

A Third-Generation Superoxide Ion Sensor

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This paper represents a third generation biosensor for superoxide anion (O_2^-) which is developed by immobilizing superoxide dismutase (SOD) on cysteine self-assembled monolayer (SAM) at gold electrode (Fig.1). A rapid and direct electron transfer of SOD was realized [1] at gold electrode by using cysteine molecule as an electron-transfer promoter and this direct electron transfer of SOD was thus utilized for the first time to develop a third-generation biosensor for sensitive and selective determination of O_2^- . At the SOD-immobilized cysteine-SAM gold electrode, O_2^- could be specifically oxidized and reduced through a catalytic reaction of SOD. The high efficiency of the direct electron transfer

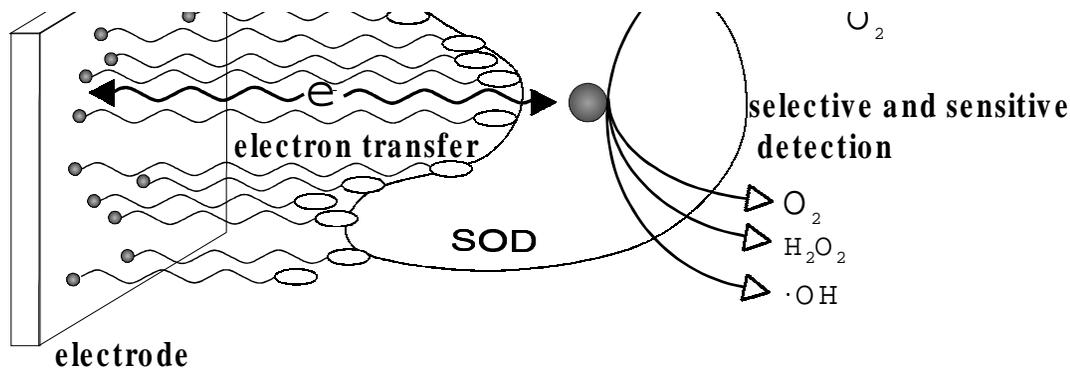
respectively. The mechanism of the sensor response and its potential dependence will be presented and discussed.

Reference:

[1] Y. Tian, M. Shioda, S. Kasahara, T. Okajima, L. Mao, T. Hisabori, and T. Ohsaka, *Biochim. Biophys. Acta*, submitted.

current measurement

Fig.1 A schematic illustration of a third-generation O_2^- sensor based on SOD-immobilized SAM modified electrodes.



of SOD and the biomolecular recognition by the exploitation of specific and significant enzyme-substrate reactivity of SOD toward O_2^- facilitated sensitive measurements of O_2^- without interference from physiological levels of ascorbic acid (AA), uric acid (UA) and metabolites of neurotransmitters. Moreover the interference from hydrogen peroxide (H_2O_2) was also minimal; for example, at +300 mV no response was observed up to 10 μM H_2O_2 and 1.7% cathodic current relative to that of 14 nM O_2^- was obtained at -200 mV. While tested with amperometry at either +300 or -200 mV the sensor response was sensitive, linear and durable; a detection limit of 5 and 6 nM was obtained with a sensitivity of 0.44 and 0.56 nA/nM and a linear range up to 200 and 220 nM at +300 and -200 mV,