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Research Article

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Selective melamine sensor based on nanoporous carbon paste/molecularly imprinted polymer

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ABSTRACT

Selective melamine sensors based on nanoporous carbon paste /molecularly imprinted polymer have been studied. The study begins with the synthesis of MIP monomer mixture of methacrylic acid (MAA), and ethylene glycol dimethacrylate (EGDMA) cross linker, benzoyl peroxide initiator, and melamine template. Membrane electrodes are fabricated by mixing nanoporous carbon and MIP with a certain ratio of inserted into the electrode surface. The optimization of sample measurements which includes melamine test of pH value of a solution pH and membrane composition. Further characterization of the electrodes was done by determining the Nernst factor, measurement range, selectivity, and lifetime. Validation method was done by determining the accuracy, precision, and the detection limit. Based on the data from FTIR, has been successfully synthesized MIP with BET analysis showing that MIP has a larger surface area , a larger pore volume, and a larger pore diameter than the MIP before extraction. The optimum conditions for the analysis of melamine using potentiometric sensor of nanoporous carbon paste/MIP electrode are the ratio of nanoporous carbon, MIP, and paraffin by 45:20:35 and the optimum pH value of 3-4. Results of melamine analysis using this sensor are the measurement range of 10^{-6} - 10^{-2} M, the detection limit is 9.51 x 10^{-7} M, the Nernst factor is 54.4 mV / decade, the accuracies of the concentration of 10^{-4} M and 10^{-3} M are respectively 106.1% and 104.3%, and this electrode is selective against melamine and is relatively undisturbed by Ca^{2+}, K^{+}, Mg^{2+}, and Na^{+} that are usually present in milk.

Keywords: selective melamine sensor, molecularly imprinted polymer, carbon nanoporous, potentiometric

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INTRODUCTION

Melamine contamination in milk is currently a problem in food safety since the milk cases in China in 2008 which resulted in thousands of kidney failure of babies and some of them died [1]. Based on these cases it is, necessary to develop analysis method of analysis of melamine which is sensitive, selective, accurate, fast, easy, and low cost.

Determination of melamine is mostly using liquid chromatographic techniques; [2] and tandem (combined) LC-MS-MS [3], [4]. However, this method takes a huge investment and high operating costs to operators as well as specialized skills.

Potentiometric analysis is a simple method that uses electrodes as sensors to identify the target analyte by measuring its potential [5]. Surface electrodes are sensors that must contain a component which reacts chemically and reversibly with the analyte [6]. This technique is inexpensive equipment, requires no special skills to operate, and easy to carry equipment that can be directly used in the field.

Currently, imprinted polymers have been developed that are useful for the development of sensors. Modified electrode with MIP sensor is selective and sensitive to analyte target [7]. *Molecularly imprinted polymer (MIP) is known as polymerization technique formed by reacting a functional monomer, crosslinker, and initiator surrounding the template molecule (analyte).* Then, the template is removed by several methods, such as the extraction process, thus forming a polymer that has been printed in accordance with the analyte molecules. This imprinted polymers specific to the analyte in sample [8]. In this technique, the template interacts with the functional monomer approach through Van der Waals bonding, electrostatic interactions, and bond hydrogen [9]. Molecularly imprinted polymer is a material that has a high sensitivity and selectivity, inert, insoluble in most organic solvents and water [10]. Melamine analysis using electrodes modified with MIP only reported by Liang et al [11]and Harsini et al [12].

In this research, is used MIP to modify the nanoporous carbon paste electrode, which would then be used as a sensor of melamine using silver wire as a conductor. MIP is used in the manufacture of monomer methacrylic acid (MAA), ethylene glycol dimethacrylate (EGDMA) as the crosslinker and benzoyl peroxide as an initiator. Election nanoporous carbon as electrodes because it is inert and has a high conductivity [13].

The Nanoporous carbon is obtained from the synthesis of wood waste processing performed by the Laboratory of Forest Products Research and Development Engineering of Forestry and Forest Products Processing, Bogor [14].

EXPERIMENTAL SECTION

Reagents, chemicals, and materials

The materials used in this study include melamine (Aldrich), methacrylic acid (Merck), chloroform (Merck), ethylene glycol dimethacrylate (EDMA)(Merck), benzoyl peroxide (BPO) (Merck), solid paraffin (Merck), methanol (Merck), glacial acetic acid (Merck), silver wire and ultra high pure water. Carbon nanoporous is obtained from Forest Products Research Center, Bogor, Indonesia.

Instrumentation

Instruments used in this study include a set of potentiometric CyberScan 510, the reference electrode Ag/AgCl, FT-IR Perkin-Elmer and glassware commonly used in analytical chemistry.

Synthesis of molecularly imprinted polymer

Molecularly imprinted polymer (MIP) was prepared by mixing 0.8 mmol of monomer methacrylic acid (MAA) in 100 mL of benzene, 2.4 mmol of the crosslinker ethylene glycol dimethacrylate (EGDMA), and 0.2 mmol melamine template. Then, it is added to 1 mmol of 1% benzoyl peroxide initiator. The mixture was heated on a hot plate at 60°C until a white precipitate formed. The precipitate that formed was dried in the open air. Further washing using acetic acid and methanol in the ratio of 2:8 and the latter with water at 70^{o}C [11]. Control polymer is made in the same way without the addition of melamine.

Preparation of nanoporous carbon paste/MIP electrode and electrochemical cells

Nanoporous carbon inserted into the watch glass is subsequently mixed with oil paraffin [15] and MIP with a certain ratio. The mixture was homogenized with a little warm and inserted into the electrode body. Electrochemical cell in this system is:

Ag, AgCl | KCl 3M || sample solution | nanoporous carbon paste /MIP| AgCl, Ag

RESULTS AND DISCUSSION

Synthesis and characterization of molecularly imprinted polymer

Melamine prints formation mechanism is shown in Figure 1. Based on Table 1, it is seen that both of the control polymer, MIP, and MIP before washing, most have a spectrum with the same vibration bands, which is CH stretching and bending of-CH_2-and-CH_3, C = O stretching, and stretching-COC -. The difference is the band at wave number 3525 cm^{-1} which shows the-OH stretching the control polymer, 3467 cm^{-1} and 3421 cm^{-1} which shows the asymmetric stretching and symmetric stretching-NH group of the melamine is still trapped on the unwashed hot water MIP. While MIP has bands at wave numbers 3506 cm^{-1} which shows the-OH stretching vibration, followed by loss of vibration-NH. This shows that melamine has been extracted from the MIP and melamine prints has formed in the MIP.

BET analysis (Table 2) shows that MIP has a surface area, pore volume, and pore diameter in a row is 48.738 m^2/g, 0.155 cc/g and 3.841 nm larger than that of MIP before extracted successively 38.766 m^2 / g , 0.144 cc / g and 3.384

nm. This suggests that the MIP was formed melamine printed. At MIP is not extractable, surface area, pore volume and pore diameter smaller.

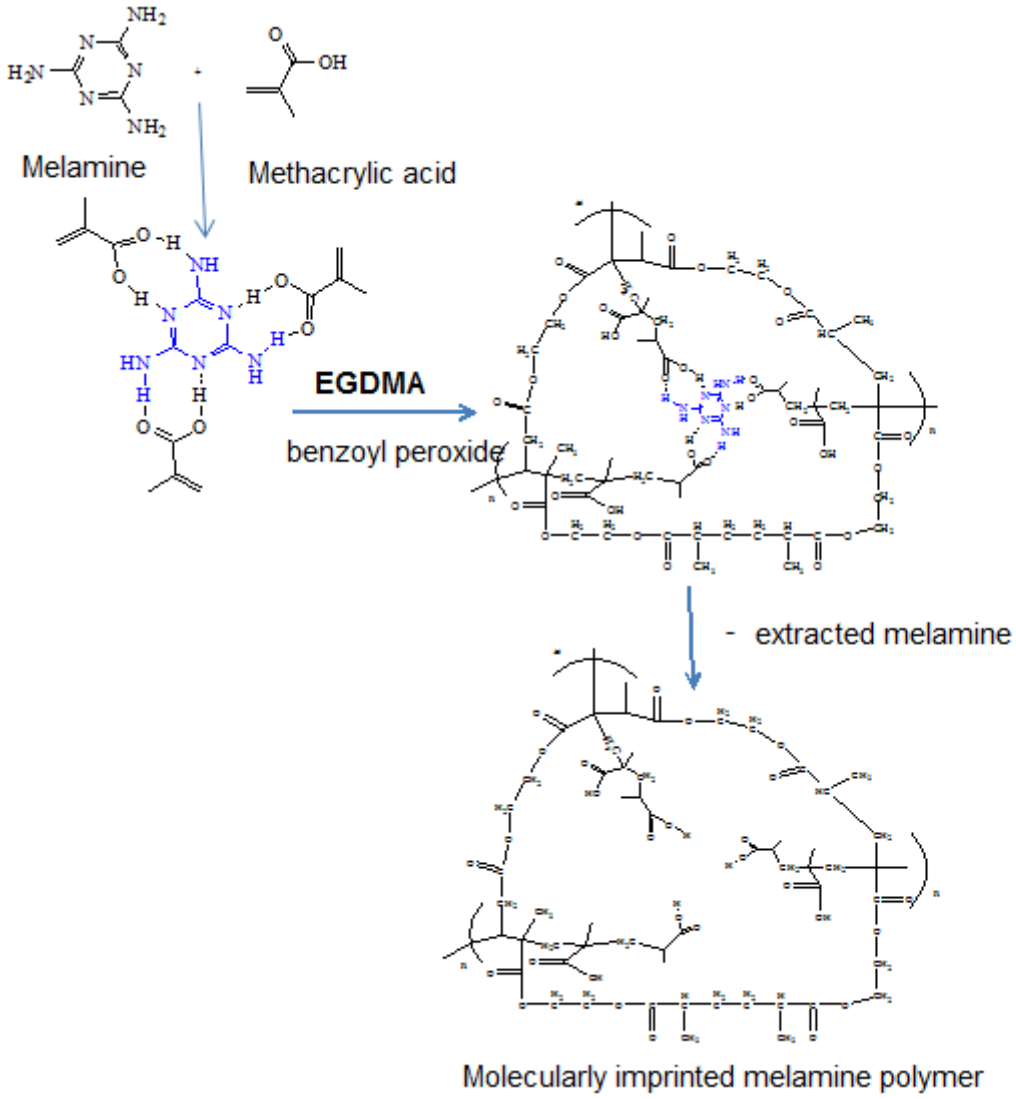

Figure 1 Formation stages of molecularly imprinted polymer of melamine [11]

Table 1. FTIR spectra data control polymer, MIP and MIP extracted before

Spectra	Wave number (cm^{-1})	Functional group vibration
Control polymer	3525	-OH stretch
	2989	C-H stretch of $-CH_3$
	1785,1760,1726	C=O stretch
	1224	C-O-C stretch
	1390-1370	-CH3 bends
	1470-1430	-CH2- bends
MIP before extracted	3467	-N-H primer asymmetric stretch
	3421	-N-H primer asymmetric stretch
	2989, 2954	-C-H- stretch of CH3/CH2
	1720	C-O stretch of C=O
	1257	C-O-O stretch
	1390	-CH3 bends
	1454	-CH2-bends
MIP	3506	-OH bends
	2989	CH stretch of -CH3
	2954	CH stretch of -CH2-
	1728, 1760, 1726	C=O stretch
	1456	$-CH_2-$ bends
	1224	C-O-C stretch

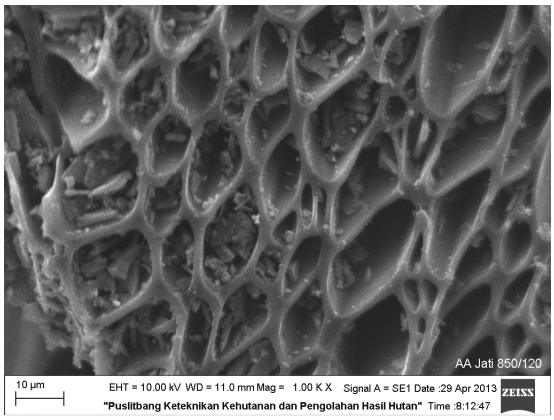

Figure 2. 1000x magnification scanning electron microscopy carbon nanopori of teak wood, which is activated by heating 850 ° C for 120 minutes

Characterization of Nanoporous Carbon

Nanoporous carbon is made of teak wood, which is activated at a temperature of 850° C for 120 minutes and have I_2 absorption of 1071 mg/g. Visual sightings nanoporous carbon using scanning electron microscopy (SEM) show that nanoporous carbon has a relatively homogeneous pore (Figure 2). Characterization of physical adsorption by the BET (Table 2) shows that this nanoporous carbon has a very wide surface area is 861.9 m^2/g, pore volume of 0.533 cc/g and a pore diameter of 123.800 nm. This suggests that the pore size is in units of nanometers. When compared with the MIP, the nanoporous carbon has a surface area, pore volume and pore diameter is much larger than the physical adsorption of MIP. Thus, MIP can be well adsorbed by the carbon.

Table 2 Data of surface area, pore volume and pore diameter MIP before extracted and MIP as well as nanoporous carbon using the BET BJH desorption data

Physical adsorption	MIP before extracted	MIP	Nanoporous carbon
Surface area (m^2/g), BET	34.604	54.486	861.900
Pore volume (cc/g) BJH desorption	0.144	0.155	0.533
Pore diameter (nm) BJH desorption	3.384	3.841	123.800

Optimization of electrode composition

Electrode composition optimization results can be seen in Table 3 which shows the Nernst factor, measurement range, and linearity at each electrode. The Curve who gives a linear relationship at each electrode is shown in Figure 3.

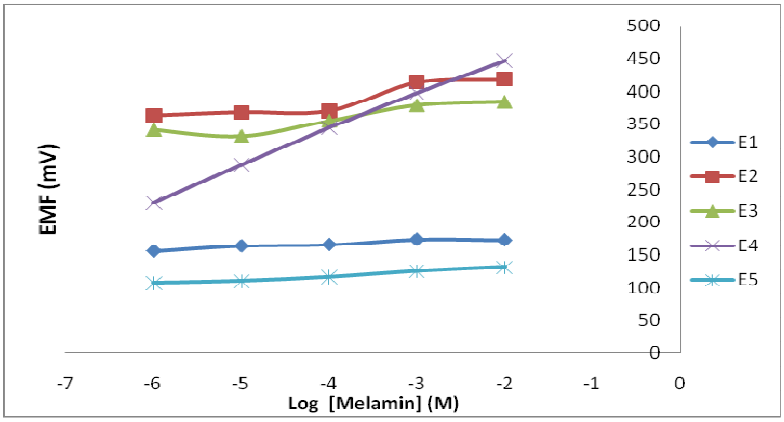

Figure 3 Relationship between log concentrations of melamine curve to the electrode potential at various compositions

Based on the data and the figure shows that the composition that gives the best results is the composition of the carbon electrode E4 nanoporous: MIP: paraffin is 45:20:35% w/w which gives the slope is closer to the Nernst factor is 54,40mV / decade. Nernst factor is expected to melamine is 59 mV / decade. In addition to the Nernst

factor, the working range and linearity are also very important. In this study, the electrode E4 gives a fairly broad range of work, ie 10^{-6} - 10^{-2} M and linearity 0.9994. While the composition of the electrode with the other giving Nernst factor smaller, shorter range and linearity measurements are less good. This is due to the absence or amount of MIP bit. However, if the amount is too large MIP (E5), will make the membranes become rigid and provide smaller response to the change in concentration of melamine.

Table 3 Composition of the carbon electrode nanopori / MIP, the Nernst factor, the measurement range and linearity

Electrode	Nanoporous carbon (%b)	MIP (%b)	Paraffin (%b)	Nernst Factor (mV/decade)	Measurement Range (M)	R^2
E1	65	0	35	8.9	10^{-8} — 10^{-5}	0.9930
E2	50	15(control polymer)	35	15.40	10^{-6} — 10^{-2}	0.9063
E3	55	10	35	24.00	10^{-5} — 10^{-3}	1.000
E4	45	20	35	54.40	10^{-6} — 10^{-2}	0.9994
E5	40	25	35	7.20	10^{-5} — 10^{-2}	0.9863

Optimization of pH melamine solution

Figure 4 shows that the optimum pH is in the range of melamine solution acidic, at the range of pH 3-4 which shows the constant potential value. At pH <3, melamine will be hydrolyzed and transformed into amelina, amelida and cyanuric acid. Whereas at pH> 4 will result in electrically neutral melamine and improve potensial [16].

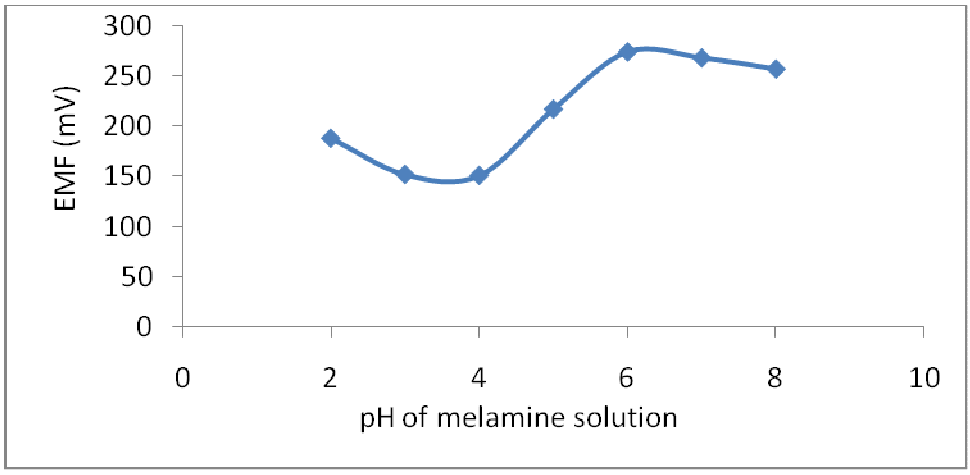

Figure 4 The effect of pH on the potential solution of melamine

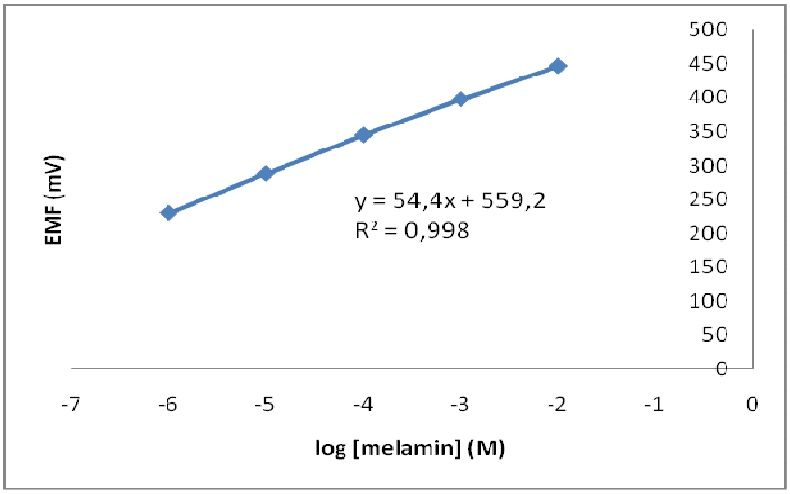

Figure 5 Calibration curve of melamine

Analytical validation method

Nanoporous carbon paste/MIP electrode (E4) gives the linear measurement range is quite wide, which is 10^{-6} - 10^{-2} by the regression equation *y = 54.4 +559.2* and the linearity is 0.998 (Figure 5). Nernst factor obtained was 54.4

mV / decade can be seen from the slope of the curve. The price indicates that the carbon electrode nanopori / MIP (E4) as melamine sensor response due to sub Nernstin have a value <59.1. Electrode response sub Nerstian < Nernstian 59.1 ± 2 < super Nernstian.

In this study, the results obtained from the detection limit of 9.51 x 10^{-7} electrodes M. These results indicate that the carbon paste electrode nanoporous / MIP good to be used, it has a low detection limit compared with the study of Liang et al [11] that has a detection limit of 6x10^{-6} M.

Determination of melamine solution precision performed at 10^{-3} M and 10^{-4} M were measured with the optimum electrode three times. The measurement results were then computed to determine the value of percent coefficient of variation (% CV). Based on calculations, the precision obtained for the solution of 10^{-4} M and 10^{-3} M respectively by 1.17% and 1.93% (Table 4). The results indicated good precision with a coefficient of variation values between 1-3 percent % [17].

Table 4 Measurement precision nanoporous carbon/MIP electrode

Concentration (M)	Potential (mV)			%CV
	Replication 1	Replication2	Replication 3	
10^{-4}	243	340	348	1.17
10^{-3}	397	384	398	1.98

The determination of accuracy is expressed by the percent recovery using comparing the measured concentration to the actual concentration in the solution. In this study, it is used a 10^{-4} M solution of melamine and 10^{-3} M to determine its accuracy. Based on calculations, the accuracy obtained for the solution of 10^{-4} M and 10^{-3} M respectively by 106.1% and 104.3% (Table 5). Good accuracy is in the range of 90-107% [18], so that the nanoporous carbon paste / MIP electrode in this study is a good electrode used to measure because melamine is still included in the allowable range.

Table 5 Measurement accuracy of the nanoporous carbon paste/MIP electrode

Concentration (M)	Potential (mV)	Accuracy (%)
10^{-4}	343	106.1
10^{-3}	397	104.3

An electrode has a selectivity to ions or molecules to be measured is determined by the coefficient selectivity [19] In this study, some of which are expected to interfere with ion analysis include $Ca^{2+,}$ K^{+}, Mg^{2+}, Na^{+}. Ions are also present in milk. Ion solution was prepared at a concentration of 10^{-3}M which is then measured by the magnitude of the electrode potential.

Table 6 Coefficient of selectivity nanoporous carbon paste/MIP electrode

Interfere ions	Selectivity Coefficient ($k_{i,j}$)
Ca^{2+}	6.77 x 10^{-3}
K^{+}	1.02 x 10^{-4}
Mg^{2+}	1.47 x 10^{-3}
Na^{+}	3.21 x 10^{-4}

If the value $k_{i,j}$ = 0, then the foreign ions do not interfere. If the value of $k_{i,j}$<1, then the ion selective electrode *i* (primary ions) than j ions (ion interference), and if the value of $k_{i,j}$> 1, the ion selective electrode ion *j* instead of *i* [6]. Since the results of the selectivity coefficient is smaller than 1, then the electrode is more selective towards melamine than on other ions, namely Ca^{2+}, K^{+}, Na^{+}, and Mg^{2+}.

CONCLUSION

Molecularly imprinted polymer (MIP) has successfully synthesized with methacrylic acid monomer material, melamine as a template and ethylene glycol dimethacrylate as crosslinker. Optimum conditions melamine analysis using nanoporous carbon paste/MIP electrode as potentiometric sensor is the ratio of nanoporous carbon, MIP and paraffin by 45:20:35 and at optimum pH 3-4. This sensor has a measurement range of 10^{-6} - 10^{-2} M, detection limit 9.51 x 10^{-7} M, the Nernst factor of 54.4 mV / decade and selective toward melamine.

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