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Carbon/Alginate/Chitosan Composite as a Sorbent for Solid-Phase Extraction and Preconcentration of Cu(II)

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Abstract: *Carbon/Alginate/Chitosan Composite (CAC) as a sorbent for solid-phase extraction of Cu(II) (pre-concentration) with atomic absorption spectrometry for analysis was successfully prepared. The sorbent was made with a mass ratio of chitosan:alginate:carbon = 1:1:1. CAC sorbents were characterized using FTIR for functional group analysis while SEM-EDX characterization was performed to determine surface morphology and percentage of elements. SPE parameters were optimized for adsorption-desorption efficiency under various conditions, i.e. of pH 5, Cu(II) concentration of 4 mg/L, the flow rate of Cu(II), and Na₂EDTA eluent at 5 mL/min, the volume of Cu(II) sample and Na₂EDTA eluent 25 mL and 1.0 M of Na₂EDTA eluent concentration. Applying CAC-based SPE for Cu(II) analysis in water samples at optimum conditions gave 80.9–83.6% recovery. In bottled drinking water samples, a pre-concentration factor of 2.5–5 times was obtained, and the Cu(II) concentration of 0.017–0.020 mg/L was detected.*

Keywords: *alginate; chitosan; solid-phase extraction; preconcentration; Cu(II)*

■ INTRODUCTION

Over the past few years, toxic metal pollution has become one of the most severe problems in ecological [1]. Cu(II) is one of the various toxic metals. Moreover, we know that Cu(II) is a vital micronutrient. Still, excess Cu(II) causes multiple diseases such as Alzheimer's [2], intestinal allergy and rectum discomfort disorder in both liver and kidney function, and hemoglobin shortage [3]. Therefore, the European Union 2014 has set the recommended limit maximum for Cu(II) in clean water to be 2 mg/L [4]. According to the Indonesian Standard SNI 01-3553-2016, the criteria for the maximum concentration of Cu(II) for clean water and drinking water are 1.0 and 0.5 mg/L, respectively [5].

As a result, accurate methods for determining Cu(II) in water and environmental samples are becoming increasingly crucial in analytical and waste chemistry [6]. However, the general procedures of heavy metals for Cu(II) removal from aqueous solutions, including chemical batch reaction and adsorption, may be ineffective and need more time [7]. Moreover, the disadvantages of both

methods are that the template ions embedded in the obtained matrices are hard to elute and have poor adsorptivity [8]. Among many techniques, solid-phase extraction (SPE) is a well-established method because of its high recovery, low cost, higher efficiency [9], large adsorption-desorption, sensitivity, simple operation, environmental friendliness, and removal of interference components from various sample matrices such as heavy metals [10]. The prerequisites for effective extraction depend on choosing a suitable adsorption material in the SPE. Sorbent for use in SPE must contain several capabilities, such as fast sorption kinetics for analyte ions, high selectivity, and suitable size to allow a continuous flow of aqueous solution [11].

Chitosan is a linear polysaccharide biopolymer obtained by the deacetylation of chitin. Chitosan consists of D-glucosamine and *N*-acetyl-D-glucosamine units linked by β-(1-4) glycosidic linkages [12]. Chitosan is a cationic polymer with sorption applications due to its lower density of amino groups [13]. One of the most popular natural biodegradable linear polysaccharides

derived mainly from gulfweed, bacteria, or brown algae and consists of (1-4) linked α-L-guluronic acid (G-unit) and β-D-mannuronic acid (M). Previous research has confirmed that as an SPE adsorbent, chitosan film can absorb Cu(II). However, Cu(II) adsorption is only 63%. Therefore, the adsorption process is less than optimal, causing the desorption process in SPE to be less optimal and decrease efficiency [14]. Adsorbed Cu(II) percentage can be optimized with an adsorbent material with a functional group as a more robust Cu^{2+} ion binder. One such functional group is the carboxylates group in alginates. The research used adsorbents from alginate-modified chitosan with a function group –COOH. Cu(II) adsorption infectivity has a percentage of more than 85%, with a contact time of 60 min [6].

Carbon is mainly used to remove organic compounds for water treatment and medical purposes. Among carbon-based materials, activated carbon has attracted increased attention with its chemical stability, high adsorption capacity, and production feasibility. Carbon has also attracted great interest in environmental concerns for use as an adsorbent in wastewater treatment [15]. Furthermore, adding materials such as activated carbon can increase adsorption capacity against metal ions because of pores and active groups [16]. Compared with other materials, chitosan modified with alginate and carbon (CAC) can also be used as a sorbent in SPE. CAC reduces lost Cu(II), and SPE speeds up analysis time.

This study aims to produce a new functional SPE with CAC as the sorbent of chitosan modified with alginate and carbon to analyze Cu(II) in an aqueous solution. The optimum condition of SPE was evaluated as optimal pH, enrichment factor, sample and eluent concentration, sample and eluent flow rate, and sample and eluent volume.

■ EXPERIMENTAL SECTION

Materials

The materials used in this study were chitosan (%DD > 75%), Na-Alginate (G/M = 2/1), and activated carbon from Sigma-Aldrich. The solvent used distilled water from PT Progo. Other ingredients are CH_3COOH, $CuSO_4 \cdot 5H_2O$, HNO_3, Na_2EDTA, and $CaCl_2$, which are

chemicals with pro-analytical quality (p.a) purchased from Merck.

Instrumentation

This study used sorbent characterization tools, such as Fourier-Transform Infrared Spectrophotometer (FTIR, Prestige-21, Shimadzu) and Scanning Electron Microscopy-Energy Dispersive X-Ray (SEM-EDX, Joel Jsm 6360LA Phenom-BSD Detector). Meanwhile, Atomic Absorption Spectrometer (AAS) (Perkin Elmer 400) was used for the analysis of Cu(II) in an aqueous solution. For the SPE process, use a glass column 1 cm × 10 cm and a peristaltic pump from Intlab.

Procedure

Synthesis of carbon/alginate/chitosan composite sorbent (CAC)

A total of 1 g of chitosan was added to a 40 mL of 2% acetic acid solution pH 3 and stirred until dissolved. Separately, 1 g of alginate was dissolved in 40 mL of 2% acetic acid. Also, 1 g of activated carbon was dispersed in 2% acetic acid (40 mL). Chitosan, alginate, and carbon solutions were mixed and stirred using a magnetic stirrer for 7 h until homogeneous. The homogeneous solution was then put into a syringe and dripped into a solution of $CaCl_2$ 2 M to form beads. Beads were stored in the refrigerator for 24 h. The beads were then filtered and dried in an oven at 50 °C for 3 h. The dried beads were then crushed to become CAC sorbent powder, sieved, and stored. The CAC mass used for each SPE experiment was 30 mg.

Parameter optimization of SPE method with CAC sorbent

In this study, the parameters of SPE conditions observed were pH, the concentration of sample and eluent, the flow rate of sample and eluent, the volume of sample and eluent, and the effect of eluent concentration on real samples.

Sample preparation of tap water and bottled drinking water

The effectiveness test was carried out with two samples, namely samples of tap water and bottled drinking water. Tap water samples were prepared by

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spiking Cu(II) to determine the percentage of recovery and the optimal value of the concentration enrichment factor. Then 5 samples with a volume of 25 mL prepared were analyzed by the SPE method under optimal conditions, with the eluent volume of Na$_2$EDTA being 5, 10, 15, 20, and 25 mL, respectively. Furthermore, 5 samples of bottled drinking water were taken with a volume of 25 mL to test the enrichment factor's effectiveness. Then the samples were analyzed using the SPE method under optimal conditions with a volume of 5 to 10 mL of Na$_2$EDTA eluent.

■ RESULTS AND DISCUSSION

Characterization of CAC Using FTIR

The FTIR analysis was aimed to determine the functional groups in the CAC sorbent. Fig. 1 shows FTIR spectra of carbon, alginate, and chitosan. The absorption peaks of activated carbon were observed at 3425, 1620, 1381, and 1118 cm^{-1}, which show –OH stretching vibrations, C=C stretching vibrations, CH$_2$OH stretching vibrations, and C–O stretching vibrations, respectively [6]. Chitosan spectra showed absorption peaks of 3448, 1620, 1419, 1327, and 1064 cm^{-1} [17-18]. The peaks indicate the presence of functional groups in chitosan, i.e.,

O–H and N–H (primary amino), C=O (secondary amide of the acetamide), N, C–N, and C–O [19]. The alginate spectra showed absorptions at 3425, 1620, 1419, and 1087 cm^{-1}. The absorptions show the –OH group vibration, the wide-stretching vibration of the asymmetric carboxylate group –COO$^-$, the symmetrical –COO$^-$ stretching vibration, and the C–O–C stretching vibration of the sugar ring functional group [20].

The CAC sorbent spectra showed the absorption of a combination of carbon, chitosan, and alginate materials. At 3448 cm^{-1} absorption, it shows the vibration of the –OH group coincides with the primary amino's N–H. Although the absorption shift was not detected significantly, the peak area formed became larger between 3000 and 3700 cm^{-1} [19] due to the formation of hydrogen bonds between chitosan, alginate, and carbon. In the stretching vibrations of –COO$^-$ asymmetric and –COO$^-$ symmetrically, there was a shift from 1620 to 1627 cm^{-1} and 1419 to 1427 cm^{-1} [21]. This vibrational shift indicates an ionic interaction between the carboxylate group –COO$^-$ of alginate and chitosan's amino group (–NH$_3^+$) [22]. In addition, a new weak absorption peak at 1527 cm^{-1} was identified as a Ca^{2+} ion as a crosslinker and electrostatically bound to the carboxyl group [23].
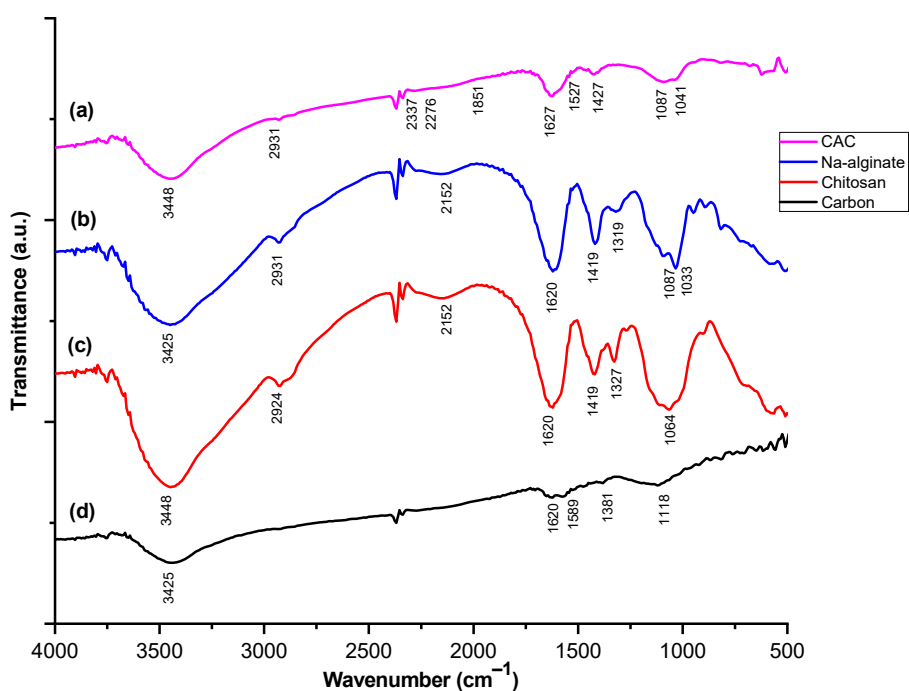

Fig 1. IR spectra of carbon, chitosan, Na-alginate and CAC sorbent

The reaction that occurs is the interaction of the active sites of alginate and chitosan that interact with each other and the added activated carbon so that the dominant active site in this compound is $-COO^-$ from alginate. The interaction happens because alginate is rich in the active site $-COO^-$ when interacting with chitosan. Therefore, it will bind with $-NH_3^+$ from chitosan to form polyelectrolyte complexes. This modification makes CAC rich in carboxyl active groups of alginate and hydroxyl from chitosan and carbon [20].

Characterization of CAC Using SEM-EDX

The aim of the characterization of bead powder using SEM-EDX was to see the characteristics of the morphology and elements of the CAC sorbent. Analyzes were performed at magnifications of 1500 and 5000 times for each sample. Fig. 2(a) and (b) show the CAC sorbent surface in SEM.

Fig. 2(c) shows the most significant percentage in a row on the sorbent CAC is an element of C, followed by O, N, and Ca at the smallest percentage. Ca was detected due to Ca as a crosslinker that connects the carboxyl group on the sorbent CAC. Before being applied with the method of SPE on the sorbent, CAC has not detected the presence of Cu(II) or other metals so that it can be used as a sorbent in the SPE for the analysis of metal Cu(II) [17]. In general, CAC sorbents consisting of chitosan, alginate, and carbon have pores prepared to adsorb Cu(II) metal and a high surface area of CAC sorbents to increase SPE efficiency.

SPE Condition

Optimal pH

The pH test of the solution is one of the essential parameters in the SPE method. In this study, the acidity of the Cu(II) solution was studied at a pH range of 2–6, with the initial sample conditions being 20 mL and the eluent of 10 mL. Then the flow rate of the sample and eluent is 15 mL/min. We chose a flow rate of 15 mL/min because this flow rate still allows sufficient contact of CAC with the sample and the analysis time is also shorter. Then the concentration of Cu(II) was 4 mg/L, and the concentration of Na_2EDTA was 0.1 M.

The effect of the pH of the Cu(II) solution can be seen in Fig. 3(a). It showed a difference in the percentage of Cu(II) absorption at each pH of the solution. The optimal percentage (%) of adsorption, desorption, and SPE efficiency is at pH 5. At pH conditions of higher than 5, Cu(II) solution will form metal speciation changes, Cu(II) to $Cu(OH)^+$, $Cu_2(OH)_2^{2+}$, and $Cu(OH)_2$ even CuO. This change causes the Cu(II) metal ion bound to the CAC sorbent in the SPE system to decrease, so the percentage (%) of adsorption decreases [6,23].

At low pH conditions (≤ 5), H^+ ions can occupy the active site of the CAC sorbent in the SPE system, which causes competition with Cu(II) ions [24]. Conversely, the lower pH of the solution will cause the number of H^+ ions increases, so the active site of the CAC sorbent will be occupied by more H^+ ions than Cu(II) ions, thereby reducing the percentage (%) of adsorption in SPE [25].

Cu(II) sample concentration

The effectiveness of SPE in the analysis of Cu(II) in water samples was determined using the concentration of the sample. The Cu(II) metal content is usually very low in real water samples. For this purpose, it was determined
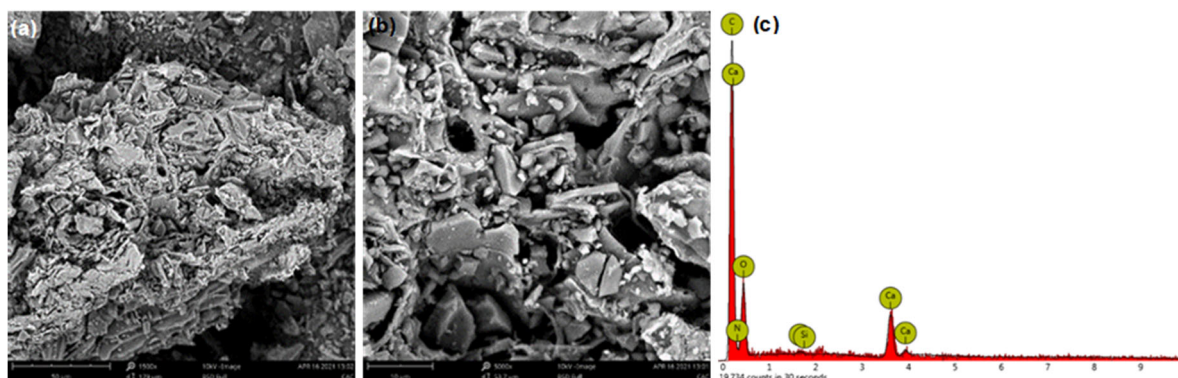

Fig 1. (a) CAC magnifications of 1500 and (b) 5000 (c) elements of CAC sorbent from EDX
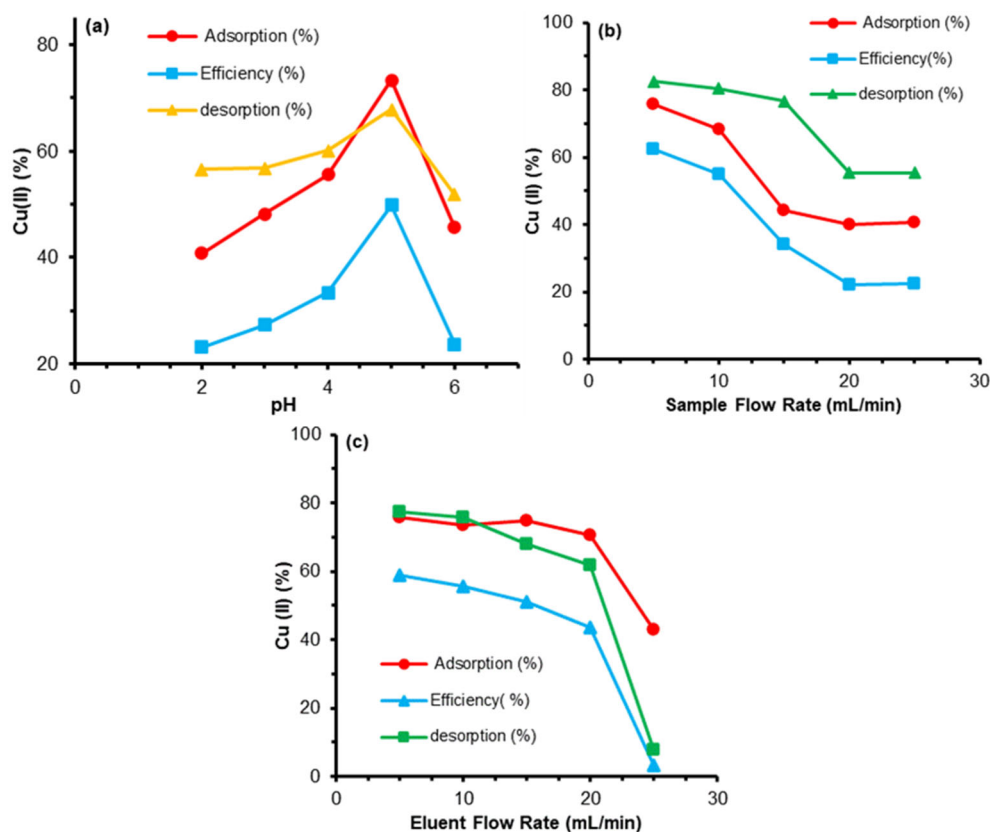
Fig 2. Effect of (a) pH, (b) sample flow rate, (c) eluent flow rate on the percentage of adsorption of Cu(II) metal on adsorption, desorption, and SPE efficiency

that the test concentration of the Cu(II) sample was 2–5 mg/L at the optimal pH of 5. The condition of the SPE was the sample and eluent flow rate of 10 mL/min. The sample volume of Cu(II) and eluent were 10 mL each, and the concentration of Na$_2$EDTA eluent was 0.1 M. The test results showed that the optimal percentage of adsorption, desorption, and efficiency of SPE Cx/Ca was at a concentration of 4 mg/L. At a concentration of 5 mg/L, the absorption has entered the saturation point to decrease slightly. Using samples with high concentrations generally gives good results because of the enrichment factor (EF), which can affect the analyte results from the SPE method [26]. However, under certain conditions, the sample concentration becomes constant because the sorbent is saturated and can no longer absorb the Cu(II) in the solution [27].

Cu(II) sample flow rate

In the SPE method, the flow rate influences the adsorption and final analysis. Therefore, the effect of the

sample flow rate on the SPE efficiency was studied by varying the sample flow rate, namely 5, 10, 15, 20, and 25 mL/min, with the sample condition at optimal pH 5, and the sample concentration of Cu(II) 4 mg/L. Then the other standard conditions were an eluent flow rate of 10 mL/min, the volume of Cu(II) sample, 10 mL eluent, and 0.5 M eluent concentration.

Fig. 3(b) showed that the flow rate of 5 mL/min was the most optimal because, at a flow rate of 10–25 mL/min, the Cu(II) sample decreased in contact time with the CAC sorbent, causing the percentage to decrease. The decrease in adsorption power on SPE indirectly affects SPE's desorption rate and efficiency because the concentration of Cu(II) adsorbed is also smaller [28].

Na$_2$EDTA eluent flow rate

In general, the effect of the eluent flow rate will directly impact the desorption percentage. Therefore, in the SPE method, the eluent flow rate was studied by varying the Na$_2$EDTA eluent flow rate of 5, 10, 15, 20,

and 25 mL/min with the sample conditions at optimal conditions, namely pH 5, sample concentration of Cu(II) 4 mg/L, and sample flow-rate of 5 mL/min. In addition, the sample volume of Cu(II) 10 mL and 0.1 M eluent concentration are other standard conditions.

Fig. 3(c) shows the desorption percentage at an eluent flow rate of 5 mL/min, which is optimal. Meanwhile, at the eluent flow rate of 10–25 mL/min, the percentage of desorption gradually decreased due to a decrease in the contact time of the Na_2EDTA eluent with the CAC sorbent, which had adsorbed Cu(II), which had an impact on the amount of Cu(II) analyte that was successfully desorbed [25].

Cu(II) sample volume

Cu(II) sample volume test is one of the parameters in the SPE method. In this study, the volume of Cu(II) samples was studied at 5, 10, 15, 20, and 25 mL under optimal conditions, namely pH 5, sample concentration of Cu(II) 4 mg/L, sample flow rate, and eluent 5 mL/min. Other standard conditions are 25 mL eluent volume and 0.1 M Na_2EDTA eluent concentration. The sample volume of Cu(II) showed an effect from 5–25 mL. As a result, there was an increase in the percentage, desorption, and efficiency of SPE. Optimal conditions were achieved at the sample volume of Cu(II) of 25 mL. The highest EF was obtained at a volume of 25 mL than at a volume of 5–20 mL. The EF at a volume of 25 mL is greater than that of a volume of 5–20 mL, as evidenced by the contact of the Cu(II) sample with the longer CAC sorbent in the SPE method. As a result, the adsorption percentage increases and is indirectly followed by the rise in desorption and SPE efficiency [29].

Eluent volume of Na_2EDTA

The eluent volume affects the extraction efficiency, which is indicated by the desorption percentage. The greater the rate of desorption, the more efficient the SPE process. In this study, the volume effect of Na_2EDTA eluent was studied at 5, 10, 15, 20, and 25 mL with optimum conditions, namely pH 5, sample concentration of Cu(II) 4 mg/L, the flow rate of sample, and eluent 5 mL/min, and sample volume of Cu(II) 25 mL. The standard condition of SPE is 0.5 M eluent concentration. The results show that the optimal volume of Na_2EDTA

eluent is at a volume of 25 mL because, at volumes below 25 mL, Na_2EDTA eluent has a less intense interaction between CAC sorbents, thereby reducing the EF, which impacts decreasing the percentage of desorption and SPE efficiency [10]. Therefore, the eluent volume is also an essential factor affecting the extraction and desorption efficiency in the SPE efficiency. Using large eluent volumes can increase the droplet surface area to achieve high SPE efficiency, but at some point, it can decrease due to the dilution factor of the sample analyte [30].

Concentration of Na_2EDTA Eluent

The effect of Na_2EDTA eluent concentration was studied by varying the concentration of 0.05, 0.1, 0.5, and 1.0 M, the difference in eluent concentration, affects the extraction efficiency as indicated by the desorption percentage. The greater the percentage of desorption, the more efficient the SPE process. In this study, the effect of Na_2EDTA eluent volume was studied under optimal conditions, namely pH 5, sample concentration of Cu(II) 4 mg/L, the flow rate of sample and eluent 5 mL/min, and volume of sample Cu(II) and eluent Na_2EDTA 25 mL.

The results showed that the eluent concentration of Na_2EDTA was 0.1, 0.5, and 1.0 M and had desorption percentages above 90% with optimal conditions. The interaction of metal ions with Na_2EDTA is relatively high because Na_2EDTA supplies binding sites for Cu(II) ions, thereby increasing the desorption and efficiency of SPE at 0.05 M of Na_2EDTA eluent concentration [31]. However, the desorption power was weaker than the higher eluent concentration due to the decreased interaction of Na_2EDTA with CAC sorbents, so the desorption process is less than optimal. These results also follow the previous research, which used EDTA salt as a desorption eluent to obtain optimal efficiency values [26-27]. Therefore, increasing the concentration of EDTA can increase the yield of analyte desorption on Cu(II) metal.

Effectiveness of SPE on Water Samples

Based on the results of the SPE parameter tests that have been carried out, various optimal parameters can be taken, including pH 5, sample and eluent flow rate of

5 mL/min, a sample volume of 25 mL, and Na₂EDTA concentration of 1 M. The SPE parameters were applied to tap water samples from PDAM; the results can be seen in Table 1.

The recovery test was carried out on tap water samples to prove the SPE method's accuracy. The character of tap water is the maximum allowed Cu(II) 1 mg/L. Other metals are iron and manganese in micro quantities. The results of the percentage recovery are shown in Table 1. The recovery value obtained is 80.9 to 83.6%, which indicates the SPE method's ability to quantify Cu(II) metal in PDAM water samples to be accurate and meet the recovery acceptance criteria. Acceptance of recovery is based on an analyte concentration of less than 1 mg/L, so the acceptance criteria for recovery accuracy are 80–110% [32]. Recovery is a measure that shows the degree of closeness of the analysis results to the actual analyte content. Therefore, a recovery test must be conducted to determine whether the SPE method with CAC sorbent has a high degree of closeness of results [33].

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To develop the SPE pre-concentration method, desorption of Cu(II) analyte from clean water samples is crucial to obtain maximum analyte concentration with low eluent usage. Therefore, using the optimal eluent volume is an optimization step that must be tested to achieve the highest efficiency in the SPE process. The results obtained using different volumes of Na₂EDTA from 5 to 25 mL with an initial sample volume of 25 mL can be seen in Fig. 4. These data indicate the absorbance value decrease with an increase in the volume of the eluent, which may be caused by the dilution of Cu(II) ions [18].

The 5 mL eluent volume has a high AAS absorbance value, while the 25 mL eluent volume produces a lower AAS absorbance value. Although a lower volume of desorption reagent is better, when the absorbance value is quantified and equivalent to the initial state, it turns out that the eluent with a volume of 25 mL gives a higher analyte concentration than other volumes. Even though the difference in concentration is
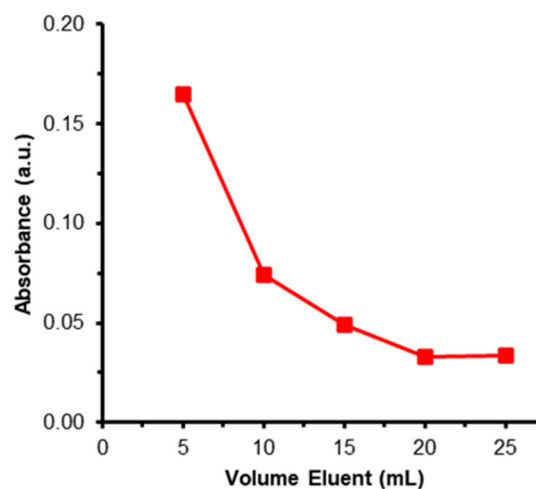

Fig 4. Effect of eluent volume on Cu(II) absorbance value in clean water sample

Table 1. Effect of Na₂EDTA eluent volume on clean water samples on percentage (%) recovery

Volume Na₂EDTA (mL)	Sample of Cu(II)			
	Ca (mg/L)	Ads(%)	Des(%)	Rec(%)
5	0.51	91.4	92.7	80.9
10	0.51	90.6	93.6	81.0
15	0.51	91.4	93.5	81.9
20	0.51	91.1	93.4	81.4
25	0.51	91.1	95.3	83.6

Table 2. Effect of enrichment factors on Cu(II) samples in bottled drinking water samples

Volume of Na₂EDTA (mL)	Cu(II) Sample			
	Ca (mg/L)	C ads (mg/L)	Cef (mg/L)	C des (mg/L)
5	-	-	0.10	0.020
10	-	-	0.04	0.017
15	-	-	-	-
20	-	-	-	-
25	-	-	-	-

not too significant, at a volume of 25 mL, the contact of the Na₂EDTA eluent with the CAC sorbent, which had absorbed Cu(II), was longer than the eluent volume, which was less than 25 mL [34].

Table 2 shows the results of the analysis of Cu(II) samples in bottled drinking water. When the 25 mL drinking water sample was analyzed without the SPE method, it did not provide an AAS absorbance value for Cu(II) metal. Cu(II) metal is not detected in bottled drinking water because the absorbance value may be close to or below the limit of quantification (LoQ) value [35]. For this reason, the samples were analyzed using the SPE method by preconcentrating 5 and 2.5 times. Pre-concentration of 5 times succeeded in quantifying Cu(II) metal of 0.10 mg/L. For 2.5 times pre-concentration, the Cu(II) metal concentration in drinking water samples was 0.04 mg/L. If these concentrations are equivalent to the initial conditions, then at concentrations of 5 and 2.5, the actual concentrations shall be 0.020 and 0.017 mg/L, respectively. Then the two results were averaged, and the concentration of bottled drinking water samples was 0.0185 mg/L. The concentration value obtained is smaller than the LoQ value, which is 0.019 mg/L, so the SPE method is not quantified when drinking water samples are analyzed without a concentration process. The use of a concentration EF is an advantage of the SPE method with CAC sorbents for Cu(II) metal concentrations that were initially undetectable to be quantified by increasing the absorbance value for Cu(II) [36].

CONCLUSION

Synthesis of alginate and carbon-modified chitosan sorbent with a ratio of chitosan:alginate:carbon is 1:1:1 to produce CAC sorbent, which is electrostatically bound and has an active side carboxyl group that acts as an adsorbent of Cu(II) metal. Comparison of chitosan:alginate:carbon is 1:1:1 obtained from optimization results. Optimal test parameters of SPE with CAC sorbent as Cu(II) metal analysis method, namely pH 5, Cu(II) concentration of 4 mg/L, flow-rate of Cu(II) sample and Na₂EDTA eluent 5 mL/min, sample volume Cu(II) and 25 mL Na₂EDTA eluent and 1.0 M Na₂EDTA eluent concentration. The effectiveness of SPE as a Cu(II) metal analysis method with CAC sorbent on clean water samples is excellent, with the results of the percentage recovery accuracy ranging from 80.9 to 83.6%, which is by the criteria for acceptance of method validation. In addition, increasing the pre-concentration EF in bottled drinking water samples to 2.5 and 5 times can increase the abortion value, which was initially undetectable. The concentration of samples that can be quantified at concentrations of 5 and 2.5 times, respectively, is 0.020 and 0.017 mg/L.

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