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Speciation of chromium by in-capillary derivatization and electrophoretically mediated microanalysis

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Abstract

An electrophoretic method for chromium speciation analysis – as Cr(III) and Cr(VI) – based on in-capillary derivatization with 1,5-diphenylcarbazide (DPC) is here proposed. As Cr(III) does not react with DPC, it was oxidized also in-capillary to Cr(VI) by Ce(IV). For this purpose, a capillary electrophoresis (CE) mode called electrophoretically mediated microanalysis (EMMA) based on sequential injection of sample and reagents – namely, DPC, sample and Ce(IV) – was employed. The conditions of both reactions – Cr(III) oxidation and Cr(VI)-DPC derivatization – were optimized in order to quantify separately the Cr(VI)–DPC complex from the original Cr(VI) in the sample and that from oxidation of Cr(III) to Cr(VI). The electrophoretic conditions were independently optimized for variables influencing the resolution and those affecting sensitivity. The method thus developed was applied to the determination of Cr(III) and Cr(VI) in glass material, for which different sample preparation methods – namely, EPA method 3060A, ultrasound-assisted leaching and microwave-assisted digestion – were tested. Microwave-assisted digestion was found to be the best sample preparation alternative in terms of efficiency of the step – 99.6 and 98.3% for Cr(VI) and Cr(III), respectively – and procedure time – 20 min. The complete method was validated with the certified reference material BAM-S004.

Keywords: Electrophoretically mediated microanalysis; Speciation; Chromium; In-capillary derivatization; Sample preparation

1. Introduction

Speciation analysis of trace elements has become important in the past decades, due to its impact on environmental chemistry, ecotoxicology, clinical toxicology and food industry. Most investigations have been focused on monitoring anthropogenic pollutants in environment, degradation processes of pharmacological drugs in human body, and determination of trace elements in foods.

A particular case is that of chromium, whose toxicity is a function of the oxidation state and concentration. Chromium occurs primarily in two valence states, hexavalent and tervalent—Cr(VI) and Cr(III), respectively. Tervalent chromium is relatively non-toxic and an essential nutrient in the human diet to maintain in healthy levels the glucose, lipid and protein metabolism [1,2]. By contrast, Cr(VI) is mainly emitted by industrial sources, such as metal plating, tanning, chromate ore processing and spray painting operations, stainless steel welding, combustion sources and fugitive dusts from contaminated soils [3,4]. Hexavalent chromium has demonstrated to be a human respiratory carcinogen in epidemiological studies when humans are exposed to relatively high levels in the workplace. The toxicity of Cr(VI) has led to the implementation of continuous monitoring in workplaces where emissions of this chromium form can be produced, as stated in the Directive 90/3941/EEC on exposure to carcinogenic substances. The occupational exposure limits (OEL) for water-soluble and certain water-insoluble compounds in indoor air are as low as 0.5 mg m^{-3} for Cr(III) and 0.05 mg m^{-3} for Cr(VI), which reflect the different toxicity of the two species and the considerable interest in the industry and regulatory community to assess the potential cancer risks of workers exposed to Cr(VI) [5].

A key aspect of the determination of Cr(VI) is that samples often have a matrix where Cr(III) ranges from 10 to 1000 times higher concentration than that of Cr(VI); thus, preservation and stabilization of the oxidation states are essential to ensure the accuracy and precision of the analysis. The pH strongly influences the relative stability of Cr(VI) and Cr(III) in aque-

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ous solutions. Alkaline media favour the stabilization of Cr(VI), while acidic pHs stabilize Cr(III) [6,7].

A great variety of methods have been proposed for the determination of chromium, being those based on atomic absorption spectrometry (AAS) the most commonly used [8,9]. Other methods for elemental chromium determination make use of inductively coupled plasma atomic emission spectrometry (ICP–AES) [10], inductively coupled plasma mass spectrometry (ICP–MS) [11] and X-ray fluorescence [12]. Only total chromium can be determined by these methods; therefore, a separation step prior to detection is required to obtain speciation information.

There are other analytical methods, which enable to determine only one of the two species. For instance, the environmental protection agency (EPA) recognises four methods for sample preparation of hexavalent chromium: 7195 (coprecipitation), 7196 (colorimetry with 1,5-diphenylcarbazide (DPC)), 7197 (chelation/extraction), and 7198 (differential pulse polarography). A study conducted by Gurknecht in 1983 evaluated the above four methods, concluding that 7195 and 7197 methods were vulnerable to effects of matrix composition [13], and that 7196 colorimetric method based on the coloured complex formed between DPC and Cr(VI) is one of the most sensitive and selective for Cr(VI) determination [14]. Nevertheless, the determination of both individual ionic forms is sometimes required. A simple alternative is the use of flow-injection (FI) methods, which enable to distinguish between both chromium species. One case in point is the inclusion of cationic and/or anionic resins in the FI manifold [15]; another is based on the sequential injection of the sample and derivatization by DPC without and with previous addition of Ce(IV), which oxidizes Cr(III) to Cr(VI) [16].

Separation techniques, such as liquid chromatography (LC), ion chromatography (IC) and capillary electrophoresis (CE), are especially attractive for speciation studies, since they can differentiate various chemical forms of the same element. A previous step such as complexation is usually required in LC and CE. The necessity for complexation in CE is due to the different charge of the two chromium species, making difficult their simultaneous determination in a single run [17].

The proposition of selective and sensitive methods for chromium speciation without any previous derivatization step is desirable. Kuban et al. proposed an electrophoretic method based on dual opposite end injection in which the anionic and cationic species, injected into opposite ends of the separation capillary, migrated towards the capillary centre where Cr(III) and Cr(VI) were monitored by a contactless conductimetric detector [18].

The objective of this research was to propose an electrophoretic method for chromium speciation based on in-capillary derivatization with DPC, with also in-capillary previous oxidation of Cr(III) to Cr(VI). For this purpose, a CE mode called electrophoretically mediated microanalysis (EMMA) based on sequential injections of the sample and derivatizing reagents has been used. The working conditions of both reactions – Cr(III) oxidation and Cr(VI)–DPC derivatization – were optimized in order to quantify separately both Cr(VI)–DPC complexes (namely, that from the original Cr(VI) contained in the sample and that from oxidation of Cr(III) to Cr(VI), representative of the Cr(III) concentration in the sample). The speciation analysis of chromium in glass material requires a key sample preparation step, which has also been an objective of this research.

2. Materials and methods

2.1. Chemicals, solutions and samples

Eighteen milliOhms deionized water from a Millipore Milli-Q water purification system was used for conditioning the capillary and preparing the stock standard solutions and buffer. All chemicals employed for this research were of reagent grade. Stock standard solutions of Cr(III) and Cr(VI) at 1 g/l were prepared from $CrCl_3 \cdot 6H_2O$ and K_2CrO_4 (Panreac, Barcelona, Spain), respectively, and stored in the refrigerator at 2 °C.

The electrolyte solution consisted of a 30 mM Na_2HPO_4 aqueous solution adjusted to pH 2 with ortophosphoric acid both from Panreac. The electrolyte solution was filtered through a 0.45 μm nylon filter prior to its use.

A 0.28 M Na_2CO_3/0.5 M NaOH solution (both reagents from Panreac) was used in all sample preparation alternatives. Hydrochloric acid was employed to adjust to pH 2 the solution from each sample preparation alternative.

A certified reference material (CRM) BAM-S004 from the Federal Institute for Materials Research and Testing (Berlin, Germany) in co-operation with the International Commission on Glass [19] was used both for the optimization of the sample treatment and for validating the proposed method. This CRM has certified values for Cr(VI) and total chromium and consists of glass container for cosmetics crushed into pieces (<10 mm in size) with a powdered fraction. Therefore, for the analysis the material was grinded in an agate mill.

2.2. Instruments and apparatus

A 3D Capillary Electrophoresis Agilent G1600A Instrument (Hewlett-Packard-Strasse, Waldbronn, Germany), equipped with a diode array detector (range 190–600 nm) and thermostated by a Peltier unit was used to separate and quantify the analytes. Agilent capillary tubing of 48 cm (effective length 40 cm) × 50 μm i.d. × 375 μm o.d. was used.

A Hobersal model HD-150 (maximum temperature 1200 °C) furnace (Forns Hobersal, Barcelona, Spain) was used to carry out the EPA method 3060A.

Ultrasonic irradiation was applied by a Branson 450 digital sonifier (20 KHz, 450 W) equipped with a cylindrical titanium alloy probe (12.70 mm diameter), which was immersed into the sample vessel positioned into a water bath.

A Microdigest 301 digestor of 200 W maximum power (Prolabo, Paris, France) was used to assist the sample preparation with microwaves as auxiliary energy.

A centrifuge (Selecta, Barcelona, Spain) was used to remove the particulates in suspension.