Trace elements – Their distribution and effects in the environment
B. Markert and K. Friese, editors

Chapter 6

Nuclear analytical techniques in atmospheric trace element studies in Portugal

M.C. Freitas, M.A. Reis, L.C. Alves, H.Th. Wolterbeek

Abstract

In this work, the nuclear techniques INAA and PIXE were used in a lichen survey using Parmelia sulcata Taylor held in Portugal in 1993. The possibilities of the two techniques are discussed, and the elements determined by both are compared. It is presented the concentration patterns of 46 chemical elements and the assignment to pollution sources attempted and compared with results on factor analysis previously published. Results are also compared with other European surveys.

Keywords: Nuclear analytical techniques; Atmospheric trace elements; Instrumental neutron activation analysis; Proton induced X-ray emission; Biomonitoring

1. Introduction

Nuclear analytical techniques (NATs) and related isotopic tracer methods are well established as important tools in a wide variety of different kinds of environmental studies. They provide a wealth of information on sources, pathways and effects of many elements of environmental and health-related interest. Apart from being regarded as of particular strength in analytical quality assurance (IAEA, 1997), nuclear and related techniques cover studies of air particulates, solid waste products, sediments, food, water, human tissues, biomonitors and other kinds of environmental samples.

Key features of most nuclear techniques are the principal absence of effects by element chemical forms, and the absence of necessary sample digestion procedures (De Goeij and Bode, 1997). Apart from being an analytical advantage, the first property should be regarded, as a drawback where information on a particular form of an element is required (viz. methylmercury rather than total mercury), the second only applies for nuclear signals of high penetrative power. However, the intrinsic accuracy of most NATs makes that they are in regular use in validation procedures of analytical methods, the development of new reference materials, and the set-up of base-line elemental information in a variety of (health-related) environmental issues.

Typical examples of the NAT family are neutron activation analysis (NAA) and proton induced X-ray emission (PIXE). NAA is one of the most robust analytical methods available in a vast range of applied sciences (Trahey, 1996; Bode and De

Goeij, 1998), hardly asks for any extensive sample preparation, but shows up to 2–4 weeks analytical turn-around time, depending on the half-lives of the involved radionuclides. PIXE is a fast analytical method, but asks for a strict sample homogeneity down to the sub-mg level, which implies laborious sample preparation procedures (IAEA/AL/095, 1996; Tadic et al., 1997). The methods are intrinsically accurate, multi-elemental and partly complement each other with respect to elements of environmental interest.

Both instrumental NAA (INAA) and PIXE are available at the Instituto Tecnológico e Nuclear (ITN), Sacavém, Portugal, and have been applied in a national study on atmospheric trace elements, using lichens as biomonitor materials. In the present paper, this survey is used to illustrate the potential of NATs in environmental studies.

2. Multi-elemental techniques versus single element techniques

The selection of elements to be analyzed and/or used in data interpretation should be closely linked to the objectives of the study (Wolterbeek and Bode, 1995). A survey may be dedicated to a single or only a limited number of elements, but it may also be set up to gather information about sources/effects based on broader points of view. The latter set-up may be regarded as most effective, because, first, the analysis of a large number of elements may generally increase the modes for interpretation, may permit a more reliable recognition of source finger-prints, and may show effects which are not a-priori anticipated (Bode and Wolterbeek, 1990; Wolterbeek et al., 1996), while, second, the resources needed for the field-work will not or hardly depend on the number of elements of interest. This indicates that the selection of a large number of elements *principally* emerges from many survey's goals: clear and unequivocal interpretation of data on specific elements may largely depend on the simultaneous presence of data on various other elements.

The above can be illustrated by data taken from several multi-element air pollution biomonitor surveys carried out at IRI: The air pollution surveys commonly include a number of soil-associated elements (e.g. Al, Fe, Sc, Cr, Th) and several rare earth elements (Kuik et al., 1993a,b). In the Factor Analysis interpretation of the data on all selected 20 elements, the soil indicator elements serve to extract a "soil-factor" (De Bruin and Wolterbeek, 1984), based on which, for *all* individual elements, site-specific soil-associated fractions of the total concentrations can be calculated (Kuik et al., 1993a,b).

The selection of a relatively large number of elements occurring in high temperature processes (e.g. V, Sb, Se, As) helped to discriminate between processes such as waste incineration, coal combustion, and other high-T sources (Kuik et al., 1993a,b), while the simultaneous analysis of elements like Br, I, and Na suggested a sea-aerosol associated and long-distance origin of As, for about 25% of its total occurrence in mosses in a 1992 survey (Kuik and Wolterbeek, 1995).

One of the most clear and simple examples can be given for the Zn smelting industries in the south-eastern part (Kempen area) of The Netherlands: here, Cd occurs as a by-product in a Zinc ore at a characteristic relative abundancy of 1 Cd:200 Zn. The analysis of a large number of elements, combined with the application of Factor

Analysis, yielded a well-defined finger-print for these Zn smelting industries: they could be characterized by a Zn/Cd factor, in which Cd and Zn were obtained in a 1:212 relative abundancy (De Bruin et al., 1987).

The data suggest that a careful selection of elements facilitates the interpretation of results on each of the individual elements; the larger the number of relevant elements involved in the eventual analysis, the more detailed information may be present in the data-set. Thus, the principal choice may be the multi-elemental analysis: the problem here is how to extract the wealth of information from the set, which may contain thousands of analytical data. A fast and functional approach may be found by the application of Factor Analysis techniques (Kuik et al., 1993a,b).

3. Nuclear analytical techniques in environmental research

Although no sharp lines can be drawn between nuclear and non-nuclear techniques (see De Goeij and Bode, 1997 for a review), the principle of the *nuclear* technique says that the analytical information on element and concentration originates from the nucleus and not from the atom. As such, chemical binding, chemical compound or matrix composition has no essential influence on the accuracy of the results (Bode and Wolterbeek, 1990; De Goeij and Bode, 1997). It should be noted here that although techniques such as particle/proton induced X-ray emission (PIXE) and X-ray fluorescence spectrometry (XRF) are basically derived from the behaviour of inner orbital electrons rather than the nucleus itself, they are often counted as a nuclear technique, primarily because inner orbital electrons do not predominate in the characteristics of the atom's chemical behaviour (but see also De Goeij and Bode, 1997 for NMR and Mössbauer techniques).

Nuclear analytical techniques such as Instrumental Neutron Activation Analysis (γ-rays oriented INAA, see Bode and De Goeij, 1998), PIXE, or XRF are all *multi-element* techniques, and *non-destructive.* Advantages of the first characteristic are discussed elsewhere in this chapter. The latter makes them different from a large number of other widely used (non-nuclear) analytical techniques, such as atomic absorption spectroscopy (AAS), inductively coupled plasma spectrometry (ICP), or mass spectrometry (MS). It also makes that INAA, PIXE or XRF are principally well suited for the routine analysis of the solid samples often encountered in environmental research: it is not necessary to bring the sample into solution, with all the associated problems ranging from incomplete digestion (elemental losses) to impurities in the applied chemicals (elemental contamination) (Bode and Wolterbeek, 1990; De Goeij and Bode, 1995). It may be clear that the absence of effects of chemical forms and the non-destructive character of the nuclear technique pays off particularly in (environmental) base-line surveys comprising large numbers of samples and/or strongly varying sample matrices (e.g. ecosystem research, including samples of soils, plants, animal tissues). It should be noted, however, that nuclear techniques may be successfully applied also in mechanistic (process) dynamic environmental studies, due to the isotope-specific responses in nuclear analytical approaches (De Goeij and Bode, 1997).

4. Nuclear analysis by INAA and PIXE

INAA stands out by non-destructivity, multi-element capability and adequate limits of detection for the majority of elements of environmental interest. The absence of digestion steps and the independence of chemical forms make that a high level of accuracy can be obtained (De Goeij and Bode, 1994, 1997). The intrinsic technique characteristics imply that this high accuracy can be maintained over a large dynamic range, from ppb to % level (Bode and Wolterbeek, 1990).

Gamma-radiation is the radiation of choice in neutron activation analysis, since it is mono-energetic and in most cases characteristic for the emitting nucleus. The other advantage of γ-radiation is that it has a high penetrating power, so that it is hardly adsorbed in the radioactive material itself (Bode and De Goeij, 1998). INAA is routinely used in a mg to g mass range of samples (IAEA/AL/095, 1996), and only when the sample matrix has a high overall atomic number Z, and/or when large samples are being analyzed, problems may arise due to self-attenuation of the induced γ-radiation, and, even more exceptional, self-shielding of the neutrons during irradiation. These phenomena are well-understood, and proper corrections can be applied, thereby making INAA applicable in a mg to kg sample mass range (Bode and Overwater, 1993; Overwater et al., 1993; Bode and De Goeij, 1998).

Although INAA may be calibrated for a large number of elements (up to 70 elements calibrated at IRI, Delft, The Netherlands, see Bode and Wolterbeek (1990)), INAA is not capable of determining low-Z elements of environmental interest, such as Be, B, Li, or high-Z elements such as Pb, Bi and Tl. Here, complementary techniques should be applied.

PIXE and INAA overlap and partly complement each other with respect to elements. The main difference between INAA and PIXE is that X-ray energies associated with PIXE are much smaller than the energies of emitted γ-rays used in routine INAA. These differences come out in differences in self-absorption (absorption of rays within the sample), which makes that PIXE should be practically regarded as a "thin-layer" (surface-related) technique, with energy-related depth-profiles, also depending on general sample matrix characteristics. This means that for all sample materials, including environmental ones such as bio-organisms, biomonitor materials, air particulate matter etc., key steps in PIXE analyses are the sample homogenization and pelleting: eventual elemental determinations are generally carried out on basis of very small (lower mg range) sample masses.

Both INAA and PIXE are intrinsically accurate, but for PIXE the small X-ray energies involved, taken together with the small sample masses in actual analysis, implies a strong dependency on both sample matrix characteristics and bulk sample homogeneity. This makes that for PIXE particular difficulties may be encountered in quantitative calibration procedures. An associated problem is that existing certified reference materials (CRMs) are generally certified for much larger sample masses: they are mostly inadequate for quality control in PIXE (IAEA/AL/095, 1996). Therefore, in both sample and CRM preparations, much effort is devoted to increase the number of sample particles in analyzed sub-samples by grinding into smaller individual particle sizes and avoiding so called "nuggets" (IAEA/AL/095, 1996).

Although high-(energy)-resolution detectors are in use in both PIXE and INAA,

peak overlaps are encountered in both techniques. The resulting doublets or multiplets can often be resolved mathematically without too many difficulties (Bode and De Goeij, 1998), but in both techniques, however, unsolvable multiplets remain, for which parallel information should be available (e.g. Si–Na–Al–Mg and P–Al in INAA, or the As (K_α), Pb (L_α) interferences in PIXE) (Gonsior et al., 1982). Here, too, both techniques may complement each other in resolving overlaps (Knoll, 1989).

5. Survey

5.1. Introduction

In Portugal, data of heavy metals in airborne particulate matter was scarce till the middle of the ninety-decade. Since then, we have published a few papers (Freitas et al., 1996, 1997, 1999a,b; Reis et al., 1996; Freitas and Nobre, 1997; Alves et al., 1998a,b) and some papers were published by the Universities of Aveiro (Pio and Feliciano, 1996; Harrison et al., 1997; Pio and Lopes, 1998, among others), Lisbon (Ruhling, 1992; Sérgio et al., 1993, among others) and Oporto (Tavares et al., 1993; Soares and Vasconcelos, 1995; Vasconcelos and Tavares, 1998, among others). The interest of the heavy metal monitoring is internationally recognised. Although it is not expected severe atmospheric pollution in heavy metals in Portugal due to the geographical position in Europe and due to the lower industrialisation, it is of the most importance to have a reference data basis. This importance is for the country itself but also for the international organisations of the atmosphere, within globalisation programs. The recognition of the problem of lack of data led our Institute to undertake some projects in this area since 1993, supported by Electricity of Portugal, International Atomic Energy Agency, and Portuguese Ministry of the Environment.

The sensibility of lichens to atmospheric pollution was demonstrated in the last two decades in various international and national publications. Recent studies showed that the survey of a large territory with lichens turns identification of atmospheric pollution sources possible (Sloof, 1993). The use of lichens is the only possibility to go through a large scale monitoring, which otherwise would be done with an enormous amount of air samplers and samples. Response of lichens to the environment is being studied through modelling very recently in our Institute (Reis et al., 1999). This is made with lichen transplants whose response to airborne particulate matter, and total deposition is followed experimentally.

5.2. Methods

5.2.1. Sampling

A lichen collection campaign was held in Portugal during the months of July and August 1993. Due to the high industrial asymmetry between the coast and the interior of the country, two different grid sizes were used for sampling site definition. Near the Atlantic coast and up to 30 to 80 km from the coast line, a 10×10 km grid was used; in the interior a 50×50 km grid was selected. A total of 228 sampling squares were thus

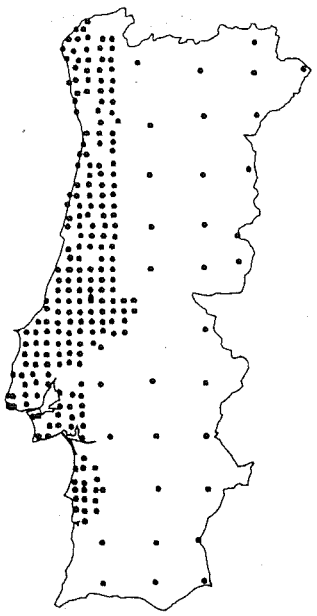

Figure 1. Grid 10 × 10 km at the Atlantic coast and 50 × 50 km at the interior.

defined; within each square and the nearest possible to the centre lichen samples were collected. The chosen lichen was *Parmelia sulcata Taylor* and it was collected from olive tree bark. In each place, this lichen was sampled from at least five different olive trees and all around the trees, at a height of 1–2 m above the ground level. Lichen samples of the same place were mixed in one bag, except in 10% of the places where they were kept separated for local variation determination. In Fig. 1 it is shown the sampling places. Spots missing are squares where it was not possible to find *P. sulcata* or olive tree.

5.2.2. Laboratory preparation

In the laboratory, remains of the tree bark and other lichen species where removed from *P. sulcata* transplants. Then each sample was put in a nylon sieve and rinsed in 18 MΩ water ejected for 30 s, freeze dried and ground in a Teflon® (balls and capsule) mill.

5.2.2.1. Instrumental neutron activation analysis (INAA) Two different size pellets were made: 500 mg was used for long irradiation (5 h) and 50–200 mg for short irradiation (30 s). The long irradiations were done in the Portuguese Research Reactor at ITN at a thermal neutron flux of 1.2×10^{13} cm^{-2} s^{-1} and the short irradiations in the Interfaculty Reactor Institute (Delft University) and the Hahn–Meitner Institut reactors at thermal neutron fluxes of 1×10^{12}–1×10^{16} cm^{-2} s^{-1}. Gamma-spectra of the irradiated samples were collected using hyperpure germanium detectors of high resolution, after 4 days and 4 weeks for long irradiation, and after 10 min. for short irradiation. Gold and zinc were used as comparators. Concentrations were determined using Gent University software for k_0-standardization method. In addition to the lichen samples, reference materials

were prepared and analysed similarly to the lichen samples, for quality control (IAEA 336 lichen material, NIST Tomato leaves, Pine needles and Citrus leaves) (Freitas and Nobre, 1997). Contents for the following elements were determined: sodium, **chlorine**, magnesium, aluminium, **potassium**, **calcium**, scandium, **titanium, vanadium**, **chromium**, **manganese**, **iron**, cobalt, **copper**, **zinc, gallium**, **arsenic**, selenium, **bromine**, **rubidium**, **strontium**, molybdenum, silver, antimony, iodine, caesium, **barium**, lanthanum, cerium, neodymium, samarium, europium, terbium, lutetium, hafnium, tantalum, tungsten, mercury, thorium, and uranium (the elements which are determined by both techniques are shown in bold type).

5.2.2.2. Proton induced X-rays emission (PIXE) Samples were pelletized inserted in a boric acid cap. Samples were irradiated with a 2.0 MeV proton beam for filtered spectra and with a 1.2 MeV for filterless conditions. Spectra were obtained with a 200 eV Si(Li) X-ray detector and analysed with AXIL computer code. Concentrations were then calculated with the DATTPIXE program. Quality control was made by analysing in similar conditions to lichen samples the IAEA 336 lichen material and the CTA-OTL-1 tobacco leaves. Contents for the following elements were determined: silicon, phosphorous, sulphur, **chlorine**, **potassium**, **calcium**, **titanium**, **vanadium**, **chromium**, **manganese**, **iron**, nickel, **copper**, **zinc**, **gallium**, **arsenic**, **bromine**, **rubidium**, **strontium**, zirconium, **barium**, and lead (the elements which are determined by both techniques are shown in bold type).

The two multi-elementary techniques determine 46 elements (16 by both techniques) in *P. sulcata.*

5.3. Results and discussion

The local variation of *P. sulcata* was published somewhere else (Freitas and Nobre, 1997) and found to be on the average ±20%.

In Tables 1 and 2 it is shown the results obtained by PIXE and INAA, respectively. There it is mentioned the number of analysed samples, the maximum and minimum values found for each element, the mean and the median of each elemental set. It is observed that the elements Cu, Ga, Mo, Ag, Nd, and W, are determined by INAA only in a few samples, although Cu and Ga are determined by PIXE in most of the samples. The elements Ca, Sr, and Ba are not determined by INAA in 50 to 100 samples, although Ca is determined by PIXE in all the samples.

In Table 3 it is presented the mean and median differences (in %) between the results obtained by both techniques, and the maximum and the minimum of these differences. In general there is a normal distribution around the mean (which is quite similar to the median) for all the elements. The value of the mean gives an idea of the existence of a systematic error. For K, Ca, V, Fe, Zn, Br, and Sr the means are lower than ±10%, and between ±10 and ±30% for Cl, Ti, Mn, and Ba, for the others the means are larger than ±30% and go as far as ±88% (Cr, Cu, Ga, and As). The elemental heterogeneity of the samples may be the main cause for the differences. It is reminded that PIXE uses very small amounts of the sample (estimated less than 1 mg) while INAA uses 500 mg at least. In the further paragraphs, it will be discussed element by element.

Table 1. Results of PIXE (mg/kg) for the 1993 lichen survey

	Si	P	S	Cl	K	Ca	Ti	V	Cr	Mn	Fe
No. of samples	299	300	300	299	300	300	300	259	266	300	300
Maximum	3.24×10^4	2.92×10^3	3.96×10^3	2.65×10^1	8.05×10^3	3.13×10^4	7.77×10^2	1.30×10^2	3.40×10^1	1.43×10^2	6.39×10^3
Minimum	1.21×10^3	3.00×10^2	9.75×10^2	$4.72 \times 10^2 2$	2.65×10^3	7.42×10^2	4.22×10^1	1.83×10^0	2.32×10^{-1}	1.56×10^1	3.57×10^2
Mean	8.99×10^3	1.34×10^3	2.03×10^3	1.08×10^3	4.65×10^3	6.18×10^3	2.42×10^2	1.45×10^1	2.17×10^0	3.96×10^1	1.94×10^3
Median	7.84×10^3	1.31×10^3	1.96×10^3	1.02×10^3	4.52×10^3	5.51×10^3	2.18×10^2	1.09×10^1	1.62×10^0	3.61×10^1	1.76×10^3
	Ni	Cu	Zn	Ga	As	Br	Rb	Sr	Zr	Ba	Pb
No. of samples	299	300	299	250	251	300	300	300	253	170	299
Maximum	3.31×10^1	2.65×10^2	5.08×10^2	3.94×10^0	1.87×10^1	5.73×10^1	7.64×10^1	1.03×10^2	8.75×10^1	7.99×10^1	1.42×10^2
Minimum	5.23×10^{-1}	3.30×10^0	2.21×10^1	3.52×10^{-1}	6.97×10^{-1}	8.71×10^0	4.71×10^0	2.84×10^0	2.52×10^0	2.01×10^0	2.03×10^0
Mean	3.92×10^0	2.88×10^1	6.91×10^1	1.13×10^0	3.82×10^0	2.29×10^1	1.75×10^1	2.29×10^1	1.32×10^1	2.29×10^1	1.84×10^1
Median	3.00×10^0	1.49×10^1	5.57×10^1	1.01×10^0	2.86×10^0	2.15×10^1	1.52×10^1	2.03×10^1	1.11×10^1	1.96×10^1	1.41×10^1

Table 2. Results of INAA (mg/kg) for the 1993 lichen survey

	Na	Mg	Al	Cl	K	Ca	Sc	Ti	V	Cr
No. of samples	301	295	297	297	301	255	297	297	297	301
Maximum	2.70×10^3	4.71×10^3	2.88×10^4	3.81×10^3	1.09×10^4	2.63×10^4	2.83×10^0	1.04×10^3	1.45×10^2	3.77×10^1
Minimum	1.31×10^2	1.90×10^2	2.17×10^0	2.48×10^2	2.10×10^3	1.15×10^3	1.35×10^{-1}	5.44×10^1	2.26×10^0	1.44×10^0
Mean	5.85×10^2	1.91×10^3	5.40×10^3	1.37×10^3	5.24×10^3	6.49×10^3	6.53×10^{-1}	3.20×10^2	1.57×10^1	5.45×10^0
Median	4.90×10^2	1.76×10^3	4.63×10^3	1.27×10^3	5.01×10^3	5.62×10^{-1}	2.48×10^2	1.14×10^1	4.69×10^0	
	Mn	Fe	Co	Cu	Zn	Ga	As	Se	Br	Rb
No. of samples	296	297	297	117	301	92	301	299	300	300
Maximum	1.47×10^2	7.57×10^3	2.90×10^0	3.45×10^2	449×10^2	7.89×10^0	3.10×10^1	1.17×10^0	9.31×10^0	$\pm$ 01
Minimum	1.87×10^1	3.90×10^2	1.36×10^{-1}	2.54×10^0	1.60×10^1	2.80×10^{-1}	4.35×10^{-1}	1.32×10^{-1}	7.08×10^0	3.97×10^0
Mean	5.02×10^1	2.10×10^3	7.75×10^{-1}	6.32×10^1	7.12×10^1	2.018×10^0	2.13×10^0	4.02×10^{-1}	2.25×10^1	1.59×10^1
Median	4.63×10^1	1.85×10^3	6.68×10^{-1}	4.72×10^1	5.61×10^1	1.78×10^0	1.34×10^0	3.73×10^{-1}	1.98×10^1	1.36×10^1
	Sr	Mo	Ag	Sb	I	Cs	Ba	La	Ce	Nd
No. of samples	219	41	11	291	297	299	250	301	301	90
Maximum	1.09×10^2	2.77×10^0	4.80×10^{-1}	6.73×10^0	1.16×10^2	3.95×10^0	1.60×10^2	1.34×10^1	2.62×10^1	1.34×10^1
Minimum	7.59×10^0	3.67×10^{-1}	5.17×10^{-2}	7.49×10^{-2}	1.05×10^0	1.03×10^{-1}	4.01×10^0	$4.75 \times 10^{-1}11$	1.06×10^0	1.42×10^0
Mean	2.35×10^1	9.41×10^{-1}	1.89×10^{-1}	3.42×10^{-1}	7.57×10^0	6.40×10^{-1}	3.35×10^1	3.00×10^0	6.56×10^0	4.28×10^0
Median	2.06×10^1	8.28×10^{-1}	1.68×10^{-1}	2.20×10^{-1}	6.15×10^0	5.12×10^{-1}	2.96×10^1	2.49×10^0	5.70×10^0	3.72×10^0
	Sm	Eu	Tb	Lu	Hf	Ta	W	Hg	Th	U
No. of samples	301	289	297	292	301	301	126	296	301	200
Maximum	1.85×10^0	7.47×10^{-1}	2.44×10^{-1}	1.14×10^{-1}	1.83×10^0	3.93×10^{-1}	1.28×10^1	1.75×10^0	6.60×10^0	1.81×10^0
Minimum	8.89×10^{-3}	3.25×10^{-2}	7.26×10^{-3}	4.44×10^{-3}	5.53×10^{-2}	9.92×10^{-3}	8.97×10^{-2}	9.17×10^{-2}	8.66×10^{-2}	7.87×10^{-2}
Mean	4.47×10^{-1}	1.84×10^{-1}	6.09×10^{-2}	2.61×10^{-2}	4.13×10^{-1}	7.51×10^{-2}	5.03×10^{-1}	3.02×10^{-1}	9.26×10^{-1}	2.71×10^{-1}
Median	3.90×10^{-1}	1.62×10^{-1}	5.12×10^{-2}	2.22×10^{-2}	3.45×10^{-1}	5.99×10^{-2}	2.62×10^{-1}	2.78×10^{-1}	6.94×10^{-1}	2.07×10^{-1}

Table 3. Comparison of INAA and PIXE results (%) for the 1993 lichen survey

	Cl	K	Ca	Ti	V	Cr	Mn	Fe
No. of samples	296.0	300.0	255.0	296.0	259.0	266.0	296.0	297.0
Maximum	143.8	105.7	177.1	164.6	183.7	188.4	146.5	144.4
Minimum	−125.1	−79.8	−131.7	−132.3	−168.3	−135.1	−97.0	−142.5
Mean	20.7	9.8	0.0	24.6	2.2	87.8	23.3	5.1
Median	21.4	8.9	4.9	27.5	2.4	94.7	25.0	6.6
	Cu	Zn	Ga	As	Br	Rb	Sr	Ba
No. of samples	117.0	299.0	86.0	251.0	300.0	300.0	219.0	170.0
Maximum	191.2	154.1	159.2	179.8	126.0	141.2	170.3	178.1
Minimum	−145.3	−136.4	−155.1	−177.8	−120.4	−136.3	−143.5	−147.1
Mean	54.6	1.9	33.4	−60.8	−6.0	−11.5	0.5	27.5
Median	58.8	−2.1	36.1	−78.0	−10.0	−13.7	−2.9	25.4

The results obtained by INAA and PIXE for the different elements were mapped as concentration data patterns for the whole country by making use of an extinction rule of $1/r^3$ (r is the distance between plot point and sampling point) without any cut-off distance artificially introduced. It means that for every grid cell of the map a plot value was calculated from the concentrations found for all sampling points as the average of these concentrations weighted with the 3rd power of the distance between plot point and sampling point (Sloof, 1993). The computer code used was Surfer®. In Figs. 2–13 it is shown the maps so-obtained for all the elements determined. The discussion is made on basis of the pollution sources associated to elements as defined by Nriagu (Nriagu, 1989).

5.3.1. Coal-fired power stations

The elements associated to the coal-fired power stations are arsenic, sulphur, and selenium. Their pattern maps are shown in Fig. 2.

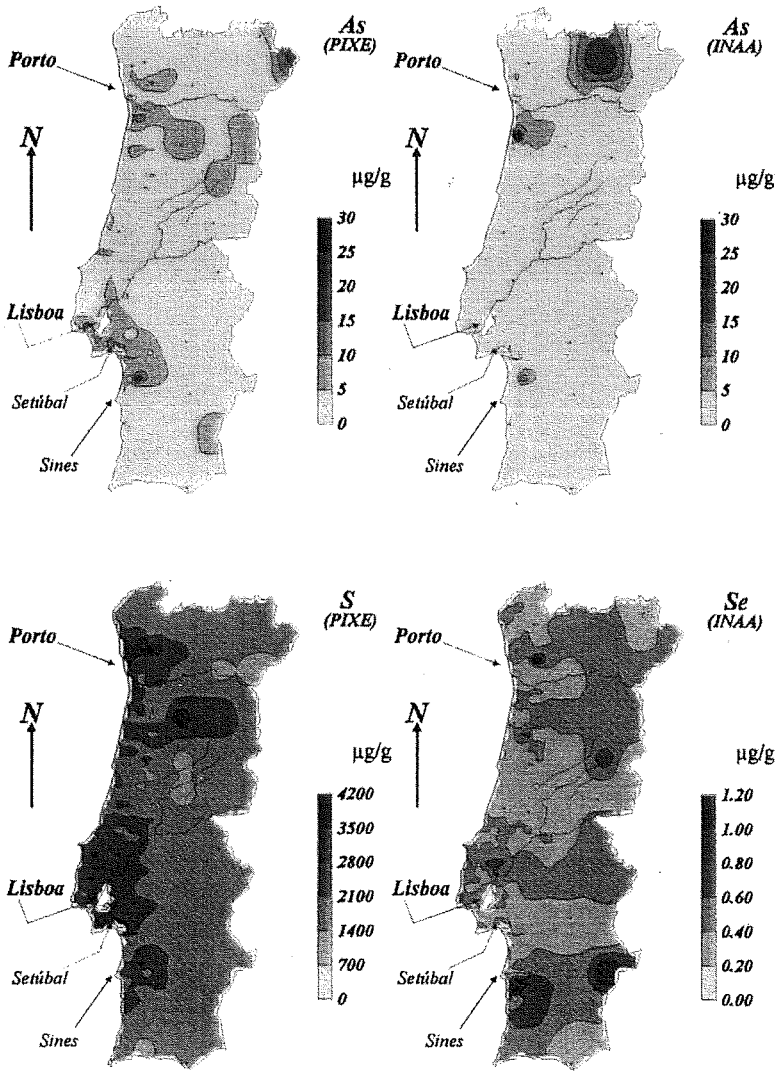

Figure 2. 1993 Survey using *P. sulcata*.

The arsenic data obtained by PIXE and INAA present quite different patterns. Whenever a large concentration of lead is present in the samples arsenic spectra peaks obtained by PIXE can be interfered, and also if arsenic is present in large quantities lead data can be also interfered. Comparing Fig. 2 with Fig. 5 we observe that in the south of the country around Lisbon the Pb-As pattern is quite similar. In the other hand Fig. 5 presents large concentrations of Pb in the north of the country which is there quite similar to the As pattern given by INAA. Therefore we are taken to conclude that the As-pattern given by PIXE should not be selected for this kind of samples. The difference between the two techniques shown in Table 3 is now explained. Therefore INAA data should be selected.

The correlation of As, S, and Se shown in Fig. 2 is hardly observed. In the north of the country, As-pattern resembles the pattern of the elements related to soil contribution and shown in Fig. 8, therefore there arsenic is predominantly originated on the ultrabasic complexes of that area, as already referred by Prudêncio et al. (1997). There is some similarity for As, S, and Se at south of Lisbon and near Oporto which are, respectively, in the neighbourhood of the Sines coal-fired power station and the Tapada do Outeiro coal-oil fired power station. Reis et al. (1996) and Freitas et al. (1997) did not identify clearly these elements to coal combustion in their factor analysis. Rahn and Huang (1999) define a coal source with a high peak of As and smaller peaks of S and Se, besides other elements.

5.3.2. *Fuel-fired power stations*

The elements associated to the fuel-fired power stations are vanadium, nickel, and rare earth elements (lanthanum, cerium, neodymium, samarium, europium, and terbium). Their pattern maps are shown in Fig. 3. Vanadium patterns obtained by both techniques are very similar which agrees with the similarity of values obtained for V and shown in Table 3 (mean difference between both techniques: 2.2%). Therefore INAA and PIXE are good techniques for V determination in lichens.

Comparing the figures we observe that the largest concentrations of vanadium are

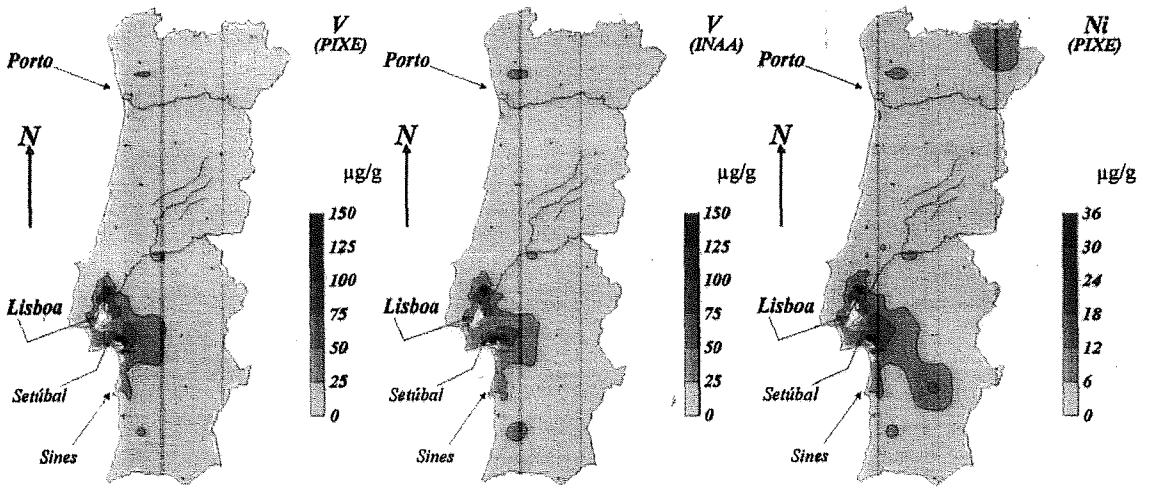

Figure 3. 1993 Survey using *P. sulcata.*

found in the same area of nickel, both indicating the oil-fired power stations of Carregado and Setúbal around Lisbon and the coal-oil power station of Tapada do Outeiro as the main sources (confirmed by factor analysis made by Reis et al. (1996), Freitas et al. (1997) and Rahn and Huang (1999)). The spot at north-east of Lisbon is not identified. Nickel has still a maximum in the north-east of the country in the same place as the elements correlated to soil contribution, which is referred by Prudêncio et al., 1997 as soil based in ultrabasic complexes.

When we look at the rare earth elements shown in Fig. 4, we conclude that there is no correlation with Ni and V. They are more similar to the elements discussed further in the soil contribution part, so we delay the discussion till that moment.

5.3.3. Mobile sources

The elements associated to mobile sources are bromine and lead. Their pattern maps are shown in Fig. 5. INAA and PIXE patterns of bromine are similar showing both a predominance of bromine at the Atlantic coast. The results obtained by the two techniques are similar as shown also in Table 3 (mean difference between the two techniques: −6%). Both maps show no correlation with Pb pattern and Br seems to have a marine origin. As referred before lead determined by PIXE may have influence on arsenic data and it was concluded that in north of Portugal lead shows a maximum (large spot in the middle of interior north) because there is a maximum of arsenic there. So we consider that spot as arsenic not lead. The largest concentrations of lead coincide with the regions of larger traffic in Portugal so we consider the mobile sources as the main origin of Pb. The spot at south-east should be considered as soil contribution since it also appears for the soil correlated elements.

5.3.4. Incinerators

The elements associated to incinerators are silver, zinc, antimony, cadmium, tin, and lead. Of these elements Zn, Sb, and Pb were determined in this work. Their pattern maps are shown in Fig. 6 (Fig. 5 for Pb). Zn obtained by both techniques gives similar information for the same large areas (near Oporto and near Lisbon), although the patterns are not so similar. In Table 3 it is presented a mean difference between the two techniques of 1.9%. The patterns of Zn, Sb, and Pb show larger concentrations of these elements in the same large areas: axis Lisbon-Setúbal-Sines and in the vicinity of Oporto, therefore it might be consequence of incineration. Officially there are no incinerators in our country, the first one started operation in February, 1999. We think that these elements appear associated due to incineration in the open air at small scale. Rahn and Huang (1999) applied the positive matrix factorisation to our data and found one factor which he identified as industry where antimony and lead are the largest components, and zinc is also present. Freitas et al. (1997) included it in one factor associated to general oil powered industries.

5.3.5. Cement production

The elements associated to cement production are calcium and magnesium. Their

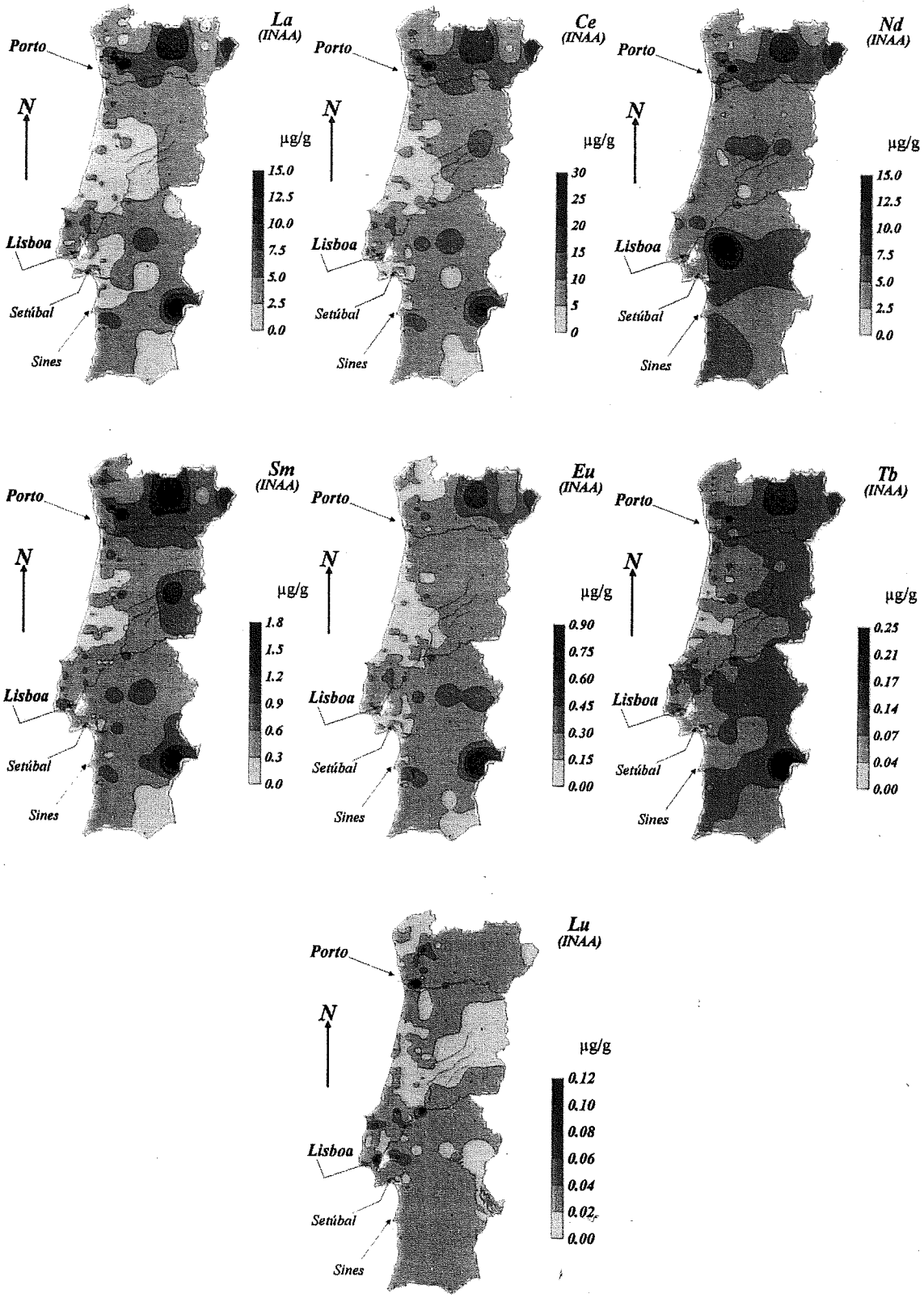

Figure 4. 1993 Survey using *P. sulcata*.

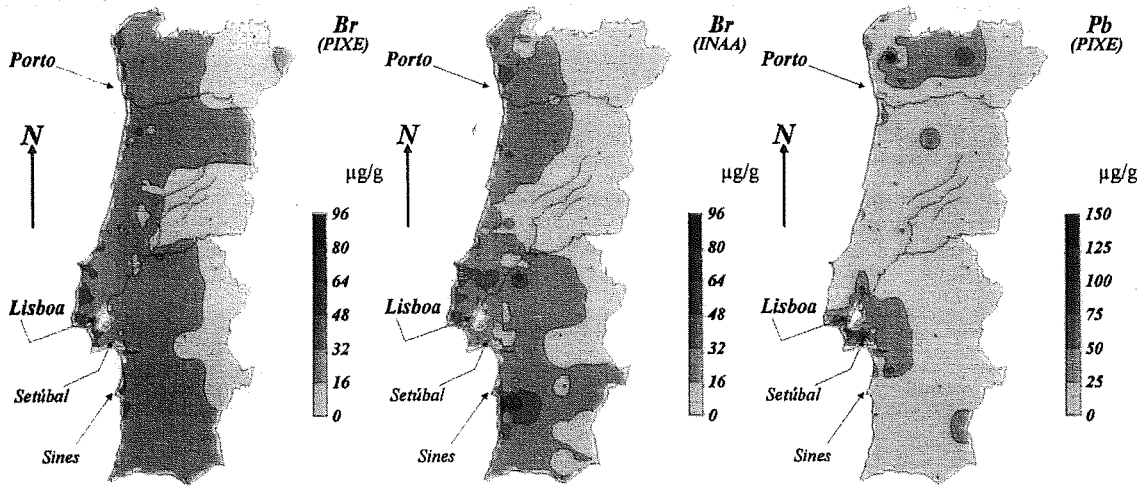

Figure 5. 1993 Survey using *P. sulcata*.

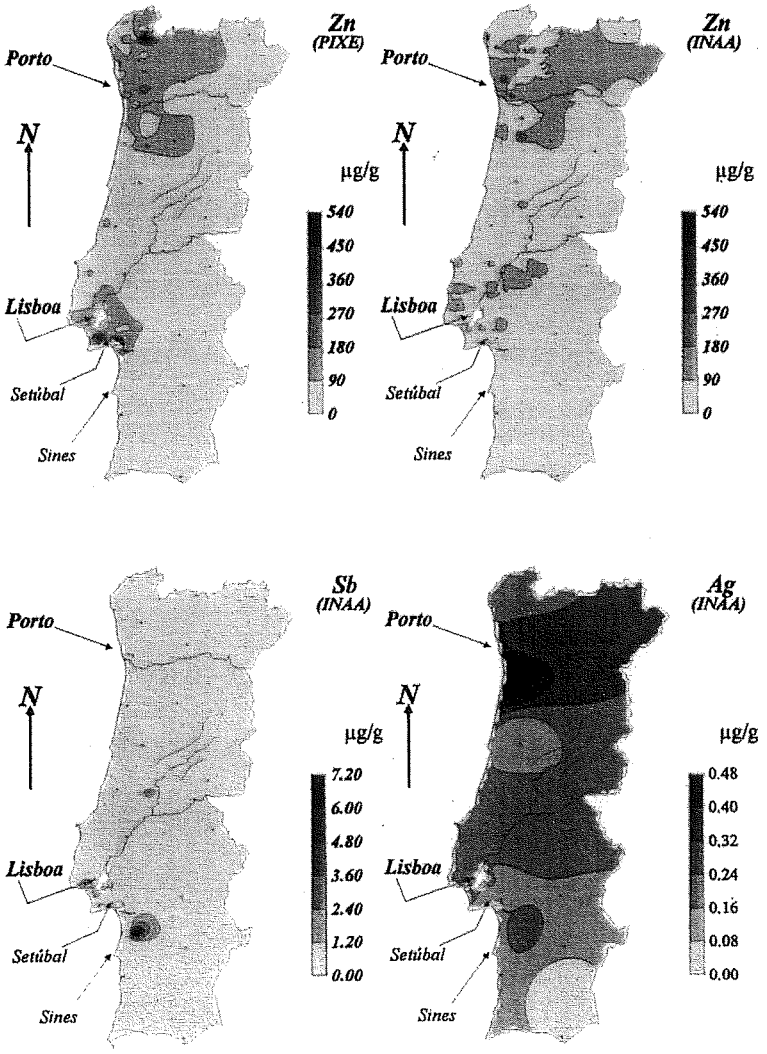

Figure 6. 1993 Survey using *P. sulcata*.

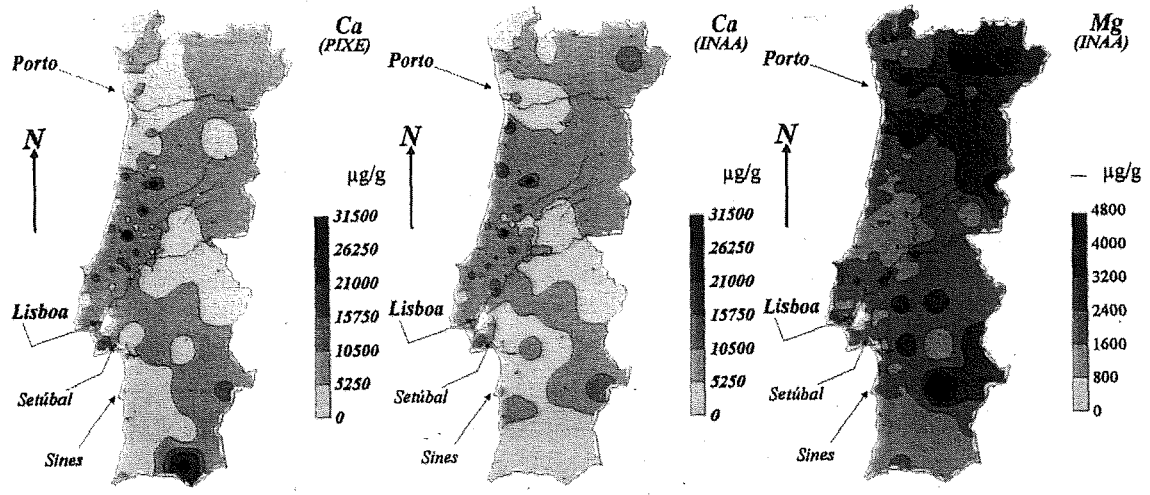

Figure 7. 1993 Survey using *P. sulcata*.

pattern maps are shown in Fig. 7. Calcium patterns determined by both techniques are similar with a few spots well pointed out, which in Freitas et al. (1997) factor analysis identified as cement sources (also Rahn (1999) identified this source after a more detailed factorisation). Ca values of INAA do not show the large spot in the south of the country as PIXE, because INAA Ca values were not measured. This is the reason why INAA values for Ca are only 255. In terms of the values itself the mean difference is 0% which is quite good (see Table 3). Magnesium is not associated to calcium and the pattern is more of one soil component (see further).

5.3.6. Soil contribution

The elements associated to soil contribution are, according to Nriagu (1989), manganese, aluminium, scandium, silicon, iron, and titanium. Their pattern maps are shown in Fig. 8. Manganese and titanium values determined by INAA are larger than the PIXE values. The mean difference found for both elements is about 20%. However, for each element the patterns are similar and the largest concentrations are found in the same areas. Iron data obtained by both techniques agree each other and also the patterns. The mean difference for Fe is 5% (see Table 3). For all the elements, the maps show high concentrations in the north-east of the country, due to the ultrabasic composition of the soils as referred previously for arsenic, nickel, the rare earth elements, lead, and magnesium. No explanation was found for the fact that only a few of the elements in discussion have heavy spots in south-east of the country. The patterns are similar for these elements demonstrating its correlation with soil contribution. The rare earth elements shown in Fig. 4 have similar patterns to the soil contribution elements. This is not surprising since the composition of soil in rare earths is soil tracer.

5.3.7. Wood burning

The elements associated to wood burning are potassium and carbon. Unfortunately

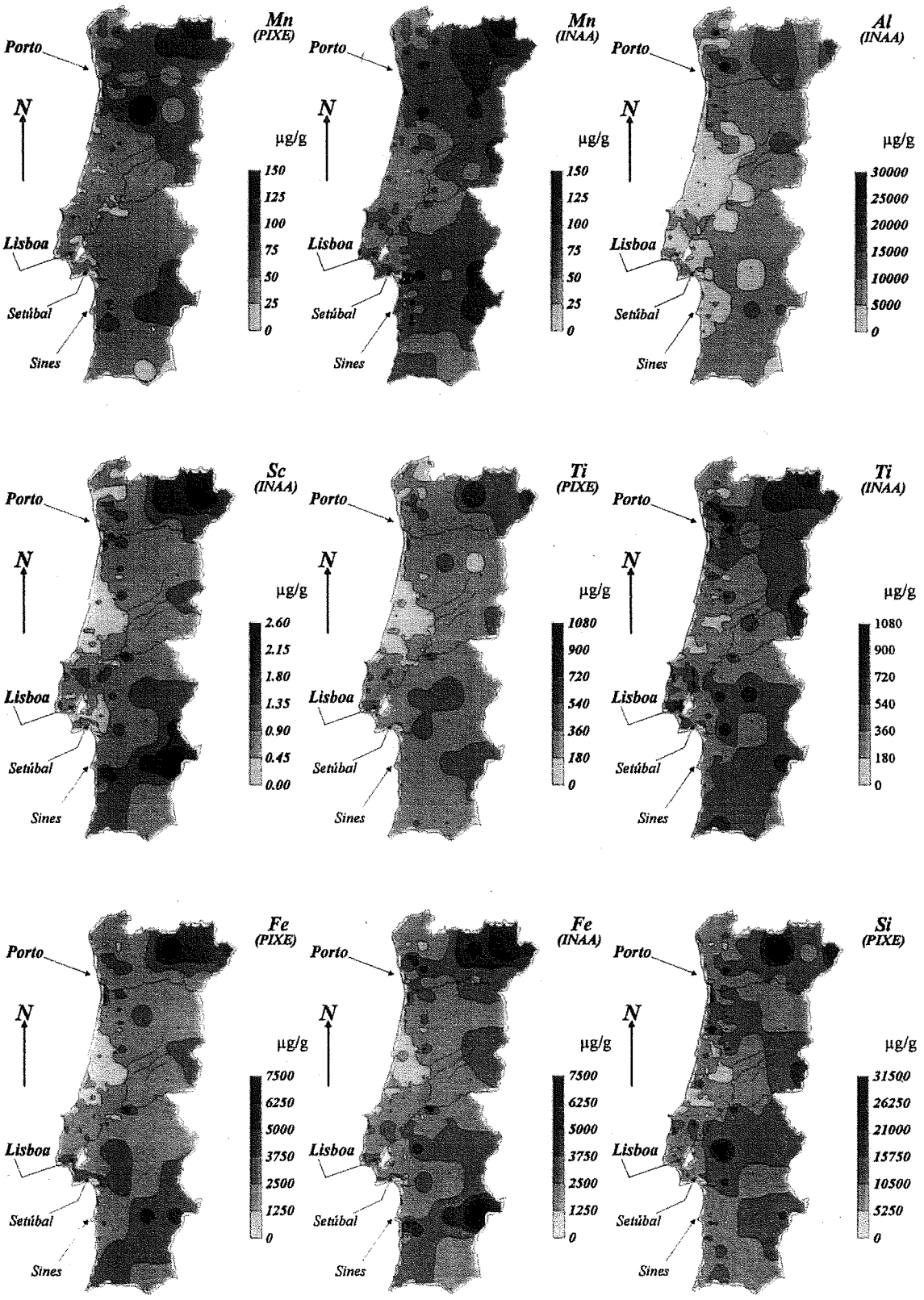

Figure 8. 1993 Survey using *P. sulcata*.

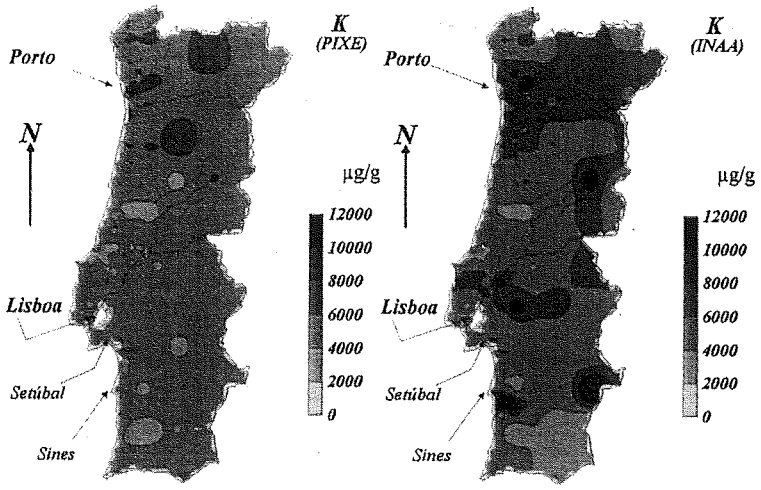

Figure 9. 1993 Survey using *P. sulcata.*

carbon is not determined by any of the techniques. Fig. 9 shows the potassium obtained by PIXE and INAA. The mean difference between both techniques is 10% (see Table 3). The patterns are similar although the PIXE values are lower. The regions with larger concentrations of potassium coincide with the areas of the country with more percentage of fires during summer, but we would need carbon to demonstrate the correlation.

5.3.8. Refineries

The rare earth elements are shown in Fig. 4. It was said previously that these elements are more soil contribution than anything else. Looking more in detail we observe that neodymium has a different pattern (except in the northern region) and samarium shows large concentrations in the centre east. The spot south of Sines might be related to the Sines refinery, as well as the small spot near Oporto.

5.3.9. Sulphide smelters

The elements associated to sulphide smelters are indium, cadmium, arsenic, selenium, and sulphur. In and Cd were not determined because of their high limits of detection with these techniques. The other elements were shown in Fig. 2 in association to coal-fired power stations. One spot which was not explained there was the one which is north-east of Sines, present in As- and S-patterns but not in Se-pattern. Might be that spot due to sulphide smelters?

5.3.10. Other elements

A few more elements were determined which are presented in Figs. 10–12. We will start to comment the similarity of the elements determined by both INAA and PIXE.

For Cl, INAA values are larger than the PIXE values and the difference is 20% on the average (see Table 3). In general the patterns indicate both more Cl near the

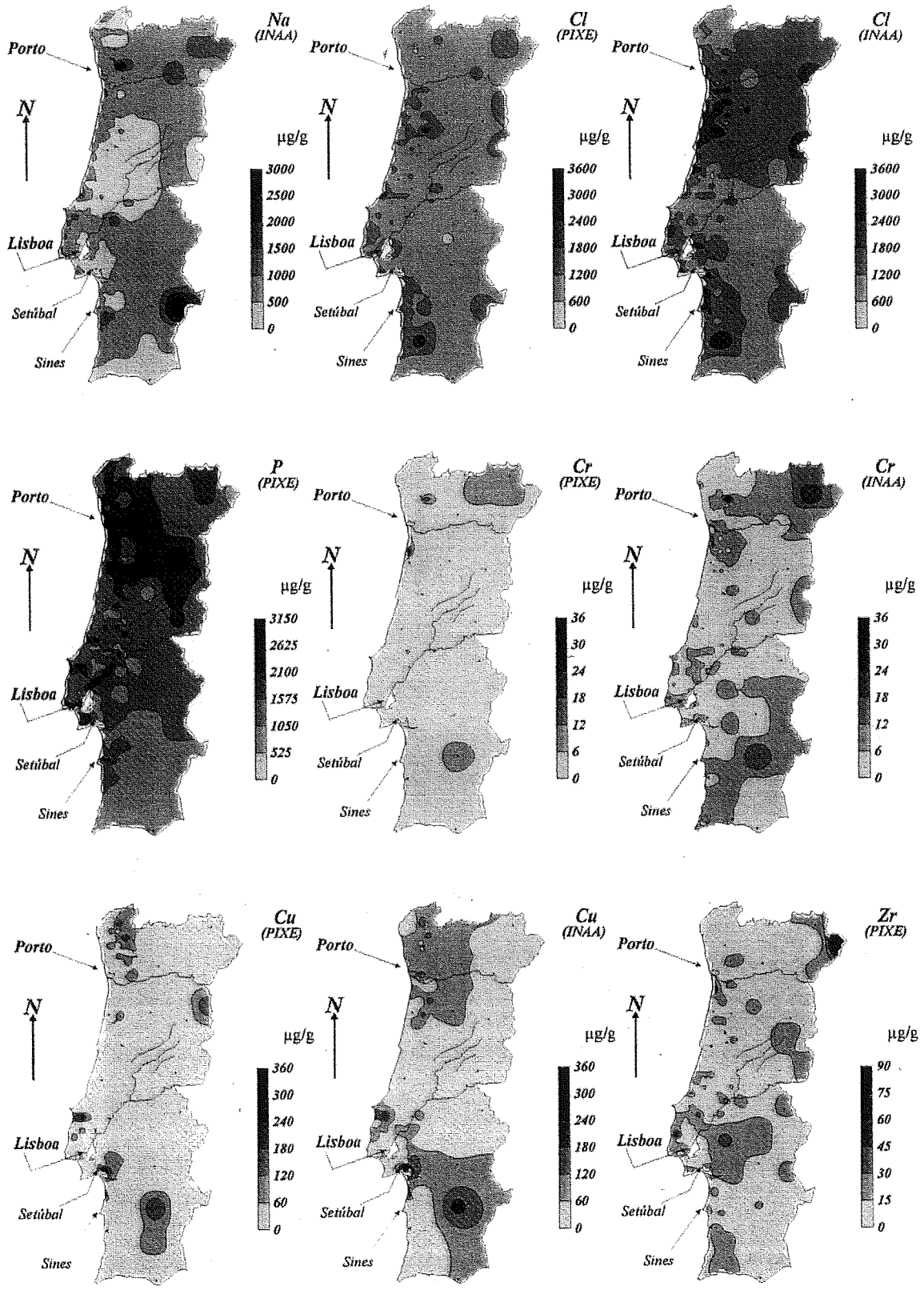

Figure 10. 1993 Survey using *P. sulcata*.

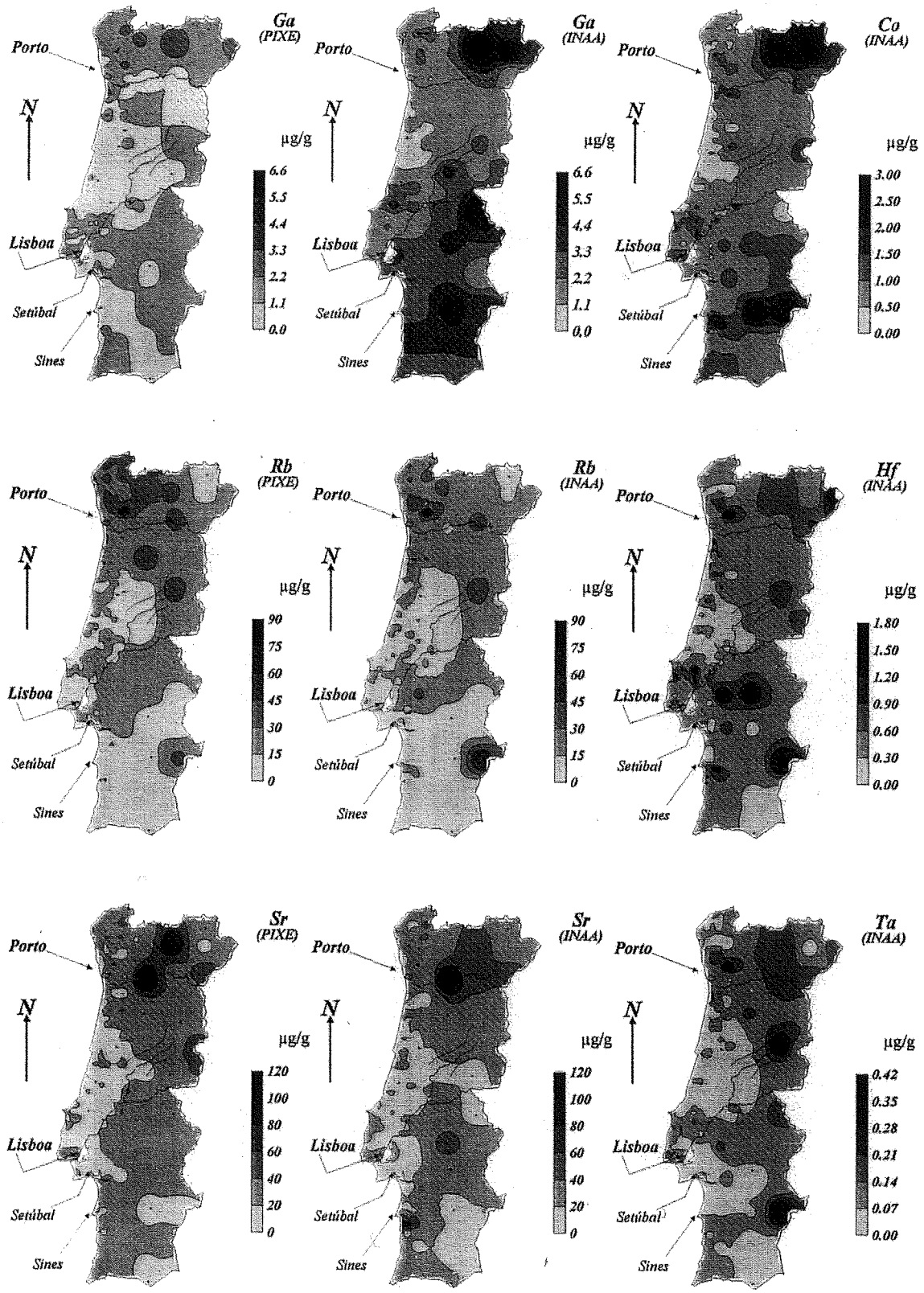

Figure 11. 1993 Survey using *P. sulcata.*

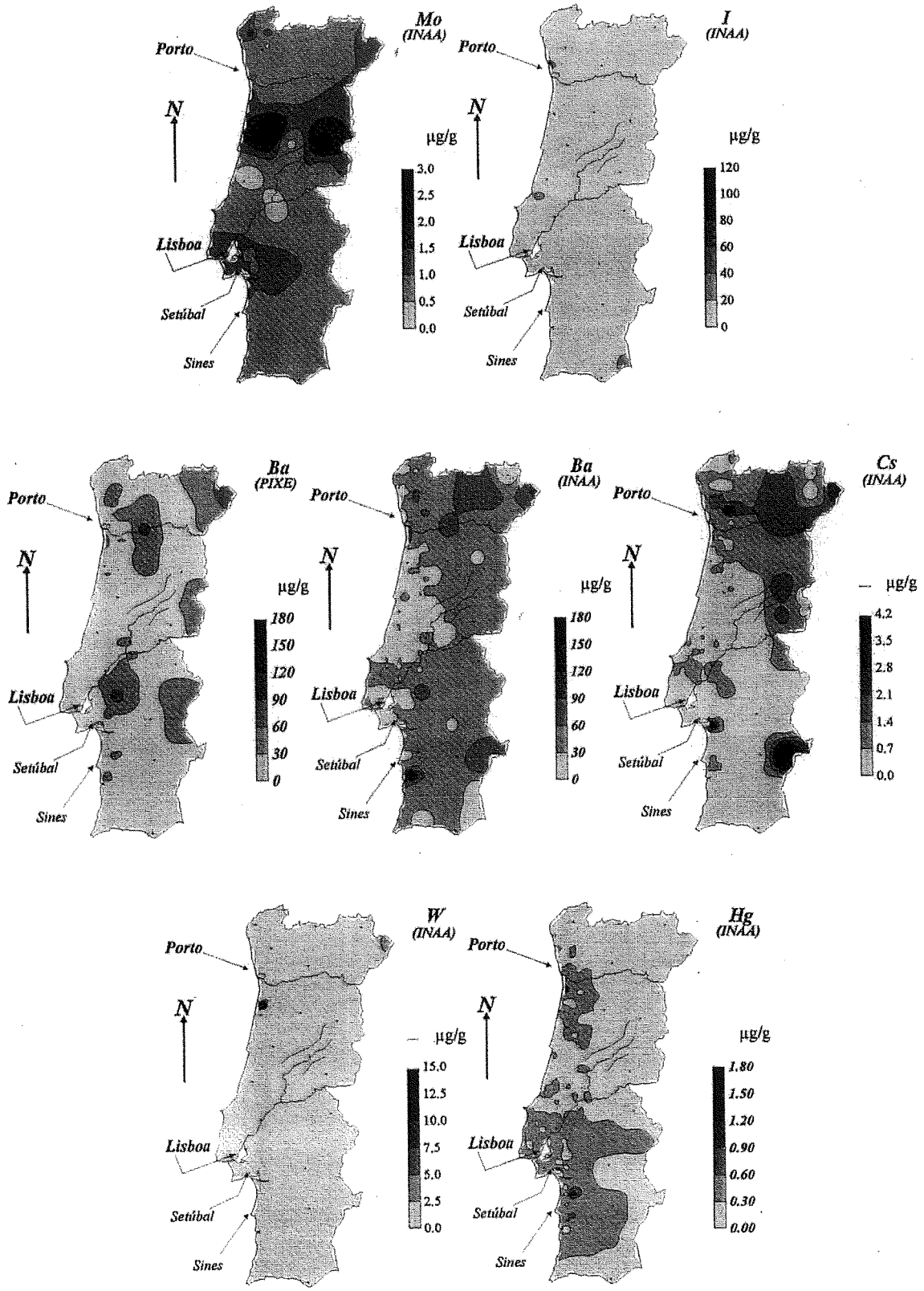

Figure 12. 1993 Survey using *P. sulcata*.

Atlantic coast as expected given the proximity of the ocean. For Cr, the mean difference (see Table 3) is quite large (88%). The INAA values are larger than the PIXE values and we think it might be due to interference of the sum peak of Sc-47 peak originated in Ca-47 ($2 \times 160 = 320$ keV). We conclude that Cr values obtained by PIXE are probably more accurate. INAA determined copper only in 117 samples in a total of 301 due to the worse detection limit. Also the mean difference is quite large (55%, see Table 3). We think PIXE is more reliable than INAA in Cu determination. For Ga, very few results were obtained by INAA. Therefore the PIXE values should be preferred. For Rb, both techniques give similar values (mean difference: 12%, see Table 3) and similar patterns. Therefore we should adopt both Rb sets. For Sr, the values of both techniques agree quite well (mean difference: 0.5%, see Table 3) and also the patterns are similar. The drawback for INAA is that Sr values of INAA are just 219 instead of 301. For Ba, the mean difference is 28% (see Table 3) and the patterns are not so similar. Ba values are interfered with U fission and correction is sometimes quite important. That may be the reason why INAA results are larger than the PIXE ones. In the other hand the PIXE results reach only 170 in a total of 300 samples. Therefore both techniques have drawbacks and it is suggested to accept both. In short, for the elements just discussed the following patterns will be adopted in Figs. 10–12.

- Both techniques: Cl, Rb, Sr, and Ba;
- Cr, Cu, and Ga: the selection will be done after patterns analysis.

We will discuss now eventual associations of the elements shown in Figs. 10–13 among themselves in one hand, and with the other elements discussed in the previous sections.

A marine source might be associated to Na (Fig. 10), Cl (Fig. 10), and I (Fig. 12) but their patterns are not similar. Na accumulates at the Atlantic coast but Cl also spreads to the north-centre interior indicating some other source than the ocean spray. Phosphorus concentrates more over the agricultural part of Portugal (north and centre) and copper in the north-west and south-west of the country might originate in copper sulphate used in the vineyards. Co (Fig. 11), Cs (Fig. 12), Hf (Fig. 11), and Ta (Fig. 11) have patterns similar to the soil elements. The values of Cr (Fig. 10), Ba (Fig. 12), and Ga (Fig. 11) obtained by INAA also have soil patterns but not the PIXE ones. In general these elements are indeed soil components therefore the INAA patterns look more plausible than the PIXE ones. Zr (Fig. 10) has a very specific pattern with spot in the north-east much probably originated in the soil. Mo (Fig. 12) shows two strong spots near the centre of the country of unknown source. Wolfram was expected to be more concentrated in the centre interior where some important W (Fig. 12) mines are located, but instead the main spot is located south of Oporto where important chemical industry is located. In this area we find also Hg (Fig. 12) contamination, as well as in the axis Lisbon-Setúbal-Sines axis where the chemical industries are also concentrated. As to Th and U (Fig. 13) the patterns are very similar with concentrated spots in the centre interior where the U mines of Urgeiriça are located. The spots in the north-east should be soil originated and the spots in the north-west are of unknown source.

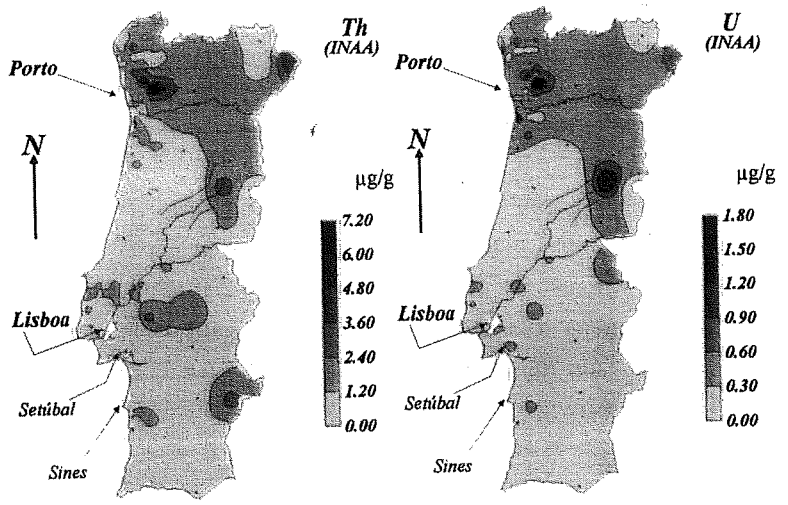

Figure 13. 1993 Survey using *P. sulcata.*

5.4. Comparison with other surveys

In Ruhling (1992) Portuguese mosses were collected from 1990 to 1992 and analysed for the elements Cd, Cr, Cu, Fe, Pb, Ni, and Zn. Comparison with Cr, Pb, and Ni was made in Freitas et al. (1999). It was found that in Portugal, surveys based on *P. sulcata* and the mosses *Hypnum cupressiforme* and *Scleropodium touretii* give similar information for these elements. We will compare now the elements Cu, Fe, and Zn. For Cu, similar large concentrations are found in the north-west and south-centre of the country. The large concentrations found in the interior centre are not visible in our survey probably because of the different grids adopted in the two surveys, finer with mosses.

Table 4. Results obtained in The Netherlands (mg/kg) in 1982/83 and 1986/87 reprinted from Sloof (1993)

Element	No. of samples analysed	Minimum	Maximum	Mean
Arsenic	110/329	1.7/0.5	17/17	5.7/5.7
Bromine	110/337	17/15	120/170	44/56
Chromium	110/337	7.6/4.0	58/270	22/26
Caesium	108/322	0.3/0.2	21/5.3	0.9/0.8
Iron	110/337	1100/720	15000/30000	4100/5800
Mercury	99/316	0.1/0.1	3.7/36	0.4/0.5
Lanthanum	99/316	0.8/0.8	16/64	4.1/6.2
Nickel	69/201	3.0/1.9	53/51	14/16
Lead	/232	/3.1	/367	/147
Selenium	98/332	0.4/0.4	4.0/7.5	1.4/1.8
Antimony	110/334	0.6/0.3	10/12	3.0/3.3
Vanadium	102/99	8.8/5.7	130/99	29/32
Wolfram	93/248	0.1/0.1	2.0/10	0.5/0.9
Zinc	110/327	80/61	630/1100	90/210

For Fe, the results are similar for both surveys: larger concentrations in the interior north and south. For Zn, the largest spots are in both surveys around Lisbon and around Oporto, although the Zn areas in the north of the country are larger for the lichen survey.

In The Netherlands similar survey was made in 1982/83 and 1986/87 using also *P. sulcata* (Sloof, 1993). In Freitas et al., (1999) comparison was made for arsenic, chromium, mercury, nickel, lead, antimony, selenium, and vanadium. It was concluded that when compared to The Netherlands, Portugal has regions with much lower contents in lichens, but the industrialised areas are as much polluted. Table 4 shows the levels of the elements which can be compared in this paper.

We observe that Br, Cr, Fe, Hg, and Zn contents in Portuguese lichens are quite lower than the Dutch lichens; V and W have similar values; Ni and Sb have lower minima; Cs, La, Pb, and Se lower maxima; and As has larger maxima. Comparing the means we observe that except for W all the elements in comparison are much lower in Portugal, for that contributing the not industrialised interior of the country. Therefore we may say that in Portugal pollutants can be as high as in The Netherlands in some regions of the country (near the Atlantic coast) but on the average the contents are still low.

6. Conclusions

In this work PIXE and INAA nuclear techniques were applied to a 1993 lichen survey held in Portugal. The two techniques determined 46 elements with 16 common elements. The common elements were compared and conclusions were attempted to select the best values, based on the concentration patterns obtained. It was concluded that:

- Ca, Cu, Ga, Sr, Mo, Ag, La, Nd, and W were determined by INAA but their detection limits are not good enough to give results in all the samples; Ca, Cu, and Ga are determined by PIXE in all the samples.
- On average: 1) K, Ca, V, Fe, Zn, Br, and Sr agree each other within $\pm 10\%$; 2) Cl, Ti, Mn, and Ba between $\pm 10\%$ and $\pm 30\%$; 3) Cr, Cu, Ga, and As between $\pm 30\%$ and $\pm 90\%$; the heterogeneity of the samples may be the main cause of the differences.
- In PIXE results whenever As and Pb are present in the samples, they interfere each other, and the values should be observed carefully.
- Some pollution sources could be regionally identified: 1) coal-fired power stations (As, Se, and S) at south of Lisbon and near Oporto; 2) fuel-fired power stations (V and Ni) in the axis Lisbon-Setúbal and near Oporto; 3) mobile sources (Pb) in regions of larger traffic (north-west and Lisbon and south Lisbon); 4) cement production (Ca); 5) soil contribution: Mn, Al, Sc, Si, Fe, and Ti, also Mg, REE, Co, Cs, Hf, and Ta (Cr, Ba, and Ga).
- It was observed two regions of large concentrations of metals in north-east of the country pointed out as originated in the ultrabasic complexes, main constituents of the soil of that region.
- The chemical complex of Estarreja (south of Oporto) is put in evidence by the Hg contents in lichens.

- Except for a few spots our results for Cr, Pb, Ni, Cu, Fe, and Zn agree quite well with the results obtained with a moss survey in a 1990/92 campaign made in Portugal using non-nuclear techniques.
- In general the pollutants determined in this work in the industrialised areas of the country near the Atlantic coast can be as highly concentrated as in The Netherlands (1982/83 and 1986/87 Dutch campaigns with the same lichen).There are regions of the country mainly located in the interior which have very low concentrations as compared with the Dutch values. Therefore on the average the elemental contents in the Portuguese lichens are still low as compared with the Dutch ones.

References

Alves, L.C., Reis, M.A., Freitas, M.C., 1998a. Air particulate matter characterisation of a rural area in Portugal. Nucl. Instrum. Methods Phys. Res. B136–138, 941–947.

Alves, L.C., Reis, M.A., Freitas, M.C., Gouveia, M.A., 1998b. Elemental analysis of particulate matter and source profile in Lisbon. X-Ray Spectrom. 27, 313–320.

Bode, P., De Goeij, J.J.M. 1998. Activation Analysis, In: R.A. Meyers (Ed.), Encyclopaedia of Environmental Analysis and Remediation, Wiley, New York, pp. 68–84, ISBN 0-471-11708-0.

Bode, P., Overwater, R.M.W., 1993. Trace element determinations in very large samples: a new challenge for neutron activation analysis. J. Radioanal. Nucl. Chem. 167, 169–176.

Bode, P., Wolterbeek, H.Th., 1990. Environmental research and instrumental neutron activation analysis: aspects of high accuracy and multi-element capability. J. Trace Microprobe Technol. 8, 121–138.

De Bruin, M., Wolterbeek, H.Th. 1984. Identification of sources of heavy metals in the Dutch atmosphere using air filter and lichen analysis. Proceedings of the Fifth International Conference on Nuclear Methods Environment Energy Resources, University of Missouri, Columbia, MO, pp. 266–276.

De Bruin, M., Van Wijk, P.M., Van Assema, R., De Roos, C., 1987. The use of multi-element concentration data sets obtained by INAA in the identification of sources of environmental pollutants. J. Radioanal. Nucl. Chem. 112, 199–213.

De Goeij, J.J.M., Bode, P., 1994. How to make nuclear methods the choice of a new generation of scientists. J. Radioanal. Nucl. Chem. 1179, 7–12.

De Goeij, J.J.M., Bode, P. 1995. Neutron Activation Analysis: Trends in Developments and Applications. Proceedings of International Conference on Neutrons and their Applications, Crete, Greece, 12–18 June 1994, SPIE-Vol. 2339, pp. 436–447.

De Goeij, J.J.M., Bode, P. 1997. Nuclear Analytical Techniques. Strong and weak points, obstacles and opportunities. In: Proceedings of the Symposium on Harmonization of Health related Environmental Measurements using Nuclear and Isotopic Techniques, Hyderabad, India, 4–7 November 1996, pp. 3–17, IAEA Proc. Series, Vienna, STI/PUB/1006, ISBN 92-0-103697-3.

Freitas, M.C., Nobre, A.S., 1997. Bioaccumulation of heavy metals using Parmelia sulcata e Parmelia caperata for air pollution studies. J. Radioanal. Nucl. Chem. 217 (1), 17–20.

Freitas, M.C., Reis, M.A., Alves, L.C., Wolterbeek, H.Th., Verdurg, T., Gouveia, M.A., 1996. Elemental accumulation in lichen transplants in the neighbourhood of thermal power stations. ANST/Nuclear Methods Environ. Res. 74, 117–118.

Freitas, M.C., Reis, M.A., Alves, L.C., Wolterbeek, H.Th., Verdurg, T., Gouveia, M.A., 1997. Monitoring of trace-element air pollution in Portugal: qualitative survey. J. Radioanal. Nucl. Chem. 217 (1), 21–30.

Freitas, M.C., Reis, M.A., Alves, L.C., Wolterbeek, H.Th., 1999a. Response of lichen Parmelia sulcata to environmental pollutants in Portugal. Environ. Pollut. 106, 229–235.

Freitas, M.C., Reis, M.A., Alves, L.C., Marques, A.P., Costa, C., 1999b. Environmental assessment in a industrial area of Portugal. Biol. Trace Elem. Res. 71/72, 471–479.

Gonsior, B., Bischof, W., Raith, B., Stratman, A., Wilde, H.R. 1982. Investigation of trace element distributions in biological structures using PIXE. In: Cesareo, R. (Ed.), X-ray Fluorescence (XRF and PIXE) in Medicine. Acta Medica, ROMA, Italy, pp. 125–153, ISBN 88-7084-002-6.

Harrison, R.M., Smith, D.J., Pio, C.A., Castro, L.M., 1997. Comparative receptor modelling study of airborne particulate pollutants in Birmingham (UK). Coimbra (Portugal) and Lahore (Pakistan), Atmos. Environ. 31, 3309–3321.

IAEA 1997. Proceedings of the Symposium on the Harmonization of Health-related Environmental Measurements using Nuclear and Isotopic Techniques, Hyderabad, India, 4–7 November 1996. IAEA Proc. Series STI/PUB/1006, ISBN 92-0-103697-3, pp. 663.

IAEA/AL/095 1996. Report of the Second Research Co-ordination Meeting on Reference Materials for Micro-analytical Nuclear Techniques; Mexico, DF, Mexico, May–June 1996, pp. 143.

Knoll, G.F. 1989. Radiation Detection and Measurement. 2nd ed. Wiley, New York, ISBN 0-471-81504-7.

Kuik, P., Wolterbeek, H.Th., 1995. Factor analysis of atmospheric trace-element deposition data in The Netherlands obtained by moss monitoring. Water, Air, Soil Pollut. 83, 323–346.

Kuik, P., Sloof, J.E., Wolterbeek, H.Th., 1993a. Application of Monte-Carlo-assisted factor analysis to large sets of environmental pollution data. Atmos. Environ. 27A, 1975–1983.

Kuik, P., Blaauw, M., Sloof, J.E., Wolterbeek, H.Th., 1993b. The use of Monte Carlo methods in factor analysis. Atmos. Environ. 27A, 1967–1974.

Nriagu, J.O. 1989. Natural versus anthropogenic emissions of trace metals to the atmosphere. In: J.M. Pacyna, B. Offar (Eds.), Control and Fate of Atmospheric Trace Metals, NATO ASI Series, Kluwer Academic Publishers, Dordrecht.

Overwater, R.M.W., Bode, P., De Goeij, J.J.M., 1993. Gamma-ray spectroscopy of voluminous sources. Corrections for source geometry and self-attenuation. Nucl. Instrum. Methods A324, 209–218.

Pio, C.A., Feliciano, M.S., 1996. Dry deposition of sulphur dioxide and ozone over low vegetation in moderate southern European weather conditions. Measurements and modelling. Ann. Geophys. Part II (Suppl. II, Col. 14), C468.

Pio, C.A., Lopes, D., 1998. Chlorine loss from marine aerosol in a coastal atmosphere. J. Geophys. Res. 103, 25263–25269.

Prudêncio, M.A., Gouveia, M.A., Freitas, M.C., Chaves, L., Marques, A.P., 1997. Soil versus lichen analysis on elemental dispersion studies (north of Portugal). IAEA TECDOC in press.

Rahn, K.A., Huang, S. 1999. A graphical technique for distinguishing plant material in biomonitors from soil and atmospheric deposition. Sci. Total Environ. 232, 79–104.

Reis, M.A., Alves, L.C., Freitas, M.C., Wolterbeek, H.Th., Verburg, T., Gouveia, M.A., 1996. Main atmospheric heavy metal sources in Portugal by biomonitor analysis. Nucl. Instrum. Methods Phys. Res. B109/110, 493–497.

Reis, M.A., Alves, L.C., Freitas, M.C., Van Os, B., Wolterbeek, H.Th., 1999. Lichens (*Parmelia sulcata*) time response model to environmental availability. Sci. Total Environ. 232, 105–115.

Ruhling, A. 1992. Atmospheric heavy metal deposition in Europe - estimations based on moss analysis, Nord 1994:9, Copenhagen, Denmark.

Sérgio, C., Sim-Sim, M., Figueira, R. 1993. Quantificação da deposição de metais pesados em Portugal através da análise de briófitos, DGA, Lisbon.

Sloof, J.E. 1993. Environmental Lichenology Biomonitoring Trace Element Air Pollution. Ph.D. Thesis, Delft University of Technology, The Netherlands.

Soares, H.M.V.M., Vasconcelos, M.T.S.D., 1995. Application of potentiometric stripping analysis for speciation of copper complexes with adsorbable ligands on the mercury electrode. Anal. Chim. Acta 314, 241–249.

Tadic, T., Jaksic, M., Bogdanovic, I., Fazinic, S., Dujmic, D. 1997. Proton micro-PIXE control of standard reference materials for PIXE environmental applications. In: Proceedings of the Symposium on the Harmonization of Health-related Environmental Measurements using Nuclear and Isotopic Techniques, Hyderabad, India, 4–7 November 1996. IAEA Proc. Series STI/PUB/1006, ISBN 92-0-103697-3, pp. 251–264.

Tavares, H.M.F., Vasconcelos, M.T.S.D., Machado, A.A.S.C., Silva, P.A.P., 1993. Application of hydride generation atomic absorption spectrometry to the determination of lead collected on air filters and sphagnum moss. Analyst 118, 1433–1439.

Trahey, N.M. 1996. NIST Standard Reference Materials Catalog, 1995–1996, NIST Special Publication 260, NIST, Gaithersburg, MD, pp. 84.

Vasconcelos, M.T.S.D., Tavares, H.M.F., 1998. Atmospheric metal pollution (Ca, Cr, Cu, Fe, Mn, Ni, Pb and Zn) in Oporto city derived from results for low-volume aerosol samplers and for moss sphagnum auricularem bioindicator. Sci. Total Environ. 212, 11–20.

Wolterbeek, H.Th., Bode, P., 1995. Strategies in sampling and sample handling in the context of large-scaled plant biomonitoring surveys of trace element air pollution. Sci. Total Environ. 176, 33–43.

Wolterbeek, H.T.H., Bode, P., Verburg, T.G., 1996. Assessing the quality of biomonitoring via signal-to-noise ratio analysis. Sci. Total Environ. 180, 107–116.