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## **THE ARBORIZATION OF AFFERENT TERMINALS IN THE BRAIN STEM AUDITORY NUCLEI**

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In the brain stem the auditory pathway originates from the cochlear nucleus (CN). The principal CN efferents ascend to the Superior Olivary Complex, to the nuclei of the Lateral Lemniscus (LL) and to the Central Nucleus of the Inferior Colliculus (CIC). The nuclei of LL - Ventral Nucleus of LL (VNLL), Dorsal Nucleus of LL (DNLL) receive ascending projection from the auditory brain stem nuclei, including the contralateral cochlear nuclei, ipsilateral MSO, ipsi- and contralateral LSO. The DNLL has connection through the commissure of Probst with the contralateral DNLL. The VNLL receives descending projection from ipsilateral CIC, while the DNLL from both the ipsi- and contralateral CIC.

The use of anterograd tracer and its combination with retrograd labeling makes it possible to determine more precisely the termination of the labelled fibers, their arborization pattern and topography. In our experiments, for anterograd labeling PHA-L (phaseolus vulgaris leucoagglutinin, Vector 2,5% in TBS, pH 7,4) and/or BDA (biotynilated dextran amine, 20% in saline), was administered with iontophoretic microinjections (5uA, 7seconds on /7seconds off cycle, 20 minutes) for retrograde labeling HRP (peroxidase from horseradish, type VI. Sigma, 30% in saline) with Hamilton syringe pressure injections were used in cat and rat. The albino rats weighing between 200-250g of either sex, were anaesthetized with a mixture of Ketamine hydrochlorid (Calypsol) and Xylazium (Rometar) and adult cats (1,5-2,5kg) with Nembutal. The survival time for PHA-L 14, for BDA 7 and for HRP experiments was 2 days. In combined (antero and retrograd labeling) experiments the second (ie. HRP) injection was carried out on the 12 and 5 days respectively. Following the survival time the animals were deeply anaesthetized and perfused transcardially with 50-100ml saline immediately followed by 300-1500ml of a mixture of 4% paraformaldehyde, 0,1% glutaraldehyde and 0,2% picric acid. The brains were cut with Vibratome into 60um thick sections and processed according to established immunocytochemical procedure and examined with light and electron microscope. The injection sites were determined in cresyl violet stained sections.

In the first group of experiments HRP was administered into the CIC. Transversally oriented fusiform and multipolar labelled cells were found in the ipsilateral VNLL. In the ipsilateral DNLL the transversally oriented fusiform and in a lesser number the multipolar, while in the contralateral DNLL the multipolar projection neurons contained the HRP granules. In the anterograde experiments, fibers of large caliber originating from the contralateral CN were observed in a topographical order: injection sites placed along the dorso-ventral axis in the ventral and dorsal CN, labelled afferents were in medio-lateral arrangement in the nuclei of the LL. In double labeling experiments, the cochlear efferents running in parallel thin layers, traversing the transversally oriented fusiform cells polar dendrites and giving off short side branches, were observed. In the VNLL the labelled terminals, containing round synaptic vesicles make contacts with both soma and dendrites of fusiform and multipolar neurons. Some terminals were in synaptic engagement with small, pale dendritic protrusions. In the DNLL the EM results were similar to those found in

the VNLL when the injections were in the same places, ie. anterograde tracer in the contralateral CN and the HRP injection in the ipsilateral CIC. In the case when the HRP was injected similarly to the anterograde tracer, into the contralateral CIC, there was no change in the distribution of labelled terminals in the VNLL, but of course without HRP labeling. In the DNLL the labelled terminals were in synaptic contact with the dendrites and soma of multipolar neurons containing the HRP reaction product, as well as with the unlabelled fusiform cells. Besides the labelled terminals, containing round synaptic vesicles, unlabelled terminals with pleomorphic and/or flattened vesicles were observed making contact with both soma and dendrites in the DNLL. The course of labelled fibers, their distribution and arborization, ascending from the CN appeared to be similar in both cat and rat.

Recent morphological and physiological evidences indicate that the nuclei of the lateral lemniscus are important structures for central processing of auditory information and the decussating axons originating from the DLL may represent the anatomical basis of the inhibitory influence on the contralateral CIC.