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ATMOSPHERIC ELEMENT POLLUTANT EVALUATION AT THE SÃO PAULO UNIVERSITY CAMPUS, SÃO PAULO USING *Canoparmelia texana* LICHEN SPECIES

Rosiana R. Rocha and Mitiko Saiki

Instituto de Pesquisas Energéticas e Nucleares (IPEN-CNEN/SP)
Av. Professor Lineu Prestes 2242
05508-000, São Paulo, SP, Brazil
rosianarocha@yahoo.com.br

ABSTRACT

The use of lichens as biomonitors of atmospheric pollutants has been considered as a very suitable tool when compared to conventional methods of direct measurements. Lichens in particular are widely used as biomonitors due to its easy sampling, low cost and resistance to environmental stresses. In this study, neutron activation analysis (NAA) was applied for element determinations in *Canoparmelia texana* lichenized fungi species. The samples were collected from tree barks in different sites at the São Paulo University Campus and in sites of areas considered non-polluted. Comparisons were made between the element concentration obtained in lichen from the study area and those from non-polluted sites. Results indicated that lichens from study area presented higher concentrations of As, Br, Ca, Cd, Co, Cr, Cs, Fe, K, La, Rb, Sb, Se and U than clean areas. The principal components analysis (PCA) was applied to the results obtained and five principal components were found as being responsible for almost 77 % of the variance. These findings suggest that element pollutants found may be associated with vehicular emissions, construction of buildings and metallurgical activities.

1. INTRODUCTION

The emission of atmospheric pollutants affects the health, well-being of the population and climatic conditions. This emission occurs mainly through processes of burning fossil fuels in motor vehicles, urbanization and industrial activities. The presence of atmospheric pollutants is one of main causes to several health disorders. Therefore is extremely important to monitor and to identify the levels of these atmospheric pollutants.

The air quality can be evaluated by direct measurement using collecting equipment. However this makes the air monitoring procedure expensive and difficult in large urban areas like São Paulo city. An alternative method is to use biological monitoring that allows an intensive assessment on a large region and over a period of time with a significant reduction in total sampling costs [1].

Various monitor materials have been applied in element air monitoring programs, such as lichens, mosses, ferns, grass, tree bark, tree rings, and pine needles [2]. Particularly, lichen species are widely used in air quality monitoring. They are slow-growing organisms and maintain a fairly uniform morphology in time [3], as well as being able to accumulate pollutants from the atmosphere or from soils [4]. Lichens are sensitive to a wide range of changes in the habitats, most of them driven by anthropogenic activities. This sensitivity makes lichens particularly advantageous for use as indicators of levels of disturbance in the environment.

The campus of the São Paulo University (USP) is considered a green urban space in the São Paulo city. However, this area is located close to highways with intense vehicular traffic, a small cement industry, chemical laboratories, buildings under construction and a hospital incinerator.

In this study, neutron activation analysis (NAA) was used for element determination in *Canoparmelia texana* (Tuck.) Elix & Hale lichenized fungi species, in order to evaluate the level of elements in atmosphere of the Campus of the São Paulo University, São Paulo city. Comparisons were made between the results obtained in lichens from this study area and those from areas considered as unpolluted.

2. MATERIALS AND METHODS

2.1. Study Area

The study area included different sampling sites at the campus of the São Paulo University (USP) in São Paulo city, located in the western part of this city (23.55 S and 46.72 W, elevation about 750 m above sea level) with land area of approximately 4, 492, 414.40 km^2 in which 843.47 km^2 are edificated.

The lichen samples from clean areas were collected in Forestry Institute in the Itirapina city located at 220 km far from São Paulo city and in Botucatu city, in the Botanic garden of the University of the State of São Paulo (UNESP) located about 230 km far from São Paulo city. These sampling sites of clean area were coded as clean region 1(CR1) of Itirapina, and clean region 2 (CR2) of Botucatu.

Figure 1: Location of the Campus of the São Paulo University (study area) in Brazilian territory

2.2. Sampling

The epiphytic lichen samples of the *Canoparmelia texana* species were collected during the period from January 2013 to June 2014, from tree barks at a height of about 1.5 m from the ground. For collection, lichens of the tree trunks were wetted with purified water and then removed from the bark using a titanium knife and stored in paper bags. The study area was divided into quadrants to standardize the collection sites. Forty lichen samples were collected in different sites at the Campus of the São Paulo University. The samples from clean areas were acquired from the University of the State of São Paulo (UNESP).

2.3. Preparation of the sample for analysis

The lichen samples were cleaned by removing extraneous materials and bark residues by examining them under a stereomicroscope model SZ 4045, Olympus. Then they were immersed into purified Milli-Q water where they remained for about 3 min. The cleaned samples were freeze-dried and then ground to fine powder using a vibratory micro mill Pulverisette 0 from Fritsch with an agate mortar.

2.4. Preparation of the synthetic elemental standards

To prepare the element standards for neutron activation analysis (NAA), standard solutions of the following elements were used: As, Br, Ca, Cd, Cl, Co, Cr, Cs, Cu, Fe, K, La, Mg, Mn, Rb, Sb, Sc, Se, U, V and Zn. These solutions containing one or more elements were prepared using certified standard solutions provided by Spex Certiprep Chemical, USA.

The synthetic standards were prepared by pippeting 50 μL of these solutions onto sheets of Whatman No. 40 filter paper. All the pippetors and volumetric flasks were verified in relation their calibration before use. These filter sheets were dried at room temperature inside a desiccator with fresh silica and then placed into clean polyethylene bags which were heat sealed.

2.5. Neutron activation analysis procedure

For NAA analysis, aliquots of about 180 mg of each sample weighed in polyethylene bags were irradiated at the IEA-R1 nuclear research reactor along with the synthetic element standards for short and long periods. Twenty five-seconds irradiations under a thermal neutron flux of about 6.6 x 10^{12} n cm^{-2} s^{-1} were carried out for Cl, Mg, Mn and V determinations. Sixteen-hours irradiations under a thermal neutron flux of about 5 x 10^{12} n cm^{-2} s^{-1} were performed for As, Br, Ca, Cd, Co, Cr, Cs, Cu, Fe, K, La, Rb, Sb, Sc, Se, U and Zn determinations.

After adequate decay times, the irradiated samples and standards were measured using a hyperpure Ge detector Model GC3020 from Canberra coupled to a DSA-1000 Multichannel Analyzer. The resolution (FWHM) of the system was of 1.0 keV for 122 keV gamma-ray peak of ^{57}Co and 1.87 keV for 1332 keV gamma ray peak of ^{60}Co. Counting times from 300 to 50,000 seconds were used, depending on the half-lives or radioactivities of the radionuclides considered. Spectra were collected and processed using Canberra Genie 2000 Version 3.1 software. The radionuclides measured were identified according to their half-lives and gamma-ray energies. The concentrations of elements were calculated by a comparative method.

For quality control of the results, the certified reference material (CRM) IAEA-336 lichen provided by the International Atomic Energy Agency (IAEA), Austria, was analyzed using the same experimental conditions of the lichen analyses. The element concentrations of the reference material were evaluated on a dry weight basis, as recommended in its certificate.

3. RESULTS AND DISCUSSION

Results obtained in the analyses of certified reference material IAEA 336 Lichen are presented in Table 1 together with values of its certificate [5]. These results indicate good accuracy and precision with relative standard deviations ranging from 3.2 to 11.2 % and relative errors lower than 9.4 %. The standardized difference or En score [6] values obtained at the confidence level of 95 % were │En-score│≤ 1, indicating the reliability of the results.

Table 1: Elements concentrations (in mg kg^{-1}) in certified material IAEA Lichen 336

Elements	Values of the certificate	This study			
		M ± SD[a] (n)[d]	RSD[b] (%)	RE[c] (%)	E_n score
As	0.63 (0.55-0.71)[e]	0.63 ± 0.06(5)	9.5	0.0	-0.00
Br	12.9 (11.2-14.6)	13.0 ± 0.5(4)	4.3	0.7	0.01
Ca	-	2555 ± 177(5)	6.9	-	-
Cl	1900 (1600-2200)*	1722 ± 136(5)	7.9	9.4	-0.14
Co	0.29 (0.24-0.34)	0.27 ± 0.02(5)	6.0	6.9	-0.08
Cr	1.06 (0.89-1.23)*	1.09 ± 0.05(5)	4.6	2.8	0.03
Cs	0.110 (0.097-0.123)	0.114 ± 0.007(5)	6.2	3.6	0.07
Fe	430 (380-480)	435 ± 18(5)	4.2	1.1	0.01
K	1840 (1640-2040)	1903 ± 159(5)	8.3	3.4	-0.05
La	0.66 (0.56-0.76)	0.61 ± 0.05(5)	8.2	7.5	-0.12
Mg	-	881 ± 99(4)	11.2	-	-
Mn	63 (56-70)	62 ± 6(5)	9.6	1.6	-0.02
Rb	1.76 (1.54-1.98)*	1.66 ± 0.11(5)	6.6	5.7	-0.10
Sb	0.073 (0.063-0.083)	0.071 ± 0.003(5)	4.2	2.7	-0.05
Sc	0.17 (0.15-0.19)*	0.18 ± 0.01(5)	5.5	5.9	0.12
Se	0.22(0.18-0.26)	0.24 ± 0.02(5)	8.3	9.0	0.12
V	1.47 (1.25-1.69)*	1.46 ± 0.12(5)	8.2	0.7	-0.01
Zn	30.4 (27.0-33.8)	31.2 ± 1.0(5)	3.2	2.6	0.03

[a] Arithmetic mean and standard deviation; [b] relative standard deviation; [c] relative error; [d] n are numbers of determinations; [e] confidence interval; * numbers with asterisks are informative values.

Results of mean concentrations of elements obtained in the analyses of lichens collected at the Campus of the São Paulo University and those in clean regions CR1 and CR2 are presented in the Figures from 2 to 4. These Figures show that the mean concentrations of As, Br, Ca, Cd, Co, Cr, Cs, Fe, K, La, Rb, Sb, Se and U obtained in lichen from Campus of USP are higher than those of clean sites, while for Sc and V they are of the same order of magnitude. On the other hand, the Cl concentration in CR1 is higher than those of the study area. For Mg and Mn the concentrations were higher for lichen from CR2. The lichens of CR2 presented Cu and Zn concentrations of the same order of magnitude of the study area.

The highest concentrations of Cl, Mg and Mn obtained in lichens from clean regions may be related to natural sources. Hauck et al., [7] suggest that high Mn concentrations are an important site factor for epiphytic lichens, and primarily soil-borne. Magnesium is pivotal and plays an important role in numerous physiological and biochemical processes affecting plant growth and development [8]. Chlorine is a common element in the terrestrial environment and this element is found in igneous rocks, sandstone, calcareous rocks, minerals as deposits of solid rock salt, as dissolved ingredient in seawater and subsurface brines [9]. The high concentration of Cl in CR1 can be attributed to natural soil composition.

The elements found in lichens from study area may be attributed to different sources. The presence of As in the environment has been related to the burning of fossil fuels [10]. The occurrence of high levels of Ca in lichen samples may be attributed to powder of a cement plant located near of the study area and from dust of the buildings under construction [11]. The content of Cd in lichens can be associated to road dust and to diesel and gasoline tailpipe emissions [12]. The Sb emissions have been attributed to brake wear [12] and can be associated to the emission of plastic material of waste incineration. Brazilian plastic materials contain Sb, since compounds of this element are used in plastic processing [13-15]. The presence of Cr in air pollution has been related to metallurgical industries.

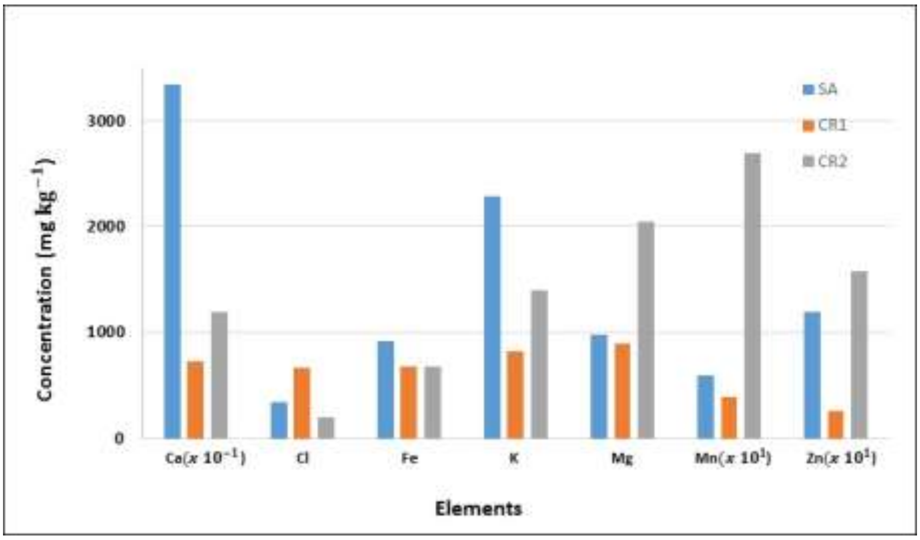

Figure 2: Mean concentrations of Ca, Cl, Fe, K, Mg, Mn and Zn in lichens from study a rea (SA) and from two clean regions (CR1 and CR2)

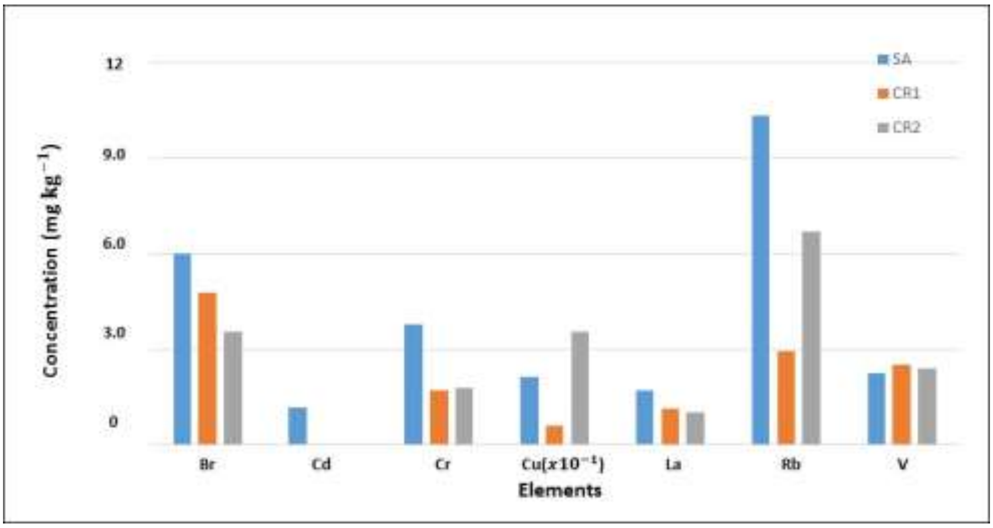

Figure 3: Mean concentrations of Br, Cd, Cr, Cu, La, Rb and V in lichens from study area (SA) and from two clean regions (CR1 and CR2)

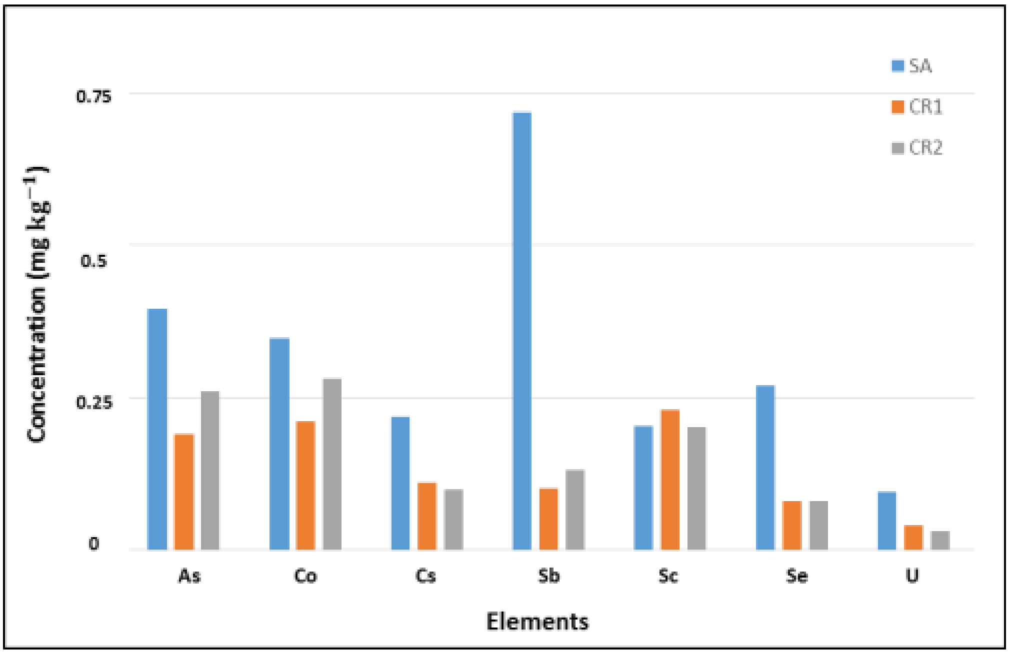

Figure 4: Mean concentrations of As, Co, Cs, Sb, Sc, Se and U in lichens from study area (SA) and from two clean regions (CR1 and CR2)

The principal component analysis (PCA) using SPSS software from IBM was applied in the results obtained in lichens collected in the Campus of the São Paulo University. This treatment was applied for the identification of the relationship between of the elements levels found with as emission sources. In Table 2 are the result of PCA where the results in bold are the elements that presented greater contribution of each factor or component. Five principal components were found accountable for almost 77% of the variance. The first component group is formed by the elements As, Co, Cr, Fe, La, Sb, Sc, Se, U and V, whereas the second group by Cd, Cl, Mn and Zn. The third group is formed by Ca, Cs, Cu and Rb, the fourth by K and the fifth by

Mg**.** PCA showed that the first group is most representative in the results of lichens from the study area. The origins of the elements As, Co, Cr, Fe, La, Sb, Sc, Se, U and V of this group may be associated to the emissions of industrial, vehicular and soil dust sources.

Table 2: Results of principal components analysis (PCA) for the results in lichens from study area

Elements	Rotating matrix component[a]				
	Components				
	1	2	3	4	5
As	**0.713**	-0.168	0.276	-0.221	-0.179
Br	0.227	0.020	0.089	-0.777	-0.074
Ca	-0.075	0.199	**-0.697**	-0.087	0.409
Cd	0.057	**0.853**	-0.280	0.019	0.025
Cl	-0.005	**0.620**	-0.059	-0.059	-0.468
Co	**0.831**	0.090	0.336	-0.149	0.016
Cr	**0.940**	0.073	0.087	0.252	0.073
Cs	0.351	-0.117	**0.678**	0.016	0.248
Cu	0.154	0.460	**0.573**	-0.363	0.226
Fe	**0.948**	0.148	0.079	0.056	0.123
K	0.383	-0.125	0.506	**0.615**	0.268
La	**0.786**	0.390	-0.205	0.010	-0.041
Mg	0.030	-0.079	0.056	0.076	**0.800**
Mn	0.011	**0.798**	0.092	-0.080	0.090
Rb	0.259	-0.165	**0.548**	0.497	0.438
Sb	**0.657**	0.070	0.132	0.571	-0.205
Sc	**0.947**	0.057	0.046	0.004	0.148
Se	**0.748**	0.074	0.265	-0.097	0.008
U	**0.822**	-0.089	0.059	0.234	-0.098
V	**0.799**	0.052	0.235	-0.246	0.199
Zn	0.197	**0.824**	-0.045	0.034	-0.167

Extraction method: Principal Component Analysis.
Rotation Method: Varimax with Standardization Kaiser
[a]Converged rotation in 32 iteractions.

4. CONCLUSIONS

The results of the analysis of certified reference material using the NAA technique were consistent with the values of certificates, indicating the accuracy of the obtained data. The relative standard deviation values obtained in this analysis also indicate good accuracy of the results certain elements.

From the results obtained in lichen from the campus of USP we can conclude that most element concentrations were higher in the study area than in clean areas. These findings indicate that the Campus of USP, a green urban space area, may be affected by air pollutants from various sources.

The principal components analysis showed that As, Co, Cr, Fe, La, Sb, Sc, Se, U and V are the most representative elements in lichen of study area. The results obtained indicate that emissions from cement plant, industrial activities and vehicular traffic may be the causes of atmospheric pollution in the study area.

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