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Electrochemical Sensor Based on Molecularly Imprinted Polymer (MIP) for Simazine Pesticide Detection

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***Abstract*— The use of pesticides and herbicides containing the active ingredient simazine must be carried out strictly so as not to cause adverse effects on humans and the environment. Measurement of contaminants in the form of *simazine* as one of the active ingredients of pesticides and herbicides is very important. An imprinted polymer molecule (MIP) has been made using methacrylic acid (MAA) as a functional monomer and ethylene glycol dimethyl acrylate (EGDMA) as a cross-linker. This research resulted in a molecularly imprinted membrane for the specific recognition of *simazine*. The electrochemical potential used with MIP coated electrodes provides both qualitative and quantitative detection of *simazine*. Polymer coated molecular-based potentiometric sensors (MIP) are promising analytical tools for developing highly selective analytical sensors. Optimal conditions for the production of *simazine* MIP were found to be 6.02 mL of chloroform, 0.025 g of *simazine*, 0.9 mL of MAA, 1.57 mL of EGDMA, and 0.07 g of benzoyl peroxide (BPO) with a heating time of 150 minutes at 70°C. The results of the electrode performance test resulted in stable and unstable conditions, with a measurement range on the surface of the double-layer electrode with a coefficient distribution of 0.9897. Compared with electrochemical procedures with MIPs sensors and spectrophotometry, it produces a significant value with a 95% confidence level with Q-test results for the 0.48 ppm level with spectrometric procedures obtained Q exp = 0.25, Q crit value is 0.71.**

***Keywords*—Simazine pesticide; molecularly imprinted polymer; electrochemical sensor.**

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I. INTRODUCTION

Pesticides and herbicides are active chemicals used to eradicate plant pests which are pollutants for the environment and humans who consume agricultural and plantation products [1]. The use of pesticides must be appropriately controlled to prevent environmental damage and reduce their impact on human health. More than 98% of insecticides and 95% of herbicides reach places other than their intended targets, including non-target species, water, air, food, and sediment [2], [3], [4]. Pesticides can reach and contaminate soil and water when sprayed from the air, allowed to flow from field surfaces, or allowed to evaporate from production and storage sites. Excessive use of pesticides can cause pest resistance to pesticides, so what happens is that the population of agricultural pests does not decrease but increases. Pesticide residues, namely pesticides that are left in large quantities from the crop are detrimental to human health [5], [6], [7].

Monitoring environmental pollution from pesticides and herbicides that affect human health and ecosystems has become a center of attention. The active ingredients in pesticides and herbicides include *simazine*, *amethrin*, *atrazine*, and others. This herbicide has low environmental persistence but a higher toxicity level [8], [9]. These pesticides and herbicides are toxic with their degradation products, such as *diethylsimazine*, which is a contaminant in surface water and groundwater [10], [11], [12].

Analysis of pesticide residues in food and the environment needs to be done to determine the level of toxicity and the risks posed to both living things and the environment. The techniques commonly used to detect pesticide residues are using gas chromatography (GC) or high-pressure liquid chromatography (HPLC) [13], [14], [15]. The weakness of the GC and HPLC analysis methods is the extraction and purification treatment in the laboratory, which requires a longer solvent and analysis time, thus allowing the risk of error. Therefore, to overcome these shortcomings, a new method is currently being developed to analyze simazine pesticide residues with electrochemistry using Molecular Imprinted Polymer (MIP) sensors coated [16], [17], [18].

Today the use of sensors is widespread in many types of modern equipment. Sensors are system elements that can capture and filter physical and chemical phenomena, then convert them into electrical signals in the form of electric current or voltage, generally known as electrodes. Currently, the use of electrochemical sensors in recognizing foreign objects in the surrounding environment has developed [7], [19], [20]. Related to previous research that has been done by other researchers, the novelty of this research is the method of making MIP and placing MIP on the surface of aluminum electrodes with the assumption that this material is a conductor that will facilitate ion transfer.

Molecularly Imprinted Polymer (MIP) is a synthetic polymer with a specific cavity for the target molecule. Cavities are obtained due to the removal of templates, where these cavities function to recognize molecules with the same size, structure, and physicochemical properties. The selectivity and affinity of the template itself will increase with increasing concentration values. The general principle of MIP is shown in Fig. 1 [21].

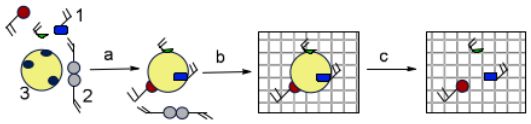


Fig. 1 General principles of MIP formation, (1) cross-linker, (2) functional monomer, and (3) template

In the synthesis of MIP, many parameters must be considered because these parameters can affect the polymer's morphology, characteristics, and usability. In MIP synthesis, the selection of chemical reagents is essential in producing an efficient functional MIP. MIPs are a crosslinking technique to produce a polymer (crosslinked polymers) that have cavities with a suitable place (template), where these cavities function to recognize molecules with the same size, shape, structure, and physicochemical properties with the presence of mechanical interactions based on molecular compatibility [22], [23], [24].

The polymer produced from this MIP technique can be applied to the surface of sensor materials with high selectivity and effectiveness, response speed, relatively low cost, and easy operation, so MIP is an alternative analysis as a detection and analysis instrument. The advantage of MIP is a sensor system that provides the results of a pollutant analysis quickly, easily, and reliably in trace amounts [25], [26].

Screen printed carbon electrode (SPCE) is an electrode that combines a carbon working electrode, a reference electrode, and a support electrode in a single compact and easy-to-use design. SPCE has been increasingly popular in electrochemical sensors in recent years. SPCE offers various advantages, including portability and ease of use, quick analysis, high efficiency, cheap cost, and small size. Because the sample is tiny, it is particularly promising for sensor development. However, because carbon-based electrodes such as SPCE are not selective, it is important to modify the electrode to improve its selectivity to the target analyte. Using molecularly imprinted polymers can improve SPCE selectivity for certain analytes (MIP). MIP has a mold molecule imprint (typically analyte molecules), increasing selectivity towards analyte molecules. Simazine MIP is a frequently used MIP for electrochemical detection (MIPs). Simazine is frequently utilized because it is conductive, readily soluble in water, and electrochemically produced. MIP was synthesized in this study using an electro-polymerization process [24].

II. Materials and Method

A. Preparation of Producing MIPs

The material used in the form of a pre-polymer solution is a mixture of *simazine* as a template, methacrylic acid (MAA) as a monomer, ethylene glycol methacrylic acid (EDGMA) as a cross-linker, and benzoyl peroxide (BPO) as an initiator. BPO was chosen because this substance has stable free radicals, so it tends to react with monomer molecules reactively. The selection of the right cross-linker and solvent will affect the resulting polymer.

The following procedure is used to synthesize MIP and NIP polymers: In brief, a simazine 0.025 gr template was prepared, and solvent chloroform 2.1 mL, cross-linker EDGMA 0.525 mL, functional monomer MAA 0.059 mL, and initiator BPO 0.05 gr were added to it, for 15 minutes, all of the components were thoroughly mixed. A simazine-free polymer was also created. After that, the reaction vial was kept in the freezer at 4°C for 1 hour and then at 70-120°C (cool and hot process) for 150 minutes to complete polymerization.

The polymer was ground after being transferred from the vial to the mortal. The polymers were collected in an acetonitrile solvent, filtered, and washed for 1 hour in methanol and water (aquabidest). The remaining filtrate residues were also collected for drying and future use. The scheme for making MIPs is shown in Figure 2.

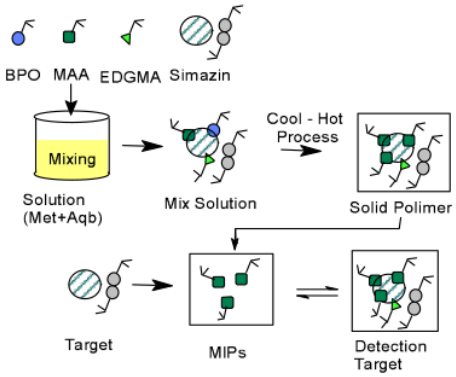


Fig. 2 The scheme for making MIPs

B. Electrochemical Sensor Preparation

Potentiostats are used to perform electrochemical experiments. The general procedure for making potentiometric sensor-based MIP from aluminum wire (1 mm in diameter and 120 mm in length) and carbon consists of several steps. In the first stage, the electrode was prepared with a wire glued to the resin as the electrode body. The surface of the wire was cleaned, sonicated for 15 minutes in distilled water, and dried in air. One end of the electrode wire is glued with aluminum and glued with unsaturated resin. In the second stage, the MIPs that have been made are then glued to the wire electrodes by coating the ends of the electrode

surfaces with MIPs and eluting them with chloroform solution on the surface of the MIPs-coated sensor. To ensure the working surface of the electrode surface is polished with sulfite paper. The schematic of MIP placement on the carbon surface is shown in Figure 3.

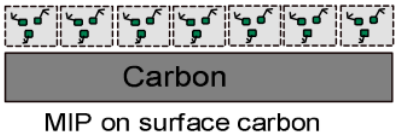


Fig. 3 MIP placement on the carbon surface

C. Analytical Technique for Determining Simazine

A potentiometric detection technique was used to investigate the electrochemical behavior of *simazine* at these electrodes. The potentiometric sensor is a sensor used to measure the voltage that responds to changes in activity in the test solution [21], [22], [23]. A micro *potentiostat* with a three-electrode electrochemical cell including an Ag/AgCl reference electrode, an aluminum wire as the counter electrode, and a carbon paste working electrode. Characterization of the electrode as a sensor using potentiometric detection measurements. The sample solution was prepared and allowed to stand for 10 seconds before detection using potentiometry and then measured and recorded in the potential range of -0.6 to +1.6 V with optimum conditions at +0.4 V. The concentration of the simazine solution sample was artificially 0.3 ppm up to 0.7 ppm, each measured potentiometrically with variations in measurement time between 30 to 90 minutes with optimum potential. Block diagram of potential measurement using MIPs coated electrodes is shown in Figure 4.

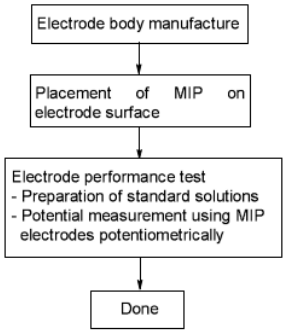


Fig. 4 Block diagram of potential measurement using MIPs coated electrodes

III. Results and Discussion

A. Optimum Conditions for Producing Molecularly Imprinted Polymer

The optimal conditions for MIPs are influenced by the correct composition of the constituents for the successful manufacture of MIP, from studies using a number of simazines, BPO with chloroform solvent, and an excess of moles of MAA and EDGMA to form simazine templates, BPO acting as an initiator or as a catalyst. MIP production is done by mixing all the ingredients in sequence and stirring. The initial mixture of methacrylic acid has a larger mole ratio than *simazine*, with the aim that when reacted, a simazine template will be formed, which is surrounded by methacrylic acid. Based on research on the manufacture of MIP simazine, data on the optimum conditions for making MIP can be seen in Table 1 as optimization of MIPS production time.

TABEL I
OPTIMIZATION OF MIPS PRODUCTION TIME

Sample	Production time (minutes)	MIP Physical Characteristic
1	30	White solid
2	90	Clear liquid
3	120	Clear liquid
4	150	Clear solid

The polymerization process is considered complete when the clear liquid heated at a 700°C turns into a clear acrylic solid like the color of methacrylic acid (MAA) attached to the vial. The MIP that has been produced is then ground until a fine MIP powder is produced. Furthermore, washing is carried out to remove the remnants of the reactants in the polymerization process and remove the simazine template. The manufacture of MIP is influenced by several factors, namely the time of the manufacturing process, the composition of the MIP-forming materials, and stirring. Based on the optimum condition data in Table 2, it can be seen that the optimum time for making MIP is 150 minutes.

TABLE II
OPTIMIZATION OF MIPS PRODUCTION COMPOSITION

Sample	Composition	MIP Physical Characteristic
1	2.01 mL chloroform; 0.3 mL MAA; 0.525 mL EDMA; BPO 0.07 g; 0.025 g *simazine*	White solid
2	6.02 mL chloroform; 0.9 mL MAA; 1.575 mL EDMA; 0.07 g BPO; 0.025 g *simazine*	Clear liquid

Making MIP requires steady stirring for successful MIP creation. Unstable stirring will cause the intermolecular bonds to become tenuous, which causes failure. The composition greatly affects the success of making MIP. Based on research, the appropriate composition for the success of making MIP is to increase each ingredient to three times the original, except *simazine* and BPO. The number of moles of MAA and EDMA was added in excess to form a simazine template surrounded by the material. The amount of BPO remains in the initial composition and is not added because BPO acts as an initiator or catalyst.

Figure 5 shows the chemical structure of *simazine* MIP, the simazine molecule surrounded by methacrylic acid, indicating that this compound can act as both a hydrogen binder and proton donor and a hydrogen binding acceptor. MIP simazine by washing has a larger number of pores because the printed simazine template will be released due to the washing process. The washing process was carried out to obtain a completely clean MIP from the simazine template, an illustration of the washing process can be seen in Figure 6. The results of this washing will produce a specific simazine cavity.

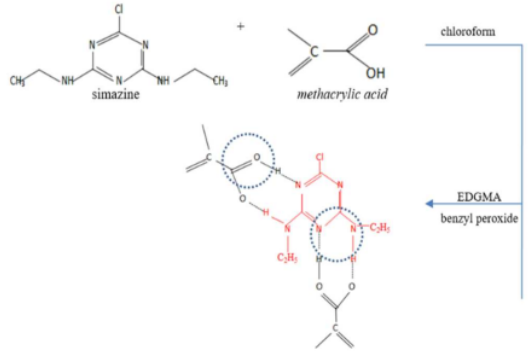
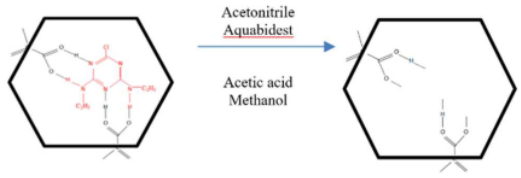


Fig. 5 The chemical structure of *simazine* MIP

Simazine template has been through a washing process so that it can function as a hydrogen binder with a higher number of pores. This washing process was carried out to obtain completely clean MIPs from the simazine template resulting in a specific simazine cavity, as shown in Figure 6.



(a) (b)

Fig. 6 (a) Before washing structure of template MIPs, (b) After washing structure of MIPs

Furthermore, the washed simazine template is placed on the surface of the electrode as a MIP sensor which has a working principle of lock and key theory on enzymes and substrates, and only simazine molecules can fill the cavity in the MIPs.

B. Determination of Steady State Measurement Conditions with MIP Sensors

Determination of steady and unsteady state profiles is carried out to determine the value of constant current at the time of measurement, which will later be used as the measurement area. This profile is determined by measuring several concentrations of simazine standard solutions at a fixed working potential of 0.7 V, using variations in simazine concentrations of $0.05x10^{-3}$ ppm, $0.2x10^{-3}$ ppm, $0.4x10^{-3}$ ppm, $0.6x10^{-3}$ ppm, $0.8x10^{-3}$ ppm, and $1.0x10^{-3}$ ppm. The current measurement at each potential work value is carried out three times, and the current recording is carried out every 2 seconds, starting from 0 seconds until it shows a steady state current value.

The measurement profile curve shows a working potential of 0.7 V produces a steady and unsteady state area. Based on the curve in Figure 7, it can be seen that the unsteady state condition is at 0-40 seconds. In this condition, simazine degradation occurs very quickly, as seen from the change in the reduction current that occurs at 0-40 seconds with a current of 2.8 A to 0.5 A. This can be explained by the fact that in the initial state (before the application of the working potential), there is only simazine equilibrium in the electrode-electrolyte-solution interface.

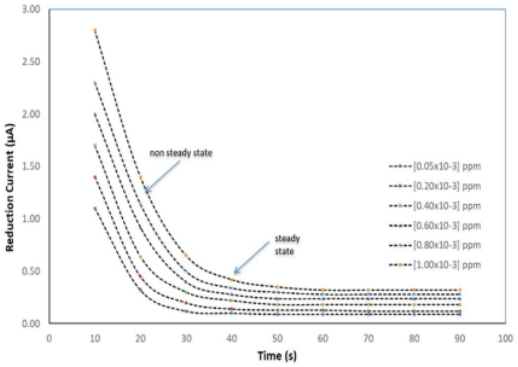


Fig. 7 Steady and unsteady state profile curves at reduced current versus time with a working potential of 0.7V

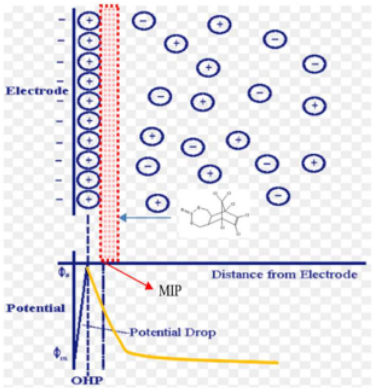


Fig. 8 Model of the electric double layer on the surface of the simazine and simazine MIP solution-electrodes

A charged electrode immersed in an electrolytic solution will repel barrier ions to its surface and form two layers of opposite polarity at the interface between the electrode and the electrolyte. The electric double layer (DL) is a molecular dielectric with an electrostatic charge, and the condition of the charge travel can be expressed in steady and unsteady conditions. Fig. 8 shows the movement of ions toward the electrode surface.

C. Validation of MIPs and UV-Vis Sensor Analysis Procedures

The MIP analysis method for contamination uses artificial samples by validating the measurement method using MIPs coated electrode sensors (MIPs sensors) and spectrophotometric methods (UV-Vis). The accuracy of the simazine level determination procedure using the MIPs sensor is determined based on the standard deviation and the average value of the measured simazine level from the results of the standard solution measurements performed. The accuracy of the procedure for determining the dissolved simazine level using the simazine MIP sensor was determined based on the standard deviation and the average value of the measured simazine level from the measurement results of the standard solution carried out. The results of calculating the accuracy of

the soluble simazine level determination procedure using MIPs and UV-Vis sensors are shown in Table 3.

TABLE III
ACCURACY OF SIMAZINE LEVEL MEASUREMENT USING MIPS SENSORS AND UV-VIS

Simazine Contents (ppm) 1 x 10^{-3}	Measurable average dissolved simazine levels (ppm)		Standard Deviation (Sd)		Accuracy (%)		Accuracy against UV-Vis (%)
	MIP	Vis	MIP	Vis	MIP	Vis	MIP
0.3	0.25 ± 0.14	0.36± 0.09	0.08	0	7.41	1.4	30.32
0.4	0.42 ± 0.12	0.44 ± 0.09	0.10	0	1.85	0.4	1.50
0.5	0.54 ± 0.14	0.53 ± 0.09	0.15	0.1	1.40	0.8	0.38
0.6	0.65 ± 0.12	0.64 ± 0.02	0.20	0.1	1.31	0.7	0.39
0.7	0.78 ± 0.33	0.74 ± 0.05	0.15	0.1	0.72	0.4	0.05

Accuracy indicates how near the measurement results are to the real value, and precision indicates how close the value difference is between repeated measurements. Comparative analysis of measurements with MIPs and UV-Vis sensors was carried out by knowing the absorbance of UV-Vis absorption and calculating the accuracy of the procedure for determining levels of dissolved *simazine* in the same concentration range ($0.30x10^{-3}$ - $0.70x10^{-3}$ ppm), determining standard deviations, and the average value of dissolved simazine levels. The results of the calculation of measurement accuracy with the MIPs sensor obtained a range of ± 0.12 - ± 0.33, and the UV-Vis procedure was obtained in an accuracy range of ± 0.02 - ± 0.09.

The range of accuracy is obtained from the alleged noise on the sensor that appears due to small readings on the measuring instrument. However, based on the results obtained, the sensor is very suitable for measurements at larger concentrations, such as at a concentration of 0.7 x 10^{-3} ppm and standard deviation MIPS sensors at 0.08 – 0.20. UV-Vis results at 0–0.1 provide accuracy values and show measurements that there are no significant differences.

Measurements' repeatability was measured using MIPs and UV-Vis sensors to see the repeatability of the measurement results, which showed the proximity of the measurement results sequentially with the same method and identical artificial simazine samples. The results of the calculation of the repetition of the measurement procedure using MIPs and UV-Vis sensors are shown in Table 4.

TABLE IV
REPEATED MEASUREMENTS USING MIPS AND UV-VIS

Simazine Contents (ppm) 1 x 10^{-3}	Measurable average dissolved simazine levels (ppm)		Repeatability (ppm)	
	MIP	Vis	MIP	Vis
0.3	0.25 ± 0.14	0.36 ± 0.09	± 0.27	±0.06
0.4	0.42 ± 0.12	0.44 ± 0.09	±0.46	±0.06
0.5	0.54 ± 0.14	0.53 ± 0.09	±0.52	±0.27
0.6	0.65 ± 0.12	0.64 ± 0.02	±0.66	±0.33
0.7	0.78 ± 0.33	0.74 ± 0.05	±0.50	±0.27

Measurement repeatability and reproducibility by ANOVA is a measurement system analysis approach that assesses the measurement system using the analysis of the variance random effects model. Evaluation of a measurement system includes all kinds of measuring instruments, test procedures, and other measurement systems, not only measuring instruments. The measurement was taken using MIP and UV Vis sensors, with the repeatability value of the MIP sensor in the minimum range of 0.27 and the maximum range of 0.66 and the repeatability value of the UV Vis sensor in the minimum range of 0.06, and the maximum range of 0.33. These results show that the measuring instrument can still be repeated and surpass the repetition standard.

The behavior of electrochemical measurements using MIPs is the accumulation of 0.4 V potential for the first time, which concentrates *Simazine* in the MIPs cavity. This can be seen in non-steady conditions where rapid degradation occurs for 40 seconds, after which the process is continued for up to 90 seconds. The measured simazine electrons experienced steady tangle conditions towards the MIP surface of the MIPs, which can be seen in the measurement time range of 40 - 90 seconds. It indicates that the measurement of artificial simazine samples after the MIPs are immersed in the sample solution can be measured in 40 to 90 seconds.

In the presence of *simazine*, only the MIP simazine sensor showed an irreversible decrease profile, as predicted. This answer implies that MIP may be used as a recognition element in sensor architecture to determine simazine levels. From each type of dissolved simazine standard solution. The results of the calculation of the repeatability of the measurement procedure using the simazine MIP sensor are shown in Table 5.

TABLE V
REPEATABLE OF MEASUREMENTS USING THE MIPS SENSOR ON UV-VIS ABSORBANCE

Simazine Contents (ppm) 1 x 10^{-6}	Measurable average dissolved simazine levels (ppm)		Repeatability (ppm)	
	MIP	Vis	MIP	Vis
0.3	0.25 ± 0.14	0.36 ± 0.09	± 0.27	±0.06
0.4	0.42 ± 0.12	0.44 ± 0.09	±0.46	±0.06
0.5	0.54 ± 0.14	0.53 ± 0.09	±0.52	±0.27
0.6	0.65 ± 0.12	0.64 ± 0.02	±0.66	±0.33
0.7	0.78 ± 0.33	0.74 ± 0.05	±0.50	±0.27

After using the accumulation potential (0.4 V) to concentrate *Simazine* in the paste's MIP cavity, the applied potential resulted in a quantitative reduction in *simazine,* and the paste was returned to solution. Using cyclic voltammetry, the electroreduction of *simazine* in HCl at pH 3.5 was examined for standard carbon paste electrodes and MIP-modified electrodes (MIPs). Fig. 9 shows the voltammograms with MIPs.

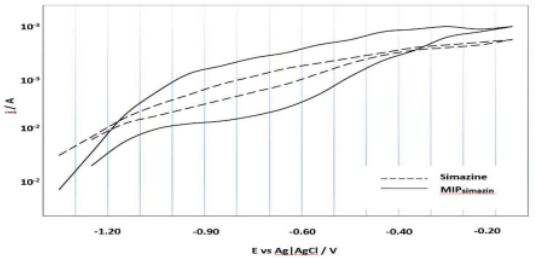


Fig. 9 Cyclic voltammograms acquired with MIP for 100 nmol L1 Simazine. simazine, simazine, simazine Measurements were performed at a scan rate of 50 mV s1 in HCl (pH 3.5)

At different pH, simazine reduction was investigated, and a drop in current was noted at pH > 3.5. Because *simazine* is electrochemically active, the effect of changing the pH from 1 to >4 was investigated. As the pH climbed, the peak height fell, and no current was measured at pH > 4. Because preconcentration and electrochemical measurement of *Simazine* yielded the best results at a pH of around 3.5, solutions with this pH value were utilized.

Application of the Method on Environmental Samples, in comparing the procedures for determining the levels of dissolved *simazine*, measurements were made of environmental samples sourced from plantation areas that used pesticides or herbicides containing the active ingredient simazine. Measurements using MIPs and UV-Vis were repeated six times each. Sample measurement data by repeated measurements six times, as shown in Table 6.

TABLE VI
REPEATABILITY OF SAMPLE MEASUREMENT WITH MIPS AND UV-VIS

Measurement	Simazine levels by MIP (ppm) $1x10^{-3}$	Simazine levels by UV-Vis (ppm) $1x10^{-3}$
X_1	0.51	0.48
X_2	0.44	0.42
X_3	0.42	0.44
X_4	0.48	0.38
X_5	0.42	0.41
X_6	0.49	0.48

From Table 4, the measurement results show that the measurement range does not appear to be significantly different, which was previously tested to determine whether there is a measurement deviation between the two methods of MIPs and UV-Vis analysis using the Q-test.

The results of the Q-test for the measurement level of 0.51 ppm with the MIPs procedure against the measurement level of 0.48 ppm with the UV-Vis spectrometry procedure obtained Q_{exp} = 0.25 while the Q_{crit} value at the 95% confidence level is 0.71, so $|Q_{exp}| < |Q_{crit}|$. The results of the Q test for 0.48 ppm levels with the UV-Vis absorbance procedure obtained Q_{exp} = 0.13, so that $|Q_{exp}| < |Q_{crit}|$, this indicates that the two data on the MIPs sensor measurement and UV-Vis absorbance are measurement data that do not deviate, measurement data from environmental samples can still be used and do not have deviant or outlier data.

IV. CONCLUSION

MIPs are polymers printed from a mixture of MAA as functional and EGDMA as crosslinking monomers. Developed a method for the determination of simazine residue. Molecularly printed polymer matrices specifically for pesticides and herbicides with *simazine* as the active ingredient can be used to manufacture MIP composite membranes. In this study, molecularly fabricated membranes were used to recognize pesticides and herbicides with the active ingredient simazine.

The results of testing the performance of the MIP sensor, referred to by UV-Vis measurements, show that the aluminum-carbon MIP sensor has a detection limit of 0.3 ppm, is sensitive in the concentration range of 0.25 to 0.78 ppm with and Nernst factor > 0,059 V/decade which has good stability.

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