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Hydride generation-headspace single-drop microextraction-electrothermal atomic absorption spectrometry method for determination of selenium in waters after photoassisted prereduction

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Abstract

A headspace single-drop microextraction (SDME) method has been developed in combination with electrothermal atomic absorption spectrometry for determination of total inorganic Se and Se(IV). SeH_2 is generated in a 40 mL volume closed-vial and trapped onto a Pd(II)-aqueous containing drop that is supported at the needle tip of a high-precision chromatographic syringe. Sample pre-treatment by UV irradiation prior to hydride generation allowed converting Se(VI) into Se(IV), hence, facilitating total Se determination. A $2_{\text{IV}}^{6\text{-}2}$ fractional factorial design was employed for characterising the effect of relevant variables over SeH_2 trapping. The variables showing the most significant effect were Pd(II) concentration in the drop and extraction time. A preconcentration factor of about 25 is achieved. The limit of detection of Se was 0.15 ng/mL using trapping onto a 3-μL drop and the precision expressed as relative standard deviation was about 3%. The limit of detection could be improved further using repeated sampling of the headspace.

Keywords: Selenium hydride; Headspace single-drop microextraction; Pd(II)-containing drop; ETAAS

1. Introduction

Single-drop microextraction (SDME) has emerged during last years as a simple, cost-effective and virtually solvent-free sample preparation technique for preconcentration and separation prior to detection [1–4]. This technique has been originally developed for organic analytes, using a drop of organic solvent suspended from the tip of a syringe needle, which was immersed in the sample solution placed in a closed vial. First, comparisons of this novel technique with solid-phase microextraction (SPME) have indicated that similar performance is reached in terms of precision, sensitivity and analysis time [2]. However, SDME could have two advantages as compared to SPME, namely, (i) the fiber lifetime is limited and prolonged usage can affect precision; unlike fibers, drops can be renewed for each extraction. (ii) SPME is limited by the availability of fiber coatings, but on the contrary, a wide variety of solvents and trapping agents can be used in SDME. When headspace sampling (HS) is feasible (i.e. analytes with large Henry's law constants), the equilibrium time is shortened as compared with the direct sampling in the solution, and in turn, potential interferences caused by the matrix are avoided [4].

Recently, the authors have reported two applications concerning preconcentration of arsine [5] and methylmercury [6] using the principles of HS–SDME in combination with electrothermal atomic absorption spectrometry (ETAAS) for detection. The method is based on the formation of hydrides derivatives, headspace sampling and trapping onto a Pd(II)-containing aqueous drop. Trapping of the hydrides onto the drop does not rely on their partitioning among the gas phase

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and the solvent phase, as typically developed for organic compounds, but on their catalytic decomposition onto the Pd(0) formed in the drop as a result of the hydrogen evolved during the derivatisation reaction. A faster mass transfer was observed for the methylmercury hydride from the headspace to the drop in comparison with SPME, which allowed equilibrium to be reached in a significantly shorter time [7].

Determination of Se by a sensitive technique, such as ETAAS in complex matrices (e.g. high salt contents) is troublesome, owing to the occurrence of both spectral and non-spectral interferences caused by salts [8]. Detection limits reported for determination of Se in seawater by ETAAS are 0.8 ng/mL with a Pd modifier [8] and 3 ng/mL with a Ni–NH_4NO_3 modifier [9]. The use of STPF conditions with Zeeman background correction yields a LOD for Se in seawater of 10 ng/mL [10]. Nevertheless, methods involving hydride generation with in situ preconcentration in the graphite furnace [11] or prior chelation and sorption [12] provide LODs in the pg/mL level. The aim of this work is to extend the SDME technique to preconcentration of Se prior to ETAAS determination in complex matrices using D_2-background correction equipment. The use of Pd in the drop serves two purposes, i.e. a matrix modifier for Se in the furnace and an effective trapping agent in the drop. Determination of total inorganic Se and Se(IV) is accomplished following pre-reduction of Se(VI) to Se(IV) by UV irradiation.

2. Experimental

2.1. Apparatus

A Unicam (Cambridge, UK) Model Solaar 939 atomic absorption spectrometer equipped with a GF-90 graphite furnace was used. A selenium hollow cathode lamp operated at 15 mA was employed as a radiation source. Atomic absorption measurements were carried out at 196.0 nm using a spectral bandpass of 0.5 nm. Pyrolytic graphite-coated graphite tubes were employed and D_2-background correction were employed throughout.

Headspace single-drop microextraction was performed with a commercially available 10-μL syringe containing a guided-PTFE plunger (Fig. 1). SeH_2 generation was carried out in a 40 mL closed-vial with a silicone rubber septum. A 705 UV-digester (Hg high-pressure lamp, 500 W) (Metrohm) was used for photoassisted prereduction of Se(VI).

2.2. Reagents

All chemicals were of analytical-reagent grade. A 1000 mg/L stock standard solution of Se(IV) from H_2SeO_3 in 1 M HNO_3 (Panreac) was employed. A 1000 mg/L stock solution of Se(VI) was prepared by dissolving the appropriate amount of Na_2SeO_4 (Sigma). Working standard solutions were made just before use by appropriate dilution of the stock standard solutions. Sodium tetrahydroborate stabilised with 0.05% (m/v) NaOH and hydrochloric acid were employed (Merck). For interference study, the following salts and organic compounds were used: NaCl, $Mg(NO_3)_2$, $MgCl_2$ $6H_2O$, oxalic acid (Merck); $CaCO_3$ (Aldrich); $Fe(NO_3)_3{\cdot}9H_2O$ (Probus); humic acid (Fluka). CRM TM-27.2 low level fortified standard for trace analysis (National Water Research Institute of Canada) was used for validation.

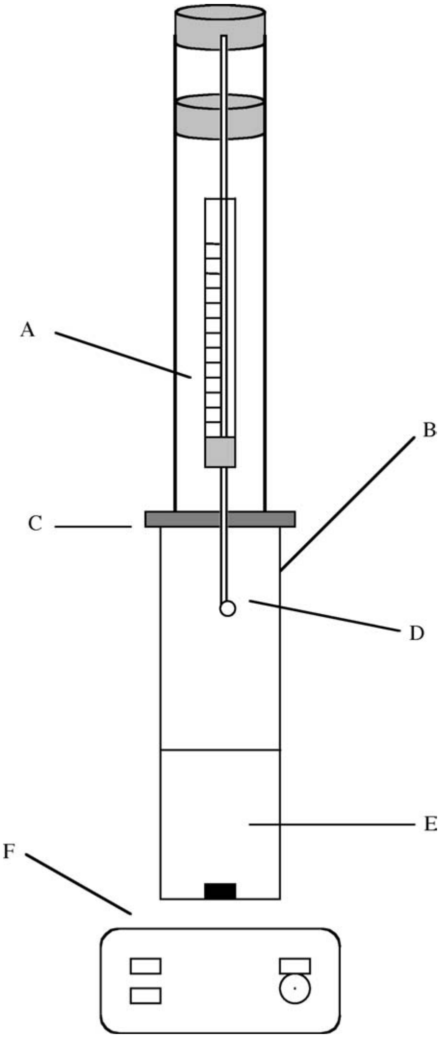

Fig. 1. Headspace single-drop microextraction device for preconcentration and matrix separation of Se prior to ETAAS detection: (A) guided microsyringe containing a PTFE plunger; (B) 40 mL-volume sampling vial; (C) septum; (D) 3 μL-volume aqueous drop containing Pd(II) (E) sample solution (F) magnetic stirrer.

2.3. Procedure

2.3.1. Determination of total inorganic Se [Se(IV) + Se(VI)] and Se(IV)

A 20 mL solution in 1.5 M HCl containing Se(IV) + Se(VI) is subjected to UV irradiation for 45 min in order to convert Se(VI) into Se(IV). The pre-treated sample is then transferred

Table 1
Thermal program for determination of selenium by ETAAS following headspace single-drop microextraction

Stage	Temperature (°C)	Hold time (s)	Ramp (°C/s)	Gas flow-rate (mL/min)
Drying	120	30	10	300
Ashing	750	20	50	300
Atomisation	2200	3	(off)	0
Cleaning	2700	3	500	300

into the microextraction vial. H_2Se is generated following injection of 0.1 mL of 1% (m/v) $NaBH_4$ into the closed vial. A 3-μL drop containing 30 mg/L of $Pd(NO_3)_2$ in 1.5% (m/v) HNO_3 is formed at the needle tip of a microsyringe and exposed to the headspace. The sample solution is stirred for 90 s so that an efficient mass transfer to the drop is facilitated. Once SeH_2 has been trapped, the drop is retracted back, the microsyringe is removed from the vial and the enriched drop with Se is injected into the graphite furnace. Se absorbance measurements are obtained after running the thermal program shown in Table 1. Peak height of the atomic absorption signal was taken as the analytical response.

For selective determination of Se(IV), the above procedure was applied a different sample aliquot (20 mL) without UV irradiation treatment.

3. Results and discussion

3.1. Optimisation of single-drop microextraction

In order to get a preliminary characterisation of this novel technique, the $2_{\mathrm{IV}}^{6\text{-}2}$ factorial fractional design was employed for screening optimisation [13]. This optimisation approach requires 16 experiments to be performed. The variables screened and their minimum (−) and maximum (+) levels were: (A) palladium concentration used as trapping agent in the drop (1–30 mg/L); (B) sodium tetrahydroborate concentration (1–3.5%); (C) volume of $NaBH_4$ injected (0.1–1 mL); (D) microextraction time (1–120 s); (E) sample volume (5–20 mL); and, (F) HCl concentration (0.25–1.5 M). Three replicates were performed for each experiment. Standardized effects obtained are shown in Fig. 2. Main effects were considered significant when they were beyond the experimental error, calculated as two times the average standard deviation of all experiments. As can be observed, the Pd(II) concentration in the hanging drop (A) and the microextraction time (D) provided the largest main effects, being positive in both cases. The sample volume (E) and HCl concentration (F), in spite of being significant, provided lower main effects than variables (A) and (D). The most significant variables (i.e. A and D) were also optimised following the univariate approach. The effect of Pd(II) concentration in the hanging drop is shown in Fig. 3. Peak absorbance increased up to ca. a 25 mg/L Pd(II) concentration. Fig. 4 shows the influence of the microextraction time on integrated absorbance. Equilibrium conditions are reached from 60 s after injecting the sodium tetrahydroborate solution into the vial. Optimal conditions for headspace single-drop microextraction of SeH_2 are: [Pd], 30 mg/L; [$NaBH_4$] concentration,

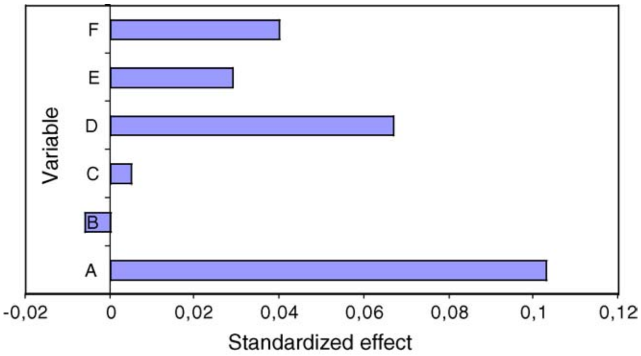

Fig. 2. Standardized effects corresponding to the optimisation of headspace single-drop microextraction of SeH_2 by the $2_{\mathrm{IV}}^{6\text{-}2}$ fractional factorial design (variables A–F as indicated in the text).

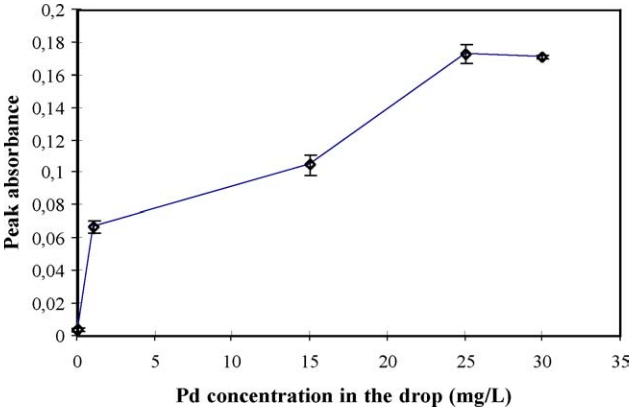

Fig. 3. Effect of the Pd(II) concentration used as trapping agent on Se absorbance (10 μg/L Se concentration).

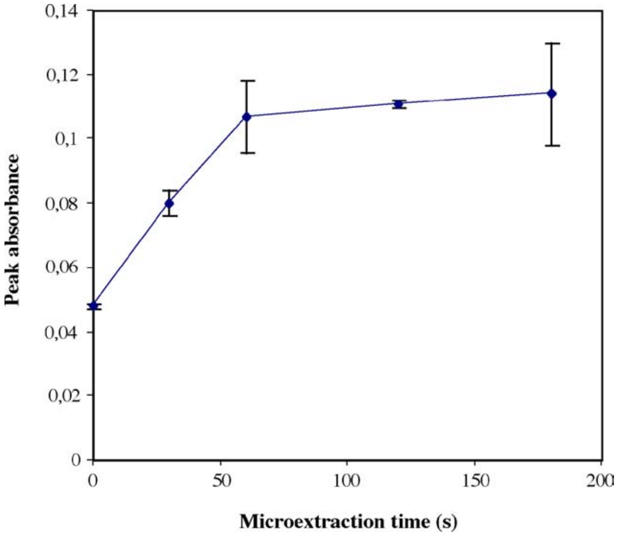

Fig. 4. Effect of the microextraction time on the Se absorbance (5 μg/L Se concentration).

1% (m/v); $NaBH_4$ volume (mL), 0.1 mL; extraction time, 90 s; sample volume in the vial, 20 mL; [HCl], 1.5 M.

3.2. Pre-reduction of Se(VI) to Se(IV) by UV irradiation

Only Se(IV) is precursor of the hydride, and therefore, a prereduction step of the Se(VI) present is mandatory for total inorganic Se determination. Prereduction of Se(VI) is usually performed by boiling with 5–6 mol/L HCl for 15–30 min under reflux. This thermal treatment has been shown to depend critically on the acid concentration, temperature and reaction time [14]. Other workers have employed microwave-assisted reduction of Se(VI) with HCl [15], HCl + HBr [16] and $KBrO_3$ + HBr [17].

Alternative treatments involving less stringent conditions (i.e. low temperature reaction, less concentrated acid) should be desirable.

UV irradiation has been applied to destroy organic matter and so remove interferences caused by dissolved organic matter when using electrochemical detection. Additionally, UV irradiation has been applied as a sample pre-treatment for determination of Se in natural waters using electrochemical detection so that Se(VI) can be reduced to Se(IV), the only electroactive Se species. A variety of conditions have been recommended for performing the determination of total inorganic Se after Se(VI) photoreduction. Thus, alkaline media are mostly recommended, e.g. pH 8 [18], pH 9 [19], pH 10 [20], pH 11 [21], pH 10.6 and degassing with nitrogen [19], but also acid media, e.g. 0.1 M HCl [22]. However, for some natural waters, precipitates may occur when working at a pH > 8, which could act as scavengers of Se(IV), and moreover, evidence exists in respect to non-reproducible photoreduction of Se(VI) in alkaline medium [23]. In this work, photoreduction was studied at different pH and time so that appropriate conditions can be chosen prior to preconcentration of Se by SDME. For this purpose, solutions containing 10 ng/mL Se(VI) were subjected to UV irradiation. Results are shown in Table 2. Conversions seem to be slightly faster in alkaline medium, hence, confirming the literature data. At pH 0.4 and lower (6 M HCl), both complete and reproducible conversions are achieved within 45 min. In alkaline medium, conversions are almost quantitative within 30 min. This time is significantly shorter as compared to those reported elsewhere for reduction of Se(VI) by UV irradiation [18,24]. Variations in the irradiation times observed in the literature for UV treatments of Se(VI) can be explained from the different characteristics of the photolysis unit employed, such as intensity of the lamp and locations of the tubes in respect to the lamp. The ability of the inner casing to reflect the UV radiation should also be considered. At pH 2, no conversion is observed for any treatment time. It has been pointed out that under UV irradiation Se(IV) can be oxidised to Se(VI) in 0.01 M HCl [25]. Experiments carried out with Se(IV) subjected to UV irradiation at pH 2 allowed us to confirm the oxidation of this species to Se(VI). Irradiation in acid medium under controlled conditions (i.e. at a HCl concentration well above 0.01 M) is preferable over alkaline medium so as to avoid the formation of precipitates. A 1.5 M HCl concentration was finally chosen for both photoreduction and single-drop microextraction so that application of the method is facilitated.

Table 2
Conversion percentages of Se(VI) into Se(IV) after UV irradiation at different pH

pH	UV irradiation time					
	5	15	30	60	120	180
0.4	33 ± 4	55 ± 1	70 ± 5	109 ± 14	97 ± 6	95 ± 7
2	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
5	63 ± 1	85 ± 1	77 ± 1	91 ± 10	98 ± 5	80 ± 4
8	59 ± 3	82 ± 4	90 ± 8	94 ± 10	96 ± 8	95 ± 7
12	55 ± 2	74 ± 2	89 ± 3	98 ± 5	110 ± 7	103 ± 6

LOQ: limit of quantification.

3.3. Effect of salts and organic matter on the Se(VI) photoreduction in acid medium

In order to apply the photoreduction of Se(VI) as a sample pre-treatment for determining total Se in natural waters, the effect of different salts has been assessed. Table 3 shows the percentage of recovery of Se(VI) and Se(IV) for different interferent-to-selenium ratios. Solutions containing Se(IV) were also UV-irradiated in order to assess the stability of this Se species in the presence of interferences. Chloride salts (NaCl, $MgCl_2$) did not display any effect even at a 10,000 mg/L concentration. However, a noticeable effect is seen to be caused by nitrate salts. Thus, when comparing the results of $MgCl_2$ and $Mg(NO_3)_2$, the depressive interference observed can be unambiguously attributed to nitrate.

Table 3
Effect of different salts on the photoreduction of Se(VI) into Se(IV) (recoveries (%) are shown)

Interferent	Se species[a]	Interferent/Se ratio			
		10^3	10^4	10^5	10^6
NaCl	Se(IV)	93 ± 14	91 ± 12	103 ± 9	104 ± 12
	Se(VI)	99 ± 14	99 ± 8	110 ± 8	96 ± 8
$CaCO_3$	Se(IV)	104 ± 13	111 ± 20	99 ± 7	90 ± 15
	Se(VI)	116 ± 8	97 ± 10	106 ± 7	91 ± 13
$Mg(NO_3)_2$	Se(IV)	91 ± 16	109 ± 9	90 ± 7	77 ± 11
	Se(VI)	92 ± 14	48 ± 5	10 ± 1	<LOQ[b]
$Fe(NO_3)_3$	Se(IV)	89 ± 16	34 ± 16	49 ± 10	20 ± 1
	Se(VI)	87 ± 14	35 ± 5	9 ± 1	<LOQ[b]
KI	Se(IV)	93 ± 14	95 ± 14	99 ± 9	<LOQ[b]
	Se(VI)	78 ± 19	38 ± 4	77 ± 11	<LOQ[b]
$MgCl_2$	Se(IV)	100 ± 17	95 ± 12	102 ± 11	101 ± 13
	Se(VI)	97 ± 16	99 ± 11	101 ± 12	106 ± 16
KNO_3	Se(IV)	121 ± 10	110 ± 10	110 ± 8	84 ± 12
	Se(VI)	112 ± 14	67 ± 3	20 ± 3	<LOQ[b]

[a] Se concentration: 10 ng/mL.
[b] LOQ: limit of quantification.

The mechanism for the photolytic reduction of Se(VI) has been shown to be similar to that of nitrate, being in both cases pH-dependent [21]. Consequently, the interference can be interpreted as a competitive reaction when samples containing both species are UV irradiated.

The tolerance of Fe(III) is at least up to a 10^3 Fe(III)/Se ratio. The effect of dissolved organic matter was also investigated. Organic matter could be oxidised by UV irradiation, hence, representing a competence in the conversion of Se(IV) into Se(IV). Oxalic acid, which is employed as model compound, did not interfere up to a 1000 mg/L concentration. However, 10,000 mg/L oxalic acid caused the complete suppression of the Se signal. This effect can be explained through volatilisation of Se compounds formed under UV irradiation in the presence of low molecular weight organic acids [26]. The effect of natural organic matter was also established by UV irradiation of Se solutions containing humic acid. No noticeable effect was observed up a 10 mg/L humic acid concentration.

In order to evaluate the applicability of UV irradiation for converting Se(VI) into Se(IV), determination of total Se and Se(IV) in two model waters (wastewater and seawater) was performed. Both model waters were spiked with 10 ng/mL Se(IV) + 10 ng/mL Se(VI). The composition of the model wastewater was: 100 mg/L NaCl + 300 mg/L $CaCO_3$ + 100 mg/L O_2 chemical oxygen demand (from oxalate). The model seawater composition was: 3% (m/v) NaCl + 0.5% (m/v) $MgCl_2$ + 0.15% (m/v) $CaSO_4$. Recoveries for the model wastewater were: 102 ± 3 % for Se(IV) and 93 ± 3% for total Se; recoveries for the model seawater were 98 ± 3% for Se(IV) and 96 ± 4% for total Se. It is remarkable to observe the absence of matrix effects due to some salts, such as nitrate, sulphate and chloride present at their typical concentration in seawater. These salts were reported to influence strongly the signal shape, integrated absorbance and thermal stabilisation of Se in the furnace [8].

3.4. Analytical characteristics

The method was characterised by establishing the relevant analytical parameters. The equation for the calibration curve of Se(IV) in 1.5 M HCl was: Abs = 0.0273 [Se] + 0.0176, where the Se concentration was expressed in ng/mL; the regression coefficient was $r = 0.995$. The calibration curve was linear up to 40 ng/mL. The limit of detection (LOD) (3 s criterion) was 0.15 ng/mL, whereas the limit of quantification (LOQ) (10 s criterion) was 0.5 ng/mL. The repeatability expressed as relative standard deviation (R.S.D.) was about 2.5% for a 10 ng/mL Se solution. LOD of Se using the same instrument operated under optimal furnace conditions and without preconcentration by HS–SDME was 4 ng/mL for a 3 μL injection volume, which represents an improvement of ca. 25 times. Given the large stability of the SeH_2 generated, the LOD could be lowered further using repeated sampling of the headspace. The maximum sample volume that can be accommodated in the graphite platform should not be exceeded when successive drops are accumulated. Repeated sampling (four times) of the headspace originated from one only sample solution yielded a LOD about 40 pg/mL. From four to five successive samplings of the headspace, a significant decrease in absorbance was observed.

Table 4
Analytical results for the determination of Se(IV) and total Se in spiked/fortified waters

Sample	Se found (μg/L)[a]		Total Se (certified)
	Se(IV)	Total Se[b]	
Spiked freshwater	5.3 ± 0.04	9.7 ± 0.12	–
Spiked seawater	4.5 ± 0.35	8.7 ± 0.3	–
NWTM-27.2 (fortified water)	–	1.5 ± 0.1	1.6 ± 0.09

[a] Average value ± standard deviation ($N = 3$).
[b] After UV irradiation for 45 min.

3.5. Determination of Se(IV) and total inorganic Se in waters

Two waters (freshwater and seawater) were spiked with 5 ng/mL Se(IV) + 5 ng/mL Se(VI) and recoveries were estimated. A freshwater was collected from the 'Gándaras' lake and another from the 'Ancoradoura' beach (Pontevedra, Spain). Analytical results for both natural waters with and without UV irradiation and HS–SDME–ETAAS determination are shown in Table 4. Calibration with Se(IV) standards prepared in 1.5 M HCl was performed. Recoveries were in the range 87–106% ($N = 3$), thereby indicating a good performance of the method. The method was also validated against CRM NWTM-27.2 (fortified water). No significant differences were found for total Se when the experimental value was statistically compared with the certified one (*t*-test). It should be noted that the total Se content of this CRM was below the LOD of ETAAS following direct injection.

4. Conclusions

Headspace single-drop microextraction combined with electrothermal atomic absorption spectrometry allows to tackle the determination of Se in natural waters in a simple way. The LOD is significantly improved as compared with direct determination of Se by ETAAS and additionally, matrix effects caused by typical salts in saline waters, such as nitrate, sulphate and chloride are overcome. Since only Se is separated from the matrix and collected onto a Pd(II)-containing aqueous drop, a Zeeman-based atomic absorption spectrometer is not required, a system equipped with D_2-background correction being adequate. Photoassisted reduction of Se(VI) into Se(IV) could be advantageous over thermal reduction with boiling 6 M HCl, since this process requires neither high temperature nor HCl acid at high concentration.

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