by electrical stimulation of the olfactory bulb. *Fedn Proc. Fedn Am. Socs exp. Biol.* **2**, 42.

SANDERS-WOUDSTRA, J. A. R. (1961). Experimental anatomisch onderzoek over de verbindingen van enkele basale telencefale hersengebieden bij de albinorat. Thesis: University of Groningen.

SHEPHERD, G. M. (1963). Neuronal systems controlling mitral cell excitability. *J. Physiol., Lond.* **168**, 101–117.

WALKER, A. E. (1938). *The Primate Thalamus.* Chicago University Press.

WHITLOCK, D. B. and NAUTA, W. J. H. (1956). Subcortical projections from the temporal neocortex in *Macaca mulatta*. *J. comp. Neurol.* **106**, 183–212.

WHITTEN, W. K. (1956). The effect of removal of the olfactory bulbs on the gonads of mice. *J. Endocrin.* **14**, 160–163.

WHITTEN, W. K. (1958). Modification of the oestrus cycle of the mouse by external stimuli associated with the male. Changes in the oestrus cycle determined by vaginal smears. *J. Endocrin.* **17**, 307–313.

YAMAMOTO, C., YAMAMOTO, T. & IWAMA, K. (1963). The inhibitory systems in the olfactory bulb studied by intracellular recording. *J. Neurophysiol.* **26**, 403–415.