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***Research paper***

# Development of Urea Sensor Based on colorimetric method and Digital RGB Analysis

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**Abstract**

A simple and low cost of urea sensor has been developed using diacetyl monoxime (DAM), Thiosemicarbazide (TSC), Sulfuric Acid and Phosphoric Acid to form pink-azo dye compoud. A urea sensor was manufactured by immobilizing reagents to the silica gel plate. The sensor was tested to determine urea in various concentrations including 0; 5; 6; 25; 26 and 80 mg/dL. The color formed on the sensor was captured by Samsung camera 12 megapixel in the black box to obtain color digital data such as red, green, blue (RGB). The Δ Mean RGB valuess were 0.0; 51.6; 51.9; 58.5; 61.4 and 75.3 respectively.

***Keywords***: *Colorimetric, kidney, RGB value, sensor, urea*

## 1. Introduction

Kidneys are the one of the human organ filtering impurities, especially urea, from blood and throw it as urine. Urea is the residual of nitrogen metabolism in the human body (Lugosi, 1972). Urea can be harmful in the certain concentration and should be released from the body in urine form (Martoharsono, 2006). The urea concentration in the blood can be used as an indicator for any kind of kidney disease (Akcay, et al., 2010). Therefore, it can be useful for monitoring the patients of chronic kidney disease (Amin et al., 2014).

The colorimetric method for determination of urea had been investigated by Fearon using diacetyl reagent in strong acidic conditions and produced a dyes compounds (Shanmugam et al., 2010). Fearon method is the basic of any method development to determine the urea concentration in biological samples such as urine and serum (Wybenga, et al., 1971). This method is less sensitive, must be operated in clinical laboratories, should be handled by experts and need much time.

The measurement of urea concentration has been developed to the sensor based on colorimetric. The sensor has an advantage in response time and ease of use. Fahmi (2015) made a urea sensor and measured its performance as a sensor. The DAM, TSC and acid reagents were immobilized in the silica gel plate. The limit of detection (LOD) and the limit of quantitation (LOQ) of urea are 0.1232 mmol / L and 0.4105 mmol / L.

Badi'ah (2015) had examined the influence of acid reagents which are DAM and TSC. As a result, the combination of reagents sulfuric acid and phosphoric acid gave 3-minutes response time for the measurement of urea. Optimization of the concentration of each reagents and techniques for sensor manufacturing process has been studied by Fauziyah, et all. 2015. The best result in immobilization technique of urea sensor fabrication was spotting. The optimum concentration of DAM and TSC were 160 mmol / L and 8 mmol / L. But there is no study of RGB value analysis of urea sensor to convert the color to the digital image data as a database to develop an android application to determine the concentration of urea base on urea sensor and the Δ Mean RGB values.

In this study, urea sensors was designed and tested by colorimetric analytical performance to determine urea in various concentrations. The concentrations of urea represent the condition of the human kidney including under normal (0 - 5 mg / dL), normal (6 mg / dL - 25 mg / dL) and abnormal (26 mg / dL - 80 mg / dL). Urea sensor used to measure each categories of urea was converted to RGB data. The RGB data obtained from a camera in a black box to adjust the lighting when taking data.

## 2. Materials and Methods

The materials were urea, DAM, TSC, $H_2SO_4$, $H_3PO_4$, $FeCl_3$, silica gel plate and distilled water.

Silica gel plate (2 x 2 cm) was immobilized by DAM solution 160 mmol/L, TSC 8 mmol/L, sulfuric acid and phosphoric acid (1: 1) with a volume ratio of 1: 1: 2. Thus the immobilized silica gel plate was dried at room temperature.

The immobilized silica gel plate was used to measure the urea concentrations 0; 5; 6; 25; 26; 80 mg/dL respectively. The color change of immobilized silica gel plate was captured by 12 MP camera in a black box with same distance and constant lighting. Digital image data stored in the RGB values and processed as Mean RGB values.

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## 3. Results and Discussion

Urea in the blood also named as a blood urea nitrogen (BUN) have normal levels of 5 to 25 mg / dL (Shanmugam et al., 2010). Urea must be excreted through the kidneys but a small portion of urea (5-25 mg / dL) are reabsorbed and become a component of the blood. The concentration of urea is less than or exceeds the normal concentration indicate health problems associated with the kidneys.
Sensor urea reagent containing DAM, TSC, sulfuric acid and phosphoric acid with a certain ratio. When the sensor is used to detect samples containing urea, the reagents will react with urea to form triazine as in Figure 1:
The intensity of pink-azo dye compound (3-hidroxi-5,6-dimetyl-1,2,4-triazin) was affected by the concentration of urea. The concentration of urea used were 0; 5; 6; 25; 26 and 80 mg / dL. The color formed on the sensor was captured with a Samsung camera 12 MP to gain the digital image data. The concentration of urea 0 and 5 mg / dL representing the below normal of urea concentration in the blood, 6 and 25 mg / dL for normal concentration, 26 and 80 mg/dL for abnormal urea concentration in the blood.
The digital image was obtained from capturing the sensor in a closed black box. On the top of the box there was a hole for the camera lens. And the side of the box there was a hole for the light source. Thus the image was taken consistently within a light and manner.
The Digital color data of urea sensor is shown in table 1. The value of R, G and B decreases generally along with increasing of urea concentration. It shows that the different concentrations affect to the value of R, G and B. The white color has the highest RGB values while the black color has the lowest the RGB values.

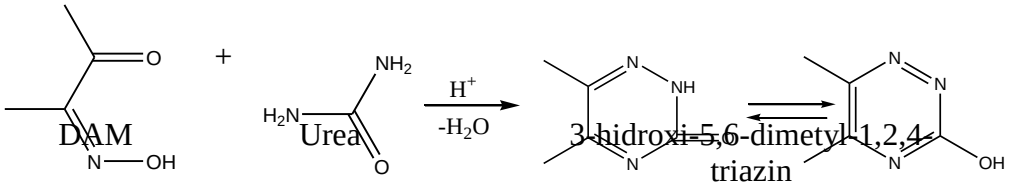



**Fig.1:** Reaction of DAM and urea to form 3-hidroxi-5,6-dimetyl-1,2,4-triazin

The intensity of pink-azo dye compound (3-hidroxi-5,6-dimetyl-1,2,4-triazin) was affected by the concentration of urea. The concentration of urea used were 0; 5; 6; 25; 26 and 80 mg / dL. The color formed on the sensor was captured with a Samsung camera 12 MP to gain the digital image data. The concentration of urea 0 and 5 mg / dL representing the below normal of urea concentration in the blood, 6 and 25 mg / dL for normal concentration, 26 and 80 mg/dL for abnormal urea concentration in the blood.
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**Table 1:** Digital color data of urea sensor

| Urea Concentration (mg/dL) | R value (n=5) | G value (n=5) | B value (n=5) |
|---|---|---|---|
| **0** | 254 | 254.4 | 252.4 |
| **5** | 220.2 | 204.8 | 181 |
| **6** | 220.8 | 201.2 | 183.2 |
| **25** | 213.8 | 195.8 | 175.6 |
| **26** | 208 | 192.8 | 175.8 |
| **80** | 196.8 | 179.6 | 158.4 |

The Δ Mean RGB was determined by calculating the differences of mean RGB of each concentration with the blank urea concentration (0 mg/dL) as shown in table 2.

**Table 2:** Δ Mean RGB of urea sensor

| Urea Concentration (mg/dL) | Mean RGB | Δ Mean RGB |
|---|---|---|
| **0** | 253.6 | 0.0 |
| **5** | 202.0 | 51.6 |
| **6** | 201.7 | 51.9 |
| **25** | 195.1 | 58.5 |
| **26** | 192.2 | 61.4 |
| **80** | 178.3 | 75.3 |

The Δ Mean RGB values of urea concentration 0; 5; 6; 25; 26 and 80 were 0.0; 51.6; 51.9; 58.5; 61.4 and 75.3 respectively. The Δ Mean RGB values can be used as a database to develop an android application to convert color on the sensor plate to the application system to determine the concentration of urea base on the Δ Mean RGB valuess.

## 4. Conclusions

A simple and rapid sensor had been successfully developed to measure the various concentration of urea and analyzed to the Δ Mean RGB valuess as a digital data. It showed that different concentration of urea affected to the value of R, G and B. The Δ Mean RGB valuess of 0; 5; 6; 25; 26 and 80 mg/dL urea concentration were 0.0; 51.6; 51.9; 58.5; 61.4 and 75.3 respectively.

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