Eurasian Journal of Physics and Functional Materials

Volume 7 | Number 3

Article 5

Pages: 175 - 184

9-22-2023

Selection of an internal standard to determine mass concentration of uranium in human urine by inductively coupled plasma mass-spectrometry

V. V. Kolbin
Institute of Radiation Safety and Ecology, National Nuclear Center RK ,Kazakhstan

M. T. Dyuisembaeva
Institute of Radiation Safety and Ecology, National Nuclear Center RK ,Kazakhstan

N. Zh. Mukhamediyarov
Institute of Radiation Safety and Ecology, National Nuclear Center RK ,Kazakhstan

A. Zh. Tashekova
Institute of Radiation Safety and Ecology, National Nuclear Center RK ,Kazakhstan

G. M. Yesilkanov
Institute of Radiation Medicine and Ecology ,Kazakhstan

See next page for additional authors

Recommended Citation

Kolbin, V. V.; Dyuisembaeva, M. T.; Mukhamediyarov, N. Zh.; Tashekova, A. Zh.; Yesilkanov, G. M.; Shakenov, Ye. Z.; and Temirzhanova, A.Ye. (2023) "Selection of an internal standard to determine mass concentration of uranium in human urine by inductively coupled plasma mass-spectrometry," *Eurasian Journal of Physics and Functional Materials*: Vol. 7: No. 3, Article 5.
DOI: https://doi.org/10.32523/ejpfm.2023070305

Selection of an internal standard to determine mass concentration of uranium in human urine by inductively coupled plasma mass-spectrometry

Authors

V. V. Kolbin, M. T. Dyuisembaeva, N. Zh. Mukhamediyarov, A. Zh. Tashekova, G. M. Yesilkanov, Ye. Z. Shakenov, and A.Ye. Temirzhanova

Eurasian Journal of Physics and Functional Materials

2023, 7(3), 175-184

Selection of an internal standard to determine mass concentration of uranium in human urine by inductively coupled plasma mass-spectrometry

V.V. Kolbin*[,1], M.T. Dyuisembaeva[1], N.Zh. Mukhamediyarov[1],
A.Zh. Tashekova[1], G.M. Yesilkanov[2],
Ye.Z. Shakenov[1], A.Ye. Temirzhanova[1]

[1]Institute of Radiation Safety and Ecology, National Nuclear Center RK, Kurchatov, Kazakhstan

[2]Institute of Radiation Medicine and Ecology, Semey, Kazakhstan

E-mail: kolbin@nnc.kz

DOI: **10.32523/ejpfm.2023070305**
Received: 15.09.2023 - after revision

The paper presents experimental studies on the choice of an internal standard in order to correction the matrix effect that occurs when determining uranium in human urine by inductively coupled plasma mass spectrometry. Monoelemental standard solutions of Sc-45, Rh-103, In-115, Ir-193, Th-232 were used as internal standards. The levels of the decrease in the sensitivity of analytical signals of internal standards and uranium spikes under the influence of the matrix effect were determined. The ratio of the measured concentration of internal standards in the background solution of 5% nitric acid and in the matrix of the urine simulator was found compared to uranium. Based on this, empirical coefficients were calculated that characterize the difference in the matrix influence on analytical signals of the internal standard and uranium signals. Adjustment applied was verified according to the internal standard using calculated empirical coefficients and measured values of analytical signals of the internal standard in samples and in the background solution. Iridium was identified as the most suitable of the elements listed as an internal standard according to various criteria. Based on experimental results, a procedure was developed for measuring uranium in urine using iridium as an internal standard.

Keywords: human urine; inductively coupled plasma mass spectrometry; internal standard; matrix effect; iridium; uranium

Introduction

According to the World Nuclear Association [1], Kazakhstan has 1.7 million tons of known reserves and resources of uranium. Accordingly, the development and application of techniques for determining the uranium content in the human body is a highly relevant task. Uranium is able to accumulate in organs-concentrators [2], including kidneys, and is excreted from the body with urine. In this way, the presence of uranium in the urine can serve as the indicator of the accumulation of this element in the body [3].

The procedure of determining the mass concentration of uranium in human urine is supposed to be applied as part of the monitoring in order to assess and control exposure doses. This is relevant to the population living in the impact zone of manufactured and natural objects as well as to control doses to personnel affected by occupational exposure and directly working with uranium-containing materials. On the example of Kazakhstan as the lead uranium mining country the demand of periodic monitoring the uranium content in the human body for workers in mining industry is legally enshrined as a highly relevant task.

The methodological scheme for determining the uranium content in human urine is based on the inductively coupled plasma quadrupole mass spectrometry (ICP-MS) technique. The topic is relevant and nowadays the choice of internal standard is being reinvestigated in ICP-MS. This technique is more effective for measuring uranium in human urine compared to a laser-luminescent, radiometric or electrochemical technique [4]. In comparison with already existing analytical methods such as alpha-spectrometric, the ICP-MS method has significant advantages in amount of time for sample preparation (3–4 hours versus >10 days), analysis (2–3 hours versus 5–7 days). Additional advantage includes the sample batch size (20–30 samples for ICP-MS method versus 1–12 for alpha-depends of the amount of chambers). Moreover, 1.5 L of sample volume is needed for equal detection limit for alpha-spectrometric analysis while ISP-MS method needs only 20 mL.

Matrix effects are some of the major causes of interference in the accurate determination of the content of elements in mass spectrometry. The significant influence of the biomatrix on the analysis results is a specific nature of the determination of uranium in urine. The matrix effect is a reason why responses of the same analyte are different when comparing samples prepared for the analysis in a biological object and in a pure solvent [5]. It is expressed in the suppression of the ion signal of the analyte and depends on the concentration and atomic mass of an element to be determined and matrix elements. This is the cause of the unaccounted matrix effect since urine is a complex biological object that contains organic and inorganic components [6]. Systematic matrix understatement of uranium concentration in urine has a linear relationship and does not exceed 20%. In accordance with the experimental data [7], it was found that the presence of salts has the greatest effect on the suppression of signals during the analysis.

Experiments on the choice of an internal standard make it possible to expand the use of analytical capabilities of laboratories, increase the accuracy and sensitivity of their methodological functionality, and effectively assess the contribution of uranium dose to the internal exposure of people. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Materials and methods

Equipment and reagents

In the course of experiments, mixed solutions of an artificial urine simulator with 0.1; 0.5; 1; 5; 10; 50 μg/dm^3 uranium additives were used. A direct 10-fold dilution of samples and adjustment by the internal standard were used to assess and neutralize the matrix effect. The artificial matrix solution of a urine simulator corresponds to the average daily composition of human urine [8], and is sufficiently stable to conduct a series of experiments without precipitating. Sc, Rh, In, Ir, Th were chosen as internal standards as they are commonly used in the global practice in accordance with the literature data [9–11].

The uranium content in urine simulator samples was diluted by a 5% nitric acid (HNO_3) solution in the ratio of 1:10. Concentration of internal standard was 1 μg/dm^3. Mass spectrometric measurements were performed with an Agilent 7700x quadrupole mass spectrometer (Agilent Technologies). In the process of experimental studies, the optimal instrumental operating mode of the mass spectrometer was chosen. Device parameters and a measurement technique are given below.

– power input to plasma: 1.450–1.550 W;
– the flow rate of aspirated gas (argon): 1.07–1.09 l/min;
– scanning type: peak scanning (3 points);
– detection mode: dual;
– number of scans per measurement: 100;
– number of replications: 3;
– delay before readout: 20 s;
– measurement time: 1 s;
– flush time: 40 s.

Urine simulator preparation

The certified reference material with urine matrix was not used. Instead of that, the urine simulator was prepared based on the artificial matrix. The solution of a urine simulator was prepared in a glass beaker continuously heated at a temperature of 35° C by sequentially spiking calculated concentrations of glucose (5% solution), urea (CH_4N_2O, pure, assay <98.5%) and macroelements (Na, K, Ca, Mg) as well as orthophosphoric and sulfuric acids and aqueous ammonia. To adding of macroelements were used Calcium Chloride 10% w/v Intravenous Infusion, Potassium nitrate, 98%, pure, Magnesium Sulfate 25% w/v solution for infusion, Isotonic solution of Sodium Chloride 9% w/v Injection. The level of pH of the solution was taken into account. Deionized water acted as a background. Once the necessary components are added, a prepared solution has to undergo infusion and filtration stages. The content of macrocomponents in the urine simulator solution was certified with an iCap 6300 atomic emission spectrometer given a 1:100 dilution and calibration lines plotted with points of 1 and 5 mg/dm^3.

Results and discussion

In the process of developing and implementing the technique, sequential series of measurements were performed in accordance with the experimental plan, and a solution of an artificial urine simulator solution was prepared and certified. Research into the effect of matrix, dilution, techniques of sample preparation, spectral cross-overs and equipment parameters on analytical results was undertaken. Various internal standards, measurements of subranges of certified uranium mixtures were selected and compared with concentrations from 0.1 to 50 µg/dm^3. The uranium detection limit, expanded measurement uncertainty and prompt control of analytical quality were defined. Statistical data were gathered for calculation the quality control parameters for measurement procedure in accordance with RMG 76-2014 GSI [12].

Preparation and certification of the matrix solution of the urine simulator

Table 1 provides the averaged certification result on the content of macrocomponents in the urine simulator solution measured in three replications.

The estimated concentration of certified values in the urine simulator solution: Na – 2.36 g/dm^3, K – 2.09 g/dm^3, Ca – 0.12 g/dm^3, Mg – g/dm^3 according to certification results, the deviation of certified values from the estimated concentration of macrocomponentss in the urine simulator solution did not exceed 8.6%, which ranges within the matrix composition of $\pm 30\%$ [4].

Evaluation of the matrix effect of the urine simulator on measured elements

The effect of urine simulator solution on various elements was evaluated to determine the most suitable element to be used as an internal standard given the matrix effect similar to that of uranium. To assess the matrix effect on the internal standard, a series of preliminary experiments were conducted using the Agilent ICP-MS technique in 5% HNO_3 and in a urine simulator solution in 12 replications (n=12). The following monoelement solutions of standard samples (SS) were used as internal standards: Sc-45, Rh-103, In-115, Ir-193, Th-232. All the elements including uranium were added to solutions of the urine simulator and measured separately. Mean, median values and standard deviation (SD) were calculated (Table 2).

Table 1.
Certified content of macrocomponents in the urine simulator solution.

Value	Ca		K		Mg		Na	
	g/dm^3	Deviation, %	g/dm^3	Deviation, %	g/dm^3	Deviation, %	g/dm^3	Deviation, %
Human biomatrix [11]	0.1–0.2		1.3–2.3		0.04–0.1		2.0–3.5	
Estimated concentration	0.12		2.09		0.16		2.36	
Certified value	0.12	0.46	2.27	0.32	0.16	0.23	2.30	0.53
Deviation, %	0.0		8.6		0.0		2.5	

Table 2.
The matrix effect of the urine simulator on analytical signals of uranium and internal standards (n =12).

Element	Concentration in 5% HNO_3		Concentration in urine simulator	
	Median, µg/dm^3	Deviation. %	Median, µg/dm^3	Deviation, %
Sc	0.89	1.69	1.09	3.60
Rh	0.94	1.49	0.65	4.90
In	0.94	1.59	0.75	4.20
Ir	1.01	1.26	0.69	2.27
Th	0.98	1.57	0.69	4.53
U	1.01	1.37	0.71	8.69

The matrix effect causes a measured signal for each element including uranium to be understated by 3–9%. Only for Sc measurements in the matrix were higher than in the background solution (in a 5% nitric acid). Overall, heavier elements are affected by the matrix similarly to uranium. The similar matrix effect is most observed for thorium and rhodium compared to uranium, about 10%. At the same time, when measuring iridium, there is hardly any sensitivity drop of the analytical signal compared to other elements. For indium, vice versa, measured intensities are overstated.

Determination of the ratio of urine simulator solution: 5% HNO_3. Calculation of the empirical coefficient

Uranium additive was measured (1 µg/dm^3) in the matrix of the urine simulator and in a 5% HNO_3 (n =12). The ratio of uranium content measured in the matrix to its content in a 5% HNO_3 was determined; obtained data was compared to measurements of internal standards. Figure 1 provides a graphical distribution of derived ratios for each individual measurement of an element.

Iridium and thorium are closest to uranium by the ratio of measurements of elements in a 5% HNO_3 and in the urine simulator solution. For rhodium, a similar ratio of measured intensities is also observed in the matrix and in a 5% HNO_3. However, a value determined for Rh in a 5% nitric acid is lower than the ones for U, Th and Ir.

Based on derived ratios, empirical coefficients C_{emp} (the difference of the matrix effect on analytical signals of the internal standard and uranium signals) were calculated for each internal standard (Table 3).

Table 3.
Empirical coefficients (the difference of the matrix effect on analytical signals of the internal standard and uranium signals).

	Sc	Rh	In	Ir	Th	U
% of deviation	58%	99%	86%	101%	96%	100%
C_{emp}	1.423	1.009	1.137	0.990	1.038	1

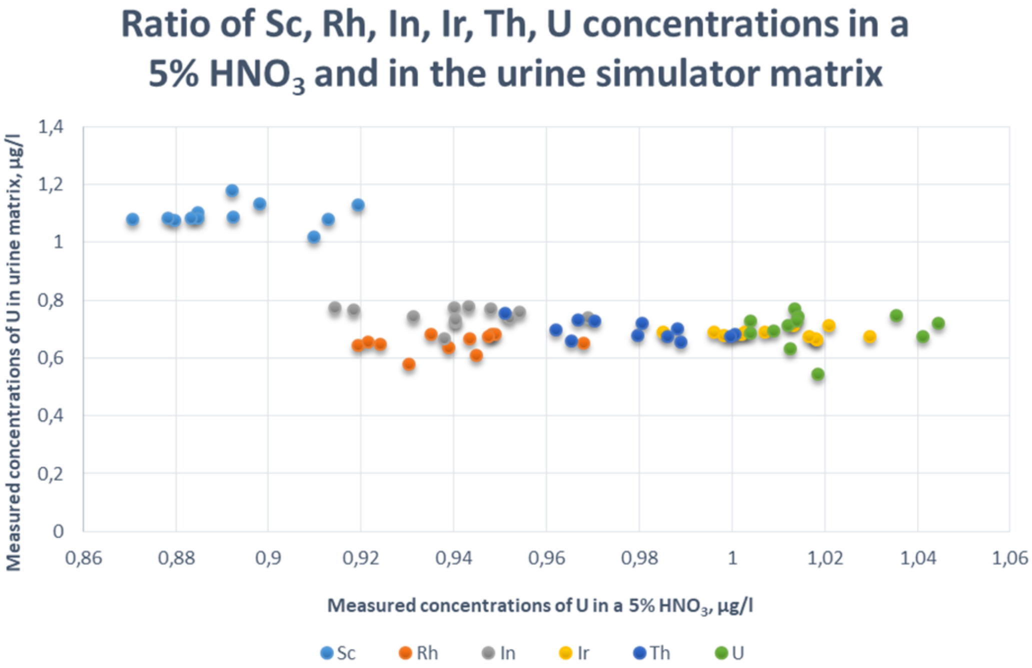

Figure 1. The ratio of concentrations of elements measured in a 5% HNO $_3$ and in the urine simulator solution.

The percentage of deviation was determined as the ratio of the change in the sensitivity of the analytical signal of uranium obtained during measurements to the change in the sensitivity of the internal standard. Thus, the empirical coefficient for uranium is equal to 1 (100%). According to the data listed in Table 3, values of rhodium and iridium are closest to unity (the matrix effect on the analytical signal of uranium) among the listed elements.

Uranium adjustment in the urine simulator solution by the internal standard

Uranium concentration in the urine simulator solution adjusted for the internal standard was determined as per the following formula:

$$X = (k * C * I_s)/I_x, \tag{1}$$

where k – empirical coefficient representing the difference of the matrix effect on analytical signals of the internal standard and uranium signals, C – uranium concentration in a sample calculated with a mass-spectrometer software, µg/dm ^{3}, I_s – measured intensity of the ion signal of the internal standard in the comparison solution prepared with a 5% nitric acid (counts per second - CPS), I_x – measured intensity of the ion signal of the internal standard in a sample solution (CPS).

According to the data of experiments, the narrowest spread of values and deviation percentage of uranium measurements in the matrix of the urine simulator when applying adjustment by the internal standard is observed when iridium is used. For thorium and rhodium, the deviation median is equal to zero, but the spread of values is wider than for iridium.

Choice of the internal standard

Based upon the experimental data, elements were determined that are the most suitable to be used as an internal standard for adjusting the matrix effect on uranium content. Determinations of uranium in the urine simulator solution with a 0.5 μg/L uranium additive closest to certified values were obtained after adjustment for iridium and thorium. For a matrix with a 1 μg/L additive – for rhodium and thorium. In this case, MSD of uranium determination in the urine simulator solution without being adjusted for the internal standard is 8.69%. The lowest averaged percentage of deviation from the certified uranium content is observed after adjustment for thorium, the highest – for rhodium. Accordingly, iridium and thorium may be the most appropriate internal standards to adjust for the matrix effect when determining uranium in urine. Nevertheless, since thorium is an element of the nuclear cycle and requires a license, Ir-193 was chosen as an internal standard for the technique being developed.

Table 4 shows adjusted results of 0.1–50.0 μg/dm^3 uranium additive added to the matrix of the urine simulator using iridium as an internal standard. This solution with uranium additive was measured in two replications, 10 measurements for each replication (in total, n =20). The process was divided into subranges of 7 measurements each adjusted for the internal standard based upon iridium intensities (CPS) in the comparison solution that was measured prior to each subrange. The empirical coefficient that represent the difference of the matrix effect on analytical signals of the internal standard and uranium signals was 0.99.

Table 4.
Adjusted concentration of uranium in the urine simulator solution (an additive of 0.1–50.0 μg/dm^3) for Ir internal standard.

Uranium additive, μg/dm^3	Measured concentration, μg/dm^3 ±Δ (n=20)	Adjusted concentration, μg/dm^3	Expanded uncertainty, $U(X)$ %
0.1	0.08±0.03	0.11	12.99
0.5	0.30±0.11	0.47	12.71
1.0	0.90±0.25	1.11	12.57
5.0	4.08±1.19	5.53	12.15
10.0	8.59±2.40	10.45	12.15
50.0	44.08±12.26	53.29	12.33

Based upon measurements the average deviation of findings on the uranium content after adjustment for the internal standard for the solution of the urine simulator matrix addied with uranium was within 10%. The size of subranges was reduced to 5 consecutive measurements, after which the iridium solution in a 5% HNO_3 was remeasured. Overall, the mean and median deviation of the uranium content in the urine simulator solution with uranium added and adjusted for the internal standard, is 5% (based on 120 measurements). In the course of measurements, an increase in pulses (CPS) is observed in each test solution, which is neutralized by occasional recalibration (every 10–14 samples).

Based upon experimental results, a technique was developed to determine uranium in human urine in the range from 0.10 μg/dm^3 to 50 μg/dm^3 by inductively coupled plasma mass spectrometry (ICP-MS) using an internal iridium standard. The relative overall standard uncertainty of measurements is calculated as the square root of the sum of squares of the standard uncertainty of type A and B [13]. The type B uncertainty to analyze uranium in urine was 5.6%. To express results of the analysis in documents providing for the use thereof, limits of the absolute bias of the uranium concentration in the urine sample, μg/dm^3, were determined at a confidence level of P=0.95 being equal to ±23%.

Conclusions

Based upon experiments with five internal standards (Sc-45, Rh-103, In-115, Ir-193, Th-232), iridium was determined as the most suitable for adjusting the effect of urine biomatrix on the uranium content. Experiments have shown that the selected internal iridium standard is defined by the matrix effect similar to that of uranium and the similar ratio in assessing the concentration being determined in the urine simulator solution and in a 5% nitric acid. The median of the deviation when adjusting for the internal standard is 1%; there is also hardly any sensitivity drop for iridium as affected by the urine simulator solution. In addition, it should be noted that iridium is not an element of the nuclear fuel cycle what should be considered as an advantage. Applying of developed measuring method will be appropriate for mass and effective monitoring procedures of detect and control exposure doses without using elements of the nuclear fuel cycle.

Among disadvantages to other internal standards, one can mention that its weight is not so close to that of uranium (compared to thorium). The deviation median when adjusting for iridium is somewhat higher than those of other internal standards.

References

[1] NEA, Uranium 2018: Resources, Production and Demand (Paris: OECD Publishing, 2018). [*Web Link*]

[2] N.V. Baranovskaya et al., Bulletin of the Tomsk Polytechnic University **339** (2010) 182–188. [*WebLink*] (In Russian)

[3] S. Jackson, G.W. Dolphin, Health Physics **12**(4) (1966) 481–500. [*CrossRef*]

[4] N.V. Agaeva et al., Bulletin of NAS RK **3**(393) (2012) 14. [*WebLink*] (In Russian)

[5] D.V. Yaroshenko, L.A. Kartsova, Journal of Analytical Chemistry **69** (2014) 311–317. [*CrossRef*]

[6] N.B. Ivanenko et al., Toxicology **13** (2012) 871–880. [*WebLink*] (In Russian)

[7] T.G. Kirillova et al., Bulletin of the NNC RK **1** (2019) 85–90. [*WebLink*] (In Russian)

[8] V.I. Pokrovsky, Small Medical Encyclopedia: in 6 volumes **3** (1992) 490. [*WebLink*] (In Russian)

[9] Haley J. Finley-Jones et al., Journal of Analytical Atomic Spectrometry **23**(9) (2008) 1214–1222. [*CrossRef*]
[10] Hector Hernandez-Mendoza et al., Quim. Nova **36**(6) (2013) 865–869. [*CrossRef*]
[11] Key-Young Choe, R. Gajek, Analytical methods **8** (2016) 6754–6763. [*CrossRef*]
[12] RMG 76-2014 GSI. State Standard 76–2014. Internal quality control of results of quantitative chemical analysis (Moscow: Standartinform Publ., 2016) 94–95. [*Web Link*] (in Russian)
[13] EVRACHIM/SITAK guide. Quantitative description of uncertainty in analytical measurements, 2nd edition, St. Petersburg: D.I. Mendeleyev VNIIMP (2002) 149. [*Web Link*] (in Russian)