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A Method in Urea Sensor Fabrication Based Immobilization Diacetylmonoxime-Thiosemicarbazide onto Silica Paper

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ABSTRACT

Background: The color changing is one of variables that can be used as an indicator of analysis by urea sensor. It can be conversed in Δmean RGB value by adobe photoshop programme. **Objectives:** This paper suggests an optimum way to fabricate urea sensor using immobilization of diacetylmonoxime and thiosemicarbazide reagents onto silica papers. The variables that tested were the immoblization techniques, the concentration of reagent diacetylmonoxime (DAM) and the concentration of reagent thiosemicarbazide (TSC). **Results:** immobilization of reagent onto silica paper was done by spraying, coating and spoting technique. The high Δmean RGB value was given by spoting techniques. The DAM variant concentration (40;100;160 mmol/L) produced 55,22, 64,89 and 69,05 for their Δmean RGB value. The TSC variant concentration (4;8;16 mmol/L) produced 57,17, 64,94 and 62,88 for their Δmean RGB value. **Conclusion:** The best immobilization tecgnique in urea sensor fabrication was spotting; the best concentrations of DAM/TSC in urea sensor fabrication were 160 mmol/L and 8 mmol/L.

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INTRODUCTION

Urea is a final result substance from metabolism which is out from body in urine fabrication through kidney. Source of urea was from final metabolism result like NH_3 which can be toxic substance if it was not excreted [2].

The determination of Urea in body liquids is one of the analyses that was often used in clinical laboratory [1]. Urea filtration by kidney in glomerulus is being excreted in urine form and a few of them is being reabsorp in tubulus to blood, So it can be one of normal component in blood. Urea in blood or usually is called *blood nitrogen urea* has normal concentration between 5-25 mg/dL [3].

Beside for being kidney disease diagnosed, the determination of urea in blood can be used to evaluate the development of chronic kidney failure sufferers in hemodialisis therapy. Hemodialisis is a therapy method to omit metabolism remnants that is stacking in blood, one of urea with diffusion principal through semipermiable membrane .

The most being used method of urea determination of blood in laboratory is urea determination method in colometri with reagent diacetylmonoxime and *enzyme assays*. Colorimetri for urea determination was first being found by Fearon in the year 1939 [4]. It based on condensation reaction between urea and diacetylmonoxime that produce a colorful substance. Then, urea is measured by spectrofotometer around 540 nm [3].

In this research, the method of urea determination using reagent diasetyl monoxime was designed in chemist sensor form. It was made in the form of paper contain diacetylmonoxime (DAM) and thiosemicarbazide (TSC). The aim of this research was to know the technique of immobilization in best silica papers and best concentration DAM / TSC in sensor urea making.

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Methodology:

This research was experimental laboratory research. The materials were urea, *diacetylmonoxime*, *thiosemicarbazide*, NaCl, sulfuric acid (H_2SO_4), phosphate acid (H_3PO_4), acetic acid (CH_3COOH), chloride acid (HCl), $FeCl_3$, aquadestilata, and silica paper.

The Immobilization Technique of Reagent Diacetylmonoxime-Thiocarbazide onto Silica Paper:

Reagent for urea identification such as reagent Diacetyl monoxime-Tiosemikarbazida and reagent acid was prepared 30 mL (1:1), also silica paper 1×1cm. Reagent for urea identification was immobilized onto silica paper 1×1 cm with adsorption technique. They were spotted, sprayed and coated. Silica papers were dried in 35^0C for 10 minutes. Silica papers were used to detect urea 0,8 mmol/L, and then we see the changing of color on to silica papers. Procedures above were repeated for each immobilization techniques.

The determination of best DAM/TSC concentration in urea sensor fabrication:

According to procedure 3.1, we prepared sensor urea in variation concentration of DAM and TSC. The variation concentration of DAM were 40 mmol/L, 100 mmol/ L and 160 mmol/ L. whereas, the concentration variation of TSC were 4 mmol/L, 8 mmol/L and 16 mmol/L. the variable that watched were time response and color changing.

RESULTS AND DISCUSSION

Immobilization of diacetilmonoxime and tiosemikarbazida in silica papers produced sensor paper white color. After it was applicated in urea sample, the paper would give pink color. The color changing can be sized in Δmean RGB value using adobe photoshop.

The result of sensor urea before urea detection

The result of sensor urea after urea detection

Fig. 1: Color comparation of urea sensor before and after urea detection

There were three ways to do immobilization. They were spraying, coating and spotting. Then, test the ability to response. The result shows that the fastest response was marked by color forming. An analysis of color changing intensity in silica papers using Δmean RGB. It was from difference between RGB blanko and RGB each immobilization technique. The big the value of Δmean RGB the strong color resulted. The best result of immobilixation technique was shown in table 1. The best technique was spotting. In spotting technique, deployment of reagent which immobilized can be prevalent in same volume per spot. This supported reaction can be maximum and the color was more prevalent despite of the others technique. So, the value Δmean RGB could be much bigger.

Table 1: Result of immobilization technique comparation

No	Techniques	Time response	Δmean RGB
1	spraying	3 minutes	65,78
2	coating	2 minutes	70,44
3	spotting	2 minutes	71,99

Concentration reagent DAM and TSC that were immobilized in silica papers was also needed to be optimized. This was related to how much is the maximum limitation of concentration reagent that can be absorbed in silica papers through sensor making process.

The variant of concentration reagent DAM that was used in sensor making were 40 mmol/L; 100 mmol/L and 160 mmol/L. Sensor urea with variant concentration used to detect urea and see the time response and also the color intensity in Δmean RGB.

From the test, Δmean RGB concentration 160 mmol/L is the biggest in 2 minutes. The result of sensor urea was shown in table 2.

Table 2: Δmean RGB Value based on DAM variant concentration

No	Concentration DAM (mmol/L)	Time response	*ΔMean* RGB
1	40	4 minutes	55,22
2	100	2 minutes	64,89
3	160	2 minutes	69,05

The variant of concentration reagent TSC that was used in sensor making were 4 mmol/L; 8 mmol/L and 16 mmol/L. Sensor urea with variant concentration used to detect urea and see the time response and also the color intensity in Δmean RGB.

From the test, Δmean RGB concentration 8 mmol/L is the biggest in 2 minutes. The result of sensor urea was shown in table 3.

Table 3: Δmean RGB Value based on TSG variant concentration

No	concentrationi TSC (mmol/L)	Time response	*ΔMean* RGB
1	4	2 minutes	57,17
2	8	2 minutes	64,94
3	16	2 minutes	62,88

Conclusion:

The best immobilization technique in urea sensor fabrication was spotting; The best concentrations of DAM/TSC in urea sensor fabrication were 160 mmol/L and 8 mmol/L.

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