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Biomonitoring of atmospheric lead, cadmium and zinc using *Eucalyptus* leaves in Rabat Salé City, Morocco

A. EL ABIDI[1], S.E. BARCHA[2*], M.FEKHAOUI [3], F. Z. EL ALAOUI-FARIS[4], A. YAHYAOUI[4], S. AYRAULT [5]

[1] *Department of Toxicology, National Institute of Health, Rabat, Morocco*

[2] *Laboratory Materials and Nanotechnology and the Environment Department of chemistry, Faculty of El Science, Mohammed V University Rabat-Agdal, Morocco*

[3] *PESR unity, Scientific Institute, University Mohammed V, Rabat, Morocco.*

[4] *Department of Biology, University Mohammed V, Rabat, Morocco.*

[5] *Laboratoire des Sciences du Climat et de l'Environnement, LSCE, CE_A-CNRS-UVSQ / IPSL, Gif-sur-Yvette, France*

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Abstract

The atmospheric metal contamination of *Eucalyptus* tree leaves was studied in several urban and rural sites of the Rabat Salé area where traffic density and intense use of metallic compounds in the potteries are major contributors in air pollution. Samples most contaminated with lead, cadmium and zinc were found around the traditional pottery manufactures of Oulja and in downtown station. The results showed that major contamination sources are particle deposits on the leaves and that the ratios of accumulation between leaves and soil are in general lower than one and the values decrease in the order Zn> Cd>Pb. The presented results suggest that *Eucalyptus* leaves are potentially good indicators of metal pollution, contributing to the biomonitoring system for cadmium and lead atmospheric inputs. Three different types of atmospheric pollution sources were inferred from the comparison of the soil and leaves contents, and of the washed and unwashed leaf contents. Due to an important zinc internalisation in *Eucalyptus* leaves, they are not convenient for atmospheric zinc biomonitoring.

Keywords: Biological monitoring; Leaves of *Eucalyptus*; Rabat Salé; Cadmium; Lead; Zinc

*E-mail: barcha.sayf@gmail.com; Tel: (+212624083716)

1. Introduction

The contamination of various ecosystems by polluting agents may cause their transfer through living organisms [1,2]. Due to the persistence of non-biodegradable compounds such as metallic pollutants in the ecosystems and their transfer and uptake toward fauna and flora, biomonitoring methods are used to estimate airborne metallic pollution. Trees have proven to be useful atmospheric pollution biomonitors under dry climates [3]. They may fix metals from the atmosphere through leaves and from soil by roots [4]. Indeed, several authors used the contamination of tree leaves by metallic compounds for the biomonitoring of atmospheric pollution in urban areas [5-10]. The *Eucalyptus* tree was selected to be a convenient bioaccumulator candidate as this species is widely spread in Morocco. It possesses non-deciduous leaves, submitted to a long-time exposure to atmospheric contaminants. *Eucalyptus* trees are used in Morocco as alignment plants on the edges of the road axes inside as well as outside the cities.

In 2004, Moutia (2004) found high metal concentrations in the barks of *Eucalyptus* tree in urban area of Rabat [19]. The lead contents were respectively 34.5 mg/kg (dry weight) in Kamra, 58.3 mg/kg, downtown and 144.3 mg/kg in BouregregneighbouringOulja, close to the most industrialised area in the Rabat Salé city.

ElAbidi (2000) showed high atmospheric lead contents in Rabat [8]. The annual mean concentration for the lead in dust was 1.12 $\mu g/m^3$ with peak concentration exceeding 2.10 $\mu g/m^3$ during rush hour. Concerning the plants, ElAbidi (2000) found the lead contents in Kamra up to 18 mg/kg (dry weight) on *Eucalyptus* and *Populus* leaves, and 51 mg/kg in *Pinus* needles. The comparison between species was difficult as the presence of the three different species on a single site was not found. On the other hand, GhizlaneMazouz et al (2007) carried out a study on the contamination of the lichens in Rabat and surroundings [11]. High lead contents were found on the area of Oulja, where their concentrations exceed 650 mg/kg (dry weight) in the lichen samples.

The purpose of this study was to evaluate contamination by trace metal elements (Pb, Cd and Zn) in urban, industrial and rural areas in and around Rabat-Salé city by using the *Eucalyptus* leaves as biomonitors. A special attention was paid on the Oulja site, which is an industrial complex where many potteries are concentrated. Finally, the role of the *Eucalyptus* leaf in atmospheric pollution biomonitoring was evaluated, by comparing the composition of the local soil and of the leaf.

2. Material and methods

2.1. Sampling sites.

Leaves were collected from *Eucalyptus* trees between December 2006 and March 2007 in ten sites, differing in traffic density and distance to potential punctual sources, such as potteries.

Site 1 (Kamra) is of high traffic density, site 2 (downtown) is of medium density, site 3 (Oulja) has an important industrial activity (potteries) and site 4 (AllalBahraoui), the furthest site from the urban center, is a rural area with the least population density and was considered as reference station in this study (Fig. 1). In addition, samples of *Eucalyptus* leaves were collected from different points close to the Oulja site (S3), in order to evaluate the dispersion of trace elements in this area, taking into account the dominant wind directions that transport the dusts emitted by the potteries. The selected sites around Oulja were: Dar Zamane (S5), Oulja forest (S6), domestic wastes disposal of Salé (S7), nearby fields (S8), Rabat pottery (S9) and Akrach (S10) (Fig. 1).

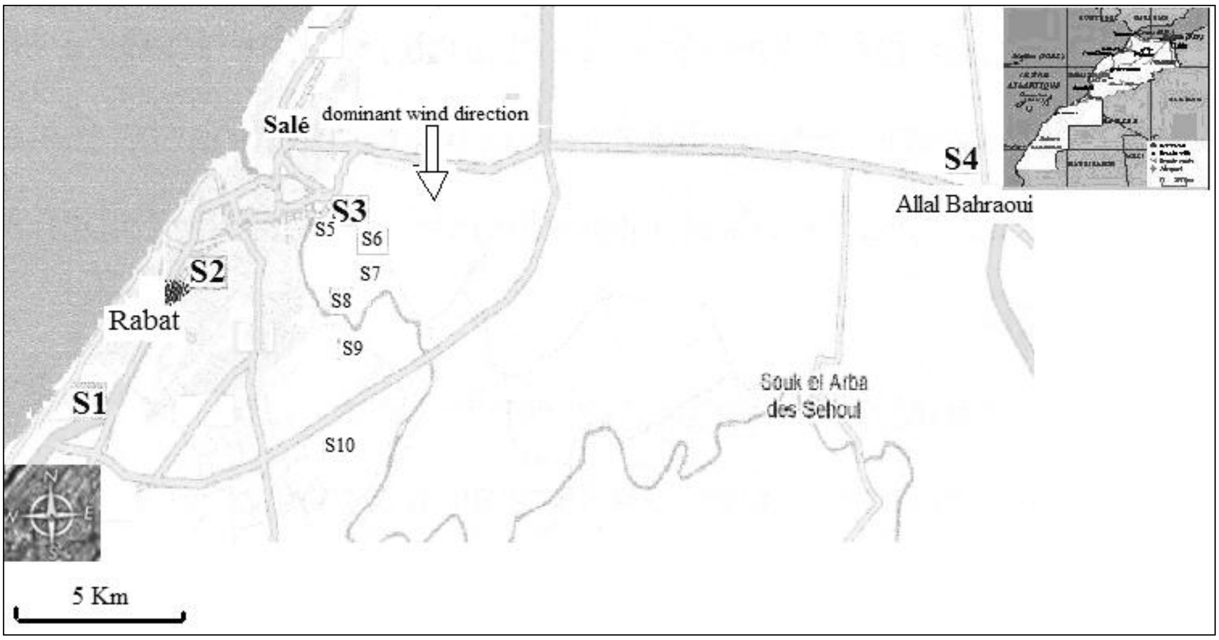

Figure 1: Location of sampling stations in Rabat Salé city

2.2. Sample collection, preparation and analysis.

The 20 cm-long leaves (approx. 3 years old) were collected with powder-free gloves and stored in clean plastic bags during 24 hours in the laboratory at 18°C before preparation for analyses. For each sample, a sub-sample was washed by aspersion with 20 ml of distilled water.

The washed and unwashed leaves were dried at 70°C to constant weight. Three aliquots of approximately 0.5 g of weighted leaves were digested at 120°C during 4 hours with 4 ml of nitric acid Suprapur (65% Merck).

The determinations of Pb and Cd were performed by Graphite Furnace - Atomic Absorption Spectrometer GF-AAS (Varian AA-Z 220); Zn concentrations were measured by flame AAS (Varian AA 20).

All trace elements analyses were performed under an internal quality control program using the reference material DORM-2: Dogfish muscle (NRC, Canada). An external control using an intercalibration exercise based on reference materials (IAEA-0140 (1997), plant and IAEA-433 (2004), marine sediment) was performed. The obtained results were in agreement with the certified values (Table 1).

Comparison between lead, cadmium and zinc contents was realised using t-Student for impaired variables.

Table 1 Results on metal levels of reference materials DORM-2 (n=3)

DORM-2	Certified value mg/kg dw	Experimental valuel mg/kg dw
Pb	0.065±0.007	0.070±0.010
Cd	0.043±0.008	0.048±0.010
Zn	25.6±2.3	24.1±1.4

3. Results

Table 2 presents mean contents and standard deviation for lead, cadmium and zinc in the leaves of *Eucalyptus* in different stations studied in Rabat-Salé.

Table 2 Mean values ± standard deviation of the concentration of lead, cadmium and zinc measured in *Eucalyptus* leaves mg/kg dry weight (n=3) from 10 sites of Rabat-Salé city.

Station reference		N	Lead	Cadmium	Zinc
S1	**Kamra**	3	14.4 ± 0.9	0.14 ± 0.02	16.3 ± 1.5
S2	**Down town**	3	22.4 ± 2.3	0.22 ± 0.02	27.5 ± 1.4
S4	**Allal Bahraoui**	3	1.7 ± 0.6	0.03 ± 0.01	14.8 ± 1.5
S3	**Oulja: Artisanal Complex**	3	23.6 ± 2.3	0.37 ± 0.03	44.3 ± 2.1
S5	**Dar Zamane**	3	6.8 ± 1.3	0.13 ± 0.02	24.0 ± 1.4
S6	**Oulja Forest**	3	4.7 ± 1.2	0.14 ± 0.01	19.9 ± 1.3
S7	**Salé disposal**	3	3.7 ± 0.5	0.11 ± 0.01	23.3 ± 4.4
S9	**Resident fields**	3	2.8 ± 0.8	0.09 ± 0.01	18.2 ± 1.9
S9	**Rabat Potteries**	3	27.0 ± 2.1	0.30 ± 0.02	38.1 ± 1.9
S10	**Akrach**	3	1.8 ± 0.7	0.05 ± 0.01	18.4 ± 1.7

These results show that *Eucalyptus* leaves collected from different stations contain relatively high levels of lead, cadmium and zinc compared to the normal values determined by Kabata-Pendias (1992) on the basis of a worldwide study [13] (Table 3).

Table 3 Normal and toxic values of lead, cadmium and zinc in plants mg/kg dry weight. (Kabata-Pendias, 1992)

	Lead	Cadmium	Zinc
Normal Value	5-10	0.05-0.2	27-150
Toxic Value	30-300	5-30	100-400

3.1. Lead

The highest levels of lead in leaves were found in the artisanal complex of Oulja (S3), near the Rabat potteries (S9) and in downtown station (S2) with respectively 23.6, 27.0 and 22.4 mg/kg of dry weight (dw). There is no significant difference between the concentrations in these three contaminated sites (p=0.56). Lead concentration in the rural area of AllalBehraoui is 1.7 mg/kg dw showing a significantly high variation compare to other stations (p<0.001).

Table 4 pH and mean values of lead, cadmium and zinc in soil samples of Rabat-Salé.

Station reference		N	pH	Lead	Cadmium	Zinc
S1	**Kamra**	3	7.05	19.2±4.7	0.38±0.07	56.4±6.9
S2	**Down town**	3	7.93	68.3±9.4	0.84±0.09	45.1±2.9
S4	**Allal Bahraoui**	3	6.15	2.5±1.3	0.20±0.06	3.3±2.7
S3	**Oulja: Artisanal Complex**	3	8.08	106.1±21.7	0.70±0.05	29.1±6.1
S5	**Dar Zamane**	3	7.05	9.2±2.7	0.51±0.08	42.3±6.6
S6	**Oulja Forest**	3	7.01	9.5±2.4	0.93±0.10	30.4±8.1
S7	**Salé disposal**	3	8.31	29.6±3.2	1.30±0.07	198.5±22.5
S9	**Nearby fields**	3	7.85	30.8±6.8	0.62±0.04	72.4±15.4
S9	**Rabat Potteries**	3	7.35	437.1±115.6	0.75±0.07	72.4±15.4
S10	**Akrach**	3	6.21	32.7±8.9	0.43±0.05	13.2±2.5

The soil metal concentrations were concurrently studied in the same stations (Table 4). The lowest value was found in AllalBehraoui (S4, 2.5 mg/kg dw) and the highest one in Rabat potteries (S9, 437 mg/kg dw), followed by Oulja artisanal complex (S3, 106 mg/kg dw). The very high lead content in soils in the S3 and S9 stations is not proportional to the leaf deposit concentration (23.6 and 27.0 mg/kg dw, respectively). The concentration ratio between the S3 and S9 stations, on one hand, and the reference S4 station, in another hand, is close to 15 for the leaf deposit, and 42 and 175 for the S3 and S9 soils, respectively. The soil lead contamination of the station S3 and S9 seems therefore poorly mobile towards the atmosphere. However, all the stations (S7, S8, S10) surrounding the S9 station (Rabat potteries) show a marked soil contamination, close to 30 mg/kg (12 times the reference S4 soil content), but corresponding to a leaf deposit which is only 1.6 times the concentration registered in the reference S4 station.

The lead contamination of the S2 station (downtown) may be explained by car traffic, as it was observed in a previous study on *Argania* tree barks [3]. The ratio S2/S4 for the leaf deposit is 22.4/1.7=13.2 and 68.3/2.5=27 for the soil, which is significantly lower than the ratio observed in S3 and S9 near the potteries.

3.2 Cadmium

The highest values of cadmium in the leaves of *Eucalyptus* are found in the artisanal complex of Oulja station S3 with 0.37 mg/kg dw. Similar values were observed in the Rabat potteries (S9, 0.30 mg/kg dw). The lowest concentration of Cd are observed in the station of AllalBehraoui (S4) with 0.03 mg/kg dw. The differences between these concentrations are highly significant between this station and the others. When comparing lead and cadmium contamination, the situation seems quite different: the AllalBehraoui station S4 remains the reference with 0.03 mg/kg for the leaf deposit. The ratio between the contaminated sites and the reference station S4 is 7 for S2 (down town) and 11 for S3 and S9 (artisanal complex and potteries) for the leaf deposit. These ratios are comparable to the ratios obtained for lead (13 and 15, respectively). However, the soil ratios for Cd are only 4.2 for S2/S4 and 3.5 and 3.8 for S3/S4 and S9/S4, respectively, a marked contrast with what was observed for lead. Furthermore, the soils of S6, S7 and S8 were heavily contaminated by Cd (mean value 0.95 mg/kg dw) with an average value for the leaves reaching 0.11 mg/kg (ratios S6, S7, S8 to S4 for the leaf deposit: 3.8). The distribution of Cd appears therefore to be dependent on other sources besides potteries or car traffic. Alternative Cd sources have to be sought out. These sources could be the urban wastes, as shown by the contamination levels observed in the waste disposal of Salé (S7).

3.3 Zinc

The highest values of zinc are recorded in the pottery complex Oulja (site S3) while the lowest ones are observed in rural station of AllalBehraoui (S4), 44.3 and 14.8 mg/ kg dw, respectively. However, the enrichment of the contaminated sites (S3, S9) near the potteries only reaches 2.8. Furthermore, the zinc content of the leaf deposit on the down town site S2 is only 1.5 higher than the zinc content observed in the reference S4 site. This suggests: 1) that the internal zinc content of Eucalyptus leaves is high, as expected, and 2) there is no massive airborne Zn contamination enabling to increase significantly the value of Zn concentration in and/or on the leaf. Moreover, Zn estimation in the soil shows a quite typical distribution, differing clearly form the leaf deposit. The most heavily contaminated site is S7, the urban wastes disposal, reaching 199 mg/kg in the soil, in contrast with S4 which exhibited low soil content. The zinc content in the leaf deposit was 4.5 times higher than in the soil in S4 (14.8 and 3.3 mg/kg dw in leaves and soil, respectively), while in S7, the zinc leaf content is 8.5 lower than the soil content. The soil of S9, very heavily contaminated by lead, reached a value of 22 for zinc when compared to the reference S4 site (S9/S4 = 174.8 for lead in soil). Moreover, the spatial distribution of atmospheric zinc contamination seems to differ from those of Cd or Pb. A main zinc soil contamination can be seen specifically in S7 (Salé waste disposal) and seems to originate from an anthropogenic deposit.

4. Discussion

The contents of metals in *Eucalyptus* leaf samples studied here can be classified by taking into account normal and toxic values of metals in the aerial parts of the plants [13] (Table 3). According to these results, although the contents of lead, cadmium and zinc, recorded in the *Eucalyptus* leaves of the urban areas and potteries of Rabat-Salé, exceeded normal values, the contents never reached the toxic values. In the studied sites, these results show how *Eucalyptus* accumulates lead, cadmium and zinc in their tissues. However, these plants were not considered as hyperaccumulator defined as possessing the ability of sampling and sequestration of exceptional concentrations of heavy metals in aerial plant parts under real conditions [23]. Many authors demonstrated that trace element concentrations in aerial parts of a plant are related with heavy metals contents in soil [7-9-25]. Consequently, heavy metals accumulation in plants could be explained by a ratio or accumulation factor (AF) of each metal. This factor, elsewhere named bioconcentration factor [17], is calculated by dividing the concentration of each element in the leaves (mg/kg dw) with the concentration of the element in the soil (mg/kg dw). According to Alloway et al. (1988), Mingorace et al. (2005) and Jung, (1995), this factor is an important and useful indicator of the relative availability of metal uptake from soil to plant [5-18-12]. For Rossini Oliva et al. (2007) the relationship between the concentration of the trace elements in the plants and soil (AF) is an index of the transfer of soil-plants, indicating the plant’s absorption capacity [20]. Ratios higher than 1 indicate that plants are enriched by trace elements from soil (accumulators). Ratios with values around 1 indicate plants which are not influenced by trace element concentrations in soil and ratios lower than 1 show weak absorption of trace elements by plants from soil.

Table 5 Accumulation factor (AF) values for lead, cadmium and zinc in *Eucalyptus* leaves in the area of Rabat Salé (Morocco).

	Station reference	N	Lead	Cadmium	Zinc
S1	**Kamra**	3	0.75	0.37	0.29
S2	**Centre town**	3	0.33	0.26	0.61
S4	**Allal Bahraoui**	3	0.66	0.15	4.50
S3	**Oulja: Complex Artisanal**	3	0.22	0.53	1.52
S5	**Dar Zamane**	3	0.74	0.25	0.57
S6	**Oulja forest**	3	0.49	0.15	0.66
S7	**Discharge Salé**	3	0.13	0.08	0.12
S9	**Nearby fields**	3	0.09	0.16	0.25
S9	**Rabat Pottery**	3	0.06	0.40	0.53
S10	**Aktrach**	3	0.05	0.12	1.39

Indeed, our results (Table 5) showed clearly that contamination sources of plant leaves were not the soil, but most probably the atmospheric dusts on these leaves for lead and cadmium (AF<1), whereas for zinc most of the trace metal elements were up taken from soil to the plant (AF>1). Furthermore, it is observed that zinc is accumulated in the plant of Eucalyptus even in the presence of low soil contents, whereas lead is slightly transferred from soil to the plant even with very high concentrations in the soil. This agrees with the results of Kerein et al. (1972) and Lee et al. (2001) [14-15].

The internal content is defined as the metal content of the washed leaves. The external content was calculated by subtracting the internal content (washed leaves) from the whole content (unwashed leaves). The difference in concentration expressed as a percentage of lead, cadmium and zinc between the external part and the internal part of the Eucalyptus plant (Fig. 2) showed that the proportion of the lead and the cadmium on the particles on the leaf (external part) was higher compared to the internal part.

Thus 65 % of lead and 52 % of cadmium were found in the external part of *Eucalyptus* leaf and only 12.8 % for zinc. According to ElAbidi et al. (2000), it suggested that plants acquire lead coming from the soil and the air [8]. Lead-bearing particles were fixed by leaves and adhered to hydrocarbons and other organic materials on the external part of leaves. Our results show clearly that the major part of the particles of lead and cadmium found in these plants are in the external part of the leaves. We may conclude that the metal content external part of the leaf (i.e. the deposit of the leaves surface) is a good indicator of the atmospheric lead and cadmium pollution. As a non-negligible part of the leaf content is internalised, which can also be attributed to soil uptake, the air pollution biomonitoring for zinc using Eucalyptus species is questionable.

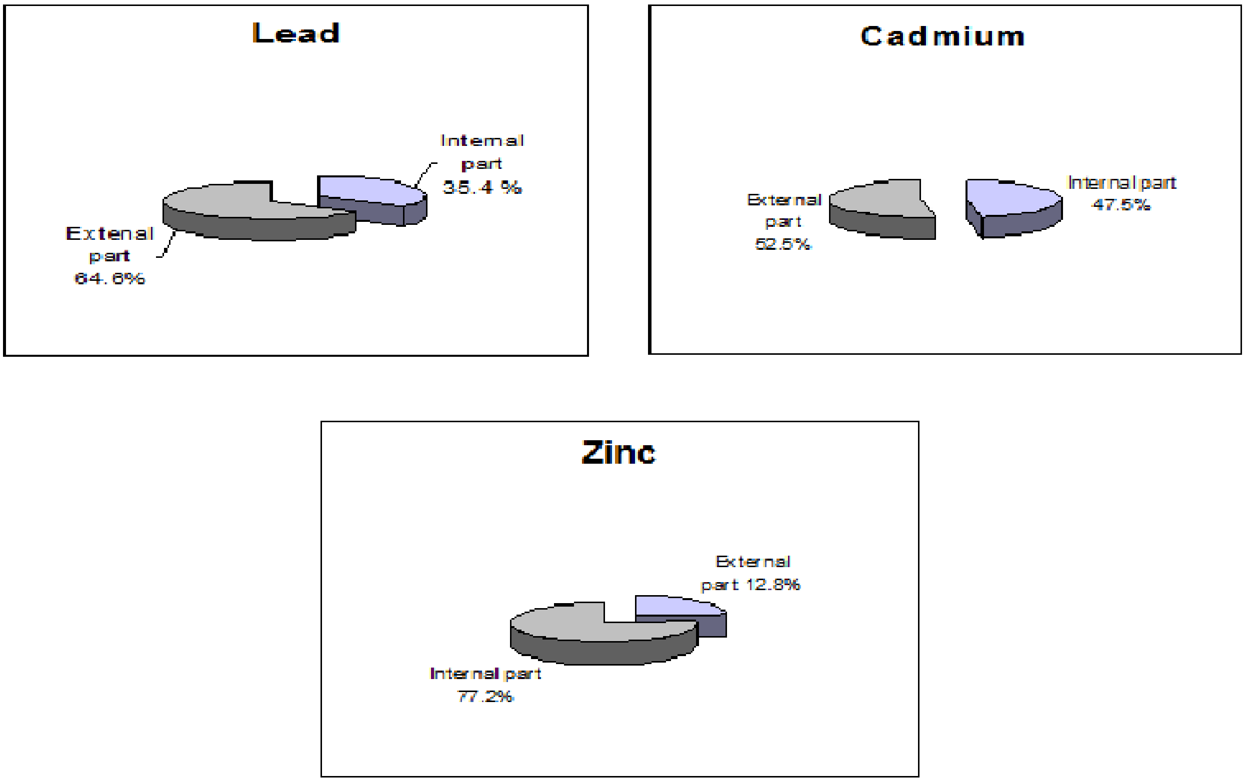

Figure 2: Difference in content lead cadmium and zinc in (%) between the internal and external part of the Eucalyptus.

5. Conclusion

The spatial variation of the metal contamination of *Eucalyptus* trees reveals that concentrations of lead, cadmium and zinc are high in aerial parts of the plant. It was mainly observed for lead close to urban stations and also to the two potteries stations of Rabat-Salé. These concentrations exceeded the normal values in plants for all the metal elements analyzed. This can explain the metal contamination of the vegetation and revealed the impact of potteries emissions and of the road traffic in Rabat-Salé city. The major part of lead and cadmium present in the leaves was issued from dusts deposits, whereas for zinc, most of contamination was issued from the soil. The values of the ratio accumulation of metals in the *Eucalyptus* tree leaves on the level of Rabat-Salé followed this order: Zn > Pb > Cd. The spatial distribution of each of the three studied metals was different between the ten studied stations. Lead seemed to be associated since a long time to car traffic and potteries. The urban waste disposal showed a specific contamination by cadmium and zinc, probably of anthropogenic origin. The zinc air contamination in the reference site suggests that it is a recent event and/or the results of a long-range transport, as the zinc soil concentration is low.

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