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Calcium Transient Registration in Response to Single Stimulation and During Train of Pulses in Mouse Neuromuscular Junction

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Abstract

© 2016, Springer Science+Business Media New York.Calcium (Ca2+) is a key ion involved in transmitter release in chemical synapses. Optical recording of fluorescence changes of Ca2+ indicators is one of the most frequently used methods to measure intracellular Ca2+ dynamics. This technique is based on use of Ca2+-binding fluorescent dyes which change their emission intensity after binding to Ca2+. The most crucial step in this type of experiments is loading of Ca2+ dye. In this paper, we present the method of Ca2+-sensitive dye loading to mammalian nerve endings through the stump of the nerve. We represent Ca2+ transient registered parameters in response to a single motor nerve stimulus. The study of Ca2+ dynamics during high frequency stimulation close to real pattern of synaptic transmission allows us to understand such fundamental process as synaptic plasticity. We describe the results obtained during the registration of Ca2+ transient caused by the rhythmic motor nerve stimulation. Intracellular level of Ca2+ estimated by the amplitude of Ca2+ transient rises with the increase of stimulation frequency. The amplitude of Ca2+ transient decreases after blocking of voltage dependent Ca2+ channels by cadmium. The obtained data showed that detected increase of fluorescence intensity is induced by Ca2+ influx through the voltage-gated Ca2+ channels to the nerve ending during an action potential. This dye-loading method is suitable for registration of presynaptic Ca2+ dynamics under both single nerve stimulus and rhythmic activity.

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Keywords

Calcium transient, Fluorescence dyes, Neuromuscular junction, Optical imaging

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