Oxidation of nucleotide derivatives by ECR sputter deposited carbon films in high potential region

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ECR sputter deposited carbon films have been reported by Hirono group [1]. Electrochemical and physical properties of ECR sputter deposited films have been also observed by You and Tsuchitani et. al, respectively [2-5]. ECR film electrodes have wide potential range, low background current and good reproducibility. The nucleotide derivatives containing nucleic base are difficult to determine the oxidation peak potential because these derivatives have generally high oxidation potential and non specific adsorption of electrode surface easily. Nucleotide derivatives play an essential role in various biological processes. The second messengers such as cAMP (Adenosine 3': 5'- cyclic monophosphate) and cGMP are recognized as key molecules for carrying information in intercellular signal transduction systems. Many hormones, cytokines, or neurotransmitters promote the cellular production of cAMP by binding to their membrane receptors. The cAMP regulates a wide variety of cellular functions such as gene transcription, secretion, neuronal plasticity, cellular proliferation, differentiation, and death by activating a cAMP-dependent protein kinase. Disorders of cAMP production are also strongly related to many diseases. Thus, monitoring of cAMP concentration is important not only for the sake of understanding how cells process information but also for clinical applications and for the validation of drugs in pharmacological industries. In this work, we report on the ECR sputter deposited firm electrodes for electrochemical detection of nucleotide derivatives including cAMP.

All electrochemical experiments were carried out using ALS-660A electrochemical analyzer (CHI instruments, Austin, TX) with a traditional three-electrode cell: a | Ag | AgCl | KCl (sat.) reference electrode (Bioanalytical Systems, USA) and a platinum wire counter electrode. The ECR sputter deposited carbon films were fabricated to the disk electrodes with 1.0 mm diameter hole by the plastic seal. The electrochemical cell was housed in a Faraday cage.

Fig. 1 shows typical CVs of 5.0 mmol dm^{-3} nucleotide derivatives (cAMP, cGMP, cCMP, cTMP) in phosphate buffer solution (pH 7.40) containing 0.1 mol dm^{-3} NaCl measured with the ECR sputter deposited carbon film electrodes. All voltammetric responses on the ECR sputter deposited carbon film electrodes showed well-defined oxidation peak potential. Fig. 2 shows relationship of electrochemical response of cAMP, NADH (nicotine adenine dinucleotide), cf-DNA (calf thymus DNA) and number of DPV on a ECR sputter deposited carbon film electrode in phosphate buffer solution (pH 7.40). As a result, well-defined DPV curves toward nucleotide derivatives were observed (data not shown). With the ECR sputter deposited carbon film electrodes, the response anodic current of all nucleotide derivatives was significantly stable with increase of scan times. These findings suggest that the ECR sputter deposited carbon film electrodes is effective to inhibiting the non-specific adsorption on electrode surface. Because of its high stability and reproducibility, the ECR sputter deposited carbon film electrodes will be applying to detection of other nucleotide derivatives with higher oxidation potential or molecular weight than cAMP.

References

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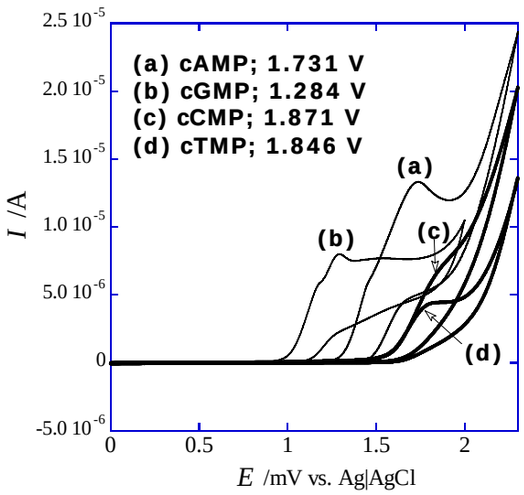

Fig. 1 Cyclic voltammograms of nucleotide derivatives on ECR sputter deposited carbon film in pH 7.40 phosphate buffer solution. (inner; oxidation peak potentials)

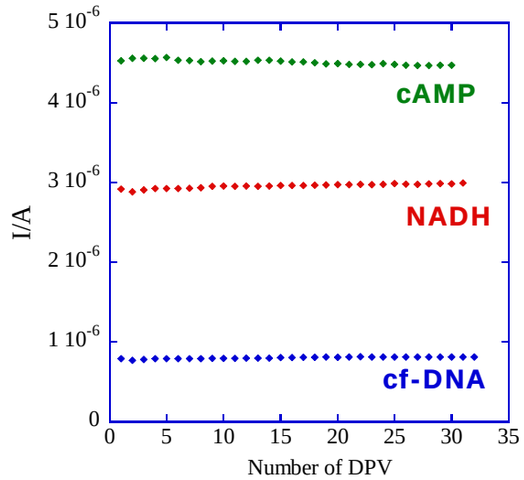

Fig. 2 Effect of adsorption for cAMP, NADH and cf-DNA on ECR sputter deposited carbon film