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Method for Determination of Neptunium in Large-Sized Urine Samples Using Manganese Dioxide Coprecipitation and ^{242}Pu as Yield Tracer

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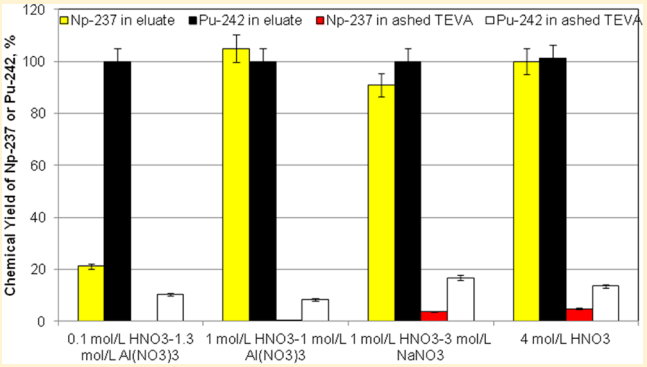
ABSTRACT: A novel method for bioassay of large volumes of human urine samples using manganese dioxide coprecipitation for preconcentration was developed for rapid determination of ^{237}Np. ^{242}Pu was utilized as a nonisotopic tracer to monitor the chemical yield of ^{237}Np. A sequential injection extraction chromatographic (SI-EC) system coupled with inductively coupled plasma mass spectrometry (ICPMS) was exploited to facilitate the rapid column separation and quantification. The analytical results demonstrated satisfactory performance of the MnO_2 coprecipitation as indicated by the high chemical yields close to 100% and high separation capacity of processing up to 5 L of human urine samples. The MnO_2 coprecipitation process is simple and straightforward in which a batch (8−12) of samples can be pretreated within 4 h (i.e., <0.5 h/sample). In connection with the automated column separation and ICPMS quantification, which takes less than 1.5 h in total, the overall analytical time was on average less than 2 h for each sample. The high effectiveness and sample throughput make the developed method well suited for urine bioassay of ^{237}Np in routine monitoring of occupationally internal radiation exposure and rapid analysis of neptunium contamination level for emergency preparedness.



Due to its considerably long half-life (2.144 Ma) compared with other actinides, ^{237}Np will be the prevailing transuranic element (TRU) in spent nuclear fuel after a few hundred years when plutonium and uranium are recycled. As an alpha emitter, ^{237}Np is highly radiologically and biologically toxic, and its relatively high bioavailability makes ^{237}Np more hazardous once it enters into the human body.[1] Thorough neptunium exposure assessment is imperative for radiation protection and medical intervention to workers or the public who are exposed to neptunium in nuclear facilities or after a radiological/nuclear incident.[2] Bioassay of urine for ^{237}Np concentration is a straightforward method for estimation of the internal radiation dose of individuals. For this purpose, it is essential to develop accurate and rapid neptunium bioassay methods for determination of ^{237}Np in urine. Over the past few decades, although a wealth of analytical methods have been developed for actinide (mostly uranium and plutonium) determination in human urine,[3−15] the neptunium urine bioassay is not well developed to meet the analytical requirements in rapid analysis for large-sized samples.[2,15]

Due to the long retention time of Np in the human body, the excretion rate of Np is very slow. Consequently, it is a requirement to measure very low levels of Np in urine to be able to meet the screening criteria of minimum internal dose of occupational and public radiation exposure. Thus, 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.

Urine has a very complicated and variable matrix composition, due to the variability in diet which differs from one individual to another with time. This matrix with a relatively large variation makes the effective separation of Np from the sample matrix sometimes complicated and difficult to predict. In order to obtain a concentrated and purified neptunium source, two steps are normally carried out, i.e., preconcentration and chemical purification.

Evaporation and coprecipitation are two often used preconcentration techniques in urine bioassay, wherein evaporation is only suitable for small volume urine samples due to its time-consuming process. On the contrary, coprecipitation is rapid and effective for processing large volumes of urine samples. Some coprecipitation methods, such as calcium phosphate ($Ca_3(PO_4)_2$),[2] bismuth phosphate ($BiPO_4$),[14] hydrous titanium oxide (HTiO),[4,16] etc., have been reported for urine bioassay of transuranics. Nonetheless, excessive PO_4^{3-} in both $Ca_3(PO_4)_2$ and $BiPO_4$ coprecipitation methods might deteriorate the separation performance when using an extraction chromatographic (EC) column (e.g., TEVA) for the further chemical purification. Despite that

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Table 1. Effect of Forms and Amount of $KMnO_4$ and $Al(NO_3)_3$ on Separation of Np and Pu by MnO_2 Coprecipitation and Extraction Chromatography[a]

group number	amount of $KMnO_4$ added, mmol[b]	reaction time, min	amount of $Al(NO_3)_3$ added,[d] mmol	chemical yield of ^{237}Np, %	chemical yield of ^{242}Pu, %	$^{237}Np/^{242}Pu$ chemical yield ratio
1	1	10	2[e]	94.2 ± 2.0	88.7 ± 11.6	1.06
2	2	10	2	107.9 ± 17.5	103.0 ± 8.5	1.05
3	2	10	0	64.3 ± 13.8	76.1 ± 8.0	0.84
4	2[c]	10	0	5.3 ± 0.6	22.5 ± 2.8	0.24
5	3	10	2	99.8 ± 6.6	95.5 ± 11.4	1.05

[a]Results are given as the average ±2SD of three replicates. [b]Unless otherwise stated, $KMnO_4$ was prepared in 0.2 mol/L of $KMnO_4$ solution. [c]$KMnO_4$ was added in solid form. [d]Unless otherwise stated, $Al(NO_3)_3$ was prepared in 2 mol/L $Al(NO_3)_3$ solution. [e]$Al(NO_3)_3$ was added in solid form.

^{237}Np was not the analyte of interest, an HTiO coprecipitation procedure reported by Dai et al.[4] was shown to be unaffected by either high contents of calcium or phosphates in urine, and actinides could be coprecipitated quantitatively with HTiO at pH 7. However, the chemical yields were on average 76 ± 8%, indicating a loss of 20% of target analyte.

Manganese dioxides coprecipitation has been utilized to capture radionuclides (e.g., Ra, Ac, Th, U, Pb, Po) from large volume environmental samples (e.g., seawater),[17−21] whereas, to the best of our knowledge, this method has not yet been utilized to handle biological samples. This is probably attributed to the complex matrix composition of urine which might greatly affect the efficiency of manganese dioxide coprecipitation.

To reduce labor intensity and analytical time, there has been an increasing interest in the employment of flow-based automated approaches for chemical separation after preconcentration of radionuclides from samples.[22−24] Nevertheless, to the best of our knowledge, all the previously published automated methods were limited to process ≤1 L of urine samples. This might be a limitation of the down-scaled automated systems developed previously which are not readily able to handle large volume sample/reagent injections for trace level radio-bioassays since compatibility with processing large-sized samples is often required.[25−28]

For monitoring the chemical yield of ^{237}Np in chemical separation, neptunium isotopic tracers, such as the long-lived ^{236}Np ($t_{1/2}$ = 5000 a) and the short-lived ^{239}Np ($t_{1/2}$ = 2.33 d) can be used. However, no commercial ^{236}Np solution with sufficient purity is currently available; the short-lived ^{239}Np can be produced by neutron irradiation of uranium in a nuclear reactor or separated from a ^{243}Am solution, but neither is easily obtained. Meanwhile, the short-lived ^{239}Np requires the individual measurement of ^{239}Np by gamma spectrometry, and ^{239}Np produced from irradiated uranium normally contains some amount of ^{237}Np, which would interfere with the measurement of very low levels of ^{237}Np in the sample.

In this work, a novel analytical method was developed for rapid and automated determination of neptunium in large-sized human urine samples. Streamlined MnO_2 coprecipitation and sequential injection extraction chromatography (SI-EC) coupled with inductively coupled plasma mass spectrometry (ICPMS) were the main cornerstones to expedite the entire neptunium urinalysis. ^{242}Pu was used as a nonisotopic tracer for monitoring the chemical yields of ^{237}Np. Analytical performance of the developed method including the effectiveness of MnO_2 coprecipitation, sorption behavior of neptunium, and separation capacity of the SI-EC platform was thoroughly investigated and evaluated.

EXPERIMENTAL SECTION

Reagents and Samples. Nitric acid (65%), hydrochloric acid (37%), phosphoric acid (85%), ammonia (25%), hydrogen peroxide (30%), ferrous sulfate, potassium disulfite, sodium hydroxide, potassium permanganate, potassium dihydrogen phosphate, calcium nitrate, and bismuth nitrate were analytical grade reagents, and all solutions were prepared with deionized water (18 MΩ·cm). ^{237}Np solution of 0.01175 Bq g^{-1} in 2 mol/L HNO_3 was diluted from a stock solution supplied by Center for Nuclear Technologies, Technical University of Denmark. ^{242}Pu standard solution (0.1037 Bq/g in 2 mol/L HNO_3) was diluted from NBL-CRM 130 purchased from New Brunswick Laboratory (Argonne, IL). TEVA extraction chromatographic resin (100−150 μm particle size) was purchased from TRISKEM International (Bruz, France).

Pooled human urine samples were collected from fifteen Danish healthy residents and preserved in clean and sealed polyethylene barrels under 5 °C and processed as soon as possible after the sample collection. One liter of urine spiked with a known amount (0.5−50 mBq) of ^{237}Np was used as a sample for method development. To investigate the separation capacity of the SI-EC system developed in this work, 2−5 L of urine samples were used and processed through the entire analytical procedure.

Sample Pretreatment. *Preconcentration with MnO_2 Coprecipitation.* To 1 L of human urine spiked with a certain amount of ^{237}Np, 10 mL of 37% HCl was added to acidify the sample to pH = 1, and then, 5 mBq of ^{242}Pu was added as a tracer to monitor the chemical yield of ^{237}Np. The pH of the sample was adjusted to 7 using 25% $NH_3·H_2O$; 1−3 mmol of $KMnO_4$ in the form of 0.2 mol/L $KMnO_4$ solution or solid (see details in Table 1) was slowly added while stirring to form MnO_2 after reacting with the reductants naturally contained in urine (e.g., urea). One to two milliliters of 25% $NH_3·H_2O$ was then added to adjust the pH to 9 to stabilize the MnO_2 precipitate. The sample was stirred for 10 min to allow for the complete uptake of Pu and Np onto MnO_2 and then transferred into four 250 mL centrifuge tubes and centrifuged at 4000 rpm for 10 min.

Decomposition of Organic Matter. The supernatant was discarded, and the precipitate was transferred into a beaker with 20 mL of concentrated nitric acid plus a few drops of 30% H_2O_2. 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 30% H_2O_2 to a total volume of 10 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.

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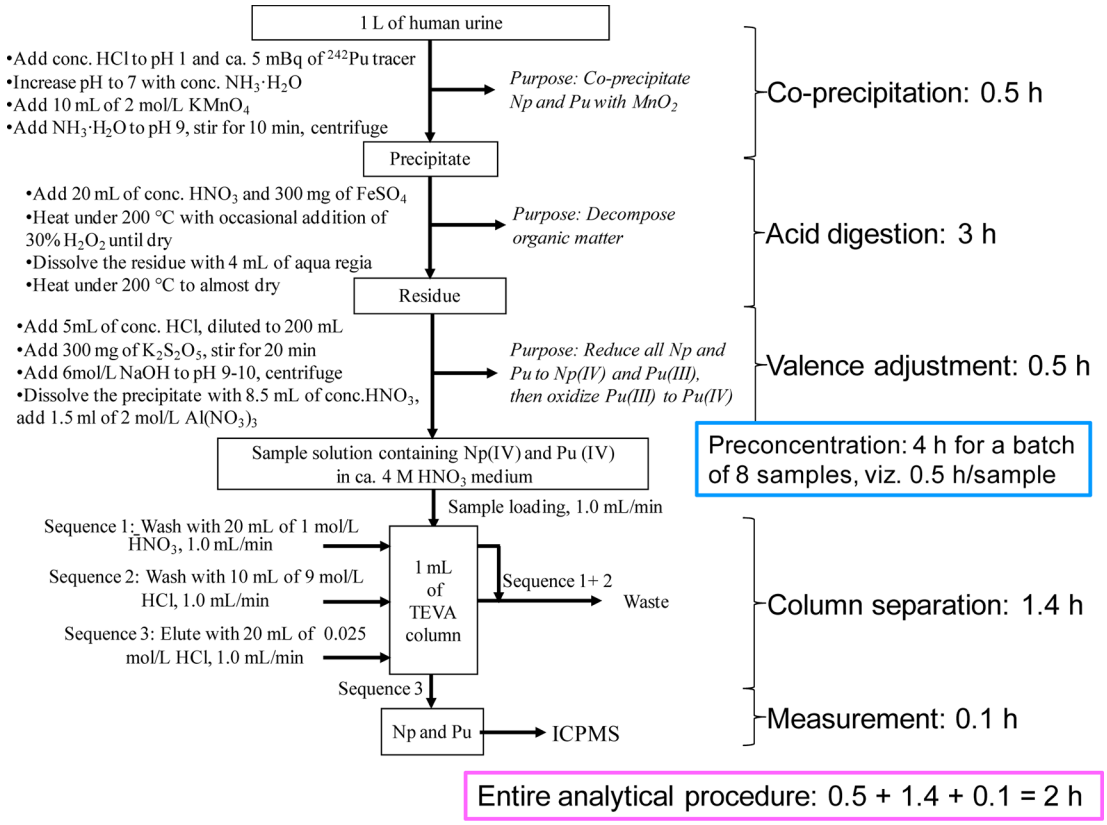

Figure 1. Optimized separation procedure for ^{237}Np determination in 1 L of human urine using MnO_2 coprecipitation.

Valence Adjustment of Np(IV) and Pu(IV) and Solution Preparation for Column Separation. $K_2S_2O_5$ (300 mg) was added, and the sample was stirred for 20 min to reduce Np and Pu to Np(IV) and Pu(III), respectively. 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, the precipitate was dissolved with 5 mL of 65% HNO_3, and the solution was transferred to a vial; the centrifuge tube was washed with 3.5 mL of 65% HNO_3, and the wash was combined with sample solution. Then, 2 mmol of $Al(NO_3)_3$ in the form of 2 mol/L $Al(NO_3)_3$ solution or solid (see Table 1) was added to the sample to complex any interfering sulfate and phosphate remaining in the sample. The sample solution was finally adjusted to ca. 4 mol/L HNO_3 (sample volume was 30 mL) for column separation using the SI-EC system.

Comparison of MnO_2 Coprecipitation with Other Traditional Preconcentration Methods. To evaluate the effectiveness of the MnO_2 coprecipitation technique relative to other traditional preconcentration methods, spiked urine samples were also processed using direct evaporation and $Ca_3(PO_4)_2$ or $BiPO_4$ coprecipitation. Within the evaporation process, 1 L of urine spiked with known amounts of ^{237}Np and ^{242}Pu was directly evaporated to dryness and then ashed in a muffle furnace at 550 °C overnight. The residue was leached with 30 mL of *aqua regia* at 150 °C for 2 h and then filtered through a GF/A filter paper (Whatman Intertional Ltd., Maidstone, England). After adding 1 mL of 0.5 g/mL $FeCl_3$, the sample pH was adjusted to 8−9 with 25% $NH_3{\cdot}H_2O$. The supernatant was discarded after centrifugation, and the residue was dissolved with diluted HCl and subject to valence adjustment and solution preparation for column separation as described above.

The $Ca_3(PO_4)_2$ and $BiPO_4$ coprecipitation procedures used in this work were based on literatures.[2,14] In brief, for $Ca_2(PO_4)_3$ coprecipitation, after the acidification of the sample solution to pH = 1 with 37% HCl and spiking ^{242}Pu tracer, 1 mL of 1.3 mol/L $Ca(NO_3)_2$ and 2 mL of 0.65 mol/L KH_2PO_4 were added, respectively. The sample was heated to 40 °C, and 25% $NH_3{\cdot}H_2O$ was added to adjust the pH to 9−10 to form the $Ca_3(PO_4)_2$ precipitate. For $BiPO_4$ coprecipitation, after acidification and spiking ^{242}Pu tracer, the sample was boiled on a hot plate for 20 min and 1 mL of 20% $NH_2OH{\cdot}HCl$ was added to reduce Pu to Pu(III) and Np to Np(IV), respectively. Five milliliters of 85% H_3PO_4 and 5 mL of 0.3 mol/L $Bi(NO_3)_3$ were added, and the sample was heated to 60 °C. Then, 25% $NH_3{\cdot}H_2O$ was slowly added to reach a pH of 1.5 to obtain the $BiPO_4$ coprecipitate. After centrifuge, both $Ca_3(PO_4)_2$ and $BiPO_4$ precipitates were subject to organic decomposition and valence adjustment steps as described above prior to the chemical purification.

Automated Purification with SI-EC System. The automated chemical purification was performed with extraction chromatography in an SI-EC system developed previously in our lab.[22] An Econo-Column (Bio-Rad Laboritories Inc., Hercules, CA) (5 mm inner diameter and 50 mm length, volume ca. 1 mL) packed with TEVA resin was assembled in the SI-EC system. The chromatographic separation was composed of the following steps: (1) cleaning the holding coil and the inlet and outlet of all tubing and preconditioning the TEVA column with 20 mL of 4 mol/L HNO_3; (2) loading sample solution onto the column; (3) rinsing the column with 20 mL of 1 mol/L HNO_3 followed by 10 mL of 9 mol/L HCl; (4) eluting ^{237}Np (along with ^{242}Pu) with 20 mL of 0.025−0.5 mol/L HCl (see the details in Elution of Neptunium in the

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Table 2. Comparison of Analytical Performance of MnO_2 Coprecipitation with Other Traditional Preconcentration Methods[a]

preconcentration method	operational time	chemical yield of ^{242}Pu, %	chemical yield of ^{237}Np, %	ratio of ^{237}Np/^{242}Pu chemical yield
MnO_2 coprecipitation	4 h	103.0 ± 8.5	107.9 ± 17.5	1.05
$Ca_3(PO_4)_2$ coprecipitation	6 h	46.8 ± 4.1	8.3 ± 5.4	0.18
$BiPO_4$ coprecipitation	8 h	39.4 ± 4.0	4.5 ± 0.5	0.11
evaporation	1.5 days	75.5 ± 7.6	81.1 ± 8.1	1.07

[a]Results are given as the average ±2SD of two replicates.

Results and Dicussion section). The flow rate for the entire chemical separation was controlled at 1.0 mL/min.

Investigation on Sorption Behavior of Neptunium and Plutonium. To investigate the sorption behavior of Np(IV) and Pu(IV) onto TEVA resin, four groups of artificial samples containing 10 mBq of ^{242}Pu and 1.0 mBq of ^{237}Np were prepared in 0.1 mol/L HNO_3−1.3 mol/L $Al(NO_3)_3$, 1 mol/L HNO_3−1 mol/L $Al(NO_3)_3$, 1 mol/L HNO_3−3 mol/L $NaNO_3$, and 4 mol/L HNO_3, respectively. Oxidation states of Np and Pu were adjusted to Np(IV) and Pu(IV) with ascorbic acid followed by $NaNO_2$. Each sample solution was loaded onto a 1 mL TEVA column, and then, Np and Pu were eluted directly with 10 mL of 0.025 mol/L HCl. The remaining TEVA resin was collected and dried under 100 °C on a hot plate and then ashed under 550 °C overnight in muffle furnace. The final residue after ashing was dissolved with 5 mL of 0.025 mol/L HCl and, together with the neptunium eluate, delivered for ICPMS measurement.

ICPMS Measurement. The detection of ^{237}Np (as well as ^{242}Pu) with ICPMS (X Series^{II}, Thermo Fisher Scientific, Waltham, MA) was performed after the addition of indium as internal standard to a final concentration of 1 μg/L. The ICPMS was equipped with an Xs-skimmer cone and a Burgener MiraMist nebulizer under hot plasma conditions. The detection limit calculated as three times the standard deviation (3σ) of the processing blank was 1.5 pg/L (equal to 0.4 nBq/L) for ^{237}Np. A least-squares regression line over the range of 0.01−100 ng/L was used for quantification of neptunium and plutonium. Prior to detection, the ICPMS instrument was tuned to maximum transmission of uranium using a 1 μg/L solution of ^{238}U and the instrumental parameters were further adjusted for neptunium using 4.243 ng/L of ^{237}Np solution to optimal detection efficiency. The typical operational conditions of the instrument have been listed in our previous work.[29] It is noteworthy that these parameters were optimized each time when the instrument was initialized. Typical sensitivities of neptunium and plutonium ranged from 1×10^5 to 5×10^5 cps per μg/L.

RESULTS AND DISCUSSION

Preconcentration of Np from Urine. MnO_2 coprecipitation was applied to preconcentrate and separate Np from the urine matrix in order to reduce the high content of inorganic species in urine, which induce a noisy background to the entire spectrum in the ICPMS measurement, lower the effective plasma temperature, and give rise to serious interferences at mass 237 through formation of polyatomic species. Parameters governing the analytical performance were investigated and optimized including the amount or form of precipitant, pH, reagents for organic matter decomposition, and adjustment of oxidation states of Np and Pu. The optimized preconcentration procedure is illustrated in Figure 1, and the detailed results for method optimization are discussed below.

MnO_2 Coprecipitation. As indicated by the chemical yields of ^{237}Np and ^{242}Pu and their corresponding ratio in Table 1, ^{237}Np and ^{242}Pu demonstrate equally high affinity onto MnO_2 particles and the addition of 2 mmol of $KMnO_4$ is effective enough to carry most of the ^{237}Np and ^{242}Pu into the precipitate (see group 2 in Table 1). The uptake process of ^{237}Np and ^{242}Pu onto the MnO_2 precipitate is very fast and can be completed within 10 min with very high chemical yields (ca. 100%) (Table 1).

It is known that sample pH is very crucial to ensure a high chemical yield, because the Mn-containing products from redox reactions depend very much on the pH of the media: barely in neutral solution, permanganate is reduced to MnO_2, while in acidic or alkaline solutions permanganate is reduced to the Mn(II) ion or MnO_4^{2-}.[30] Therefore, practically, the pH value of the sample solution was first adjusted to 7−8 to allow the formation of MnO_2 through the redox reaction between $KMnO_4$ and the reducing agents contained in urine, such as urea ($(NH_2)_2CO$) and other organic substances, followed by increasing the pH to 9−10 to promptly entrap the Np and Pu by MnO_2 and allow settling of the precipitate.

At this point, it is also noteworthy that appropriate storage (under <5 °C in sealed barrel) of urine sample is important to guarantee the stabilization of the urine matrix, since urea is easily converted to ammonium by urease under a ventilated air condition.[31] We have observed an intensive formation of foams (assumed as the formation of ammonium from urea) in urine samples stored under room temperature for 2−3 days after collection, and for these samples, MnO_2 coprecipitation was not obtained quantitatively even though the same experimental operations as mentioned above were followed. To ensure the consistent formation of MnO_2, it is also advisable to add an extra amount of reducing reagents such as $MnCl_2$, ascorbic acid, or hydroxylamine hydrochloride after the addition of $KMnO_4$ solution.

Another important parameter affecting the MnO_2 coprecipitation efficiency is the form of $KMnO_4$ reagent added into the urine sample. When solid $KMnO_4$ was directly added into the urine sample, extremely low chemical yield (ca. 5% for ^{237}Np) was obtained (group 4, Table 1). This might be attributed to the highly concentrated $KMnO_4$ dissolved in a partial region of the sample solution which acts as a strong oxidant and reacts aggressively with the reducing substances in urine wherein MnO_2 is not formed.

To verify the effectiveness of MnO_2 coprecipitation over other preconcentration techniques, including evaporation and $Ca_3(PO_4)_2$ and $BiPO_4$ coprecipitation, analytical performances of these techniques were compared and the analytical results are summarized in Table 2. Unfavorably low chemical yields of ^{237}Np (5−10%) and poor synchronism between the Np and Pu sorption behavior (as indicated by the ^{237}Np/^{242}Pu chemical ratios (0.1−0.2) deviating far from 1.0) were encountered when using $Ca_3(PO_4)_2$ and $BiPO_4$ coprecipitation processes, which might be consequences of (1) low and unequal uptake

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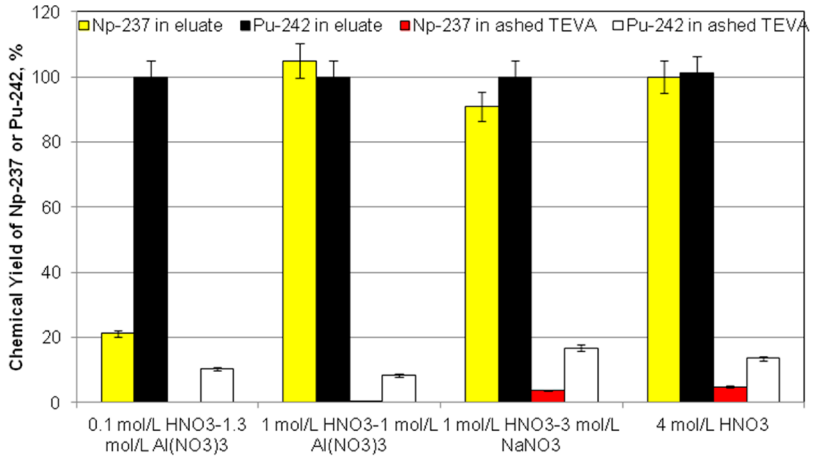

Figure 2. Sorption behavior of Np(IV) and Pu(IV) onto TEVA in nitric acid solutions with different composition.

efficiency of Np and Pu onto the phosphates precipitate surface within a limited reaction time (ca. 10 min); (2) deterioration of the separation performance of TEVA column for Pu and Np absorption caused by the highly competitive absorption of phosphates.[32] Although with the evaporation technique, equally satisfactory chemical yields were obtained for ^{237}Np (ca. 80%) and ^{242}Pu (ca. 75%), relatively long analytical time (1.5 day) is needed since this approach often requires overnight ashing to remove the large amount of organic matter. This prolongs the analytical procedure and thus impairs the sample throughput which is especially important in emergency situations. Therefore, on the basis of the overall consideration regarding the analytical time, chemical yields of ^{237}Np, and the separation behavior coherence between ^{242}Pu and ^{237}Np, MnO_2 coprecipitation is selected as the optimal technique for preconcentration of large-sized urine samples.

Decomposition of Organic Matter. During the coprecipitation step, a certain amount of organic substances would also follow Np and Pu entering into the MnO_2 coprecipitate, which might deteriorate the column performance in following separation. Therefore, an acid digestion was exploited herein to decompose the organic matter contained in the precipitate.

The preliminary investigation has shown that a mixture of HNO_3–H_2O_2 solution performed better than *aqua regia*–H_2O_2 with respect to the decomposition of organic matter contained in urine, as indicated by the residue color after digestion and dryness (white residue for HNO_3–H_2O_2 digestion vs dark residue for *aqua regia*–H_2O_2 digestion). Moreover, the addition of ferric ions promoted the effectiveness of organic matter decomposition by forming Fenton's reagent in the Fe–H_2O_2–HNO_3 system, wherein iron acts as a catalyst (see eq 1 and 2) to form hydroxyl and peroxide radicals, therefore accelerating the decomposition of organic matter.[33] Besides, the Mn(IV)/Mn(II) redox pair also perform as a catalyst in a similar way during the acid digestion process which assists in the production of hydroxyl and peroxide radicals.

$$Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + OH\cdot + OH^- \quad (1)$$

$$Fe^{3+} + H_2O_2 \rightarrow Fe^{2+} + OOH\cdot + H^+ \quad (2)$$

Therefore, 20 mL of conc. HNO_3 with occasional addition of 30% H_2O_2 in total volume of 10 mL was used for the acid digestion, and a few hundred milligrams of $FeSO_4$ was added to form Fenton reagents with H_2O_2 to stimulate the organic decomposition reaction. The sample was heated to dryness at 200 °C so as to completely decompose the remaining organic matter and then redissolved with a few milliliters of *aqua regia* (since we observed that the dried residue was not easily dissolved with other single acid solutions (e.g., HNO_3 or HCl)) and heated to almost dryness.

Valence Adjustment of Np(IV) and Pu(IV). As per the equally highest uptake of Np(IV) and Pu(IV) onto TEVA resin, it is imperative to adjust both Np and Pu to tetravalent states before loading onto the column for further chemical separation. Therefore, herein, we exploited a two-step valence adjustment using Fe–$K_2S_2O_5$–HNO_3 as reported previously,[34] in which the presence of iron will also perform as a catalyst to prompt the reduction of high valence Np and Pu to Np(IV) and Pu(III), respectively. Different from our previous work, the sample solution was prepared in 4 mol/L HNO_3 because Np(IV) and Pu(IV) have lower distribution factors onto the TEVA column in highly concentrated nitric acids (e.g., 8 mol/L). The addition of $Al(NO_3)_3$ to the sample solution before loading onto the TEVA column show a significant effect on the separation performance of Np and Pu in the extraction chromatographic process (see group 2, Table 1). This might be attributed to the masking of phosphate through formation of complexes with Al, consequently eliminating the competitive adsorption of phosphate onto TEVA resin.[32] Phosphate and calcium are components of human urine; calcium contained in urine may form precipitates with phosphate and is coprecipitated along with MnO_2 when the pH value of the sample solution was raised to 8–9 in the coprecipitation process and therefore enters into the final solution loaded onto the TEVA column. Compared with the solid form, $Al(NO_3)_3$ solution is more effective for masking phosphate. However, addition of a highly excessive amount of $Al(NO_3)_3$ should be avoided, because of high viscosity of $Al(NO_3)_3$, which increases the viscosity of sample solution, consequently impairing the analytical performance of the SI-EC system by leakage or clog problems.

Sorption Behavior of Neptunium and Plutonium on TEVA. It is well-known that Np(IV) and Pu(IV) have similar sorption behavior onto TEVA resin in nitric acid medium, and their affinities to the resin strongly depend on the concentration of HNO_3 with the highest distribution coefficiency at about 3 mol/L.[33] To investigate the effect of $[NO_3^-]$ and $[H^+]$ on the adsorption of Np(IV) and Pu(IV) on TEVA resin, four Np(IV)–Pu(IV) solutions were prepared in an identical 4.0 mol/L of $[NO_3^-]$ but different $[H^+]$ (0.1–4

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mol/L); the solutions were loaded onto the TEVA column followed by direct elution. The amounts of Np and Pu in eluate and resin were measured (see details in Experimental Section). The results (Figure 2) show that Pu(IV) has a consistently strong sorption onto TEVA in all solutions investigated, regardless of the variation in acidity and composition. Np(IV) was also retained strongly onto TEVA in 1 mol/L HNO_3−1 mol/L $Al(NO_3)_3$, 1 mol/L HNO_3−3 mol/L $NaNO_3$, and 4 mol/L HNO_3 solutions. However, a significant low yield of Np was observed for solution of 0.1 mol/L HNO_3−1.3 mol/L $Al(NO_3)_3$. Np(IV) might be readily oxidized in diluted nitric acid under aerobic conditions; the valence changes of Np(IV) to most likely Np(V) (which has lower distribution coefficiency onto TEVA) might be the main reason of the low Np yield for 0.1 mol/L HNO_3−1.3 mol/L $Al(NO_3)_3$ solution, and the low acidity might have less effect on the adsorption of Np(IV) onto TEVA resin. It is, therefore, assumed that the key factor affecting the sorption of Np(IV) and Pu(IV) onto TEVA is the concentration of NO_3^-, which forms anion complexes with Np(IV) and Pu(IV) to be adsorbed on the exchangeable sites of TEVA resin. However, the oxidation states of Pu and Np have to be adjusted and remain in Np(IV) and Pu(IV) before loading onto the column. This investigation also confirms the feasibility of applying ^{242}Pu as a nonisotopic tracer for neptunium determination in consideration to ensure the similar sorption behavior of Np(IV) and Pu(IV) on the TEVA column.

Elution of Neptunium. Lariviere et al.[23] have compared five different eluents, including 0.1 M $(NH_4)_2C_2O_4$, H_2O, 0.1 M HCl, and 0.01 M HNO_3 for plutonium elution from TEVA column, and concluded that both 0.01 M $(NH_4)_2C_2O_4$ and 0.1 M HCl could be used for plutonium elution. Because oxalates have a potential risk of forming black carbon grains in the torch due to the incomplete decomposition during the ICPMS measurement,[22] we chose diluted HCl as an eluent for neptunium (along with ^{242}Pu) elution and investigated the effect of concentration of HCl on the efficiency of Np and Pu elution. To double-check the elution efficiency, the final remaining TEVA resin was analyzed to measure the retention of Np and Pu on the resin by ashing the resin at 550 °C in Muffle furnace overnight. The results (Figure 3) show that, in the range of 0.025−0.5 mol/L, the lower the concentration of HCl, the slightly better is the elution efficiency for neptunium. With the first 20 mL of eluents, ≥90% of Np and Pu can be eluted from the column in most cases. Therefore, 0.025 mol/L HCl was finally selected as an eluting solution in this work.

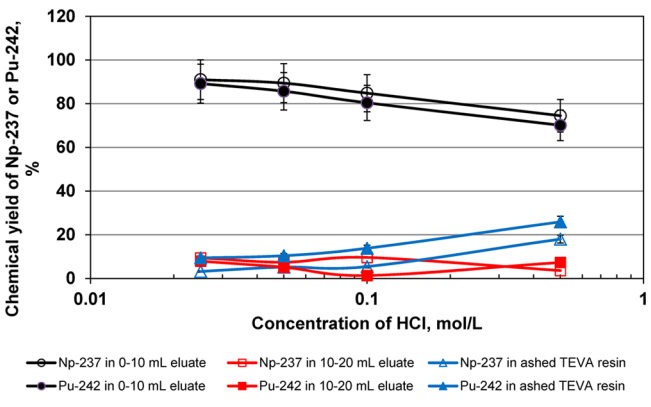

Figure 3. Efficiency of different concentrations of HCl on elution of Np and Pu from TEVA column.

Compared with neptunium, the elution of ^{242}Pu was relatively slow since the remaining proportion in the final ashed TEVA was higher in most cases. This might be due to tetravalent Pu having a relatively higher distribution factor in diluted HCl medium.[32] Although asynchronic eluting behavior between neptunium and plutonium can be observed in some cases, the sum of neptunium in the first 20 mL of eluates is consistent with plutonium recovery with an average $^{237}Np/^{242}Pu$ chemical yield ratio of 1.07 ± 0.07, which means ^{242}Pu can be used as a nonisotopic tracer for Np urine analysis.

Separation Capacity and Sample Throughput. To assess the capacity of the 1 mL extraction chromatographic column integrated in the SI system for separation of Np, 2−5 L of urine samples were processed through the entire procedure. The results (Table 3) indicate that the SI-EC method showcases a high separation capacity of up to 5 L of urine with satisfactory chemical yield (70%). Compared with the previously reported values (≤1.0 L),[15,35] the concentration factor and thus the minimum detectable concentration of neptunium could be ≥5-fold better.

Table 3. Evaluation of Separation Capacity of the Developed Method for Np and Pu in Urine[a]

volume of urine, L	chemical yield of ^{242}Pu, %	^{237}Np spiked	^{237}Np measured	difference, %
1	103.0 ± 8.5	0.803 ± 0.017	0.860 ± 0.113	7.10
2	96.8 ± 9.1	1.126 ± 0.154	1.179 ± 0.177	4.71
3	81.4 ± 4.1	1.270 ± 0.038	1.208 ± 0.072	4.88
5	77.6 ± 7.8	1.337 ± 0.135	1.340 ± 0.062	0.22

[a]Results are given as the average ±2SD of two replicates.

With the developed method, processing a single sample takes about 2 h, offering a daily throughput approaching 12 samples. Compared with the traditional manual methods which take 2−3 days,[15] the separation efficiency is here 20−30-fold improved. The proposed method is therefore better suited for the rapid neptunium urine bioassay as endorsed in assessment of internal radiation dose of humans exposed to Np contamination and for radiobioassay in radiological emergencies.

Validation of the Developed Method. To evaluate the accuracy of the proposed analytical method for handling urine samples, seven aliquots of 1 L of urine spiked with different amounts of ^{237}Np ranging from ~0.005 to 0.6 mBq were analyzed. The measured activities of ^{237}Np for the overall samples agree well with the expected values as illustrated Figure 4. The correlation between the measured activity of ^{237}Np (A_{mea}) and the expected activity of ^{237}Np (A_{exp}) is A_{mea} = 1.01 A_{exp}, with R^2 = 0.9969, demonstrating satisfactory accuracy and precision of the analytical method developed in this work.

CONCLUSIONS

In this paper, a novel SI-EC method using a downscaled TEVA column (1 mL) has been proposed for rapid determination of neptunium in large-sized (up to 5 L) urine samples. The streamlined MnO_2 coprecipitation-based analytical procedure features attributes including effective scavenging of neptunium, simplicity, and the short analysis time required (on average <2 h per sample). This in turn affords the developed SI-EC analytical method with reduced labor intensity and enhanced sample throughput, which is well suited in the

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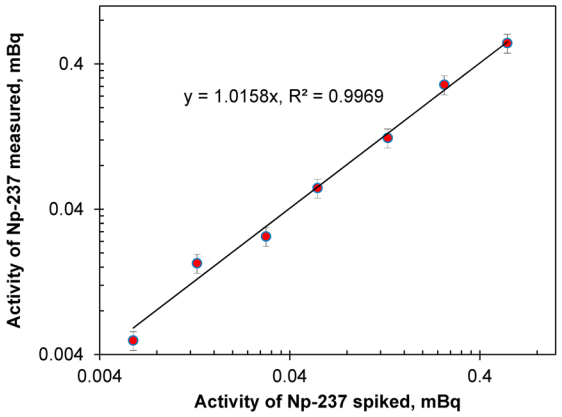

Figure 4. Analytical results for urine samples (1 L of each) spiked with different levels of ^{237}Np.

application of routine healthy monitoring and emergency preparedness programs.

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Notes

The authors declare no competing financial interest.

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