ULTRASTRUCTURAL STUDY OF THE SYNAPTIC GLOMERULI IN THE RAT'S COCHLEAR NUCLEUS

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Summary

The granule cell domain of the cochlear nucleus complex contains interneurons, which are the targets for nonprimary auditory inputs from the superior olivary complex, inferior colliculus, auditory cortex, cuneate and trigeminal nuclei of the somatosensory system. The cellular targets of the non-primary projections are unknown due to a lack of information regarding postsynaptic profiles in the granule cell areas. In the present paper, we examined the synaptic relationships between a heterogeneous class of large synaptic terminals, called mossy fibers and their targets within subdivisions of the granule cell domain. During the late stage of postnatal development, we observed heterogenous groups of complex synaptic glomeruli. Using electron microscopy, we provide evidence for ultrastructural features of dendrites that receive input from the mossy fibers. The distinct synaptic relations between mossy fibers and dendrites of microneurons further imply fundamentally separate roles in processing of acoustic signals.

Key wards: cochlear nucleus, granule cell domain, synaptic glomerulus

Introduction

The granule cell domain of the cochlear nucleus comprises up to seven different subdivisions [1]. These subdivisions are found around the core of the ventral cochlear nucleus (VCN) and the dorsal cochlear nucleus (DCN), including the superficial layer of the VCN, the lamina dividing DCN from VCN, the subpeduncular dorsal corner of the VCN, the dorsal strial corner of the DCN, and layer II of the DCN [2, 3].

The granule cells and other microneurons (unipolar brush cells – UBCs, chestnut and Goldgi cells) integrate a wide spectrum information about attention, head position, sound localization, and
sound recognition [1, 4-6]. The afferent organization of these inputs and the identity of their postsynaptic targets are important for understanding the neuronal mechanisms, underlying such diverse operations. The granule cells and other microneurons do not receive terminals from the myelinated auditory nerve fibers but, instead, are the target for a variety of non-primary inputs [6]. The granule cell areas receive projections from neurons in higher auditory nuclei, including the inferior colliculus [7, 8] and the primary auditory cortex [3, 9], as well as from the olivocochlear neurons [10]. The type II auditory nerve fibers, which carry information from the outer hair cells of the cochlea also terminate inside the granule cell domain, while the myelinated type I nerve fibers do not [2]. The granule cell domain also receives non-auditory inputs, including projections from the somatosensory cuneate nucleus [11, 12], the trigeminal nuclei [13, 14], and the vestibular organ [15, 16].

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The aim of our study was to describe morphological diversity of synaptic glomerulus in granule cell lamina between VCN and DCN, and the strial corner of granule cells located external to the dorsal acoustic stria at the dorsomedial pole of the DCN, in cochlear nucleus complex of the white rat.

Materials and Methods

Ten Wistar rats aged from 15 to 25 days were used. The experiment was approved by the Animal Care and Use of Laboratory Animals section of the Ethical Committee of the Medical University – Pleven, based on the principles described in the Guide for the Care and Use of Laboratory Animals. All procedures were performed under deep anesthesia with pentobarbital sodium (50 mg/kg i.p.). Transcardiac perfusion with a mixture of 2% glutaraldehyde, 1% paraformaldehyde and 0.015% CaCl2, in 0.1 M cacodylate buffer (pH 7.2-7.4) was performed. The brain tissue were removed and postfixed for 2 h in the same solution. The material was immersed in 1% OsO4 for 1 h, then was dehydrated in a graded series of ethanol concentration, embedded in Durcupan resin and orientated for radial plane of sectioning. Adjacent ultrathin sections (thickness 500 Å) were obtained by LKB Ultratome and material was collected on mesh grids. Ultrathin sections were double-stained with 4% uranylacetate and 0.1% lead citrate, and observed with Tesla 500 BS transmission electron microscope.

Results

The mossy fiber glomerulus was a clearly defined structure of the granule cell domain. We focused on one distinct class of synaptic endings in the granule cell domain – the mossy fiber endings, located in the center of each synaptic glomerulus. Mossy fibers endings were large, vesicle-filled terminals, surrounded by postsynaptic dendrites and distributed in granule cell areas. The most common type of mossy fiber profile (mf) in granule cell areas consisted of a large central mossy fiber terminal surrounded by small, round dendritic profiles (D) containing microtubules and mitochondria (Figure 1). The dendritic profiles were occasionally seen in longitudinal section (D1), revealing that they were actually elongated claw-like structures (Figure 1-A; Figure 1-B). In cross section, these claws appeared as oblong profiles around the perimeter of the mossy fiber (indication D, Figure 1). The dendrites were smooth and without spines, usually had round-to-oblong shapes when viewed in transverse section. Terminals displayed asymmetric membrane specializations, where the postsynaptic density was more prominent than the presynaptic thickening (Figure 1, arrowhead). This asymmetry and the presence of numerous round synaptic vesicles in the central terminal suggest an excitatory synapse. Other elements in this interaction were small mossy fiber terminals (mf1) (Figure 1-B) or axons (Ax) (Figure 1-A). Most of glomeruli were enveloped in glial processes (gl) (Figure 1-C; Figure 1-D).
Figure 1. Electron micrograph of a mossy fiber glomerulus. The central mossy fiber (mf) is surrounded by granule cell dendrites (D), and axon terminals (Ax) in the periphery small mossy fibers (mf1). There are synaptic contacts (arrowheads) between the mossy fiber and granule cell dendrites. Myelinated axons (Ax) are prominent but do not interact with the glomerulus. Figure 1-A, B, C Mag. 10000x; Figure 1-D Mag. 12000x

Discussion

On the basis of structural features of the relationships between mossy fibers and these small dendrites [17, 18] and especially in the cases, where the postsynaptic target is clearly a stereotypic granule cell dendrite, the most logical interpretation is that predominant postsynaptic targets are granule cell dendrites, as viewed from various perspectives.

These dendritic claws are characteristic of the granule cells [18]. The presence of these claws plus the morphology of the dendrites strongly suggest that they are granule cell dendrites [1, 17]. The morphology of the postsynaptic dendrites is not consistent with UBCs, which contain ribosomes and dense core vesicles in their dendritic stalks [3], and emit large, tufted dendrites that completely enclose a single mossy fiber terminal. Chestnut cells are not likely candidates, because such cells are mostly adendritic, and their postsynaptic processes are void of mitochondria and thread-like tendrils [18].

The mossy fiber glomeruli described in this paper were similar to those described in the cerebellum, and we concluded that the major postsynaptic targets are the granule cell dendrites.

The large endings, located in centre of each synaptic area in the cochlear nucleus, were named mossy fiber endings because of their similarity to the mossy fiber rosettes of the cerebellum [17, 19-21].

The numerous structural similarities between the granule cell systems of the cochlear nucleus and the cerebellum have led to speculation about possible similarities in function. For example, both the cerebellum and the dorsal cochlear nucleus have a primary sensory input from the inner ear that is supplemented by a large population of mossy fibers from diverse sources.
In both systems, the mossy fibers synapse on granule cell dendrites; the granule cells then send their axons toward the pial surface, where they branch and run as parallel fibers, perpendicular to the planar orientation of the dendrites, residing in the molecular layer. In the cerebellum, the parallel fibers synapse on the inhibitory Purkinje cells, whose axons, in turn, synapse on excitatory neurons in the deep cerebellar nuclei [22]. In the cochlear nucleus, the parallel fibers synapse on both the inhibitory cartwheel neurons and the excitatory pyramidal neurons [4, 17, 23]. The cartwheel neurons, which have many anatomical, developmental, and neurochemical similarities to the Purkinje neurons [24], project onto the pyramidal neurons [23]. The pyramidal neurons, therefore, may be analogous to the deep cerebellar nucleus neurons, because both cell types are the primary output neurons for the DCN and the cerebellum, respectively.

References

12. Kanold PO, Young ED. Proprioeptive infor-