THE PRIMARY OPTIC CENTRES OF THE RAT. AN EXPERIMENTAL STUDY BY THE 'BOUTON' METHOD

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A great number of investigations on the central termination of the optic nerve in mammals has firmly established the fact that retinal fibres end in the superior colliculus and in the dorsal nucleus of the lateral geniculate body. In addition, the subthalamic nucleus of Luys and the nucleus opticus tegmenti have been claimed to receive optic fibres in several lower animals. Lastly, physiological data (Ranson & Magoun, 1933) strongly suggest that the pretectal region contains another primary optic centre with special importance for the pupillo-constrictor reflex.

Hitherto morphological data in this field have almost exclusively been obtained by the study of normal material and by experimental studies conducted either by the Marchi technique or by the method of transneuronal cell atrophy.

In recent years the study of the end-feet of Held-Auerbach (boutons), which become unusually conspicuous after section of afferent tracts, has yielded valuable information regarding synaptic relations in the lateral geniculate body (Glees & Clark, 1941; Glees, 1941). No complete determination of their distribution in the central nervous system after section of the optic nerve has, however, been attempted so far. For this reason we publish our findings, which are based on the bouton method and which differ in some points from the results obtained by the more usual methods.

MATERIAL AND METHODS

Three albino rats were employed. Both eyes were removed from one, and the left eye only from each of the others. After survival for 5 days the animals were killed by ether and perfused through the left ventricle with saline, followed by 80 % alcohol. The brains were carefully removed and placed in 96 % alcohol for 3 days. The brain of one of the unilateral cases was then prepared by Bodian's protargol method and those of the other two according to Ranson's pyridine silver technique.

The value of the bouton method has been much discussed. Phalen & Davenport (1937) have shown that the size and shape of normal boutons in the spinal cord vary widely in different individuals and in different species. Their conclusion is that only those changes in the usual appearance of boutons which are massive in extent can be relied upon for localizatory purposes. In our experiments, however, this restriction played no important part, since we constantly failed to find more than a very few solitary boutons in six normal rat brains stained either with the pyridine silver or with the protargol technique, whereas the optic centres in our experimental series showed a wealth of heavily staining ring- and bulb-shaped structures, in all respects comparable to those described by Hoff (1932), Glees & Clark (1941) and others (Pl. 1, fig. 2).

Meyer & Meyer (1945) have recently suggested that the appearance of heavily staining boutons in some particular region depends on physico-chemical changes in that region causing an increased argentophilia of pre-existent structures. Since they were able to demonstrate abnormally argentophil boutons in normal biopsy material, various not necessarily degenerative factors seem to be able to induce such changes. There can, however, be no doubt that at least the great majority of the boutons in our experimental material represented degenerative changes of the optic system, since after unilateral eye removal the ratio between ipsi- and contralateral boutons was found to correspond roughly with the results of previous experiments regarding the distribution of retinal fibres, in so far as more were found in the contralateral than in the ipsilateral optic centres. Although we have tried to establish this ratio by means of counting the boutons, it is doubtful whether the absolute number of boutons found in a nucleus is of value for estimating the number of fibres ending in it, since it is not yet possible to state that every degenerating fibre develops one or more degenerating boutons, and we cannot exclude the possibility that some of the boutons were not 'boutons terminaux' but bouton-like structures developing in the course of degenerating fibres ('boutons de passage', recently depicted by Meyer & Meyer, 1945).

RESULTS

Removal of both eyes gave results essentially similar to those following removal of one eye: it is the results of the latter operation that are described.

Lateral geniculate body

(*a*) *Nucleus dorsalis*. A large number of heavily staining ring- and bulb-shaped structures were found in the dorsal nucleus. The right nucleus contained roughly twice as many as the left. No definite localization of the boutons in the nucleus could be detected. On both the right and left side their number was largest in the superficial part of the nucleus and regularly decreased from without inwards, while the boutons were rather evenly distributed over the dorsoventral diameter of the nucleus. On this point our results do not correspond with those obtained in rodents and marsupials by other methods (Brouwer, Zeeman & Mulock Houwer, 1922; Jefferson, 1940; Bodian, 1937), which seems to prove that uncrossed fibres end chiefly in the dorsomedial part of the caudal half of the nucleus.

(*b*) *Ventral nucleus*. Extremely few boutons could be found in the ventral nucleus of the lateral geniculate body. Previous investigations have already indicated that the Marchi reaction in the ventral nucleus following eye removal is due to degeneration of fibres of passage and not of terminal fibres.

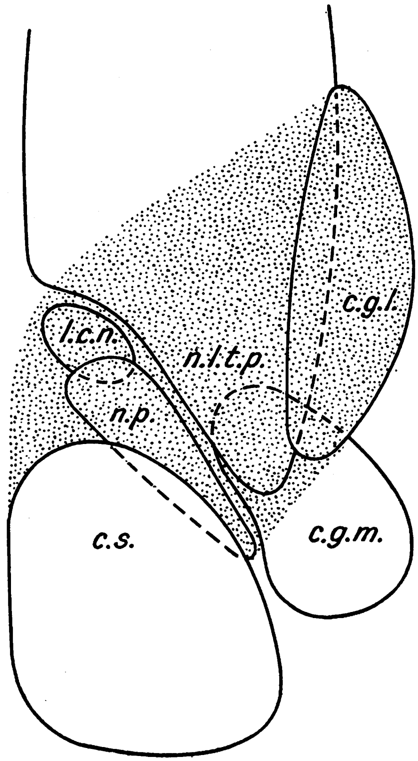
Pretectal area

After passing the lateral geniculate body and the lateral nucleus of the thalamus a considerable portion of the optic tract comes into close relation to the two main nuclei of this region, viz. (Clark's nomenclature, 1932):

(1) *The large-celled nucleus of the optic tract* (Cajal's (1911) noyau de la voie optique bigéminale), a small ovoid cell group occupying a rostromedial position in the pretectal area, just dorsal to the fibres of the commissura posterior (Text-fig. 1; Pl. 1, fig. 1).

(2) *The pretectal nucleus*, a much larger group of small cells situated immediately underneath the optic tract (Text-fig. 1; Pl. 1, fig. 1).

The rostral pole of the pretectal nucleus overlies the caudal one-third of the large-celled nucleus. The pretectal nucleus extends in a caudal and lateral direction. Its middle part lies between the rostral part of the superior colliculus medially and the nucleus lateralis thalami pars posterior (pulvinar) laterally, while its caudal part extends beyond the caudal tip of the pulvinar, and is thus situated between the colliculus medially and the medial geniculate body ventrolaterally. On the medial side the nucleus is continuous with the stratum opticum of the superior colliculus. Many optic fibres apparently pass through the pretectal nucleus only to enter the stratum opticum.



Text-fig. 1. Free-hand schematic reconstruction of the relative position of the nucleus lateralis thalami pars posterior (*n.l.t.p.*); lateral and medial geniculate bodies (*c.g.l.* and *c.g.m.*); large-celled nucleus of the optic tract (*l.c.n.*); pretectal nucleus (*n.p.*) and superior colliculus (*c.s.*), and their relation to the optic tract (shaded).

Previous anatomical studies have adduced only meagre evidence for the termination of retinal fibres in the pretectal region. In normal material Cajal (1911) observed optic collaterals entering the 'noyau de la voie optique bigéminale' (i.e. the large-celled nucleus) of the mouse, and Tsai (1925) reported similar findings for the pretectal nucleus in the opossum. The Marchi method has often yielded negative results (Bodian (1937) in the opossum; Jefferson (1940) in the ferret; Packer (1941) in the phalanger; Nichterlein & Goldby (1944) in the sheep). Clark (1931), however, observed some Marchi reaction in the large-celled nucleus of the rat following eye removal, and Lashley (1934) reported similar findings, without, however, being certain that optic fibres ended in the nucleus. It is possible that the cellular atrophy in the thalamus opticus of the goat and cat, observed by Minkowski (1920) after eye removal, involved the pretectal area also. As a whole, the available data do not seem to offer a satisfactory morphological basis for the activity of the pretectal area in the pupillo-constrictor reflex (Ranson & Magoun, 1933; Magoun, Atlas, Hare & Ranson, 1936).

Large-celled nucleus

In our experimental material the large-celled nucleus contained such an enormous number of heavily staining boutons (Pl. 1, fig. 2 *c*) that we may safely state that no comparable concentration of these structures can be demonstrated in any of the other primary optic end-stations after eye removal. The neuropil, which apparently consists chiefly of unmyelinated collaterals of fibres of the overlying optic tract, stains much more heavily than in normal preparations. Although the ipsilateral nucleus contains a very considerable number of boutons, the changes are greatest in the contralateral nucleus. By virtue of its greater content of argentophil elements the contralateral nucleus stains more darkly than its fellow of the opposite side, a phenomenon which is already evident under low magnification (Pl. 1, fig. 1). In general, the boutons are somewhat smaller than in the other optic centres (Pl. 1, fig. 2).

Pretectal nucleus

A considerable number of boutons had appeared in the contralateral pretectal nucleus. Boutons were most numerous in the dorsal part of the nucleus. It was striking that practically no boutons could be found in the ipsilateral pretectal nucleus.

Nucleus lateralis thalami pars posterior

This part of the thalamus is bordered by the lateral geniculate body on its lateral side and by the pretectal area on its medial side; it is covered with a great number of optic fibres. The attenuated hindpart of the nucleus extends beyond the caudal pole of the lateral geniculate body to a point dorsal to the medial geniculate body, the pretectal nucleus being applied to its medial side (Text-fig. 1).

Although a large part of the thalamus is covered with optic tract fibres, degenerating boutons were found only in that portion of the lateral thalamic nucleus which lies on a level with the pretectal nucleus. Their number was considerable in the contralateral and only slightly less in the ipsilateral nucleus. Marchi granules following retinal lesion have been described by Brouwer *et al.* (1922) in the lateral thalamic nucleus of the rabbit, and Minkowski (1920) reported bilateral cell atrophy in the posterior part of the same area of the goat and cat 6 months after enucleation of one eye. It seems that recent studies with the Marchi method have failed to confirm these observations.

Superior colliculus

Our findings in this centre largely correspond with those reported by Jefferson (1940) from Clark's laboratory. There was a large number of ring and bulb formations in the crossed colliculus, especially in the stratum griseum superficiale, but to a minor extent also in the stratum opticum and stratum zonale. Very few were present in the ipsilateral colliculus, but we cannot confirm Jefferson's statement that it was entirely devoid of abnormal boutons. Those that were present were inconsistently distributed over the three superficial layers.

Nucleus opticus tegmenti

A small number of degenerating boutons was found in this cell group and a few were present in the tractus peduncularis transversus also, very near its entrance into the nucleus. There were fewer in the ipsilateral than in the contralateral nucleus.

'Noyau de la bandelette optique'

We could identify this small cell group which Cajal (1911) described in the mouse as a centre receiving collaterals from the optic tract. It is a small nucleus situated in a shallow triangular depression of the basal surface of the cerebral peduncle. Only in one section did we observe one single ring structure, and it hardly seems possible to attach any value to this solitary finding. Our preparations suggest that the nucleus is related to the ansa peduncularis rather than to the tractus opticus.

Nucleus subthalamicus (Luys)

A connexion of the retina with the subthalamic nucleus by means of the anterior accessory optic tract has been claimed for the rabbit by Bochenek (1908) and Loepp (1912). In our preparations no boutons could be found in the subthalamic nucleus of either side. This seems to confirm the results of many later observations (see Jefferson, 1940) which tended to exclude the existence of retinal fibres ending in the corpus Luysii.

Nucleus supraopticus

In the guinea-pig, Collin (1935) described unmyelinated collaterals from the chiasma and the optic tracts, which ramified and ended with end-rings or bulbs on the surface of cells of the nucleus supraopticus. Our preparations did not show any change of the intercellular fibres from their normal appearance, and no boutons could be found in the nucleus.

DISCUSSION

The most impressive feature of our experimental material was the abundance of degenerating boutons in the pretectal area, indicating a much more important relation of the retina to this area than previous anatomical findings had led us to believe, and substantiating the physiological evidence of optic synapses in the pretectal region adduced by Ranson & Magoun (1933) and Magoun *et al.* (1936). It is of interest that while the large-celled nucleus, which contained the greatest concentration of these structures, apparently receives a considerable number of uncrossed retinal fibres, the pretectal nucleus seems to be almost exclusively related to the contralateral retina. In this respect the pretectal nucleus is comparable to the superior colliculus, which seems to favour Bodian's (1939) conception of the nucleus as a mesencephalic rather than a diencephalic structure. The large-celled nucleus, on the other hand, would seem to be more closely related to the diencephalic optic centres, since the termination of the optic nerve in the lateral geniculate body and the pulvinar is also strongly bilateral.

It was a matter of some surprise to us that we failed to find any definite localization of crossed and uncrossed boutons in the dorsal nucleus of the lateral geniculate body. It is possible that a number of degenerative ring-shaped or solid nodular swellings in the preterminal course of optic fibres masked the absence of true optic terminals in the dorsomedial part of the contralateral nucleus. It would be of interest to repeat our experiments in mammals possessing a laminated geniculate body, because more precise localization of crossed and uncrossed optic endings has proved to be possible in such forms. Although there is thus a possibility that a point-for-point relationship between related centres cannot be established by the bouton method, its value for gross localization is unchallenged, since bouton-like structures do not develop along the entire cellulo-fugal course but only in the end-stations of interrupted pathways. A striking example of this rule is furnished by the fact that the ventral nucleus of the lateral geniculate body contains practically no boutons after uni- or bilateral eye removal, although the nucleus is penetrated by a large number of optic fibres destined for the dorsal nucleus.

SUMMARY

1. Five days after removal of one eye degenerating boutons were found in the dorsal nucleus of the lateral geniculate body, the nucleus lateralis posterior thalami, the pretectal area, the superior colliculus and the nucleus opticus tegmenti.

2. Scarcely any boutons could be found in the ventral nucleus of the lateral geniculate body or in the 'noyau de la bandelette optique' (Cajal), and none at all in the corpus Luysii or in the nucleus supraopticus. In the large-celled nucleus of the optic tract (pretectal area) degenerating boutons were exceedingly numerous.

3. A considerable number of uncrossed fibres appear to end in the lateral geniculate body, the nucleus lateralis posterior thalami, and the large-celled nucleus of the optic tract, while the superior colliculus and the pretectal nucleus evidently receive very few fibres from the ipsilateral retina.

4. The material studied did not allow of any definite localization of crossed and uncrossed optic terminations in the dorsal nucleus of the lateral geniculate body. Areas receiving either crossed or uncrossed optic fibres have been demonstrated in the nucleus of rodents with other methods.

5. It remains for further study to decide whether the bouton method is of value for establishing point-for-point relationships between centres.

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EXPLANATION OF PLATE

Fig. 1. Part of the pretectal area, 5 days after unilateral eye enucleation. The large-celled nucleus (*l.c.n.*) has stained darker on the crossed (left half of the figure) than on the uncrossed side. Bodian's technique. × 50.

Fig. 2. Degenerating boutons in several of the contralateral optic centres, 5 days after enucleation of one eye. *a*, dorsal nucleus of the lateral geniculate body; *b*, superior colliculus; *c*, large-celled nucleus of the optic tract; all three taken at same magnification (× 1100). Pyridine-silver technique.

Key to lettering

c.p. commissura posterior.
l.c.n. large-celled nucleus of the optic tract.
n.p. pretectal nucleus.

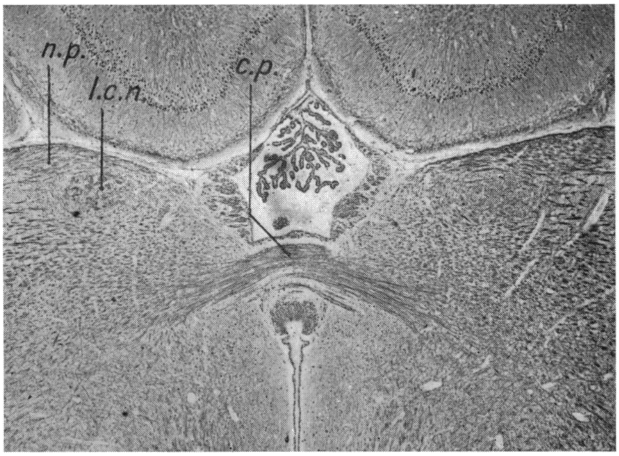
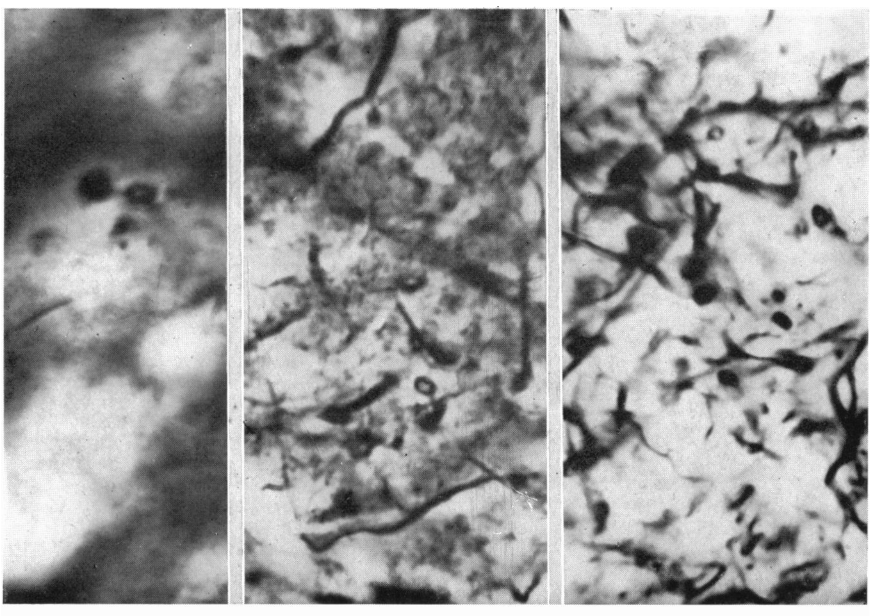


Fig. 1.



a b c

Fig. 2.

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