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Microwave-assisted digestion using diluted acid and base solutions for plant analysis by ICP OES

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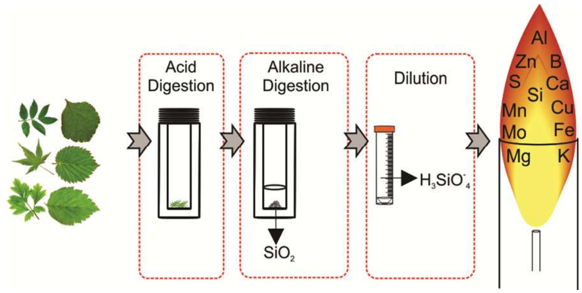
Graphical Abstract



Highlights

A reliable, safe and green microwave-assisted digestion procedure employing diluted solutions of HNO_3 and NaOH was developed for determining silicon and eleven nutrients in plants by ICP OES.

Abstract

A new sample preparation procedure for the determination of Si in plant materials by ICP OES is proposed. A two-step digestion procedure employing diluted solutions of HNO_3 (1.0 mol L^{-1}) and NaOH (1.5 mol L^{-1}) was applied for digesting plant samples. Limits of detection and quantification for Si were 56 and 186 μg g^{-1} respectively. A comparative study was done to evaluate the accuracy of the developed procedure by comparing the results obtained for Si in five sugar cane leaves samples with those obtained by micro energy dispersive X-ray fluorescence (μED-XRF) and according to a *t*-test the results agreed at a 95% confidence level. To verify the versatility of the procedure, Si, Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, and Zn were simultaneously determined in plant materials. Analytes were quantified in four certified reference materials: apple leaves (NIST 1515), tomato leaves (NIST 1573), white cabbage powder (BCR-679) and bush branches and leaves (NCS DC 73349) for accuracy assessment. All recoveries were in the range of 91.0-109% and all results agreed at a 95% confidence level (*t*-test) with certified concentrations. Foliar diagnosis was performed to demonstrate the applicability of the developed procedure for leaves of sugar cane, corn, soy and alfalfa. The proposed procedure is simple, versatile, reliable and safe for determination of Si, macro and micronutrients in plants by ICP OES.

Keywords: Foliar analysis; silicon; silicic acid; acid digestion; sample preparation; plant nutrients; alkaline digestion.

1. Introduction

Silicon is an element of utmost importance in numerous biochemical, geochemical, and surface processes. Plants that grow in silicate rich soils, can exceed up to 100-fold the usual concentration of phosphorus[1], therefore plants can absorb Si in the same amount in which macronutrients are absorbed[2,3]. Plants obtain Si in the form of monosilicic acid (H_4SiO_4), which is formed from the reaction between mineral silica (SiO_2) in the soil and water[1,4].

Some of the beneficial effects of using Si as an additive in plants include: increased mechanical strength and resistance to biotic and abiotic stress[3,5], such as exposure to ions formed by Al, Fe, and Mn[6], increased absorption of phosphorus and resistance to climatic conditions[7]. Silicon can be considered as a potent nutritional additive to plants, however its absence does not prevent its growth or natural development. There is a growing trend in analytical chemistry towards the determination of Si in plants and several investigations have been presented[8–11].

Several procedures are described in the literature for sample preparation of plant materials. The use of open vessel systems are commonly described[12–14], however, the use of those systems have some drawbacks, being prone to contamination and generally high volumes of concentrated acids are needed. On the other hand, there are numerous advantages reported in the literature regarding to the use of microwave-assisted procedures[10,11,15,16] in which closed vessels are used. The main advantage of microwave-assisted procedures is the possibility of working with low volumes of diluted acid solutions and consequently better control of the analytical blank solutions are achieved[16].

After microwave-assisted acid digestion of plant materials, precipitated silica (SiO_2) is found at the bottom of the digestion vessel the use of hydrofluoric acid (HF) during the sample preparation procedure for dissolving SiO_2 is usually reported[9,11,17,18]. The addition of HF implies on a second digestion step, in which boric acid (H_3BO_3) is added to the digestate for masking the remaining fluoride ions. This procedure has been applied for determining 30 elements in plant materials by ICP-MS, but results were not satisfactory for Si and it was impossible to check its accuracy because of the lack of CRMs with certified Si concentrations[11].

The solubility of SiO_2 does not change significantly at the pH range of 2-9, however, it increases abruptly at pH higher than 9 due to the formation of silicate ions[4,19–21]. Considering the behaviour of Si in alkaline medium, an alternative to the use of HF is alkaline solubilization of SiO_2.

Some authors[8,22] have described the use of a combination of concentrated NaOH solutions and H_2O_2 to digest plant materials. A two-step procedure, in which acid digestion of plant materials was followed by alkaline dissolution of Si and analyte determination via ICP OES was investigated[10]. However, it was also reported in the literature[8] difficulties in applying the procedure proposed by Haysom and Zofia[10].

Even though some of the above-mentioned procedures were effective on Si solubilization; the use of concentrated NaOH solutions difficult their implementation in routine analysis because of deterioration of quartz components of ICP OES, and, depending on the configuration of the torch, central tube tip blockage due to salt deposits may be a limitation[16]. The use of high concentrations of NaOH also could lead to contamination of the digests leading to inaccurate results for some important elements, such as Ca, Fe, K, Mg and Mn. Sodium is well-known as an easily ionisable element (EIE) and introducing large quantities of Na into an argon plasma could lead to suppression or enhancement of emission signals compromising the accuracy[23,24].

Thus, the main goal of the study here described was the development of a sample preparation procedure, which allows quantitative Si digestion and solubilisation in plant materials followed by determination using ICP OES. The capability of the developed procedure for multielement analysis was also evaluated.

2. Experimental

2.1. Equipment

Acid and alkaline decomposition were carried out in TFM™-PTFE digestion vessels (DAP-100®) by using a microwave oven (Speed Wave Four, Berghof Analytik, Chemnitz-Germany). All measurements were performed with an Agilent 5100 ICP OES with Dichroic Spectral Combiner (DSC) technology (Agilent Technologies, Mulgrave, Australia). The Synchronous Vertical Dual View (SVDV) mode was selected for data acquisition; in this mode, data from both axial and radial views are simultaneously obtained. Plasma operating conditions and parameters of the sample introduction system are shown in Table 1. The method accuracy was checked by comparing the results with those obtained by direct analysis using micro energy dispersive X-ray fluorescence (µED-XRF). A µEDX-1300 micro fluorescence spectrometer (Shimadzu, Kyoto, Japan) was used and the operational conditions are described in Table 1.

Table 1. Operational parameters adopted for Si determinations by ICP OES and µED-XRF in plant materials.

Method	Instrument parameter	Operational condition
ICP OES	RF applied power (kW)	1.5
	Argon auxiliary flow rate (L min^{-1})	1.0
	Argon plasma flow rate (L min^{-1})	12
	Argon nebulizer flow rate (L min^{-1})	0.60
	Nebulizer type	Seaspray®
	Nebulization chamber	Single-pass cyclonic
	Reading time (s)	20
	Replicates	3
	Sample uptake delay (s)	15
	Stabilization time (s)	15
µED-XRF	Acquisition time (s)	10
	Pass (µm)	100
	Number of spots	30
	Beam diameter (µm)	50
	Current (µA)	100
	Tension (kV)	50
	Detector	Si(Li) semiconductor
	Si line (keV)	Kα=1.74
	X-Ray	Rh tube
	Acquisition region (keV)	0-40

2.2. Reagents and reference solutions

All solutions were prepared with ultrapure water, (resistivity ≥18.2 MΩ cm - Millipore, Bedford, MA, USA), in polypropylene flasks and all flasks used was previously decontaminated in 10% v/v HNO_3 for 24 h and rinsed with deionized water before use. Sub-boiling nitric acid was obtained from a distillation apparatus (Milestone, Sorisole, Italy). Alkaline solutions were prepared by dissolving the proper amount of NaOH (PA-ACS ≥ 97%, Synth, Diadema, SP, Brazil) pellets with deionized water. Monoelement stock solutions of 1000 mg L^{-1} Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, Si and Zn (Fluka, Buchs St. Gallen, Switzerland) were diluted and used to prepare multielement analytical calibration solutions.

Calibration solutions for Ca, K, Mg, P, S and Si were prepared with analyte concentrations in the range of 1.0 to 75.0 mg L^{-1}. A volume of 375 µL of HNO_3 14 mol L^{-1} and 2.5 mL of NaOH 1.5 mol L^{-1} were added to each standard. Calibration solutions in the range of 1.0 to 3000 µg L^{-1} for Al, B, Cu, Fe, Mn, Mo and Zn were prepared and 750 µL aliquot of HNO_3 14 mol L^{-1} followed by 5 mL of NaOH 1.5 mol L^{-1} were added to each standard.

2.3. Certified reference materials and samples

The following certified reference materials (CRM) were used to evaluate the accuracy of the developed procedure: apple leaves NIST 1515, tomato leaves NIST 1573 (National Institute of Standards and Technology, Gaithersburg, MD, USA), white cabbage powder BCR-679 (Institute for Reference Materials and Measurements, Geel, Belgium), and bush branches and leaves NCS DC 73349 (National Analysis Center for Iron & Steel, Beijing, China).

Six different species of sugar cane leaves were provided by the Sugarcane Technology Center in Piracicaba (SP, Brazil). These samples have Si concentrations previously determined by LIBS[25] and μED-XRF. Samples of leaves and corn roots, alfalfa, and soy leaves were obtained from Embrapa Southeast Livestock in São Carlos (SP, Brazil). For each sample, forty leaves were collected. Each leaf had the central nervure removed and discarded. The leaves were washed with deionized water and dried at 65 °C for 72 h in a forced air oven. Samples were ground in a cutting mill fitted with a 20-mesh sieve.

2.4. Sample preparation

In Figure 1 a schematic representation of the proposed sample preparation procedure is shown. The digestion procedure was carried out in two steps. Firstly, sample masses of 100 mg were microwave-assisted digested using 5 mL of HNO_3 1 mol L^{-1} plus 5 mL of H_2O_2 (30% v/v). The heating program was performed in six steps: (1) 5 min to reach 120 °C; (2) 5 min at 120 °C; (3) 5 min to reach 160 °C; (4) 5 min at 160 °C; (5) 3 min to reach 230 °C; and (6) 5 min at 230 °C. The vessels were then removed from the microwave rotor and cooled down at room temperature. After cooling, vessels were open and a volume of 5 mL of NaOH 1.5 mol L^{-1} was added to each vessel. Vessels were then closed and a second heating program was applied in four steps: (1) 5 min to reach 150 °C; (2) 5 min at 150 °C; (3) 5 min to reach 230 °C; and (4) 10 min at 230 °C. A 1305 W applied power was used in both heating programs. After cooling down vessels were open and digests were quantitatively transferred to 50 mL polyethylene tubes and 750 μL of HNO_3 14 mol L^{-1} was added. Final volumes were made up to 50.0 mL. Aluminium, B, Cu, Fe, Mn, Mo and Zn were determined in these solutions. A second set of solutions were prepared with a further 2-fold dilution. Calcium, K, Mg, S and Si were determined in these more diluted solutions.

For the µED-XRF analysis, 0.5 g of cryogenically ground sugar cane leaves samples were pelleted applying 8 t cm^{-2} of pressure for 5 min. Pellets of 15 mm of diameter by 2 mm of thickness were obtained.

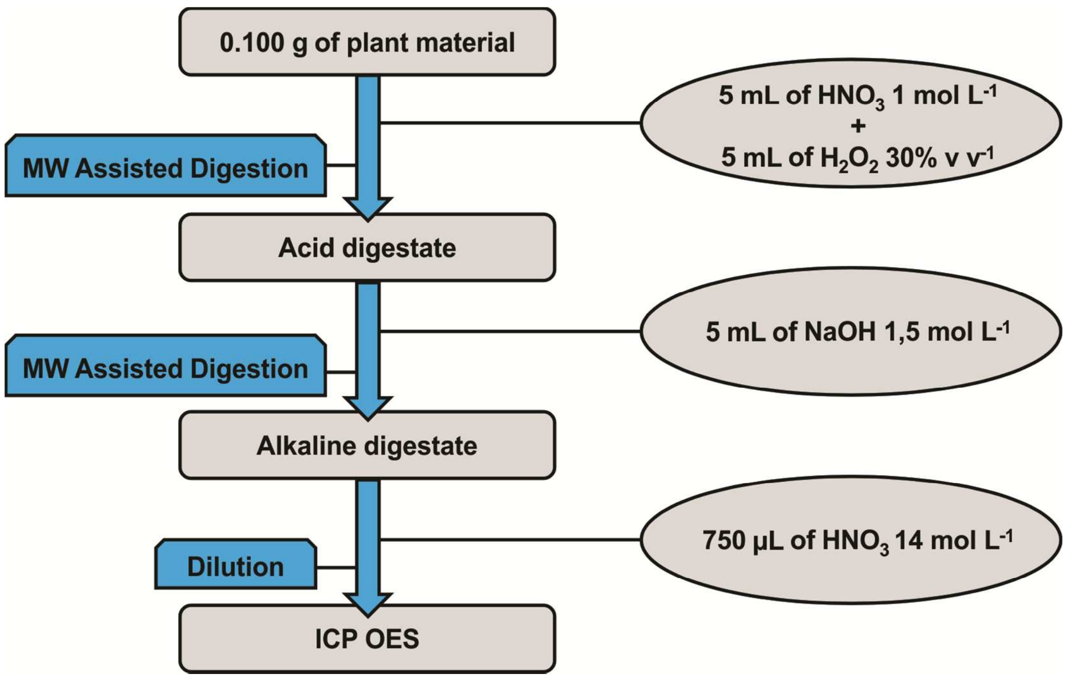


Figure 1. General scheme of the developed sample preparation procedure for Si determination in plant materials.

3. Results and discussion

3.1. Optimizing sample preparation procedures

Considering all negative effects caused by introduction of high concentrations of Na into the plasma, six experiments were performed in order to optimize the sample preparation procedure aiming at maximum efficiency on SiO_2 solubilization by using low concentrations of HNO_3 and NaOH solutions. Another important aspect evaluated was the need of conducting the procedure in two steps, being the first step an acid decomposition and the second one an alkaline dissolution.

Former procedures described in the literature usually are performed in one-step with highly concentrated NaOH solutions and H_2O_2 for sample decomposition[22,26], however we have experienced problems with these procedures. In more than one occasion, uncontrolled exothermic reactions occurred leading to melting of the microwave vessel walls. We also have tried a single step digestion with low concentration of NaOH, *i.e.* 2.0 mol L^{-1}, in this case the digestion was ineffective for digesting organic compounds.

One sugar cane leaves sample with 9.77±0.07 mg g^{-1} Si was selected and used in the optimization experiments. Experiments 1, 2 and 3 were performed using a solution of 2 mol L^{-1} HNO_3 in the first digestion step and solutions containing 7.5; 3.75 and 2.5 mol L^{-1} NaOH in the second step. Experiments 4, 5 and 6 were done using 1 mol L^{-1} HNO_3 in the first digestion step and 2.0; 1.5 and 1.0 mol L^{-1} NaOH solutions in the second step. In each experiment, samples were digested in triplicate. Table 2 contains Si concentrations obtained in experiments 1 – 6 and the experimental conditions applied in each experiment. Silicon concentrations in experiments 1 – 5 were in the range of 9.67 – 10.0 mg g^{-1} and the RSD were in the range of 0.02 – 0.16%. The conditions used in experiment 6 led to a lower concentration for Si (8.05 mg g^{-1}). This is related to the low concentration of NaOH in the final digests when compared to the high concentration of Si in the sample; however, it seems possible to further reduce NaOH concentration when working with plant materials containing low concentrations of Si. Since there were no differences (*t*-test; 95% confidence level) among the results obtained in experiments 1 to 5, we adopted conditions used in experiment 5 because it requires the lowest concentration of NaOH to dissolve SiO_2 and led to better precision. A comparison among the procedures described in the literature regarding to the determination of Si in plant matrices and the procedure here proposed is shown in Table 3.

Table 2. Determination of Si in a sugar cane leaves samples applying six different strategies during the optimization of the sample preparation procedure. Concentration values represented as (mean ± confidence interval, n = 3 and *t* = 0.05).

Experiment	Reagents volume and concentrations	Si concentration (mg g^{-1})	Determined concentration (mg g^{-1})
1	5 mL of HNO_3 2 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 7.5 mol L^{-1}		9.90 ± 0.20
2	5 mL of HNO_3 2 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 3.25 mol L^{-1}		9.79 ± 0.14
3	5 mL of HNO_3 2 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 2.5 mol L^{-1}		10.0 ± 0.18
4	5 mL of HNO_3 1 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 2.0 mol L^{-1}	9.77 ± 0.07[a]	9.87 ± 0.41
5	5 mL of HNO_3 1 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 1.5 mol L^{-1}		9.67 ± 0.10
6	5 mL of HNO_3 1 mol L^{-1}; 5 mL of H_2O_2 30% v v^{-1}; 5 mL of NaOH 1.0 mol L^{-1}		8.05 ± 0.16

[a]Obtained by µED-XRF

Table 3. General overview of procedures described in the literature for Si determination in plant materials and the proposed procedure.

Sample	Procedure	Determination method	Reference
NIST SRM 1515; NIST SRM 1575	Microwave-assisted digestion of 0.5 g of sample with 5.0 mL of HNO_3 14 mol L^{-1} and 0.1 mL of HF	ICP-MS	11
Rice straw samples	Autoclave-induced digestion of 0.1 g with 2 mL of H_2O_2 and 4.5 mL of NaOH 12.5 mol L^{-1}	UV-Vis spectrophotometry	22
Rice straw and sugar cane leaves	A mass of 0.1 g of plant material was oven-induced digested with 2 mL of H_2O_2, 4 mL of NaOH 12.5 mol L^{-1} and 5 drops of octyl-alcohol. After digestion, 1 mL of NH_4F $5x10^{-3}$ mol L^{-1} was added to the digestates	UV-Vis spectrophotometry and ICP OES	8

Table 3 (*continued*)

Sample	Procedure	Determination method	Reference
Rice straw, sugar cane, mixed pasture	A mass of 0.2 g of plant material was microwave-assisted acid digested in two steps using 3 mL of HNO_3 14 mol L^{-1} and 2 mL of H_2O_2 in the first step, and alkaline digested with 10 mL of NaOH 2.5 mol L^{-1} in the second step	ICP OES	[10]
Sugar cane leaves, soy leaves, corn leaves and roots.	A mass of 100 mg of plant material were microwave-assisted acid digested with 5 mL of HNO_3 1 mol L^{-1} and 5 mL of H_2O_2 30% v v^{-1} in the first step and alkaline digested in the second step with 5 mL of NaOH 1.5 mol L^{-1}	ICP OES	This work

3.2. Analytical figures of merit

The performance of the proposed procedure was evaluated by analysing reference solutions of Si. Short time intervals in each analysis were achieved when the SVDV mode is adopted. In the so called SVDV mode, emission data from both radial and axial views are obtained simultaneously.

The calibration curve was linear in the range of 1 to 75 mg L^{-1} and a linear correlation coefficient of 0.9999 was attained. The correspondent linear equation to the calibration graph was I = 2408.40 C + 598.53 (where I is the intensity in counts per second and C is the concentration of Si in mg L^{-1}). Considering the background equivalent concentrations (BEC) and relative standard deviations (RSD) for 10 consecutive measurements of the blanks, a limit of detection (LOD) of 0.06 μg L^{-1} and a limit of quantification (LOQ) of 0.21 μg L^{-1} were established. Ten digestions blanks were prepared and considering the mass of sample and dilutions, a LOD of 56 μg kg^{-1} and a LOQ of 186 μg kg^{-1} were obtained.

3.3. Evaluation of multielement determination capability

Although the main goal of this work was the development of a simple and green procedure for determining Si in plants, foliar diagnosis cannot be restricted to one element. Considering its attractiveness for application in routine analysis laboratories, an analytical procedure must be versatile, allowing the analyst to determine important macro and micronutrients. Thus, we evaluated the applicability of the developed procedure for determining Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, and Zn in plants. Table 4 contains LODs and LOQs for these elements. It is noticeable that high limits of detection (258 µg g^{-1}) and quantification (861 µg g^{-1}) were obtained for K; this is caused by the high content of K in the NaOH reagent (≥ 0.3% m/m), however, K is a macronutrient and the normal concentration of K in healthy plants is in the range of 15,000-55,000 µg kg^{-1} [27].

Table 4. Emission lines, background equivalent concentration (BEC), limits of detection and quantification for Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, Si and Zn (standard deviation for 10 digestion blanks).

Element	Emission Line	BEC	LOD (µg g^{-1})	LOQ (µg g^{-1})
Al	237.312	77.8	9.5	31.8
B	249.678	8.15	1.6	5.2
Ca	422.673	0.14	137	456
Cu	223.009	4.10	0.8	2.6
Fe	259.940	23.0	3.8	12.7
K	404.721	0.49	2585	8615
Mg	285.213	0.01	3.5	12
Mn	293.305	1.71	1.5	5.1
Mo	281.615	2.70	0.3	1.0
S	181.972	3.14	164	547
Si	250.690	0.06	56	186
Zn	213.857	15.0	3.0	9.9

3.4. Comparative study: silicon determination in sugar cane samples

A comparative study was done to evaluate the accuracy of the developed sample preparation procedure. Five sugar cane samples (SCL 1–5) had Si concentrations determined by µED-XRF and by ICP OES using the proposed sample preparation procedure. The linear regression method was used to associate the results obtained by both methods, and a graphical representation as well as the obtained equation are shown in

Figure 2. The calculated correlation coefficient of 0.9934 evidenced good correlation among the results when comparing ICP OES with μED-XRF (Fig. 2). The confidence intervals at a 95% confidence level for the slope and linear coefficient for the equation were calculated as being 1.35 ± 0.30 and -1.90 ± 1.96, respectively, showing that systematic errors did not occur when comparing both methods. According to a *t*-test there were no significant differences among the results at a 95% confidence level. The coefficient variation (CV) obtained for the proposed procedure (n = 3 digests) ranged from 0.2 to 1.80%, while the CV obtained with μED-XRF (n = 30 spots, 100 μm pass) ranged from 0.7 to 1.80%.

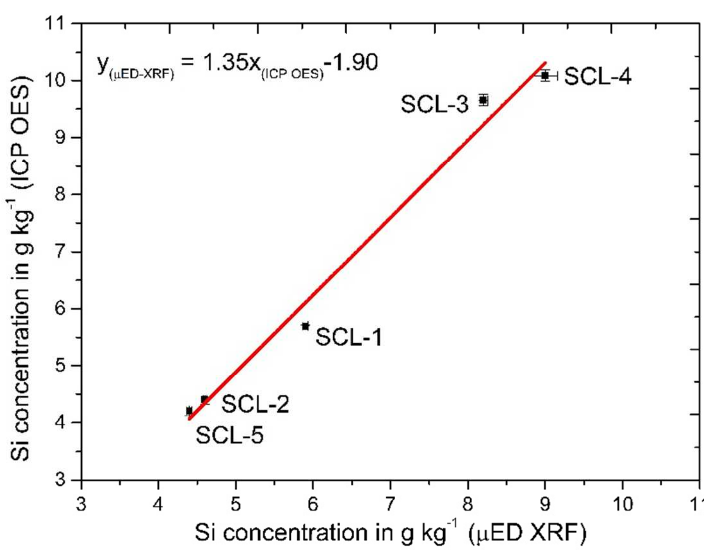


Figure 2. Correlation among Si concentrations in five sugar cane leaves (SCL 1-5) determined by ICP OES using the developed digestion procedure and μED-XRF.

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3.5. Determination of Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, Si and Zn in plant materials

Silicon was determined in three CRMs, *i.e.* NIST 1515, NCS DC 73349 and NIST 1573a, and a wide range of concentrations were found (400 - 6000 µg g^{-1}), however it was not detected in the BCR 679. Only the NCS DC 73349 CRM had a certified concentration of Si, and the recovery for this analyte was 95.2%, also the standard deviation obtained with the proposed procedure was lower than the one certified. According to the certificate of analysis; the reference value was obtained by colorimetry (molybdenum blue method), gravimetry and X-ray fluorescence spectrometry.

To verify the accuracy of the procedure for determining Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, and Zn in plants, these analytes were also determined in the above mentioned CRMs and results are shown in Table 5. Analyte recoveries were in the range 91.0 to 109 % for all CRMs. There is no statistical difference among certified concentrations and those ones obtained applying the proposed procedure (*t*-test; 95% confidence level).

The procedure here proposed was applied to the determination of several analytes in leaves of corn, sugar cane, soy, alfalfa, and corn roots (Table 6). It is important to mention that these samples were cultivated in a dark-red latosoil with a high content of organic matter. Soils had pH corrected with limestone. Samples were obtained from farms located in São Carlos (SP, Brazil) and Piracicaba (SP, Brazil); both cities are within a distance of 105 km apart so the regional setting in both of these locations is similar. The weather conditions from both cities are characterized as tropical of altitude, since both cities are located at approximately 550-900 m above sea level.

Table 5. Determination of Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, P, S, Si and Zn and analyte recoveries in NIST 1515, NIST 1573a, NCS DC 73349 and BCR 679 certified reference materials by ICP OES after a two-step microwave-assisted digestion procedure. Results for NCS DC 73349 represented as mean ± standard deviation, n = 3. Results for NIST 1515, NIST 1573a and BCR 679 represented as mean ± confidence interval, n = 3 and *t* = 0.05.

Analyte	NIST 1515			NCS DC 73349		
	Found (µg g^{-1})	Certified (µg g^{-1})	Recovery (%)	Found (µg g^{-1})	Certified (µg g^{-1})	Recovery (%)
Al	330±4.0	286±9	115	1999±80	2000±300	99
B	29.3±0.3	27±2	108	41±1	38±6	108
Ca	14411±106	15260±150	94.4	15594±89	16800±1100	92.6
Cu	5.80±0.2	5.64±0.24	103	6.57±0.1	6.6±0.08	99.6
Fe	83±7	83±5	100	1036±27	1070±57	96.8
K	15108±927	16100±200	94	9934±185	9200±1000	108
Mg	2854±6.0	2710±80	102	4839±219	4800±400	101
Mn	53±4	54±3	98	66±1.7	61±5	109
Mo	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
S	1786±51	1800*	99.2	6877±50	7300±600	94.2
Si	400±0.04	NA	---	5610±5.61	6000±700	95.2
Zn	12.5±0.6	12.5±0.3	100	56.6±0.99	55±4	103

Table 5 *(continued)*

Analyte	NIST 1573a			BCR 679		
	Found (μg g^{-1})	Certified (μg g^{-1})	Recovery (%)	Found (μg g^{-1})	Certified (μg g^{-1})	Recovery (%)
Al	629±37	598±12	105	125	NA	---
B	32.8±1.6	33.3±0.7	98.6	30.2±2.0	27.7±1.9*	109
Ca	49960±302	50500±900	99	8068±177	7768±655*	104
Cu	4.61±0.30	4.70±0.14	98.0	3.00±0.1	2.89±0.1	105
Fe	332±5	368±7	90.2	58.3±0.4	55±2.5	106
K	29273±785	27000±500	108	36421±690	NA	---
Mg	12043±1173	12000*	104	1399±53	1362±127*	103
Mn	230±7	246±8	93.7	14.4±0.02	13.3±0.5	109
Mo	<LOD	<LOD	<LOD	14.8±2.4	14.8±0.5	100
S	9195±376	9600*	95.8	7.2±0.1	NA	---
Si	1800±0.2	NA	---	<LOD	NA	<LOD
Zn	28.1±0.6	30.9±0.7	91.0	80.6±0.4	79.7±2.7	101

*Indicative values

Table 6. Determination of Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, Si and Zn in plant samples (mean ± standard error of the mean, n = 3).

Sample	Analyte concentration in µg g^{-1}					
	Al	B	Ca	Cu	Fe	K
Sugarcane leaves (1)	323±0.3	5.74±0.1	5252±186	4.42±0.1	225±4.6	13072±165
Sugarcane leaves (2)	236±0.1	2.78±0.1	2178±31	3.72±0.1	129±1.5	13634±199
Sugarcane leaves (3)	206±0.01	3.75±0.2	5890±40	3.54±0.1	153±2.4	11452±125
Sugarcane leaves (4)	300±0.02	3.04±0.1	4286±21	4.01±0.1	223±4	9350±27
Sugarcane leaves (5)	113±0.01	2.34±0.1	2640±104	4.95±0.2	99.4±5.4	13410±2003
Sugarcane leaves (6)	194±0.01	3.61±0.04	3220±33	4.35±0.05	112±1.8	9780±130
Corn roots	48±0.01	16.0±0.3	4961±64	3.28±0.1	16±0.3	39177±1247
Corn leaves	63±0.08	46.3±1.2	3021±13	2.95±0.1	46±1.2	30881±2996
Soy leaves	300±12	29.4±0.03	12838±128	11.2±0.1	277±12	22581±391
Alfalfa leaves	1183±58	39.7±0.3	10858±43	12.2±0.3	458±13	30092±1685

Sample	Analyte concentration in µg g^{-1}					
	Mg	Mn	Mo	S	Si	Zn
Sugarcane leaves (1)	1810±42	47.5±1.4	8.70±4.2	2033±4.3	5701±12	17.3±0.3
Sugarcane leaves (2)	907±13	76±1.6	2.48±1.1	1314±13	4404±46	14.1±0.2
Sugarcane leaves (3)	2825±30	36.7±0.6	1.17±0.3	2257±38	9654±58	19.9±0.2
Sugarcane leaves (4)	2762±8	50.2±0.2	5.36±0.7	1720±10	10083±63	19.3±0.1
Sugarcane leaves (5)	1514±61	53.6±2.4	2.62±0.8	1604±6	4206±64	16.8±0.3
Sugarcane leaves (6)	1350±10	70.3±2.5	3.98±1.7	1600±6.6	56.3±4.7	16.9±0.2
Corn roots	2467±45	28.8±8.5	3.85±0.6	2802±141	444±9.0	12.5±0.7
Corn leaves	1420±20	46.7±0.7	7.59±2.0	1478±33	450±6.0	16.2±2.2
Soy leaves	3199±59	53.6±0.5	1.88±0.2	2460±22	5725±99	34.5±0.1
Alfalfa leaves	2770±96	36.2±0.1	5.18±2.0	2245±61	465±39	32.8±0.6

4. Conclusions

A new procedure for determining Si in plants using microwave-assisted two-step digestion and ICP OES was proposed. Best results were obtained when HNO_3 and NaOH solutions contained as low as 1.0 and 1.5 mol L^{-1} concentrations, respectively.

Considering the figures of merit, the use of diluted solutions of NaOH for sample preparation is a feasible alternative since it minimizes contaminations from NaOH reagent, allowing the determination of other important macro and micronutrients.

A comparison among the results obtained with µED-XRF and the proposed procedure demonstrated that there is a high correlation among the Si concentrations obtained by both methods. The procedure was successfully applied to the simultaneous determination of Si, Al, B, Ca, Cu, Fe, K, Mg, Mn, Mo, S, and Zn in four CRMs and in a variety of ten different plant samples. Analyte recoveries and RSDs were in acceptable ranges. Low standard errors were obtained when the above-mentioned analytes were determined in all samples.

The developed procedure is simple, safe and reliable. It shows significant improvements when compared to the procedures described in the literature without adding any amount of HF.

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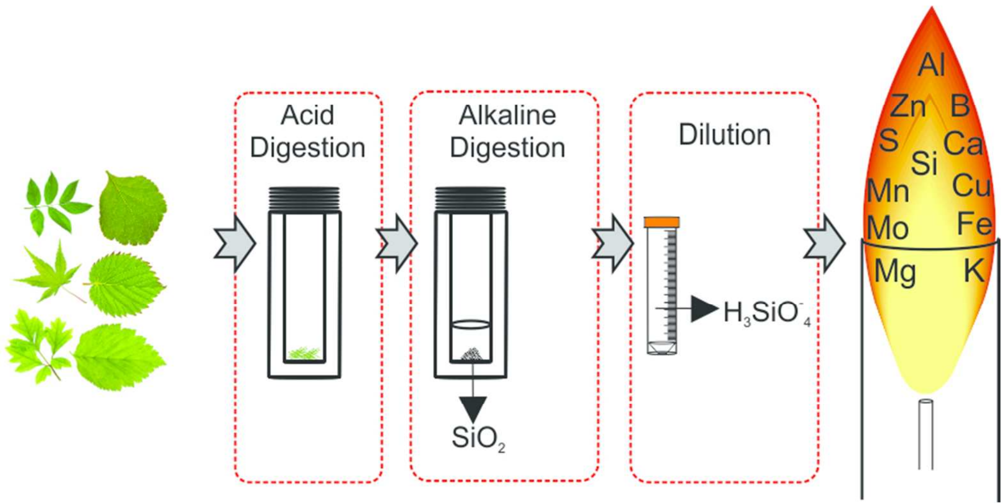

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