PAMELA DA ROCHA PATRÍCIO

MECANISMO DE PARTIÇÃO E QUANTIFICAÇÃO DAS ESPÉCIES DE CROMO USANDO SISTEMA AQUOSO BIFÁSICO

Tese apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Agroquímica, para obtenção do título de *Doctor Scientiae*.

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APROVADA: 5 de julho de 2016.

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Maria do Carmo Hespanhol da Silva
(Orientadora)

Aos meus pais Neusa e José Patrício,

Aos meus irmãos Elaine e Orley

Ao Rodrigo

Dedico

Para tudo há uma ocasião, e um tempo para

cada propósito debaixo do céu:

Há tempo de nascer e tempo de morrer;

tempo de plantar e tempo de arrancar;

tempo de matar e tempo de curar;

tempo de derrubar e tempo de construir.

Há tempo de ficar triste e tempo de se alegrar;

tempo de chorar e tempo de dançar;

tempo de espalhar pedras e tempo de ajuntá-las;

tempo de abraçar e tempo de afastar.

Há tempo de procurar e tempo de perder;

tempo de economizar e tempo de desperdiçar;

tempo de rasgar e tempo de remendar;

tempo de ficar calado e tempo de falar.

Há tempo de amar e tempo de odiar;

tempo de guerra e tempo de paz.

Eclesiastes 3, 1-8

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BIOGRAFIA

Pamela da Rocha Patrício, filha de José Patrício Filho e Neusa de Rocha Patrício, nasceu em 07 de Julho de 1987, na cidade de Divinópolis, Minas Gerais.

Em maio de 2006 ingressou no curso de Química da Universidade Federal de Viçosa. Neste mesmo ano iniciou suas atividades como estagiária voluntária no grupo de pesquisa QUIVECOM, onde foi bolsista da Fapemig, no período de março de 2007 a dezembro de 2010 sob orientação da Profª. Maria do Carmo Hespanhol da Silva no Departamento de Química da UFV.

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SUMÁRIO

LISTA DE SÍMBOLOS E ABREVIATURAS

A_{MRP} – absorbância da fase rica em macromolécula

A_{ERP} – absorbância da fase rica em eletrólito

$\mathrm{C}_{\mathrm{Cr(III)}}^{\mathrm{ABS}}$ – concentração de Cr(III) no sistema aquoso bifásico

$\mathrm{C}_{\mathrm{Cr(VI)}}^{\mathrm{ABS}}$ – concentração de Cr(VI) no sistema aquoso bifásico

$\mathrm{C}_{\mathrm{E}}^{\mathrm{FRE}}$ ou $\mathrm{C}_{\mathrm{E}}^{\mathrm{ERP}}$ – concentração de eletrólito na fase rica em eletrólito

$\mathrm{C}_{\mathrm{E}}^{\mathrm{FRM}}$ ou $\mathrm{C}_{\mathrm{E}}^{\mathrm{MRP}}$ – concentração de eletrólito na fase rica em macromolécula

$\mathrm{C}_{\mathrm{P}}^{\mathrm{FRE}}$ ou $\mathrm{C}_{\mathrm{P}}^{\mathrm{ERP}}$ – concentração de polímero na fase rica em eletrólito

$\mathrm{C}_{\mathrm{P}}^{\mathrm{FRM}}$ ou $\mathrm{C}_{\mathrm{P}}^{\mathrm{MRP}}$ – concentração de polímero na fase rica em macromolécula

CFRE – composição da fase rica em eletrólito

CFRM – composição da fase rica em macromolécula

CG – composição global

CLA ou TLL – comprimento da linha de amarração

CONAMA – Conselho Nacional do Meio Ambiente

$\mathrm{Cr}_{\mathrm{T,PRP}}$ – concentração de cromo total na fase rica em polímero

$\mathrm{Cr}_{\mathrm{T,ERP}}$ – concentração de cromo total na fase rica em eletrólito

D – coeficiente de distribuição

DPC – 1,5-difenilcarbazida

EF – fator de enriquecimento

ELT – extração líquido-líquido tradicional

EO – óxido de etileno

EPA - Agência de proteção ambiental

ET-AAS – espectrometria de absorção atômica com atomização eletrotérmica

FAAS – espectrometria de absorção atômica com chama

FRM ou MRP – fase rica em macromolécula

FRE ou ERP – fase rica em eletrólito

HPLC – cromatografia líquida de alta eficiência

ICP-MS – espectrometria de massas com plasma indutivamente acoplado

ICP-OES – espectrometria de emissão óptica com plasma indutivamente acoplado

K – constante de estabilidade ou constante de formação

K_{DPC} – coeficiente de partição da 1,5-difenilcarbazida

LA – linha de amarração

LOD – limite de detecção

LOQ – limite de quantificação

MM – massa molar

m_{PRP} – massa da fase rica em polímero

m_{ERP} – massa da fase rica em eletrólito

m_{ABS} – massa do sistema aquoso bifásico

$n_{Cr\ total}^{ERP}$ – número de mol de cromo total na fase rica em eletrólito

$n_{Cr\ total}^{PRP}$ – número de mol de cromo total na fase rica em polímero

$n_{Cr(III)}^{ERP}$ – número de mol de Cr(III) na fase rica em eletrólito

$n_{Cr(III)}^{PRP}$ – número de mol de Cr(III) na fase rica em polímero

$n_{Cr(VI)}^{ERP}$ – número de mol de Cr(VI) na fase rica em eletrólito

$n_{Cr(VI)}^{PRP}$ – número de mol de Cr(VI) na fase rica em polímero

n_{M}^{MRP} – número de mol do íon metálico na fase rica em macromolécula

n_{M}^{total} – número de mol do íon metálico no sistema

Pc – ponto crítico

PEO – poli(óxido de etileno)

PF – fator de pré-concentração

PRP – fase rica em polímero

PPO – poli(óxido de propileno)

PRP – fase rica em polímero

RSD – desvio padrão relativo

SAB, ABS ou ATPS – sistema aquoso bifásico

$S_{Cr(VI), Cr(III)}$ – fator de separação entre Cr(III) e Cr(VI)

TBAB – brometo de tetrabutilamônio

UV-Vis – ultra violeta – visível

WHO – Organização Mundial da Saúde

%E – porcentagem de extração

% (m/m) – porcentagem massa/massa

%R – porcentagem de recuperação

$\Delta_{mix}H$ – variação da entalpia de mistura

$\Delta_{mix}G$ – variação da energia livre de mistura

$\Delta_{mix}S$ –variação da entropia de mistura

λ – comprimento de onda

γ – coeficiente de atividade

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RESUMO

PATRÍCIO, Pamela da Rocha, D.Sc., Universidade Federal de Viçosa, julho de 2016. **Mecanismo de partição e quantificação das espécies de cromo usando sistema aquoso bifásico**. Orientadora: Maria do Carmo Hespanhol da Silva. Coorientadores: Luis Henrique Mendes da Silva e Luis Antônio Minim.

Os dois principais estados de oxidação do elemento cromo, Cr(III) e Cr(VI), divergem bastante nas suas propriedades biológicas e toxicológicas, assim, é muito importante distingui-los. As técnicas analíticas mais usuais para quantificação permitem a determinação da quantidade total de cromo, em vez do teor de cada espécie. Portanto, é uma contribuição para área desenvolver novos métodos para a especiação deste elemento. No presente trabalho foi avaliada a partição de Cr(III) e Cr(VI) utilizando sistema aquoso bifásico (SAB) formado por polímero (PEO1500) ou copolímero tribloco (L64), um eletrólito (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, ou $C_4H_4Na_2O_6$) e água, sem a presença de qualquer extratante. O íons Cr(III) particionaram favoravelmente para a fase rica em eletrólito (FRE) do SAB, enquanto os íons Cr(VI) se concentraram preferencialmente na fase rica em macromolécula (FRM) na maioria dos SAB estudados. A porcentagem de extração (%E) das espécies de cromo da FRE para FRM foi afetada pelo pH, comprimento da linha de amarração (CLA), natureza do eletrólito e hidrofobicidade da macromolécula. O SAB PEO1500 + Na_2SO_4 + H_2O em pH 2,0 à 25,0 °C revelou ser um excelente sistema para especiar cromo sendo obtida $\%E_{Cr(III)} = 0{,}0100$ %, $\%E_{Cr(VI)} = 99{,}9$ % e fator de separação ($S_{Cr(VI),Cr(III)}$) = $1{,}00 \times 10^8$. Diante desses resultados foi desenvolvido um método analítico para a determinação das espécies Cr(III) e Cr(VI). O SAB PEO1500 + Na_2SO_4 + H_2O em pH 2,0 e CLA = 46,97 % (m/m) à 25,0 °C foi utilizado para a separação e, ou pré-concentração das espécies, enquanto a espectrometria de absorção atômica com chama (EAAC) foi usada para determinação das espécies. As espécies não sofreram interconversão durante o procedimento de separação e, ou pré-concentração. O método proposto foi aplicado para determinação de Cr(III) e Cr(VI) em amostras de água de torneira, estação de tratamento, rio e resíduo de galvanoplastia. As figuras de mérito foram limite de detecção 0,0538 mg kg^{-1}, limite de quantificação 0,163 mg kg^{-1}, faixa dinâmica 0,0500 - 4,00 mg kg^{-1}, precisão 0,802 % para Cr(VI) e 3,53 % para Cr(III) e exatidão 13,6 %. O método proposto apresenta vantagens em relação a outros métodos existentes, como ser ambientalmente seguro, pois utiliza componentes

atóxicos e biodegradáveis, além de ser robusto, simples, de baixo custo e permitir uma rápida separação de fase sem a formação de emulsões estáveis.

ABSTRACT

PATRÍCIO, Pamela da Rocha, D.Sc., Universidade Federal de Viçosa, July, 2016. **Partition mechanism and quantification of chromium species using aqueous biphasic system**. Adviser: Maria do Carmo Hespanhol da Silva. Co-advisers: Luis Henrique Mendes da Silva and Luis Antônio Minim.

The two principal oxidation states of the chromium element, Cr(III) and Cr(VI), are very different in their biological and toxicological properties, so, it is very important to distinguish between them. The usual analytical techniques for quantification allow the determination of the total amount of chromium rather than each species content. Consequently, it is a contribution to the field the development of new methods for this element speciation. In the present work, it was investigated the Cr(III) and Cr(VI) partition using aqueous biphasic systems (ABS) formed by polymer (PEO1500) or a triblock copolymer (L64), an electrolyte (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, or $C_4H_4Na_2O_6$) and water, without the presence of any extractant. Cr(III) ions were favorably partitioned to the electrolyte-rich phase (ERP) of the ABS, whereas the Cr(VI) ions were preferentially concentrated in the macromolecule-rich phase (MRP) in the most analyzed ABS. The extraction percentage (%E) of chromium species from the ERP to the MRP was affected by pH, tie-line length (TLL), electrolyte nature, and macromolecule hydrophobicity. The PEO1500 + Na_2SO_4 + H_2O ABS, TLL = 46.97 % (m/m), at pH 2.0 and 25.0 °C has proved to be an excellent system to speciate chromium being obtained $\%E_{Cr(III)} = 0.0100$ %, $\%E_{Cr(VI)} = 99.9$ % and the separation factor ($S_{Cr(VI),Cr(III)}$) = 1.00×10^8. Considering these results it was developed an analytical method for Cr(III) and Cr(VI) determination. PEO1500 + Na_2SO_4 + H_2O ABS at pH 2.0, 25.0 °C and TLL = 46.97 % (m/m) was used to separate and, or pre-concentrate the species, while flame atomic absorption spectrometry (FAAS) was used to determine the species. The species do not interconvert during the separation and, or pre-concentration procedure. The proposed method was applied to determine Cr(III) and Cr(VI) in water samples of tap, treatment plant, river and electroplating waste. The figures of merit were limit of detection 0.0538 mg kg^{-1}, limit of quantification 0.163 mg kg^{-1}, dynamic range 0.0500 - 0.500 or 0.100 - 4.00 mg kg^{-1}, precision 0.802 % for Cr(VI) and 3.53 % for Cr(III) and accuracy 13.6%. The proposed method has advantages over other existing methods, as to be environmentally safe, since it uses nontoxic and biodegradable components, besides being robust, simple, low cost and allows fast phase separation without the formation of stable emulsions.

CAPÍTULO 1

Revisão de literatura

1.1. INTRODUÇÃO

O desenvolvimento de novas técnicas analíticas tem propiciado cada vez mais a obtenção de melhores limites de detecção, quantificação, seletividade e sensibilidade. Todavia, estas nem sempre eliminam a necessidade de etapas de pré-tratamento da amostra para a quantificação das espécies de interesse. Neste contexto, um processo analítico que se destaca no pré-tratamento de amostras é a extração líquido-líquido tradicional (ELT).

A ELT é muito importante para separação e pré-concentração de analitos [1]. Estes são processos auxiliares empregados quando técnicas não podem alcançar, diretamente, os objetivos de uma análise: identificação de componentes e determinação de seus valores [1]. A ELT é caracterizada pela transferência de solutos contidos numa solução aquosa para uma fase orgânica [2]. Além disso, ela permite o emprego de diferentes solventes, extratantes e consequentemente a determinação de uma grande variedade de analitos [3]. Contudo, uma desvantagem presente neste processo analítico é o uso de solventes orgânicos que, normalmente, são tóxicos, cancerígenos e, ou inflamáveis [3].

Uma técnica ambientalmente segura e alternativa à ELT é o sistema aquoso bifásico (SAB) [3,4]. Este sistema é formado majoritariamente por água e seus demais componentes são geralmente atóxicos, não inflamáveis e muitas vezes recicláveis e biodegradáveis [4,5]. A característica do SAB de ser ambientalmente seguro aliado a simplicidade da técnica [6], baixo custo [5], rápida separação de fase [5], facilidade de operação [5,6] e possibilidade de trabalhar em larga escala [5-7] o torna atrativo para a separação, pré-concentração e determinação de biomoléculas [8-10], fenóis [11,12], corantes [13-16] e íons metálicos [5,7,17-20].

1.2. SISTEMA AQUOSO BIFÁSICO

1.2.1. Sistema Aquoso Bifásico: Termodinâmica de Formação

O SAB consiste de duas fases líquidas imiscíveis formadas espontaneamente pela combinação de soluções aquosas de polímero-polímero [21], polímero-eletrólito [22], líquido iônico-eletrólito [23], ou líquido iônico-polímero [4], sob condições termodinâmicas específicas de temperatura, composição e pressão [4]. Cada fase do sistema heterogêneo resultante é rica em um dos componentes, mas a água é o componente majoritário das duas fases.

A formação de duas fases durante o processo de mistura das soluções aquosas dos componentes formadores do SAB dependerá das interações intermoleculares entre os reagentes, expressas em termos da variação da energia livre de mistura ($\Delta_{mix}G$), Eq. (1.1) [24]:

$$\Delta_{mix}G = \Delta_{mix}H - T\,\Delta_{mix}S \tag{1.1}$$

onde, $\Delta_{mix}H$ é a variação da entalpia de mistura, $\Delta_{mix}S$ é a variação da entropia de mistura e T é a temperatura em Kelvin. Segundo a termodinâmica, a mistura de três componentes, resultará em um sistema homogêneo sempre que $\Delta_{mix}G < 0$, caso contrário, o sistema buscará uma nova configuração (a produção de duas fases em equilíbrio), que terá uma menor energia livre [24].

Os SAB explorados neste trabalho são os sistemas formados pela combinação de polímero (ou copolímero), eletrólito e água. Os estudos calorimétricos realizados por da Silva e Loh [25] evidenciaram que a $\Delta_{mix}H$ de sistemas com composição semelhante é positiva, ou seja, a interação entre polímero, eletrólito e água ocorre com absorção de energia. Assim, acredita-se que ao misturar soluções aquosas de polímero (ou copolímero) e eletrólito, os cátions oriundos do eletrólito solvatam os segmentos de óxido de etileno da macromolécula e liberam moléculas de água que antes solvatavam a cadeia polimérica, aumentando a entropia do sistema [25]. Este processo acarreta diminuição no valor de $\Delta_{mix}G$ e mantém o sistema homogêneo [25]. Considera-se que a solvatação dos segmentos da macromolécula pelos cátions do eletrólito ocorre até que haja uma saturação energética dos segmentos das macromoléculas. Nesta situação, a adição de eletrólito acarretará na interação dos íons com as moléculas de água, organizando-as em camadas de solvatação e reduzindo a entropia translacional do sistema. Assim, o sistema segrega-se em duas fases para minimizar a energia livre de Gibbs [25]. Esse processo segregativo resulta em um sistema heterogêneo composto por duas fases imiscíveis, sendo uma fase rica em macromolécula (FRM), e a outra fase rica em eletrólito (FRE) (Fig. 1.1).

COMPOSIÇÃO GLOBAL

[PEO1500] = 26,61 % (m/m)

[Na_2SO_4] = 10,76 % (m/m)

[H_2O] = 62,63 % (m/m)

FASE RICA EM MACROMOLÉCULA

[PEO1500] = 46,29 % (m/m)

[Na_2SO_4] = 1,20 % (m/m)

[H_2O] = 52,51 % (m/m)

FASE RICA EM ELETRÓLITO

[PEO1500] = 0,59 % (m/m)

[Na_2SO_4] = 23,58% (m/m)

[H_2O] = 75,83 % (m/m)

Fig. 1.1. Composição do SAB formado por PEO (poli(óxido de etileno) 1500 g mol^{-1}) + sulfato de sódio + água a 25 ºC [26].

Presume-se que após a separação de fase do sistema, cátions do eletrólito formador do SAB estabelecem uma forte interação com os segmentos de óxido de etileno das cadeias poliméricas e conduz a formação de uma estrutura carregada positivamente chamada de pseudopolicátion (Fig. 1.2) que interage eletrostaticamente com analitos aniônicos [7,25].

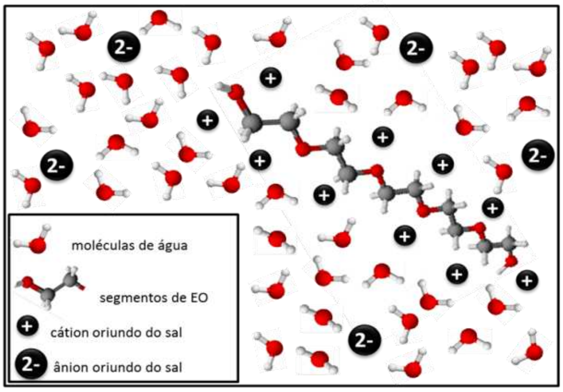


Fig. 1.2. Representação esquemática de formação do pseudopolicátion em solução aquosa devido à interação cátion-PEO.

1.2.2. Sistema Aquoso Bifásico: Princípios e Propriedades

O sistema aquoso bifásico pode ser caracterizado por um diagrama de fases retangular como exibido na Fig. 1.3. Neste exemplo o SAB é formado por polímero, eletrólito e água.

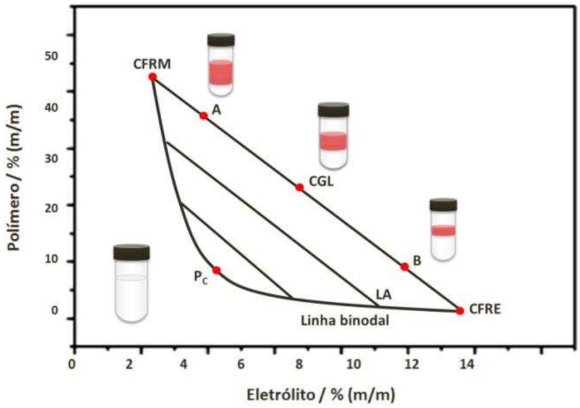


Fig. 1.3. Diagrama de fases expresso em coordenadas retangulares de um SAB constituído por polímero, eletrólito e água. (CFRM = composição da fase rica em macromolécula, CFRE = composição da fase rica em eletrólito, CGL = composição global, LA = linha de amarração, Pc = ponto crítico).

No diagrama de fases, normalmente a concentração do eletrólito do sistema se encontra no eixo das abscissas e a concentração do polímero no eixo das ordenadas. Estas concentrações geralmente são expressas em porcentagem mássica, % (m/m). O diagrama (Fig. 1.3) delineia a potencial área de trabalho do sistema aquoso bifásico, pois, apresenta as composições globais (CGL) que irão formar duas fases em equilíbrio, a composição da fase rica em macromolécula (CFRM) e composição da fase rica em eletrólito (CFRE) [24].

No diagrama de fases (Fig. 1.3) também é exibido a linha binodal que representa a concentração limite que separa a região monofásica da região bifásica [4]. Sua posição varia de acordo com o tipo e a massa molar do polímero, natureza química do eletrólito e temperatura [24]. Já as linhas de amarração (LA) são retas que ligam pontos no diagrama que representam a composição das duas fases em equilíbrio, CFRM e CFRE [24]. Ao longo de uma mesma LA, todas as propriedades termodinâmicas intensivas de cada fase são constantes (densidade, condutividade), enquanto as propriedades termodinâmicas extensivas (volume, massa) variam [24]. Na Fig. 1.3, por exemplo, os pontos de composição global A e B terão uma mesma composição de fase rica em macromolécula e eletrólito, enquanto os volumes das fases variam ao longo da linha de amarração. O comprimento da linha de amarração (CLA) é um indicador numérico da diferença de composição entre as duas fases em equilíbrio [4]. Este parâmetro é calculado a partir dos valores de concentração dos solutos em cada fase, como mostra a Eq. (1.2):

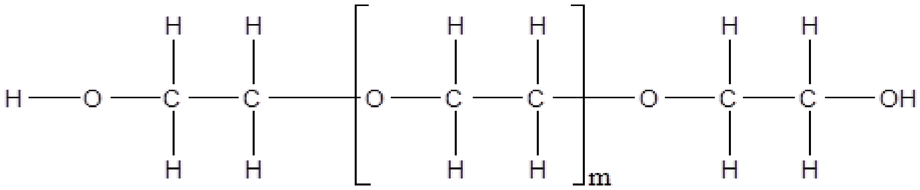
$$CLA = \left[\left(C_P^{FRM} - C_P^{FRE} \right)^2 + \left(C_E^{FRM} - C_E^{FRE} \right)^2 \right]^{1/2} \tag{1.2}$$

onde, C_P^{FRM} e C_P^{FRE} são as concentrações do polímero (% (m/m)) na fase rica em macromolécula e eletrólito, respectivamente, enquanto C_E^{FRM} e C_E^{FRE} são as concentrações do eletrólito (% (m/m)) na fase rica em macromolécula e eletrólito, respectivamente [24]. Assim, quanto maior o valor do CLA, maior a diferença de composição entre as fases, elevando portanto a eficiência na extração e, ou partição do soluto de interesse, desde que esse apresente migração preferencial para uma das fases [24].

1.2.3. Sistema Aquoso Bifásico: Histórico

Os sistemas aquosos bifásicos foram descobertos por Beijerinck em 1896 [27,28] que ao misturar soluções aquosas de gelatina e ágar ou gelatina e amido solúvel, percebeu que o sistema se segregava espontaneamente em duas fases. Todavia, esta técnica apenas se expandiu após os estudos de Albertsson [29] na década de 50 que empregou SAB formados por soluções aquosas de dois polímeros estruturalmente distintos na partição de biomateriais.

A aplicação destes sistemas na extração de íons metálicos iniciou-se em 1984 [30] e desde então os sistemas formados por misturas de polímero e eletrólito inorgânico predominam, em relação àqueles formados pela mistura de dois polímeros. O principal polímero utilizado é o poli(óxido de etileno), PEO, cuja estrutura molecular é exibida na Fig. 1.4. Trata-se de uma macromolécula linear que contém em sua cadeia a unidade monomérica (-CH_2-CH_2-O-) e grupos terminais R= OH e R'= H [3].



$$H-O-CH_2-CH_2-[O-CH_2-CH_2]_m-O-CH_2-CH_2-OH$$

Fig. 1.4. Estrutura molecular do polímero PEO.

Em 1984 Zvarova e colaboradores [30] empregaram o SAB constituído por

PEO2000 + $(NH_4)_2SO_4$ + H_2O na extração de metais e avaliaram a influência de extratantes orgânicos e inorgânicos solúveis em água no comportamento de partição de cobre, zinco, cobalto, ferro(III), índio e molibdênio(V). Desde então, surgiram alguns trabalhos em que o comportamento de extração de íons metálicos incluindo lantanídeos [31], actinídeos [31-33] e outros metais de transição foi estudado, em diferentes SABs e na presença de diferentes extratantes.

Alguns trabalhos têm reportado a utilização de SAB na extração de íons metálicos na ausência de extratante [19,20]. Todavia, normalmente estes sistemas de extração empregam extratantes solúveis em água como iodeto [17,34-36], tiocianato [35,36] e arsenazo III [31]. Por outro lado, o emprego de extratantes hidrofóbicos como 1-(2-piridilazo)-2-naftol [18] e 1-nitroso-2-naftol [5,36], tornou-se possível apenas em 2008 [36] com o emprego de SAB formados por copolímeros triblocos de poli(óxido de etileno)-poli(óxido de propileno)-poli(óxido de etileno) da série Pluronic que são constituído por três blocos de polímeros e designados por PEO-PPO-PEO (Fig. 1.5) [3,36].

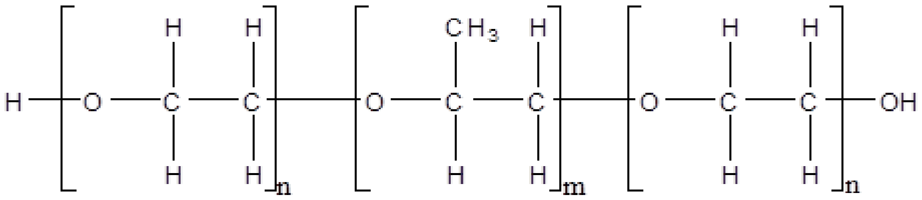


Fig. 1.5. Estrutura molecular do copolímero L64 [3].

Quando o copolímero tribloco é solubilizado em água, em uma determinada temperatura e concentração, ocorre um processo de micelização [36]. As micelas formadas possuem um núcleo hidrofóbico composto por PPO, rodeado por uma parte hidrofílica constituída por PEO [36]. Assim, os núcleos hidrofóbicos são capazes de solubilizar complexantes hidrofóbicos e particionar seus respectivos complexos metálicos para a fase rica em copolímero (Fig. 1.6) [36] e na coroa hidrofílica (unidades de óxido etileno) se formam pseudopolicátions que podem extrair analitos aniônicos [37].

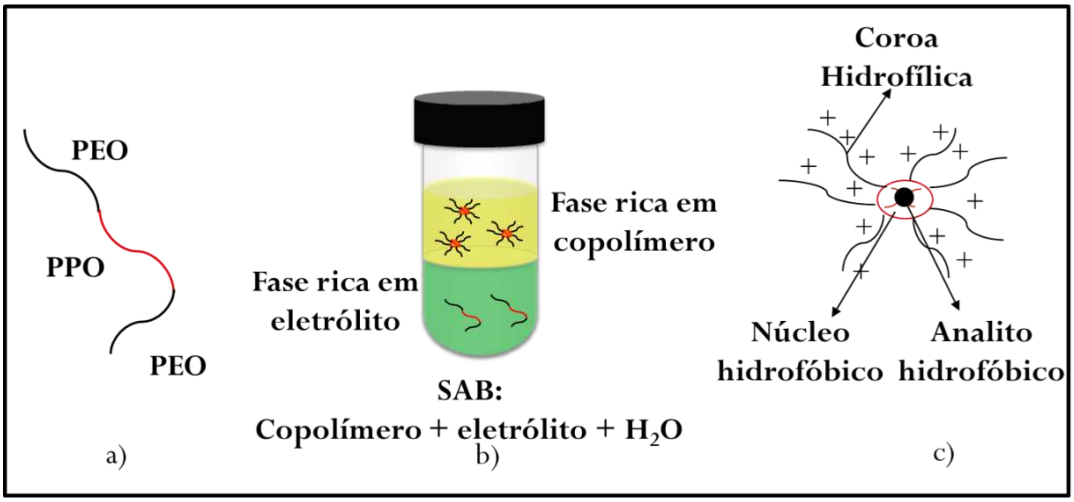


Fig. 1.6. (a) Molécula de copolímero tribloco PEO-PPO-PEO; (b) sistema aquoso bifásico formado por copolímero, eletrólito e água; (c) analito hidrofóbico (complexo metálico ou complexante hidrofóbico) solubilizado na parte hidrofóbica das micelas.

Outra atraente característica deste sistema é a possibilidade de reciclar os polímeros e copolímeros normalmente empregados. Estes reagentes são termosseparáveis e sob aumento de temperatura da fase rica em polímero/copolímero esta se segrega em uma fase rica em água e outra rica em macromolécula [38-40]. Todavia, essa termosseparação ocorre em menores temperaturas para os copolímeros constituídos por PEO-PPO-PEO (cerca de 50 ºC) quando comparado a temperatura de reciclagem dos polímeros formados por PEO (acima de 100 ºC) [38].

1.3. OBJETIVOS

1.3.1. Objetivo geral

O trabalho proposto tem como objetivo desenvolver um método de especiação de cromo empregando sistema aquoso bifásico.

1.3.2. Objetivos específicos

✓ Estudar o comportamento de partição das espécies Cr(III) e Cr(VI) utilizando diferentes sistemas aquosos bifásicos: PEO1500 + eletrólito (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, $C_4H_4Na_2O_6$) + H_2O, L64 + eletrólito (Li_2SO_4, Na_2SO_4).

✓ Avaliar a influência dos parâmetros: efeito da quantidade de extratante, pH, hidrofobicidade da macromolécula formadora do SAB, natureza do eletrólito formador do SAB, composição do SAB e concentração do metal, sobre a porcentagem de extração dos íons Cr(III) e Cr(VI).

✓ Verificar se o SAB converte Cr(III) em Cr(VI) ou Cr(VI) em Cr(III).

✓ Desenvolver um procedimento de pré-concentração utilizando SAB.

✓ Determinar Cr(III) e Cr(VI) em altos e baixos teores em diferentes matrizes aquosas utilizando SAB e espectrometria de absorção atômica com chama.

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CAPÍTULO 2

Chromium speciation using aqueous biphasic systems: Development and mechanistic aspects

2.1. ABSTRACT

The two principal oxidation states of chromium diverge significantly in their biological and toxicological properties, and it is very important to distinguish between them. Develop procedures that are efficient, fast, easier and cheaper for chromium speciation is an analytical challenge. Chromium speciation was investigated using aqueous biphasic systems (ABS) formed by polyethylene oxide (PEO1500) or a triblock copolymer (L64), an electrolyte (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, or $C_4H_4Na_2O_6$) and water, without the presence of any extractant. Cr(III) was favorably partitioned to the electrolyte-rich phase (ERP) of the ABS, whereas the Cr(VI) was preferentially concentrated in the macromolecule-rich phase (MRP) in most analyzed ABS. The fluorescence spectroscopy data revealed that the Cr(III) and Cr(VI) separation process is governed by specific interactions between the ethylene oxide segments in the macromolecules and the Cr(VI) species, which do not occur for Cr(III) species. The extraction percentage (%E) of chromium species from the ERP to the MRP was affected by pH, tie-line length (TLL), electrolyte nature, and macromolecule hydrophobicity. For the ABS comprising of PEO1500 + Na_2SO_4 + H_2O, TLL = 50.89% (m/m), 25°C and pH 2.0, the %E values for Cr(III) and Cr(VI) were 0.0100 and 99.9%, respectively. The speciation of Cr(III) and Cr(VI) that was achieved in this ABS resulted in an excellent separation factor ($S_{Cr(VI),Cr(III)}$) of 1.00×10^8.

Keywords: chromium; speciation; aqueous biphasic system; fluorescence; green chemistry

2.2. INTRODUCTION

Chromium is a trace element and highly reactive pollutant which spreads into ecosystems from a variety of industrial activities, such as electroplating, leather tanning, steel works, refractories, pigments, mining, textile dyeing, chromate preparation and timber processing companies [1-3]. Cr(III) and Cr(VI) are the most stable and prevalent forms of this element in the environment [3]. The toxicological as well as biological properties of chromium are known to depend strongly on its oxidation state [4]. Thus, the quantification of total chromium does not provide sufficient information for understanding its toxicity, bioavailability, biotransformation and dissemination routes [4].

Cr(III) is an essential nutrient for humans, playing a vital role in the metabolism of glucose, lipids, and proteins [1,2,4-6]. In contrast, the toxic form, Cr(VI), is a suspected carcinogen, inhibits enzymatic sulfur uptake in the cell and is harmful to the lungs, liver, and kidneys [1,7]. Because of their toxicity differences, precise and reliable methods for the quantification of each species of chromium is more important than the total chromium measurement [4].

Many analytical techniques such as flame atomic absorption spectrometry (FAAS) [5,8-11], inductively coupled plasma–atomic emission spectrometry (ICP-AES) [12-14], inductively coupled plasma–mass spectrometry (ICP-MS) [15-17], electrothermal atomic absorption spectrometry (ET-AAS) [18-20] and luminescence spectroscopy [21,22] have been used for chromium determination. However, because these techniques determine only the total chromium, several additional procedures have been developed to separate the different chromium species and to determine each species. These include liquid-liquid extraction [23-25], solid-phase extraction [8,9], solid-phase extraction using chelating resins [1,10], co-precipitation [5,11,26], high performance liquid chromatography (HPLC) [4,15,27], electromembrane extraction [27], chronoamperometric biosensors [27,28] and cloud-point extraction [2,18,20].

As some of these procedures have high operational costs, are time-consuming or make use of solvents that cause environmental problems, the development of a simple, inexpensive, and efficient procedure for the speciation of chromium is essential. The use of aqueous biphasic systems (ABS) or aqueous two-phase systems (ATPS) has the potential to be an interesting approach for chromium speciation. These systems do not use organic solvents; rather, they are based on green chemistry principles, are predominantly formed by water, and their minority components (polymers and electrolytes) are nonvolatile, recyclable, nonflammable and offer safety and cost benefits [29,30]. ABS have been applied in the separation of proteins [31,32], the determination of phenol compounds [33,34] and extraction of the dyes [35,36] and metallic ions [29,30,37-39].

Despite the importance of chromium speciation, only two studies have proposed the ABS as sample preparation step for the separation of chromium species. Akama and Sali [37] used the ABS formed by tetrabutylammonium bromide (TBAB) and $(NH_4)_2SO_4$ to suggest a selective extraction of Cr(VI) in the presence of Cr(III). The system was efficient to extract Cr(VI) for the TBAB rich phase while Cr(III) remained at $(NH_4)_2SO_4$ rich phase. The system pH from 1.0 to 9.0 was investigated and the pH range from 1 to 5 was efficient to extract Cr(VI) without interference from the coexisting Cr(III). The Cr(VI) extraction was attributed to the presence of $HCrO_4^-$ ions and the ion pair formation as $HCrO_4^-.TBAB^+$. However, the driving force of Cr(VI) extraction behavior was slightly investigated. The quantity of $(NH_4)_2SO_4$ added to the system was also examined and the results showed that the increase in the

$(NH_4)_2SO_4$ amount improved the extraction of Cr(VI). This electrolyte addition results in an unknown ABS phase composition. Then, the authors not worked in defined tie-line lengths and could not determine the effect of ABS components concentration in chromium speciation procedure. Finally, Akama and Sali [37] did not evaluate the effects of the ABS components nature and restricted the investigation to only one system that included a toxic and expensive component (TBAB).

The other investigation of ABS as speciation tool was performed by Roy and Lahiri [39]. The authors used systems formed by a polymer poly(ethylene oxide) with average molar mass (MM) of 4000 g mol^{-1} and five ABS-forming electrolytes (Na_2CO_3, NaOH, $Na_2S_2O_3$, Na_2HPO_4, Na_2SO_4). The system constituted by Na_2SO_4 leads to the best condition to selective separation of chromium species. It was verified a great influence of the electrolyte anion in modulate the chromium speciation, but it was not studied the effect of electrolyte cations. The effect of pH range from 1.0 to 7.0, was also examined and at pH 3.0 Cr(VI) was quantitatively extracted to the macromolecule rich-phase (MRP) and Cr(Il) remains at electrolyte rich-phase (ERP). The authors attributed the Cr(VI) extraction behavior to the formation of $Cr_2O_7^{2-}$ and the transference of Na^+ and K^+ as counter ion for the MRP. This hypotheses confronts the extraction mechanism proposed by Akama and Sali [37]. Then, it is necessary new researches to understand the mechanistic aspects of chromium extraction in ABS for future robust applications. Moreover, similarly to Akama and Sali [37], Roy and Lahiri [39] proposed the electrolyte addition to improve the Cr(VI) extraction. Despite the increment of the percentage extraction of Cr(VI) the present procedure leads to an ABS with indefinite phases composition and the authors also not assessed the effect of the phase components concentration in chromium speciation. Another research gap was the absence of ABS-forming polymer investigation.

The present work studies the influence of two new polymers, five electrolytes, and discusses the driving force behind the chromium speciation process in ABS. The extraction behaviors of Cr(III) and Cr(VI) species were investigated in different ABS composed of (i) polyethylene oxide with an average molar mass (MM) of 1500 g mol^{-1} (PEO1500), an electrolyte (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, or $C_4H_4Na_2O_6$), and water, or (ii) (polyethylene oxide)$_{13}$-(polypropylene oxide)$_{31}$-(polyethylene oxide)$_{13}$ with an average MM of 2900 g mol^{-1} (L64), an electrolyte (Li_2SO_4 or Na_2SO_4) and water. The influence of a wider pH range, tie-line length (TLL), electrolyte nature (cations and anions) and polymer hydrophobicity on the %E of the ions were systematically investigated in order to study a chromium speciation in ABS. The procedure for chromium speciation is robust, and a quantitative separation of chromium species can be performed with different ABS systems without the aid of any extractant.

2.3. EXPERIMENTAL

2.3.1. Chemicals

All reagents were of analytical grade. Milli-Q deionized water (Millipore Corp., Massachusetts, USA) was employed in the preparation of all solutions. Copolymer L64, $K_2Cr_2O_7$ and metallic chromium were acquired from Aldrich (Wisconsin, USA). Cr(III) was obtained dissolving 0.0200 g of chromium metal in 0.500 g of 1:1 HCl with gentle heating. The solution was cooled and diluted to 2.000 g to give 10,000 mg kg^{-1} Cr(III) stock solution. The polymer PEO1500 was purchased from Synth (Sao Paulo, Brazil). $Li_2SO_4 \cdot H_2O$, Na_2SO_4, $MgSO_4 \cdot 7H_2O$, $C_6H_5Na_3O_7 \cdot 2H_2O$ and $C_4H_4Na_2O_6 \cdot 2H_2O$ were obtained from Vetec (Rio de Janeiro, Brazil). HCl and 1,5-diphenylcarbazide (DPC) were purchased from Merck (Darmstadt, Germany) and Fluka (Missouri, USA), respectively.

2.3.2. Aqueous biphasic system construction

The ABS compositions — L64 + Li_2SO_4 + H_2O [40], L64 + Na_2SO_4 + H_2O [40], PEO1500 + Li_2SO_4 + H_2O [41], PEO1500 + Na_2SO_4 + H_2O [41], PEO1500 + $MgSO_4$ + H_2O [41], PEO1500 + $C_6H_5Na_3O_7$ + H_2O [42], and PEO1500 + $C_4H_4Na_2O_6$ + H_2O [42] — were obtained from the literature, and the TLLs are reported in Tables A1 to A7 (Annex). HCl and NaOH solutions in the pH range 1.0 to 11.0 were prepared from 12 mol L^{-1} HCl or 1.0 mol L^{-1} NaOH. The pH of the ABS was assumed to be equal to the pH of the HCl or NaOH solution. The pH was adjusted according to a pH meter (model Hi 221, Hanna Instruments, Brazil). To obtain the ABS L64 + Li_2SO_4 + H_2O with TLL = 21.51 % (m/m), an NaOH or HCl aqueous solution was used as the solvent to prepare stock solutions of L64 at 25.49 % (m/m) and Li_2SO_4 at 13.02 % (m/m). All macromolecule and electrolyte stock solutions were prepared in this way, but at different pH and macromolecule and electrolyte concentrations, depending on the desired ABS and TLL. The ABS was obtained by mixing 2.00 g of the appropriate macromolecule stock solution and 2.00 g of electrolyte stock solution.

2.3.3. Extraction of the chromium species

To study the %E of the chromium species, each ABS was obtained by mixing 2.00 g of the macromolecule stock solution and 2.00 g of the electrolyte stock solution (containing chromium) with the assistance of an analytical balance (Model AY-220, Shimadzu, Brazil). The total chromium concentration in the ABS was 4.00 mg kg^{-1}. After mixing the macromolecule and electrolyte stock solutions, the system was manually stirred for 3 min, centrifuged (Heraeus Megafuge 11R, Thermo Scientific, Germany) at 9503 *g* for 5 min and placed in a thermostatic bath (Model MQBTC 99-20, Microquimica, Brazil) at 25.0°C for 20 min. Two clear phases were observed. Aliquots from the MRP and/or ERP were collected and diluted, and the concentrations of Cr(III) and Cr(VI) were determined in triplicate by FAAS (Model AA240, Varian, Australia) against previously determined calibration curves ($R^2 \geq 0.999$). The chromium ions were determined with a deuterium background correction, using an air–acetylene burner at a wavelength of 357.9 nm, lamp current of 7 mA, and slit width of 0.2 nm.

The %E of each species was calculated using Eq. (2.1):

$$\%E = \frac{n_M^{MRP}}{n_M^{Total}} \times 100 \tag{2.1}$$

where n_M^{MRP} is the quantity of metal ions in the MRP in mol and n_M^{total} is the total number of moles of metal ions added to the system.

2.3.4. Influence of the quantity of extractant added on the extraction of chromium species

The L64 + Li_2SO_4 + H_2O ABS, TLL 41.86 % (m/m), 25.0°C and pH 2.0, was used to investigate the influence of the quantity of extractant added on the %E of the chromium species. Working DPC solutions at concentrations between 0 to 800 mg kg^{-1} were obtained by dissolving the DPC in the copolymer stock solution (as described in section 2.3.2), and the working chromium solution at a concentration of 8.00 mg kg^{-1} was obtained by dissolving the chromium species in the electrolyte stock solution (as described in section 2.3.2). The DPC:chromium species molar ratio varied from 0:1 to 22:1. The blank assays were performed by mixing the working DPC solutions and Li_2SO_4 stock solution. The extraction procedure was similar to that described in section 2.3.3.

2.3.5. Determination of partition coefficient of 1,5-diphenylcarbazide (K_{DPC})

The determination of K_{DPC} was performed in the system formed by L64 + Li_2SO_4 + H_2O, TLL = 41.86 % (m/m), 25.0°C and pH 2.0. To determine K_{DPC}, in a tube was added 4.00 g of DPC solution (800 mg kg^{-1}, and using the copolymer stock solution as solvent) and 4.00 g of electrolyte stock solution. After mixing, the tube was stirred for 3 min, centrifuged at 9503 *g* for 20 min and placed in a thermostatic bath at 25.0°C for 72 h. In this way, the ABS was obtained and the DPC partitioned. Aliquots of the MRP and ERP were collected and diluted. The molecular absorption spectrum in the UV-visible region of each phase was obtained using an UV/Vis spectrophotometer (Model UV–2550, Shimadzu, Japan). The partition coefficient of 1,5-diphenylcarbazide was obtained using Eq. (2.2):

$$\mathrm{K_{DPC}} = \frac{\mathrm{A_{MRP}}}{\mathrm{A_{ERP}}} \tag{2.2}$$

where A_{MRP} and A_{ERP} are the absorbance (at 281 nm) of DPC in the macromolecule- and electrolyte-rich phases, respectively.

2.3.6. Determination of the binding constant between the chromium species and ethylene oxide (EO) segments (K_b)

The binding constant of the chromium species with the pseudopolycation surface was investigated by fluorescence spectrophotometry (Cary Eclipse Fluorescence Spectrophotometer, Agilent Technologies, Malaysia). To obtain the K_b values, fluorescence measurements were performed in solutions with increasing concentrations of EO segments varying from 1.14 to 6.82 mol kg^{-1}. Then, the same concentrations of EO segments were prepared and measured in the presence of constant Cr(VI) or Cr(III) (5.00×10^{-4} mol kg^{-1}) and Na_2SO_4 (0.103 mol kg^{-1}) concentrations. The emission spectra were recorded from 300 to 550 nm with an excitation wavelength of 300 nm. The slit widths were 5 and 10 nm for excitation and emission, respectively.

The binding constants were calculated using Eq. (2.3), derived from the Benesi–Hildebrand method [43]:

$$\frac{1}{\Delta F} = \frac{1}{\Delta F_{max}} + \frac{1}{(\Delta F_{max}\, K_b\ [EO])} \tag{2.3}$$

where ΔF is the difference in fluorescence intensity between samples with and without chromium, ΔF_{max} is an estimated parameter which corresponds to the maximum ΔF value in the curve, $[EO]$ is the concentration of the EO segments (mol kg^{-1}) and K_b is the binding constant between the chromium species and the EO segments.

2.4. RESULTS AND DISCUSSION

2.4.1. Influence of the quantity of extractant added on the extraction of chromium species

In general, ions spontaneously concentrate in the ERP when partitioned in an ABS, so it is necessary to add an extractant to promote their transfer from the ERP to the MRP [29,44]. Due to the selective complexation of Cr(VI) by DPC at pH 2.0 [45,46], it was chosen as the extractant for chromium speciation in the ABS.

The %E values for the chromium species in the presence of different amounts of DPC in the ABS L64 + Li_2SO_4 + H_2O, TLL = 41.86 % (m/m), at 25.0°C and pH 2.0 are presented in Fig. 2.1.

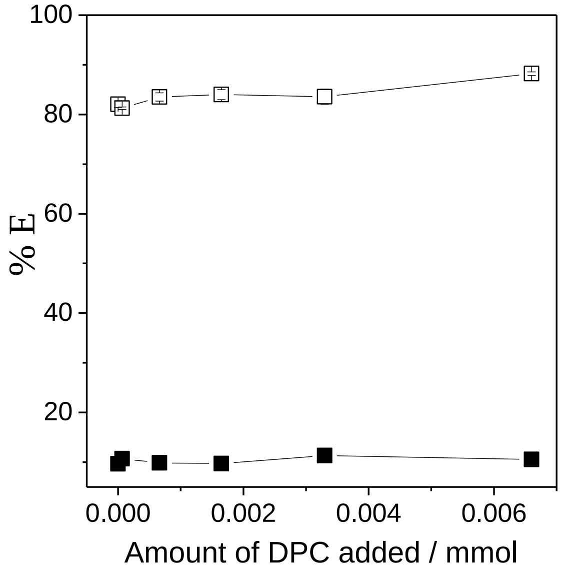

Fig. 2.1. Effect of the DPC concentration on the extraction percentage (%E) of Cr(III) and Cr(VI) in the ABS formed by L64 + Li_2SO_4 + H_2O, TLL = 41.86 % (m/m). [metal] = 4.00 mg kg^{-1} ; T = 25.0 °C; pH 2.0. (■) Cr(III) and (□) Cr(VI).

In the absence of DPC, the extraction percentages for Cr(VI) ($\%E_{Cr(VI)}$) and Cr(III) ($\%E_{Cr(III)}$) were 82.1 % and 9.65 %, respectively. The increasing addition of DPC did not affect the Cr(III) ion transfer from the ERP to the MRP, since the %E remained constant. On the other hand, the Cr(VI) partition was slightly enhanced when the DPC concentration was increased, mainly due to the transfer of the hydrophobic Cr(VI)–DPC complex to the MRP is related to a specific characteristic of the L64. In aqueous solution at specific temperature and concentration, L64 macromolecules forms micelles with a core consisting of the hydrophobic units of the polypropylene oxide (PO), delimited by a crown of the hydrophilic units of the EO [30]. The hydrophobic micelle cores solubilize the Cr(VI)–DPC complexes. Evidence for this phenomenon is the high partition coefficient of the DPC (K_{DPC} = 69.6) in this ABS.

The DPC is unequally distributed between the ABS phases, concentrating 69.6 times more in the MRP than in the ERP. This behavior indicates a preferential interaction of the hydrophobic substance DPC with the MRP components (mainly copolymer) and it should favored the Cr(VI)–DPC complexes partition.

The small increase in $\%E_{Cr(VI)}$ due DPC presence demonstrated that it is not necessary to use an extractant for efficient chromium speciation. The Cr(VI) and Cr(III) separation could be an important advance in analytical chemistry from both economic and environmental perspectives. In the absence of an extractant, other metal ions studied in ABS, with the exception of pertechnetate [47] and nitroprusside [48] ions, exhibited low distribution coefficients (D) (e.g., $D_{Ba} = 0.0035$ [49]; $D_{Ca} = 0.0022$ [49]; and $D_{Sr} = 0.0043$ [49]), which make possible to separate Cr(VI) from a complex mixture.

Because DPC was not soluble in the ABS formed with PEO, the %E of the chromium species was not evaluated in those systems.

The partitioning of ionic species in the ABS is due the different interactions between the ions and the components present in both ABS phases. It is well known that these interactions are affected by the ABS' hydrogen ion concentration [29,50]. Therefore, it would be expected that the chromium species' concentration would be dependent on the system pH.

2.4.2. Influence of pH on the extraction of chromium species

The extraction behaviors of Cr(III) and Cr(VI) as a function of pH in the ABS formed by L64 + Li_2SO_4 + H_2O (TLL = 41.86 % (m/m)), L64 + Na_2SO_4 + H_2O (TLL = 47.82 % (m/m)) or PEO1500 + Na_2SO_4 + H_2O (TLL = 46.97 % (m/m)) were investigated (Fig. 2.2).

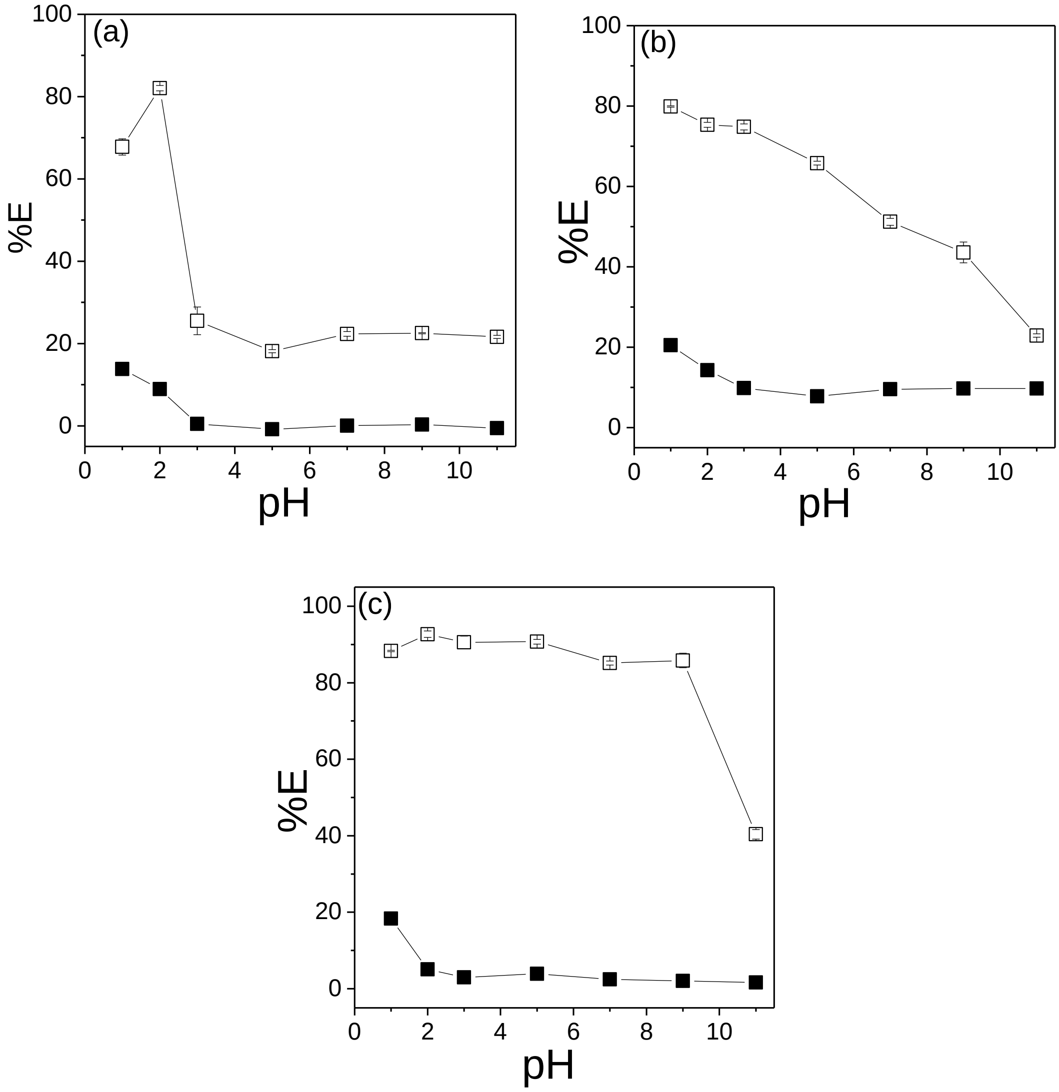

Fig. 2.2. Effect of pH on the extraction percentage (%E) of Cr(III) and Cr(VI) in the ABS formed by: (A) L64 + Li_2SO_4 + H_2O, TLL = 41.86 % (m/m); (B) L64 + Na_2SO_4 + H_2O, TLL = 47.82 % (m/m); (C) PEO1500 + Na_2SO_4 + H_2O, TLL = 46.97 %(m/m); [metal] = 4.00 mg kg^{-1}; T = 25.0 °C. (■) Cr(III) and (□) Cr(VI).

Fig. 2.2 shows that the chromium extraction is influenced by the hydrogen ion concentration, macromolecule structure, and electrolyte nature. However, for all the ABS studied, the highest differences between the %E values for Cr(VI) and Cr(III) were achieved in ABS systems at pH 2.0. The chromium species partition can be rationalized by the following mechanism.

The small extraction efficiency of Cr(III) species is related to the interaction of the metal ions with the sulfate anions present in the ERP, which promote the formation of the following complexes (Eq. (2.4)–(2.9)) [51].

$$\mathrm{Cr^{3+} + H^{+} + SO_4^{2-} \rightleftharpoons [Cr(HSO_4)]^{2+}} \tag{2.4}$$

$$\mathrm{Cr^{3+} + SO_4^{2-} \rightleftharpoons [Cr(SO_4)]^{+}} \tag{2.5}$$

$$\mathrm{Cr^{3+} + 2SO_4^{2-} \rightleftharpoons [Cr(SO_4)_2]^{-}} \tag{2.6}$$

$$\mathrm{Cr^{3+} + 3SO_4^{2-} \rightleftharpoons [Cr(SO_4)_3]^{3-}} \tag{2.7}$$

$$\mathrm{2Cr^{3+} + 3SO_4^{2-} \rightleftharpoons [Cr_2(SO_4)_3]^{0}} \tag{2.8}$$

$$\mathrm{2Cr^{3+} + OH^{-} + 3SO_4^{2-} \rightleftharpoons [Cr_2(OH)(SO_4)_3]^{-1}} \tag{2.9}$$

The reaction of a metal cation (M^{m+}) with an anion (A^{a-}) to form a complex can be generally represented as in Eq. (2.10):

$$\mathrm{M^{m+}{}_{(aq)} + xA^{a-}{}_{(aq)} \rightleftharpoons M(A)_x{}^{(ax-m)-}{}_{(aq)}} \tag{2.10}$$

The global stability constant for the formation of the metal–anion complex can be represented by Eq. (2.11):

$$\beta_{M(A)_x^{(ax-m)^-}} = \frac{\gamma_{M(A)_x^{(ax-m)^-}} \cdot [M(A)_x^{(ax-m)^-}]}{\gamma_{M^{m+}} \cdot [M^{m+}] \cdot \gamma_A \cdot [A^{a-}]^x} \tag{2.11}$$

where, $\beta_{M(A)_x^{(ax-m)^-}}$ is the standard thermodynamic constant and γ is the activity coefficient of the ionic species (M^{m+}, A^{a-}, or $M(A)_x^{(ax-m)-}$). The stability constant is governed by the reaction conditions, and under defined conditions, it is expected that Cr(III)–sulfate complexes will be preferentially formed in the following order: $[Cr_2(OH)(SO_4)_3]^{-1}$ (log $\beta_{Eq.2.9}$ = 11.1) [51] > $[Cr_2(SO_4)_3]^0$ (log $\beta_{Eq.2.8}$ = 3.5) [51] > $[Cr(SO_4)_3]^{3-}$ (log $\beta_{Eq.2.7}$ = 2.5) [51] > $[Cr(HSO_4)]^{2+}$ (log $\beta_{Eq.2.4}$ = 2.1) [51] > $[Cr(SO_4)_2]^{-}$ (log $\beta_{Eq.2.6}$ = 2.0) [51] > $[Cr(SO_4)]^{+}$ (log $\beta_{Eq.2.5}$ = 1.3) [51] . However, the low %E of Cr(III) (< 21 %) in all the evaluated ABS and pH levels can be attributed to the predominant formation of the cationic complexes ($[Cr(HSO_4)]^{2+}$ or $[Cr(SO_4)]^{+}$), which preferably interact with species present in the ERP and are electrostatically repulsed when interacting with positively charged chemical species in the MRP. Da Silva and Loh [52] suggested the existence of a pseudopolycation on the MRP that is formed during the phase-separation process. Based on microcalorimetric measurements, the authors proposed that the ABS are formed via specific interactions between the cations and macromolecule segments in a process driven by enthalpic and entropic increases in the system. Thus, before phase separation in the ABS, the Gibbs free energy minimization due to salt-polymer interaction is attributed to the system entropic increase which occurs due to the release of the water of solvation from the ions and macromolecules. However, at specific temperatures and concentrations, this ion–segment interaction is blocked when the macromolecules become energetically saturated, preventing the entropic gain obtained from the increase in the translational degrees of freedom of the water. At this saturation point, phase separation necessarily occurs to allow a reduction in the system Gibbs free energy of the ABS. The authors suggested that, after phase separation, the polymer-rich phase contains macromolecules with adsorbed cations, resulting in a positively charged polymer surface (a pseudopolycation), which can interact with negatively charged species [52]. If the ABS is composed of copolymers, the formation of micelles occurs, as previously described, and the pseudopolycations are formed in the hydrophilic crowns of the PEO units.

On the other hand, the preferential transfer of Cr(VI) from the ERP to the MRP can be attributed to specific interactions (electrostatic, van der Waals, charge transfer, etc.) between the Cr(VI) species (anionic structures) and pseudopolycation segments.

The Cr(VI) species present in aqueous solution are chromic acid (H_2CrO_4), bichromate ($HCrO_4^-$), dichromate ($Cr_2O_7^{2-}$), and chromate (CrO_4^{2-}) [53-55]. In concentrated acidic media, H_2CrO_4 is the principal species that is deprotonated at pH > 1.0 to form $HCrO_4^-$, which is present depending on the concentration and system pH. It is well known that above 10^{-3} mol L^{-1} of total chromium $HCrO_4^-$ starts to condense, yielding $Cr_2O_7^{2-}$ and that above pH 7, $HCrO_4^-$ is converted to CrO_4^{2-} (Eq. (2.12)–(2.14)) [53-55]:

$$H_2CrO_4 \rightleftharpoons HCrO_4^- + H^+ \tag{2.12}$$

$$HCrO_4^- \rightleftharpoons CrO_4^{2-} + H^+ \tag{2.13}$$

$$2\ HCrO_4^- \rightleftharpoons Cr_2O_7^{2-} + H_2O \tag{2.14}$$

According the discussion above, all the Cr(VI) species are anions at pH 2.0, suggesting that all the anionic chromium species could interact electrostatically with the pseudopolycation surface, leading to high %E values. However, there is a large range of extraction yields for Cr(VI), with a minimum %E of 18.1 % for L64 + Li_2SO_4 + H_2O and a maximum %E of 92.7 % for PEO1500 + Na_2SO_4 + H_2O. To explain this extreme extraction behavior for Cr(VI), we must consider that in the different ABS there are distinct relative concentrations of Cr(VI) species and the Cr(VI)–pseudopolycation interactions depend on the chromium anion structure. To verify the dependence of %E on the Cr(VI) anion structure, UV-Vis molecular absorption spectra of the Cr(VI) species in the macromolecule- and electrolyte-rich phases of three ABS [L64 + Li_2SO_4 + H_2O (TLL = 41.86 % (m/m)), L64 + Na_2SO_4 + H_2O (TLL = 47.82 % (m/m)), and PEO1500 + Na_2SO_4 + H_2O (TLL = 46.97 % (m/m))] at pH 2.0, 5.0 and 11.0 were obtained. Fig. 2.3 shows the electronic spectra of the Cr(VI) species at various pH and the phases in which its concentration is higher due to preferential extraction. The UV-Vis spectra in Fig. 2.3A were obtained in the MRP at pH 2.0, whereas in Fig. 2.3B, the spectra were obtained in the ERP at pH 11.0, for the same systems.

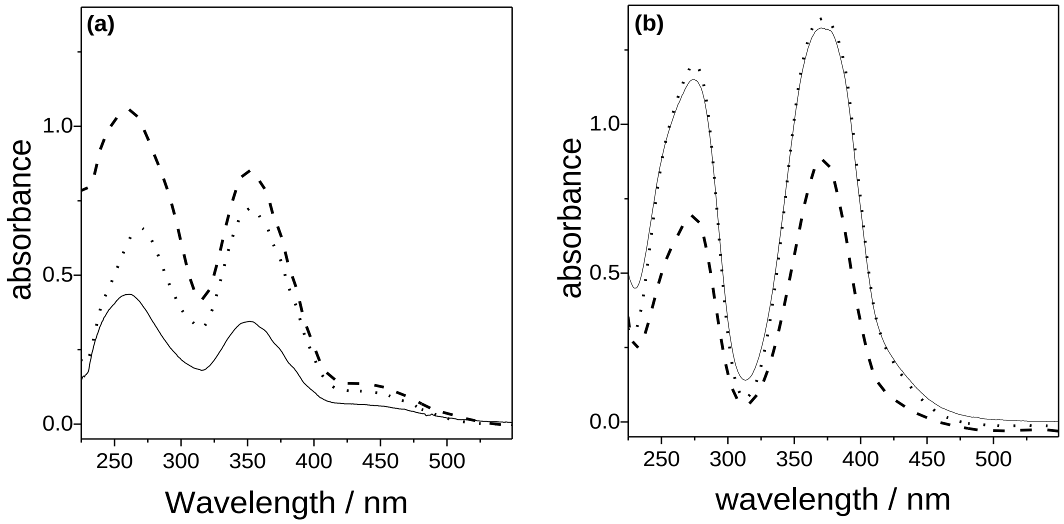

Fig. 2.3. UV-Vis molecular absorption spectra of chromium species in (A) macromolecule-rich phase at pH 2.0; and (B) electrolyte-rich phase at pH 11.0. (....) L64 + Li_2SO_4 + H_2O, TLL = 41.86 % (m/m); (▬) L64 + Na_2SO_4 + H_2O, TLL = 47.82 % (m/m); (– – –) PEO1500 + Na_2SO_4 + H_2O, TLL = 46.97 %(m/m)); [metal] = 15.0 mg kg^{-1}; T = 25.0°C.

According to the literature, the CrO_4^{2-} spectrum consists of two bands with maximum absorbance wavelengths (λ_{max}) at 273 and 371 nm, with the band at shorter wavelength having lower intensity [53,56-58]. The spectrum for $HCrO_4^-$ also displays two bands, with λ_{max} equal to 255 and 350 nm, although the band at shorter wavelength is more intense [53,56-58]. Our spectroscopic data shows that $HCrO_4^-$ is the predominant species in the MRP in the ABS L64 + Na_2SO_4 + H_2O and PEO1500 + Na_2SO_4 + H_2O, since they exhibited bands around 261 and 353 nm with more intense first bands. In the MRP of the system constituted by L64 + Li_2SO_4 + H_2O, two bands with similar intensities and λ_{max} values intermediate (270 and 353 nm) to those observed for the pure $HCrO_4^-$ and CrO_4^{2-} species were observed. As the pH increased, the %E values decreased and the $HCrO_4^-/CrO_4^{2-}$ equilibrium was simultaneously shifted toward CrO_4^{2-} (Fig. A1 to A3 - Appendix), demonstrating that the presence of the $HCrO_4^-$ species determines the higher %E for Cr(VI) due its stronger interaction with the pseudopolycation surface. In the ERP at pH 11.0, two well-defined bands with λ_{max} near 274 and 371 nm were observed, enabling the conclusion that CrO_4^{2-} is the principal species in this ABS phase. Thus, the small %E values obtained at some pH levels can be explained by considering that $HCrO_4^-$ is converted to CrO_4^{2-}, which weakly interacts with the pseudopolycation and remains in the ERP, reducing the %E. In addition, the preferential concentration of CrO_4^{2-} in the electrolyte-rich phase can be attributed to the favorable interaction of these ions with the Na^+ cations and to the higher system entropy produced when this anion is present in this phase [32]. Thus, the decrease in the %E for Cr(VI) with the increase of the medium pH in the ABS consisting of L64 + Li_2SO_4 + H_2O, L64 + Na_2SO_4 + H_2O and PEO1500 + Na_2SO_4 + H_2O can also be attributed to the conversion of $HCrO_4^-$ to CrO_4^{2-} and its unfavorable interaction with the pseudopolycation surface.

In order to confirm this EO-chromium interaction dependence on the anion structure is important to determine its biding constant. Fluorescence spectroscopy is a strategic technique for investigating the intermolecular interactions between the EO segments and chromium species. There are powerful fluorescence methods based on the measurement and interpretation of the quenching of fluorophores under controlled experimental conditions. One of the most used approaches to determine binding parameters is the Benesi-Hildebrand method. In this approach, the fluorophore fluorescence intensity is reduced by the presence of a quencher that interacts with the fluorophore. The magnitude of this emission intensity decrease expresses the extent of the intermolecular interaction. In our specific case, the fluorophore consisted of PEO1500, as shown in Fig. 2.4.

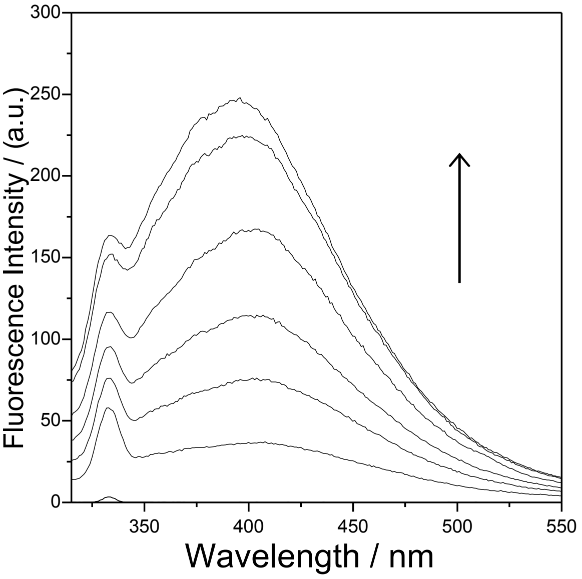

Fig. 2.4. Emission spectra of different concentrations of PEO1500 (1.14, 2.27, 3.41, 4.55, 5.68 and 6.82 mol kg^{-1} of EO segments) in Na_2SO_4 medium (0.103 mol kg^{-1}). The arrow indicates the increasing concentration of PEO1500.

Fig. 2.4 presents the typical fluorescence emission spectra of PEO1500 at different concentrations (1.14, 2.27, 3.41, 4.55, 5.68 and 6.82 mol kg^{-1} of EO segments) in Na_2SO_4 medium (0.103 mol kg^{-1}). The fluorescence spectrum consists of two bands: one around 335 nm attributed to the raman scattering of water [59] and another near 395 nm, associated with the PEO1500 [60].

Paik et al. [60] showed for the first time that aqueous solutions of PEO of different molecular weights were capable of producing fluorescence emissions when excited near 300 nm. The fluorescence emission was attributed to the structural orientation of the double-ended hydrophilic head groups which arrange themselves in vesicular organization and offer electron-dense regions [60]. The authors also discovered that the addition of metal ions to these polymeric solutions can modify the polymer structure, separating it into monomeric segments, and hence, decreasing the fluorescence intensity.

Fig. 2.5 shows the maximum fluorescence intensity as function of PEO1500 concentration, expressed in relation to the EO segment concentration in Na_2SO_4 medium in the presence and absence of Cr(VI) species at pH 2.0 ($HCrO_4^-$). Similar results were obtained at pH 11.0 (Fig. A4 - Appendix).

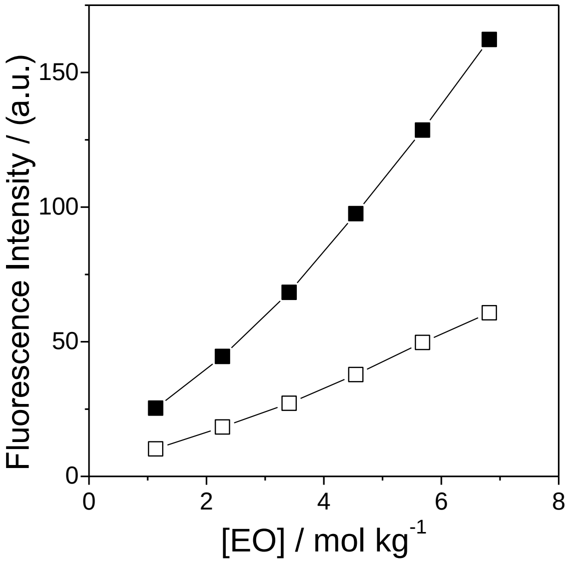

Fig. 2.5. Effect of Cr(VI) on the fluorescence intensity of the polymer PEO1500. [Na_2SO_4] = 0.103 mol kg^{-1}; [Cr (VI)] = 5.00 × 10^{-4} mol kg^{-1}; pH = 2.0. (■) in the absence of Cr(VI); (□) in the presence of Cr(VI).

The results in Fig. 2.5 and Fig. A4 show that the $HCrO_4^-$ species at pH 2.0 quenches the PEO1500 fluorescence intensity, suggesting an interaction between the polymer and chromium species. The PEO–Cr(VI) species ($HCrO_4^-$ at pH 2.0 and CrO_4^{2-} at pH 11.0) binding constants can be calculated using Eq. (2.3), by considering the interaction as a 1:1 ligand type (i.e., Cr(VI) to binding site (EO segment)). The double reciprocal plots of $1/\Delta F$ as a function of $1/[EO]$ for PEO1500 are shown in Fig. 2.6.

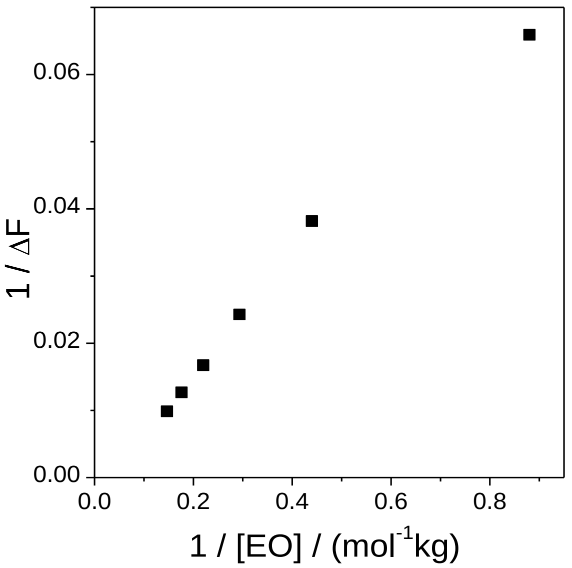

Fig. 2.6. Double-reciprocal graph of the concentration of EO segments versus the difference in fluorescence intensity between the sample without and with Cr(VI). [Na_2SO_4] = 0.103 mol kg^{-1}; [Cr(VI)] = 5.00 × 10^{-4} mol kg^{-1}; pH = 2.0.

A simple linear regression was applied to the data in Fig. 2.6 to estimate the binding parameters. The parameter ΔF_{max} was estimated using the linear regression coefficient, and the value of the binding constant, K_b, for the EO–Cr(VI) interaction was calculated through the angular regression coefficient (Table 2.1).

Table 2.1. Binding constants (K_b) and maximum fluorescence intensities (ΔF_{max}) for the binding of chromium species to EO segments.

Sample	ΔF_{max}	K_b (µmol^{-1} kg)	R^2
Cr(VI) pH 2.0 ($HCrO_4^-$)	2.06×10^3	6340	0.985
Cr(VI) pH 11.0 (CrO_4^{2-}).	3.96×10^3	4250	0.997

Researchers have previously demonstrated the existence of interaction between curcumin and casein micelles [43,61]. Yazdi and Corredig [43] determined the K_b for binding of curcumin to casein micelles in different conditions and obtained K_b values as 0.22 ± 0.02 and 0.26 ± 0.01 µmol^{-1} L. In the present work, constants with greater magnitude order was obtained. The obtained binding constant reflects the ratio between the EO segment–Cr(VI) complexes and the free concentrations of EO segments and Cr(VI) species. The higher K_b value for the Cr(VI)–EO interaction at pH 2.0 (6340 µmol^{-1} kg) compared to pH 11.0 (4250 × µmol^{-1} kg) suggests a higher binding equilibrium for $HCrO_4^{-}$ than CrO_4^{2-}. This preferential interaction between EO–$HCrO_4^{-}$ is the cause of the higher %E at pH 2.0 than pH 11.0.

It was observed that the addition of Cr(III) species did not suppress the PEO1500 fluorescence intensity. This suggests a weak interaction between the Cr(III) and EO segments of the macromolecule, and thus, the poor transfer of this chromium species to the MRP. Due the small effect of Cr(III) on the PEO1500 fluorescence intensity, it was not possible to determine the binding parameters for this chromium specie.

As at pH 2.0, the predominant species is $HCrO_4^{-}$, which is spontaneously concentrated in the MRP and affords a higher %E for the Cr(VI) species, this pH was adopted for use in further experiments. In addition to pH, other properties of ABS phases (e.g., their composition and type of components) can modulate the interaction between $HCrO_4^{-}$ and the pseudopolycation, which will determine the distribution of the chromium species in the ABS. Therefore, these factors were also evaluated to determine their effects on chromium speciation.

2.4.3. Influence of ABS TLL values on the extraction of chromium species

The preferential partitioning of chemical species between ABS phases is due to the different intensive thermodynamic properties in the macromolecule- and electrolyte-rich phases, thus, promoting different intermolecular interactions between the species that will be partitioned and ABS components. In general, a parameter used to express the difference between the intensive thermodynamic properties of the phases is the TLL. The TLL is a parameter that numerically expresses the composition difference between two phases and it is calculated as a function of the concentration differences of the components in each phase of the ABS, as indicated in Eq. (2.15) [62]:

$$\mathrm{TLL} = \left[\left(C_{\mathrm{P}}^{\mathrm{MRP}} - C_{\mathrm{P}}^{\mathrm{ERP}}\right)^2 + \left(C_{\mathrm{E}}^{\mathrm{MRP}} - C_{\mathrm{E}}^{\mathrm{ERP}}\right)^2\right]^{1/2} \tag{2.15}$$

where $C_{\mathrm{P}}^{\mathrm{MRP}}$ and $C_{\mathrm{P}}^{\mathrm{ERP}}$ are the polymer concentrations, % (m/m), in the macromolecule- and electrolyte-rich phases, respectively, and $C_{\mathrm{E}}^{\mathrm{MRP}}$ and $C_{\mathrm{E}}^{\mathrm{ERP}}$ are the corresponding electrolyte concentrations, % (m/m), in the macromolecule- and electrolyte-rich phases, respectively.

Fig. 2.7 shows the %E for the chromium species plotted against the TLL in the ABS composed of L64 + Na_2SO_4 + H_2O or L64 + Li_2SO_4 + H_2O. As is generally found, the increase in TLL values improves the extraction efficiency of the analytes in the ABS, i.e., Cr(VI) is more concentrated in the MRP and Cr(III) is increasingly transferred to the ERP. As the increment in TLL values increases the polymer and electrolyte concentrations in the macromolecule- and electrolyte-rich phases (Tables A1-A7), respectively, the driving forces that determine the separation between the chromium species are intensified. Then, the TLL increment leads to ABS condition where Cr(VI) is quantitatively recovery in MRP and Cr(III) remains in ERP.

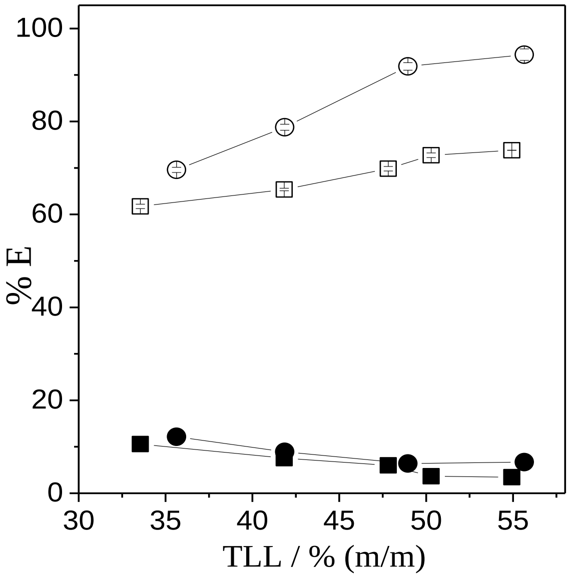

Fig. 2.7. Effect of tie-line length (TLL) on the extraction percentage (%E) of Cr(III) and Cr(VI) using the L64 copolymer. [metal] = 4.00 mg kg^{-1}; T = 25.0°C; pH 2.0. (■) Cr(III), Na_2SO_4; (□) Cr(VI), Na_2SO_4; (●) Cr(III), Li_2SO_4; (○) Cr(VI), Li_2SO_4.

2.4.4. Effect of hydrophobic/hydrophilic balance of the macromolecules on the extraction of chromium species

The partitioning of $HCrO_4^-$ and Cr(III) from the ERP to the MRP might also be influenced by hydrophobic interactions. To verify the hydrophobic driving force for this ion transfer, partitioning experiments were performed with distinct ABS formed by macromolecules of different hydrophobic/hydrophilic balances. The ABS formed with L64 has a more hydrophobic macromolecular phase because its molecule structure have a large content of poly(propylene oxide) which is water insoluble and it is capable of solubilizing compounds that are insoluble in water, whereas systems formed using PEO1500 have hydrophilic phases. Fig. 2.8 shows the %E of chromium species as a function of the TLL in ABS with different hydrophobicities.

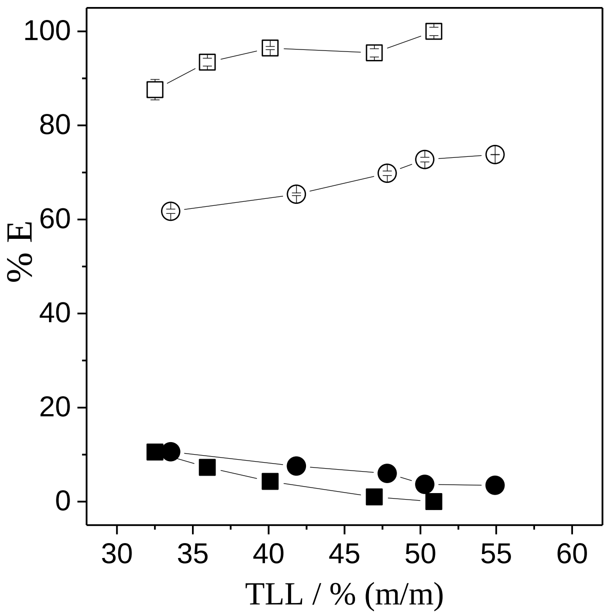

Fig. 2.8. Influence of the hydrophobic/hydrophilic balance of the macromolecules on the extraction percentage (%E) of Cr(III) and Cr(VI) in ABS formed with Na_2SO_4 and PEO1500 or L64. [metal] = 4.00 mg kg^{-1}; T = 25.0°C; pH 2.0. (■) Cr(III), PEO1500; (●) Cr(III), L64; (□) Cr(VI), PEO1500; (○) Cr(VI), L64.

The %E of Cr(III) was little affected by differences in the hydrophobicity of the ABS. Cr(III) was preferentially concentrated in the ERP in the ABS formed by either PEO or L64. In contrast, the Cr(VI) extraction behavior was dependent on the structure of the macromolecules that formed the ABS. At all TLL values, the transfer of Cr(VI) ions from the ERP to the MRP was higher in the ABS formed with PEO than in the L64 systems.

The ABS constituted with PEO afforded the maximum %E for Cr(VI) of 99.9 % at a greater TLL, 50.89 % (m/m), whereas, for the ABS formed with L64, the maximum %E for Cr(VI) of 73.8 % was obtained for a higher TLL, 54.93 % (m/m). This difference in the Cr(VI) extraction processes in the two ABS can be justified on the basis of the small content of EO groups in L64 (only 40 wt%), thus, the number of sites for interactions between the Cr(VI) and EO segments in such systems is lower than in ABS consisting of PEO.

2.4.5. Influence of electrolyte nature on the extraction of chromium species

Previous studies have shown that ion-partitioning behaviors in different ABS are very dependent on the nature of the electrolyte [29,44,63]. These electrolyte effects can be attributed to the different interactions between the analytes and the ABS-forming cations and anions. Besides the effect of these ion–ion interactions on the ion–ABS distribution, distinct ions can promote different interactions between the partitioned metallic ion and the EO segments, because distinct cation produce pseudopolycations with different positive charge densities.

Fig. 2.9A shows the %E for Cr(III) and Cr(VI) versus the TLL values for systems formed by PEO1500 + Li_2SO_4 + H_2O, PEO1500 + Na_2SO_4 + H_2O, and PEO1500 + $MgSO_4$ + H_2O. The smallest TLL value of the PEO1500 + Li_2SO_4 + H_2O ABS (29.43 % (m/m)) was responsible for the highest %E values for Cr(VI) and Cr(III) (99.9 % and 12.9 %, respectively). Because of the efficient extraction of both chromium species, this ABS would be inappropriate for chromium speciation. On the other hand, in the PEO1500 + Na_2SO_4 + H_2O ABS, the highest TLL (50.89% (m/m)) afforded the lowest %E for Cr(III) (0.0100 %) and the maximum %E for Cr(VI) (99.9 %), which makes this ABS the better choice for Cr(III) and Cr(VI) separation. Unfortunately, in the PEO1500 + $MgSO_4$ + H_2O ABS, independent of the TLL, Cr(VI) extractions were lower, whereas the extraction behaviors of Cr(III) for all TLL were similar to the extractions in the PEO1500 + Na_2SO_4 + H_2O ABS, which makes this ABS inefficient for chromium speciation.

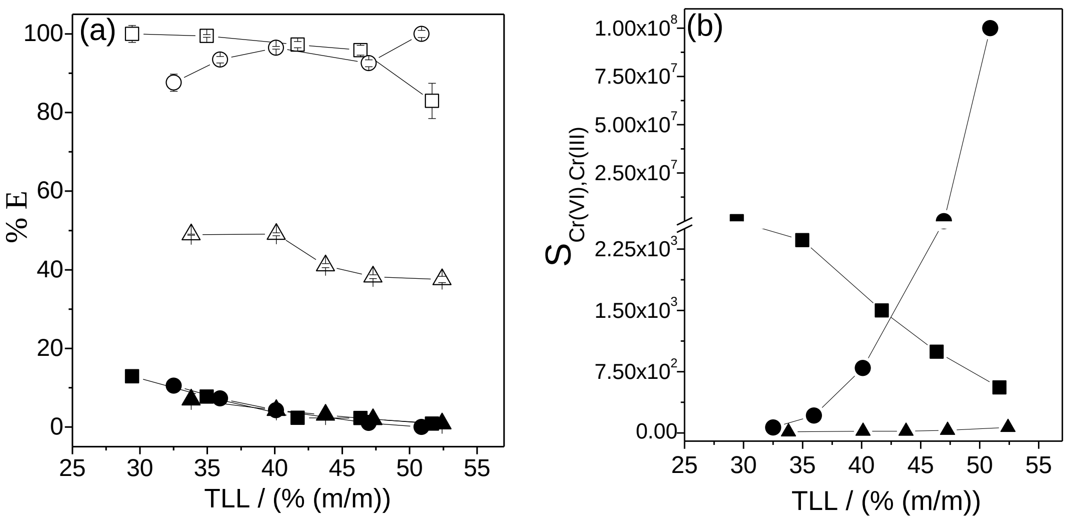

Fig. 2.9. The influence of the ABS-forming cation on the extraction percentage (%E) of Cr(III) and Cr(VI) in the PEO1500-based ABS. [metal] = 4.00 mg kg^{-1}; T = 25.0°C; pH 2.0. (A) Percentage extraction of the chromium species: (■) Cr(III), Li_2SO_4; (●) Cr(III), Na_2SO_4; (▲) Cr(III), $MgSO_4$; (□) Cr(VI), Li_2SO_4; (○) Cr(VI), Na_2SO_4; (Δ) Cr(VI), $MgSO_4$; and (B) Chromium species separation factor: (■) Li_2SO_4; (●) Na_2SO_4; (▲) $MgSO_4$.

This cation effect on the Cr(VI)/Cr(III) separation can be understood based on the microcalorimetric results of da Silva and Loh [52]. These results demonstrated that PEO – cation interaction intensity and consequently chains have a positive charge density follow the order: Li_2SO_4 > Na_2SO_4 > $MgSO_4$. Thus, at the lower TLL values (29.43% (m/m)) in the ABS formed by Li_2SO_4, the pseudopolycation establishes strong attractive electrostatic interactions with the Cr(III) species (anionic sulfate complex) and the Cr(VI) species ($HCrO_4^-$), providing higher %E values for both species. Consequently, this ABS does not offer the best environment for the selective extraction of Cr(VI). The ABS formed by $MgSO_4$ leads to the formation of a pseudopolycation with a low positive charge density, which reduces its interactions with the Cr(III) and Cr(VI) anionic species. In contrast, the pseudopolycations formed in systems containing Na_2SO_4 have an intermediate positive charge density that favors their interaction with the $HCrO_4^-$ ions, but it is insufficient to effectively interact with the anionic species of Cr(III) and extract them from the ERP into the MRP.

The separation factors for Cr(VI) and Cr(III) ($S_{\mathrm{Cr(VI),Cr(III)}}$) promoted by Li_2SO_4, Na_2SO_4, and $MgSO_4$ as function of TLL are presented in Fig. 2.9B. The separation factor is given by Eq. (2.16) [38]:

$$S_{Cr(VI),Cr(III)} = \frac{D_{Cr(VI)}}{D_{Cr(III)}} \tag{2.16}$$

where $D_{Cr(VI)}$ and $D_{Cr(III)}$ are the distribution coefficients of Cr(VI) and Cr(III) in the ABS. The distribution coefficient can be expressed by Eq. (2.17) [38]:

$$D_{Cr} = \frac{\%E}{100 - \%E} \tag{2.17}$$

According to Fig. 2.9B, the most selective extraction of Cr(VI) with respect to Cr(III) occurs in the ABS consisting of PEO1500 + Na_2SO_4 + H_2O, TLL = 50.89 % (m/m), with an exceptional separation factor value, $S_{\mathrm{Cr(VI)/Cr(III)}} = 1.00 \times 10^8$. Based on literature data, for an efficient separation of metal ions, it is sufficient that the separation factor value has a magnitude of at least 10^3 [38]. Thus, considering economic and environmental aspects, other systems that use smaller electrolyte and polymer quantities could be chosen for chromium speciation, especially the systems formed by PEO1500 + Na_2SO_4 + H_2O (TLL = 46.97 % (m/m)) with $S_{\mathrm{Cr(VI)/Cr(III)}} = 5.82 \times 10^4$, PEO1500 + Li_2SO_4 + H_2O (TLL = 29.43 % (m/m)) with $S_{\mathrm{Cr(VI)/Cr(III)}} = 6.76 \times 10^4$, and PEO1500 + Li_2SO_4 + H_2O (TLL = 34.97 % (m/m)) with $S_{\mathrm{Cr(VI)/Cr(III)}} = 2.36 \times 10^3$.

To explore the contribution of the anion nature to the %E of the chromium species, ABS formed by a common cation (Na^+) and different anions were studied in the chromium speciation process. Fig. 2.10A illustrates the %E of chromium species versus the TLL for systems consisting of PEO1500 + $C_6H_5Na_3O_7$ + H_2O and PEO1500 + $C_4H_4Na_2O_6$ + H_2O.

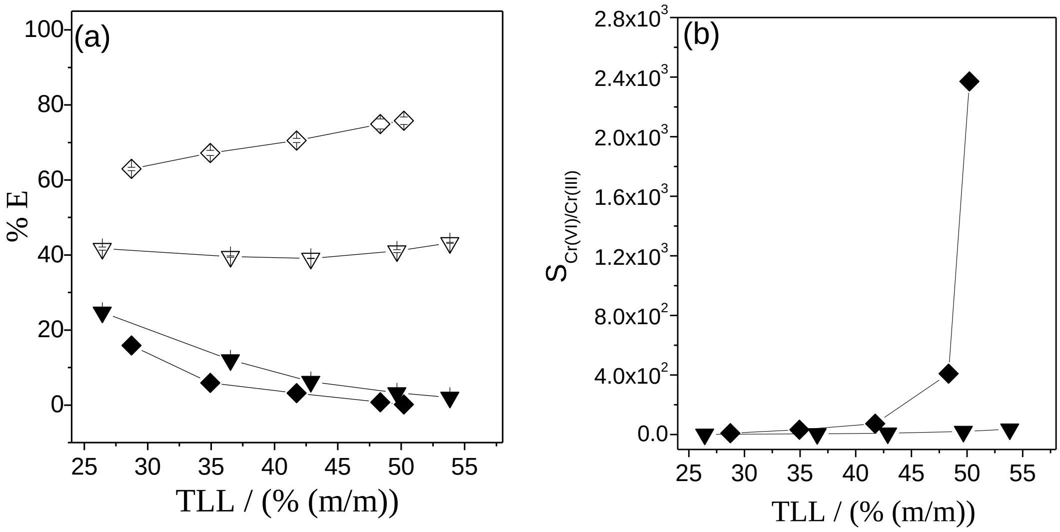

Fig. 2.10. The influence of the ABS-forming anion on the extraction percentage (%E) of Cr(III) and Cr(VI) by. PEO1500 + organic salt + water. [metal] = 4.00 mg kg^{-1}; T = 25.0°C; pH 2.0. (A) (♦) Cr(III), $C_4H_4Na_2O_6$; (▼) Cr(III), $C_6H_5Na_3O_7$; (◊) Cr(VI), $C_4H_4Na_2O_6$; (∇) Cr(VI), $C_6H_5Na_3O_7$; (B) Separation factor values for metal ions in systems formed by: (♦) $C_4H_4Na_2O_6$; (▼) $C_6H_5Na_3O_7$.

The ABS formed with sodium citrate ($C_6H_5Na_3O_7$) or tartrate ($C_4H_4Na_2O_6$) were less efficient in Cr(VI)/Cr(III) separation than those formed with Na_2SO_4, mainly because the ABS constituted by these organic electrolytes achieved lower %E for Cr(VI) and higher %E for Cr(III). As a consequence, lower separation factor values were obtained (Fig. 2.10B). Under the best separation conditions for the PEO1500 + $C_6H_5Na_3O_7$ + H_2O ABS, TLL = 53.84 % (m/m), a small $S_{Cr(VI)/Cr(III)} = 36.6$ ($\%E_{Cr(III)} = 2.04$ % and $\%E_{Cr(VI)} = 43.2$ %) was obtained. The highest separation factor, $S_{Cr(VI)/Cr(III)} = 2.37 \times 10^3$ ($\%E_{Cr(III)} = 0.132$ % and $\%E_{Cr(VI)} = 75.8$ %), was obtained in the ABS formed by PEO1500 + $C_4H_4Na_2O_6$ + H_2O, TLL = 50.21 % (m/m).

The lower %E of Cr(VI) in the ABS formed with the organic electrolytes than in the ABS composed of sulfate salts can be attributed to the presence of CrO_4^{2-} in the ERP of the PEO1500 + organic electrolyte + water ABS, as shown by the electronic spectra of the ERP (Fig. 2.11).

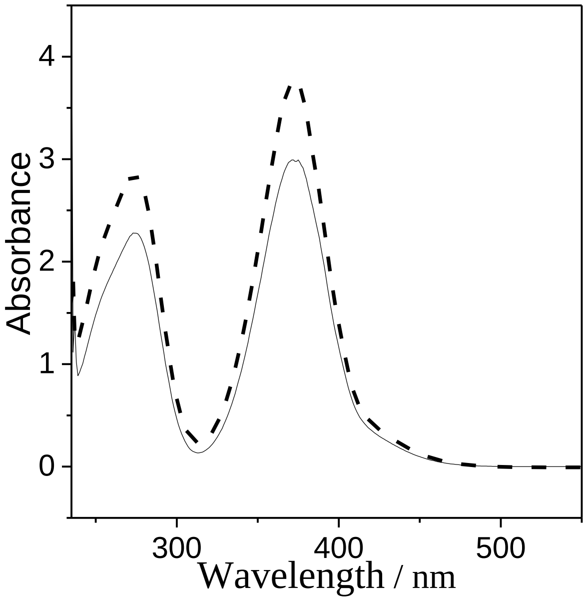

Fig. 2.11. UV-Vis molecular absorption spectra of Cr(VI) species in the electrolyte-rich phase of the ABS composed of PEO1500. [metal] = 40.00 mg kg^{-1}; T = 25.0°C; pH 2.0. (---) CrO_4^{2-}, $C_4H_4Na_2O_6$ (TLL = 50.21% (m/m)) and (▬) CrO_4^{2-}, $C_6H_5Na_3O_7$ + H_2O (TLL = 53.84% (m/m)).

As discussed in section 2.4.2, CrO_4^{2-} concentrates in the ERP due its weak interaction with the pseudopolycation surface. The Cr(III) extraction behavior is related to the formation of $Cr(A)_x^{(ax-3)-}$ complexes (where A = citrate or tartrate) with tartrate ($\log \beta_1 = 2.267$, $\log \beta_2 = 2.0$, $\log \beta_3 = 3.59$) [64] and citrate anions ($\log \beta_1 = 19.01$, $\log \beta_2 = 16.34$, $\log \beta_3 = 12.54$, $\log \beta_4 = 8.18$, $\log \beta_5 = 2.56$, $\log \beta_6 = -3.51$, $\log \beta_7 = 6.13$, $\log \beta_8 = -13.30$) [65].

2.5. CONCLUSION

In the present work it was demonstrated that ABS formed by macromolecule and an electrolyte is capable of separating Cr(III) and Cr(VI) without the use of organic solvents or any extractant. The Cr(III) was partitioned to the electrolyte-rich phase, while the Cr(VI) was preferentially concentrated in the macromolecule-rich phase in the many analyzed systems. The fluorescence spectroscopy results showed that the Cr(III) and Cr(VI) separation process is based on the specific interactions that occur between the EO segments of the polymers and the Cr(VI) species, which are not observed for Cr(III). The proposed system is a green, simple, and fast liquid–liquid extraction process to separate chromium species.

The extraction behaviors of Cr(III) and Cr(VI) were affected by pH, TLL, polymer hydrophobicity and the electrolyte, because these parameters modulate the chromium–EO interactions. Although all the ABS used in this study showed the potential for the speciation of chromium species, the system comprising PEO1500 + Na_2SO_4 + H_2O (TLL = 50.89% (m/m)) at pH 2.0 afforded the highest $S_{Cr(VI)/Cr(III)}$ value, 1.00×10^8, with $\%E_{Cr(VI)} = 99.9\%$ and $\%E_{Cr(III)} = 0.0100\%$.

The use of the ABS can be extended to the speciation of other metal ions and the PEO1500 + Na_2SO_4 + H_2O ABS can be applied in the quantification of the chromium species.

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CAPÍTULO 3

Aqueous biphasic system combined with flame atomic absorption spectrometry for chromium speciation in wastewater samples

3.1. ABSTRACT

The wide use of chromium species in industrial processes and the Cr(VI) toxicity has stimulated the chromium speciation analysis. However, several analytical techniques only quantify the total chromium concentration requiring tedious steps of sample preparation in chromium speciation analysis. In the present work, it was developed a new and efficient analytical method for Cr(III) and Cr(VI) determination using aqueous biphasic system (ABS). Cr(III) and Cr(VI) species were separated and, or pre-concentrated by PEO1500 + Na_2SO_4 + H_2O ABS at pH 2.0, 25.0 °C and TLL = 46.97 % (m/m) and flame atomic absorption spectrometry (FAAS) was used to determine the species. The proposed method was validated by Cr(III) and Cr(VI) determination in water samples of tap, treatment plant, river and electroplating waste. Recovery percentages between 87.9 ± 3.7 and 107 ± 4 were obtained. Under the optimum experimental conditions were achieved limit of detection 0.0538 mg kg^{-1}, limit of quantification 0.163 mg kg^{-1}, dynamic range 0.0500 - 0.500 or 0.100 - 4.00 mg kg^{-1}, precision 0.802 % for Cr(VI) and 3.53 % for Cr(III) and accuracy 13.6%.

Keywords: chromium speciation, aqueous biphasic system, pre-concentration

3.2. INTRODUCTION

It has been recognized that the impact of heavy metals on the ecological system, biological organisms, as well as human health, not only is determined by the total amount of the element, but also is governed by its chemical forms [1]. Consequently, there is a great interest in speciation analysis. Speciation analysis, according IUPAC definition, are analytical activities of identifying and/or measuring the quantities of one or more individual chemical species in a sample [2].

The chromium element exists mainly as Cr(III) and Cr(VI) and they are extensively used in industrial processes such as metallurgical, chemical industries, electroplating, refractories, pigments and leather tanning, so chromium compounds are released into the water bodies [3-5]. Effective speciation analysis is usually indispensable to differentiate chromium species since the two oxidation states**,** Cr(III) and Cr(VI), are drastically different in toxicity, reactivity, bioavailability and biotransformation [3,4]. While Cr(III) is relatively non-toxic and an essential nutrient for humans, Cr(VI) species are potentials carcinogenic agents and it is also harmful to lungs, liver and kidney [1,6]. In this context, Brazilian National Council for the Environment (CONAMA), recommended the maximum allowable concentrations of Cr(III) and Cr(VI) in effluent discharges in aquatic bodies of 1.0 mg L^{-1} and 0.1 mg L^{-1}, respectively [7]. Due the Cr(VI) carcinogenicity, the World Health Organization (WHO) [8] proposed the current guideline value of 0.05 mg L^{-1} for Cr(VI) in drinking water whereas the US Environmental Protection Agency (EPA) [9] recommend a total chromium concentration in drinking water of 0.1 mg L^{-1} and CONAMA determined a total chromium concentration in drinking water of 0.05 mg L^{-1} [7].

In usual conditions, there is no selectivity for different oxidation states of elements by traditional instrumental analysis for metallic ions as flame atomic absorption spectrometry (FAAS) [10], electrothermal atomic absorption spectrometry (ETAAS) [11], inductively coupled plasma optical emission spectrometry (ICP-OES) [12,13] and inductively coupled plasma–mass spectrometry (ICP–MS) [6]. These techniques only determine the total quantity of the chemical elements. Then, to perform a chromium speciation analysis it is necessary to do many tedious steps of sample preparation as solid-phase extraction [14,15], cloud point extraction [16], co-precipitation [17], and liquid-liquid extraction (LLE) [11,18-20] or using expensive technique like HPLC-ICP-MS [21,22]. Among them, LLE has attracted substantial interest in the separation and pre-concentration of substances because of its advantages as cost, simplicity and the use of different solvents and extractant allowing that several analytes are separated and, or pre-concentrated [23]. Although of wide use of LLE, it poses several problems as slow phase separation when emulsions are formed, the use of large sample volumes and toxic organic solvents that makes LLE an expensive, time consuming and environmentally unfriendly technique [24].

An environmentally safe alternative to LLE is the aqueous biphasic system (ABS) or aqueous two-phase systems (ATPS). ABS has the same advantages of LLE, but not has many drawbacks. This system is formed by two immiscible aqueous-rich phases based on the combination of polymer–polymer, polymer–electrolyte or electrolyte–electrolyte under specific conditions of temperature, pressure and concentration [25]. The use of nontoxic, nonflammable and biodegradable components, low-cost and rapid phase separation are some advantages of the ABS technique [26,27].

In 2016, Simonova et al. [28] proposed a method for the chromium speciation species in waste water samples using ABS. The authors employed specific extractants for both chromium species to obtain satisfactory extractions of Cr(III) and Cr(VI). To speciate the chromium species, initially, the authors proposed the use of 1,5-diphenylcarbazide extractant since it makes a selective complexation and extraction of Cr(VI) to the polymer-rich phase. They also proposed the use of 4-(2-pyridylazo)-resorcinol as selective extractant for Cr(III) to determine Cr(III) concentration in the presence of Cr(VI). The authors suggested that besides polymer-salt ABS, the speciation can be performed in systems formed by organic solvents (ethanol and isopropanol) and electrolyte (ammonium sulfate). In our previous work, the driving force behind the chromium species partition in ABS systems was reported [29]. We demonstrated that ABS composed of polymer and an electrolyte is capable of separating Cr(III) and Cr(VI) without the use of organic solvents or any extractant. This discovery represents an economical and environmental gain.

Thus, in the present work was developed a chromium speciation analytical method, without the use of organic solvents or any extractant, using ABS consisting of poly(ethylene oxide) polymer with average molar mass of 1500 g mol^{-1} (PEO1500), Na_2SO_4 electrolyte and water. To determine low concentrations of Cr(III) and Cr(VI) in water samples by the non-selective and low cost technique, FAAS, it was proposed a preliminary ABS separation and pre-concentration procedure. Factors affecting the chromium speciation as interconversion of chromium species, chromium species concentration in the system, effect of water matrices and concomitant ions were investigated. The proposed method is green, robust, simple, inexpensive and allows the speciation of Cr(III) and Cr(VI) in matrices with a wide range of chromium concentration as tap, treatment plant, river and electroplating wastewater using FAAS.

3.3. EXPERIMENTAL

3.3.1. Chemicals

All reagents were of analytical grade quality and were used as received without further purification. Deionized water (Milli-Q, Millipore) was employed in the preparation of all used solutions. The poly(ethylene oxide) polymer with average molar mass of 1500 g mol^{-1} was acquired from Synth (São Paulo, Brazil). The chemicals sodium sulfate and chromium chloride were obtained from Vetec (Rio de Janeiro, Brazil). The reagents potassium dichromate, phosphoric acid, sulfuric acid and the extractant 1,5-diphenylcarbazide were purchase from Sigma-Aldrich (Missouri, USA). Hydrochloric acid was acquired from Merck (Darmstadt, Germany) and acetone was bought from Alphatec (Rio de Janeiro, Brazil).

3.3.2. Instrumentation

Chromium quantification was performed with a flame atomic absorption spectrometer Model 240 (Varian, Australia) or with an UV/visible spectrometer Model UV–2550 (Shimadzu, Japan). All pH values were measured using a pHmeter Hi 221 (Hanna Instruments, Brazil). Analytical balance AY-220 (Shimadzu, Brazil), centrifuge Heraues Megafuge 11R (Thermo Scientific, Germany) and thermostatic bath MQBTC 99-20 (Microquímica, Brazil) were used in the samples preparation.

3.3.3. ABS composition

ABS consisting of PEO1500 + Na_2SO_4 + H_2O at the tie-line length (TLL) equal to 46.97 % (m/m) was used in the chromium speciation method. The ABS liquid–liquid equilibrium data was obtained from literature [30]. The system is formed by a polymer-rich phase (PRP) constituted by 43.41 % (m/m) of PEO1500 and 1.46 % (m/m) of Na_2SO_4 and an electrolyte-rich phase (ERP) composed by 1.03 % (m/m) of PEO1500 and 21.71 % (m/m) of Na_2SO_4.

3.3.4. Liquid–liquid extraction procedure

Stock solutions of PEO1500 and Na_2SO_4 were prepared solubilizing appropriate masses in HCl solution 0.0100 mol L^{-1} (pH = 2.0). The solutions of Cr(III) and Cr(VI) in the desired concentrations (8.00 – 1.20 x 10^3 mg kg^{-1}) were prepared by solubilizing the respective salts in the Na_2SO_4 stock solutions at pH 2.0. In a centrifuge tube were mixed proper quantities of PEO1500 aqueous stock solution with appropriate quantities of chromium species solutions to obtain the desired systems. The blank assays were performed by mixing the proper masses of PEO1500 and Na_2SO_4 stock solutions. The resulting systems were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min to accelerate the phase separation and were placed in a thermostatic bath at 25.0 ºC for 20 minutes to reach the thermal equilibrium. Then, two clear phases were observed and aliquots from the PRP and ERP were collected, diluted, and the concentrations of Cr(III) and Cr(VI) were determined in triplicate by FAAS after the obtainment of analytical curves ($R^2 \geq 0.999$). Chromium ions were determined with a deuterium background correction, an air–acetylene burner at wavelength of 357.9 nm, lamp current of 7 mA and slit width of 0.2 nm.

The percentage extraction (%E) of the metallic ions were calculated by Eq. (3.1)

$$\%\mathrm{E} = \frac{\mathrm{n}_{\mathrm{M}}^{\mathrm{phase}}}{\mathrm{n}_{\mathrm{M}}^{\mathrm{total}}} \mathrm{X}\ 100 \tag{3.1}$$

where n_M^{phase} is the quantity of metallic ions in the PRP or ERP in mol and n_M^{total} is the total number of moles of metallic ions in the system.

3.3.5. Influence of the quantity of chromium species on the %E of Cr(III) and Cr(VI)

Stock solutions of PEO1500 44.4 % (m/m) and Na_2SO_4 23.2 % (m/m) were prepared by solubilizing appropriate masses in HCl solution 0.0100 mol L^{-1} (pH = 2.0). Working Cr(III) and Cr(VI) solutions at concentrations between 8.00 and 1.20 x 10^3 mg kg^{-1} were obtained dissolving the respective salts in Na_2SO_4 stock solution at pH 2.00. 2.00 g of Cr(III) or Cr(VI) working solution were mixed with 2.00 g of PEO1500 stock solution in order to get an ABS with 2.00 g of each phase. The formed ABS were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min and were placed in a thermostatic bath at 25.0 ºC for 20 minutes. Then, two clear phases were observed and aliquots from the PRP and ERP were collected, diluted, and the concentrations of Cr(III) and Cr(VI) were determined in triplicate by FAAS

3.3.6. Interconversion of chromium species in the ABS

Stock solutions of PEO1500 44.4 % (m/m) and Na_2SO_4 23.2 % (m/m) were prepared solubilizing appropriate masses in HCl solution 0.0100 mol L^{-1} (pH = 2.0). Cr(III) and Cr(VI) solutions at 3.20 x 10^3 mg kg^{-1} were prepared dissolving the respective salts in Na_2SO_4 stock solutions. A working solution simultaneously containing Cr(III) and Cr(VI) 16.0 mg kg^{-1} was prepared diluting the chromium solutions at 3.20 x 10^3 mg kg^{-1} with Na_2SO_4 stock solutions. 4.50 g of chromium working solution were mixed with 4.50 g of PEO1500 stock solution. The resulting systems containing 4.50 g of each phase were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min and were placed in a thermostatic bath at 25.0 ºC for 20 minutes. Aliquots of PRP and ERP were collected, diluted and the total quantity of chromium, in mol, in the ERP ($n_{Cr\ total}^{ERP}$) and PRP ($n_{Cr\ total}^{PRP}$) were obtained. The term $n_{Cr\ total}^{ERP}$ is the sum of Cr(III) quantity in ERP ($n_{Cr(III)}^{ERP}$) and Cr(VI) quantity in ERP ($n_{Cr(VI)}^{ERP}$), as shown in Eq.(3.2):

$$n_{Cr\ total}^{ERP} = n_{Cr(III)}^{ERP} + n_{Cr(VI)}^{ERP} \tag{3.2}$$

By colorimetric standard method for Cr(VI) water analysis [31] was possible to determine the Cr(VI) concentration in ERP ($n_{Cr(VI)}^{ERP}$) (PRP was not submitted to the colorimetric standard method for Cr(VI) since it interferes in the analytical method response). By difference, it was determined the Cr(III) quantity in ERP ($n_{Cr(III)}^{ERP}$). Knowing the number of moles of Cr(VI) in ERP it was also possible to determine the content of Cr(VI) in PRP ($n_{Cr(VI)}^{PRP}$) (Eq. (3.3)):

$$n_{Cr(VI)}^{added} = n_{Cr(VI)}^{ERP} + n_{Cr(VI)}^{PRP} \tag{3.3}$$

where $n_{Cr(VI)}^{added}$ is the number of moles of Cr(VI) added in the system.

The $n_{Cr\ total}^{PRP}$ determined in PRP by FAAS can be expressed by Eq. (3.4):

$$n_{Cr\ total}^{PRP} = n_{Cr(III)}^{PRP} + n_{Cr(VI)}^{PRP} \tag{3.4}$$

where $n_{Cr(III)}^{PRP}$ is the number of moles of Cr(III) partitioned to the PRP.

Using the Eq. (3.4) it was calculated the content of $n_{Cr(III)}^{PRP}$. So, by the Eq. (3.1) it was determined the %E of Cr(III) (%$E_{Cr(III)}$) and Cr(VI) (%$E_{Cr(VI)}$) in ERP and PRP when they were simultaneously added to the system. These results were compared with the %E obtained when the chromium species were individually added to the system.

3.3.7. ABS pre-concentration speciation procedure

Synthetic water samples containing both chromium species were prepared. Deionized water at pH 2.00 was spiked with Cr(III) and Cr(VI) at different concentrations (Cr(III) = 0.500 and 1.00 mg kg^{-1}, Cr(VI) = 0.0500 and 0.100 mg kg^{-1}). In vessels were added 0.682 g of PEO1500, 13.1 g of Na_2SO_4 and 46.2 g of synthetic water sample to obtain systems with 1.40 g of PRP and 58.6 g of ERP. The resulting ABS were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min and were placed in a thermostatic bath at 25.0 ºC for 20 minutes. Aliquots from the PRP and ERP were collected, diluted, and the total chromium concentration in the PRP ($C_{T,PRP}$) and ERP ($C_{T,ERP}$) were measured by FAAS. Similar ABS were prepared adding only one chromium specie (Cr(III) or Cr(VI)) and the concentration of Cr(III) or Cr(VI) in the PRP and ERP were obtained.

3.3.8. Determination of chromium species in real samples by ABS speciation procedure

3.3.8.1. Sample collection and preparation. Water samples were collected in clean polyethylene bottles from our laboratory tap (Viçosa, Minas Gerais, Brazil), local wastewater treatment plant (Viçosa, Minas Gerais, Brazil) and Piracicaba River near industrial disposal (Ipatinga, Minas Gerais, Brasil). The samples were filtered and its pH were adjusted to 2.00 with HCl 12.0 mol L^{-1}. Wastewater from electroplating industry (Ubá, Minas Gerais, Brazil) were also collected in clean polyethylene bottles and it has the pH only measured. All samples were stored at 4 ºC.

3.3.8.2. Samples with low chromium content. The water samples of laboratory tap, wastewater treatment plant and Piracicaba River were submitted to the procedure described in section 3.3.8.1 and were quantified by FAAS. Then, the water samples were simultaneously spiked with Cr(III) and Cr(VI) at 0.500 mg kg^{-1} and 0.0500 mg kg^{-1}, respectively. In vessels were added 0.682 g of PEO1500, 13.1 g of Na_2SO_4 and 46.2 g of water samples to obtain systems with 1.40 g of PRP and 58.6 g of ERP. Then, the two formed phases were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min and were placed in a thermostatic bath at 25.0 ºC for 20 minutes. Aliquots of PRP and ERP were collected, diluted and the total chromium concentration in the PRP ($C_{T,PRP}$) and ERP ($C_{T,ERP}$) were measured by FAAS.

3.3.8.3. Samples with high chromium content. Total chromium concentration and possible interfering ions as iron, zinc, copper and nickel were determined in electroplating industry wastewater by FAAS. Cr(VI) quantity was determined by the colorimetric standard method for Cr(VI) water analysis [31] and by difference it was determined Cr(III) concentration. In centrifuge tubes were mixed 1.48 g of PEO1500, 2.12 g of Na_2SO_4 and 4.40 g of electroplating industry wastewater to obtain systems with 4.00g of each phase. The formed systems were manually stirred for 3 minutes, centrifuged at 9503 g for 5 min and were placed in a thermostatic bath at 25.0 ºC for 20 minutes. PRP and ERP aliquots were collected diluted and the concentration of the interfering ions and the total chromium concentration in the PRP ($C_{T,PRP}$) and ERP ($C_{T,ERP}$) were also measured by FAAS.

3.4. RESULTS AND DISCUSSION

3.4.1. Influence of the quantity of chromium species added in the ABS on the %E of Cr(III) and Cr(VI)

It was previously reported, the dependence of Cr(VI) concentration in the abundance of the species H_2CrO_4, $HCrO_4^-$, $Cr_2O_7^{2-}$ and CrO_4^{2-} [32,33]. Thus, the present study investigated the effect of the quantity of chromium species added in the PEO1500 + Na_2SO_4 + H_2O ABS at pH 2.0 on the %E of Cr(III) and Cr(VI) using the TLL = 46.97 % (m/m) (Fig. 3.1).

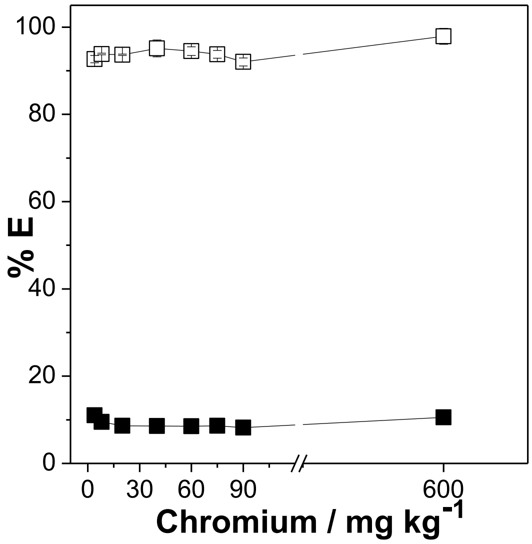

Fig. 3.1. Effect of chromium concentration in the system on the extraction percentage (%E) of Cr(III) and Cr(VI) using PEO1500 + Na_2SO_4 + H_2O ABS, pH=2.00, TLL = 46.97 % (m/m), T = 25.0 ºC. (■) Cr(III) in polymer-rich phase and (□) Cr(VI) in polymer-rich phase.

The chromium concentration in the system varied from 4.00 to 600 mg kg^{-1}. In the investigated range of concentration, Cr(III) and Cr(VI) were successfully separated and it was observed little changes in the percentage extraction as function of the quantity of metallic ions added in the system. Percentage extraction for Cr(III) ($\%E_{Cr(III)}$) varied from (10.6±0.1) to (11.0±0.6)

%, while the percentage extraction for Cr(VI) (%$E_{Cr(VI)}$) oscillated from (92.7±0.8) to (97.9±1.8) %.

Patrício et al. [29] discussed the driving force behind the chromium species partition in ABS. The authors investigated the Cr(III) and Cr(VI) partition behavior in systems composed by PEO1500 polymer or a triblock copolymer (L64), an electrolyte (Li_2SO_4, Na_2SO_4, $MgSO_4$, $C_6H_5Na_3O_7$, or $C_4H_4Na_2O_6$) and water, without the presence of any extractant. The influence of pH, TLL, electrolyte nature and macromolecule hydrophobicity on the %E of the ions were examined. It was demonstrated that ABS formed by macromolecule and an electrolyte is capable of separating Cr(III) and Cr(VI) since Cr(III) partitioned to the electrolyte-rich phase, while the Cr(VI) preferentially concentrated in the polymer-rich phase. Cr(III) partition behavior was attributed to the predominant formation of cationic sulfate complexes which preferably interact with species present in the ERP and are electrostatically repulsed by pseudopolycation in the PRP. The preferential transference of Cr(VI) from ERP to the PRP, occurs at pH 2.0. The Cr(VI) partition was related to the predominant occurrence of $HCrO_4^-$ ions at pH 2.0 and the specific interactions that occur between $HCrO_4^-$ and the pseudopolycation surface. The extraction behavior of the chromium species was affected by pH, TLL, polymer hydrophobicity and the electrolyte since these parameters modulate the chromium–EO interactions. Although several ABS can be used to separate Cr(III) and Cr(VI), the authors observed the potential of PEO1500 + Na_2SO_4 + H_2O ABS at pH 2.0 for the speciation of chromium species. So, in the present work, such conditions were used to investigate the influence of the quantity of chromium species added in the ABS on the %E of Cr(III) and Cr(VI).

Based on da Silva and Loh [34] model, several works [29,35-37] have proposed that the partition behavior of metallic ions in the ABS is related to the formation of a pseudopolycation during the phase separation process. Microcalorimetric data obtained by da Silva and Loh [34] suggest the following mechanism: when polymer and electrolyte solutions are mixed, occurs specific interactions between the cations and the segments of ethylene oxide (EO) of the polymer that increase the system enthalpy. This cation–macromolecule interaction promotes desolvation of the ions and macromolecules, increasing the entropy and minimizing the Gibbs free energy of the system. However, at specific conditions of temperature and concentration, the cation-polymer binding become unfavorable since the polymer surface become energetically saturated avoiding the entropy gain by desolvation process. In order to decrease the system Gibbs free energy, phase separation occur producing a polymer-rich phase enriched with polymer chain adsorbed by cation (pseudopolycation) and an electrolyte-rich phase. The pseudopolycation can interact with negatively charged species.

Thus, in the present work, Cr(III) extraction behaviour was attributed to the predominant formation of cationic sulphate complexes which favourable interact with species present in the ERP and are electrostatically repulsed by pseudopolycation in the PRP. The Cr(VI) partition can be attributed to the presence of $HCrO_4^-$ ions and the specific interactions that occur between $HCrO_4^-$ and the pseudopolycation surface. The low change in $HCrO_4^-$ abundance in the investigated concentration range possibilities the current method application in samples with wide concentration range.

3.4.2. Interconversion of chromium species in the ABS speciation procedure

It is recurrent in the literature reports about the possible interconversion of chromium species during analytical procedures [38,39]. Thus, it was evaluated the possible conversion of chromium species during the analytical procedure using ABS. The %E of Cr(III) and Cr(VI) when the chromium species were individually added to the system were compared to the %E obtained when the chromium species were simultaneously added to the ABS (Table 3.1).

Table 3.1. Percentage extraction of Cr(III) ($\%E_{Cr(III)}$) and Cr(VI) ($\%E_{Cr(VI)}$) obtained when the chromium species were individually or simultaneously added in PEO1500 + Na_2SO_4 + H_2O ABS at pH 2.00, T = 25.0 ^{0}C, TLL = 46.97 % (m/m) and metal = 8.00 mg kg^{-1}.

Addition mechanism	$\%E_{Cr(III),PRP}$	$\%E_{Cr(III),ERP}$	$\%E_{Cr(VI),PRP}$	$\%E_{Cr(VI),ERP}$
Individual	8.05±0.14	91.3±0.3	92.5±0.6	7.70±0.37
Simultaneous	4.67 ± 1.34	96.0 ± 2.3	93.3 ± 0.2	6.71 ± 0.23

Similar %E values were obtained when the chromium species were individually added and simultaneously added in the system. The mass balance ($\%E_{X,PRP}$ + $\%E_{X,ERP}$) of each chromium specie close to 100 % for the addition by distinct mechanism shows that ABS speciation procedure does not cause interconversion between Cr(III) and Cr(VI) species.

3.4.3. ABS pre-concentration speciation procedure

FAAS technique has advantages as simple operation, low cost and good selectivity [40]. However, FAAS technique only determine the total quantity of the element and not determined very low concentrations due to the insufficient sensitivity [5]. Thus, an effective separation and pre-concentration procedure is usually indispensable to differentiate between Cr(III) and Cr(VI). In this work, ABS pre-concentration speciation procedure was applied at determination of low chromium quantities at different ratios using, pioneeringly, first degree equations (Eq. (3.5) and Eq.(3.6)) in two variables coupled to FAAS in order to speciate metallic species:

$$\%\mathrm{E}^{\mathrm{PRP}}_{\mathrm{Cr(III)}}\mathrm{x}\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(III)}} + \%\mathrm{E}^{\mathrm{PRP}}_{\mathrm{Cr(VI)}}\mathrm{x}\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(VI)}} = \frac{\mathrm{m}_{\mathrm{PRP}}\mathrm{x}\ \mathrm{Cr}_{\mathrm{T,PRP}}}{\mathrm{m}_{\mathrm{ABS}}} \qquad (3.5)$$

$$\%\mathrm{E}^{\mathrm{ERP}}_{\mathrm{Cr(III)}}\mathrm{x}\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(III)}} + \%\mathrm{E}^{\mathrm{ERP}}_{\mathrm{Cr(VI)}}\mathrm{x}\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(VI)}} = \frac{\mathrm{m}_{\mathrm{ERP}}\mathrm{x}\ \mathrm{Cr}_{\mathrm{T,ERP}}}{\mathrm{m}_{\mathrm{ABS}}} \qquad (3.6)$$

where $\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(III)}}$ and $\mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(VI)}}$ are the Cr(III) and Cr(VI) concentration in the system. $\mathrm{m}_{\mathrm{PRP}}$, $\mathrm{m}_{\mathrm{ERP}}$ and $\mathrm{m}_{\mathrm{ABS}}$ are the masses of polymer-rich phase, electrolyte-rich phase and of the system, respectively. $\mathrm{Cr}_{\mathrm{T,PRP}}$ and $\mathrm{Cr}_{\mathrm{T,ERP}}$ are the total chromium concentrations obtained by FAAS in the polymer- and electrolyte-rich phase, respectively. The %E of Cr(III) and Cr(VI) in the PRP and ERP ($\%E^{PRP}_{Cr(III)}$, $\%E^{PRP}_{Cr(VI)}$, $\%E^{ERP}_{Cr(III)}$, $\%E^{ERP}_{Cr(VI)}$) were obtained by the individual addition of Cr(III) and Cr(VI) in the system and they were substituted in the Eq. (3.7) and Eq. (3.8):

$$0.000866xC^{ABS}_{Cr(III)} + 0.204\ xC^{ABS}_{Cr(VI)} = \frac{1.40\ x\ Cr_{T,PRP}}{58.6} \qquad (3.7)$$

$$1.02\ xC^{ABS}_{Cr(III)} + 0.732\ xC^{ABS}_{Cr(VI)} = \frac{1.40\ x\ Cr_{T,ERP}}{58.6} \qquad (3.8)$$

After to obtain the concentrations of Cr(III) and Cr(VI) in the ABS ($C^{ABS}_{Cr(III)}$ and $C^{ABS}_{Cr(VI)}$) it was calculated the concentration of each specie in the synthetic samples from the Eq. (3.9) and Eq.(3.10):

$$\mathrm{C}_{\mathrm{Cr(III)}}^{\mathrm{ABS}} \times \mathrm{m}_{\mathrm{ABS}} = \mathrm{C}_{\mathrm{Cr(III)}}^{\mathrm{Found}} \times \mathrm{m}_{\mathrm{Add}} \tag{3.9}$$

$$\mathrm{C}_{\mathrm{Cr(VI)}}^{\mathrm{ABS}} \times \mathrm{m}_{\mathrm{ABS}} = \mathrm{C}_{\mathrm{Cr(VI)}}^{\mathrm{Found}} \times \mathrm{m}_{\mathrm{Add}} \tag{3.10}$$

where $C_{Cr(III)}^{Found}$ and $C_{Cr(VI)}^{Found}$ are the concentrations of Cr(III) and Cr(VI) in the water samples and $\mathrm{m}_{\mathrm{Add}}$ is the mass of water sample added in the ABS.

The equation system (Eq. (3.7) and Eq. (3.8)) was applied to determine the chromium concentration in water deionized samples with different quantities of chromium species (Table 3.2). Recovery percentages (%R) between 90.3 and 112 % were obtained, i.e., quantitative recovery of each chromium specie was obtained at different chromium species ratios.

3.4.4. Determination of chromium species in real samples by ABS speciation procedure

3.4.4.1. Samples with low chromium content. The viability of the proposed speciation method was explored to determine the concentrations of Cr(III) and Cr(VI) in real samples as tap water, wastewater treatment plant and river water. The samples were quantified by FAAS and the chromium element was not found in the samples. The samples were spiked with chromium quantities that satisfies World and Brazilian organizations [7-9]. The equation system (Eq. (3.7) and Eq. (3.8)) and Eq. (3.9) and Eq.(3.10) were also used to determine the chromium species in spiked water samples (Table 3.3). The recovery percentage of the analytes were higher than 88 % suggesting a small water matrices interference.

Table 3.2. Determination of Cr(III) and Cr(VI) concentration in deionized water sample at different concentrations ratio by the proposed method (N = 3).

Cr(III):Cr(VI) ratio	Added / (mg kg^{-1})		CPRP/ (mg kg^{-1})	CERP/ (mg kg^{-1})	Found / (mg kg^{-1})		Recovery / (%)	
	Cr(III)	Cr(VI)			Cr(III)	Cr(VI)	Cr(III)	Cr(VI)
5:0.5	0.500	0.0500	0.317±0.009	0.476±0.042	0.560±0.052	0.0452±0.0013	112±10	90.3±2.7
5:1	0.518	0.100	0.652±0.013	0.469±0.007	0.516±0.008	0.0955±0.0019	99.5±1.5	95.5±1.9
10:1	0.996	0.100	0.644±0.029	0.866±0.004	1.01±0.01	0.0922±0.0044	92.2±4.4	102±0.6

Table 3.3. Determination of Cr(III) and Cr(VI) concentration in spiked real water samples by the proposed method (N = 3).

Sample	Added /(mg kg^{-1})		CPRP/ (mg kg^{-1})	CERP/ (mg kg^{-1})	Found / (mg kg^{-1})		Recovery / (%)	
	Cr(III)	Cr(VI)			Cr(III)	Cr(VI)	Cr(III)	Cr(VI)
Tap	-	-	n.d[a]	n.d[a]	n.d[a]	n.d[a]	-	-
water	0.498	0.0506	0.333±0.009	0.387±0.0216	0.4471±0.027	0.0480±0.001	89.8±5.4	94.9±2.7
WTP[b]	-	-	n.d[a]	n.d[a]	n.d[a]	n.d[a]	-	-
	0.517	0.0501	0.321±0.011	0.472±0.020	0.554±0.023	0.0457±0.00154	107±4	91.2±3.1
River	-	-	n.d[a]	n.d[a]	n.d[a]	n.d[a]	-	-
water	0.505	0.0501	0.307±0.012	0.405±0.005	0.472±0.007	0.0440±0.002	93.5±1.4	87.9±3.7

[a] n.d, not detected, [b] WTP, wastewater treatment plant

3.4.4.2. Samples with high chromium content. The proposed method was also applied to determine the Cr(III) and Cr(VI) concentrations in electroplating waste that contains high chromium quantity. Besides chromium species, the wastewater can contain other metallic ions. Chromium and foreign ions as nickel, zinc, copper and iron were determined by FAAS and the results are given in Table 3.4.

Table 3. 4. Determination of metallic ions concentration in electroplating waste by FAAS (N=3).

Metallic ion	Concentration / (mg kg^{-1})
Chromium	(9.85±0.19) x10^{3}
Copper	106±1
Iron	45.8±0.3
Nickel	224±2
Zinc	2.54±0.03

In order to evaluate the possible analytical applications of the ABS speciation procedure, the %E of the interfering ions in the ABS system were achieved (Fig. 3.2).

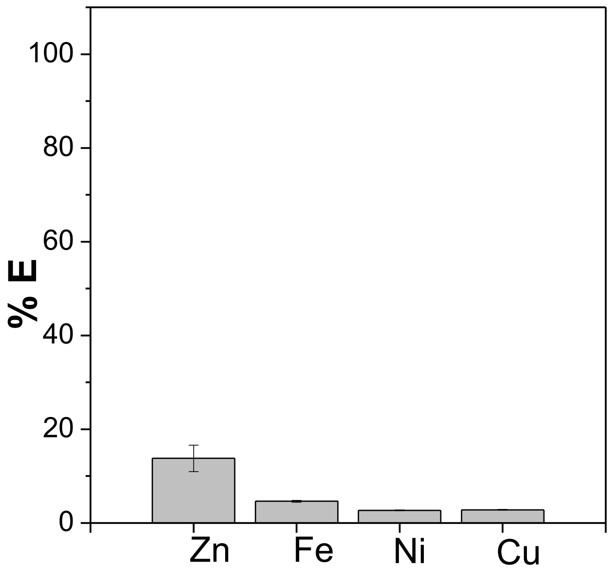

Fig. 3.2. Percentage extraction of Zn, Fe, Ni and Cu for the polymer-rich phase in the presence of Cr(III) and Cr(VI) in the PEO1500 + Na_2SO_4 + Electroplating waste ABS at T = 25.0 ºC, TLL = 46.97 % (m/m).

The metallic species exhibited small extraction percentages as $\%E_{Zn}$= (13.8±2.8) %, $\%E_{Fe}$= (4.64±0.20) %, $\%E_{Ni}$= (2.69±0.08) % and $\%E_{Cu}$= (2.80±0.08) %. As generally found, in the absence of extractant, the metallic ions spontaneously concentrate in electrolyte-rich phase due the weak interaction with the macromolecule segments [27,35,41]. Thus, the EO segments of the macromolecules were free to interact with the high content of Cr(VI) species. In the presence of foreign ions, Cr(III) and Cr(VI) concentration in electroplating waste were determined by the ABS speciation procedure. The chromium species concentration was obtained by the following equation system (Eq. (3.11) and (3.12)) and by the equations Eq. (3.9) and Eq.(3.10).

$$0.925 \times C_{Cr(III)}^{ABS} + 0.0805 \times C_{Cr(VI)}^{ABS} = \frac{2.00 \text{ x } Cr_{T,PRP}}{4.00} \tag{3.11}$$

$$0.0770 \times \mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(III)}} + 0.913 \times \mathrm{C}^{\mathrm{ABS}}_{\mathrm{Cr(VI)}} = \frac{2.00 \text{ x } \mathrm{Cr}_{\mathrm{T,ERP}}}{4.00} \qquad (3.12)$$

The colorimetric standard method of Cr(VI) water analysis [31] was also performed in order to validate the suggested method accuracy. The Cr(VI) concentration was determined by diphenylcarbazide method, total chromium concentration by FAAS and by difference Cr(III) concentration was found. The proposed and standard method results were compared as shown in Table 3.5.

Table 3.5. Determination of Cr(III) and Cr(VI) concentration in electroplating waste by the standard and proposed method (N = 3).

Specie	Standard method / mg kg^{-1}	C_{PRP}/ (mg kg^{-1})	C_{ERP}/ (mg kg^{-1})	Proposed method / mg kg^{-1}	Relative error / %
Cr(III)	(1.92±0.25) x10^3	(10.5±0.06) x10^3	(2.86±0.05) x10^3	(1.66±0.05) x10^3	13.6
Cr(VI)	(7.92±0.05) x10^3			(8.41±0.05) x10^3	-6.21

The results revealed good concordance between the ABS and the standard method results. The interfering ions did not have significant effect on the separation and determination of Cr(III) and Cr(VI) ions.

3.4.5. Analytical figures of merit

Table 3.6 shows the analytical features of the proposed method. External calibration plots were constructed and excellent linearity were obtained with minimal correlation coefficients of 0.999. Analytical curves with linear range of 0.05 to 0.500 mg kg^{-1} were used to determine the detection and quantification limits. The detection limit (LOD) was calculated as 3.3 x s/S, where s is the standard deviation of the ordinate intercept and S is the slope of the analytical curve. The quantification limit (LOQ) was calculated as 10 x s/S where s is the standard deviation of the ordinate intercept and S is the slope of the calibration curve. The enrichment factor (EF) or preconcentration factor (PF) was determined based on the relation $C_{PRP}/C_{initial}$ where C_{PRP} is the concentration of analyte in the macromolecule-rich phase and $C_{initial}$ is the concentration of analyte in the spiked samples. The percentual relative error (RSD) for five Cr(VI) extractions (n = 5, c = 15.0 mg kg^{-1}) and for five Cr(III) extractions (n = 5, c = 23.0 mg kg^{-1}) were obtained. The recovery studies for Cr(III) and Cr(VI) were performed in wastewater of treatment plant, tap and river water. known amounts of chromium species were added in the distinct water samples in order to estimate the accuracy of the presented method by the recovery percentage (%R) and to evaluate interference caused by different matrices.

Table 3.6. Analytical figures of merit

Parameter	
Linear range / (mg kg^{-1})	0.0500 to 0.500 or 0.100 to 4.00
LOD / (mg kg^{-1}) (n=14)	0.0538
LOQ / (mg kg^{-1}) (n=14)	0.163
RSD / % (n=5)	0.802 (Cr(VI)) and 3.53 (Cr(III))
EF	6.69
%R / %	87.9

3.5. CONCLUSION

An environmentally safe analytical method to determine Cr(III) and Cr(VI) in aqueous matrices was developed. The proposed procedure is simple and involves the separation and, or pre-concentration of the chromium species using PEO1500 + Na_2SO_4 + H_2O ABS at pH = 2.0, 25.0 ºC and TLL = 46.97 % (m/m) and

FAAS technique to quantify them. The %E of Cr(III) and Cr(VI) was not affected by the quantity of chromium species added in the system, indicating the applicability of the proposed method in samples with wide concentration range. The ABS speciation procedure does not cause interconversion between the chromium species and therefore, could be satisfactorily applied to determine Cr(III) and Cr(VI) in water samples of tap, treatment plant, river and electroplating waste.

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CONCLUSÕES GERAIS

Um método analítico ambientalmente seguro foi desenvolvido para a determinação das espécies Cr(III) e Cr(VI) em matrizes aquosas. Sistemas aquosos bifásicos formados por macromolécula e um eletrólito foram eficientes para separar Cr(III) e Cr(VI) sem a utilização de solventes orgânicos ou qualquer extratante. O comportamento de partição de Cr(III) e Cr(VI) foi afetado pelo pH, CLA, hidrofobicidade da macromolécula e natureza do eletrólito. Os resultados mostram que em pH 2,0 os íons Cr(III) foram particionados para a fase rica em eletrólito, enquanto os íons Cr(VI) concentraram-se na fase rica em macromolécula. Para determinar a força motriz deste comportamento de partição utilizou-se espectroscopia de fluorescência. Apreciável redução da intensidade de fluorescência do polímero PEO1500 na presença de íons Cr(VI) em pH 2,0 sugere que a partição desta espécie para fase rica em macromolécula é regida por interações específicas que ocorrem entre os segmentos de óxido de etileno das macromoléculas e as espécies $HCrO_4^-$ predominantes neste pH. Essas interações não foram observadas para os íons Cr(III). Maiores comprimentos de linha de amarração geram SAB com maior concentração de macromolécula na fase rica em macromolécula e por isso, favoreceram a partição de Cr(VI) para a fase rica em macromolécula e Cr(III) para fase rica em eletrólito. De modo geral, este comportamento de partição também é mais favorecido por sistemas formados por PEO1500 que L64 e por sistemas constituídos por sais inorgânicos (Li_2SO_4, Na_2SO_4 e $MgSO_4$) que sais orgânicos ($C_6H_5Na_3O_7$ e $C_4H_4Na_2O_6$).

Os fatores de separação obtidos neste trabalho mostram que alguns SAB investigados apresentaram potencial para separar Cr(III) e Cr(VI). Entre eles podemos citar os sistemas aquosos bifásicos PEO1500 + Na_2SO_4 + H_2O (CLA = 50,89 %

(m/m)) com $S_{\mathrm{Cr(VI)/Cr(III)}}$ = 1,00 × 10^8, PEO1500 + Na_2SO_4 + H_2O (CLA = 46,97% (m/m)) com $S_{\mathrm{Cr(VI)/Cr(III)}}$ = 5,82 × 10^4, PEO1500 + Li_2SO_4 + H_2O (CLA = 29,43% (m/m)) com $S_{\mathrm{Cr(VI)/Cr(III)}}$ = 6,76 × 10^4 e PEO1500 + Li_2SO_4 + H_2O (CLA = 34,97% (m/m)) com $S_{\mathrm{Cr(VI)/Cr(III)}}$ = 2,36 × 10^3.

O SAB PEO1500 + Na_2SO_4 + H_2O em pH = 2,0, 25,0 ºC e CLA = 46,97 % (m/m) foi utilizado no desenvolvimento de um método analítico para especiação de Cr(III) e Cr(VI). Este sistema foi utilizado para separar e, ou pré-concentrar as espécies e a técnica EAAC foi empregada para quantifica-las. O limite de detecção obtido satisfaz o teor exigido para estes analitos em amostras ambientais. O procedimento proposto não causa interconversão das espécies e, portanto, pode ser aplicado em diferentes matrizes aquosas com resultados satisfatórios.

Os resultados obtidos neste trabalho foram importantes para ajudar esclarecer os mecanismos que governam a partição de espécies no SAB e permitiram o desenvolvimento de um método verde, barato, simples e sensível usando SAB–EAAC para a determinação das espécies de cromo.

ANEXO

Table A1. Percent composition (m/m) for L64 (w_{L64}), Li_2SO_4 (w_E) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{L64}	w_E	w_W	w_{L64}	w_E	w_W	w_{L64}	w_E	w_W
21.51	12.75	6.51	80.75	23.40	5.07	71.53	2.09	7.95	89.96
31.25	16.60	6.31	77.09	32.08	4.14	63.78	1.12	8.48	90.39
35.63	18.17	6.76	75.08	35.77	4.01	60.23	0.56	9.50	89.94
41.86	21.07	6.64	72.30	41.72	3.27	55.00	0.41	10.01	89.57

Table A2. Percent composition (m/m) for L64 (w_{L64}), Na_2SO_4 (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{L64}	w_E	w_W	w_{L64}	w_E	w_W	w_{L64}	w_E	w_W
33.55	17.76	5.84	76.40	34.31	3.10	62.59	1.21	8.58	90.20
41.83	21.14	5.92	72.95	41.73	2.23	56.05	0.55	9.60	89.85
47.82	23.98	6.11	69.91	47.49	1.76	50.75	0.47	10.46	89.08
50.29	25.19	6.35	68.46	49.83	1.33	48.84	0.55	11.37	88.08
54.93	27.25	6.85	65.91	54.12	1.19	44.69	0.37	12.50	87.13

Table A3. Percent composition (m/m) for PEO1500 (w_{PEO}), Na_2SO_4 (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W
32.42	17.37	9.24	73.40	32.26	2.84	64.80	2.47	15.63	81.90
35.96	18.32	9.97	71.72	34.51	2.16	63.33	2.12	17.77	80.11
40.10	19.90	10.70	69.40	37.94	1.95	60.11	1.86	19.45	78.69
46.97	22.22	11.59	66.20	43.41	1.46	55.13	1.03	21.71	77.26
50.89	23.44	12.39	64.17	46.29	1.20	52.51	0.59	23.58	75.83

Table A4. Percent composition (m/m) for PEO1500 (w_{PEO}), Li_2SO_4 (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W
29.43	20.65	10.42	68.94	34.44	5.30	60.26	6.85	15.54	77.61
34.97	22.89	10.50	66.61	39.18	4.15	56.67	6.60	16.85	76.55
41.71	25.16	11.29	63.56	44.40	3.25	52.35	5.91	19.32	74.77
46.36	27.03	11.61	61.37	48.38	2.58	49.04	5.68	20.63	73.69
51.67	29.43	12.14	58.44	53.26	2.15	44.59	5.60	22.12	72.28

Table A5. Percent composition (m/m) for PEO1500 (w_{PEO}), $MgSO_4$ (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W
33.81	22.01	8.91	69.09	37.27	1.63	61.10	6.75	16.18	77.07
40.12	24.55	9.48	65.98	42.80	1.16	56.04	6.29	17.79	75.92
43.76	25.78	9.82	64.41	45.72	0.81	53.47	5.84	18.82	75.34
47.28	26.65	10.80	62.56	47.95	0.56	51.49	5.34	21.04	73.62
52.40	28.54	11.54	59.93	52.24	0.38	47.38	4.83	22.70	72.47

Table A6. Percent composition (m/m) for PEO1500 (w_{PEO}), $C_6H_5Na_3O_7$ (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W
26.43	13.51	12.57	73.93	25.17	6.35	68.48	1.84	18.78	79.37
36.54	16.39	13.34	70.28	32.31	4.39	63.30	0.46	22.29	77.25
42.87	18.62	13.95	67.43	37.06	3.02	59.92	0.18	24.88	74.94
49.67	21.13	15.21	63.67	42.24	2.13	55.63	0.01	28.28	71.71
53.84	22.82	16.00	61.19	45.63	1.70	52.67	0.01	30.29	69.70

Table A7. Percent composition (m/m) for PEO1500 (w_{PEO}), $C_4H_4Na_2O_6$ (w_e) and water (w_w) in an aqueous biphasic system (ABS) at 25.0 ◦C.

TLL	Overal			Macromolecule-rich phase			Electrolyte-rich phase		
	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W	w_{PEO}	w_E	w_W
28.73	15.68	12.85	71.48	28.62	6.60	64.78	2.73	19.09	78.18
34.94	17.10	13.47	69.44	32.65	5.51	61.84	1.54	21.43	77.03
41.74	19.28	14.20	66.53	37.65	4.30	58.05	0.90	24.10	75.00
48.34	21.89	15.29	62.83	42.92	3.38	53.70	0.85	27.20	71.95
50.21	22.48	15.65	61.87	44.25	3.15	52.59	0.71	28.15	71.13

APÊNDICE

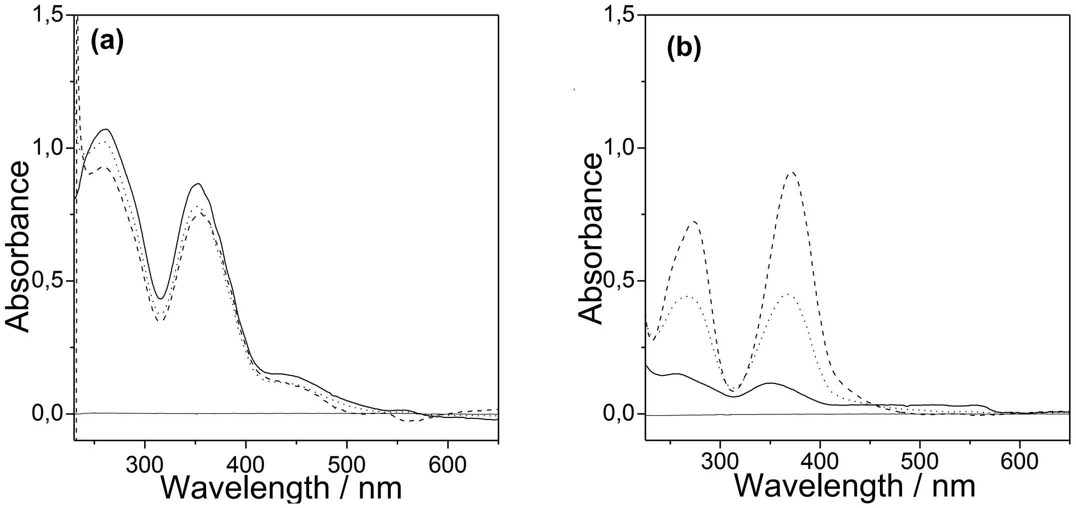

Fig. A1. UV-vis molecular absorption spectra of chromium species in (a) macromolecule-rich phase; (b) electrolyte-rich phase in the ABS formed by PEO 1500 + Na_2SO_4 + H_2O, TLL = 46.97 %(m/m)); [metal] = 15.0 mg kg^{-1}; T = 25.0 ºC. (__) pH 2.0; (….) pH 5.0; (---) pH 11.0.

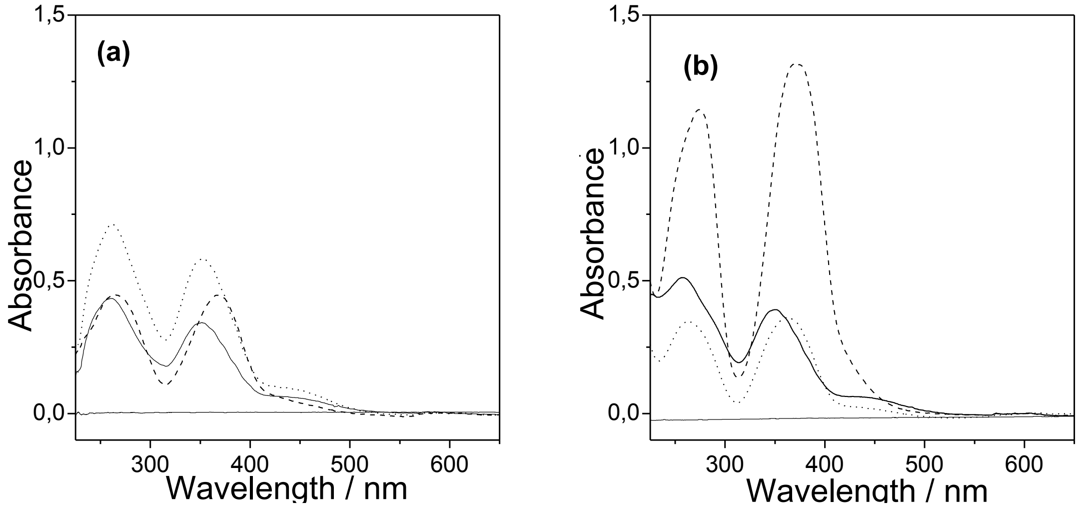

Fig. A2. UV-vis molecular absorption spectra of chromium species in (a) macromolecule rich phase; (b) electrolyte-rich phase in the ABS formed by L64 + Na_2SO_4 + H_2O, TLL = 47.82 %(m/m)); [metal] = 15.0 mg kg^{-1}; T = 25.0 ºC. (__) pH 2.0; (….) pH 5.0; (---) pH 11.0.

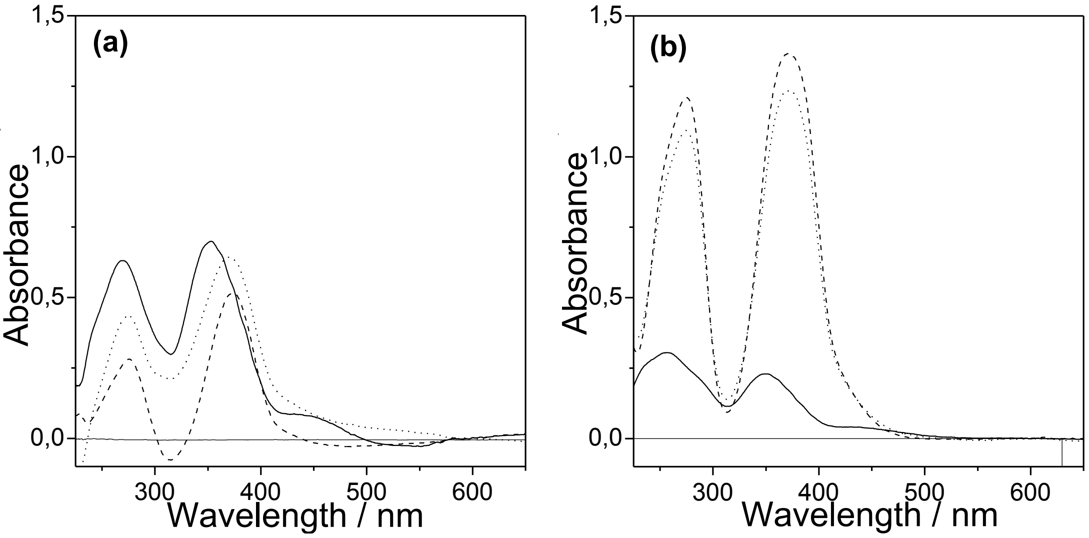

Fig. A3. UV-vis molecular absorption spectra of chromium species in (a) macromolecule rich phase; (b) electrolyte-rich phase in the ABS formed by L64 + Li$_2$SO$_4$ + H$_2$O, TLL = 41.86 %(m/m)); [metal] = 15.0 mg kg^{-1}; T = 25.0 ºC. (__) pH 2.0; (….) pH 5.0; (---) pH 11.0.

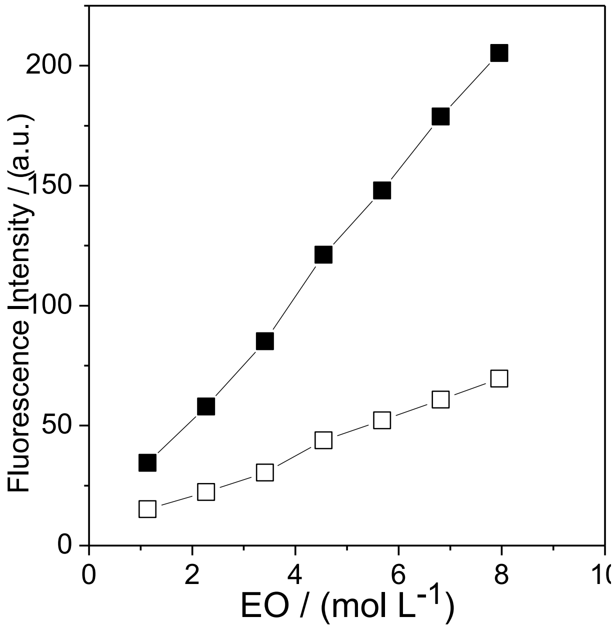

Fig. A4. Effect of Cr(VI) in fluorescence intensity of the polymer PEO1500. [Na$_2$SO$_4$] = 0.103 mol L^{-1}; [Cr (VI)] = 5 x 10^{-4} mol L^{-1}; pH = 11.0. (■) in the absence of Cr(VI); (□) in the presence of Cr(VI).