EFFECT OF DIRECT ELECTRICAL STIMULATION ON SYNAPSE ULTRASTRUCTURE IN THE CEREBRAL CORTEX

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After electrical stimulation of the cortical surface in dogs and cats, marked swelling of dendritic (apical) terminals was found in the neuropil of the superficial layers. Presynaptic terminals, even those forming synapses with the swollen dendrites, were unchanged in size and in their content of submicroscopic structures.

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Ultrastructural changes in synapses have now been studied during degeneration (after proximal division of the corresponding axons) [2, 3, 5-8, 14], in cerebral edema [11], local cooling [9], changes in the surrounding temperature [3], radiation injury to the brain [12, 13], during electrical stimulation of nerve tissue cultures [4], and so on. Investigations have revealed a number of characteristic changes in the ultrastructure of synapses. For example, with degeneration the primary changes were observed in the medullated fibers and their presynaptic terminals. In response to electrical stimulation an increase was observed in the number and diameter of the synaptic vesicles and their accumulation near the presynaptic membrane was observed in synapses of the ciliary ganglion in birds [1].

In this paper the results of an investigation of the ultrastructure of synapses in the upper layers (I-III) of the cortex of adult cats and dogs in response to direct electrical stimulation of the cortical surface are compared.

EXPERIMENTAL METHOD

The current used was of the normal voltage for experiments involving stimulation of the cortical surface (10-12 V for 3 animals and 15 V for another 3 animals); stimulation was applied several times in succession for 3-5 sec at intervals of 2-3 sec. The stimulated area of cortex (together with the site of the electrodes) was excised at the time of or after stimulation as a cylinder 1-2 mm in diameter. The pieces of brain were fixed in 1.5% osmium tetroxide solution in phosphate buffer (pH 7.4). After dehydration in alcohols of increasing concentration the material was embedded in araldite. Sections cut with glass knives on an ultratome were stained with lead citrate by Reynolds' method and examined without a supporting film in the electron microscope with an accelerating voltage of 80 kV.

EXPERIMENTAL RESULTS

The neuropil of the superficial layers of the motor cortex was investigated; the corresponding area of the opposite hemisphere, which was not stimulated electrically, and also areas of the same hemisphere adjacent to the point of application of the electrodes, acted as control. The great majority of "boutons" of presynaptic terminals outlined in layer I-III of the cortex were usually 1.5-2 μ in diameter, circular or angular in shape, or less frequently slightly elongated (Fig. 1). The outlines of the dendritic terminals were approximately the same as those of the presynaptic terminals, but more frequently they were elongated and irregular in configuration. An area of neuropil from layer II of a control portion of the dog's brain is illustrated in Fig. 1. In it the presynaptic terminals were filled with synaptic vesicles, and some of them contained from 1 to 4 mitochondria. The dendritic terminals were often flattened, and one of them

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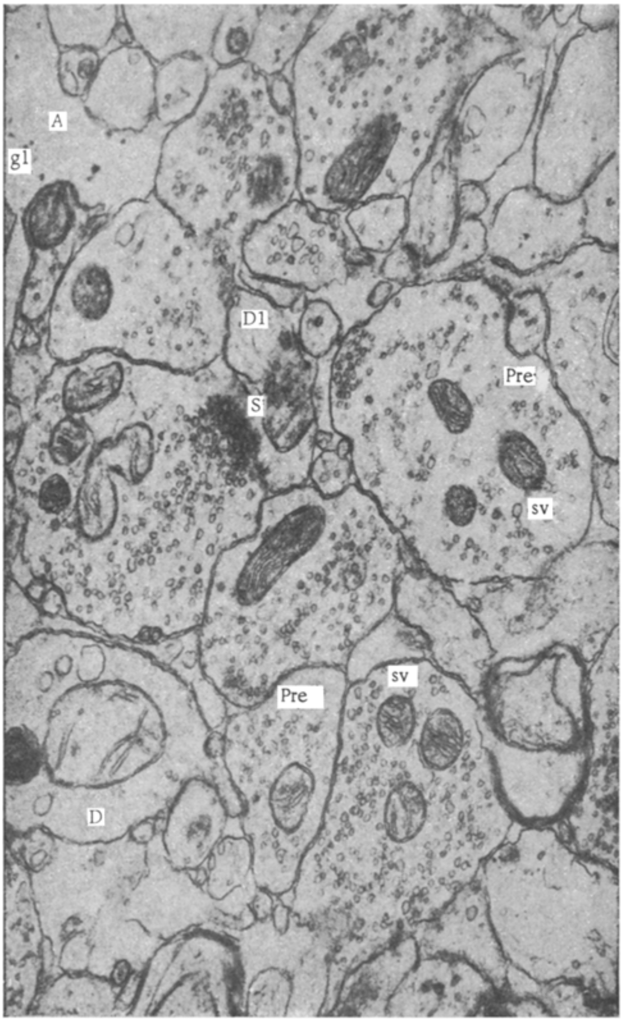

Fig. 1. Electron-microscopic picture of an area of neuropil from layer II of the motor cortex of a dog (control). Pre) presynaptic processes; sv) synaptic vesicles; D) dendrites; D1) dendrite on which a synapse (S) is formed; A) astrocytic process; gl) particles of glycogen. 30,000×.

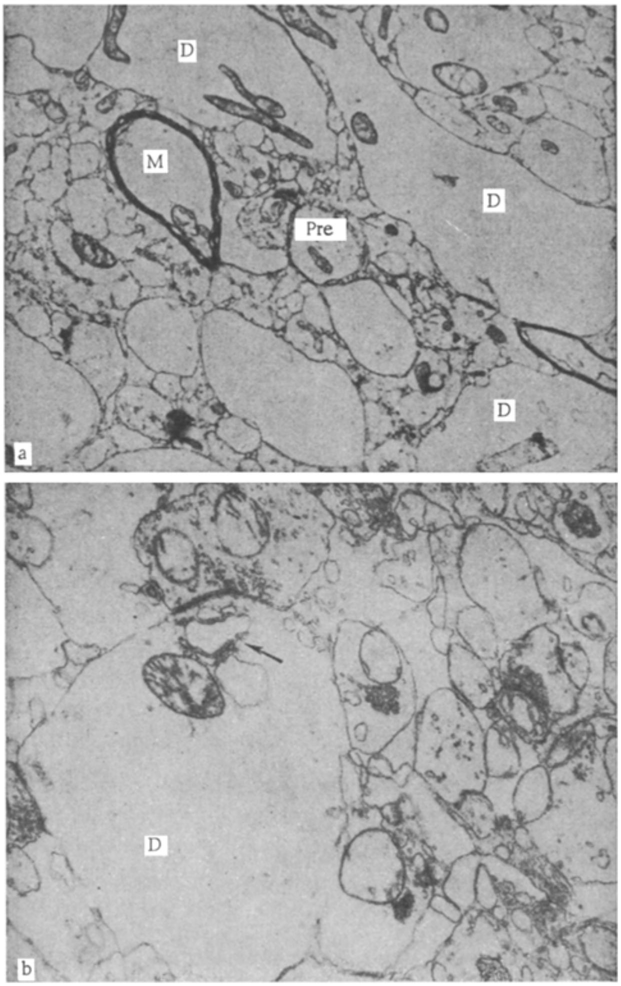

Fig. 2. Area of neuropil from layer II of the motor cortex of a dog stimulated by electrodes. a) Swollen outlines of dendrites (D). Medullated fiber (M). Presynaptic terminals (Pre). 15,000×; b) grossly swollen dendrite (D). Subsynaptic structures located in postsynaptic area (marked by arrow). 15,000 ×.

(D1) was surrounded by presynaptic terminals with the formation of a synapse. Processes of protoplasmic astrocytes of low electron density frequently contained glycogen granules.

Even under low power, after electrical stimulation many greatly widened outlines of terminals could be seen in the cortical neuropil, several times larger than in the control. These outlines (Fig. 2a and b) were discovered to be greatly swollen terminal (apical) dendrites, sometimes attaining a large size. Their cytoplasm was clear, and their organelles (mitochondria, multivesicular bodies, vacuoles of different sizes) were scattered among the widened cross-section of the terminal. Nevertheless, the subsynaptic saccules and their vesicles, and also the osmiophilic mass spreading from the postsynaptic membrane had not moved from their usual place. This possibly suggests a close connection between these postsynaptic structures and the postsynaptic membrane (Fig. 2b). In some cases the divided structures which were swollen after electrical stimulation were astrocytic processes. The first report of swelling of dendritic terminals during electrical stimulation was given by the author at the Jubilee Session of the Institute of Physiology, Academy of Sciences of the Georgian SSR, in June, 1967. Van Harreveld and Khattab [10] also observed swelling of some structures in the neuropil of mice during cooling and stimulation of the cortex; in their opinion dendrites or their spines were swollen, because synapses were found on some of them.

Despite the gross swelling of the dendritic terminals, the presynaptic terminals, even those forming synapses with swollen dendritic terminals (Fig. 2b), remained normal in size. The nature of the submicroscopic structures contained in the presynaptic terminals likewise showed no appreciable changes. The intercellular spaces were widened here and there. The nonmedullated fibers were unchanged. Hence, in response to direct electrical stimulation of the cortex, those processes of the neurons (the dendrites) in the neuropil on which synapses are formed swell, but those processes (axons) which, under natural conditions, usually carry impulses are unchanged.

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