Using QuEChERS sample preparation technique followed by ICP-Ms method to determination Cr(III) and Cr(VI) in seawater

Le Thi Thuy Nguyen[1*], Dinh Hong Thang[2], Vu Thi Hao[1],
Nguyen Khac Manh[1], Tran Tuan Viet[1]

[1]Institute for Tropical Technology and Environmental Protection (VITTEP);
[2]Viet Nam University Ho Chi Minh City – University of Science (HCMUS).
[*]Corresponding author: lethithuynguyen89@gmail.com

Received: 30 Aug 2022; Revised 7 Nov 2022; Accepted 28 Nov 2022; Published 20 Dec 2022.
DOI: https://doi.org/10.54939/1859-1043.j.mst.FEE.2022.266-271

ABSTRACT

Currently, Cr(III) and Cr(VI) are analyzed directly at trace level using anion-exchange liquid chromatography inductively coupled plasma mass spectrometry (LC – ICP – MS), which is well-known as an expensive and complicated method. Therefore, the determination of Cr(VI) and Cr(III) by spectrometer or combined with ICP (or AAS) is seemly preferred to choose in most laboratories, although this method presents a higher limit of detection (LOD) and is influenced by salt ions in seawater. In this study, after forming a reddish-violet complex by adding diphenylcarbazide (DPC) to seawater samples, a QuEChERS extraction technique was applied to separate Cr from the high salt level matrix and transfer the Cr(VI)-DPC structure to the acetonitrile phase. In the case of determination of total Cr, the Cr(III) in samples had to convert to Cr(VI) by oxidate with $KMnO_4$ before adding DPC. Then, a digestion process with HNO_3 – H_2O_2 mixture was implemented before analyzing Cr(VI) by ICP-MS. The Cr(III) is calculated indirectly based on total Cr and Cr(VI) results. This method had a linear concentration range of Cr(VI) from 0.02 to 50 µg/L with a detection limit of 0.015µg/L, the relative standard deviations under 3,03%, and the recovery was 92-108 % for seawater. According to the outcome of the parallel experiment with the common method, which used a spectrometer and did not apply QuEChERS extraction step, the ICP-MS method presented a better result with a smaller LOD, RSD, and recovery range.

Keywords: Cr(VI); Cr(III); QuEChERS; ICP-MS; Sea water.

1. INTRODUCTION

Chromium metal (Cr) occurs naturally in the environment in the oxidation states, with Cr(III) and Cr(VI) being the oxidation states that exist mainly in the environment because the redox conditions and pH determine the designation of chromium in the aquatic environment [6, 7]. Trivalent chromium Cr(III) is an essential nutrient for the body while Cr(VI) is one of the most toxic, and it is many times more toxic when compared to Cr(III) [9]. When Cr(VI) concentration exceeds the permissible level, it can cause skin rashes, kidney and liver damage, internal hemorrhage, teeth abnormalities, respiratory ailment, cancer, and other diseases [8, 10]. Cr(VI) is a base compound used in various chemical industries, including leather, to produce tannery, wood treatment, pigment products, and corrosion inhibitors.

In recent years, when quantifying Cr in solution, most researchers have used a color comparison method with 1,5-diphenylcarbazide (DPC) complex, which in acidic conditions forms reddish-violet species. For photometry methods, the coloration of dark colors directly affects the sensitivity of the method. The DPC method was chosen because of the intense color development at the 540 nm region. The Standard method for the determination of Cr(VI) [18-20].

In another study, based on the use of ICP-MS and instrumental neutron activation analysis (INAA) together with a Chelex 100 resin column studied Cr(III) and Cr(VI) in surface waters for concentrations of Cr(III) and Cr(VI) was found in the range of 0.24 - 52.28 µg/L and 2.24 -

11.13 µg/L [25]. Although this method has a higher detection limit (LOD) but is affected by salt ions in seawater.

Purpose of this study, Cr(VI) is determined by: After forming reddish-violet complexing by adding 1,5-diphenylcarbazide (DPC) to seawater samples, a QuEChERS extraction technique was applied to separate Cr from the salt level matrix and transfer the Cr(VI) - DPC structure to the acetonitrile phase and the Cr(VI) - ICP-MS analyzed DPC structure. This combination of methods was chosen because of its more straightforward, cheaper process, possible removal of salt ions from seawater, and sample clean-up step in the QuEChERS extraction technique. The results obtained after analysis by ICP-MS are higher sensitive and more reliable.

2. METHODS AND MATERIALS

2.1. Chemicals

Potassium chromate (K_2CrO_4 - 99.5%), Chromium chloride ($CrCl_3$) 1000mg/amp, magnesium chloride ($MgCl_2$ - 99%), sodium sulfate (Na_2SO_4 - 99%), nitric acid (HNO_3 - 99%), sulfuric acid (H_2SO_4 - 99%), orthophosphoric acid (H_3PO_4 - 99%), hydroperoxide (H_2O_2 - 99%). All were purchased from Merch (Darmstadt, Germany). 1,5-diphenylcarbazide (Kanto - Japan - 98%).

Acetonitrile (CH_3CN) were purchased from Schalaur (Spain). Deionized water purified by Thermo Scientific (USA) was used to prepare solutions of standards and samples.

2.2. Instrument

A Shimadzu 1800 UV-Visible spectrophotometer with Hellma 50 mm quartz cuvettes for the absorbance measurements. The Agilent 7700 series (USA) is coupled with induced plasma with mass spectrometry (ICP-MS). The setting parameters for the ICP-MS analysis are studied by the team and presented in table 1.

Table 1. *Parameters of ICP-MS setting.*

ICP-MS parameter	Value
Forward power	1550 W
Plasma gas	15 L/min
Kinetic energy discrimination (KED)	3 V
Helium collision gas	4.5 mL/min
Integration time/point	0.2 s
Replicates	3

2.3. Color development

Seawater was collected at Cangio District into 5L glass bottles, chilled, and transported to a field laboratory according to standards [27]. All samples were filtrated through a glass fiber membrane and stored in the dark in a refrigerator. To determine Cr(VI), mix a 50 mL seawater sample into a conical flash of 100 mL, and add 250 µL of H_3PO_4 concentrated and 100 µL of H_2SO_4 concentrated. Add 1.0 mL of 1,5-diphenylcarbazide solution (1% - w/v), mixture and let stand 10 min for full color development. Transfer to a 50 mm absorption cell and measure its absorbance at 540 nm. To determine total Cr, the Cr(III) in the samples had to convert to Cr(VI) by oxidating with 0.5mL of $KMnO_4$ 0.01M solution, warm in bath water for 40 min at 60^0C. Cool, add a few drops NaN_3 4% w/v solution, and warm again at 60^0C for 3 min. It was cooling before color development the same way above. The Cr(III) is calculated indirectly based on total Cr and Cr(VI) results.

2.4. Extraction methods

Sea water containing high salt content is not measured directly by ICP-MS because of the high background signal and block holes that clog the system. The chelate-resin column was used to retain metals and salt in the sample to resolve these problems. The high cost of chelate-resin materials commercials is a significant obstacle when applied in laboratories. The spectrometric method uses some standard methods widely for measuring Cr(VI). The steps in these standards method were set up in this study and did not require more research. We focus on techniques such as solid phase extraction and liquid-liquid extraction to transfer the Cr(VI)-DPC complex out of the water matrix. These techniques have some advanced flexibility and are cheaper and easier to work. Some materials sorbent, such as C18, and HLB, were used to load Cr(VI)-DPC complex solution, but the reddish-violet complex moved throughout the cartridge observed and did not show the results.

Figure 1. *Liquid-liquid extraction to extract Cr(VI)-DPC complex by (1) acetonitrile - above layer, (2) n-butanol - above layer, (3) ethylacetate above layer, (4) chloroform – bottom layer, and (5) dichloromethane - bottom layer at Cr(VI) 10 ng/mL.*

In LLE, solvents dichloromethane, chloroform, ethylacetate, n-butanol, and acetonitrile were used to extract Cr(VI)-DPC complex. In Fig.1, we can see the reddish-violet move to the polar solvents such as acetonitrile (Fig.1(1)) and n-butanol (Fig.1(2)) phase more entirely compared to other solvents. Spectrometry scanned these extracts from 420 - 650 nm and has the same absorbance at maximum peak, about 0.33 Abs. n-butanol evaporates more slowly than acetonitrile in the solvent drying step prior to ICP-MS measurement, so acetonitrile was the best choice in this experiment.

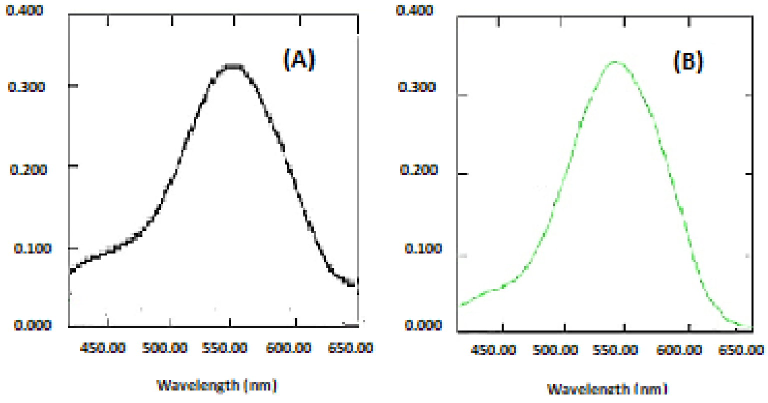

Figure 2. *Spectrometry scanned these extracts from 420-650nm (A) Acetonitrile, and (B) n- Butanolat Cr(VI) 10 ng/mL.*

2.5. QuEChERS Extraction

Take 5 mL of Cr(VI)-DPC complex solution to a 15 mL centrifuge tube. Add 5 ml ACN, 1 g of $MgCl_2$, and 1 g Na_2SO_4, shaking for 30 min, and centrifuge for 5 min at 5000 rpm. After centrifugation, collection of the ACN phase into tube 15 mL and blown to dryness by N_2 gas flow. Then, 0.2 mL of HNO_3 – H_2O_2 (1:1, v/v) mixture was added, tightened the cap, and put it on the COD reactor set to 150°C for 30 min or until colorless. Finally, fill up to 10 mL with Deion water for ICP-MS analysis.

2.6. ICP-MS method validation

Eight-point calibrations, over the concentration range 0.02 - 50 µg/L, were made $R^2 > 0.99$. The method's accuracy and trueness were performed in 7 replicate sub-sample of seawater containing Cr(VI) at 10 µg/L and assessed through the percent of relative standard deviation (% RSD) and percent of recovery efficiency (% RE) value, which was made %RSD < 30% and %RE in the range 60 - 120%, respectively. The limit of detection (LOD) was performed in 7 replicates sub-sample seawater containing Cr(VI) at 0.5 µg/L. The formula calculates the LOD values:

$$\mathrm{LOD} = t_{0.99,\, f=7} * \mathrm{SD}$$

Where SD is the standard deviation of the ratio of the analyte signal to the IS signal and $t_{0.99,\, f=7}$ is the Student's *t*-value appropriate for the single-tailed 99 percentile *t*-statistic and an SD estimate with $n - 1$ degree of freedom [26].

3. RESULTS AND DISCUSSION

3.1. Extraction method

As can be seen, the two solvents acetonitrile and n-butanol for the best extraction performance are shown in figure 1. Based on the properties of the two solvents, we can see that acetonitrile has a mild odor that is not as dark as n-butanol, the small boiling point is convenient for faster drying and especially the cost of acetonitrile is cheaper. Therefore, acetonitrile solvents are suitable for further service for this study.

3.2. QuEChERS Extraction and Digestion

QuEChERS extraction: To enhance the ability of the Cr(VI)-DPC complex to move to the acetonitrile phase, time extract was experimental. In Fig.3, the efficient recovery of Cr(VI) increased in the 5 to 30 min ranges and decreased in the 30 to 60 min ranges. Chromium investigations at the ppb level and prolonged time can degrade the Cr(VI)-DPC complex and make the ICP-MS signal unstable. Due to measurement, 30 min is the optimal time for this survey step and was used for the subsequent experiments.

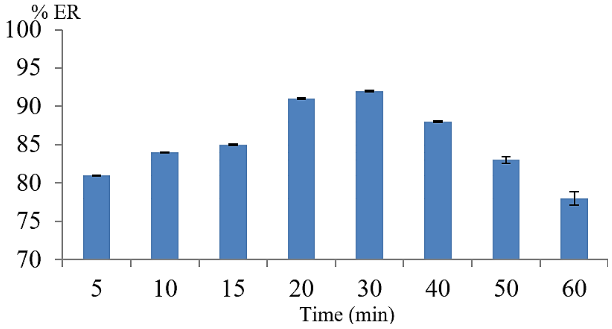

Figure 3. *Time extract in Optimal QuEChERS Extractionat Cr (VI) 10 ng/mL.*

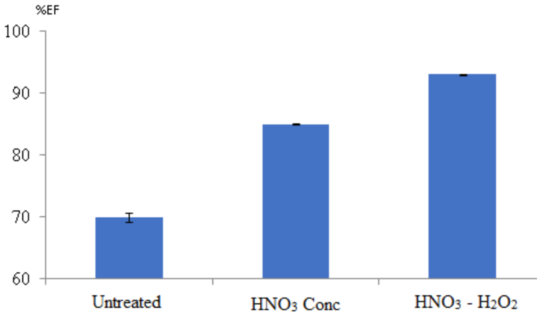

Figure 4. *Parameters of the digestion systems.*

Digestion: The extracts were blown by N_2 gas flow to dryness. Next, the residue was dissolved in 10 mL HNO_3 1% for ICP-MS analysis. Furthermore, the residue was digested with HNO_3 concentrate and a mix of HNO_3 - H_2O_2 (1:1 v/v). The efficiency recovery of Cr(VI) at 10 ng/mL was shown in Fig.4. In Fig.4, the efficiency recovery was highest in digested by a mix of HNO_3 - H_2O_2 (1:1, v/v) and was used for the subsequent experiments.

3.3. ICP-MS validation

ICP-MS validation. Following the optimal conditions, Eight-point calibrations over the concentration of Cr(VI) in the range 0.02 - 50 µ*g*/L were made $R^2 > 0.99$. The method's accuracy and trueness were performed in 7 replicate sub-sample of seawater containing Cr(VI) at 10 µg/L and assessed through the percent of relative standard deviation (% RSD) and percent of recovery efficiency (% RE) value, which was made %RSD < 30% and %RE in the range 60 - 120%, respectively. The limit of detection (LOD) was performed in 7 replicates of sub-sample seawater containing Cr(VI) at 1 µg/L. The results were shown in table 2.

Table 2. *Standard curve parameters of two methods ICP-MS and Spectrometry.*

Method	Calibration Curve	R^2	%RSD	Recovery efficiency (%)	LOD (µg/L)
ICP-MS	Y= 6.317X	0.999	3.0	95.2	0.015
Spectrometry	Y= 0.079 X	1.000	2.3	106.4	0.66

Field samples

The 10 coastal seawater samples were collected in the coastal area of Can Gio - Can Gio District - Ho Chi Minh City. Total Cr and Cr(VI) were analyzed in these seawater samples, and Cr(III) was calculated indirectly by the difference of the total Cr content minus the Cr(VI) content, shown in table 3.

Table 3. *Parameter results between the two methods ICP-MS and Spectrometry.*

N^o	**Sample**	**Results (µg/L)**				
		ICP-MS				**Spectrometry**
		Total Cr	**Cr (VI)**	**Cr (III)**	**% Recovery****	
1	SW 1	0.35	0.34	0.007	92.1	ND
2	SW 2	0.40	0.29	0.106	93.0	ND
3	SW 3	0.30	0.29	0.026	96.0	ND
4	SW 4	0.43	0.42	0.008	92.3	ND
5	SW 5	0.30	0.29	0.016	98.6	ND
6	SW 6	0.44	0.14	0.303	103.1	ND
7	SW 7	0.33	0.29	0.066	99.2	ND
8	SW 8	0.34	0.14	0.203	99.3	ND
9	SW 9	0.36	0.29	0.088	107.8	ND
10	SW 10	0.45	0.44	0.009	101.2	ND

**ND: Not detected*
*** Calculated at samples spiked 10 µg/L Cr (VI)*

According to the outcome of parallel experiment with common method, which used spectrometer and did not apply QuEChERS extraction step with (%RSD 2.3% - LOD 0.66 µg/L), the ICP-MS method presents a better result with smaller (%RSD 3.0% - LOD – 0.015 µg/L), and recovery range.

4. CONCLUSIONS

The determination of Cr(III) and Cr(VI) can be done very effectively in the DPC method combined with QuEChERS extraction technique and analysis by ICP-MS at trace concentrations in seawater samples. The proposed method has been successfully applied to determine Cr(III) and Cr(VI) in seawater samples and has a recovery efficiency of over 92% in all cases, an advantage that can remove the salt background height in seawater, with lower cost, higher sensitivity and more reliability. According to the experimental results, this proposed method gives better results with a smaller LOD – 0.015 µg/L, RSD – 3.0% and recovery range.

REFERENCES

[1]. Guertin, J., Jacobs, J. A., Avakian, C. P., *"Chromium (VI) Handbook"*, 1st ed., CRC Press: Boca Raton, FL, USA; p. 784, (2005).

[2]. McNeill, L. S., McLean, J. E., Parks, J. L., Edwards, M. A., *"Hexavalent chromium review, part 2: Chemistry occurrence and treatment"*. J. Am. Water Works Assoc. 104, E395–E405, (2012).

[3]. Martone, N., Rahman, G. M. M., Pamuku, M., and Kingston, H. M. S., *"Determination of chromium species in dietary supplements using speciated isotope dilution mass spectrometry with mass balance,"* Journal of Agricultural and Food Chemistry, vol. 61, no. 41, pp. 9966–9976, (2013).

[4]. Korshoj, L. E., Zaitouna, A. J., and Lai, R. Y., *"Methylene bluemediated electrocatalytic detection of hexavalent chromium,"* Analytical Chemistry, vol. 87, no. 5, pp. 2560–2564, (2015).

[5]. Achmad, R., Budiawan, B., Auerkari, E., *"Effects of Chromium on Human Body"*. Annu. Res. Rev. Biol, 13, 1–8, (2017).

[6]. ASTM D1687-17, *"Standard Test Methods for Chromium in Water"*, ASTM International, West Conshohocken, PA, (2017).

[7]. ISO 11083:1994, *"Qualite de l'eau - Dosage du chrome(VI) - Methode par spectrometrie d'absorption moleculaire avec la 1,5- diphenylcarbazide"*, Comite technique ISO/TC 147/SC 2, (1994).

[8]. NF T90-043, *"Essais des eaux - Dosage du chrome (VI) - Methode par spectrometrie d'absorption moleculaire"*, AFNOR, 1988.

[9]. Kimbrough, D. E., Cohen, Y., Winer, A. M., Creelman, L., Mabuni, C. A., *"Critical Assessment of Chromium in the Environment"*. Crit. Rev. Environ. Sci. Technol. 29, 1-46, (1999).

[10]. Hassani, H., Ghodsi, M., and Howell, G., *"A note on standard deviation and standard error".*

[11]. Tiêu chuẩn Việt Nam TCVN 5998:1995 (ISO 5667-9: 1992).

TÓM TẮT

Xác định Cr(III) và Cr(VI) trong nước biển bằng phương pháp ICP-MS kết hợp với kỹ thuật chiết QuEChERS

Hiện nay, Cr(III) và Cr(VI) tồn tại ở dạng vết được phân tích trực tiếp bằng cách sử dụng sắc ký lỏng trao đổi anion - khối phổ plasma kết hợp cảm ứng (LC-ICP-MS), được biết đến là một phương pháp phức tạp và tốn kém. Do đó, để xác định Cr(III) và Cr(VI) thì hầu hết các phòng thí nghiệm thường sử dụng là phương pháp quang phổ kế hoặc kết hợp với ICP (hoặc AAS) mặc dù phương pháp này có giới hạn phát hiện (LOD) cao hơn và bị chịu ảnh hưởng trực tiếp bởi các ion muối trong nước biển. Trong nghiên cứu này. Sau khi Cr(VI) được tạo phức màu tím đỏ bằng cách thêm 1,5-diphenylcarbazide (DPC) vào các mẫu nước biển, và kỹ thuật chiết QuEChERS đã được áp dụng để tách cấu trúc Cr(VI)-DPC khỏi chất nền có hàm lượng muối cao và chuyển sang pha Acetonnitrile. Trong trường hợp xác định tổng Cr, Cr(III) trong mẫu phải được oxy hóa thành Cr(VI) bằng cách thêm $KMnO_4$ vào trước khi thêm DPC. Sau đó, quá trình phân hủy bằng hỗn hợp $HNO_3 – H_2O_2$ được thực hiện trước khi đưa vào phân tích Cr(VI) bằng ICP-MS. Hàm lượng Cr(III) được tính gián tiếp dựa trên kết quả của tổng Cr và Cr(VI). Phương pháp này có khoảng nồng độ tuyến tính của Cr(VI) từ 0.02 đến 50 µg/L với giới hạn phát hiện là 0.015 µg/L, độ lệch chuẩn tương đối dưới 3.03% và độ thu hồi là 92 đến 108% đối với các mẫu nước biển đã được phân tích. Theo kết quả so sánh đối với thí nghiệm song song và phương pháp thông thường, sử dụng quang phổ kế và không áp dụng kỹ thuật chiết QuEChERS thì phương pháp quang phổ kế với sự kết hợp của ICP-MS cho kết quả tốt hơn với LOD, RSD và dải tuyến tính nhỏ hơn.

Keywords: Cr(VI); Cr(III); QuEChERS; ICP-MS; Nước biển.