INFLUENCE OF Ni(OH)$_2$ NANOPARTICLES ON INSULIN SENSOR SENSITIVITY

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ABSTRACT

The influence of nickel hydroxide (Ni(OH)$_2$) nanoparticles on sensitivity of insulin sensor was investigated. Ni(OH)$_2$ nanoparticles that have an average diameters of 60 nm was characterized by Transmission Electron Microscopy (TEM). Insulin sensor was fabricated from silica gel/chitosan (SiO$_2$/Chit) electrode that was modified with Ni(OH)$_2$ nanoparticles. A mixture of silica gel and chitosan in weight ratio of 4 : 6 was prepared. This mixture was heated at 65 ºC and stirred, then Ni(OH)$_2$ nanoparticles solution (2 mL/100 mg of the mixture) and paraffin (15 % from the mixture weight) were added. The mixture was stirred continuously until the mixture becomes solid (paste). The paste was inserted in the bottom of glass tube and conected to Ag wire from the other side of the glass tube. The surface of the electrode was polished by abrasion paper grade 2000. The response of SiO$_2$/Chit modified by Ni(OH)$_2$ nanoparticles (SiO$_2$/Chit/Ni(OH)$_2$nps) electrode to insulin concentration was observed. The addition of 2 mL Ni(OH)$_2$ nanoparticles could increase the sensitivity of the insulin sensor as much as sixfold compared to the electrode that was not modified by Ni(OH)$_2$ nanoparticles. A linear graph was obtained when anodic (i_{pa}) and cathodic (i_{pc}) current were plotted at varied scan rate.

Keywords: nickel hydroxide nanoparticles, sensor, insulin.

INTRODUCTION

Insulin is a hormone that produced in the pancreas to control blood glucose levels. Human insulin has a molecular weight of 5800 g/mol which consists of 51 amino acids. Disorders of insulin production in pancreas causes diabetes disease [1]. According to the data from the international diabetes federation (IDF) in 2014 there were 387 million diabetics in the world, and 9 million in Indonesia. The information about insulin level in the blood may be used to predict a person's health.

Determination of organic compound by electrochemical method have been reported, e.g insulin [2], DNA [3], cholesterol [4], glucose [5] [6] [7] and dopamine [8]. The electrochemical method has several advantages such as samples can be analyzed directly without any sample preparation, does not require chemical reagents, and sample analysis takes less than 5 minutes [9]. It is different to chromatographic and immunoassays methods which require sample preparation, analysis takes more than 2 hours and requires a lot of chemical reagent [10][11]. Several researches in insulin analysis by electrochemical method with modified electrode have been reported. Modified electrode of silica gel/carbon [12] and nickel oxide/guanine/glassy carbon [13] were used to measure insulin.

Ni(OH)$_2$ particles have been applied for some modified electrode sensors such as glucose [14] [15], and Vitamin D sensors [16]. Modified electrode of over-oxidized polypyrrole nanowire that was modified with Ni(OH)$_2$ nanoflakes has been applied for glucose sensor. This electrode has sensitivity of 1049.2 μAmM^{-1}cm^{-2} [14]. Ni(OH)$_2$ particles that modified on silicon microchannel plates has been applied for glucose sensor with sensitivity of 0.25 mAmM^{-1}cm^{-2} [15]. Silica and graphene oxide that was modified with Ni(OH)$_2$ particles has been applied for vitamin D determination with a good sensitivity [16].

The modification of electrode by Ni(OH)$_2$ increases the sensitivity of glucose and vitamin D sensor due to its electrocatalytic activity and role as electron transfer agent in the reduction and oxidation of the sample [17]. Ni(OH)$_2$ nanoparticles can be synthesized in fast and more simple method by electrolysis of nickel metal using high voltage method [18]. Thus, in this present work, the Ni(OH)$_2$nps that produced by electrolysis of nickel metal at high voltage was mixed to Si/Chit electrode and applied to measure insulin in 0.1 M phosphate buffer at pH 7.4. The influence of Ni(OH)$_2$ nanoparticles on sensitivity of insulin sensor was investigated by cyclic voltammetry.

MATERIALS AND METHODS

Materials

Silica gel G.60, K$_2$HPO$_4$, and KH$_2$PO$_4$ were received from Merck, and used without any further purification. Insulin 100 IU/mL (R. Lantus) was used as received without any further purification. Glass tube (inner diameter = 0.2 cm, outer diameter = 0.5 cm, and length = 5 cm), silver (Ag) wire (outer diameter = 0.8 mm and length = 7 cm), solid paraffin, and demineralized water were purchased from local market. Chitosan and nickel hydroxide (Ni(OH)$_2$) nanoparticles were prepared at Instrumentation and Analytical Sciences Laboratory, Institut Teknologi Sepuluh Nopember, Indonesia. Ni(OH)$_2$ nanoparticles was synthesized with high potential method [18]. Briefly, two nickel metals were used as anode and cathode, then electrolyzed in 300 mL sodium citrate solution at 100 ºC and potential of 55 V was applied. During electrolysis the solution was kept in stirring for 30 minutes, then the Ni(OH)$_2$ nanoparticles that obtained were used for the fabrication of SiO$_2$/Chit/Ni(OH)$_2$nps

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electrode. The Ni(OH)$_2$ nanoparticles were characterized by transmission electron microscopy (TEM).

All electrochemical measurements have been performed on potentiostat/galvanostat from eDaQ (potentiostat E161 and e-corder 410 which is equipped with e-chem software vs 2.0.1). A conventional three-electrode cell system was used for this work. It consist of Ag/AgCl (KCl 3 M) as reference electrode (RE), platinum wire as counter electrode (CE), and SiO$_2$/Chit/Ni(OH)$_2$nps electrode as working electrode (WE).

Fabrication of SiO$_2$/Chit/Ni(OH)$_2$nps electrode

The SiO$_2$/Chit/Ni(OH)$_2$nps electrode was prepared using the following procedure. Preparation of silica gel and chitosan composite, a mixture of silica gel and chitosan in the weight ratio of 4: 6 was prepared. This mixture was heated at 65 ºC and stirred, then Ni(OH)$_2$ nanoparticles solution (2 mL/100 mg of the mixture) and paraffin (15 % from the mixture weight) were added. The mixture was stirred continuously until the mixture becomes solid (paste). The paste was inserted in the bottom of glass tube, then conected to Ag wire from the other side of the glass tube. The electrode surface was polished by abrasion paper grade 2000. The influence of Ni(OH)$_2$ nanoparticles was investigated. The volume of Ni(OH)$_2$ nanoparticles added were varied at 1, 2, 3, 4, and 5 mL. The SiO$_2$/Chit/Ni(OH)$_2$nps electrode was used to measure 1.0 µM insulin in 0.1 M phosphate buffer pH 7.4 with cyclic voltammetry in the potential range of -1.0 to 1.0 V, and at scan rate of 100 mV/s. The responses of SiO$_2$/Chit/Ni(OH)$_2$nps electrode at different insulin concentrations and scan rate were observed. Insulin concentrations was varied at 1, 2, 3, 4, and 5 nM, meanwhile scan rate was varied at 20, 40, 60, 80, 100, 120 and 140 mV/s.

RESULTS AND DISCUSSIONS

Ni(OH)$_2$ nanoparticles has an oval shape with average diameters of 60 nm. The TEM characterization is shown in Figure-1. The increase of potential in the electrolysis process is proportional to the electron transfer in the anode. It causes the process of oxidation of Ni metal become Ni(OH)$_2$ nanoparticles occurs faster.
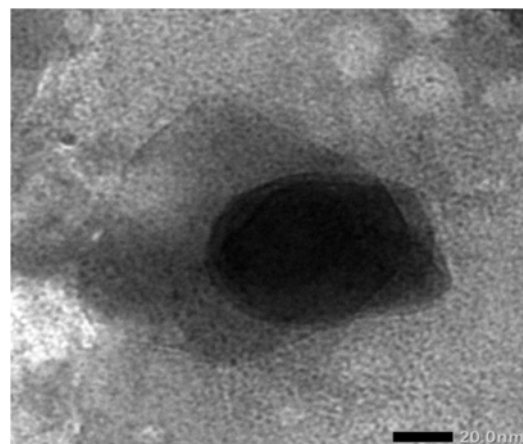

Figure-1. TEM image of Ni(OH)$_2$ nanoparticles.

The influence of Ni(OH)$_2$ nanoparticles in the SiO$_2$/Chit electrode to detect insulin was investigated. The response of SiO$_2$/Chit electrode with and without Ni(OH)$_2$ nanoparticles to measure 1.0 µM insulin in phosphate buffer pH 7.4 solution were shown in Figure-2.

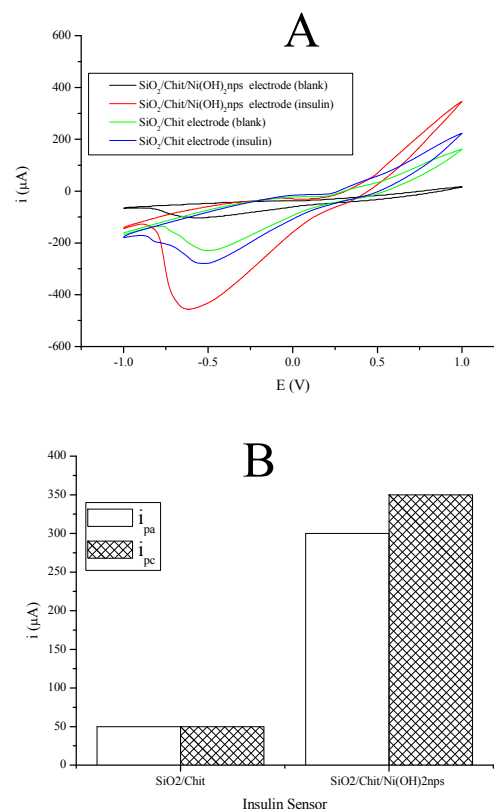**Figure-2.** (A) The Cyclic voltammogram of phosphate buffer pH 7.4 solution (blank) and 1.0 µM insulin in phosphate buffer pH 7.4 solution with scan rate 100 mV/s (insulin sample) for SiO$_2$/Chit and SiO$_2$/Chit/Ni(OH)$_2$nps electrodes. (B) The i$_{pc}$ of SiO$_2$/Chit and SiO$_2$/Chit/Ni(OH)$_2$nps electrodes.

Figure-2 shows that the oxidation of insulin occur at 0.8 V (E_{pa}) and the reduction of insulin occur at -0.57 V (E_{pc}). The anodic peak current (i_{pa}) of $SiO_2/Chit/Ni(OH)_2$nps electrode is 300 µA. Meanwhile the chatodic peak current (i_{pc}) of $SiO_2/Chit/Ni(OH)_2$nps electrode is -350 µA. The i_{pa} and i_{pc} of blank solution by $SiO_2/Chit/Ni(OH)_2$nps electrode are lower than $SiO_2/Chit$ electrode. These mean that the addition of $Ni(OH)_2$ nanoparticles in insulin sensor can improve sensitivity of the sensor as much as sixfold as shown Figure- 2(B). This is because $Ni(OH)_2$ nanoparticles has high reactivity and electrocatalytic activity [13]. $Ni(OH)_2$ nanoparticles cause the electron transfer occurs faster on the electrode surface. A possible reaction between $Ni(OH)_2$ nanoparticles with insulin on electrode surface is described in the equation (1) and (2) below: [19]

$$Ni(OH)_2 \leftrightarrow NiOOH + e^- + H^+ \qquad (1)$$

$$NiOOH + Insulin \rightarrow Ni(OH)_2 + product \qquad (2)$$
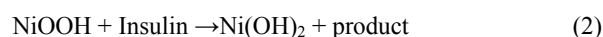
The influence of $Ni(OH)_2$ nanoparticles to the electrode response toward insulin was observed. The $Ni(OH)_2$ nanoparticles in the varied volume of 1- 5 mL in 100 mg of SiO2/Chit mixture was investigated. The results are shown at Figure-3.
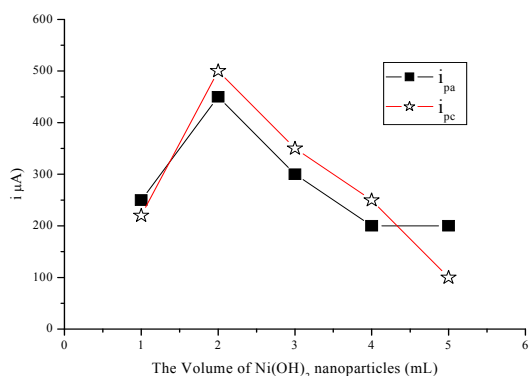
Figure-3. Influence of $Ni(OH)_2$ nanoparticles (mL/100 mg composition of electrode) in $SiO_2/Chit/Ni(OH)_2$nps electrode to measure 1.0 µM insulin in phosphate buffer pH 7.4 solution with scan rate 100 mV/s.

Figure-3 shows that i_{pa} and i_{pc} at 2 mL $Ni(OH)_2$ nanoparticles addition are higher than i_{pa} and i_{pc} of 1, 3, 4, or 5 mL addition. The addition of $Ni(OH)_2$ nanoparticles more than 2 mL causes the physical properties of the electrode were decrease. In this condition, the electron transfer between $Ni(OH)_2$ nanoparticles and insulin on the electrode surface was inhibited becasuse the electrode compactness was decrease.

The response of $SiO_2/Chit/Ni(OH)_2$nps electrode at different scan rate (20, 40, 60, 80, 100, 120 and 140 mV/s) was investigated . The cyclic voltammogram of $SiO_2/Chit/Ni(OH)_2$nps electrode to measure 1.0 µM insulin in phosphate buffer pH 7.4 solution can be seen at

Figure-4. The results show that both anodic and cathodic current proportional to increasing the scan rate.
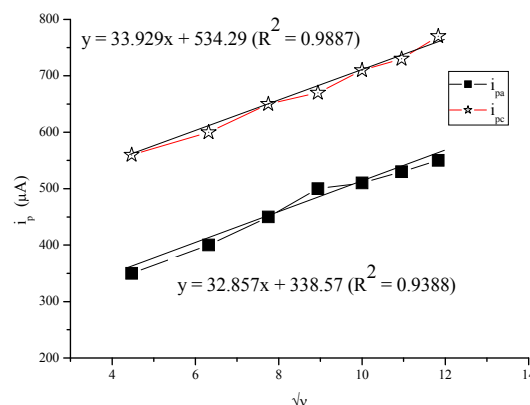
Figure-4. The response of $SiO_2/Chit/Ni(OH)_2$nps electrode to measure 1.0 µM insulin in phosphate buffer pH 7.4 solution at different √scan rate (√v).

The $SiO_2/Chit/Ni(OH)_2$nps electrode was used to measure insulin at different concentration. Linear responses were observed both in anodic (i_{pa}) and cathodic (i_{pc}) current, Figure-5. The cathodic (i_{pc}) current response was higher than the anodic (i_{pa}) current, but no significant differences observed in their sensitivity.
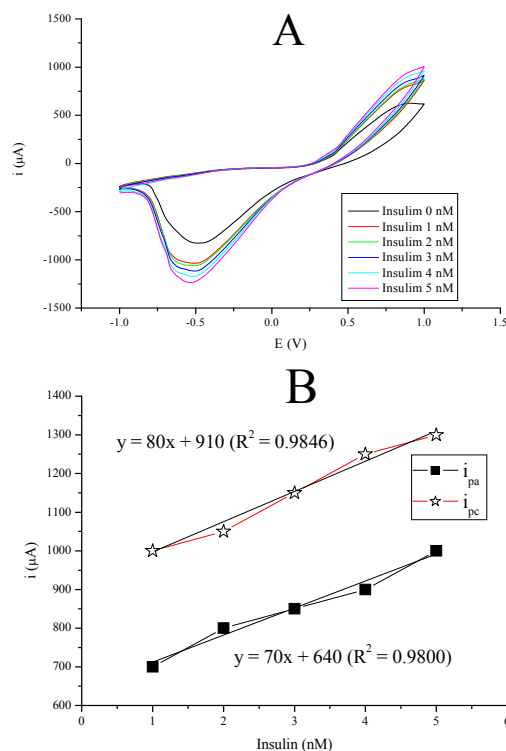
Figure-5. (A) Cyclic voltammogram of insulin different concentration in phosphate buffer pH 7.4 solution with scan rate 100 mV/s. (B) The calibration curve of insulin.

CONCLUSIONS

The modification of silica gel and chitosan electrode by 2 mL of $Ni(OH)_2$ nanoparticles could increase the sensitivity of the electrode to insulin as much as sixfold. The anodic (i_{pa}) and cathodic (i_{pc}) current obtained were linear at varied squareroot of scan rate. Linear responses of anodic (i_{pa}) and cathodic (i_{pc}) current were observed at measurement of insulin from 1 mM to 5 mM. The cathodic (i_{pc}) current response was higher than the anodic (i_{pa}) current, but no significant differences observed in their sensitivity.

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