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Application of Microbiosensors to Neuroscience

KENJI YOKOYAMA

RECAST University of Tokyo,
4-6-1 Komaba, meguro-ku, Tokyo 153, Japan

An in vivo acetylcholine sensor is a powerful tool for elucidating the action of a neuron. A smaller electrode causes less damage to the tissue during insertion. The carbon fiber electrodes are considered to be one of the most useful transducers for in vivo biosensors. Acetylcholine is not directly oxidized on a carbon fiber, so the immobilized enzymes are necessary to detect actylcholine. In this study, electrochemical operations for pre-electrolysis and measurement were employed for the sensitive determination of hydrogen peroxide. A carbon fiber electrode was modified with acetylcholine esterase and choline oxidase to fabricate an acetylcholine sensor.

The sensitivity of the carbon fiber electrode to hydrogen peroxide is insufficient to be applied to neuroscience. In this study, a platinized carbon fiber electrode was fabricated and employed for the highly sensitive detection of hydrogen peroxide. Glutamate oxidase was immobilized on the platinized carbon fiber electrode and was applied to the determination of glutamate, which is one of the most important neurotransmitters. The calibration graph for glutamate was evaluated. A calibration range of the ultramicroglutamate sensor was from 2 μM to 1.2 mM.

Glutamate is one of the most important neurotransmitters. Released by presynaptic neurons, it binds to specific receptors on the postsynaptic neuron. Interactions between glutamate and glutamate-receptor are strongly related to the memory storage phenomena known as long-term potentiation in hippocampus and long-term depression in cerebellar. Quantitative analyses of released glutamate are needed to clarify the molecular mechanism, especially in the long term depression phenomena. In vivo and in vitro glutamate sensors are powerful tools to elucidate the action sites in the brain. In this study, a microglutamate sensor was applied to the determination of glutamate released from cerebellar neurons.

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The response obtained after potassium stimulation was investigated. A rapid response was observed to be a peak value.

The dependence on calcium ion was also explored. In the normal Ringer solution, a high response was obtained. However, in the calcium-free Ringer solution, a very small response was observed. This small response was owing to electroactive species, such as catecholamine, because the denatured glutamate sensor responded at the same place. These results supported that the voltage-dependent calcium ion channel was related to glutamate release from cerebellar synaptic junctions.

Finally, the response to the electric pulse stimulation was investigated. The electric stimulation was applied to the white matter 1, 2, 3, 6, and 10 times every 10 s. With the increase in the pulse number, glutamate release increased. The quasi-real-time monitoring of glutamate released from cerebellar neurons could be achieved using the microglutamate sensor. This method has strong potential to be applied to analyses of interactions between glutamate and glutamate-receptor which are related to neuronal functions and phenomena like an excitatory transmission, synaptic plasticity, and ischemic neuronal damage.