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Highly selective electrochemical biosensor for the determination of folic acid based on DNA modified-pencil graphite electrode using response surface methodology

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ABSTRACT

An electrochemical DNA biosensor was proposed as a screening device for the rapid analysis of folic acid using a pencil graphite electrode modified with salmon sperm ds-DNA. At first, immobilization of the ds-DNA on pencil graphite electrode was optimized using response surface methodology. Solution pH, DNA concentration, time of DNA deposition and potential of deposition was optimized each at three levels. The optimum combinations for the reaction were pH 4.8, DNA concentration of 24 μg mL^{-1}, deposition time of 304 s, and deposition potential of 0.60 V, by which the adenine signal was recorded as 3.04 μA. Secondly the binding of folic acid to DNA immobilized on a pencil graphite electrode was measured through the variation of the electrochemical signal of adenine. Folic acid could be measure in the range of 0.1–10.0 μmol L^{-1} with a detection limit of 1.06×10^{-8} μmol L^{-1}. The relative standard deviations for ten replicate differential pulse voltammetric measurements of 2.0 and 5.0 μmol L^{-1} folic acid were 4.6% and 4.3%, respectively. The biosensor was successfully used to measure folic acid in different real samples.

1. Introduction

One of the water-soluble vitamin B groups is folic acid that is a tasteless and odorless yellowish orange crystal. Folate has been identified as one of the most important vitamin for normal human metabolic function. The amount of folic acid perception is proper to neural tube defects (NTD) in newborns, cardiovascular diseases, colon cancer and certain anemia [1]. Also, folic acid is essential for cell growth and division and participates in lots of reactions in body and mainly in synthesis of nucleic acid and some important substances. It promotes the synthesis of protein from amino acid, in human body. Folate is not synthesized in humans, thus it should be obtained from dietary sources. In many countries mean folate intake was found to be lower than recommended or desire. To reduce the risk of woman of childbearing age to have a child with neural tube defect, some countries mandated folate fortification of staple food [1]. Recently, high-performance liquid chromatography (HPLC) [2,3], HPLC–MS [4], colorimetry [5], spectrophotometry [6], chemiluminescence [7], flouorimetric [8] and microbial methods [9] have been reported for the determination of folic acid. However, most of these methods are expensive and time consuming and/or suffer from many interfering compounds, and thus restrict their applications in food analysis.

It is well known that electrochemical methods are simple and inexpensive, in which analytical techniques require small amount of sample [2,10]. Electrochemical devices are easy to miniaturize, simple, and inexpensive compared with optical instrumentation [11]. Electrochemistry can overcome on the problem of coupling tiny chips with large readout optical systems. In addition, turbidity of sample is not a matter of concern and powerful sources of energy, for example lasers, are not required [12]. Indeed the use of electrochemical techniques instead of fluorescence allows for simpler and smaller detectors [13].

There are some studies about utilizing electrochemical methods for folic acid detection, but many of them are not selective, and sometimes are not sensitive sufficiently. Biosensors have higher selectivity than other sensors. One of the natural polymers is deoxyribonucleic acid (DNA) that has gained increasing attention in biosensor designing [14]. Since the discovery of electrochemical activity of nucleic acids by Palececk, huge promotion particularly by the development of electrochemical DNA biosensors based on the concept of chemically modified electrodes have been developed [12]. DNA is one of the most important biological molecules targeted by many small molecules [15]. Small molecule could bind with DNA by making covalent or non covalent binding. Intercalation between the stacked base pairs of native DNA, electrostatic interactions with the negative-charged sugar-phosphate at the out of DNA structure and binding interactions with both minor and major grooves of DNA double helix are three modes of non-covalent DNA interactions with small molecules [16,17]. The compound–DNA interaction can be irreversible, causing subsequent destabilization, breaking of the hydrogen bonds, and opening of the double helix [18]. This will lead to exposure of the DNA purine bases

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