

DETERMINATION OF Cr(VI) BY USING CHITOSAN-1,5- DIPHENYL CARBAZIDE RESIN MODIFIED AT THE PRECONCENTRATION SYSTEM WITH COLUMN METHOD

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**DETERMINATION OF Cr(VI) BY USING CHITOSAN-1,5- DIPHENYL
CARBAZIDE RESIN MODIFIED AT THE PRECONCENTRATION
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ABSTRACT

A chelating resin of chitosan-1,5-diphenylcarbazide modified with the alginate salts and utilization as a filler material column has been synthesized and the sorption properties have been investigated for preconcentration and determination of Cr(VI) at trace levels. The optimal composition of the formation of Ca-alginate-chitosan-1,5-diphenylcarbazide microcapsules were 1% Na-alginate, 0.1M CaCl₂ and 0.6 g resin. The retention capacity of resin modified was 2.2124 mg Cr (VI) /g resin at optimum pH 4-6. The analytical performance this method is good which are shown by the limit of detection was 1.14 µg/L and the reproducibility level shown by the percentage of the coefficient variance was 1.9563 %. This method was applied for Cr(VI) determination in river water samples collected from Karang Mumus and Mahakam Rivers, East Kalimantan, Indonesia.

KEYWORDS: Chelating resin, Chitosan, Diphenylcarbazide, Cr(VI), Preconcentration.



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INTRODUCTION

Chromium occurs in nature at very low concentrations predominantly in two oxidation states, which differ significantly in their toxicity. While Cr (VI) is toxic and carcinogenic upon inhalation, Cr (III) is known to be an essential nutrient for humans supplied also in pharmaceutical products as a dietary supplement. Increasing production of Cr (VI), due to oxidation of Cr (III) in industrial plants or from its spontaneous oxidation in soils, may pose pollution problems to drinking and surface waters¹. In recent years, the determination and absorption of chromium, especially Cr (VI) by using biomass is becoming very important in environmental samples². One technique of determining Cr (VI) developed in recent years is preconcentration technique based chelating resin, using a spectrophotometer detector. The important role of preconcentration technique and separation of trace elements according to the nature of the samples, the concentrations of the analytes and the measurement techniques used^{3, 4}. Preconcentration step by sorption technique not only increases the concentration of the analyte, but also can eliminate the matrix effects which may interfere with the analysis. This technique is able to provide enrichment factor up to several hundred times, separation of interfere component, high efficiency and easy to combine with a variety of detector of instrumental analysis⁵. Chelating resin basically is consisted of two components that are functional of chelating group and polymer matrix as supported. Nature of from both this components will determine usage and performance from a chelating resin. The selectivity will be determined by type chelating group, while capacities, mechanic strength and the chemistry resistance determined by supporter polymer type who applied⁶. This resin also can provide selectivity, mechanical stability and good binding and ease the process of regeneration for sorption-desorption cycle^{7,8}. Pure chitosan is generally used as an absorbent for heavy metals in flakes and powder form. Adsorption ability of chitosan against heavy metals is strongly influenced by physical-chemical properties of chitosan. Chitosan which not cross-linked have an adsorption capacity is greater than

the cross-linked chitosan, but the cross-linked chitosan have the physical endurance to acid better than not cross-linked chitosan⁹. Chelating resin of chitosan-1.5-diphenylcarbazine can be synthesized by the diazo coupling reaction and can be modified to form microcapsules Ca-alginate-chitosan-1.5-diphenylcarbazine by reaction between resins with Na-alginate in CaCl₂ solution^{9,10}. Retention characteristics of Ca-alginate-chitosan-1.5-diphenylcarbazine resin toward Cr(VI) ions is good, shown that the resin can be used as a filler material column in the preconcentration step of Cr(VI) ions.

MATERIALS AND METHODS

Instrumentation

Equipment used in this research are a set of reflux system, three-neck flask, glass columns (1 cm i.d x 30 cm length), manifold equipment for preconcentration stages. Other equipment such as analytical balance, the volume pipettes, stopwatch, oven, hot plate with stirrer, and Spectrophotometer FT-IR (Fourier Transform - Infra Red) Prestige 21-D, Spectronic UV-Vis (Bachman DU 7500), atomic absorption spectrophotometer (GBC-Avanta 6506), was used for all measurements.

Reagents

The materials used in this research were; potassium bichromate, chitosan-1.5-diphenylcarbazine resin synthesis, sodium hydroxide, hydrochloric acid, sulfuric acid, nitrate acid, calcium chloride, sodium alginate, buffer pH 4, 7 and 10; all reagents were of analytical-reagent grade (E. Merck), and aquabidest.

Procedure of Chitosan-1,5-diphenyl carbazine Resin

2 g of chitosan added to 100 mL 1M HCl alternating with addition of 75 mL 1M NaNO₂ until resin was azotated in the presence of iodine paper color change, by keeping the temperature between 1-3 °C. Then add dropwise 100 mL of 10% 1,5-diphenylcarbazine and stirred for 1 h with

temperature between 1-3 °C and left for 24 h in the refrigerator. The mixture was then filtered and rinsed with distilled water until neutral pH and dried in oven at 60 °C¹⁰.

Preparation of Ca-Alginate-Chitosan-1,5-diphenylcarbazine Microcapsules Resin

25 mL CaCl₂ solution prepared with various concentration of 0.1 - 1 M into each 100 mL glass beaker, then added 0.1 - 1.0 g Chitosan-1,5-diphenylcarbazine resin synthesis and stir until homogeneous with magnetic stirrer. 1% Na-alginate was added dropwise with a burette while stirring with a magnetic stirrer on to form of Ca-alginate-chitosan-1,5-diphenylcarbazine resin, adjust the flow rate in

the buret and the rotational speed of the magnetic stirrer and Ca-alginate-chitosan-1,5-diphenylcarbazine microencapsulated granules dried at room temperature for ± 24 hours.

Preparation and Conditioning of the Column for Preconcentration Technique

2 g Ca – alginate – chitosan -1.5 – diphenyl carbazine chelating resin suspended in aquabidest was slurry packed into column (1 cm i.d x 60 cm length). Before the use, the column was precondition with aquabidest adjusted to pH 6. The Prototype module of the preconcentration system based on column is shown in Figure 1.

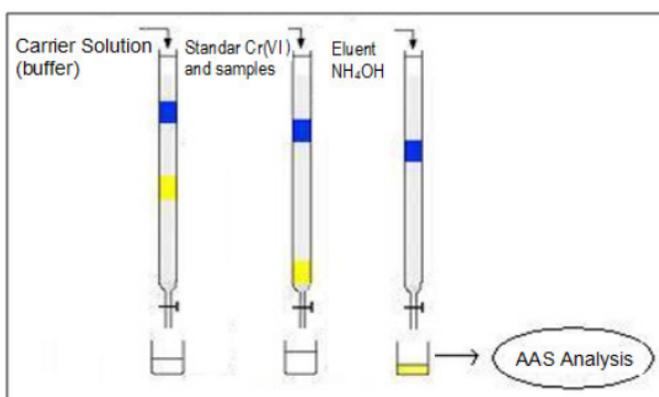


Figure 1
Prototype module design preconcentration technique based on column

Optimization of the Preconcentration Techniques Based Column Method

The preconcentration steps⁸ is done by dynamic techniques to know the effect of pH on the retention of Cr(VI) ions and capacity retention. A number of the Cr(VI) standard at different pH was passed into the column containing chelating resin. The amount of ions retained calculated from measurements of the concentration by using atomic absorption spectrophotometer. The retention capacity was determined at the pH optimum retention by using varying concentrations of Cr(VI) ions. The influence of the sample volume, type and volume of NH₄OH eluent, performed by using with the same procedures, as well as the above protocol.

Evaluation of analytical performance preconcentration technique developed

Based on the optimum conditions were obtained, the analytical performance of preconcentration technique by column method has been studied. Important parameters are specified

- a. Determination of the linearity of Cr(VI) in order to obtain the calibration curve.
- b. Determination of the reproducibility of Cr(VI). Measurements were performed repeatedly with the Cr(VI) standard solution with the same concentration. Reproducibility showed with % CV (Coefficient of Variance).
- c. Determination of the limit detection (LOD). The LOD showed as a comparison standard signal (S) to the blank signal (N) or S/N = 3.

d. Determination of the enrichment factor (EF), by comparing measurements of the same concentrations standar Cr(VI) between direct measurement method with preconcentration techniques developed.

Determination of samples

To determine the influence of matrix on the determination of Cr(VI) ions in water samples from nature, the spike method was done. In this method a certain volume of Cr(VI) was pipetted and diluted with the samples. The treatment is then performed with the optimum conditions and the absorbance was measured using a preconcentration technique based on column-AAS were developed.

group into a diazo group using concentrated acid solution¹⁰. In this process chitosan reacted with 1M HCl and 1M NaNO₂ alternately at 1-3°C until the resin azotated. Reaction temperature was maintained at below 3°C because the reaction is highly exothermic. Hydrochloric acid is a strong acid which serves as a catalyst and forming chitosan chloride salt. NaNO₂ serves as forming nitrosonium ion together with HCl to produce diazonium salt. The mixture then reacted with 1,5- diphenyl carbazide 5 % dropwise and stirred for ± 1 hours at 1-3 °C. Diazonium salt is stable at low temperature (0-4 °C) and sensitive to light and can easily be damaged at ultraviolet wavelength and visible light¹⁰. Diazonium salt has coupling reaction with 1,5- diphenylcarbazine to produce chitosan- 1,5- diphenylcarbazine compound (Figure 2). The obtained Chitosan-1,5-diphenylcarbazine resin after diazotation is reddish brown with a yield of 39.43%.

RESULTS AND DISCUSSION

Synthesis of Chitosan-1,5-Diphenylcarbazine Resin

Synthesis of chitosan-1,5-diphenylcarbazine resin is done through the diazotation reaction. Diazotation is a way to change the amine

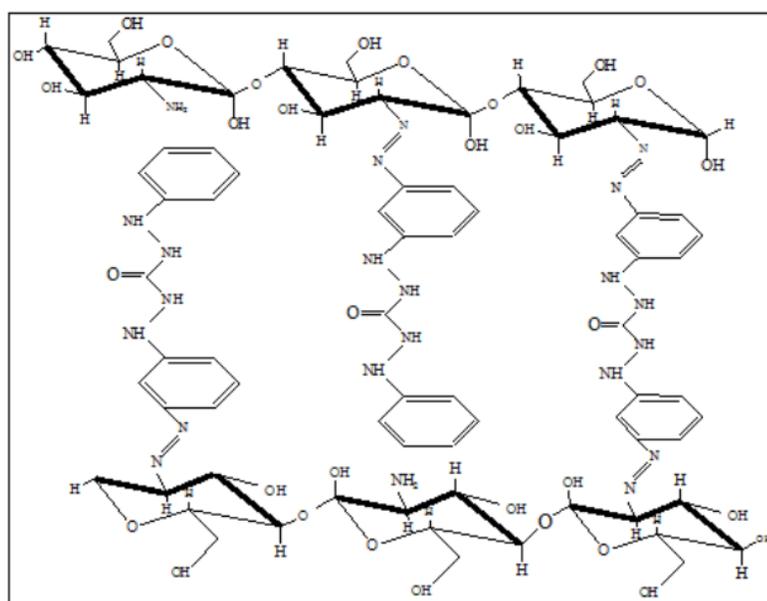


Figure 2
Structure of Chitosan-1,5-Diphenylcarbazine

Microcapsulation of Ca-alginate-chitosan-1.5-diphenylcarbazine resin

At this stage of the study varied between CaCl₂ and the Na-alginate concentration to obtain the best composition of the Ca-alginate

to absorb Cr(VI) ions. Chitosan-1.5-diphenylcarbazine resin synthesis can be modified to form microcapsules of Ca-alginate-chitosan-1.5-diphenylcarbazine by reaction between Na-alginate solutions with

resin synthesis in CaCl_2 solution. The optimum conditions were obtained in the composition of 0.1 M CaCl_2 , 1% Na-alginate and 0.6 g resin synthesized. Chelate complex formation between chitosan Ca-alginate-chitosan-1,5-diphenyl carbazide resin with Cr(VI) ions occurs because the donation of a lone pair of the amine group and the oxygen atoms from the resin compounds¹¹. The bond was formed between the microcapsules of resin with Cr(VI) ions is a Van der Waals bonds. The metal ions bounded through the formation of a weak bond with the O group on the electron pair on alginate compounds¹². The weak bonding that occurs between Cr(VI) and Ca-alginate-chitosan-1,5-diphenyl

carbazide is used as a sorption-desorption technique in the preconcentration stages in this research.

Analysis Functionalized group of Ca-alginate-chitosan-1,5-diphenylcarbazide resin

Ca-alginate-chitosan-1,5-diphenylcarbazide microcapsule were analyzed by FT-IR spectroscopy. Spectrum analysis of Ca-alginate-chitosan-1,5-diphenyl carbazide microcapsule before interacted with Cr(VI) compared to the spectrum of Ca-alginate-chitosan-1,5-diphenylcarbazide microcapsule that after interacted with Cr(VI) can be seen in Figure 3.

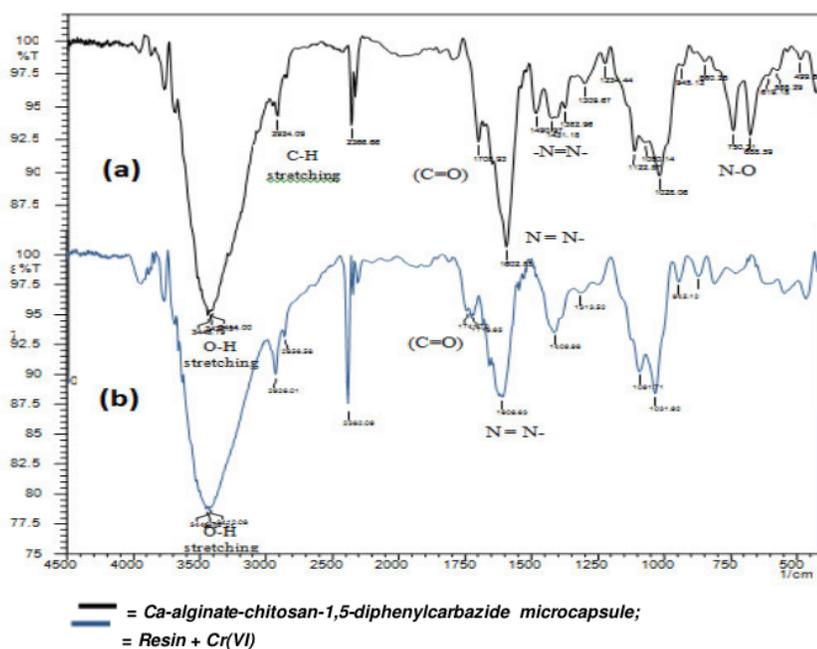


Figure 3
FT-IR spectrum of (a) Ca-alginate-chitosan-1,5-diphenylcarbazide microcapsule; (b) resin + Cr(VI)

FT-IR spectrum in Figure 3(a) shows a broad absorption peaks at wave number 3446.79; 3427.51 and 3414.00 cm^{-1} indicate typical absorption peak of O-H stretching vibration. Absorption peaks at wave number 2924.09 cm^{-1} is the CH stretching vibration of alkanes. Absorption peaks at wave number 1708.93 cm^{-1} indicate typical absorption of stretching vibrations of carbonyl group (C=O) where is possibility of amide groups at 1.5 DPC and

carboxylic acid salt of alginic. Absorption peaks at 1062.85 and 1490.97 cm^{-1} is typical of the stretching vibration peak symmetry and asymmetry -N=N- groups that is likely to come from diazo bond between chitosan and 1,5-diphenylcarbazide or 1,5-diphenylcarbazone. Absorption peaks at wave number 1028.06; 1080.14 and 1122.57 cm^{-1} showed C-O stretching vibration from the compound chitosan and alginate. Absorption peaks at

wave number 750.31 cm^{-1} , is typical absorption of stretching vibration of N-O. FT-IR spectrum Ca-alginate-chitosan-1,5-diphenylcarbazide icrocapsule has interacted with Cr(VI) (Figure 3.b), shows that most of the wave number has changed absorption intensity. It can be seen at 1741.72 and 1716.65 cm^{-1} absorption peaks showed reduced uptake in group C=O and C-O followed by increased uptake in the wave number 1031.92 cm^{-1} . This is expected because there has been a change in the structure of 1,5-diphenylcarbazide to 1,5-diphenylcarbazone binds to Cr(VI). Intensity

decrease also occurred in the -N=N- stretching vibration in wave numbers 1608.63 cm^{-1} and also the NH- bending vibration at wavenumber 727.16 cm^{-1} , this may occur due to the interaction between the 1,5-diphenylcarbazone complexing with Cr(VI).

Optimization of Preconcentration Technique

Influence of pH on Cr(VI) retention

pH influence at Cr(VI) of retention done with batch method. Figure 3 showing pH influence Ca-alginate-chitosan-1,5-diphenyl carbazide resin to retention of Cr(VI) ion.

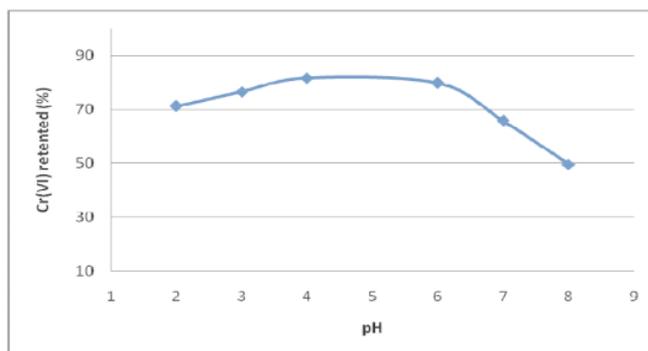


Figure 4
Effect of pH toward Ca-alginate-chitosan-1,5-diphenylcarbazide resin

The result of research showed, pH 4-6 gave good results compared to measurements at other pH. This is due to the formation of pH 4-6, stability of the complex between Ca-alginate-chitosan-1,5-diphenylcarbazide with Cr(VI) ions maximal and the metal ions can be absorbed optimally. Whereas at $\text{pH} > 7$, the absorption of Cr(VI) by resin decreased because in the alkaline solution formed hydroxide compound $\text{Cr}(\text{OH})_3$ and can't be

detected by atomic absorption spectrophotometer.

Retention Capacity of Ca-alginate-chitosan-1,5-diphenylcarbazide

Retention capacity of resin is measure of the ability of Ca-alginate-chitosan-1,5-diphenylcarbazide to retain Cr(VI). Determination of retention capacity of Cr(VI) to chelating resin synthesis done at optimum pH with contact time during 30 minutes.

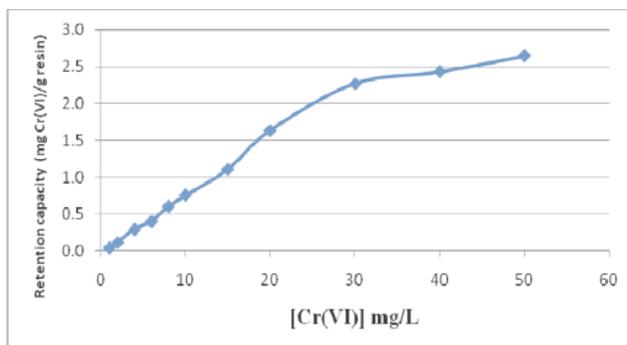


Figure 5
Retention capacity of Ca-alginate-chitosan-1.5-diphenylcarbazine resin

The result showed, retention capacity from synthesized chelating resin are 2.2124 mg Cr(VI)/g resin, means that for every 1 g of Ca-alginate-chitosan-1.5-diphenylcarbazine resin can adsorb optimally 2.2124 mg Cr(VI) ions. This retention capacities value shows the chelating resin can be applied as filler material of column to preconcentration technique for determination of trace Cr(VI) in samples.

¹²
Effect of the concentration and volume of the eluent

NH₄OH useful as eluent to release the analyte bound to the resin. The concentration and volume of eluent must be optimized because it affects the performance of the analytical performance of measurement and resins that exist in the column can be used repeatedly¹³. Figure 6 shows, in this research obtained concentration of NH₄OH > 0.5 M, the ions of Cr(VI) were eluted did not differ significantly with greater concentration. To make sure all the Cr(VI) ions was eluted in the 1M NH₄OH eluent used for further work.

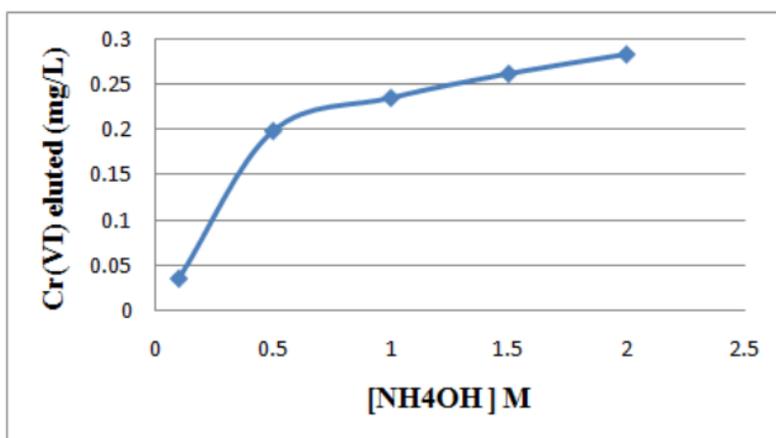


Figure 6
Effect of eluent NH₄OH concentration

²
The influence of the eluent volume in the step of Cr(VI) desorption from the column was also investigated. It can be concluded with the addition of 4 mL volume of eluent were already maximally elute all Cr(VI) ion retained by the resin in the column.

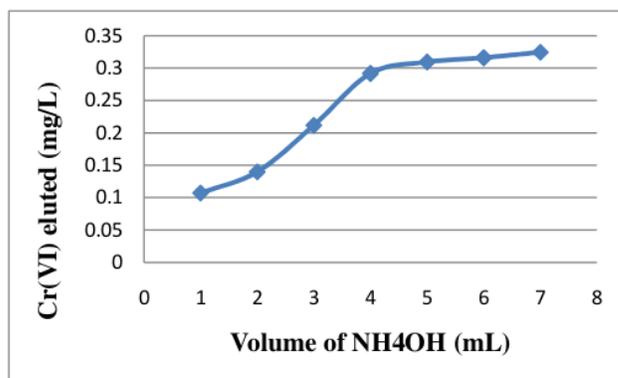


Figure 7
Effect of eluent volume

Effect of the Cr(VI) volume

Total volume of the Cr(VI) ions need to determine of the optimum volume of samples were able to captured by the resin. The concentration of Cr(VI) ion measurement of

0.1 mg/L by using a variation of the volume of standard from 1 mL to 10 mL and the concentration of 1M NH₄OH with the volume of 5 ml. The eluted of samples measured by atomic absorption spectrophotometer.

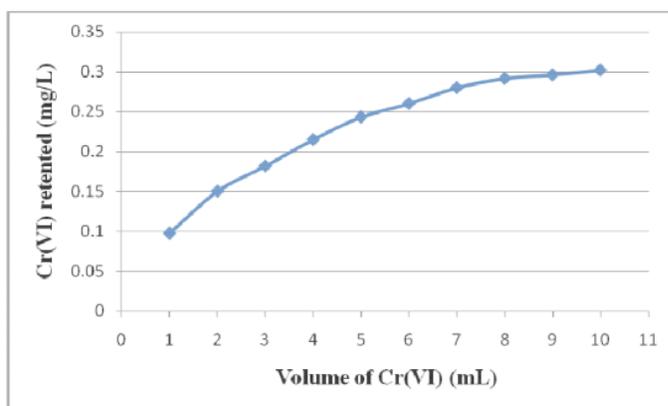


Figure 8
Effect of Cr(VI) volume

The amount of Cr(VI) ion eluted with NH₄OH increases with increasing sample volume. The addition of 9 - 10 mL volume standard, the amount of Cr(VI) did not differ significantly eluted with a standard volume of 8 mL. This is due to the number of ions of Cr (VI) was added too much, the amount of the resin contained in the column can't to retain all the Cr(VI). Furthermore, the volume of Cr(VI) obtained optimum at 8 mL used for further work.

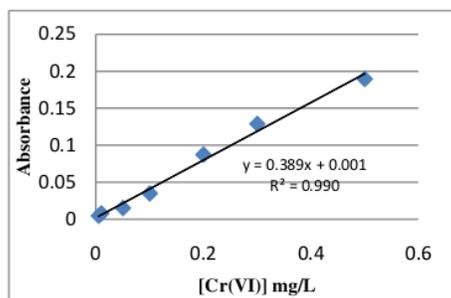
11 Analytical Performance of the preconcentration system

Reproducibility

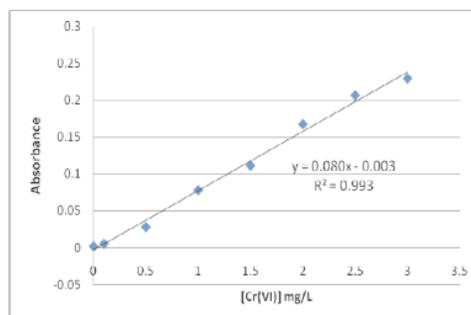
The reproducibility of the method was examined under the optimum experimental conditions described above by using the model solution. Reproducibility level shown as percentage of the coefficient of variance (%CV) value obtained from the seven measurement standard samples at the same condition. The result of measurement shows for the samples standard 50 µg/L Cr(VI), %CV value was 1.9563 %.

Linearity

A calibration curve was made to search the linearity of measurement area between the concentrations of the analyte in the sample with the resulting of regression linear. Calibration curve was obtained by plotting concentration of Cr(VI) standard series with absorbance value. Based on the measurement results were obtained by the linear range before preconcentration (Figure 9.a) at a concentration of 0.1 – 3.0 mg/L.



(a)



(b)

Figure 9

Calibration curve of Cr(VI) (a) before; (b) after preconcentration

Limit of Detection (LOD)

Detection limit is the concentration or mass minimum analyte that was detected with a high level of confidence. Detection limit is expressed as the ratio between the standard signal (S) of the blank signal (N) or $S/N = 3$. Based on the result of measurements carried out for preconcentration of Cr(VI) ion by using Ca-alginate-chitosan-1.5-diphenyl carbazide resin, the detection limits are 1.14 $\mu\text{g/L}$. The LOD values obtained before preconcentration are 0.053 mg/L (53.0 $\mu\text{g/L}$). Preconcentration method developed very well when compared with the direct measurement by using atomic absorption spectrophotometer and can improve the results of measurements in the range of $\mu\text{g/L}$. The detection limit of the measurement preconcentration method developed increased 46 times from the determination the Cr(VI) directly by AAS.

Enrichment Factor (EF)

Enrichment factor, or often called the sensitivity of increase is done by comparing the absorbance of the ion Cr(VI) measured before and after preconcentration done. The

Equation of the regression linear is $y = 0.080x - 0.003$ with correlation coefficient values (R) are good that is equal to 0.993. The calibration curve obtained for Cr(VI) ion after passed into the column containing Ca-alginate-chitosan-1.5-diphenyl carbazide resin, the linear range at a concentration of 0.05 – 0.5 mg/L (Figure 9.b). Equation of the regression linear is $y = 0.389x + 0.001$ with correlation coefficient values (R) are good that is equal to 0.990.

determination of EF value is very important because it could illustrate the succes of the developed method. The greater EF value, the better of the developed method because can be used to determine samples at the trace levels. Based on the results of this research, the EF value is 6.82 times. This Mean that the developed method can increase the absorbance value 7 times than before preconcentration. The increase shows the Cr(VI) ion at trace concentrations can be detected using this method.

5

Determination of Cr(VI) in Water Samples

The proposed preconcentration method had been applied to water samples from the nature for determination of Cr(VI). In this research, to determine the % recovery was conducted by the spike method. Water samples and blanks were added with a certain amount of standard solution of the analyte. The concentration of analyte determined in the water samples and the blank, and compared in order to obtain % recovery.

Table 1
Result of determination of Cr(VI) ions in water samples

Samples	Cr(VI), $\mu\text{g L}^{-1}$		
	Added	founded	% Recovery
Mahakam river	0	37.48±4.27	-
	50	88.77±3.71	98.55 ± 0.74
Karang Mumus river	0	36.44±13.72	-
	50	89.62±17.65	96.45 ± 0.56
Water dam	0	92.44±13.72	-
	100	194.62±17.65	98.87 ± 0.92

The result, which are shown in Table 1, have been calculated on the assumption of >95 % recovery of Cr(VI) ions, indicate that proposed method can be used for Cr(VI) determination in water samples.

CONCLUSION

Based on these results found in the present study, it can be concluded that using Calcium alginate-chitosan-1.5-diphenyl carbamide resin is an effective material for the preconcentration of Cr(VI) ions in the river water samples. The combination of column method systems with FAAS using polymeric resins functionalized with complexing reagents can be a promising way in the preconcentration and determination of Cr (VI) in a wide range of samples. The optimum of results using of this research are the retention of capacity was 2.2124 mg Cr(VI)/g resin. The

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best sorption at pH 4-6, the eluat was 1M NH_4OH with the volume of sample was 8 mL and the volume of eluat was 4 mL. The analytical performance this method is good which are shown by the limit of detection was 1.14 $\mu\text{g/L}$, the reproducibility level shown by the percentage of the coefficient variance was 1.9563 %, and the enrichment factor of resin was 6,82 times. The technique of preconcentration developed can be applied to determine the concentration of Cr(VI) in water samples, shown by the percentage recovery are >95%.

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