A STUDY ON THE MOSSY FIBER ENDINGS IN THE HIPPOCAMPAL FORMATION BY ELECTRON MICROSCOPIC AUTORADIOGRAPHY.

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Since Maske (4) reported that hippocampal formation of mammals gave red color in response to injection of dithizon in vivo, depending on the presence of zinc, zinc in this area has received attention as the problem for study. Based on studies by using the light microscopic autoradiography (1,3) and the sulfide-silver method as a histochemical method of demonstration of heavy metal (6), appearance of zinc in agreement with the area of distribution of mossy fiber endings in the hippocampal formation was demonstrated. Relationship between the ultrastructure of mossy fiber ending and zinc was studied only by the modification of sulfide-silver method for electron microscopy (2, 3).

We have therefore injected Zn^{65}Cl in mouse intraperitoneally and sacrificed

FIG. 1. Electron microscopic autoradiograph of the mossy fiber ending.
E: mossy fiber ending, D: dendrite of the pyramidal cell. $\times 34,000$.

the animal 18 hours later, when Zn^{65} is concentrated most intensely in the area of mossy fiber endings according to the light microscopic autoradiographic findings (5). The brain was removed and the hippocampal formation resected. Thereafter, the specimen was fixed in glutaraldehyde solution saturated with hydrogated sulfate. After dehydration and embedding in epon, the specimen was prepared in ultrathin slices to prepare specimens for electron microscopic autoradiography. As the result, accumulation of large amount of Zn^{65} at the mossy fiber ending was demonstrated. As to the ultrastructure in the nerve ending, synaptic vesicle and mitochondria were observed. Grains of Zn^{65} were found on synaptic vesicles, and scarcely any grains were found on the mitochondria. At this stage, Zn^{65} was not found to be distributed in the granular cells of the dentate gyrus and their processes, pyramidal cells of the hippocampus and glia cells. These findings were in agreement with the findings in the study using sulfide-silver method for the demonstration of heavy metal modified for electron microscopy (2, 3). The positive substance in this reaction was distributed on the synaptic vesicles in the mossy fiber endings. This would confirm the presence of zinc, one of the heavy metals, as the substance giving positive reaction.

The chemical mode of existence of Zn^{65} which is present in the synaptic vesicle of the mossy fiber ending is entirely unknown. Zn^{65} is probably found as some form of neurotransmitter or substance intimately related with neurotransmitter.

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