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Bead Injection Extraction Chromatography Using High-Capacity Lab-on-Valve as a Front End to Inductively Coupled Plasma Mass Spectrometry for Urine Radiobioassay

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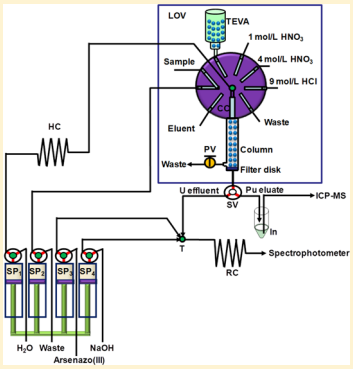
ABSTRACT: A novel bead injection (BI) extraction chromatographic microflow system exploiting a high-capacity lab-on-valve (LOV) platform coupled with inductively coupled plasma mass spectrometric detection is developed for rapid and automated determination of plutonium in human urine. A column attached to the LOV processing unit is loaded online with a metered amount of disposable extraction chromatographic resin (up to 330 mg of TEVA (abbreviation for tetravalent actinides)) through programmable beads transport. Selective capture and purification of plutonium onto the resin beads is then performed by pressure driven flow after preliminary sample pretreatment. The analytical results demonstrate the large capacity of bead surfaces for uptake of Pu within the tailor-made LOV platform that fosters processing of large-sized biological samples, e.g., 1 L of human urine, along with good reproducibility for automatic column renewal (0.319 ± 0.004 g, n = 5). The chemical yields of plutonium were averagely better than 90% under the optimal experimental conditions, and the entire analytical procedure could be accomplished within a short time frame (<3 h) as compared to manual counterparts (1−2 days). Therefore, the developed system is well suited for expedient analysis of low-level plutonium in urine of exposed individuals as required in emergency situations.



The unexpected nuclear power plant accidents, such as those at Chernobyl and Fukushima, and emergency events associated with radiological/nuclear terrorist threats have raised the request of quick assessment of internal radioactive contamination in a potentially exposed population. Plutonium is regarded as a highly radiological and biological toxic element as a result of the harmful α particle emission of its most predominant isotopes (^{238}Pu: 87.7 year; ^{239}Pu: 24 110 year; ^{240}Pu: 6561 year),[1] its readily deposition in the liver and/or bones if introduced into human body, and lengthy retention in the body within the order of decades. Urine bioassay has been regarded as a reliable approach to estimate the radiation dose of internal exposure for people occupationally or incidentally exposed to radioactive contamination. Over the past few decades, a wealth of analytical methods have been developed and applied to determine plutonium in human urine.[2−9]

Due to long-term retention of Pu in the human body, the excretion rate of Pu to urine is very slow. As a consequence, it is required to measure very low levels of Pu in urine to be able to meet the screen criteria of minimum internal dose of occupational and public radiation exposure. The typical concentrations of plutonium in urine are extremely low, at the micro-Becquerel level per liter for nonexposed population.[10] Large sample volumes (e.g., ≥1 L) are normally required to cope with the sensitivity demands of either radiometric or mass spectrometric techniques, especially in the case of occupational health monitoring.

In the majority of plutonium urinalysis methods reported in the literature, the overall experimental operations were normally performed in a manual fashion. The resulting procedures are therefore time-consuming and labor intensive with lengthy and poorly repeatable chromatographic separations due to the low (<0.5 mL/min) and variable flow rate caused by the large density and viscosity of the loading solution. This is originated from the matrix components of human urine (e.g., calcium, phosphate, sulfate, organic substances) and the extra addition of elements to facilitate the initial coprecipitation of actinides (e.g., manganese, iron, calcium, phosphate) and/or complexing reagents (i.e., aluminum nitrate) to mask potential interferences from phosphates and sulfates.

To overcome the above shortcomings, an analytical method has been reported lately using vacuum box technologies to facilitate a fast multimodal separation on stacked columns of tetravalent actinides (TEVA), uranium and tetravalent actinides (UTEVA), and diglycolamide (DGA) for determination of ^{237}Np and plutonium isotopes in small-sized (10 and 100 mL) urine samples.[11] Although the reported column separation system is simple and cost-effective, it is semiautomated and still

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needs manual operations, such as stacking/splitting columns, dispensing sample/reagents, and collecting eluates.

There has been recently an increasing interest in the usage of flow-based automated approaches for plutonium assays in urine samples. Most of these automated methods resorted to extraction chromatographic (EC) procedures capitalized on Eichrom (or TRISKEM) TEVA or transuranic (TRU) resins.[12−14] Although fast and straightforward analysis with minimum labor intensity are to be achieved via automation, several important drawbacks are still not well overcome, which, in turn, would limit the widespread application of flow-based approaches for radioassays: (1) the majority of works were evaluated for small urine volumes (1−100 mL) that might not suffice for detection of low abundance Pu;[12−14] (2) though a 1 L urine sample was also directly loaded onto the TEVA column for chemical separation,[12] this method was tedious and prone to render biased results because of clogging of the chromatographic column; (3) for TRU resin, the separation between Pu and U was not quantitative, resulting in the coelution of U in the course of Pu retrieval;[13] (4) flow injection (FI) was the methodology of choice among most of the previously published works for urine radiobioassays, but complex manifolds have been assembled consisting of several injection valves and peristaltic pumps.[14] In addition, FI consumes large amounts of sample/reagents as a result of its continuous-flow nature. In contrast, the second and third generations of flow analysis, which are so-called sequential injection (SI) and lab-on-valve (LOV), respectively, allow for more accurate handling of solutions via bidirectional and discontinuous programmable flow.[15,16]

With the inherent versatility to implement unit operations at will, without the need of manifold reconfiguration, SI has been consolidated as a powerful approach in the radio-analytical chemistry field.[1] The concept of renewable surfaces, the so-called bead injection (BI), in SI and LOV microfluidic platforms or related configurations for online disposable solid-phase extraction beads, has also gradually been spread out to overcome the performance deterioration of permanently packed sorbent columns in flow systems and applied in several analytical fields.[17−21] The state-of-the-art of BI has been presented in a number of review articles,[15,22−26] where we can observe that BI-based platforms developed up to date and exemplified in practical applications for radio-assays are rather uncommon. Notwithstanding the fact that Egorov et al.[27] determined ^{90}Sr, ^{241}Am, and ^{99}Tc recoveries by exploiting a BI renewable separation column in SI with a nominal volume of 250 μL, this work was dedicated to analyze small-sized nuclear wastes (e.g., 0.3 g of vitrified glass samples), and the final sample solution injected into the SI−BI system was at the microliter level (namely, 100 μL). Avivar et al.[28−30] used miniaturized renewable columns internally assembled in the LOV module for the determination of uranium and thorium in water, but again, a mere 30 mg of sorbent material (i.e., UTEVA) was handled in the flow setup.

To the best of our knowledge, no BI-based renewable system has been reported so far for determination of anthropogenic transuranium elements (e.g., Pu) in biological samples. This might be due to the fact that down-scaled BI networks proposed to date are not readily applicable to biological assays of trace levels of radionuclides for which compatibility of processing large-sized samples is most often required.

Hereto, BI extraction chromatography exploiting high-capacity LOV devices is proposed in this work for rapid and automated separation of plutonium from human urine, whereafter inductively coupled plasma mass spectrometry (ICPMS) is harnessed for expedient quantification of plutonium isotopes. The analytical performance of the developed LOV-BI manifold including column renewability, separation efficiency, and sample throughput is investigated and evaluated in detail.

EXPERIMENTAL SECTION

Flow Manifold. The flow system is composed of a multisyringe pump (BU4S; Crison Instruments, Barcelona, Spain) with programmable flow rate and a polymethylmethacrylate LOV conduit encompassing 7 integrated microchannels of 1.2 mm i.d./14.0 mm length and a large-sized channel of 4.0 mm i.d./14.0 mm length (to prevent valve clogging) to which a methacrylate column (5 mm i.d./50 mm length) is nested for containment of beads (see Figure 1). A 25 μm-pore size glass-

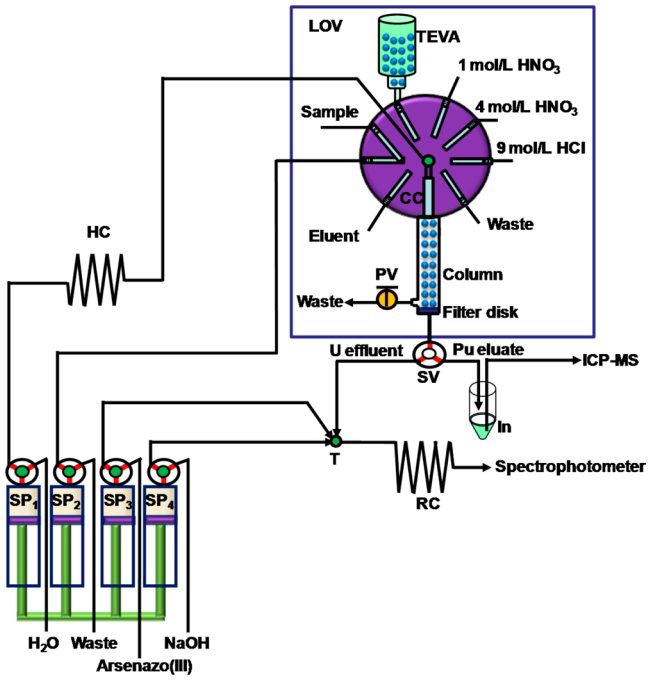

Figure 1. Lab-on-valve bead injection system for rapid extraction chromatographic separation of plutonium with at-line ICPMS measurement and in-line detection of uranium removal (CC: central channel; HC: holding coil; PV: pinch valve; RC: reaction coil; SV: solenoid valve; T: confluence point).

fiber filter disc was placed between the column and the bottom cap to allow for all solutions to flow freely while trapping the beads effectively. A bead suspension of 1: 3 (w/v) was prepared in ultrapure water (18 MΩ·cm) and contained in a 10.0 mL polyethylene syringe which was mounted vertically onto one port of the LOV conduit. The LOV sample processing unit working as stator is mounted atop an eight-port multiposition selection valve (MSV, Valco Instruments, Houston, TX). The central port of the LOV is made to address the peripheral ports of the unit (1−8) for sequential aspiration of the various constituents for the BI-based solid-phase extraction procedure, via the central communication channel (CC) in the MSV.

The syringe pump is equipped with two 10 mL and two 5 mL gastight glass syringes (Hamilton, Switzerland) operating as liquid drivers. Each syringe has a three-way solenoid valve (N-

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Research, Caldwell, NJ, USA) at the head, which facilitates the application of multicommutation schemes (on: in-line flow; off: to reservoirs). There is also one external three-way solenoid valve (SV, N-Research) connected to the bottom end of the external column to divert at will (on: to eluate collector; off: to detect U) and one external pinch valve (PV, PK-0305-NC, Takasago Electric, Inc., Nagoya, Japan) connected to the side port of the column assisting in the replenishment of the column and withdrawal of beads (on: beads to waste; off: trap the beads inside the column).

The flow network is constructed with polytetrafluoroethylene (PTFE) tubing, including a 5.1 m-holding coil (HC) of 1.5 mm i.d. with an inner capacity of 9 mL for temporary storage of different solutions prior to delivery into the column via flow reversal, and a 3 m-knotted reaction coil (RC) of 0.8 mm i.d. for mixing the color developing reagent with column effluents for in-line spectrophotometric detection of uranium in decontamination studies. Instrumental control and acquisition of spectrophotometric data are performed using the software package AutoAnalysis 5.0 (Sciware, Palma de Mallorca, Spain). Solenoid and pinch valves are connected to the syringe pump through an interface so as to feed them with 12 V.

Reagents and Samples. Nitric acid (65%), hydrochloric acid (37%), ammonia (25%), hydrogen peroxide (30%), arsenazo-III, chloroacetic acid, sodium chloroacetate, ferric chloride, potassium disulfite, sodium hydroxide, and potassium permanganate were analytical grade reagents, and all solutions were prepared with ultrapure water. Solutions of ^{242}Pu (0.1037 Bq/g in 2 mol/L HNO_3) were diluted from NBL-CRM 130 which was purchased from New Brunswick Laboratory (Argonne, IL). A ^{239}Pu standard solution of 0.100 Bq/g in 2 mol/L HNO_3 was supplied by the Center for Nuclear Technologies at the Technical University of Denmark. A standard solution of uranium of 1000 mg/L in 1 wt % HNO_3 was purchased from Sigma-Aldrich (St. Louis, MO). TEVA extraction chromatographic resin (100−150 μm particle size) was obtained from TRISKEM International (Bruz, France).

Pooled human urine samples were collected from 15 Danish healthy residents and preserved in clean and sealed polyethylene barrels under 5 °C. One liter of urine spiked with a known amount (5−50 mBq) of ^{239}Pu was used as a sample for method development.

Sample Pretreatment. To 1 L of human urine, 10 mL of concentrated HCl was added to acidify the sample to pH = 1 and then 5 mBq of ^{242}Pu was added. The pH of the sample was adjusted to 7 using concentrated $NH_3{\cdot}H_2O$; 5 mL of 0.2 mol/L $KMnO_4$ and 5 mL of 0.3 mol/L $MnCl_2$ were added with stirring to form MnO_2 precipitate, and 1−2 mL of concentrated $NH_3{\cdot}H_2O$ was then added to adjust the pH to 9 to stabilize the MnO_2 precipitate. The sample was stirred for 10 min and then transferred into four 250 mL centrifuge tubes and centrifuged at 4000 rpm for 10 min.

The supernatant was discarded, and the precipitate was transferred into a beaker with 20 mL of concentrated nitric acid plus a few drops of hydrogen peroxide. After adding 300 mg of $FeSO_4$, the sample was digested and evaporated to dryness on a hot plate at 200 °C with the occasional addition of hydrogen peroxide in a total volume of 5 mL to decompose the organic matter. The residue was dissolved with 4 mL of *aqua regia*, heated to almost dryness at 200 °C, and then diluted to 200 mL with deionized water.

$K_2S_2O_5$ (300 mg) was added, and the sample was stirred for 20 min to reduce overall plutonium to Pu(III). NaOH (6 mol/L) was added to adjust the pH to 9−10, and the sample was centrifuged at 4000 rpm for 10 min. After discarding the supernatant, 8.5 mL of concentrated HNO_3 was added to dissolve the precipitate, meanwhile oxidizing Pu(III) to Pu(IV). Then, 1 mL of 2 mol/L $Al(NO_3)_3$ was added, and the sample solution was finally diluted to ca. 4 mol/L HNO_3 (sample volume was 20−30 mL) for column separation using LOV-BI.

Automated Column Separation Exploiting LOV-BI. The automated EC column separation in the LOV-BI format was composed of the following steps: (1) automatic packing of the BI column with ca. 300 mg of TEVA resin, as detailed in Table 1; (2) preconditioning the TEVA column with 20 mL of 4 mol/L HNO_3; (3) loading the sample solution (20−30 mL) onto the column; (4) rinsing the column with 20 mL of 1 mol/L HNO_3 followed by 10 mL of 9 mol/L HCl; (5) eluting plutonium with 20 mL of 0.025 mol/L HCl; (6) automatic removal of TEVA beads (see Table 1) and cleaning the HC and the inlet tubing for sample loading with 10 mL of water. The flow rates for sample loading, column washing, and plutonium elution were fixed to 1.0 mL/min. Further details of the analytical sequences for BI analysis are given in Table 1.

Table 1. Optimized Procedure for Automatic On-Line Packing and Disposal of the Extraction Chromatographic TEVA Column in LOV

column packing	
step number	operation
1	fill the holding coil with deionized water
2	dispense 1 mL of water into the beads container at 10 mL/min to resuspend TEVA resin (soaked previously in water)
3	aspirate 200 μL of TEVA beads at 0.6 mL/min into holding coil
4	dispense 200 μL of TEVA beads at 5 mL/min into the column
5	dispense 300 μL of deionized water (by means of 3 consecutive pulses, 100 μL each) into the column at 10 mL/min
6	repeat 12 times, steps 2−5
column emptying	
step number	operation
1	fill the holding coil with 9 mL of deionized water and then keep the pinch valve open
2	dispense 3 mL of deionized water (by means of 3 consecutive flushes, 1 mL of each) into column at 7.5 mL/min for bead disposal
3	back-flush the column by aspirating 1 mL of the mixture of water and remaining beads at 5 mL/min
4	dispense 1 mL of deionized water/beads into waste at 10 mL/min
5	repeat three times, steps 3−4
6	dispense 3 mL of deionized water into the column at 10 mL/min

Detection of Plutonium with ICPMS. The detection of plutonium with ICPMS (X Series^{II}, Thermo Fisher Scientific, Waltham, MA) was performed after collecting the Pu eluate and the addition of internal standard of indium to a final concentration of 1 μg/L. The ICPMS instrument was equipped with an Xs-skimmer cone and a Burgener nebulizer under hot plasma conditions. The detection limits calculated as three times the standard deviation of the processing blank ranged from 1.0 to 1.5 pg/L for both ^{239}Pu and ^{242}Pu. A least-squares regression line over the 0.01−100 ng/L range was used for quantification of plutonium in the specific aliquots introduced into the ICPMS. Experimental results confirmed that the mass fractionation was insignificant, and the sensitivities (cps per μg/L) of ^{239}Pu and ^{242}Pu were identical within 1% uncertainty.

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Table 2. Investigation of Experimental Operations for on-Line Column Packing with TEVA Beads

operations	investigated variables	optimum condition
(1) beads resuspension	the use of a burst of air or water to resuspend the beads	water
	flow rate of dispensing water, 5−20 mL/min	10 mL/min
	volume of water, 0.5−2 mL	1 mL
(2) aspirate beads into holding coil	the use of air or water plug in holding coil before aspiration	water
	flow rate of aspirating beads, 0.57−1.5 mL/min	0.6 mL/min
	volume of beads aspiration, 0.1−0.5 mL	0.2 mL
(3) transfer of beads into column	flow rate for dispensing of beads, 0.6−30 mL/min	5 mL/min
	the use of water surplus for completion of beads transfer into column bed, 0.1−1 mL	0.1 mL (three times)
	flow rate for pumping water surplus, 2−30 mL/min	10 mL/min
(4) repeat operations 2−3	number of cycles for aspiration and trapping of beads	12

Practically, we harnessed ^{242}Pu as a standard for the quantification of both ^{239}Pu and ^{242}Pu (as tracer) in urine throughout. Prior to detection, the ICPMS instrument was tuned to maximum transmission of uranium using 1 μg/L ^{238}U solution, and the instrumental parameters were further adjusted for plutonium using a 5 ng/L ^{242}Pu solution for optimal detection efficiency. The typical operational conditions of the instrument have been given in our previous work.[31] It is important to note that these parameters were optimized each time when the instrument was initialized. Typical sensitivities of plutonium ranged from 1×10^5 to 5×10^5 cps per μg/L.

In-Line Spectrophotometric Detection of Uranium. The in-line post-BI detection system for investigation of U decontamination is composed of a deuterium-halogen light source (Mikropack, Germany), two optical fibers of 400 and 600 μm ID (Ocean optics, USA), a 10 mm long Teflon AF liquid core waveguide capillary flow-cell (World Precision Instruments, FL, USA), and a USB 2000 miniaturized fiber-optic spectrometer (Ocean Optics), connected to a PC via the USB interface. The spectrophotometric detection of uranium was carried out on the basis of the derivatization reaction of uranyl ions with the chromogenic reagent Arsenazo (III) forming a stable 1:1 blue complex at pH 2. Whenever the acidic uranium effluent was driven out of the TEVA column at 0.6−2.0 mL/min, 0.01% (w/v) Arsenazo (III) in chloroacetic acid/sodium hydroxide buffer (pH = 2) and NaOH (ranging within 0.38−1.99 mol/L depending on the acidity of uranium effluent) in syringes of SP3 and SP4, respectively, were simultaneously dispensed at a rate of half as much as that of the uranium effluent, toward the confluence point T. The composite zone was driven by 5 mL of carrier through the RC to allow uranium to react at pH = 2 for ca. 2 min. The U(VI)−arsenazo (III) complex was detected downstream by flow-through spectrophotometry at 653 nm (analytical wavelength) using 760 nm as a reference wavelength. The RC was finally washed with 10 mL of water before starting a new analysis cycle.

Caution. Safety concerns in handling radioactive plutonium should be addressed hereby. In this work, a semi-hot laboratory was specifically used for storage of the radioactive stock solutions (namely, ^{239}Pu and ^{242}Pu) and care was paid whenever performing analytical procedures for Pu determination.

RESULTS AND DISCUSSION

Automated LOV Bed Column Disposal. In preliminary investigations, we observed that the analytical performance of the LOV-based EC method was prone to deteriorate significantly when reusing the online packed TEVA beads, as indicated by low chemical yields (<60%), notable cross-contamination (>10%), and increase of back pressure. Especially when processing large-sized (e.g., 1 L) urine samples, the organic matter contained in the matrix might not be completely decomposed and thus remains strongly bound to the functional groups of TEVA resin which makes column reusability impracticable. Besides, unneglectable remnants of Mn oxides originated from the coprecipitation step were also difficult to be completely removed from TEVA entities. Therefore, it is necessary to dispose the spent beads and replenish the extraction chromatographic column with fresh sorbent material in each single assay for reliable and satisfactory results.

Critical variables in the automated packing of the BI column were investigated in detail. These include the use of a burst of air or water to resuspend the resin beads in the external container, the use of air or water plugs prior to aspiration of the beads, the use of short cycles of aspiration and pumping to prevent beads settlement, and the use of a water volume surplus for transportation of beads into the column. The analytical operational steps are summarized in Table 2. Optimal conditions for online packing of the TEVA resin bed encompass resuspending the beads in the container with a fast water flush, whereupon short cycles of aspiration and dispensing are to be programmed to prevent clogging of the central channel of the LOV-BI platform (see Table 1). In each individual short cycle, reliable bead packing is accomplished with slowly aspirating a small amount (200 μL) of sorbent suspension into HC followed by a quick pulse of carrier to dispense the sorbent toward the column. It is proven that a metered volume of water (300 μL) is needed to bring all of the aspirated beads quantitatively into the column. On the basis of the optimal experimental conditions, online column packing repeatability was better than 1.3% with beads amounting to 319 ± 4 mg (n = 5). Compared with previous BI-related works for radionuclides,[28,30] this is the first LOV-BI renewal system able to handle >100 mg of sorptive phase.

In a typical LOV-BI sequence, after each analytical run, the sorbent particles are disposed by flow reversal. However, due to the compact settlement of the large amount of sorbent packed in the column and the asymmetrical shape of the bead-transporting path in LOV, online disposal of large-sized columns is proven troublesome. Therefore, in our design, an external pinch valve was added to the manifold so as to open a smooth way for discharging the beads from the bottom end of the column with several quick and short forward-flow pulses. In combination with back-flushes to clean up the beads remaining at dead column volumes (see Table 1), these series of operations fulfill the demands of handling the bead material reproducibly in the BI format within the microfluidic platform.

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Removal of Interferences. Accurate mass spectrometric determination of plutonium requires a thorough removal of isobaric and polyatomic interferences, namely, $^{238}U^{1}H$, and nonspectral interferences (e.g., thorium), which cause a noisy background in the ICPMS spectrum, especially when dealing with determination of low-abundance plutonium. The concentration of dietary uranium in human urine varies greatly depending on the population's geographic location and diet. According to Shi et al.,[32] a wide range of uranium concentrations (3−310 pg/mL) have been reported as a normal background level of uranium concentration in human urine. To evaluate the efficiency of the proposed protocol for the removal of uranium, artificial solutions containing a known amount of uranium in nitric acid medium were subjected to EC in the LOV-BI mode, and the elution curves were recorded online with spectrophotometric detection. The minute operational maintenance and superior simplicity of online spectrophotometric detection make it more appealing than ICPMS for qualitative measurement of uranium as low detection limits are not necessarily required in this case due to the high content of uranium in the column rinsing effluents. The effects of the concentration, volume, and flow rate of washing solutions on uranium decontamination can be straightforwardly monitored through the variation of the elution curve profiles.

Effect of Flow Rate. The greater the flow rate of the rinsing solution, the better is the sample throughput but at the expense of potentially jeopardizing the analytical performance. The effect of flow rates on the removal of uranium was investigated within the range of 0.6−2.0 mL/min. As seen in Figure 2, the

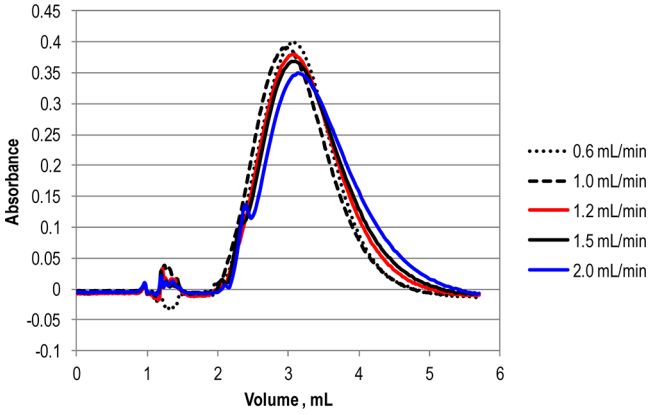

Figure 2. Effect of the flow rate of rinsing solution on the removal of uranium (sample solution: 1 mL of 6 μg/mL uranium in 1 mol/L HNO_3; separation sequence: after loading the sample solution at 1.0 mL/min, 5 mL of 1 mol/L HNO_3 was passed through the column to strip out uranium at different flow rates as indicated in the figure).

efficient stripping out of uranium is proven to be deteriorated at high flow rates (e.g., 2 mL/min), but the separation process is sped up, whereas lower flow rates (e.g., 0.6 mL/min) make the column separation tedious, although better uranium removal can be achieved as observed by the increase in absorbance peak area in Figure 2. Practically, the flow back pressure increased exponentially with the increase of flow rate; therefore, a compromise flow rate of 1.0 mL/min was chosen to reduce the risks of leaking in the LOV-BI system.

Effect of the Concentration of Nitric Acid. Figure 3 shows the effect of the concentration of nitric acid on the removal of uranium from TEVA. It can be seen that the elution curves

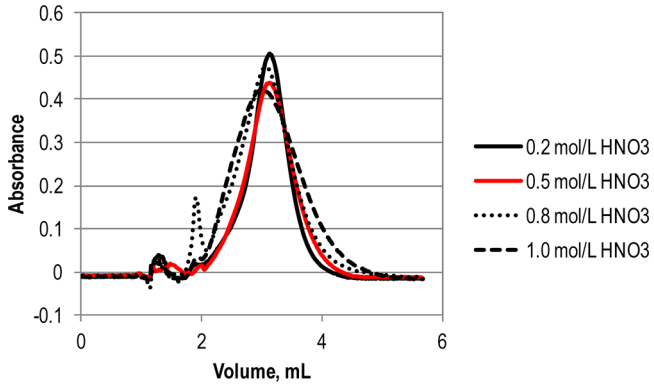

Figure 3. Effect of the concentration of nitric acid on the removal of uranium (sample solution: 1 mL of 6 μg/mL uranium in 1 mol/L HNO_3; separation sequence: after sample loading, 5 mL of different concentrations of HNO_3 was passed through the column to strip out uranium; flow rate of the whole sequence is 1.0 mL/min).

become sharper when decreasing the concentration of nitric acid, which brings a faster flushing of uranium from the column. However, in <1 mol/L HNO_3, the risk of plutonium preelution might be increased due to the lower distribution coefficient of plutonium onto TEVA at decreasing concentrations of nitric acid. Besides, the absorbance peak areas of uranium shown in Figure 3 differ slightly with the variation of nitric acid concentration in the range of 0.2−1.0 mol/L, revealing that most of the uranium can be removed from the TEVA column under the overall investigated conditions. Therefore, 1 mol/L HNO_3 was selected for uranium removal in the remainder of the work according to our previous work.[31]

Uranium Removal Pattern from TEVA in 1 mol/L HNO_3. Figure 4 illustrates the removal behavior of uranium from

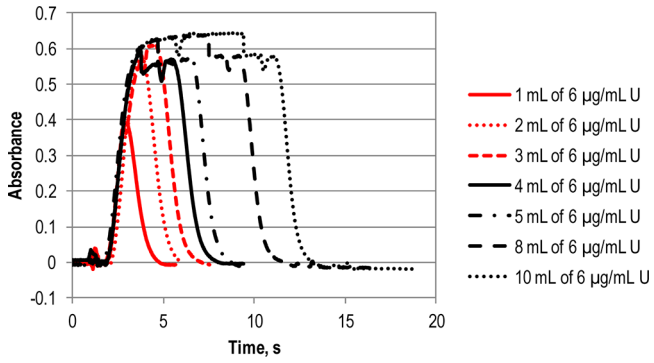

Figure 4. Effect of the amount of uranium on the removal efficiency (sample solutions: 6 μg/mL of uranium in 1 mol/L HNO_3 with different volumes loaded onto TEVA column as illustrated in the figure; separation sequence: after sample loading, 5 mL of 1 mol/L HNO_3 solution was passed through the column to strip out uranium; flow rate of the whole sequence is 1.0 mL/min).

TEVA column in 1 mol/L HNO_3 under dynamic flow-through conditions. It can be seen that the slopes (the right-hand decreasing lines of each curve in Figure 4) for uranium removal after loading 1−10 mL of 6 μg/mL U solutions are in all cases almost identical, which reveals the steady uranium removal pattern regardless of the sample volume and the effectiveness of 1 mol/L HNO_3 rinsing for cleaning up uranium from TEVA column in the BI format.

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Table 3. Analysis of Samples (1 L of Urine) Spiked with ^{239}Pu Using the Proposed LOV-BI Extraction Chromatographic Microflow Device

sample ID	^{239}Pu added, mBq	^{239}Pu measured, mBq	deviation, %	chemical yield of ^{242}Pu, %
1	0.48	0.40 ± 0.04	−16.7	91.2 ± 0.9
2	2.42	1.88 ± 0.10	−22.3	84.3 ± 5.2
3	5.94	5.77 ± 0.41	−2.9	90.6 ± 8.9
4	6.57	7.43 ± 0.56	13.1	103.5 ± 9.5
5	7.01	7.22 ± 0.81	3.0	89.9 ± 10.6
6	7.21	7.15 ± 0.70	−0.8	87.4 ± 7.7
7	7.24	6.28 ± 0.50	−13.3	111.9 ± 6.9
8	12.08	10.11 ± 0.99	−16.3	80.7 ± 9.8
9	25.57	19.55 ± 1.31	−23.5	77.6 ± 8.0
10	47.83	42.72 ± 3.95	−10.7	81.8 ± 7.4
11	468	447 ± 41	−4.5	98.0 ± 9.0
average			−8.2	90.6
SD				10.3

Elution of Plutonium. In our previous work, 0.1 mol/L $NH_2OH{\cdot}HCl$ dissolved in diluted HCl was used for efficient plutonium elution from the TEVA column.[31] However, the high content of $NH_2OH{\cdot}HCl$ in the eluate demands for reagent decomposition, but sometimes the removal is far from quantitative, thus deteriorating the sensitivity of the ICPMS measurement and also hampering direct injection of the eluate after column separation. In order to facilitate direct at-line injection of the final eluate into ICPMS, 0.5 mol/L HNO_3 was initially evaluated as eluent for plutonium elution. However, the experimental results indicate (results are not shown here) that less than 1% of Pu can be eluted with 10 mL of 0.5 mol/L HNO_3. This might be a consequence of the high distribution coefficient of Pu(IV) (ca. 100) in 0.5 mol/L HNO_3 onto TEVA. Thereafter, we investigated the effectiveness of HCl solution for Pu elution, and the results demonstrated (results are not shown here) that >90% of Pu can be eluted with 20 mL of diluted HCl ranging within 0.025−0.5 mol/L. Therefore, 20 mL of 0.025 mol/L HCl was finally selected as the eluent solution for Pu in this work.

Analytical Performance of the LOV-BI System. To investigate the analytical performance of the proposed LOV-BI system, 11 pooled urine aliquots (1 L of each) spiked with different amounts of ^{239}Pu were processed following the entire analytical procedure (see Table 3). The relative deviation (D_i) was calculated according to the equation $D_i = (A_i - A_{ei})/(A_{ei}) \times 100\%$, where, A_{ei} and A_i are the expected and measured values for individual samples, respectively.

The measured ^{239}Pu activities ranging from 0.48 to 468 mBq agreed well with the expected values with a mean relative deviation of −8.2%, demonstrating the satisfactory trueness of the proposed analytical method. Chemical yields of plutonium ranged from 77.6 to 111.9% with an average value of 90.6%, which compare favorably well to the ones (50−70%) reported in previous publications.[2,14]

The chemical separation method involving column renewal and executed within the LOV-BI platform is rather rapid and takes approximately 1.5 h. Notwithstanding the fact that the preliminary sample treatment is relatively time-consuming (ca. 4 h), a batch (4−8) of samples or replicates can be at least performed concurrently along with the automated column separation. Therefore, our proposed method could have an analysis turn-around time of ∼6 h, and practically we were able to analyze 6 samples in a continuous 24-h run period.

CONCLUSIONS AND PERSPECTIVES

A LOV-BI microfluidic network was developed for the first time in this work for determination of low abundance plutonium in large-sized (1 L) urine samples. Distinct analytical merits of the LOV system include automated extraction chromatography in a bead-injection format encompassing online column packing and renewal, rapid separation of interferences and precise control of bead materials and sample/solution delivery at will via versatile programmable flow. The developed flow setup is miniaturized and shows a great potential for in-line conjunction with many detectors, e.g., inductively coupled plasma atomic emission spectroscopy (ICP-AES) or ICPMS, which can be integrated in hot cells or glove boxes for fully automated processing of highly radioactive materials. Nevertheless, further improvements are still needed in future work to overcome the restricted flow rate (<1 mL/min) of flowing streams as a consequence of the build-up of back-pressure and the progressive settlement of sorptive beads within the communication channel of the multiposition valve and between the valve rotor and the LOV stator, which might pose problems of leakage and/or manifold malfunction on a long-term operation.

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Notes

The authors declare no competing financial interest.

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