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Succinate dehydrogenase activity in the telencephalon of crocodiles correlates with the projection areas of sensory thalamic nuclei

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Telencephalic auditory and visual regions in crocodiles have been identified by following the course and distribution of degenerating axons that result from stereotaxic lesions of certain neuronal aggregates in the thalamus. One such area, nucleus reuniens pars centralis, which receives bilateral auditory input from the central nucleus of the torus semicircularis[7], projects to a caudomedial portion of the dorsal ventricular ridge[8]. Another diencephalic region, nucleus rotundus, which receives bilateral visual afferents from the optic tectum[2], projects to an anterolateral portion of the dorsal ventricular ridge[9]. Furthermore, the dorsal ventricular ridge of crocodiles, as well as that of other reptiles and birds, shares certain properties in common with neocortical areas in mammals[5,10]. In fact, some have suggested that the dorsal ventricular ridge of birds and reptiles is homologous to certain neocortical areas in mammals[5]. Regardless of its similarities with mammalian neocortex, the dorsal ventricular ridge of crocodiles merits study because it is the highest sensory integration center in this group of reptiles.

Unfortunately, distinct cyto- and myeloarchitectonic properties which readily distinguish auditory and visual regions in the midbrain[7] and thalamus[7,9] of *Caiman* could not be demonstrated in the dorsal ventricular ridge[8,9]. While subdivisions of the dorsal ventricular ridge have been identified in crocodiles[11,12], they do not correspond precisely with the distribution of sensory thalamic efferents in the telencephalon of *Caiman*[8,9]. Thus, an approach based on properties other than cyto- or myeloarchitecture was sought.

The importance of the identification of such sensory areas in the dorsal ventricular ridge of *Caiman* is 3-fold. First, since small thalamic lesions were made in previous experiments[8,9], damage to nucleus reuniens pars centralis and to nucleus rotundus was subtotal. Consequently, only a portion of each respective telencephalic projection area was identified. Thus, a technique that would correlate with the telencephalic projection areas of these thalamic nuclei might demonstrate the full extent of these regions in the dorsal ventricular ridge. Second, the ability to determine such areas in descriptive material might allow similar identification in other planes of section, e.g. sagittal, which

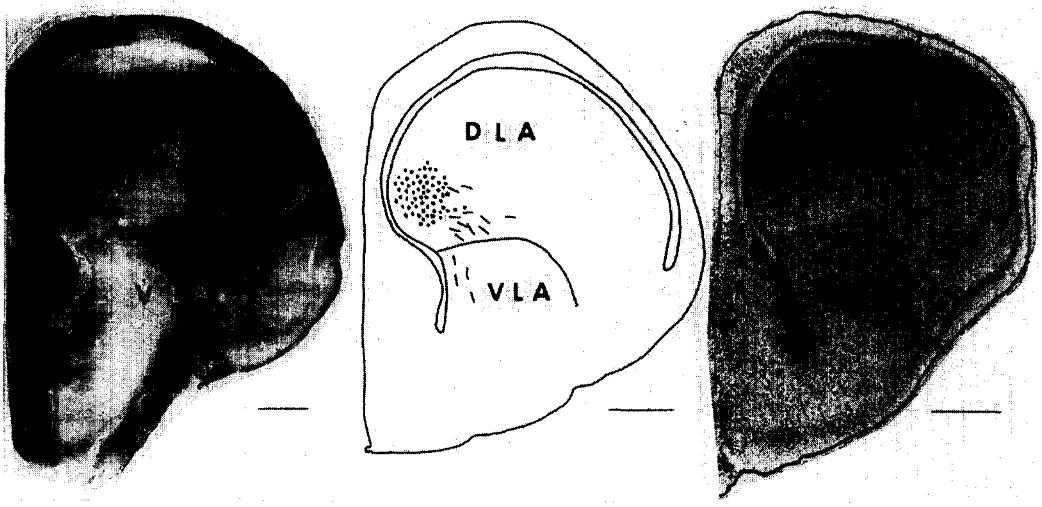

Fig. 1. Telencephalic auditory area. Sections stained for succinate dehydrogenase and with cresyl violet are shown in the left and right panels, respectively. The middle panel is a charting of a comparable section stained with the Fink–Heimer method to demonstrate the telencephalic projection area of nucleus reuniens pars centralis. Dots in this middle panel represent axon terminals while short line segments represent degenerating axons. Note the correspondence between regions high in succinate dehydrogenase activity (dark area in left panel) and the locus of termination of reuniens axons (middle panel). The lack of distinct cytoarchitectonic boundaries that correspond precisely with the telencephalic projection area of nucleus reuniens pars centralis can be seen in the photograph of the section stained with cresyl violet. DLA and VLA are abbreviations for the dorsolateral and ventrolateral area. The dorsolateral area corresponds to the dorsal ventricular ridge. Bar scale represents 1 cm.

might disclose relationships not previously appreciated. Third, a method based on non-experimental techniques that might be generalizable to other reptilian groups would circumvent the tedious and difficult process of the anterograde determination of thalamo-telencephalic connections. For these reasons, we investigated several techniques that might correlate with the telencephalic distribution of afferents from nucleus reuniens pars centralis and nucleus rotundus. We found that regions high in succinate dehydrogenase activity correlate with these telencephalic sensory areas. The present report describes our results.

Caiman crocodilus were prepared and stained for succinate dehydrogenase activity according to techniques described in detail elsewhere[6]. Selected sections were then projected by a microprojector and were compared with Fink–Heimer stained material of lesions of nucleus reuniens pars centralis[8] and nucleus rotundus[9] as well as with sections at similar brain levels that were stained for cells and fibers. This analysis demonstrated that areas rich in succinate dehydrogenase activity located in the caudomedial and anterolateral dorsal ventricular ridge correlated with the telencephalic projection sites of nucleus reuniens pars centralis and nucleus rotundus. Of the two sensory areas in the dorsal ventricular ridge, the telencephalic projection site of nucleus rotundus proved the most readily identifiable. A comparison between histochemical sections and those stained with cresyl violet and by the Fink–Heimer method is shown in Figs. 1 and 2.

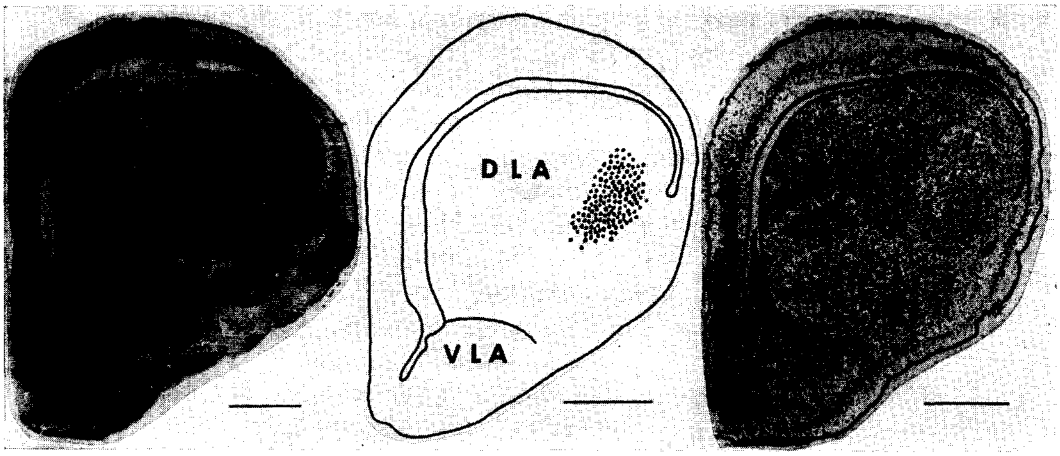

Fig. 2. Telencephalic visual area. Sections stained for succinate dehydrogenase and with cresyl violet are shown in the left and right panels, respectively. The middle panel is a charting of a comparable section stained with the Fink–Heimer method to demonstrate the telencephalic projection area of nucleus rotundus. Dots in the middle panel represent axon terminals. Note the correspondence between regions high in succinate dehydrogenase activity (dark area in left panel) and the locus of termination of rotundal axons (middle panel). The lack of distinct cytoarchitectonic boundaries, especially dorsad, that correspond precisely with the telencephalic projection area of nucleus rotundus can be seen in the photograph of the section stained with cresyl violet. DLA and VLA are abbreviations for the dorsolateral area and ventrolateral area. The dorsolateral area corresponds to the dorsal ventricular ridge. Bar scale represents 1 cm.

The present report demonstrates that certain areas in the dorsal ventricular ridge of crocodiles high in succinate dehydrogenase activity correlate with the distribution of auditory and visual efferents from the thalamus. Thus, while several studies[1,3,4] have investigated the histochemical properties of various reptilian telencephalons, the present experiment points out the significance of at least one histochemical property in a single group of reptiles. The findings of the present analysis led us to investigate histochemical properties of other brain regions in *Caiman* as well as to examine histochemical properties of the telencephalon of other reptiles. With respect to the former, difficult boundaries between certain thalamic groups in *Caiman*, e.g. nucleus reuniens pars centralis and pars diffusa, become distinct when sections are stained for succinate dehydrogenase. With respect to the latter, a caudomedial and an anterolateral area rich in succinate dehydrogenase activity are present in the dorsal ventricular ridge of other reptiles such as *Iguana iguana*. Based on our knowledge that these regions in *Caiman* represent the telencephalic projection sites of nucleus reuniens pars centralis and nucleus rotundus, these latter findings suggest that similar relationships occur in other reptiles. Furthermore, other regions rich in succinate dehydrogenase activity are present in the dorsal ventricular ridge of crocodiles. Although the source of afferents to these areas is unknown, some of these regions may represent unidentified targets of other sensory areas in the thalamus. However, additional studies will be necessary to determine two important facts. One is the correlation between histochemical properties and telencephalic distribution of efferents from thalamic nuclei homologous to nucleus reuniens pars centralis and nucleus rotundus in reptiles other than *Caiman*. The other is to determine the relationship between telencephalic regions whose afferents are unknown and histochemically distinct areas of the dorsal ventricular ridge in crocodiles as well as in other reptiles and birds. Should a close correlation be found between histochemically distinct telencephalic regions and the projection sites of thalamic and non-thalamic nuclei, then histochemistry may prove a valuable tool in the following ways. First, histochemistry might be used to circumvent the difficult and tedious studies of thalamo-telencephalic connections of nuclei in other reptiles and birds that are homologous to nucleus reuniens pars centralis and nucleus rotundus of *Caiman*. Second, histochemistry might serve to predict previously unidentified sensory and non-sensory areas in the dorsal ventricular ridge of non-mammalian amniotes.

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1 Baker-Cohen, K. F., Comparative enzyme histochemical observations on submammalian brains. I. Striatal structures in reptiles and birds, *Ergebn. Anat. Entwickl.-Gesch.*, 40 (1968) 1–41.
2 Braford, M. R., Jr., Ascending efferent tectal projections in the South American spectacled caiman, *Anat. Rec.*, 172 (1972) 275–276.
3 Kusunoki, T., The chemoarchitectonics of the turtle brain, *Yokohama med. Bull.*, 22 (1971) 1–29.
4 Masai, H., Kusunoki, T. and Ishibashi, H., The chemoarchitectonics in the forebrain of reptiles, *Experientia (Basel)*, 22 (1966) 745–746.
5 Nauta, W. J. H. and Karten, H. J., A general profile of the vertebrate brain with sidelights on the ancestry of cerebral cortex. In F. O. Schmitt (Ed.), *The Neurosciences; Second Study Program*, Rockefeller University Press, New York, 1970, pp. 7–26.
6 Northcutt, R. G., Some histochemical observations on the telencephalon of the bullfrog, *Rana catesbeiana* Shaw, *J. comp. Neurol.*, 157 (1974) 379–390.
7 Pritz, M. B., Ascending connections of a midbrain auditory area in a crocodile, *Caiman crocodilus*, *J. comp. Neurol.*, 153 (1974) 179–198.
8 Pritz, M. B., Ascending connections of a thalamic auditory area in a crocodile, *Caiman crocodilus*, *J. comp. Neurol.*, 153 (1974) 199–214.
9 Pritz, M. B., Anatomical identification of a telencephalic visual area in crocodiles: ascending connections of nucleus rotundus in *Caiman crocodilus*, *J. comp. Neurol.*, 164 (1975) 323–338.
10 Pritz, M. B., Parallels in the organization of auditory and visual systems that synapse in the midbrain of crocodiles. In S. O. E. Ebbesson (Ed.), *Comparative Aspects of Telencephalic Organization*, Plenum Press, New York, in press.
11 Riss, W., Halpern, M. and Scalia, F., The quest for clues to forebrain evolution — the study of reptiles, *Brain Behav. Evol.*, 2 (1969) 1–50.
12 Rose, M., Histologische Lokalisation des Vorderhirns der Reptilien, *J. Psychol. Neurol. (Lpz.)*, 29 (1923) 219–272.