Beryllium and its inorganic compounds – Determination of beryllium in urine by atomic absorption spectrometry

Biomonitoring Methods

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Abstract

The working group „Analyses in Biological Materials" of the Permanent Senate Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area verified the present biomonitoring method. This procedure describes a method for the determination of beryllium in urine by means of atomic absorption spectrometry using the graphite furnace technique with a pyrolytically coated graphite tube and Zeeman background correction. The acidified samples are homogenised and injected into the graphite tube without further sample preparation using an autosampler. Magnesium nitrate is used as a matrix modifier. After drying, pyrolysis and atomisation, the absorbance is measured at 234.9 nm. The beryllium concentration is quantified using either a calibration curve (method I) or the standard addition method (method II).
The method was extensively validated and the reliability data were confirmed by independent laboratories, which have established and cross-checked the whole procedure.

Keywords

beryllium; urine; biomonitoring; Analyses in Biological Materials; atomic absorption spectrometry; AAS

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Matrix:	Urine
Hazardous substances:	Beryllium and its inorganic compounds
Analytical principle:	Atomic absorption spectrometry
Completed in:	November 2011

Overview of the parameters that can be determined with this method and the corresponding hazardous substances:

Substance	CAS	Parameter	CAS
Beryllium and its inorganic compounds	7440-41-7	Beryllium	7440-41-7

Summary

This procedure describes a method for the determination of beryllium in urine by means of atomic absorption spectrometry using the graphite furnace technique with a pyrolytically coated graphite tube and Zeeman background correction. The acidified samples are homogenised and injected using an autosampler into the graphite tube without further sample preparation. Magnesium nitrate is used as a matrix modifier. After drying, pyrolysis and atomisation, the absorbance is measured at 234.9 nm.

The concentration of the analyte is determined using either a calibration curve (method I) or the standard addition method (method II).

Reliability of the method

As the calibration procedure can be accomplished with both, calibration curve (method I) or standard addition method (method II), validation data for both procedures were determined.

Beryllium

Within day precision (method I):	Standard deviation (rel.)	s_w = 7.85%
	Prognostic range	u = 17.3%
	at a spiked concentration of 40 ng Be per litre urine and n = 12 determinations	
Within day precision (method II):	Standard deviation (rel.)	s_w = 7.41%
	Prognostic range	u = 16.1%
	at a spiked concentration of 40 ng Be per litre urine and n = 13 determinations	
Day to day precision (method I):	Standard deviation (rel.)	s_w = 14.3%
	Prognostic range	u = 31.9%
	at a spiked concentration of 40 ng Be per litre urine and n = 11 determinations	
Day to day precision (method II):	Standard deviation (rel.)	s_w = 5.83%
	Prognostic range	u = 13.2%
	at a spiked concentration of 40 ng Be per litre urine and n = 10 determinations	
Accuracy (method I):	Recovery rate (rel.)	r = 87.5%
	at a spiked concentration of 40 ng Be per litre urine and n = 12 determinations	
Accuracy (method II):	Recovery rate (rel.)	r = 102%
	at a spiked concentration of 40 ng Be per litre urine and n = 13 determinations	
Detection limit:	1.8 ng Be per litre urine	
Quantitation limit:	6.8 ng Be per litre urine	

General information on beryllium and its inorganic compounds

Beryllium (molecular weight: 9.0122 g, atomic number: 4, melting point: 1287 °C) is a silvery-white, very hard and brittle light metal. Its chemical properties are very similar to those of aluminium. Beryllium is a relatively rare metal in the Earth's crust, with an average content of about 2 to 5 mg/kg. It is most commonly extracted from beryl and bertrandite ores, whose beryllium contents are considered sufficient for production.

Beryllium and its compounds are used in a wide range of applications in many modern industrial sectors. Beryllium-containing alloys are extremely lightweight yet stable. They combine high flexural stiffness and fatigue resistance with an excellent thermal and electrical conductivity [IARC 1993].

Pure beryllium metal and alloys with a higher beryllium content are mostly used in aerospace and defence technology, nuclear power engineering, scientific instrumentation and similar applications.

For the most part, beryllium is used in copper alloys usually containing less than 2% beryllium. These alloys are mainly used to manufacture a variety of components for electrical and electronic applications such as switches, plugs, cable connectors, springs, etc. Another important application of beryllium is as beryllium oxide in ce-

ramics in radio and mobile communications engineering, laser technology and scientific instrumentation. The Monograph of the International Agency for Research on Cancer (IARC) [IARC 1993] provides a good summary of the industrial use and the physicochemical properties of beryllium and its compounds.

Beryllium and its compounds are toxic and present serious health hazards. Epidemiological data showed a relationship between occupational beryllium exposure and the incidence of lung cancer. Therefore Beryllium and its inorganic compounds have been classified in category I of carcinogenic substances by the Commission. Moreover, there is the danger of respiratory and skin sensitisation [DFG 2017]. Beryllium and its inorganic beryllium compounds have been classified within the EU as Group 1B carcinogens [EG 2008] and by the International Agency for Research on Cancer (IARC) as carcinogenic to humans, based on epidemiological data [IARC 1993].

Naturally occurring concentrations of beryllium in the environment are very low. The US EPA estimated the total daily beryllium intake through food and air to be 423 ng, with the largest contributions from food and drinking water (420 ng) and only smaller contributions from inhalation of air and dust (3 ng) [US EPA 1998]. As tobacco contains beryllium, smokers may be exposed to higher levels of beryllium than non-smokers [ATSDR 2002; Minoia et al. 1990; Morton et al. 2011].

Utmost significance is attached to the occupational exposure to beryllium with inhalation being the most important route of occupational exposure. The absorption of beryllium through intact skin contributes only marginally to the overall exposure of occupationally exposed persons. Absorption after oral intake is low [ATSDR 2002; WHO 1990]. Single exposure to high concentrations of beryllium (> 100 µg beryllium/m^3) can cause acute beryllium disease. Repeated exposure to low concentrations of beryllium presents a risk factor for developing chronic beryllium disease (berylliosis), characterised by functional restrictions of the lung as well as systemic effects [Drexler and Greim 2005; Greim 2005].

The smelting and subsequent industrial processing of beryllium ores present the greatest health hazards. Beryllium, however, is not produced in the European Union [Cherrie et al. 2011], but Beryllium and semi-finished beryllium products are imported into the EU and processed. A study conducted in France points to the industries and technologies that are likely to have the highest exposure levels [Vincent et al. 2009]. These primarily include thermal processes such as melting and casting of non-ferrous metals, but also mechanical processes during which beryllium-containing dust may be generated.

Beryllium is primarily excreted in the urine and only to a small extent in the faeces. Animal studies performed on guinea pigs showed that the maximum renal beryllium excretion is reached between 10 and 30 hours after the end of exposure [Stiefel et al. 1980]. As the resorption of beryllium depends on numerous factors, no general correlations between beryllium in the air and in the urine have been derived so far. However, an increased urinary beryllium concentration is an indicator for a previous exposure to beryllium and thus a suitable biomarker of an occupational exposure to beryllium [Drexler and Greim 2005; Schaller 2016].

In urine samples collected from non-occupationally exposed adults in Europe, beryllium could only be detected in individual cases even when using the highly sen-

sitive ICP-MS method. Based on these measurements, a biological reference value (BAR) of 0.05 µg beryllium/L urine was established [DFG 2017; Schaller 2016].

For detailed evaluation of beryllium and its inorganic compounds, see "Toxikologisch-arbeitsmedizinische Begründung von MAK-Werten" and the English translation "Occupational Toxicants" by the Commission [Greim 2005]. A detailed description of the diseases caused by beryllium and its compounds is also given in the "Toxikologisch-arbeitsmedizinische Begründung von MAK-Werten" and the English translation "Occupational Toxicants" by the Commission [Greim 2005] as well as in the documentation of the Scientific Committee on Occupational Exposure Limits (SCOEL) [SCOEL 2017].

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1 General principles

This procedure describes a method for the determination of beryllium in urine by means of atomic absorption spectrometry using the graphite furnace technique with a pyrolytically coated graphite tube and Zeeman background correction. The acidified samples are homogenised and injected using an autosampler into the graphite tube without further sample preparation. Magnesium nitrate is used as a matrix modifier. After drying, pyrolysis and atomisation, the absorbance is measured at 234.9 nm. The concentration of the analyte is determined using either a calibration curve (method I) or the standard addition method (method II).

2 Equipment, chemicals and solutions

2.1 Equipment

- Atomic absorption spectrometer with Zeeman background correction (e.g. Thermo Solaar MQZ, Thermo Scientific)
- Autosampler for automatic dosing of reagents and samples (e.g. autosampler FS95, Thermo Scientific)
- Beryllium hollow cathode lamp or electrodeless discharge lamp depending on instrumentation (e.g. Thermo Scientific)
- Pyrolytically coated graphite tube with or without platform (e.g. Omega or ELC cuvette, Thermo Scientific)
- Microliter pipettes, adjustable between 100 µL and 1000 µL (e.g. Eppendorf, Germany)
- Various volumetric flasks (e.g. Brand, Germany)
- Vortex mixer (e.g. VTX-3000 L Mixer UZUSIO, LMS, Japan)
- Laboratory centrifuge (e.g. Heraeus Megafuge, Thermo Scientific)
- Ultrasonic bath for sample homogenisation (e.g. Bandelin Sonorex RK 102, Germany)
- Vials for the sample changer (vials for samples and reagents, device-specific)
- Plastic vials for sample and reagent storage

2.2 Chemicals

Unless otherwise stated, the quality of all used chemicals must at least be of p.a. grade.

- Beryllium standard solution CertiPUR 1000 mg/L (e.g. Merck, No. 170207)
- Magnesium matrix modifier 10 ± 0.2 g/L (e.g. Merck, No. 105813)
- Ultrapure water
- Nitric acid 65% Suprapur (e.g. Merck, No. 100441)
- Acetic acid 96% for analysis EMSURE (e.g. Merck, No. 100062)
- Argon ≥ 99.999 mol-% (e.g. Alphagaz 1, Air Liquide)

2.3 Solutions

- HNO_3 cleaning solution
  To prepare the HNO_3 cleaning solution, equal volumes of ultrapure water and 65% nitric acid are carefully mixed.
- Nitric acid 2%
  In a 1000 mL volumetric flask, 31 mL of 65% HNO_3 are pipetted to 500 mL ultrapure water. The solution is made up to the mark with ultrapure water.
- Matrix modifier 0.1%
  100 µL magnesium matrix modifier (c(Mg) = 10 ± 0.2 g/L) are diluted with 900 µL of 2% HNO_3 in a sample vial and intensively mixed. The solution must be freshly prepared every day.
- Quality control material
  Quality control material, e.g. Recipe ClinChek® Urine Controls for Trace Elements Level 1 and Level 2 (Recipe, No. 8849)

2.4 Calibration standards

Stock solutions

- Stock solution 1 (1 mg/L)
  100 µL of the beryllium standard solution are pipetted into a 100 mL volumetric flask. The flask is made up to the mark with 2% nitric acid. This solution is stable at 4 °C for at least eight weeks.
- Stock solution 2 (1 µg/L)
  100 µL of the stock solution 1 are pipetted into a 100 mL volumetric flask and made up to the mark with 2% nitric acid. This solution is stable at 4 °C for at least eight weeks.

Working solution (for the autosampler)

- Working solution (250 ng/L):
  200 µL of the stock solution 2 are diluted with 600 µL of 2% HNO_3 in a sample vial and intensively mixed. This solution must be freshly prepared every day.

The programmed dilution stages for both, the calibration by calibration curve (method I) and standard addition (method II), are automatically adjusted by the graphite furnace autosampler using the sample, the working solution and the reagents (2% HNO_3, matrix modifier).

The calibration standards for method I might be optionally prepared manually according to the pipetting scheme in Table 1. For the standard addition method the concentrations of the added solutions have to be established on the basis of the sample absorbance (see Section 6.2). The addition steps shall be chosen such that the absorbance after the last step is approximately twice to triple the absorbance of the unspiked urine sample.

Table 1 Pipetting scheme for the preparation of calibration standards (method I).

Calibration point	Spiked conc. [ng/L]	HNO_3 2% [µL]	Stock solution 2 [µL]
0	0	1000	0
1	10	990	10
2	20	980	20
3	40	960	40
4	60	940	60
5	70	930	70
6	90	910	90

2.5 Cleaning of the equipment and testing for contamination

For cleaning the laboratory equipment, the HNO_3 cleaning solution prepared according to Section 2.3 is used. The vials used should be soaked for at least two hours before washing with ultrapure water and drying.

Random sampling is performed to ensure that the vials and single-use devices (e.g. pipettes, vials, etc.) are free of contamination. Laboratory glassware can be a potential source of contamination and should therefore not be used. Moreover, all chemicals used have to be tested for blank values.

3 Specimen collection and sample preparation

3.1 Specimen collection

The prevention of contamination during specimen collection and analysis is of crucial importance for the quality of the analytical results. So far, no beryllium contamination has been detected with customary urine containers.

Urine samples are taken from spontaneous urine. The urine samples are collected at the end of exposure, at the end of a work shift or in the case of several preceding shifts with exposure, in the morning following the last shift [Schaller 2016]. It is imperative that the urine samples are collected in an appropriate and clean environment and with the greatest care.

The collected samples are acidified with acetic acid (1 mL acetic acid per 100 mL urine) and stored in the refrigerator at 4 °C. Under these conditions, the samples are stable for at least four weeks. For long-term storage, the samples must be frozen at –24 °C.

3.2 Sample preparation

For sample preparation frozen urine samples are thawed at room temperature or at approx. 40 °C. Prior to removing aliquots, the samples must be homogenised in an

ultrasonic bath. 1 mL of each urine sample is pipetted into a sample vial and placed into the autosampler.

4 Operational parameters

4.1 Atomic absorption spectrometer

Wavelength:	234.9 nm
Spectral slit width:	0.5 nm
Lamp current:	50 to 80% (according to the manufacturer's instructions)
Inert gas:	Argon
Measuring mode:	peak height
Graphite furnace:	with platform (Omega)
Background correction:	Zeeman
Matrix modifier:	10 µL
Standard preparation:	constant volume

The instrumental settings are identical for both methods, I and II, except for the sample volume.

Sample volume method I:	35 µL
Sample volume method II:	25 µL

The temperature program and inert gas flow must be optimised for each individual instrument. The program given in Table 2 was optimised for the Solaar MQZ system with graphite furnace and autosampler FS95, Thermo Scientific. Other instrument settings are possibly required for instruments of different manufacturers.

Table 2 Graphite furnace program.

Analytical step	Hold time [s]	Temperature [°C]	Ramp [°C/s]	Gas type	Gas flow [L/min]
Drying	45	135	2	Argon	0.2
Pyrolysis	20	1400	150	Argon	0.2
Atomisation	3	2700	0	Argon	off
Heating	3	2700	0	Argon	0.2

5 Analytical determination

For the analytical determination the prepared samples according to Section 3.2 as well as the working solution (Section 2.4), nitric acid 2% and the matrix modifier (Section 2.3) are placed into the autosampler. Dosing of the solutions and reagents and the transfer of the samples into the graphite tube is done by the autosampler. The beryllium level of each sample is determined by atomic absorption spectrometry.

6 Calibration

6.1 Calibration curve (method I)

The use of the calibration curve method is recommended for measurements at the lower end of the concentration range (10 to approx. 75 ng/L) or to get an overview of the sample concentrations. If sample concentrations are above the calibration range, the sample can be diluted, another calibration can be carried out for the extended measuring range or the sample can be analysed using the standard addition method. Upon introduction of this method and at regular intervals, however, at least once a year, the analytic function is determined using a six-point calibration. For that purpose, the working solution (Section 2.4) is placed into the graphite furnace autosampler. The concentrations of 10, 20, 40, 60, 70 and 90 ng/L are automatically dosed by the autosampler. At each calibration point, measurement is carried out in triplicate. Alternatively, the calibration standards for the six-point calibration can be prepared manually (Table 1). Figure 1 (in the Appendix) shows an example of the calibration graph.

In daily routine, for each run a three-point calibration (e.g. 10, 30 and 80 ng/L) is carried out with each concentration and the blank being measured three times. The calibration graph has to be checked at the end of each analytical run.

The calibration graph method has the advantage that larger sample volumes (35 µL) can be used, whereas the standard addition method only allows the use of volumes of up to 25 µL.

6.2 Standard addition (method II)

The standard addition procedure is carried out by adding working solution in stepwise increasing amounts. The three addition steps shall be chosen such that the absorbance after the last step is approximately twice to triple the absorbance of the unspiked urine sample. Afterwards, the standard addition function is derived from those measurements. The autosampler of modern atomic absorption spectrometers can normally be programmed to perform standard addition calibrations.

Figure 2 in the appendix shows a chart of a standard addition function that was processed using a urine sample of a non-ferrous founder with a beryllium level of 58 ng/L. Furthermore, in Figures 3 and 4 atomisation curves of the standard addition method of both, spiked urine as well as a urine sample of a beryllium exposed worker, are given.

The calibration curve and the standard addition function are calculated and plotted by the software. Apart from the analytic function, the coefficient of determination is recorded. The coefficient of determination must be greater than 0.995. Non-linear analytic functions are not accepted.

7 Calculation of the analytical results

7.1 Calibration curve (method I)

The calibration standards are measured and a calibration graph is obtained by plotting the beryllium concentration of the standards on the abscissa and the absorbance on the ordinate. Using the calibration function corresponding to the analytical run, the analyte concentration in ng/L urine can be calculated.

7.2 Standard addition (method II)

The absorbance of each solution is measured. The concentration of the added beryllium is plotted on the abscissa and the absorbance on the ordinate. The calibration graph obtained by linking the plotted points is extended and the analyte concentration of the unknown sample is taken from the intersection of the calibration graph on the abscissa.

Normally, the analytical results are calculated automatically by the software.

8 Standardisation and quality control

The quality control requirements for biomonitoring studies are laid down in the AMR (Arbeitsmedizinischen Regel) 6.2 [AfAMed 2013]. In addition, the guidelines of the Bundesärztekammer (German Medical Association) and a general chapter of the MAK-Collection for Occupational Health and Safety Part IV: Biomonitoring Methods [Bader et al. 2010; Bundesärztekammer 2008] must be observed.

For internal quality control at least one QC-sample is analysed within each analytical run. The concentration of this control material should lie within the relevant concentration range. Control urine is for example commercially available at two concentration levels from Recipe or can be prepared in the laboratory. For that purpose, urine of healthy, non-exposed persons (the urine must not contain any beryllium) is spiked with a defined quantity of beryllium. The urine can be collected from several persons and pooled. Aliquots of this material can be stored frozen at –24 °C for at least one year.

The nominal value and the tolerance ranges of this quality control material are determined in a pre-analytical period (one analysis each of the control materials on ten different days) [Bader et al. 2010].

External quality control can be done by participating in round robin tests. Round robin analyses in occupational and environmental toxicology are carried out on a regular basis on behalf of the German Society of Occupational and Environmental Medicine (Deutsche Gesellschaft für Arbeits- und Umweltmedizin e. V., DGAUM). This scheme also encompasses the determination of beryllium in urine.

9 Evaluation of the method

9.1 Precision

To determine the within day precision, urine was spiked with beryllium at 10 ng/L and 80 ng/L, respectively, prepared and analysed. The quantitation was carried out using a six-point calibration (method I) according to Section 6.1. Based on the ten-fold determination of these urine samples, the precision data given in Table 3 were obtained.

Table 3 Within day precision for the determination of beryllium in urine (n = 10) (method I).

Spiked concentration [ng/L]	Standard deviation (rel.) [%]	Prognostic range [%]
10	7.89	17.8
80	12.5	28.3

In routine practice a three-point calibration including the blank value was performed. Tables 4 and 5 show the within day and day to day precision data achieved under these conditions for both, method I and method II. The beryllium solutions used were prepared by spiking clear urine that did not contain measurable levels of beryllium.

Table 4 Within day precision data for the determination of beryllium in urine under routine analysis conditions using method I (n = 12) or method II (n = 13).

	Spiked concentration [ng/L]	Standard deviation (rel.) [%]	Prognostic range [%]
Within day precision (method I)	40	7.85	17.3
Within day precision (method II)	40	7.41	16.1

Table 5 Day to day precision data for the determination of beryllium in urine under routine analysis conditions using method I (n = 11) or method II (n = 10).

	Spiked concentration [ng/L]	Standard deviation (rel.) [%]	Prognostic range [%]
Day to day precision (method I)	40	14.3	31.9
Day to day precision (method II)	40	5.83	13.2

9.2 Accuracy

The accuracy of the method was tested by recovery experiments. For this purpose, urine was spiked at a level of 40 ng/L with beryllium and analysed under routine

conditions. The mean relative recovery rates for quantitation with method I and method II were found to be 87.5% and 102%, respectively.

9.3 Detection limit and quantitation limit

The limit of detection and the limit of quantitation, respectively, were determined based on DIN 32 645 using six-point calibrations. Based on the standard deviation of the obtained calibration graph a detection limit of 1.8 ng/L and a quantitation limit of 6.8 ng/L was evaluated.

As the sensitivity of the method depends on the wear of the graphite tube, the effective limit of detection and quantitation must be calculated for each run or standard addition using the calibration curve. For example, the developer of the method determined the mean limit of detection and the mean limit of quantitation in everyday routine using a three-point calibration to be 3.17 ng/L (range: 0.2–9.0 ng/L) and 12.5 ng/L (range: 0.8–35.3 ng/L), respectively (Table 6).

Table 6 Detection and quantitation limit for the determination of beryllium in urine.

	Six-point calibration (n = 3) [ng/L]	**Three-point calibration in routine analysis (n = 59) [ng/L]**
Detection limit	1.8	3.17 (range 0.2–9.0)
Quantitation limit	6.8	12.5 (range 0.8–35.3)

9.4 Sources of error

The sensitivity of the method depends on the graphite furnace and on the urine matrix.

The quality and the wear of the graphite furnace determine whether an adequate precision and accuracy at the lower end of the concentration range is achieved. Generally, optimal results are achieved after a new graphite tube is installed and conditioned by several standard measurements. After a relatively small number of measurements (approx. 300) the procedure's sensitivity considerably decreases and the graphite tube has to be replaced. For this reason, three-point calibration including the blank is used in everyday routine.

With individual samples, results may vary considerably due to matrix interferences. In this case, an UV digestion using hydrogen peroxide is recommended, under which the reagents and vials used have to be tested for blank values.

Method I and II may be applied for mutually checking results. The use of the Zeeman background correction is part of the method.

10 Discussion of the method

As only very low beryllium concentrations are present in the environment, the background exposure level of the general population is very low and cannot be detected with the given methods. However, the optimised and cross-checked methods described here, can be reliably used for the occupational medical and toxicological concentration range.

Care must be taken to avoid contaminations in taking samples, during transport and in storage. The risk of contamination from the environment is very low, as only very low beryllium concentrations are present in here. Contamination at the workplace, however, must be prevented. The urine samples should therefore be collected after showering and changing into street clothes.

The analytical determination of beryllium in urine must meet the criteria for analysis at ultratrace level. Especially, no glassware but pre-cleaned plastic vessels should be used for urine sampling and storage and for storage of the needed solutions.

To achieve the low detection and quantitation limit described within the method, high-performing instrumentation is needed. The pyrolytically coated graphite tube can be used with or without platform. Comparative studies with atomic absorption spectrometers of various manufacturers showed that improvements could be achieved by choosing an appropriate tube. Moreover, the addition of Triton X-100 (0.1% in water) to the samples was tested. The use of the detergent did not improve the performance characteristic of the methods.

The advantage of the calibration curve method (method I) over the standard addition method (method II) is that a larger number of samples can be analysed with the same graphite tube. In case of scattering measured values or of obvious matrix effects, method II can be used to verify the results. Using the described methods, the measurement conditions are constantly changing due to the wear of the graphite tube. As a new graphite tube produces optimal results only after a conditioning phase, quality assurance measures are of particular importance.

These include re-calibration at the end of an analytical run when using the calibration curve method and the analysis of at least one quality control sample per run.

Among other things, the sensitivity of the method depends on the wear of the graphite tube. As a result, the limit of detection and quantitation vary for each run and standard addition, respectively. The developer of the method found a mean limit of quantitation of 12.2 ng beryllium per litre urine (range: 0.8–35.3 ng/L; n = 59) in routine analyses (see Section 9.3), which enables the reliable determination of the biological reference value (BAR) of 0.05 µg beryllium/L urine. The difference between the limit of quantitation established based on DIN 32 645 and the one established as part of routine is small (factor 2). Therefore, the method is to be considered reliable and suitable for routine applications.

Instruments used:

Atomic absorption spectrometer Thermo Solaar MQZ with Zeeman background correction and autosampler FS95, Thermo Scientific

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12 Appendix

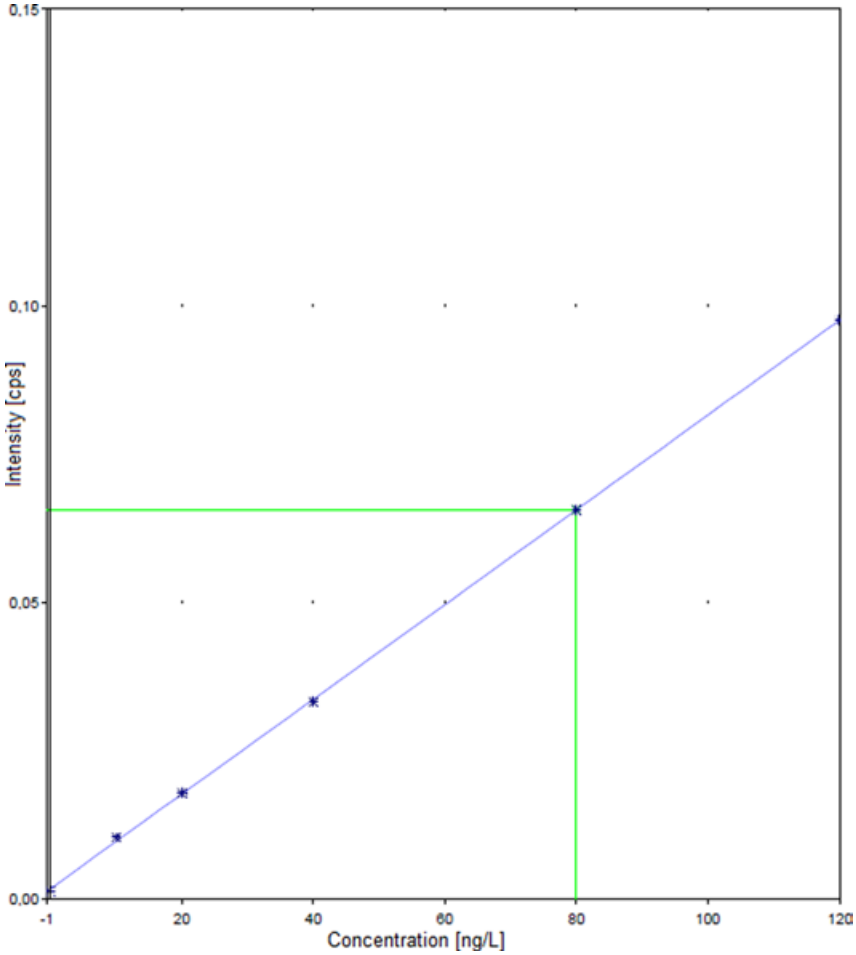

Figure 1 Calibration graph for the determination of beryllium in urine (method I) in a concentration range of up to 120 ng/L (Omega cuvette).

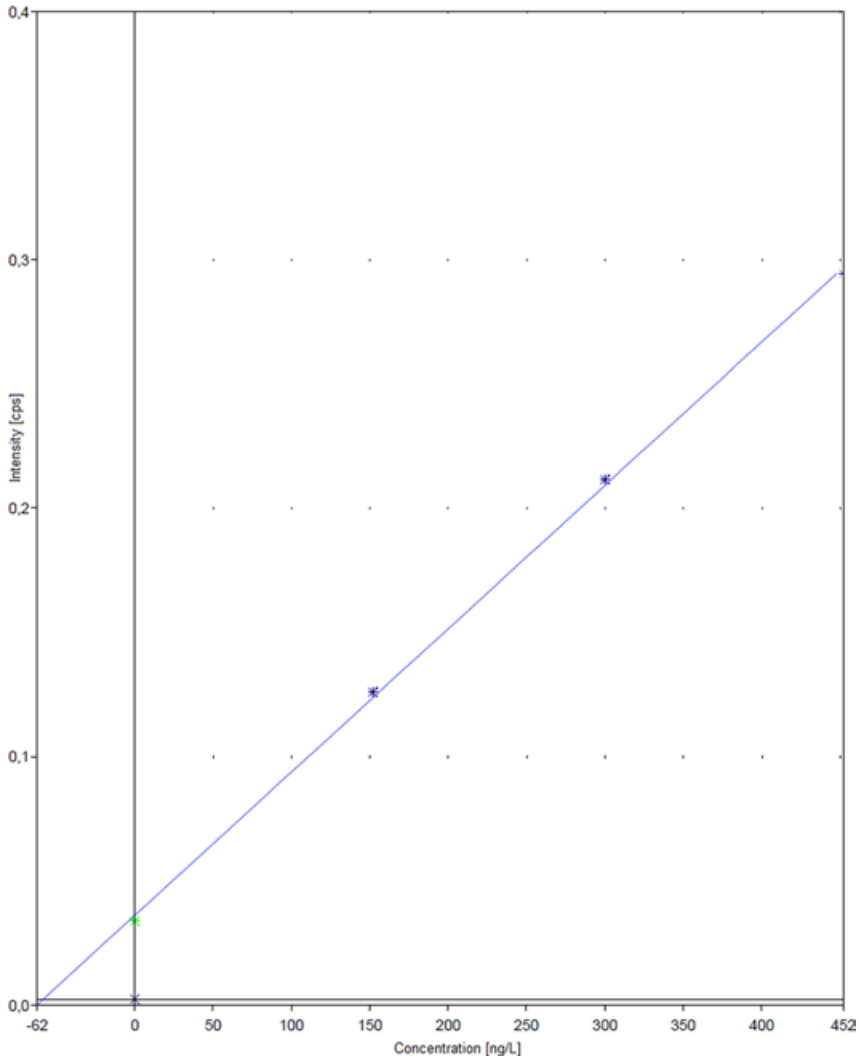

Figure 2 Plot of a standard addition to urine of an exposed non-ferrous founder with a quantified beryllium level of 58 ng/L (ELC cuvette).

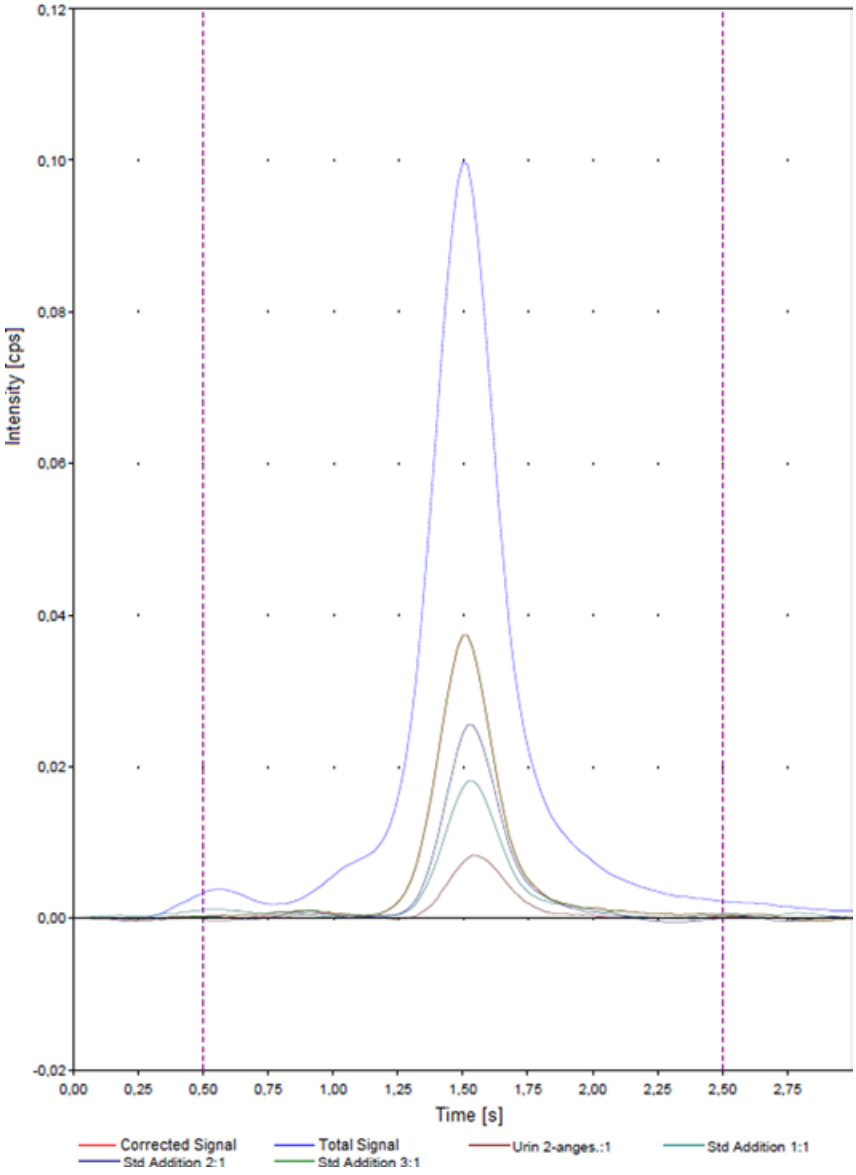

Figure 3 Peak profiles by standard addition using a urine sample spiked with 15 ng/L beryllium (Omega cuvette).

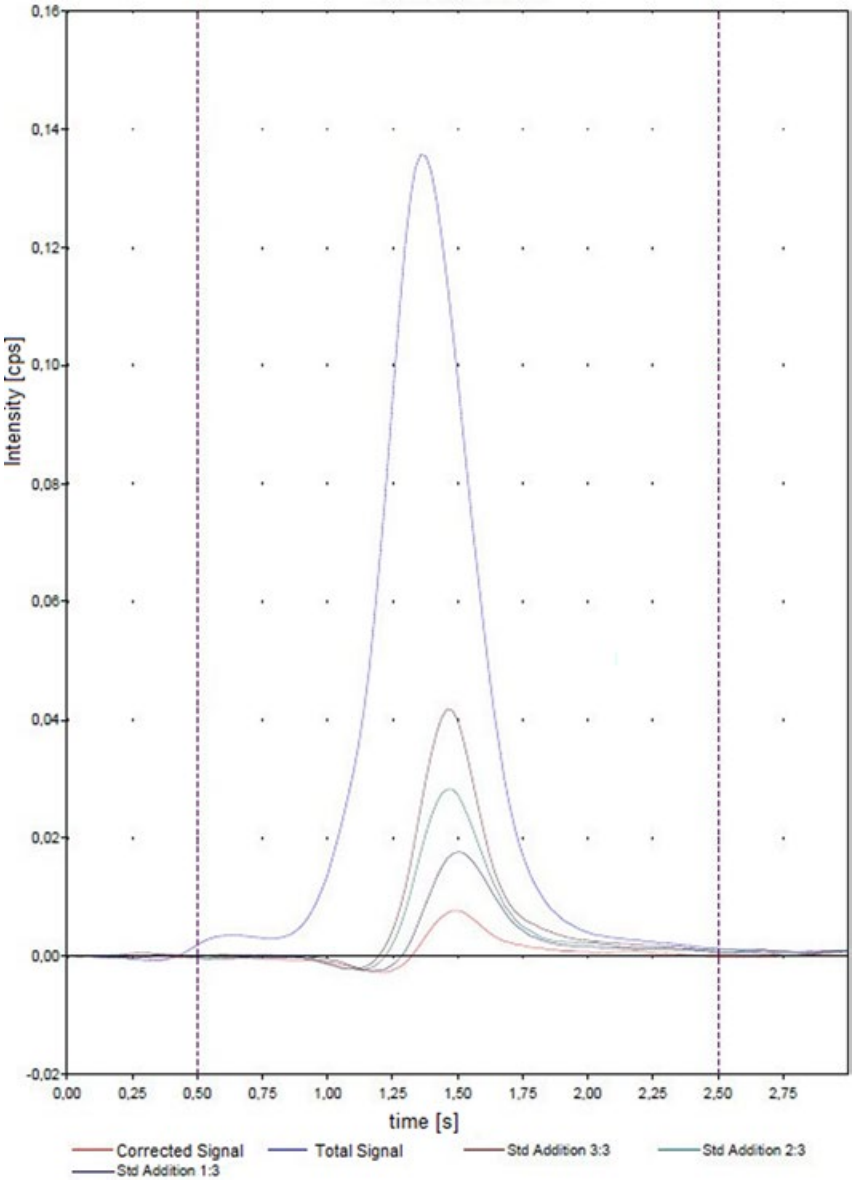

Figure 4 Peak profiles by standard addition using urine of an exposed non-ferrous founder with a quantified beryllium level of 13 ng/L (Omega cuvette).