

Preparation of enriched isotopic and natural abundance standard solutions

⁵⁰Cr(VI) standard solution. 0.0029 g of Cr₂O₃ (⁵⁰Cr enriched isotope) was transferred to a platinum beaker and 0.4 g of NaKCO₃ and 0.1 g of NaOH were added. The contents were melted by a Bunsen burner until a yellow-orange melt was obtained. During the melting procedure quantitative oxidation of Cr by air oxygen was achieved only in an alkaline media. The melt was cooled to a room temperature and 0.7 mL of concentrated HCl was carefully added to dissolve the deposit. The clear solution was transferred to a Teflon tube and diluted to 10 mL with water.¹ The concentration of Cr in the stock isotopic spike solution was determined by reverse ID-ICP-MS and found to be $197.2 \pm 0.8 \mu\text{g mL}^{-1}$.

⁵³Cr(III) standard solution. 0.0029 g of Cr₂O₃ (⁵³Cr enriched isotope) was transferred to a Teflon vessel and 4 mL of concentrated HNO₃ was added. The Teflon vessel was subjected to microwave assisted digestion performed at maximal power of 1200 W, ramp to temperature 20 min, 190 °C, pressure 10 bar, holding 20 min, cooling 20 min. The clear solution was quantitatively transferred to a platinum beaker and the contents carefully evaporated to approximately 0.2 mL. Then 0.7 mL of concentrated HCl was added, the solution transferred to a Teflon tube and diluted to 10 mL with water.¹ The concentration of Cr in the stock isotopic spike solution was determined by reverse ID-ICP-MS and found to be $200.3 \pm 0.9 \mu\text{g mL}^{-1}$.

Cr(III) and Cr(VI) standard solutions. For preparation of working standard solutions of natural abundance Cr(III) and Cr(VI) standards purchased from Merck were appropriately diluted with water.

Isotopic-enriched-certified standard solution. 31 mg of SRM 979 (Cr(NO₃)₃ x 9H₂O) was weighed in a Teflon container and diluted with 1 % HNO₃ to 400 g. This solution was used for determination of detector dead time.

Determination of total concentrations of enriched $^{53}\text{Cr(III)}$ and $^{50}\text{Cr(VI)}$ standard solutions

A solution containing approximately 20 ng mL^{-1} $^{50}\text{Cr(VI)}$ or $^{53}\text{Cr(III)}$ was spiked with 20 ng mL^{-1} Cr. Then the isotope ratios $^{53}\text{Cr}/^{52}\text{Cr}$ and $^{50}\text{Cr}/^{52}\text{Cr}$ were determined. The concentration of the spike (C_{spike}) was calculated using the following equation:^{2,3}

$$C_{\text{spike}} = N_s \times M_s / V_{\text{mix}}$$

$$N_s = N_x \times (R_{a/52} \times {}^{52}\text{A}_x - {}^a\text{A}_x) / ({}^a\text{A}_s - R_{a/52} \times {}^{52}\text{A}_s)$$

Regarding the total concentration determined, “a” represents either mass 50 or 53 and $R_{a/52}$ the corresponding ratio. N_s and N_x are the amounts (mole) of the isotopic spike and the standard with natural isotopic abundance in the solution, respectively. M_s is the average atomic weights of the isotopic spike and the standard in g mol^{-1} , respectively. V_{mix} is the volume of the solution of which the concentration is being determined. ${}^a\text{A}_x$ and ${}^a\text{A}_s$ are the atomic fractions of ${}^a\text{Cr}$ for the standard and spike, respectively. ${}^{52}\text{A}_x$ and ${}^{52}\text{A}_s$ are the atomic fractions of ^{52}Cr for the standard and spike, respectively. The average atomic weight was calculated as: sum (atomic weight of the isotope \times atomic fraction).

Determination of Cr(VI) concentration in the enriched $^{50}\text{Cr(VI)}$ standard solution

A solution containing 20 ng mL^{-1} $^{50}\text{Cr(VI)}$ and 20 ng mL^{-1} Cr(VI) was prepared, acidified to pH 3 and chromatographically separated. After separation the ratio of $^{50}\text{Cr}/^{52}\text{Cr}$ in Cr(VI) species was determined ($R_{50/52}^{\text{VI}}$). The concentration of Cr(VI) in the $^{50}\text{Cr(VI)}$ solution was calculated using the equation given in:^{2,3}

$$C_{\text{spike}}^{\text{VI}} = N_{\text{s}}^{\text{VI}} \times M_{\text{s}} / V_{\text{mix}}$$

$$N_{\text{s}}^{\text{VI}} = N_{\text{x}}^{\text{VI}} \times (R_{50/52}^{\text{VI}} \times {}^{52}\text{A}_{\text{x}} - {}^{50}\text{A}_{\text{x}}) / ({}^{50}\text{A}_{\text{s}} - R_{50/52} \times {}^{52}\text{A}_{\text{s}})$$

where $C_{\text{spike}}^{\text{VI}}$ is the concentration of Cr(VI) in the spike. N_{s}^{VI} and N_{x}^{VI} are the amounts (mole) of Cr(VI) in the isotopic spike and the standard with the natural isotopic abundance in the solution, respectively.

It was proven that the concentration of Cr(VI) corresponded to the total concentration of ${}^{50}\text{Cr(VI)}$, confirming the purity of the isotopic spike solution.

For ${}^{53}\text{Cr(III)}$ no calibration was needed as only Cr(III) was present in this solution.

Determination of detector dead time and mass bias

Detector dead time was determined by measuring the isotope ratio $R_{50/52}$ of SRM 979 at concentrations ranging from 10 to 120 ng Cr mL⁻¹. $R_{50/52}$ at a dead time of 46 ns was the closest to the certified value. Therefore, in all experiments a dead time of 46 ns was used. Mass bias was determined daily by chromatographic separation of Cr(VI) at concentrations that were close to Cr concentrations in the sample investigated. To compensate for shifts during the day, mass bias was determined before and after a series of six samples and the average mass bias was then used to calculate Cr(VI) concentrations. The measured ratio $R_{50/52}$ at a dead time of 46 ns was corrected for the mass bias factor calculated for the corresponding concentration.³

$$\text{mass bias factor} = R_{\text{t}} / R_{\text{m}}$$

$$R_{\text{c}} = \text{mass bias factor} \times R_{\text{m}}$$

where R_{t} is the certified ratio, R_{c} is the corrected isotope ratio for dead time and mass bias and R_{m} is the measured isotope ratio.

References (related to Electronic Supplementary Information)

- 1 B. Novotnik, T. Zuliani, A. Martinčič, J. Ščančar and R. Milačič, *Talanta*, 10.1016/j.talanta.2012.05.022
- 2 P. Rodríguez-González, J.M. Marchante-Gayón , J. I. García Alonso and A. Sanz-Medel, *Spectrochim. Acta Part B*, 2005, **60**, 151-207.
- 3 USEPA, Method 6800, 2007, Elemental and speciated isotope dilution mass spectrometry, Test methods for evaluating solid waste, physical/chemical methods, SW 846, Update IVA. US Government Printing Office (GPO), Washington DC.