

DISTRIBUTION OF MONOAMINERGIC NEURONS IN THE BRAINSTEM OF THE LITTLE SKATE,
RAJA ERINACEA

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We re-investigated the localization of monoaminergic neurons in the brainstem of cartilaginous fish. Questions of specific interest to us were (1) What are the regional distribution patterns of catecholaminergic and serotonergic neurons? (2) What is the morphology of the monoaminergic neurons and how are they positioned with respect to the brain ventricles? (3) How does the chemoarchitectonic organization of the elasmobranch brain compare to that of other vertebrates?

Skates (*Raja erinacea*) of both sexes weighing two to three pounds were used in this study. Anesthetized (12 mg/kg sodium pentobarbital, i.v.) skates were perfused through the conus arteriosus with approximately 800 ml of phosphate buffered (pH 7.2) elasmobranch (E)-Ringer containing 4% paraformaldehyde. The brains were removed from the cartilaginous skull, postfixed in perfusion fluid for 2 to 4 hours, and transferred to E-Ringer containing 28% sucrose. Serial frozen sections were cut at 30 μ m in transverse, horizontal, and sagittal planes. Adjacent series were processed for Nissl staining and immunocytochemistry. The primary antisera was either anti-tyrosine hydroxylase (TH; Eugene Int.) for the detection of catecholaminergic neurons, or anti-serotonin (5-HT; Immunonuclear) which labels indolamine-containing neurons and their processes. The indirect peroxidase anti-peroxidase staining procedure was carried out employing biotinylated secondary antisera (Vector Inc.).

Numerous groups of monoamine containing neurons were localized in the ventral brainstem of the skate. Catecholaminergic groups predominated rostrally, whereas caudal to the level of the third cranial nerve only 5-HT positive neurons were observed (Fig. 1). A small population of monoaminergic neurons occurred in the telencephalon. In the diencephalon several clusters of TH positive neurons were mapped. Dorsally, a small aggregate of cells was located ventral to the habenula and fasciculus retroflexus within the dorsomedial thalamus. Four additional groups were observed in the hypothalamus (Fig. 1). Two groups of TH positive cells were found in the mesencephalon. The largest of the two was localized in the ventral midline region and it distributed primarily within nucleus interpeduncularis, pars dorsalis (Fig. 1b). Caudally, this midline group of TH positive cells extended up to the level of the exiting third nerve. Neurons within this group formed a sagittally placed sheet of small, closely spaced cells that projected their dendrites primarily in dorsoventral and rostrocaudal planes. The second midbrain group extended laterally from the first. It was bordered medially by the brachium conjunctivum and nucleus ruber, ventrally by fasciculus basalis telencephali and laterally by nucleus lateralis tegmentalis. The relationship to the ventricular system differed between indolamine and catecholamine containing neurons. Only 5-HT positive cells were found in the ventricular layer. These were groups of specialized neurons that were bottle shaped and projected processes into the lumen of the third ventricle (Fig. 2a). TH positive cells occurred in subventricular regions and they had simple bipolar or triangular shapes (Fig. 2b).

Monoaminergic neurons of mammals distribute primarily in the "paracore" (Nieuwenhuys, R.: Chemoarchitecture of the Brain. Springer-Verl., Berlin, 1985). This neurochemical segregation also occurred in skates. The monoaminergic neurons of the ventricular core, which project dendritic processes into the cerebral spinal fluid, form an exception. (Supported by NIH grant NS-13747).

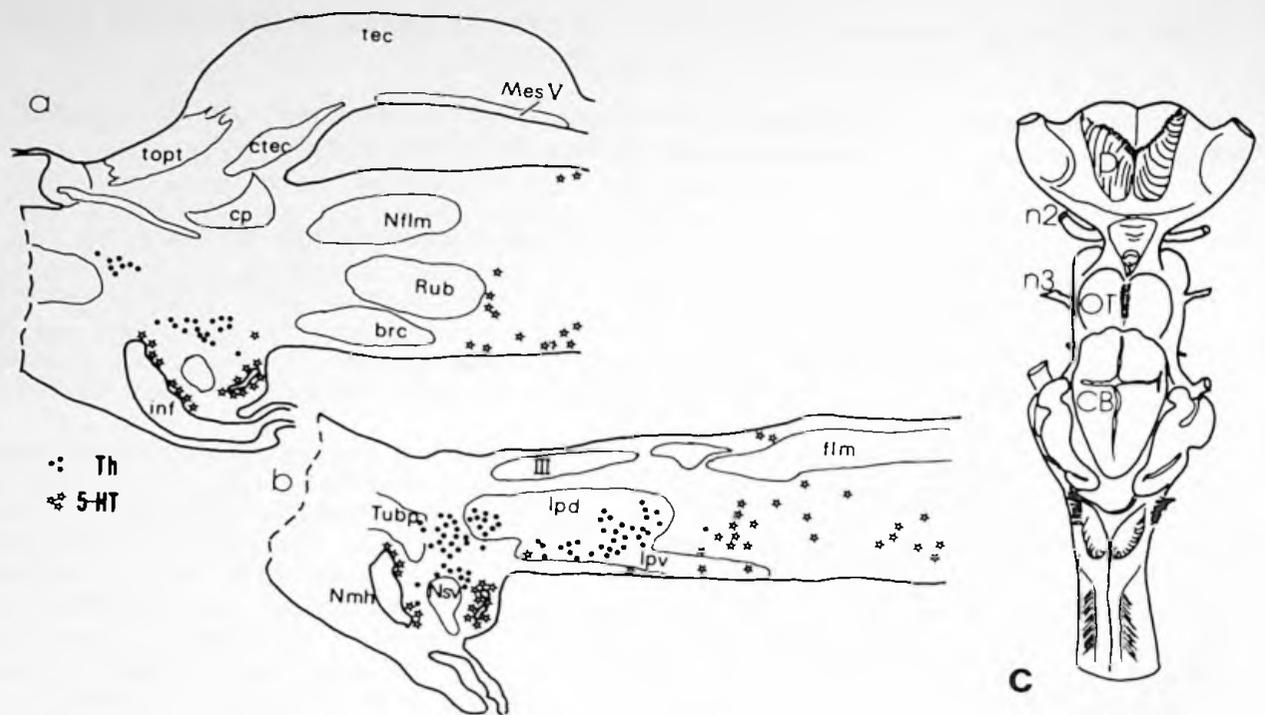


Figure 1. Camera lucida tracings of monoaminergic neurons in the brainstem of *Raja erinacea*. (a) parasagittal, (b) sagittal plane. (c) Dorsal view of the skate brain. brc= brachium conjunctivum; CB= cerebellum; cp= commissura posterior; ctec= commissura tecti mesencephali; D= dorsal pallium; flm=fasciculus longitudinalis medialis; Ipd= nucleus interpeduncularis, pars dorsalis; Ipv= nucleus interpeduncularis, pars ventralis; inf= infundibulum; MesV= nucleus mesencephalicus nervi trigemini; Nflm= nucleus fasciculi longitudinalis medialis; NmH= nucleus medius hypothalami; Nsv= nucleus sacci vasculosi; n2= nervus opticus; n3= nervus oculomotorius; OT= tectum mesencephali; Rub= nucleus ruber; topt= tractus opticus; Tubp= nucleus tuberculi posterior.

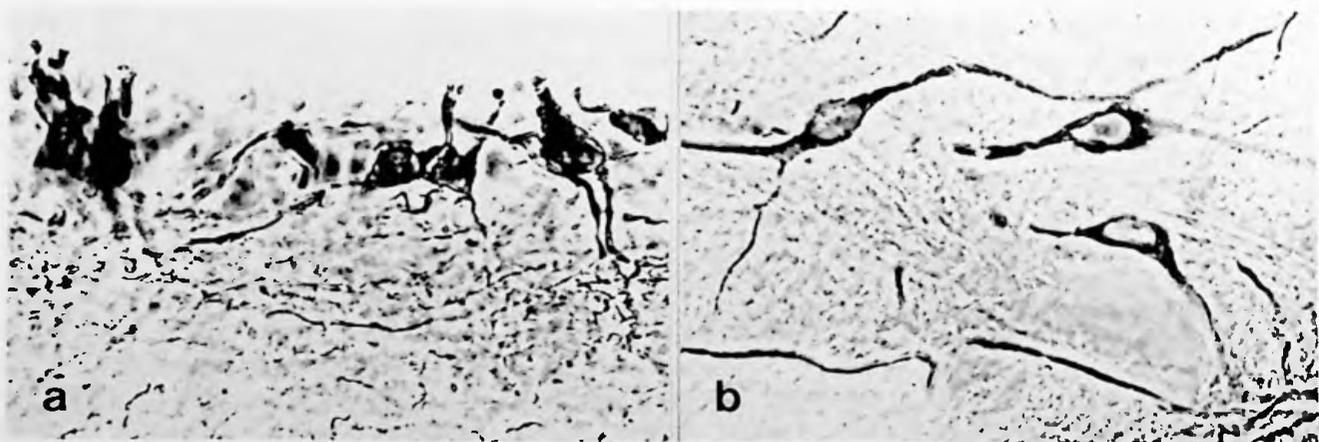


Figure 2. Photomicrographs of two types of monoaminergic neurons in the skate. (a) Bottle shaped 5-HT positive neurons in the ventricular layer of the hypothalamus. (b) Bipolar shaped Th positive neurons in the lateroventral mesencephalon.