# CHARACTERISTICS OF ELECTRODE MEMBRANE PVAENZIME COATING PVC-PLASTICIZER KTPCLPB FOR UREA BIOSENSOR

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# CHARACTERISTICS OF ELECTRODE MEMBRANE PVA-ENZIME COATING PVC-PLASTICIZER KTPCLPB FOR UREA BIOSENSOR

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**ABSTRACT**

*This study aims to determine the effect of the composition comparison of the PVA-Enzyme urease coating PVC-Plastisizer KTpClPB membrane with memmbran characterization using FTIR and analyze membrane electrodes with XRD. The method used is the potentiometric method using the Electrode Selective Ion (ISE), where the polymer used is PVA and PVC as a coating with a composition ratio of PVA: PVC is 1: 1. The same PVA and PVC were used, namely 0.0350 g, with 2 mg of enzyme and 0.0500 g and 0.0120 g of KTpClPB plasticizer. The best composition result is the PVA-Enzyme PVC-Plastisizer coating enzyme KTpClPB 0.0500 g membrane electrode characterization using XRD shows the highest intensity value is at an angle of 2θ = 44.020 which is 1,626 count second (cts) and membrane characterization using FTIR shows that there is a functional group OH, CH, C≡C, NH, CO and CH. The addition of PVA-Enzyme coating will reduce or even eliminate the amount of Wolframm and can reduce the amorphous properties of the membrane electrodes.*

***Keywords:*** ***Electrode Selective Ion (ISE), Potentiometric, PVA-Enzyme, PVC-Plastisizer KTpClPB***

## INTRODUCTION

Biosensors are analytical tools that convert biological reactions into measurable signals such as electrical signals that allow it to analyze concentrations. The working principle of the biosensor is based on the immobilization of biological components (enzymes, bacteria, etc.) on the polymer membrane matrix which is integrated with the signal transducer on the analyte. The biological component functions as an electroactive sensor that plays a role in electrochemical half-cell reactions so that the potential generated is sensitive and selective to certain ions (Corcuera & Cavalieri, 2003).

The device developed based on the biological reaction (biocatalyst) "Urease" for analyzing urea is known as the urea biosensor. A urea biosensor is a device / biosensor that is made based on a signal from the urease “biocatalyst” reaction to urea to analyze urea levels in solution (Gupta, et al. 2010). The method of determining urea levels usually uses the spectrophotometric method, which is a method that uses the reaction between urea and diacetylmonoxime to produce a yellow color and the absorbance value is measured. The determination of urea by spectrophotometry is quite accurate, but it requires a relatively long time and a lot of chemicals. Therefore it is necessary to have a better method used in the determination of urea.

Another way to find out urea levels is by the potentiometric method. In the potentiometric method, the measurement of the equilibrium potential difference between the indicator electrode and the reference electrode will be measured

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(Tamba, 2016). The indicator electrode is an electrode that is able to determine the change in concentration for a particular ion that corresponds to the enzyme on the electrode. One type of indicator electrode is the membrane indicator electrode, which is commonly called the Ion Selective Electrode (ISE), which is a membrane that allows certain types of ions to pass through but repels other types of ions (Arora, 2013). The problem that often occurs in the potentiometric method is the manufacture of membrane components to become biosensor samples. The electrode membrane (ISE) is the place where the potential or current is generated from the enzyme-catalyzed reaction on the electrode membrane with the solution being analyzed, so the electrode membrane parameters are very important because it will determine the biosensor's performance (Goncalves, 2014).

The electrode membrane parameters are determined by the type of supporting material or matrix chosen (Fauziyah, 2012). Therefore, various studies have been carried out using various types of polymers as urease immobilization matrix for membrane electrodes. One of them is the use of PVA polymer. PVA (Polyvinyl alcohol) is a water-soluble polymer that has great promise for biomedical applications. PVA is now known as a synthetic polymer which has been used as an interesting research material for immobilization of the "biocatalyst" in the form of a membrane. It is non-toxic and biocompetible with good chemical and thermal stability. The large number of hydroxyl groups in PVA provides a biocompetible microenvironment for the presence of enzymes (eg urease enzymes) (Kale, et al, 2016).

However, PVA is water soluble, so a waterproof but well-pore coating membrane is needed as a coating membrane. Research conducted by Tamba (2016) used PVA as the urease immobilization matrix. PVA was used as much as 0.5044 grams dissolved in water and mixed with 6 mg of urease. The coating membrane used was a 0.5044 gram PVC solution with 0.0120 KTpClPB plasticizer and dissolved in THF. However, in his research, the correlation coefficient was 0.896. For this reason, changes will be made to the composition of the electrode membrane material, both in the composition of the PVA-enzyme membrane and the composition of the PVC-plasticizer membrane.

PVC is water-resistant and has good pores so it is suitable for use as a membrane. The use of plasticizers is needed to add elastic properties to PVC. Comparison of the composition of the PVC membrane and the plasticizer will affect the sensitivity and selectivity of the ISE membrane because the PVC membrane will coat the PVA membrane. The electrode membrane characterization was performed using X-ray diffraction (XRD) and the membrane characterization was performed using Fourier transform infrared spectroscopy (FTIR). FTIR is used to identify functional groups contained in a compound, and XRD is used to see the ratio of membrane intensity and characterize the resulting membrane (Hakim S, et al. 2018).

## METHODS

The equipment used is XRD-6100 (Shimazu), FTIR (Shimadzu), tungsten indicator electrode (W) and other supporting tools. The materials used in this study were 0.0350 g polyvinylalcohol (PVA), 2 mg of Urease enzyme (1%), 0.0350 g of polyvinylchloride (PVC), Pottasium tetrakis p-clorophenyl barate (KTpClPB)

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0.0120 g and 0, 0500 g, Wolframm electrode with a diameter of 1 mm and a length of 4 cm, and the method used is the potentiometric method.

Comparison of the composition of the membrane used (W PVA: W PVC-plasticizer = 1: 1) where the mass of PVA and PVC are the same and the ratio of PVC: Plastisizer = 1: 2 (Hakim S, et al, 2018). In this study, researchers used PVC: KTpClPB plasticizer with a ratio of 1:2. The preparation of the PVA-Enzyme solution comes from a mixture of the PVA solution and the enzyme solution. 0,0350 g PVA is dissolved in 10 mL of hot water to cold water, after which 2 mg of urease enzyme is dissolved in 0.5 mL consisting of water and alcohol with a ratio of 50% water: 50% alcohol. While the 0.0120 g KTpClPB PVC-Plastisizer solution comes from mixing 0.0350 g PVC and 0.0120 g KTpClPB dissolved into 10 mL THF in a beaker glass, and 0.0500 g KTpClPB PVC-Plastisizer solution from mixing 0.0350 g PVC and 0.0500 g KTpClPB which were dissolved in 10 mL THF in a beaker. So the PVA Enzyme solution and the PVC-Plastisizer KTpClPB solution are used to form membranes and membrane electrodes. The membrane was characterized using FTIR, while the membrane electrode was characterized by XRD.

## RESULT & DISCUSSION

### Characterization of Membrane Electrodes Using XRD

The membrane electrode is made using a tungsten electrode rod with a different PVA-Enzyme coating. Sample I, sample II, sample III, respectively, are one layer, two layer membrane and three layers of PVA-Enzyme coated with composition I (PVC-Plastisizer KTpClPB 0.0500 g). Sample IV, sample V and sample VI, respectively, are one layer, two layer membrane and three layers of PVA-Enzyme coated with composition II (PVC-Plastisizer KTpClPB 0.0120 g). The characterization of membrane electrodes using XRD was carried out to determine the spectrum of wavelengths and the highest intensity values as well as the effect of adding a PVA-Enzyme layer from each sample of membrane electrodes in each composition.

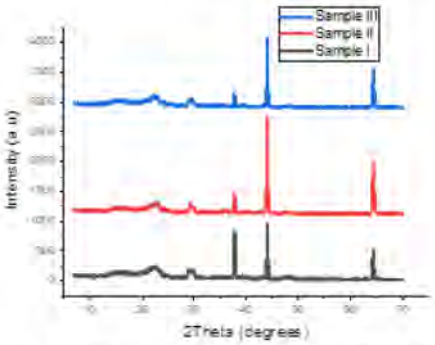


**Figure 3.1 Diffraction pattern Samples of one, two and three layers of PVA-Enzyme coating composition I (PVC-Plastisizer KTpClPB 0.0500 g)**

In the diffraction pattern of Figure 3.1, there are five dominant diffraction peaks in each sample. In sample I (1 PVA-Enzyme layer) has diffraction peaks at 2θ = 22.560; 28.940; 37.800; 44.020 and 64.340 with the highest intensity value at the angle of 2θ = 44.020, namely 1,262 count second (cts). Sample II (2 PVA-Enzyme layers) had

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diffraction peaks at 2θ = 22.660; 29.280; 37.780; 44.020 and 64.380 with the highest intensity value found at the angle of 2θ = 44.020, namely 1,626 count second (cts). And sample III (3 layers of PVA-Enzyme) has diffraction peaks at 2θ = 22.220; 29.560; 37.780; 44.020 and 64.380 with the highest intensity value at the angle of 2θ = 44.020, namely 1,174 count second (cts).

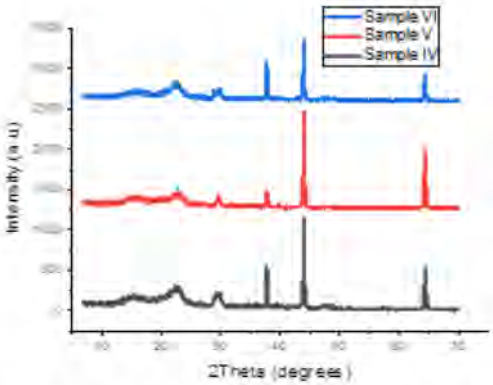


**Figure 3.2 Diffraction pattern Samples of one, two and three layers of PVA-Enzyme coating composition II (PVC-Plastisizer KTpClPB 0.0120 g)**

In the diffraction pattern of Figure 3.2, there are five dominant diffraction peaks in each sample. In sample IV (1 PVA-Enzyme layer) has diffraction peaks at 2θ = 22.400; 29.420; 37.800; 44.040 and 64.400 with the highest intensity value found at the angle of 2θ = 44.040 which is 1,104 count second (cts). Sample V (2 PVA-Enzyme layers) had diffraction peaks at 2θ = 22.600; 29.640; 37.780; 44.000 and 64.380 with the highest intensity values found at the angle of 2θ = 44,000, namely 1,208 count second (cts). And sample VI (3 layers of PVA-Enzyme) has diffraction peaks at 2θ = 22.520; 29.660; 37.760; 44.000 and 64.320 with the highest intensity value at the angle of 2θ = 44.020, namely 770 count second (cts).

In general, polymer materials are semicrystalline, which means they have an amorphous and a crystalline part (Rohaeti, 2009). The diffraction pattern has peaks that are not sharp indicating a structure where this diffraction pattern is generally owned by semicrystalline materials (Yulianti, et al, 2013). In Figure 3.1 (PVA-Enzyme coating composition I), the diffraction pattern of the three samples has sharp peaks indicating crystalline properties and has non-sharp peaks indicating amorphous properties so it can be said that the sample is a semicrystalline material. Figure 3.2 (PVA-Enzyme coating composition II) also shows crystalline and amorphous properties so that the three samples are also semicrystalline materials.

In the diffraction pattern of sample I in Figure 3.1, it can be seen that the amorphous pattern at the beginning is higher than sample II and sample III. And so also in the diffraction pattern of sample IV in Figure 3.2 which shows a higher initial amorphous pattern than sample V and sample VI. This shows that the addition of a PVA-Enzyme layer in each composition reduces the amorphous properties of the membrane electrodes.

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A good diffraction pattern can be seen from the intensity value obtained. The greater the intensity value obtained, the better the sample will be used (Hakim S, et al, 2019). The analysis above shows that the diffraction pattern that has the highest intensity value is sample II where the highest intensity value is 1,626 count second (cts) for the PVA-Enzyme coating membrane electrode composition I (PVC-Plastisizer KTpClPB 0.0500 g) and sample V where the intensity value The highest was 1,208 count second (cts) for the electrode of the PVA-Enzyme coating composition II (PVC-Plastisizer KTpClPB 0.0120 g).

**Table 3.1 Compounds and Elements on PVA Membrane Electrodes-Enzyme coating composition I (PVC-Plastisizer KTpClPB 0.0500 g)**

| Membrane Electrodes | Formula | Total (%) | Crystal System | Element |
|---|---|---|---|---|
| **Sample I (1 PVA-Enzyme layer)** | $C_6Cl_8N_{10}O_8$ | 44.6 | Monoclinic | C, Cl, N, O |
| | $B_{10}ClH_{13}$ | 36.1 | Monoclinic | B, Cl, H |
| | $KNO_3$ | 16.6 | Orthorhombic | K, N, O |
| | $H_2O_7W_2$ | 1.8 | Cubic | H, O, W |
| | $Cl_5P$ | 0.9 | Tetragonal | Cl, P |
| **Sample II (2 PVA-Enzyme layer)** | $C_6Cl_8N_{10}O_8$ | 40.4 | Monoclinic | C, Cl, N, O |
| | $Cl_{10}N_5P_5$ | 31.1 | Orthorhombic | Cl, N, P |
| | $B_{10}Cl_2H_8P_2$ | 14.4 | Orthorhombic | B, Cl, H, P |
| | $KNO_3$ | 13.6 | Orthorhombic | K, N, O |
| | $O_3W$ | 0.5 | Triclinic (anorthic) | Cl, K, O |
| **Sample III (3 PVA-Enzyme layer)** | $C_6Cl_8N_{10}O_8$ | 38.1 | Monoclinic | C, Cl, N, O |
| | $B_{10}ClH_{13}$ | 37.8 | Monoclinic | B, Cl, H |
| | $KNO_3$ | 22.8 | Orthorhombic | K, N, O |
| | $Cl_5P$ | 1.3 | Tetragonal | Cl, P |

The components that are expected to be seen on the PVA-Enzyme PVC-Plastisizer coating enzyme KTpClPB electrode are C, O, H, Cl, B, P, K, and W (Hakim S, et al, 2017). In table 3.1, samples I and II of the PVA-Enzyme coating coating electrode compoaiai I (PVC-Plastisizer KTpClPB 0.0500 g) have complete elements, namely elements C, O, H, Cl, B, P, N, K, and W Likewise with samples IV and V of the PVA-Enzyme coating coating electrode composition II (PVC-Plastisizer KTpClPB 0.0120 g) has complete elements, namely elements C, O, H, Cl, B, P, N, K, and W shown in table 3.2. This shows that the membrane electrode is formed from the PVA-Enzyme membrane, PVC-Plastisizer KTpClPB and tungsten rods.

Based on the characterization carried out for the membrane electrode with three layers of PVA-Enzyme (sample III and sample VI), no W element was seen. This indicated that the membrane was made too thick so that the tungsten rod was covered by the membrane. The addition of the PVA-Enzyme layer on the tungsten rod can affect the amount of W element present in the membrane electrodes.

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**Table 3.2 Compounds and Elements on the PVA Membrane Electrode-Enzyme coating composition II (PVC-Plastisizer KTpClPB 0.0120 g)**

| Membrane Electrodes | Formula | Total (%) | Crystal System | Element |
|---|---|---|---|---|
| Sample IV (1 PVA-Enzyme layer) | $C_{12}N_{18}O_8$ | 77.6 | orthorhombic | C, N, O |
| | $BH_9N_3P$ | 9.6 | Monoclinic | B, H, N, P |
| | P | 8.4 | Triclinic (anorthic) | P |
| | $N_4W_3$ | 2.5 | Cubic | N, W |
| | $ClKO_4$ | 1.9 | orthorhombic | Cl, K, O |
| Sample V (2 PVA-Enzyme layer) | $C_{12}N_{18}O_8$ | 80.6 | orthorhombic | C, N, O |
| | $K_4O_7P_2$ | 9.7 | Hexagonal | K, O, P |
| | $BH_9N_3P$ | 4.8 | Monoclinic | B, H, N, P |
| | $ClKO_4$ | 2.6 | orthorhombic | Cl, K, O |
| | $O_7P_2W$ | 2.2 | Cubic | O, P, W |
| Sample VI (3 PVA-Enzyme layer) | $C_{12}N_{18}O_8$ | 70.9 | orthorhombic | C, N, O |
| | $O_7P_4$ | 17.5 | Monoclinic | O, P |
| | $BHO_2$ | 11.1 | Cubic | B, H, O |
| | $ClKO_4$ | 0.5 | orthorhombic | Cl, K, O |

**Membrane Characterization Using FTIR**

Based on the results of the analysis using XRD, a good sample is sample II (2 layers of PVA-Enzyme PVC-Plastisizer coating 0.0500 g KTpClPB) for composition I and sample V (2 layers of PVA-Enzyme PVC-Plastisizer coating 0.0120 g KTpClPB) for composition II. The membrane that has been made based on a good sample of each composition will be characterized using FTIR to determine the functional groups contained in the membrane. Figure 3.3 below shows the FTIR spectrum graph of the PVA-Enzyme PVC-Plastisizer KTpClPB membrane (0.0500 g and 0.0120 g).

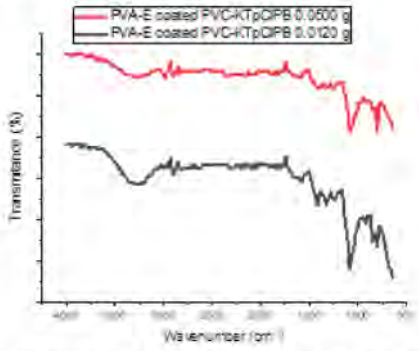


**Figure 3.3 FTIR spectrum on a 2 layer PVA membrane-enzyme coating PVC-Plastisizer KTpClPB (0.0500 g and 0.0120 g)**

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Based on Figure 3.3, on the PVA-E 2X membrane 0.0500 g KTpClPB PVC-P and 2X PVA-E membrane. PVC-P KTpClPB 0.0120 g contained the CH functional group seen in the wave numbers 2,850-2,970 $cm^{-1}$, 1,340-1,470 $cm^{-1}$, and 3,010-3,100 $cm^{-1}$ groups. The OH functional group is seen in the wave number group 3,200-3,650 $cm^{-1}$ and the NH functional group is seen in the 3200-2400 $cm^{-1}$ group. Groups of wave numbers at 1,000-1,250 $cm^{-1}$ and 1,500-1,750 $cm^{-1}$ indicate the presence of a CO functional group. It can be seen that there is a C≡C functional group in the wave number group 2,100-2,300 $cm^{-1}$, but not very prominent.

At 1,000-1,250 $cm^{-1}$ and 1,340-1,470 $cm^{-1}$ wavelengths, the CH and CO functional groups were seen on the PVA-E 2X membrane. 0.0500 g KTpClPB PVC-P stands out more than the PVA-E 2X membrane. PVC-P KTpClPB 0.0120 g. Then the OH functional group was also more prominent on the PVA-E 2X membrane. PVC-P KTpClPB 0.0500 g. This shows that the addition of the PVC-Plastisizer KTpCBlB composition will increase the number of transmittances (%) on several functional groups found on the PVA-Enzyme PVC-Plastisizer KTpClPB membrane.

## CONCLUSION

Based on the results of the analysis of membrane electrode characterization using XRD, it is known that the best intensity is found in the PVA-Enzyme coated with composition I (PVC-Plastisizer KTpClPB 0.0500 g) with the highest intensity value at the angle of 2θ = 44.020, namely 1,626 count second (cts). And based on the analysis of membrane characterization on composition I and II using FTIR, the functional groups of CH, OH, NH, CO, and C≡C were obtained. It was concluded that the addition of the PVA-Enzyme layer would reduce the amorphous properties of the membrane electrodes, and the addition of the PVC-Plastisizer KTpCBlB composition would increase the number of transmittances (%) in several functional groups found on the membrane.

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