Characterisation of the First “Speciated” Chromium Enriched Organically Bound Yeast Reference Material: ERM-BD213a

Sarah Hill, John Entwisle, Panayot Petrov and Heidi Goenaga Infante

LGC Limited, Queens Road, Teddington, Middlesex, TW11 0LY, UK sarah.hill@lgcgroup.com

Background

• Chromium (Cr) food supplements account for >6% of mineral supplements sales
• Cr can act as an essential element depending on its speciation
• Trivalent chromium, Cr(III), is regarded as essential
• Hexavalent chromium, Cr(VI), is classified as a Category 1 carcinogen
• European Council maximum limit of <0.2% of Hexavalent chromium, Cr(VI)
• Cr can act as essential element depending on its speciation
• Chromium (Cr) food supplements account for

Introduction

• There is a lack of suitable certified reference materials available for Cr species in food and food supplements
• Therefore a new yeast-based reference material was produced: ERM-BD213a
• Both total Cr and Cr(III) are important target parameters
• Isotope dilution analysis (IDA) is the method of choice for reference value assignment:
  • SI traceability
  • high accuracy measurements
  • fit-for-purpose uncertainty
• Total Cr was determined by double IDA in combination with inductively coupled plasma mass spectrometry (ICP-MS)
• Cr species have complex chemistry and their determination is hampered by bidirectional conversion between Cr(III) and Cr(VI)
• Experiments with Cr(III) enriched yeast have shown ~68% of Cr(III) converts to Cr(VI) in basic media
• Single spike double IDA is not sufficient to account for these changes
• Therefore, a species-specific double spike single IDA method was developed
• It is based on isotope pattern deconvolution which accounts for bidirectional species transformation in the analytical process
• Combined with chromatography and ICP-MS, Cr(III) can be accurately determined

Methods - Double IDA

• A highly enriched isotope spike ($^{53}\text{Cr}$Cr at 98.7% abundance) was added to the sample prior to sample preparation
• 0.2 g sample mixed with the $^{53}\text{Cr}$ enriched spike to give a gravimetric ratio ($^{52}\text{Cr}^{53}\text{Cr}$Cr) of 1
• 7 mL HNO$_3$ and 3 mL H$_2$O$_2$
• Microwave digestion @ 180°C
• Diluted to 50 g with water
• The primary calibration standard (NIST SRM 3113a) was also prepared in the same manner
• Analysis using helium collision/KED mode (7700, Agilent Technologies)

Methods - Species-Specific Double Spike Single IDA

• Two highly enriched Cr isotopes with different oxidation states were added to the sample prior to alkaline hydrolysis: $^{52}\text{Cr}$(III) and $^{53}\text{Cr}$(VI)
• This ensured species transformation was captured through the whole measurement process
• Highly selective separation of Cr(III) and Cr(VI) species within 20 mins using reversed phase ion pairing liquid chromatography (Bio-inert 1260, Agilent Technologies)
• Peak area determination of $^{52}\text{Cr}$Cr, $^{53}\text{Cr}$Cr using ICP-MS/MS in ammonia reaction mode (8800, Agilent Technologies)
• Isotope pattern deconvolution (IPD) was utilised to establish the degree of interconversion and quantification of the Cr species

Results – Total Cr

$$w_{\text{total Cr}} - w_{\text{sample}} = m_{\text{sample}} - m_{\text{total Cr}} = R_{\text{sample}} - R_{\text{total Cr}}$$

• The characterisation was undertaken using 12 bottles in duplicate, with the analysis split over 2 days
• Quantification via double IDA using the equation above in accordance with ISO/IEC 17025 accreditation

Results – Cr Species

• The Cr species were determined using the species-specific double spike single IDA in combination with IPD calculation
• 12 bottles in duplicate, fully nested design comprising of three runs
• Cr(VI) was not detected under strong reducing conditions induced by the matrix
• The major uncertainty contribution was the batch-to-batch variation (94%) with the remainder attributed to homogeneity

Conclusion

• ERM-BD213a: first reference material for total Cr and Cr species in supplements
• Two different IDA methods were used highlighting the power of the technique
• The total Cr value (305.5 mg/kg) was certified with methodology accredited to ISO/IEC 17025 with low uncertainty (1.6% relative)
• The IPD approach ensured (III)–(VI) transformations were accounted for and the Cr(III) species accurately determined (302 mg/kg) with fit-for-purpose uncertainty (15.6% relative), demonstrating its capability for complex species
• This material represents an important step forwards to support challenging EU regulations

References