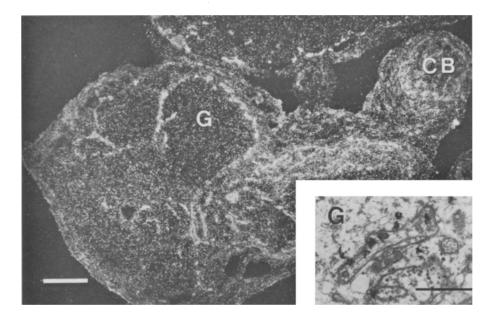
Fig. 2. Dark field radioautograph of paraffin section of Aplysia punctata buccal ganglia incubated in 5×10^{-7} M 3 H-5 HT. Intensely labelled fibres from cerebrobuccal connective (CB) in contact with a large neurone (G). Inset: Electron microscope radioautograph of a labelled fibre invaginated in the cytoplasm of G. (Bar = 50 µm); (Inset Bar = $2 \mu m$).



by incubation in 5.10⁻⁷ M ³H-NA. In both cases, the diffuse reaction persisted but preferential localizations on cell bodies and fibres disappeared. Thus these preferential reactions result from a selective uptake process, not excluding a possible labelling of 5-HT receptors. However, in a vertebrate model administration of ³H-LSD results in a diffuse labelling ¹².

Similar incubation experiments recently demonstrated that radioactivity observed in the ganglia may correspond to ³H-5 HT and/or to 2 of its derivatives (sugar conjugates)¹³. Concerning the identification of labelled structures, the serotonergic nature of C₁ cells has been well established by electrophysiological¹⁴ and biochemical methods⁵, but N₂ cells had not yet been described as serotonergic. However, identical results of test control experiments for both types of cells, together with the previously demonstrated existence in other models of a specific in vitro uptake of 5 HT at low concentration, strongly suggest that labelled neurones are truly serotonergic¹⁵. Thus labelled 5 HT terminals described in buccal ganglia may originate from either of these cells. The contacts between 5 HT terminals and buccal neurones could provide the morphological basis for the transmitter action of 5 HT at this level.

- Acknowledgment. We should like to thank Mr H.W. Cooper for correction of the English manuscript. With the help of the INSERM grant No. 77.4.178.6.
- D.J. Goldberg, J.E. Goldman and J.H. Schwartz, J. Physiol. 259, 473 (1976).
- J.E. Goldman and J.H. Schwartz, J. Physiol. 242, 61 (1974).
- D. Paupardin-Tritsch and H.M. Gerschenfeld, Brain Res. 58, 529 (1973).
- D. Weinreich, M.W. McCaman, R.E. McCaman and J.E. Vaughin, J. Neurochem. 20, 969 (1973).
- K.R. Weiss and K. Kupfermann, Brain Res. 117, 33 (1976).
- H.M. Gerschenfeld, M. Hamon and D. Paupardin-Tritsch, J. Physiol. 274, 265 (1978)
- G.K. Aghajanian and F.E. Bloom, Science 153, 308 (1966).
- A. Calas, G. Alonso, E. Arnauld and J.D. Vincent, Nature 250, 241 (1974).
- M. Lafon-Cazal, A. Calas and S. Bosc, J. Microsc. 17, 223 (1973).
- F. Larra and B. Droz, J. Microsc. 9, 845 (1970).
- J.G. Richards, Brain Res. 134, 151 (1977).
- J.E. Goldman and J.H. Schwartz, Brain Res. 136, 77 (1977).
- H.M. Gerschenfeld and D. Paupardin-Tritsch, J. Physiol. 243, 457 (1974).
- E.G. Shaskan and S. Snyder, J. Pharmac. exp. Ther. 175, 404 (1970).

Cerebellar decussation of fibres from the nucleus reticularis tegmenti pontis in the brain of the albino rat¹

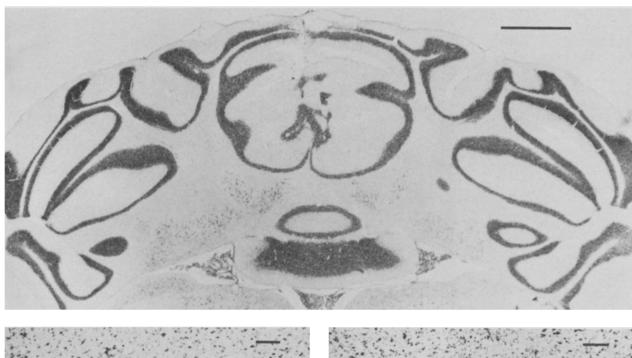
P. A. Brown² and J. B. Carman

Department of Anatomy, University of Auckland Medical School, Auckland (New Zealand), 2 January 1978

Summary. The existence of a cerebellar decussation of fibres from the medial portion of each nucleus reticularis tegmenti pontis (Rtp) of the albino rat is indicated. Definite cell loss in the medial aspect of the most rostral third of Rtp is detectable after cerebellar hemisection involving parts or the entire depth of sublobule VIb. Cell loss in the medial aspect of the caudal half of Rtp is evident as a consequence of experimental lesions which damage both sublobules IIb and III.

The nucleus reticularis tegmenti pontis (Rtp) of the mammalian brain is part of a group of nuclei within the brainstem reticular formation which is considered to project exclusively to the cerebellum³. The Rtp nucleus in particular is a relay station for impulses en route to the cerebellum from numerous regions including both the cerebral cortex and the spinal cord³.

The distribution of projection fibres from Rtp to the cerebellum has been described as both contralateral and ipsilateral⁴. The effect of unilaterally sectioning the middle



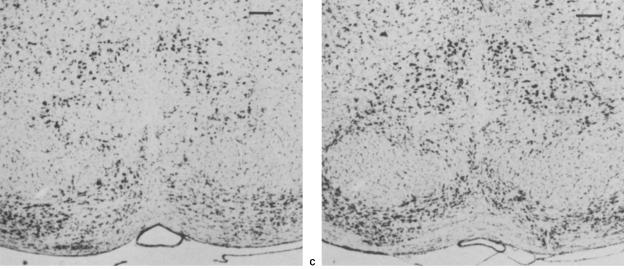


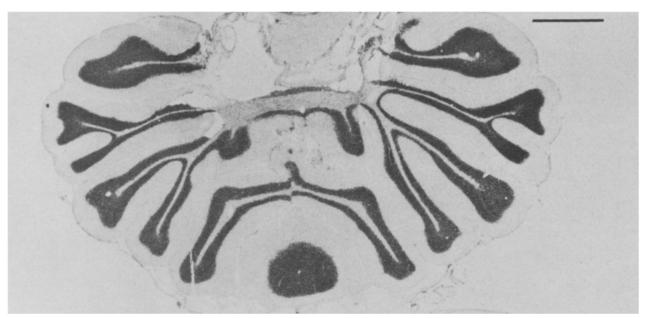
Fig. 1. A Coronal section of rat cerebellum with experimental lesion at the midline. Bar equals 1 mm. B Obvious cell loss in the medial aspect of nucleus Rtp resulting from the above lesion. Bar equals 0.1 mm. C Normal nucleus Rtp from a non-lesioned rat brain. Bar equals 0.1 mm.

cerebellar peduncle of the rat is cell loss in the contralateral lateral portion and in the ipsilateral medial portion of the Rtp nucleus⁵.

A midline decussation of Rtp fibres at the level of this bilateral nucleus as well as a decussation in the cerebellar lobules VIb and VIc of fibres which originate in Rtp have been revealed by fibre degeneration techniques⁵. Consequently, in order to elucidate the additional point of whether the decussation in the cerebellum consists of fibres from both the medial and lateral portions of Rtp or from only one division, a retrograde cell degeneration study has been undertaken.

Materials and methods. 12 male Wistar albino rats 6 weeks old were used. The animals were deeply anesthetized with ether prior to the operation. After removal of a portion of the interparietal and supraoccipital bones at the midline, lesions were made by hemisecting the underlying midline

lobules of the cerebellum with a fine scalpel to differing levels, as confirmed by histological examination. After a survival period of 8 weeks, the animals were deeply anesthetized and perfused through the aorta with 10% formalin. The brains were removed from the crania and immersed in a solution of 2% acetic acid in 70% ethanol for 3 weeks. The brains were then dehydrated in a graded series of ethanol, transferred to chloroform for 48 h, and vacuum embedded in paraffin wax. Each brain was sectioned symmetrically after determining optimum plane orientation. Coronal or horizontal sections 15 µm thick were cut. A series of 1-in-5 sections was stained with thionin⁶. A 2nd series from each brain of all sections containing the Rtp nucleus (rostrocaudal length of 1 mm) was collected and also stained with thionin. Photomicrographs of Rtp at comparable levels in each brain were taken to compare the relative cell population densities. Rat brains without lesions were used for comparison controls.



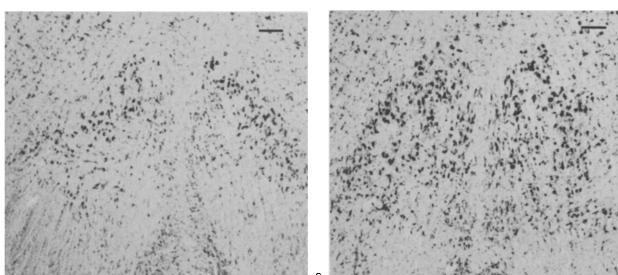


Fig. 2. A Horizontal section of rat cerebellum with experimental lesion at the midline. Bar equals 1 mm. B Obvious cell loss in the medial aspect of nucleus Rtp resulting from the above lesion. Bar equals 0.1 mm. C Normal nucleus Rtp from a non-lesioned rat brain. Bar equals

Results and discussion. Of the 12 lesioned brains, 8 brains which had received lesions mainly to sublobule VIb showed a definite cell loss in the medial aspect of Rtp involving the most rostral third of the rostrocaudal extent of the nucleus, as compared to the control brains (figure 1, A-C). In addition, 4 brains with lesions to sublobules IIb and III exhibited cell loss in the medial aspect of the caudal half of the Rtp nucleus (figure 2, A-C).

Since brains with lesions to the midline lobules show cell loss in mainly the medial portion of Rtp rather than the lateral aspect, these results indicate that there are differences in the origin of fibres which make up the Rtp cerebellar fibre decussation. In addition, because lesions to sublobule VIb result in cell loss in the rostral portion of Rtp, whereas lesions involving sublobules IIb and III result in cell loss mainly to the caudal portion of Rtp, a topographical pattern of fibre projections from Rtp to the cerebellum may exist. It is known that the Rtp fibres which

cross at the level of the nucleus originate in the lateral and/or middle portions of Rtp5.

- This work was supported by the Medical Research Council of New Zealand.
- Acknowledgments. A special acknowledgment to Mrs Heather Reid for her excellent preparation of the histological specimens. Grateful thanks to Mrs Maureen Owen for her careful typing assistance.
- A. Brodal, in: The Reticular Formation of the Brain Stem: Anatomical Aspects and Functional Correlations, p. 87. Oliver and Boyd, Edinburgh 1957.
- A. Brodal and J. Jansen, J. comp. Neurol. 84, 31 (1946). J.M. Church, Thesis, University of Auckland, New Zealand
- 5 1975.
- H.J. Conn, M.A. Darrow and V.M. Emmel, in: Staining Procedures, p. 94. Williams and Wilkins, Baltimore 1965.