

Analytical Possibilities of Total Reflection X-ray Spectrometry (TXRF) for Trace Selenium Determination in Soils

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Selenium content of soils is an important issue due to the narrow range between the nutritious requirement and toxic effects upon Se exposure. However, its determination is challenging due to low concentrations within complex matrices that hamper the analysis in most spectroscopic techniques. In this study, we explored the possibilities of several analytical approaches combined with total reflection X-ray (TXRF) spectrometry for soil Se determinations. The direct analysis of a solid suspension using 20 mg of fine ground material (<50 μm) has a relatively high Se limit of detection (LOD) of 1 mg/kg (worldwide Se average in soils = 0.4 mg/kg) and is therefore only suitable for seleniferous soils. Several fast and simple analytical strategies were developed to decrease matrix effects and improve the LOD for Se determination in soil digests. On one hand, the application of a liquid–liquid extraction procedure using ethyl ether and the introduction of a Cr absorbent in the instrument configuration were carried out to avoid the associated problems on TXRF analysis of soil extracts due to the high Fe concentrations (~ 700 mg/L). On the other hand, a dispersive liquid–liquid microextraction procedure (DLLME) before the TXRF analysis of the soil digest was also developed. The effects of various experimental parameters such as sample volume, effect of major elements present in the soil matrix (Fe), and Se concentration in the sample were investigated. The LOD using this analytical methodology (0.05 mg/kg of Se) was comparable to or lower than those obtained in previous works using other popular spectrometric techniques such as GFAAS, ICPMS, and AFS. The calculated Se concentration for JSAC-0411 ([Se] = 1.32 ± 0.27 mg/kg) using the combination of DLLME and TXRF ([Se] = 1.40 ± 0.23 mg/kg) was in agreement with the certified value.

Selenium (Se) determination in soils is a critical issue in geochemistry due to the active role of soils in the selenium

biogeochemical cycle and the narrow range between the nutritious requirement and toxic effects upon exposure.¹

However, Se determinations in soil samples are difficult because of the low concentrations (e.g., worldwide mean concentration of Se is ~ 0.4 mg/kg)² and the complexity of the matrix. Therefore, it was not until the past decade that analytical advances have allowed the detection of Se at low concentrations to be of real interest to environmental studies.

Common techniques for Se determination include inductively coupled plasma mass spectrometry (ICPMS), inductively coupled plasma atomic emission spectrometry (ICPAES), graphite furnace atomic absorption spectrometry (GFAAS), and atomic fluorescence spectroscopy (AFS). These types of instruments are basically designed for the analysis of liquid samples, and thus, soil samples have to be brought into solution by means of a wet digestion procedure before the spectroscopic analysis. In Table 1, the most relevant analytical procedures published in the past decade for Se determination in soil samples are summarized. The main drawback concerning the use of these techniques when dealing with Se determination in soil samples is the presence of severe interferences that significantly hamper the analysis. For instance, Ar-based polyatomic ions overlapping Se isotopes interfere with Se measurements by ICPMS. Moreover, in complex matrixes such as geological materials, additional spectral interferences might occur, and the use of a collision/reaction cell or high-resolution ICPMS instruments is mandatory to obtain accurate results.^{4,21} During the hydride generation step often used in ICP-OES, AAS, and AFS, the reaction between the reducing agent and Se is subject to acute interferences caused by the presence of transition metals and nitric acid (commonly used for soil digestions) leading to a significant signal suppression.²² In addition, hydride generation is only sensitive for Se(IV), and thus, a reduction step is necessary if total Se has to be determined. AAS can also be performed using a graphite furnace, but with this approach strong interference occurs during the atomization step

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Table 1. Analytical Procedures Published in the Last Decade for Se Determination in Soil Samples

technique	details ^a	sample treatment	LOD (ppm)	sample (g)	comments	ref	
ICP	ICPMS	octopole reaction cell (H ₂)	0.07	2	spectral interference	3	
		DRC (H ₂) + ETV	<0.03 ^b	0.4	(reaction cell needed);	4	
AAS	ICP-OES	slurry sample	0.02 ^b	0.2	high argon consumption	5	
		wet digestion + reduction	<0.2 ^c	0.5	interference with	6	
	HGAAS	flame	<0.2 ^c	0.5	hydride formation (HNO ₃ ,	7	
		graphite furnace	0.02		transition metals), only		
		flow injection	<0.14	1	Se(IV)	8	
		using solid reagents	0.53	1		9	
	GFAAS	quartz furnace	wet digestion	<0.05 ^c	0.5		10
		matrix modifier	wet digestion + thiol cotton precon.	0.02	0.25	interference in atomization step	11
		Mg(NO ₃) ₂ + Pd matrix modifier	slurry sample	<0.5 ^c	0.25	(background correction needed)	12
		Mg(NO ₃) ₂ + Ir matrix modifier	wet digestion				13
	Ni(NO ₃) ₂	slurry sample	<0.12			13	
AFS		wet digestion	0.005		interference with	14	
		wet digestion	0.015		hydride formation, only Se(IV)	15	
INAA	irradiated 7 h; cooling 10 days; measurements 1–10 h	solid sample	<2.8 ^c	0.1	long analysis time	16	
		irradiated 20 h; cooling 7 days; measurements 2070 s	0.5	0.1–0.2		17	
	irradiated 6 h; cooling 21 days; measurements 1800 s	<0.09 ^c	0.3		18		
XRF	field portable (EDXRF), miniature X-ray tube	solid sample	6		limited sensitivity	19	
		laboratory EDXRF	8			20	
	field portable (EDXRF), radioisotope source	solid sample	40				

^a DRC, dynamic reaction cell; ETV, electrothermal vaporization. ^b Procedural blank. ^c Lowest reported concentration.

and background correction and the use of chemical modifiers are needed. Preconcentration procedures can be included as an additional sample preparation step to solve the problems related to the low Se concentrations and complex matrix.¹¹

Another alternative to determine Se in soil samples is the use of solid-state techniques, such as instrumental neutron activation analysis (INAA) and X-ray fluorescence spectrometry (XRF), that entail less sample manipulation avoiding the risk of contamination and Se volatilization (see Table 1). Despite the high selectivity and sensitivity of INAA, the high costs, the need for a nuclear reactor for irradiation, and the rather long time of analysis imposed by the long waiting (cooling) periods for the decay of short-lived radioisotopes has restricted its use for Se analyses. On the other hand, XRF has been a popular technique for major elemental analysis in geological samples to avoid complicated acid-digestion procedures. In particular, the speed, accuracy, and versatility of XRF are the most important features among the many that have made it a very mature analytical tool in this field.²³ In addition, the possibility to perform in situ analysis with field-portable XRF equipment has become a common and standardized technique for on-site screening and fast turnaround analysis of contaminant elements in environmental samples.^{20,24} Nevertheless, the major shortcoming of conventional XRF has been the poor elemental sensitivity, which is mainly a consequence of high background noise levels, resulting from instrumental geometries and sample matrix effects.²⁵ A reduction of spectral background can be effectively achieved by using total reflection X-ray fluorescence (TXRF) geometry. In this configuration, the primary beam strikes the sample at a very small angle (~0.1°) and the solid-state energy-

dispersive detector is accommodated very close to the sample (~0.5 mm). Consequently, an improvement of power of detection is achieved compared with conventional XRF.²⁶

To perform analysis under total-reflection conditions, samples must be provided as thin films, depositing 5–50 μL of sample on a reflective carrier with a subsequent drying by applying heat or vacuum. Preparation of samples as thin layers excludes matrix effects, such as absorption and secondary excitation, and thus, the quantification in TXRF analysis can be done directly by the addition of an internal standard to the sample.

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Up to now, TXRF has been mostly applied for the determination of trace elements in water samples (drinking water, rainwater, streamwater).^{27,28} Suitable limits of detection (LOD) have also been achieved for the determination of Se in biological and fluid samples with and without recourse of preconcentration strategies.^{29–31}

In view of the significance of Se determination in geological samples and the complexity of determining this element using atomic spectroscopic techniques, the main goal of the present research was to test the possibilities of TXRF for trace selenium determination in soils as a possible analytical alternative. To our knowledge, relative few papers exist about the TXRF investigations of geological samples³² and none for Se determination in soils.

In this study, we compared direct analysis of soil suspensions (using different dispersant agents) with the analysis of the digested soil sample for total Se determination. In the latter case, several fast and simple chemical strategies were developed to decrease matrix effects and improve the LOD for Se determination. On one hand, the application of a liquid–liquid extraction procedure using ethyl ether and the introduction of a Cr absorbent in the instrument configuration were carried out to avoid the associated problems on TXRF analysis of soil extracts due to the high Fe concentrations (~700 mg/L). On the other hand, a dispersive liquid–liquid microextraction procedure (DLLME) was used to isolate Se from the soil matrix. The great advantage of this procedure compared with conventional liquid–liquid extraction procedures is that the equilibrium is reached quickly, due to the large surface area between extraction solvent (a few microliters) and aqueous sample (a few milliliters), so that the extraction is almost independent of time.³³ It is a simple, inexpensive, fast, and effective pretreatment technique that has been mostly applied

to the analysis of organics³⁴ and, recently, for the determination of trace metals in water or biological samples using GFAAS spectrometry.³⁵ Since the microanalytical capability of TXRF spectrometry is very suitable for this approach, we have tested the applicability of a DLLME procedure using ammonium pyrrolidinedithiocarbamate (APDC) as chelating agent, carbon tetrachloride as extraction solvent, and ethanol as dispersant solvent as sample preparation strategy for Se determination in soil samples. To our knowledge, it is the first time the DLLME procedure has been combined with TXRF analyses. For all used approaches, limits of detection and accuracy studies were carefully evaluated to test the real capability of the developed TXRF methodologies for the intended purpose.

EXPERIMENTAL SECTION

Reagents and Materials. Stock solutions of 1000 ± 0.5 mg/L (Spectroscan, TECKNOLAB A/S, Norway) of appropriate elements were used to prepare standard solutions and spiked samples. High purity water used for dilution of stock solutions and samples was obtained from a Milli-Q purification system (Millipore Corp., Bedford, MA). The commercial nonionic surfactant Triton X-114 (poly(ethylene glycol) *tert*-octylphenyl ether), ethyl ether (>99.9%), ethanol (absolute, >99.5%), carbon tetrachloride (>99.5%), and APDC (~99%) were purchased from Sigma-Aldrich (Spain). Concentrated hydrochloric acid (Trace Select) was obtained from Fluka, Germany. For sample microwave digestion, we used analytical grade suprapur quality nitric acid (67–69% Romil SpA, Se < 0.1 mg/kg) and hydrogen peroxide (30%, Merck SpA).

In this work, quartz glass discs with a diameter of 30 mm and a thickness of 3 ± 0.1 mm were used as sample holders for introducing the sample into the TXRF equipment. A chromium foil of 25 mm × 25 mm, 10 μm thickness, 99.9% purity, and a permanent support of polyester was tested as absorber between the sample and the detector to decrease the Fe signal entering the detector when analyzing soil extracts.

The certified reference material JSAC-0411 (“Volcanic ash soil”, [Se] = 1.32 ± 0.27 mg/kg, Japan Society of Analytical Chemistry, Shinagawa, Tokyo, Japan) was employed to test the LOD and accuracy of the developed TXRF methodologies. Although we focus our study on a volcanic soil, the major and trace elements composition are similar to those from the upper continental crust.³⁶ Therefore, the developed analytical methodologies can be extended to the analysis of other soil types.

Sample Treatment Procedures. *Solid Suspension of Soil Sample.* To prepare slurries containing 20, 50, and 100 mg of soil, samples were brought into polypropylene tubes, and 1 mL of the dispersant solution was added. It is important to remark that to obtain reproducible and quantitative results in the TXRF analysis of soil suspensions, finely ground soil material is necessary. In the present study, solid suspensions using high purity water or diluted solutions (1% and 10% in high purity water) of a commercial nonionic surfactant (Triton X-114) were tested as dispersing agents. Then, Ge was added to the slurry sample for internal

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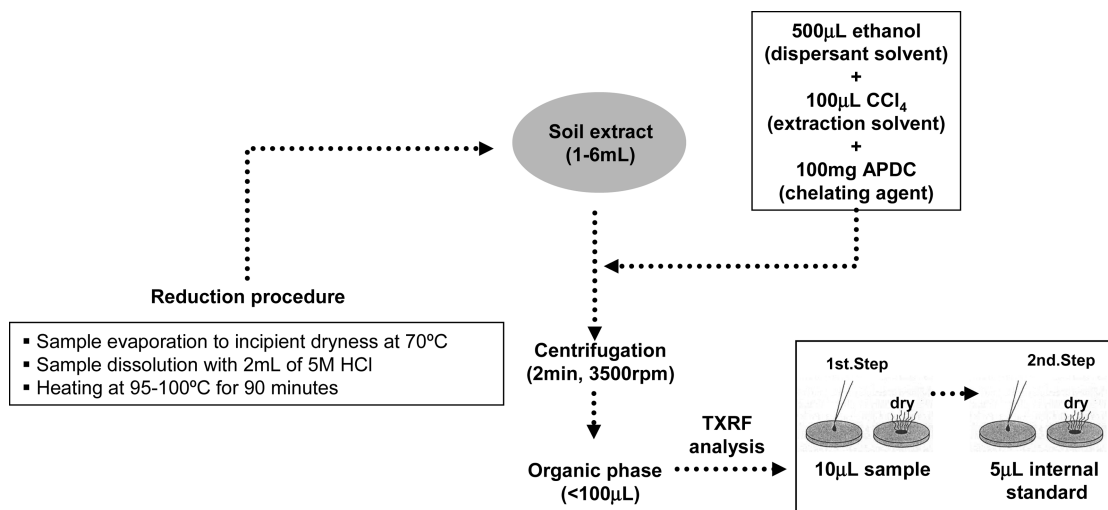


Figure 1. Scheme of the dispersive liquid–liquid microextraction procedure (DLLME) used for Se determination in soil digests by TXRF.

standardization (final Ge concentration 0.5 mg/L). The resulting solution was thoroughly homogenized (Vortex device) and an aliquot of 5 μL was transferred onto a quartz glass sample carrier and left to dry at room temperature under a laminar flow hood before TXRF analysis.

Soil Sample Digestion and TXRF Analysis. Microwave acidic digestion of soil samples was performed according to the EPA method 3051 (method 3051, US Environmental Protection Agency, 2008) with an acid mixture of 10 mL of HNO_3 and 2 mL of H_2O_2 . In order to take advantage of the microanalytical capability of TXRF, the above-mentioned microwave program was also adapted for mass-limited samples using 0.1 g of soil, 2.5 mL of HNO_3 , and 0.5 mL of H_2O_2 in quartz vessels and the same microwave program. ICPMS analysis following the protocol developed by Floor et al.⁴ showed that there were not statistical differences between the two methods.

For the direct TXRF analysis of soil extracts, Ge was used again for internal standardization and added to 0.25 mL of the soil digest (final Ge concentration of 0.5 mg/L). After homogenization, an aliquot of 5 μL was transferred onto a quartz glass sample carrier and dried as for solid suspensions before TXRF analysis.

Ether Extraction Procedure. To remove Fe from soil extracts, a liquid–liquid extraction procedure using ethyl ether was carried out.³⁷ For that, 0.5 mL of soil extract was acidified by addition of 100 μL of concentrated hydrochloric acid to form the corresponding Fe–chloro complex. Then, 0.5 mL of ethyl ether was added to the acidified extract, and the mixture was manually shaken for 2 min. Following the extraction, the mixture was allowed to stand for 10 min for separation of phases. Then, the upper organic layer (containing the Fe–chloro complexes) was discarded. This extraction procedure was carried out twice to improve the extraction efficiency.

Dispersive Liquid–Liquid Microextraction Procedure (DLLME). A dispersive liquid–liquid microextraction procedure (DLLME) using APDC as complexing agent was employed to separate and preconcentrate Se from soil extracts. Taking into account that the

developed DLLME procedure is only effective for Se(IV), a reduction step of the sample was necessary.¹ For that, soil digests were evaporated to incipient dryness at a temperature of 70 $^\circ\text{C}$ in Teflon beakers. Afterward, a 6 M hydrochloric acid solution was added, and the sample was heated to >90 $^\circ\text{C}$ for 90 min. After cooling, the sample was diluted with water to the initial volume (final matrix 1 M HCl).

To perform the DLLME procedure, 1–6 mL of the reduced soil extract was placed in a precleaned conical glass. Then, 0.5 mL of ethanol (dispersant solvent) was mixed with 0.1 mL of carbon tetrachloride (extraction solvent) and 100 mg of ammonium pyrrolidinedithiocarbamate, APDC (chelating agent) and injected rapidly into the sample solution. The mixture was then centrifuged at 3500 rpm for 1 min to achieve phase separation. After this step, 10 μL of the carbon tetrachloride sedimented at the bottom of the conical test tube, which contains the Se–APDC complex, was deposited onto a quartz glass sample carrier and left to dry at room temperature under a laminar flow hood. Finally, 5 μL of a 1000 ± 0.5 mg/L Y solution was added on the dried sample and left to dry at room temperature under a laminar flow hood before TXRF analysis. In Figure 1, a scheme of the DLLME procedure used is displayed.

Equipment and Instrumentation. A quadrupole-based ICPMS system (Agilent 7500c, Agilent Technologies, Tokyo, Japan) equipped with an octapole collision cell was used as reference technique for Se determination. Instrumental parameters used were published in a previous work.⁴

The analysis by TXRF was performed using a TXRF 8030C spectrometer (Atomika Instruments GmbH), equipped with a 3 kW X-ray tube with a Mo/W alloy anode and a double-W/C multilayer monochromator, adjusted to obtain an excitation energy of 17.4 keV (Mo $K\alpha$). In this equipment, the characteristic radiation emitted by the elements present in the sample is detected by a Si(Li) detector with an active area of 80 mm^2 with a resolution of 150 eV at 5.9 keV. The measurements were performed working at 50 kV, and the current was adjusted automatically as a trade-off between the detector dead time and total analysis time. A fixed acquisition time of 500 s was used.

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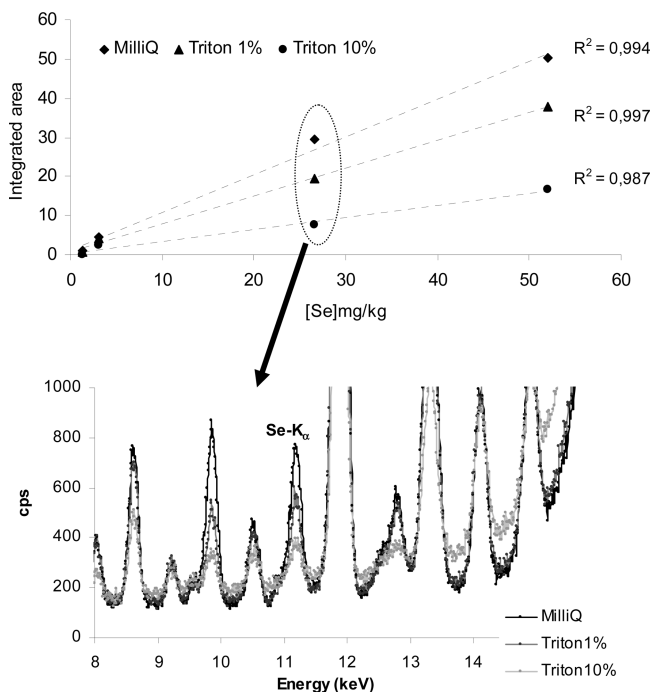


Figure 2. Relationship between Se signal and Se concentration in spiked soil suspensions using different suspension agents with a ratio of 20 mg of soil to 1 mL (measuring conditions: 5 μ L, 500 s).

RESULTS AND DISCUSSION

Analysis of Soil Suspensions. One of the most interesting features of TXRF in the analytical field is the possibility of quantitative direct analysis of solid samples without previous chemical treatment. In a first approximation, the direct solid analysis can be reached by means of an adequate suspension followed by internal standardization.³⁸ This analytical approach has been reported in a few studies only including the direct analysis of Al_2O_3 ³⁹ and boron nitride⁴⁰ powders.

In the present study, three dispersants (high-purity water and 1% Triton X-114, and 10% Triton X-114 in high-purity water) were tested for soil suspension preparation. As can be seen in Figure 2, the best sensitivity using a ratio 20 mg of soil to 1 mL of suspension agent was obtained by means of high-purity water. As reported in the literature, nonionic surfactants such as Triton X-114 could be used to adjust the viscosity of solutions and enhance the homogeneity of the analyzed samples.⁴¹ However, as is shown in the obtained results, an improvement of the results when using such reagent as a diluting agent did not occur. On the contrary, the background of the obtained TXRF spectra increased considerably when dilute solutions of Triton X-114 (1% and 10%) were used, and the sensitivity and linearity for Se determination decreased considerably. Therefore, the use of high-purity water as dispersant was considered appropriate for further experiments.

The influence of the slurry concentration on the background intensities and the signal-to-background ratios was also studied.³⁹

For that, 20, 50, and 100 mg of the JSAC-0411 certified reference material was mixed with 1 mL of water. Similar results were obtained when 20 or 50 mg of sample was deposited on the reflector. However, significantly lower signal-to-background ratios were found when 100 mg of sample was deposited on the quartz disk. This demonstrates that with 100 mg of sample, the condition of thin layer is broken. In view of the obtained results, a soil amount of 20 mg was established as optimum. The calculated LOD (3 times the square root of the background⁴²) for Se when using the best analytical conditions studied to prepare the slurry (20 mg of soil per 1 mL of high-purity water) was found to be around 1.0 mg/kg.

For quantification purposes, Ge was used as internal standard. Appreciable amounts of Ga (15.0 mg/kg) and Y (38.0 mg/kg) were found in the analysis of the reference soil sample, and therefore these elements, which are commonly used in TXRF analysis as internal standards, could not be employed. By direct analysis of the slurry of the JSAC-0411, it was not possible to quantitatively determine the Se content due to the proximity of the certified Se concentration ($\text{Se} = 1.32 \pm 0.27$ mg/kg) and the calculated Se LOD (~ 1.0 mg/kg Se). Nevertheless the direct measurement of soil slurries could be applied in order to get a first approximation of the concentration range for Se and element composition of the sample (multielemental information). This fast and relatively simple methodology can be successfully applied to analyze seleniferous soil samples with Se concentrations in the milligram per kilogram range. It is especially suitable for mass-limited samples since only 20 mg of sample is required. However, in all cases microhomogeneity of the soil powder has to be ensured in order to achieve reliable analytical results.

Analysis of Soil Digests. *Direct TXRF Analysis of Soil Digests.* In a former test, the analysis of a water-based sample with a low salinity and hardness (ARS 29 groundwater, indicative Se concentration 12 ± 3 $\mu\text{g/L}$)⁴³ was performed to evaluate the instrumental Se LOD using TXRF. It was found that the LOD was 0.4 $\mu\text{g/L}$ when 5 μL of sample, a measuring time of 500 s, and an excitation current of 47 mA were used.

However, the determination of Se at a few micrograms per liter level in soil digests was not possible due to the presence of high amounts of Fe (~ 700 mg/L) in the matrix that increased the dead time of the detection system. Consequently, a reduction of the excitation current of the X-ray tube was necessary to decrease the detector dead time with an associated loss of emission intensity and instrumental sensitivity. The calculated Se LOD in spiked soil digests using an excitation current of 12 mA (current decreased from 47 to 12 mA) was found to be 18.4 $\mu\text{g/L}$ corresponding to 0.76 mg/kg of Se in the soil.

For quantitation purposes, Ge was used as internal standard since, as for the solid suspensions, Ga and Y are present in the soil digest. It was not possible to quantify the Se content in the direct analysis of the CRM soil digest due to the proximity of the certified Se concentration and the calculated LOD for this element. However, in order to test the capability of the direct TXRF analysis, a fortified soil extract with 115 $\mu\text{g/L}$ of Se

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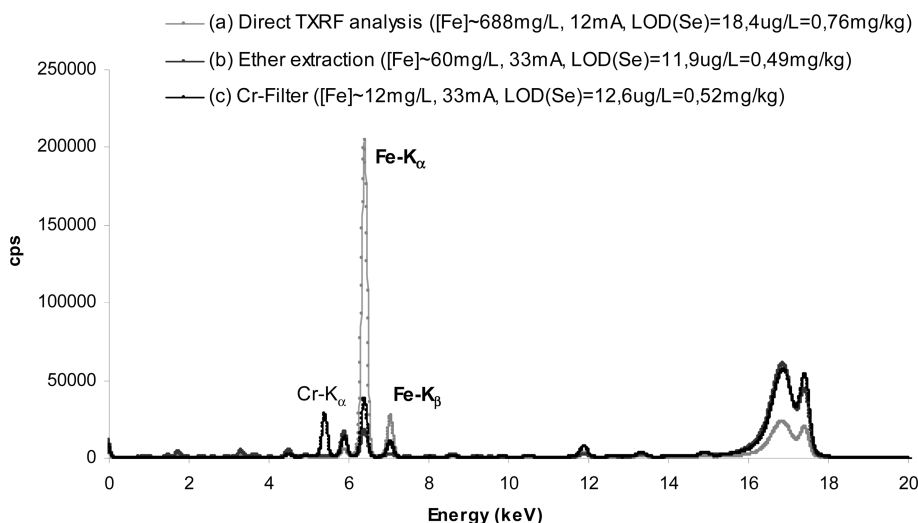


Figure 3. Comparison of TXRF spectra obtained for (a) the direct analysis of a soil extract, (b) after an ether liquid–liquid extraction procedure, and (c) with a Cr detector filter of 10 μm .

(corresponding to 4.6 mg/kg of Se in the soil sample) was analyzed, and a recovery of $105\% \pm 14\%$ for duplicate determinations was obtained. Therefore, direct analysis of soil digest can be used for screening of the multielemental composition of the soil digest or the quantification of Se in soils in the milligram per kilogram range.

A further enhancement of analytical quality of TXRF results can be achieved using more sophisticated sample treatments in order to reduce the Fe content in the soil digest or to isolate Se from the soil matrix. In the present study, both analytical strategies were studied to improve the LOD for Se in soil digests by TXRF spectrometry.

Reduction of the Fe Interference on Se Determination in Soil Digests. In order to reduce the Fe interference on Se determination when analyzing soil digests, two different approaches were tested. First, the extraction of Fe from soil digest solutions was carried out by liquid–liquid extraction using ethyl ether. In this case, it was necessary to treat the soil digest with concentrated hydrochloric acid to form the Fe–chloro complex, which was later extracted in the organic phase (ethyl ether).

Second, a physical approach introducing a suitable absorber between the sample and the detector was adopted to decrease the Fe signal entering the detector. Taking into account the energy of the emission lines of Fe ($K\alpha$ 6.403 keV; $K\beta$ 7.059 keV) and Se ($K\alpha$ 11.221 keV, $K\beta$ 12.501 keV), it was found that a Cr foil was the best option to be used as absorber since the absorption edge for this element (Cr K_{abs} 5.988 keV) is suitable to decrease considerably the Fe peak without a significant reduction of Se signal. With use of a chromium foil of 10 μm thickness, the Fe signal was reduced to 3.2%, whereas for Se the signal was reduced only to 49.7%, according to the exponential law of the attenuation photons in homogeneous matter.⁴²

In Figure 3, a comparison of the spectra obtained for the direct analysis of a soil extract after the application of the ether extraction procedure and with a Cr foil of 10 μm between the sample and the detector is displayed. As is shown, a significant reduction of the Fe peak (>90%) is achieved in both analytical approaches with a subsequent increase of the excitation current (33 mA) and an

improvement of the Se LOD ($\sim 12 \mu\text{g/L}$ corresponding to $\sim 0.5 \text{ mg/kg}$ of Se in the soil) compared with the direct analysis of the soil extract.

To evaluate the accuracy for Se determination, three replicate analyses of the JSAC-0411 soil were performed using both methodologies. In the case of the Cr absorber approach, Ge was used as internal standard for quantitation purposes. The determined Se concentration for three replicate measurements was $21.0 \pm 2.1 \mu\text{g/L}$ Se corresponding to $0.87 \pm 0.10 \text{ mg/kg}$ Se in the soil. As can be seen, significant differences were obtained between the calculated Se content and the reference value. Although the Se peak was detected, the Se concentration in the sample was below the quantification level. To check the reliability of the measurements, recovery tests on a fortified soil extract containing $115 \mu\text{g/L}$ of Se (corresponding to 4.6 mg/kg of Se in the soil sample) were carried out. A quantitative recovery was obtained for duplicate analysis of soil digests with a mean value of $97.4\% \pm 1.6\%$.

When the Se content in the soil extract was analyzed after the ether extraction, Ge could not be used as internal standard due to the formation of Ge volatile anionic chloro complexes.⁴⁴ In this case, a standard addition procedure was employed for quantification purposes using four Se additions in the range of 0–150 $\mu\text{g/L}$. The determined Se concentration in the JSAC-0411 soil digest following this analytical approach was $61.6 \pm 3.5 \mu\text{g/L}$ Se corresponding to $2.55 \pm 0.15 \text{ mg/kg}$ Se in the soil. This shows that also in this approach Se concentrations at these levels cannot be successfully quantified. Both the introduction of a Cr absorber between the sample and the detector and the ether extraction have similar improvement of the Se LOD for samples with high Fe contents. However, the Cr adsorber is the best strategy since no treatment of the soil digest before the TXRF analyses is needed.

Extraction of Se from the Soil Digest by DLLME. Taking into account the microanalytical capability of TXRF spectrometry, the combination with a DLLME procedure to extract the Se content from the soil matrix can significantly improve the analytical

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performance. We tested, for the first time, the applicability of a DLLME procedure combined with TXRF spectrometry (Figure 1). The DLLME procedure uses APDC (100 mg) as complexing agent and carbon tetrachloride (0.1 mL) as extraction solvent due to the high efficiency of carbamates to form metal complexes that are quantitatively extracted using chloride-based organic solvents.⁴⁵ Ethanol was selected as dispersant since it was miscible in both the extraction solvent and the soil digest. A volume of 0.5 mL of ethanol was used as the optimum volume in order to achieve a better and more stable cloudy solution without reducing the extraction efficiency, as has been demonstrated by Bidari et al using a similar DLLME system for Se determination in water samples.¹ In the particular case of analyzing soil extracts, it was not necessary to previously acidify the sample to form the Se-APDC complexes since the matrix of the soil digest after the reduction procedure was 1 M HCl.

An additional study was conducted to test the effect of sample volume on the Se extraction to the organic phase (100 μ L of carbon tetrachloride). Four aliquots (2, 4, 5, and 6 mL) of two standard solutions containing 10 and 30 μ g/L of Se were submitted to the DLLME procedure, and the amount of Se extracted in the organic phase was estimated from the difference of the initial Se concentration in the standard solution and the concentration measured after the sample preparation treatment by ICPMS. Higher percentages of Se extraction were obtained when 2 mL of sample (90–95%) was used compared with those obtained when 4–6 mL of sample (70–75%) was used for both Se concentration levels studied. Moreover, results achieved for the TXRF analysis of the sedimented organic phase showed that the Se signal-to-noise ratio improved with decreasing sample volume. Therefore, 2 mL of sample was used in the subsequent experiments. In Figure 4, a comparison of TXRF spectra obtained for the direct analysis of a soil digest and analysis after the DLLME procedure (2 mL of sample, 0.1 g of APDC, 0.1 mL of carbon tetrachloride, 0.5 mL of ethanol) is displayed. As is shown, a considerable reduction of Fe peak and a significant enhancement of Se peak were obtained after the DLLME procedure. The calculated LOD for Se using the described sample preparation procedure was 1.1 μ g/L of Se corresponding to 0.05 mg/kg of Se in the soil. Therefore, the Se LOD was improved more than one order of magnitude when the DLLME procedure was used to treat the soil extract before the TXRF analysis. Moreover, this LOD is one order of magnitude below the worldwide average of Se soil concentrations.

Germanium could not be used as internal standard in this approach due to the formation of Ge volatile anionic chloro complexes.⁴⁵ However, since several matrix elements including Y were removed from the soil digest sample in the DLLME treatment, Y was used as internal standard.

The effect of major elements present in the soil matrix on Se extraction was tested by investigating the extraction efficiency using Se standard solutions with and without the presence of Fe ([Fe] = 200 mg/L) in the range 25–100 μ g/L of Se. It was found that the extraction efficiency for Se (~75–79%) was independent of the initial metal concentration and the Fe content present in the initial solution. This demonstrates the feasibility of the use of the DLLME procedure for quantification purposes, since the metal

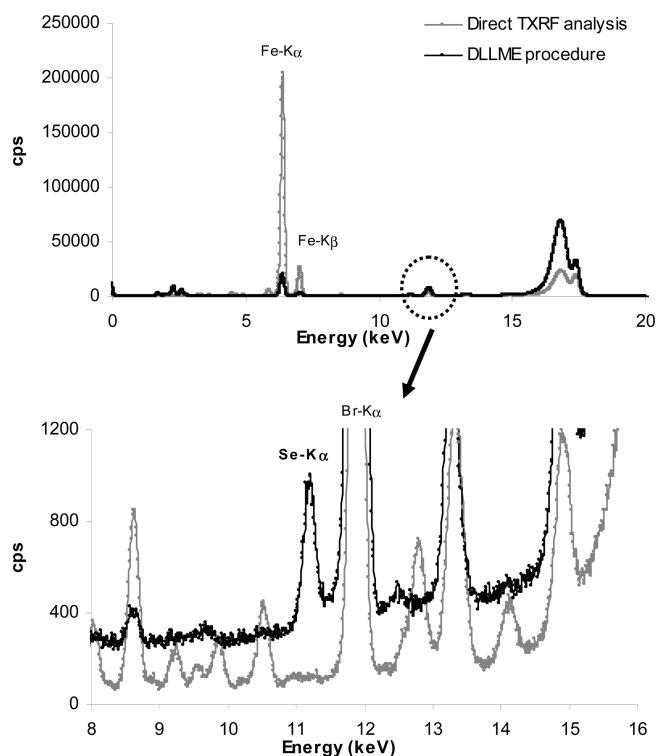


Figure 4. TXRF spectra obtained for the direct analysis of a soil extract and after the dispersive liquid–liquid microextraction procedure (experimental conditions: 2 mL of sample, 100 mg of APDC, 0.5 mL of ethanol, 0.1 mL of carbon tetrachloride).

extracted in the organic phase is related to the initial Se content in the soil digest. A similar trend was observed when the same experiment was performed on spiked soil extracts in the range 25–100 μ g/L of Se. However, in this case, despite the extraction efficiency for Se also being constant in the studied Se concentration range (53% \pm 7%), it was lower than that obtained for Se standard solutions. Therefore, for quantification purposes, it was not possible to use external calibration with aqueous Se standards, and a standard addition procedure was necessary. Moreover, to compensate for small differences in sample deposition on the reflector, the relative Se signal (Se signal/Y signal) was considered. Following this analytical approach four Se additions in the range of 0–100 μ g/L were performed, and Se determination in the CRM soil digest was 33.7 \pm 5.6 μ g/L Se corresponding to 1.40 \pm 0.23 mg/kg Se in the soil. The Se concentration determined was in very good agreement with the certified value (1.32 \pm 0.27 mg/kg Se), and no significant differences at the 95% confidence level were found. Thus, the accuracy of the developed DLLME procedure and its suitability for the determination of trace amounts of Se in soil digests was confirmed.

CONCLUSIONS

In this work, we have demonstrated the possibilities of several analytical approaches combined with TXRF spectrometry for Se determination in soils (see Table 2). An important part of the use of these methodologies is to understand and recognize their capabilities and limitations and to evaluate their suitability and complementarity depending on the objective of the analyses and the sample characteristics.

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Table 2. Limits of Detection and Accuracy of Results Obtained for Se Determination in Soils Using the Developed TXRF Analytical Methodologies

	limit of detection		accuracy ^a	
	[Se], $\mu\text{g/L}$	[Se], mg/kg	[Se], $\mu\text{g/L}$	[Se], mg/kg
(20 mg/1 mL Milli-Q water)		Solid Sample		
		0.91	<i>b</i>	<i>b</i>
		Digested Sample		
direct	18.40	0.76	<i>b</i>	<i>b</i>
Cr filter	12.61	0.52	21.0 ± 2.1	0.87 ± 0.09
ether extraction	11.99	0.49	61.6 ± 3.5	2.55 ± 0.15
DLLME	1.11	0.05	33.7 ± 5.6	1.40 ± 0.23

^a JSAC-0411 Volcanic soil ([Se] = 1.32 ± 0.27 mg/kg). ^b Not determined.

The use of direct analysis of soil suspensions by TXRF can be interesting as a fast and relatively simple methodology to get a first approximation of the multielemental composition of the soil. Moreover, it can be successfully applied to determine the Se content when an extremely small amount of sample is available (only 20 mg of soil is required) provided that the Se content is sufficiently high. A further enhancement of Se LOD can be achieved using more sophisticated sample treatment on the digested soil sample by the application of a liquid–liquid extraction procedure using ethyl ether or simply the introduction of a Cr absorber in the instrument configuration. With both approaches, the LOD for Se is reduced around 35% compared with the direct analysis of the soil digest. However, if Se content in soil samples is below the milligram per kilogram level, the applicability of a DLLME procedure to extract the Se content from the soil matrix and improve the LOD for Se by TXRF is a good alternative. By this simple and low-cost sample preparation strategy, accurate results can be obtained at the low milligram per kilogram range. The Se LOD is almost 10 times lower than the worldwide mean concentration of Se in soils, and it is also competitive with those determined using other popular spectrometric techniques such

as GFAAS, ICPMS, and AFS (see Table 1). This application could be also interesting to be extended to mass-limited samples with similar composition such as soil dust and volcanic ashes.

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