Urine strontium-90 (Sr-90) manual and automated pre-analytical separation followed by liquid scintillation counting

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Abstract

Responding to a radiological or nuclear incident may require assessing tens to hundreds of thousands of people for possible radionuclide contamination. The measurement of radioactive Sr is important because of its impact on people's health. The existing analytical method for urine Sr-90 analysis using crown ethers is laborious and involves possible exposure to concentrated acids; therefore, this work is devoted to the development of the automated Sr-90 separation process, which became possible with the prep*Fast* pre-analytical system (Elemental Scientific, Inc).

Keywords Radiostrontium separation · Liquid scintillation counting · PrepFast system

Introduction

Sr-90 is commercially produced through nuclear fission for medical or industrial use. It is also found in the environment from nuclear testing and in nuclear reactor waste. From the environment, it can be easily transferred to humans through the food chain [1]. Because of its chemical similarity to calcium, Sr tends to concentrate in bones and teeth. Internal exposure to Sr-90 has also been linked to bone cancer and leukemia [2]. As a result, a rapid, accurate, and precise analytical method for urine Sr-90 analysis is necessary to analyze urine samples of people potentially contaminated in a radiological accident or nuclear terrorism incident.

The manual method which uses a vacuum box and Sr resin cartridges was developed by Horwitz [3], improved by

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¹ Inorganic and Radiation Analytical Toxicology Branch, Division of Laboratory Sciences, National Center for Environmental Health, Centers for Disease Control and Prevention, 4770 Buford Hwy, MS S-110-5, Atlanta, GA 30341, USA Maxwell [4] and developed further by our group [5, 6]. It is dependable, but labor intensive and requires constant handson time from the analyst; therefore, automating this method is very appealing. We present here the progression of method development from the Sr-90 manual method using a vacuum box and Sr resin cartridges, to an automated 1-probe/4-Sr resin column prep*Fast* system, to an automated 5-probe/5-DGA column prep*Fast* system (both prep*Fast* systems are from ESI—Elemental Scientific, Inc.). We also discuss the advantages and disadvantages of each method.

Experimental

Reagents and materials

For these experiments, we used double distilled nitric acid (GFS Chemicals) to prepare solutions of various concentrations. For methods involving Sr resin cartridges or columns, oxalic acid dihydrate (Fisher Scientific) was used for preparation of a mixture of 0.05 M oxalic acid with 3 M nitric acid and aluminum nitrate nonahydrate, 99% (ACROS Organics) was used for preparation of the 2 M aluminum nitrate solution by mixing with deionized water.

For the method involving DGA columns, we used Triton X (Alfa Aesar) to prepare a mixture of 0.002% Triton X with 5% nitric acid (v/v), hydrochloric acid (GFS Chemicals) to prepare 0.5 M HCl solution, and ammonium oxalate



monohydrate (Alfa Aesar) to prepare the 0.1 M ammonium bioxalate by mixing with oxalic acid and deionized water.

For the inductively coupled plasma mass spectrometry (ICP-MS) Sr recovery method, 1000 µg/mL strontium standard in 0.1% v/v nitric acid (Inorganic Ventures) was used to prepare Sr-88 calibrators and quality control materials while 1000 µg/mL rhodium standard in 2% nitric acid (Inorganic Ventures) was used for diluent preparation. Ultima Gold AB cocktail (UGAB) (PerkinElmer) was used for liquid scintillation counting (LSC) analysis. \geq 18 M Ω cm deionized (DI) water was used for all solutions (Aqua Solutions).

Base urine was collected through anonymous human donations in accordance with Centers for Disease Control and Prevention Institutional Review Board protocol 3994 and acidified to 1% v/v nitric acid. All radioactive source solutions were traceable to the National Institute for Standards and Technology (NIST) (Gaithersburg, MD, USA). U-GAB_Base_2015 is non-spiked base urine. Urine gross alpha/beta quality control (QC) materials U-GAB_ Low_2015 and U-GAB_High_2015 were bought from Eckert & Ziegler Analytics. These materials are base urine spiked with NIST traceable Am-241 and Sr-90/Y-90. Reference material (GAB-RM) and high calibration range material (GAB-HCR) are Am-241 and Sr-90/Y-90 urine spikes at different levels, prepared in our laboratory with Am-241 and Sr-90/Y-90 NIST traceable reference solutions.

Table 1 shows the original gross alpha/beta urine spikes and QC material characterized by LSC. After purification, these solutions became U-GAB_Base-Sr90, U-GAB_Low-Sr90, U-GAB_High-Sr90, Sr90-RM, and Sr90-HCR respectively. To estimate the limit of detection (LOD), we prepared four samples near the LOD with the total Sr-90 activity range of 0–65 Bq/L in base urine. We purchased Am-241 and Sr-90/Y-90 reference materials, used for quench curves and urine spikes preparation, from Eckert & Ziegler Analytics. Co-60 and Cs-137 standard solutions (Eckert & Ziegler Analytics) were used for urine samples spiking before purification procedure to check the method selectivity.

Instrumentation and labware

Sr resin cartridges (2 mL) with a vacuum box for 24 samples were purchased from Eichrom Technologies while Sr resin columns with the 1-probe/4-column prepFast MC computercontrolled system and DGA (DGA-Normal) columns with the 5-probe/5-column prepFast MC computer-controlled system were purchased from ESI. The ultra-low-level liquid scintillation spectrometer Quantulus1220 and 20-mL plastic LSC vials (all from PerkinElmer) were used for LSC analysis. For Sr-88 ICP-MS analysis (as Sr recovery method), a NexION®300 inductively coupled plasma dynamic reaction cell mass spectrometer (ICP-DRC-MS) (PerkinElmer) was used. Additional supplies included 15-mL and 50-mL conical polypropylene tubes from Beckton Dickinson Labware, a high precision analytical balance capable of accuracy to 0.0001 g (Mettler Toledo), a Brinkman bottle top dispenser with capacity from 5 to 25 mL (Brinkman Instruments) for LSC cocktail dispensing, and a set of four automatic pipettes with total volume range from 5 μ L to 5 mL (Eppendorf).

Sample preparation

The sample preparation procedure for each method/instrument is presented in the next three diagrams. We used stable Sr recovery by ICP-MS [6] for all three methods. For different recovery methods such as stable strontium gravimetric recovery or Sr-85 recovery by gamma spectrometry [7], the sample preparation procedure would be slightly different.

Sample preparation for the manual process with vacuum box and Sr-resin cartridges (Eichrom Technologies)

Following our previously developed procedure [6], we mixed Sr-90/Y-90 and Am-241 urine spikes (5 mL) with concentrated nitric acid (5 mL), 2 M aluminum nitrate (1 mL), and stable Sr standard 1000 μ g/mL (100 μ L) in 15 mL polypropylene tubes. Then we performed the next several steps as described in Fig. 1.

Sample ID	Gross alpha/be	ta specifi	Alpha bias (%)	Beta bias (%)				
	Found alpha	SD	Found beta	SD	Target alpha	Target beta		
U-GAB_Base_2015	0.1	0.8	44.9	6.2	0	N/A	NA	N/A
U-GAB_Low_2015	76.8	8.4	1770	47.0	80.0	1740	-4.0	1.7
U-GAB_High_2015	5270	166	105,100	1880	5350	106,000	-1.5	-0.8
GAB-HCR	15,100	450	153,000	2490	15,000	150,000	0.7	2.0
GAB-RM	4070	160	50,100	733	4000	50,000	1.8	0.2

Table 1 LSC results for gross alpha/beta original urine spikes using Quantulus 1220 analyzed more than 20 times on different days



Fig. 1 Flow chart for urine Sr-90 manual sample preparation using vacuum box and Sr-resin cartridges (2 mL) (Eichrom Technologies)

Sample preparation during the automated process with 1-probe/4-Sr-resin column prep*Fast* MC system (ESI)

The samples were prepared in 15-mL tubes the same way as with the manual method (see above). The tubes were placed on a tray on the autosampler deck, and the next several steps were performed automatically as illustrated in Fig. 2. The 1-probe/4-column syringe-driven system allows sample loading, multiple acid washes, column conditioning, and elution cycles all at user-defined intervals (time, volume, and flow rate). One probe transfers the solution one by one with the intermediate probe wash for initial sample loading and final eluting.



Fig. 2 Flow chart for urine Sr-90 automated sample preparation on 1-probe/4-Sr resin column prep*Fast* system (ESI)

Sample preparation during the automated process with 5-probe/5-DGA column prep*Fast* MC system (ESI)

For the automated method with DGA columns we used a purification procedure for Sr-Ca separation [8] with some changes. Sr-90/Y-90 and Am-241 urine spikes (5 mL) were mixed with 12 M nitric acid (1 mL) and stable Sr standard 1000 μ g/mL (100 μ L) in 15 mL polypropylene tubes, which were placed on the tray on the autosampler deck. The next several steps were performed automatically as shown in Fig. 3. The 5-probe/5-column syringe-driven system allows sample loading, multiple acid washes, column conditioning and elution cycles simultaneously for all 5 columns at user-defined intervals (time, volume, and flow rate).

LSC parameters for radioactivity measurements

For LSC analyses, we used the Quantulus1220 in alpha/ beta mode and our liquid scintillation counting approach [9], which includes pulse shape analysis (PSA) setting optimization and Sr-90/Y-90 quench curve preparation. Count time for the sample was 5 min (min) with 1 min for external standard counting, resulting in a total analysis time of approximately 7.5 min per sample. As shown previously [9], this time provides reasonable minimum detectable activity and good counting statistics.

ICP-MS method parameters for stable Sr recovery

ICP-MS method parameters are described in our 2021 article in the Journal of Radioanalytical and Nuclear Chemistry



Fig. 3 Flow chart for urine Sr-90 automated sample preparation on 5-probes/5-DGA column prep*Fast* system (ESI)

[6]. The diluent in the method is an aqueous solution of 10 μ g/L internal standard (rhodium) in 1% v/v nitric acid and the method rinse solution is an aqueous solution of 1% v/v nitric acid. With each set of samples, the instrument software collects data on 4 external calibrators in the range of 0–200 μ g/L prepared in 0.1% v/v nitric acid and generates a simple linear calibration curve for Sr-88. The Sr-88 ICP-MS method QC materials (60 μ g/L and 150 μ g/L) are analyzed within each analytical run to show that the instrument and method are under control.

Results and discussion

We prepared samples manually in 15 mL tubes for all 3 methods. The rest of the purification procedure was conducted either manually using a vacuum box or automatically using the prep*Fast* computer-controlled systems (ESI). Each stage was optimized with the appropriate solvent and solvent amount. The desired method criteria were stable strontium recovery by ICP-MS and LSC specific activity results within limits for known urine spikes.

Sr recovery results for each sample preparation method

We purified five urine spikes containing Am-241 and Sr-90/Y-90 at different levels (QC, GAB-RM, and GAB-HCR) using the vacuum box and Sr resin cartridges (Eichrom Technologies). The recovery results are presented in Table 2. Average Sr recovery is 91–92%.

To automate this process, we used the 1-probe/4-column prep*Fast* MC system with Sr resin columns (ESI) for purifying urine gross alpha/beta QC materials and blank urine (BU). The procedure was similar the procedure that uses a vacuum box. Table 3 shows the recovery results with average strontium recovery of 76–77%.

Finally, we purified the original five urine spikes (QC, GAB-RM, and GAB-HCR) with the automated 5-probe/5-column prep*Fast* system with DGA columns (ESI). For that

 Table 2
 Stable Sr recovery by ICP-MS for manual method using Sr resin cartridges and vacuum box (Eichrom Technologies)

Sample ID	Sr recovery (%) (±SD)
U-GAB_Base-Sr90	94 (±0.9)
U-GAB_Low-Sr90	88 (±0.9)
U-GAB_High-Sr90	93 (±0.9)
Sr90-HCR	89 (±0.9)
Sr90-RM	93 (±0.9)
Average	91.4

 Table 3
 Stable Sr recovery by ICP-MS for automated method with 1-probe/4-Sr resin column prepFast system (ESI)

Sample ID	Sr resin column #	Sr recovery (%) (± SD)
U-GAB_Base-Sr90	1	74 (±0.7)
U-GAB_Low-Sr90	2	75 (±0.8)
U-GAB_High-Sr90	3	77 (± 0.8)
BU	4	$76(\pm 0.8)$
Average		75.5
BU	1	$80(\pm 0.8)$
BU	2	$76(\pm 0.8)$
BU	3	$74(\pm 0.7)$
BU	4	$76(\pm 0.8)$
Average		76.5

we changed Sr-Ca separation procedure [8]. The first step was changed to double wash with 0.1 M ammonium bioxalate to remove some alpha nuclides from the columns. The second step was changed to 0.5 M hydrochloric acid wash. This step and 2 M nitric acid wash allow separation of Sr-90 from such nuclides as Y-90, K-40, Co-60, and Cs-137. To increase strontium recovery, 6 mL of 0.2 M nitric acid was used for the final elution. Table 4 shows the results with the average strontium recovery of 93–94%.

Sr-90 carry-over observation

As indicated, recovery in the 1-probe/4-Sr resin column automated method is the lowest (76–77%), while recoveries for the Sr resin cartridges manual method and 5-probe/5-DGA columns automated method are comparable and higher

 Table 4
 Stable Sr recovery by ICP-MS for the automated method with 5-probe/5-DGA column prepFast system (ESI)

Sample ID	DGA column #	Sr recovery (%) (±SD)
U-GAB_Base-Sr90	1	92 (±0.9)
U-GAB_Low-Sr90	2	89 (±0.9)
U-GAB_High-Sr90	3	94 (±0.9)
Sr90-HCR	4	96 (±1.0)
Sr90-RM	5	93 (±0.9)
Average		92.8
BU	1	92 (±0.9)
BU	2	$93(\pm 0.9)$
BU	3	93 (±0.9)
BU	4	98 (±1.0)
BU	5	96 (±1.0)
Average		94.4

than 90%. The lower recovery means that Sr-90 was partially lost. If Sr-90 stays on the columns, it can cause carry-over.

The manual method uses one cartridge for each sample; therefore, no carry-over occurs from sample to sample. However, the automated methods use the same columns for purification many times, so carry-over is a potential problem. To check for carry-over, we purified non-spiked blank urine (BU) samples after gross alpha/beta urine spikes. LSC analysis of these blank samples shows that carry-over takes place with both automated systems. The carry-over for Sr resin columns is about 3-5% of the original activity, while for DGA columns, the carry-over is about 100 times less (0.05%). These results demonstrate that only samples with high specific activity (50,000 Bq/L or more) will create a carry-over problem for DGA columns. To prevent carry-over from such samples, 2 M nitric acid solution will be injected after them into the columns. We showed that normally one injection will clean the columns.

Sr-90 activity calculation and urine stable Sr effect

Sr-90 activity for each purification method was calculated by LSC as gross beta in alpha/beta mode. This activity was

Sample ID

Column #

corrected for Sr recovery and Y-90 in-growth, calculated through the Bateman equation [10], as described earlier [6]. The start time for Y-90 in-growth should be recorded in each method. Y-90 in-growth will be calculated as the percentage from Sr-90 activity. Knowing the Sr recovery and Y-90/Sr-90 ratio, we can find current Sr-90 activity.

Normal stable Sr content in urine is in the range of $27-178 \ \mu g/L$ [11]. As we discussed [6], the analytical error introduced from naturally occurring stable Sr in urine is no more than 1% when 200-fold dilution was applied prior ICP-MS analysis.

Table 5 presents the final LSC results. The LSC gross alpha/beta analysis shows that the original QC, GAB-HCR, and GAB-RM samples contained gross alpha/beta emitters (Table 1 and Fig. 4). LSC spectra of gross alpha/ beta QC after purification shows a large Sr-90 peak with a small Y-90 peak that increased with time (Fig. 5) and does not show a signal for Am-241. Column # shows which column is used to purify blank urine samples after urine spikes to see carry-over. The worst correlation between found and target data (up to -10%) was observed for the automated method with Sr resin columns because of Sr loss issues.

Beta bias (%)

Sr-90 method/instrument

		(Bq/L)				
		Found	SD	Target		
U-GAB_Base-Sr90		8.3	9.3	0	<lod< td=""><td>Manual</td></lod<>	Manual
U-GAB_Low-Sr90		837	23.4	870	-3.8	Manual
U-GAB_High-Sr90		52,000	1300	53,000	-1.9	Manual
Sr90-HCR		73,600	1700	75,000	- 1.9	Manual
Sr-90 RM		24,850	746	25,000	-0.6	Manual
U-GAB_Base-Sr90	#1 Sr	5.4	3.7	0	<lod< td=""><td>Automated 1/4</td></lod<>	Automated 1/4
U-GAB_Low-Sr90	#2 Sr	782	29	870	- 10.1	Automated 1/4
U-GAB_High-Sr90	#3 Sr	49,000	2080	53 000	-7.5	Automated 1/4
BU	#4 Sr	8	1.7	0	<lod< td=""><td>Automated 1/4</td></lod<>	Automated 1/4
BU after Base-Sr90	#1 Sr	7	1.6	0	<lod< td=""><td>Automated 1/4</td></lod<>	Automated 1/4
BU after Low-Sr90	#2 Sr	39	4.8	0	2–3 LOD	Automated 1/4
BU after High-Sr90	#3 Sr	1388	31.1	0	80–90 LOD	Automated 1/4
BU after BU	#4 Sr	19	3.1	0	<lod< td=""><td>Automated 1/4</td></lod<>	Automated 1/4
U-GAB_Base-Sr90	#1 DGA	2.4	4.2	0	<lod< td=""><td>Automated 5/5</td></lod<>	Automated 5/5
U-GAB_Low-Sr90	#2 DGA	860	25.0	870	-1.1	Automated 5/5
U-GAB_High-Sr90	#3 DGA	52,200	1240	53,000	-1.5	Automated 5/5
Sr90-HCR	#4 DGA	75,300	1160	75,000	0.4	Automated 5/5
Sr-90 RM	#5 DGA	24,600	976	25,000	-1.6	Automated 5/5
BU after Base-Sr90	#1 DGA	2	0.4	0	<lod< td=""><td>Automated 5/5</td></lod<>	Automated 5/5
BU after Low-Sr90	#2 DGA	7	1.5	0	<lod< td=""><td>Automated 5/5</td></lod<>	Automated 5/5
BU after High-Sr90	#3 DGA	39	4.8	0	2–3 LOD	Automated 5/5
BU after HCR-Sr90	#4 DGA	44	5.1	0	2–3 LOD	Automated 5/5
BU after RM-Sr90	#5 DGA	3	0.7	0	<lod< td=""><td>Automated 5/5</td></lod<>	Automated 5/5

Sr-90 specific activity

Table 5LSC results for grossalpha/beta urine spikes afterpurification accounted for Srrecovery and Y-90 in-growthreceived on Quantulus1220



Fig.4 Spectrum of high gross alpha (Am-241)/beta (Sr-90/Y-90) urine QC before Sr-90 purification from Quantulus1220



Fig. 5 Spectra of high gross alpha (Am-241)/beta (Sr-90/Y-90) urine QC after Sr-90 purification in one hour and in 24 h from Quantulus1220

Limit of detection

The limit of detection was statistically determined for the manual and 5-probe/5-DGA column methods in accordance

with established protocols for determining the limit of detection and limit of quantification [12]. This LOD estimation is based on the results of four Sr-90 LOD urine samples analyzed at least 20 times each. Table 6 shows the LOD results as well as comparison results of these three systems. The LODs for the manual method and the automated method with 5-probe/5-DGA column are comparable, although the LOD is slightly lower for the automated method. We did not estimate the LOD for the 1-probe/4-Sr resin column system because due to carry-over, this method with Sr resin columns is not practical for Sr-90 analysis.

Advantages and disadvantages of each systems

Table 6 also lists the advantages and disadvantages of each system. The Sr recovery for both the manual and the automated 5-probe/5-DGA column methods is high (88-98%) and comparable as is the correlation between found and target Sr-90 activities, which is in the range of $\pm 5\%$. The manual method creates less waste than the automated one, but the automated system is less laborious and can be left unattended during the sample preparation step. The disadvantage of the 5-probe/5-column system is the additional step: transferring the eluted solution from 15-mL tubes into LSC vials for LSC analysis. This additional step occurs because of the 5-probe system design: the same trays and, as a result, the same tubes are used for original and purified samples. For 1-probe/4-column system, the different trays can be used for the original and eluted samples, which allows the purified samples to be dispensed directly into LSC vials. However, the 5-probe/5-column system allows the loading of samples to the columns and dispensing the final solutions simultaneously decreasing the preparation time per sample in comparison with the 1-probe/4-column system. The main disadvantage of the 1-probe/4-Sr resin column system is the lowest Sr recovery which can be probably explained by the effect of aging Sr resin. As we showed earlier [6], the older the age the lower the recovery even for ICP-MS recovery method. Sr resin became less effective from run to run under the chosen conditions and, as a result. Sr resin columns can be reused about 20 times while Sr resin cartridges about 5 times with lower Sr recovery each time. The price per sample, based on column/cartridge use, is less for the 5-probe/5-column system.

Conclusion

This work described the development of the automated sample prep system for Sr-90 analysis from a vacuum box through a 1-probe/4-Sr resin column prep*Fast* system to a 5-probe/5-DGA column prep*Fast* system. The 5-probe/5-DGA column system with four trays (60 tubes per tray) can

Table 6	Comparison	of three systems	used for urine	Sr-90 sample preparation
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Parameters	Vacuum box	1-probe-4-column	5-probe/5-column DGA columns	
	Sr resin cartridges	Sr resin columns		
Method LOD (Bq/L)	20	N/A	15.9	
Time for 24 samples (h)	2.5	6	3.3	
Sr recoveries (%)	88–97	74–80	89–98	
Samples per day	48	60	120	
Number of people	1	1	1	
Time for labor (h)	5	1	1	
Number of units	1	1	1	
Samples/column, cartridge	1	20	60	
Price per column, cartridge (\$)	1330/box of 50	490/column	490/column	
Price per sample (\$)	26.6	24.5	8.17	
Price per system (\$)	3,000	60,000	80,000	
Sr recoveries	High, but depends on cartridges age [6]	Changing with column use	High and constant	
Acidic waste per 20 samples (L)	0.5	About 1.0	About 2.0	
Additional step for transferring the final solution	No	No	Yes	
Carry-over	No	Yes, about 3–5%	Yes, about 0.05%	
Analyst requirement	Should be present at all times	Present only for initial sample mixing and taking samples at the end for ICP-MS	Present only for initial sample mixing, taking samples at the end for ICP-MS, and transfer- ring from the tubes into LSC vials	
Found Sr-90 activities versus target (%)	±5	- 10	±5	

accommodate 120 tubes with original solutions as well as 120 tubes with purified solutions with the current method. 5 samples can be purified in about 40 min; therefore, purifying 120 samples will take approximately 16 h. In addition, DGA columns can be reused about 60 times without a change in strontium recovery and column performance.

Although each method has its own advantages and disadvantages, the main advantage of 5-probe/5-DGA-column system is that the Sr-90 separation can be done automatically. This is important when analysis is time sensitive and a large number of samples needs to be analyzed non-stop. Exchanging Sr resin columns for DGA columns in 1-probe/4-column system allows the use of this system for Sr-90 sample separation as well.

ICP-MS works accurately and precisely for stable Sr-88 analysis as a recovery method. Gross alpha/beta LSC analysis confirmed that after the given urine spikes purification (and not counting crosstalk from high Sr-90 activity samples), only Sr-90 is in the final solution when the original solutions contained Am-241, Co-60, and Cs-137.

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Declarations

Conflict of interest The authors declare that they have no competing financial interest.

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