Synthesis and Characterization of Metal-Organic Framework (MOF) CoBTC as Non-Enzymatic Electrochemical Biosensor for Glucose

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Abstract. Diabetes mellitus is one of the highest causes of death in the world caused by high levels of glucose in the blood. Thus, monitoring glucose levels in the blood becomes very important. Non-enzymatic glucose detection is currently an emerging research area, one of which is to use Metal-Organic Material (MOF), which has advantages such as having a large surface area and having a large amount of micropore. The purpose of this study was to synthesize MOF CoBTC and see its performance as a glucose sensor. The synthesis method used is the solvothermal method, while the sensor performance testing method uses an electrochemical method. The solvothermal method of the CoBTC was carried out at a temperature of 100°C and duration of synthesis of 10 hours CoBTC materials. The structural characterization of the synthesized samples by X-Ray Diffraction (XRD), Scanning Elektron Microscopy (SEM), Fourier Transform Infrared (FTIR) Spectroscopy, Brunauer Emmett Teller (BET) Surface Area Analyzer showed that we successfully synthesized CoBTC. Furthermore, the electrochemical characterization via cyclic voltammetry (CV) and amperometry results showed the optimal working potential of the sensor at 0.4 V, the sensitivity of 53.455 μA mM⁻¹ cm⁻² and the limit of detection (LOD) at 5.49 μM.

1. Introduction

One of the diseases suffered by humans today is diabetes mellitus. Diabetes mellitus is a condition in which the glucose content in the blood exceeds the average threshold [1]. Diabetes mellitus is a metabolic disease caused by the inability of the pancreas to produce enough insulin or the inability of the human body to use the insulin effectively, resulting in an uncontrollable increase in glucose levels in the blood [2]. The International Diabetes Federation (IDF) estimates that in 2013 there were over 380 million people with diabetes, and it is estimated that this number will increase to 592 million people in 2035, wherein 2013, 175 million of them have not been diagnosed, so they are threatened with progressive development into complications without realizing it [3]. Furthermore, without prevention, Diabetes mortality alone reaches 80% in countries with lower-middle-income levels. Thus, the detection of diabetes mellitus is very much needed to be able to prevent and avoid developing diabetes. Until recently, diabetes detection was carried out using enzymatic electrochemical glucose biosensors, where...
85% of the glucose sensors currently on the market are used for diabetes detection, and the remainder is used for the detection of glucose content in medicines, foods, and others [4].

Since fast and reliable glucose detection has become essential in many fields such as biotechnology, clinical diagnosis, and the food industry, the development of electrochemical based glucose sensors has attracted much attention. In the past, research on enzymatic glucose biosensors based on the oxidation reaction of glucose compounds was prevalent because of its sensitivity [5]. However, testing using modified enzyme electrodes has disadvantages such as relatively low stability due to environmental conditions, high cost of the enzymes, critical operational conditions, and complicated immobilization procedures. Therefore, glucose detection without the use of enzymes or non-enzymatic detection is highly desirable with the advantages of stability, simple, sustainable production, and low cost [6]. Currently, the development of non-enzymatic electrodes has achieved the modification of precious metal compounds, metal alloys, and metal nanoparticles. However, suitable detection materials are required for glucose detection.

One of the materials that can be used as active material in biosensors is material based on the metal-organic framework. Metal-organic frameworks (MOF) or coordination polymers are metal-ligand compounds whose structures will expand or extend in one, two, or three dimensions and where the ligands will become connecting organic groups [7]. MOF compounds that have microporous properties are interesting material to develop. The space between metals connected to organic ligands makes MOF material have a large number of micropores and a large surface area. This large surface area causes MOF to have the potential to be developed into a biosensor material. Therefore, in this study, the development of a non-enzyme sensor based on Metal-Organic Framework (MOF) was chosen, namely the Co material with BTC. Co is used because it has strong magnetic properties, does not readily react with oxygen in the air and does not dissolve easily in water, and is very popular as a glucose biosensor in its oxide formation. MOF with a Co metal base will be of particular interest due to the Co metal and the MOF material itself so that the material is considered to have excellent performance as an active material for glucose detection biosensors. Therefore, further study is needed on the performance of CoBTC and how the synthesis formulation of this material is to produce active glucose detection biosensor material with optimum performance so that it can be used as a glucose detection biosensor.

2. Experimental

2.1. Materials

Cobalt(II) nitrate hexahydrate (Co(NO$_3$)$_2$.6H$_2$O), trimesic Acid (H$_3$BTC), dimethylformamide (DMF), glucose, Nafion, and other common reagents were acquired from Sigma-Aldrich, Singapore. The organic solutions such as ethanol were purchased from Merck, Singapore. All chemical materials were of analytical grade and used without any further purification.

2.2. Synthesis of CoBTC

The synthesis of CoBTC was carried out by mixing metal precursors and linkers at heating temperature or what can be called the solvothermal method. First, we dissolved the cobalt dinitrate hexahydrate (1.454 grams) in 10 mL distilled water and trimesic acid precursor (0.525 grams) in 10 mL DMF and 10 mL ethanol, then stirred separately for ± 20 minutes. Next, the two solutions were mixed by dropping the trimesic acid solution drop by drop into the cobalt metal solution and stirred for ± 45 minutes. After the mixture is homogeneous, the mixture was heated thermally in the autoclave at 100 °C for 10 hours. The mixture will produce a product in the form of powder. The as-collected powder then washed several times with ethanol while sonicated. Then the product was dried at a temperature of 100 °C for 10 hours. Finally, the sample is ready to be used and characterized. The synthesis of CoBTC will follow the following formula:

$$3\text{Co(NO}_3\text{)}_2.6\text{H}_2\text{O} + 2\text{H}_3\text{BTC} \rightarrow \text{Co}_3(\text{BTC})_2.12\text{H}_2\text{O}+ 6\text{NO}_3^- + 6\text{H}_2\text{O} + \text{H}^+$$

(1)
2.3. Material Characterization
Bruker D8 Advanced collected the X-Ray Diffraction (XRD) patterns with CuKα x-ray generator, \( \lambda = 1.5418 \text{ Å} \). The morphologies property of the CoBTC were analyzed using Hitachi Scanning Electron Microscopy (SEM) SU3500 series. The surface area of the CoBTC was analyzed using a Surface Area Analyzer with the Brunauer, Emmet, and Teller (BET) analysis method and Quantachrome Instrument Nova 1000, while Fourier Transform Infrared (FTIR) spectrum was collected by Prestige 21 Shimadzu. We employed CorrTest CS Series Electrochemical Station to carry out electrochemical measurements. All electrochemical measurements were carried out in a standard three-electrode system in an aerated condition at room temperature. In this work, we employed a glassy carbon electrode (GCE, 3 mm in diameter) modified with the CoBTC samples as the working electrode. Meanwhile, we used an Ag/AgCl standard electrode (3 M KCl) as the reference electrode and a Pt wire as the counter electrode. NaOH solution 0.1M was employed as the supporting electrolyte for the electrochemical characterization. The Cyclic Voltammograms (CVs) test of the samples was carried out in the potential range of -0.2-0.8 V with a scan rate of 50 mV/s. Meanwhile, the amperometry test was carried out at 0.4 V. All potential are versus the Ag/AgCl standard reference electrode.

3. Results and Discussion
3.1. Structural Characterization
The graph of the synthesized CoBTC diffraction peaks compared to the reference can be seen in Figure 1 (a). Graph of FTIR spectroscopy results are shown in Figure 1 (b) shows the classification of the analysis from the top of the graph to the range of the infrared spectrum absorbed from the covalent bond. Based on Figure 1(a), the synthesized CoBTC has peaks that are the same as the CoBTC synthesized by Crane et al., 2014 [8], and Aguiar et al. 2017 [9], namely at 17.3, 18.4, and 26.7 2θ (deg), which indicates the material is Co₃(BTC)₂.12H₂O material. In Figure 1 (b), it can be seen that all forms of chemical bonds that make up CoBTC compounds based on its chemical structure can be detected, which also confirms that the CoBTC material is perfectly formed.

![Figure 1](image1.png)

**Figure 1** (a) XRD patterns of the CoBTC in this work and its reference (b) FTIR spectrum of the CoBTC

SEM on the synthesized CoBTC material was carried out at two different magnifications, namely 2,000 times and 15,000 times. The result of magnification of 2,000 times can be seen in Figure 2 (a), while for magnification of 15,000 times can be seen in Figure 2 (b). Based on SEM results at a magnification of 2,000 times, it is known that the morphological formation of the CoBTC material synthesized tends to clump. At a magnification of 15,000 times, it appears that CoBTC has a cubical shape, and it appears that CoBTC has a length between 25-30 μm and a width of 21-24 μm. Based on
the references found, CoBTC has various forms of morphology, depending on the synthesis method used and the variations in the method concerned. The morphological forms of CoBTC include a distorted octahedral form [10] and rod [9]. The synthesized CoBTC has a cubical shape that has six surfaces with uneven conditions, and the resulting CoBTC tends to be agglomerated, and the size is not homogeneous.

The large surface area is one of the advantages of MOF materials. MOF is a porous material that has so far been used and holds great promise for storing hydrogen and methane, which has already been applied to fill car tanks. As a sensor, a large surface area is an advantage of MOF material, because the more significant the surface area, the higher the active area of interaction between MOF as a sensor with glucose molecules. CoBTC itself, which has been characterized by a BET Surface Area Analyzer, has a surface area of 557.862 m²/g. However, these results are much better than the synthesis results from Aguiar et al. 2017 [9], which is only 57 m²/g with the same XRD characterization results and higher than Co in its oxidation form, Co₃O₄ which is only 260 m²/g [11].

### 3.2. Electrochemical Characterization

The process of cyclic voltammetry testing is carried out in alkaline conditions where a 0.1 M NaOH solution is used as the analysis solvent. The use of an alkaline environment is because glucose is not reactive in an acidic environment and the instability of the transition metal/metal oxide-based electrode materials in acidic conditions. This instability can cause the oxidation process not to run optimally, and the resulting voltammetry peak will be small. Glucose in a neutral environment tends to form a double layer region where the oxidation peak will be reformed so that the peak from the maximum oxidation period of glucose is challenging to know. The ideal alkaline environment becomes a detection environment because glucose will be easily oxidized and become gluconic acid, which is indicated by the peak value of oxidation from the cyclic voltammetry test.

This voltammetry test is carried out in a voltage range from -0.2 to 0.8V with a positive test direction and a scan rate of 50 mV / s. The oxidation equation that occurs is as follows:

- CoBTC + 2OH⁻ → [CoBTC(OH)₂]⁻ (2)
- [CoBTC(OH)₂]⁻ + e⁻ → Co(OH)₂ + BTC (3)
- Co(OH)₂ + OH⁻ → CoOOH + e⁻ + H₂O (4)
- CoOOH + e⁻ + glucose → CoO + OH⁻ + gluconic acid (5)
- CoO + OH⁻ → CoOOH + e⁻ (6)

Equations (2), (3), and (4) are the material activation process. In this process, the material is tested in 0.1 M NaOH solution without glucose so that the material is active. Equations (3) and (4) also prove that the sensor material will produce current even in the absence of glucose. After the material is activated, it is tested with glucose. During the testing process with glucose, reactions such as equations...
(2), (3), and (4) are added with the reactions according to equations (5) and (6), which will cause an increase in the peak flow when without glucose and when tested at the glucose concentration higher. This process occurs when the oxidation of Co becomes a catalyst for the oxidation of glucose to gluconic acid. The cyclic voltammetry testing process can be seen in Figure 3. It can be seen that CoMOF can increase glucose oxidation current compared to bare GCE. The oxidation of glucose by CoMOF occurs at a potential of 0.4 V.

Furthermore, amperometric testing was carried out to determine the sensitivity and limit of detection (LOD) of the selected CoBTC material to glucose. In this study, a concentration variation from 0 to 5 mM of glucose was used, and potential value of 0.4 V was used where this potential value was obtained from cyclic voltammetry experiments because, at a potential of 0.4 V, the material was able to oxidize glucose completely. The graph of the results of this amperometric test can be seen in Figure 4.

From Figure 4, it can be seen that the increase in the value of glucose concentration causes an increase in the resulting current value, where the difference in the increase in flow is directly proportional to the difference in the increase in glucose. A graph of the relationship between concentration and the resulting current is drawn, as shown in figure 4(b), to find out quantitatively about this comparison. From the graph, it can be seen the linearity of the material in detecting glucose from a range of 0 to 5 mM with a
value of $R^2 = 0.9859$. The sensitivity value can also be seen from the slope value of the line on the graph of the ratio of concentration to current. The sensitivity value obtained was 53.455 μA mM$^{-1}$ cm$^{-2}$ and the LOD was 5.49 μM. The sensitivity of the CoBTC biosensor of 53.455 μA mM$^{-1}$ cm$^{-2}$ was still lower than the non-enzymatic biosensor based on Co$_3$O$_4$, which was 1377 μA mM$^{-1}$ cm$^{-2}$ by Wang et al., 2017 [12] so that the modification of this material was to exceed the sensitivity of Co$_3$O$_4$ will need to be done in the future.

4. Conclusions
In this work, we successfully synthesized CoBTC material using the solvothermal method. The CoBTC was characterized using XRD, SEM, FTIR, and BET and showed similar features to those previously synthesized. The CoBTC was then used as a sensing material for non-enzymatic electrochemical detection of glucose. The results showed that the optimal working potential of the sensor at 0.4 V, the sensitivity of 53.455 μA mM$^{-1}$ cm$^{-2}$ and the limit of detection (LOD) at 5.49 μM. This result indicates that CoBTC is promising to be further studied as a non-enzymatic electrochemical sensor material.

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