



## Human population exposure to a heavy metal contamination (Cd, Cu, Pb and Zn) from a mining site in Marrakech-Morocco

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Received: 16<sup>th</sup> September, 2008 ; Accepted: 21<sup>st</sup> September, 2008

### ABSTRACT

In this work, the epidemiologic study showed that more than 63 % of the working population of this zone works in agriculture. This activity puts directly the population in contact with wastewater and the grounds dust enriched in certain trace elements contents, whose can pass in the human organism by cutaneous way or/and by inhalation. The impregnation of the studied population by the trace elements (particularly children of less than 10 years), is alarming. In addition to the direct contamination by food, these children are not safe from an additional (indirect) contamination by the smoke of the tobacco, their play activities inside and especially outside the habitats associated with a defective hygiene. The study also showed that the habits (use of kohl) play a very significant role in the increase of metal concentration in population, especially in women.

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### KEYWORDS

Trace elements;  
Human blood;  
Hair;  
Exposure;  
Mine area.

### INTRODUCTION

In recent years, there has been an increased awareness in the several countries about the health effects of toxic and other trace elements in relation to nutritional disorders, environmental or occupational exposure, pathological status and medical therapy<sup>[15,62]</sup>.

A variety of human activities, notably industrial and mining process have been responsible for the wider diffusion of heavy metals into the environment (Loumbourdis, 1997).

Several toxic metals, notably Cd and Pb, have been released in large quantities into the environment<sup>[57]</sup> in industrial areas, and large population groups may be

exposed to toxic metals from the polluted environment, as is the case in the district of DARAA LASFAR, located in the North-West of the Mrabatine zone, approximately 10 km to the west of Marrakech city (Morocco).

Thus, exposure to these metals, and other environmental pollutants, are considered as severe environmental pollution and occupational hygiene problems with possible short- and long-term adverse effects on human health.

The threat of heavy metals to human health is aggravated by their long-term persistence in the environment<sup>[29,17]</sup>, they may be transferred and accumulated in the bodies of animals or human beings through food

chain, which will probably include teratogenesis, anomalies in reproduction (Loumbourdis and Wray, 1998, Loumbourdis et al., 2006), cause DNA damage and carcinogenic effects by their mutagenic ability (Das, 1997; Knasmuller et al., 1998; Mc Laughlin et al., 1999, Yang et al., 2005).

Biological monitoring of the general population and of groups considered at risk provides essential information for both the evaluation of exposure and risk characterization. In this study we measured the concentrations of Cd, Cu, Pb and Zn in blood and hair of 'healthy' subjects (men, women, girls and boys) using analytical methods based on electro-thermal atomic absorption spectrometry.

In this paper, we present the preliminary results of this survey and evaluate them in the light of data obtained in a group of healthy subjects (controls) from a rural area, and data previously reported in the literature for the general population living in other countries.

## METHOD AND MATERIAL

### Sampling

Blood samples were collected from 67 subjects. The majority of them was born and/or lived at least one year on the spot. They are distributed as following:

- 26 men of more than ten years.
- 18 women of more than ten years.
- 16 boys of less than ten years.
- 8 girls of less than ten years.

All the subjects receive the same food ration composed especially on local animal foodstuffs and vegetables irrigated from the same source: the Tensift River receiving the Drâa Lasfar mine wastewater.

Control blood samples were collected from 30 subjects from a rural area. The rural area selected was at approximately 35Km in the southern part of the Marrakech region (Amzmiz region), to minimize the possible influence of atmospheric deposition from the industrial region (the prevailing wind direction in Marrakech is from the northwest).

Before sampling blood, a questionnaire was managed with both population groups in order to collect variables relating to the individual factors likely to influence the rate of the elements traces in blood (and

hair) and the variables relating to the various factors of studied risks.

Sampling blood was carried out by puncture after careful disinfection using the cotton soaked with surgical alcohol. Approximately 5 ml of blood were collected aseptically with needles of single use and stored in heparinised tubes. Tubes are numbered and placed in a refrigerator.

Approximately 10 g of hair of each subject were taken on from the nape of the neck then placed in plastic bags and numbered.

### Preparation of samples

All samples obtained from the smokers and from the alcohol drinkers were eliminated. The literature has reported that these two factors can intervene in the evolution of the metal concentrations.

### Blood

Approximately 2 ml of blood sub-samples were dried to constant weight at 85°C; [6,42,43,52,66,67,46]. Dried samples were cold digested in 1.5 ml of concentrated nitric acid overnight<sup>[46]</sup>.

### Hair

The measurement of trace elements in hair is not without its own inherent problems. In fact, analysis must only take into account the internal (endogenous) fraction [396,61] which highlights the importance of removing the external (exogenous) contaminant fraction coming from the metal-rich dust deposited on the hair<sup>[60,61]</sup>.

The hair procedure washing was carried out as it's described by several authors<sup>[15]</sup>: nitric acid (10%) 4 × 10 min.

Approximately 2g sub-samples were dried to a constant weight at 80°C. A precisely weighed 50 mg test specimen was carefully dried at constant temperature to constant weight. This specimen was introduced into a polystyrene tube, and 1ml nitric acid 8N was added<sup>[42]</sup>.

The corked tube was preserved at ambient temperature for 24h. During this preliminary phase, most of the sample dissolved in the acid. To perfect dissolution, the corked tubes were placed for 1h in a boiler at 60°C<sup>[42]</sup>. Corks were maintained in place by pressure (plate plus weight).

The recovered liquids were diluted in a suitable

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amount of bi-demineralised water for trace element analysis.

### Method validation

Since the quality of analytical results is an essential pre-requisite for the assessment of health risks, methods were validated in-house according to the EURACHEM guideline<sup>[26]</sup> and the requirements of ISO/IEC 17025<sup>[32]</sup> and ISO 15189<sup>[31]</sup>, including estimates of uncertainty of measurement according to the EURACHEM/CITAC Guide<sup>[26]</sup>.

### Analysis

Trace metal concentrations were determined by flame atomic absorption spectroscopy for Copper and zinc, and by graphite furnace atomic absorption

spectroscopy for cadmium and lead. Metal concentrations were expressed as mean individual values  $\pm$  standard deviation.

## RESULTS

Descriptive statistics on blood and hair concentrations of cadmium, copper, lead and zinc analysed in the 67 subjects from the mining area and in the 30 subjects from a rural area are shown in TABLES 1-4.

Results show that mining area population has significantly higher trace elements levels than rural area one in both blood and hair ( $p < 0.05$ ).

### Trace elements in hair

155 samples of hair and 67 samples of blood were

**TABLE 1: Cd, Cu, Pb and Zn blood levels in mining population and statistical study of influence of age on their accumulation**

Element	Sex	IP		AP		F	test	Z	t	Conclusion
		M	SD	M	SD					
Cd ( $\mu\text{g/l}$ )	F	1.95	1.11	1.76	1.06	1.09	t	0.25	2.02	H <sub>0</sub>
	M	1.28	0.81	1.67	0.83	1.07	t	1.56	2.07	H <sub>0</sub>
Cu ( $\mu\text{g/l}$ )	F	1.46	0.27	1.33	0.48	1.55	t	0.47	2.02	H <sub>0</sub>
	M	1.59	0.34	1.55	0.42	3.24	t	0.08	2.02	H <sub>0</sub>
Pb ( $\mu\text{g/l}$ )	F	186.17	28.41	226.31	57.27	3.66	NA	1.27	2.02	H <sub>0</sub>
	M	194.00	36.46	194.68	75.39	4.27	NA	0.04	2.07	H <sub>0</sub>
Zn ( $\mu\text{g/l}$ )	F	3.69	0.87	4.00	0.85	1.05	t	0.55	2.02	H <sub>0</sub>
	M	3.37	0.86	3.71	1.06	1.50	t	1.14	2.07	H <sub>0</sub>

IP: Infantile population; AP: Adult population; F: Females; M: males

**TABLE 2: Cd, Cu, Pb and Zn blood levels in mining population and statistical study of influence of sex on their accumulation.**

Element	Sex	F		M		F	test	Z	t	Conclusion
		M	SD	M	SD					
Cd ( $\mu\text{g/l}$ )	AP	1.76	1.06	1.67	0.83	1.61	t	0.36	2.02	H <sub>0</sub>
	IP	1.95	1.11	1.28	0.81	1.89	t	0.97	2.09	H <sub>0</sub>
Cu ( $\mu\text{g/l}$ )	AP	1.33	0.48	1.55	0.42	1.31	t	1.66	2.02	H <sub>0</sub>
	IP	1.46	0.27	1.59	0.34	1.58	t	0.38	2.09	H <sub>0</sub>
Pb ( $\mu\text{g/l}$ )	AP	226.31	57.27	194.68	75.39	1.92	t	0.63	2.02	H <sub>0</sub>
	IP	186.17	28.41	194.00	36.46	1.65	t	0.32	2.09	H <sub>0</sub>
Zn ( $\mu\text{g/l}$ )	AP	4.00	0.85	3.71	1.06	1.57	t	0.98	2.02	H <sub>0</sub>
	IP	3.69	0.87	3.37	0.86	1.01	t	0.51	2.09	H <sub>0</sub>

IP: Infantile population; AP: Adult population; F: Females; M: males

**TABLE 3: Cd, Cu, Pb and Zn hair levels in mining population and statistical study of influence of age on their accumulation.**

Element	Sex	AP		IP		F	test	Z	t	Conclusion
		M	SD	M	SD					
Cd ( $\mu\text{g/g}$ )	F	1.91	0.64	1.76	0.41	2.46	t	0.40	2.02	H <sub>0</sub>
	M	2.03	0.77	1.69	0.86	1.23	t	1.42	2.02	H <sub>0</sub>
Cu ( $\mu\text{g/g}$ )	F	23.36	1.86	21.22	3.43	3.41	NA	1.26	2.13	H <sub>0</sub>
	M	23.07	5.90	26.24	3.26	3.27	t	0.12	2.02	H <sub>0</sub>
Pb ( $\mu\text{g/g}$ )	F	22.44	6.37	17.06	5.76	1.26	t	1.34	2.02	H <sub>0</sub>
	M	19.36	5.67	15.01	7.18	1.61	t	2.17	2.02	H <sub>1</sub>
Zn ( $\mu\text{g/g}$ )	F	157.48	37.55	161.06	48.20	1.65	NA	0.13	2.02	H <sub>0</sub>
	M	159.56	57.94	158.68	27.46	4.45	t	0.07	2.01	H <sub>0</sub>

IP: Infantile population; AP: Adult population; F: Females; M: males

**TABLE 4: Cd, Cu, Pb and Zn hair levels in mining population and statistical study of influence of sex on their accumulation**

Element	Sex	F		M		F	test	Z	t	Conclusion
		M	SD	M	SD					
Cd (µg/l)	AP	1.91	0.64	2.03	0.77	1.48	t	1.24	2.02	H <sub>0</sub>
	IP	1.76	0.41	1.69	0.86	4.47	NA	0.35	2.02	H <sub>0</sub>
Cu (µg/l)	AP	23.36	1.86	23.07	5.90	10.09	NA	0.24	2.03	H <sub>0</sub>
	IP	21.22	3.43	26.24	3.26	1.11	t	2.01	2.07	H <sub>0</sub>
Pb (µg/l)	AP	22.44	6.37	19.36	5.67	1.27	t	1.71	2.02	H <sub>0</sub>
	IP	17.06	5.76	15.01	7.18	1.60	t	0.47	2.07	H <sub>0</sub>
Zn (µg/l)	AP	157.48	37.55	159.56	57.94	2.38	NA	0.14	2.02	H <sub>0</sub>
	IP	161.06	48.20	158.68	27.46	3.08	t	0.10	2.07	H <sub>0</sub>

IP: Infantile population; AP: Adult population; F: Females; M: males

analyzed.

However, all the results were not exploitable, so we eliminated some values those were obviously too high and were apart from an acceptable zone of variation and as a consequent were representing either a particular exposure to metals or a contamination during the preparation of the samples.

### Cadmium

Statistical analysis of the obtained results shows that the mean Cd contents found in the men ( $2.03 \pm 0.77 \mu\text{g/g}$ ) living in mining zone are higher than those found in women ( $1.91 \pm 0.64 \mu\text{g/g}$ ). These two values are not significantly different ( $P < 0.05$ ) but these are significant with reference to the mean contents noted in the control population,  $0.04 \pm 0.15 \mu\text{g/g}$  and  $0.43 \pm 0.12 \mu\text{g/g}$  in men and women respectively.

Moreover, this statistical analysis equally shows that the girls of age less than 10 years accumulate more Cd ( $1.76 \pm 0.41 \mu\text{g/g}$ ) in their hairs compare to the boys ( $1.69 \pm 0.86 \mu\text{g/g}$ ), although the difference between the two values is not significant ( $P < 0.05$ ).

In all the studied cases (mining and rural populations), the statistical analysis did not highlight the effects of age and sex on the accumulation of the Cd in the hair of individuals.

Several authors have made different observations for the populations living in the neighbourhood of deserted mines. In their study, Zumkley et al.<sup>[81]</sup> have found that Cd cadmium contents are higher in men ( $8.2 \mu\text{g/g}$ ) than in women ( $6.4 \mu\text{g/g}$ ). Same has been noted by Medeiros and Pellum<sup>[50]</sup>  $9.5 \mu\text{g/g}$  vs  $7.9 \mu\text{g/g}$  in the USA and Nishiyama and

Nordberg<sup>[56]</sup>  $6.9 \mu\text{g/g}$  vs  $4.5 \mu\text{g/g}$  in Japan. The Cd concentrations obtained in our study are less than to those noted in USA and in Japan, but are higher than to

those noted by Pereira et al.<sup>[59]</sup> in Portugal for the population living in the vicinity of an unused Cu mine ( $0.89 \mu\text{g/g}$  in men and  $0.62 \mu\text{g/g}$  in women), by Barlow and Al<sup>[2]</sup>  $0.22 \mu\text{g/g}$  observed in general population in Great Britain and by Leotsinidis and Kondakis<sup>[44]</sup>  $0.19 \mu\text{g/g}$  in Greece. Remembering that the contents level noted in our study remain inferior to the reference values cited in by Iyenger and Woittiez<sup>[34]</sup> of the order of  $2.4 \mu\text{g/g}$ .

### Copper

The statistical analysis of the results showed that the difference between mean Cu contents in the two sexes hair ( $23.1 \pm 5.9 \mu\text{g/g}$  at the men and  $23.4 \pm 1.9 \mu\text{g/g}$  among women) is not significant ( $P < 0.05$ ). Thus, these mean contents remained higher than those noted in the rural population ( $11.1 \pm 1.6 \mu\text{g/g}$  at the men and  $12.9 \pm 1.8 \mu\text{g/g}$  among women). The same report was made in children.

The obtained levels in our study are lower than those found by Suzuki et al.<sup>[71]</sup> ( $46 \mu\text{g/g}$ ) in exposed female population in Japan and those noted by Jamall and Allen<sup>[35]</sup> ( $30.7 \mu\text{g/g}$ ) in a total population in Pakistan.

However, they are lower than those published by Gibson<sup>[29]</sup> ( $14.9 \mu\text{g/g}$ ) in a general population) and by Leotsinidis and Kondakis<sup>[44]</sup> ( $10.6 \mu\text{g/g}$  in women and  $10.5 \mu\text{g/g}$  in men).

### Lead

Statistical analysis of the results obtained showed that contrary to cadmium, the mean Pb contents are higher in female hairs than those of the males. Thus, the Pb contents noted respectively in women and girls ( $22.4 \pm 6.4 \mu\text{g/g}$  and  $17.1 \pm 5.7 \mu\text{g/g}$ ) are higher than those observed in men and boys ( $19.4 \pm 5.7 \mu\text{g/g}$  and  $15.0 \pm 7.2 \mu\text{g/g}$ ) respectively. However, the difference is not



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significant ( $P < 0.05$ ). Moreover, the contents noted in the two sexes are largely higher than those of the rural population ( $4.0 \pm 1.3 \mu\text{g/g}$  in women and  $5.3 \pm 1.2 \mu\text{g/g}$  in men). A significant difference was noted between girls and women (TABLE 3).

The Pb contents noted in this study are higher than those published by Waqar and al.<sup>[77]</sup> in Pakistan in a total population ( $10.6 \mu\text{g/g}$ ) and by Leotsinidis and Kondakis<sup>[44]</sup> for the population in Greece ( $4.4 \mu\text{g/g}$ ). However, they are lower than those noted by other authors; in the United States,  $54 \mu\text{g/g}$  in women and  $23 \mu\text{g/g}$  in men<sup>[50]</sup>, in Venezuela,  $49.9 \mu\text{g/g}$  in a total population exposed to Pb<sup>[10]</sup> and in Pakistan ( $31.7 \mu\text{g/g}$ ) in a total population<sup>[35]</sup>.

A technical explanation can be used to justify this increase in the Pb contents in the women hairs in our study. In fact, the samplings of hairs were carried out on the last centimetres at the end of hairs which remained a long time exposed to a contamination<sup>[12,55]</sup>. The studies undertaken by Sedki<sup>[66]</sup> highlighted that the Pb contents in the hair increases gradually from the hair root to the point. Moreover, the age of a hair segment is proportional to the distance which separates it from the scalp, more a segment is far from the root more it remained a long time exposed to the contaminants (sweat, water, cosmetics, airborne dusts...) which supports its impregnation by these elements traces<sup>[66]</sup>.

### Zinc

Statistical analysis of the obtained results shows that the mean Zn contents found in the men of the mining zone ( $159.6 \pm 57.9 \mu\text{g/g}$ ) are higher than those found in women ( $157.5 \pm 37.5 \mu\text{g/g}$ ), but the difference is not significant ( $p < 0.05$ ). However, these Zn contents are higher than those of the rural population ( $96.8 \pm 9.6 \mu\text{g/g}$  in men and  $104.2 \pm 17.2 \mu\text{g/g}$  in women). The same report was made in children.

Moreover, the study of the age effect on the accumulation of zinc in the hair revealed that there is no significant difference between the mean contents of this element in children and in adults hair ( $p < 0.05$ ).

The absence of significant difference between the two sexes (absence of a sex effect on the accumulation of zinc in hair) for this metal was noted by several authors<sup>[65]</sup>. The Zinc levels, which we noted, were lower than those published by Waqar and al.<sup>[77]</sup> in Pakistan

( $254.4 \pm 16.6 \mu\text{g/g}$  in men and  $236.1 \pm 22.5$  in women) and by Leotsinidis and Kondakis<sup>[44]</sup> ( $182.5 \mu\text{g/g}$  in men and  $187.5 \mu\text{g/g}$  in women) in Greece.

### Trace elements in blood

#### Cadmium

Contrary in the hair, blood cadmium levels (CdB) in women ( $1.76 \pm 1.06 \mu\text{g/l}$ ) were higher than those in men ( $1.67 \pm 0.84 \mu\text{g/l}$ ), but the difference was not significant ( $P < 0.05$ ). On the other hand, a significant difference was noted between the studied zone and rural areas ( $0.71 \mu\text{g/l}$  and  $0.73 \mu\text{g/dl}$  respectively in men and women).

For infants, a higher Cd level was noted in girls blood ( $1.95 \mu\text{g/l}$ ) compared to the boys ( $1.28 \mu\text{g/l}$ ). No significant difference was noted between these levels ( $p < 0.05$ ).

The obtained Cd levels in this study were lower than those published by Maravelaias et al.<sup>[47]</sup> in an exposed Greek population ( $3.7 \mu\text{g/l}$  in men and of  $3.4 \mu\text{g/l}$  in women), by Bergomi et al.<sup>[4]</sup> in an Italian general population ( $4.7 \mu\text{g/l}$ ), by Senft et al.<sup>[68,69]</sup> (CdB =  $5.9 \mu\text{g/l}$ ) in a total population in Czech, and by Koreckova<sup>[37,38]</sup> in the same population ( $2.36 \pm 0.26 \mu\text{g/l}$ ). However, our values were higher than those noted by Kowal et al.<sup>[40]</sup> in USA population ( $1.1 \mu\text{g/l}$  in men and  $0.7 \mu\text{g/l}$  in women). Remembering that the contents level noted in our study remain inferior to the reference values ( $0.3$ - $0.7 \mu\text{g/dl}$ ) cited in by Iyenger and Woittiez<sup>[34]</sup>.

#### Copper

Normal values of copper in blood (CuB) vary between  $0.8$  to  $1.4 \text{ mg/l}$ <sup>[33]</sup>.

Statistical analysis of the obtained results shows that the mean Cu contents found in the studied population are higher than those found in rural population.

Moreover, men and boys present higher contents ( $1.6 \pm 0.7 \text{ mg/l}$  and  $1.6 \pm 0.3 \text{ mg/l}$ ) than those in women and girls ( $1.3 \pm 0.5 \text{ mg/l}$  and  $1.5 \pm 0.3 \text{ mg/l}$ ). No significant difference was noted by the statistical tests.

These CuB values are higher than those reported by Cui et al.<sup>[17]</sup> in two Chinese exposed populations ( $1.06 \text{ mg/l}$  and  $0.89 \text{ mg/l}$  in men,  $1.02 \text{ mg/l}$  and  $0.90 \text{ mg/l}$  in women), by Dlhopolcek and Laurincova<sup>[22]</sup> in a total population in Slovak ( $0.94 \text{ mg/l}$ ), by Vlcek et al.<sup>[74]</sup> in Czech ( $1.3 \text{ mg/l}$ ) and by Burguera et al.<sup>[10]</sup> ( $1.0 \text{ mg/l}$

in women and 1.4 mg/l in men) in a population in Venezuela.

### Lead

In the balance state, the blood lead (PbB) represents 2% of the total lead quantity present in the organism<sup>[13,16]</sup>.

Contrary to the results of many studies (higher PbS in men than in women), our results confirmed the reverse, with  $226.3 \pm 57.3 \mu\text{g/l}$  and  $186.2 \pm 28.4 \mu\text{g/l}$  in women and girls blood respectively, whereas those noted in men and boys are only  $194.7 \pm 75.4 \mu\text{g/l}$  and  $194.0 \pm 36.5 \mu\text{g/l}$  respectively.

Statistical analysis of these results highlighted that neither sex nor age effect on the accumulation of this element in blood. Moreover, these Pb levels are higher than those in the rural subjects ( $40.0 \pm 4.9 \mu\text{g/l}$  and  $38.0 \pm 2.3 \mu\text{g/l}$  in men and women respectively).

Our results are higher than those published by Koreckova and Skopkova<sup>[37,38]</sup> in a Czech population exposed to metal risks (respectively 71.3 and 42.7  $\mu\text{g/l}$  in men and women), by Bittnerova<sup>[5]</sup> in a Czech population (165,8  $\mu\text{g/l}$ ) and by Roels and Lauwerys<sup>[64]</sup> in a Belgian population (163  $\mu\text{g/l}$  at the men and 114  $\mu\text{g/l}$  among women).

However, our results are less than those noted by Vysckocil et al.<sup>[75]</sup> in a Czech population (296.3  $\mu\text{g/l}$ ), by Kriz and Kodl<sup>[41]</sup> 267  $\mu\text{g/l}$  in boys and 255,9  $\mu\text{g/l}$  in girls respectively.

### Zinc

Our results showed that women and girls present higher zinc contents in blood (ZnB) ( $4.0 \pm 0.8 \text{ mg/l}$  and  $3.7 \pm 0.9 \text{ mg/l}$  respectively) than men and boys ( $3.7 \pm 1.1 \text{ mg/l}$  and  $3.4 \pm 0.9 \text{ mg/l}$  respectively).

These values were largely higher than those published by Cui et al.<sup>[17]</sup> in two exposed populations in China (1.18 and 1.12  $\text{mg/l}$  in men and 1.11 and 1.14  $\text{mg/l}$  in women), by Burguera et al.<sup>[10]</sup> (ZnB = 0.98  $\text{mg/l}$ ) in a total population in Venezuela, by Lepsi et al.<sup>[10]</sup> (ZnB = 1.1  $\text{Mg/L}$ ) in a male population in Czech, by Derzsiova et al.<sup>[20]</sup> (ZnB = 1.1  $\text{mg/l}$ ) in a total population living in a mine zone in Slovak, by Kosut<sup>[39]</sup> (ZnB = 1,1  $\text{mg/l}$ ) in an exposed total population in Czech. Dastyh et al.<sup>[18]</sup> have noted higher values for the same population, but remain lower than those noted in our study (ZnB = 1,4  $\text{mg/l}$ ). However, these are lower than those found

by Astrug et al.<sup>[1]</sup> ( $6.9 \pm 3.7 \text{ mg/l}$ ), Djingova<sup>[21]</sup> ( $8.5 \pm 0.8 \text{ mg/l}$ ), Dover et al.<sup>[23]</sup> ( $6.6 \text{ mg/l}$ ) and by Sedki<sup>[66]</sup> ( $6.2 \pm 2.5 \text{ mg/l}$  in women and  $8.5 \pm 3.2 \text{ mg/l}$  in men).

The obtained values in our study remained in general inferior to the reference values (6 to 7  $\text{mg/l}$ ) cited by Iyengar<sup>[33]</sup>.

## DISCUSSIONS

The evaluation of the risks constitutes today a major tool for decision making regarding management of the environmental situations.

At international level, several ecotoxicological researches reported that the use of wastewater to irrigate arable lands contributes to considerable concentration of metals in grounds<sup>[79,11,28]</sup>.

Several works<sup>[53,27,49]</sup> reported that surface and underground watery ecosystems those are enriched in metal contents, cause the contamination of the agricultural soils<sup>[14,24]</sup>, of the spontaneous higher plants<sup>[73]</sup> and of local fauna: earthworms, birds, bovines and sheep<sup>[66]</sup>. The results of this work indeed shows that there can be a real transfer of mineral micropollutants through the polluted soils towards the links of the trophic chains to reach the Man finally, living sedentarily in these zones, via the food products, animals and vegetables<sup>[42,24,66]</sup>. In this context, several authors<sup>[43,30,66]</sup>, underlined in their works that the impregnation of human body by the trace elements does not depend only on the transfer of these micropollutants through the polluted soils via the foodstuffs to the Man, but also on the sex of the individual, his age, his professional statute, his passive nicotinism<sup>[43]</sup>; and on the factors related to the habitat and its situation compared to the mine<sup>[30]</sup>.

The results of the trace elements measuring in hair show that the mean contents of Cd (as out of Cu) are higher in male sex than in female sex. These results perfectly coincide with those of several works<sup>[66,59]</sup>. This report can be justified by the nature of their activities (more than 63% are farmers), of their frequent contact with the ground and wastewater and of their physical efforts required causing an increase in the organism metabolism and consequently an increase in the quantity of introduced contaminated food.

In blood, the Cd concentrations obtained in men's hair are contrary less than in women. This light increase

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in women can be explained by a longer biological half-life of Cd than in the men<sup>[9]</sup>. This great capacity of conservation of blood Cd could be due to a more effective synthesis of metallothioneic proteins in women<sup>[78]</sup>.

Contrary to Cd, the results obtained show that the mean contents of Cu in blood are higher in men than in women. These results coincide perfectly with work of several authors<sup>[66,45]</sup>, which justify this difference by the fact that 90 to 95 % (according to the authors) of trace elements in blood are related to red blood corpuscles.

The number and the volume of red blood corpuscles play a main role in the fluctuation within each individual. However, precisely the age and the sex induce significant modifications, either through the number of red blood corpuscles or through their mean volume. Several authors confirmed that the number of red blood corpuscles is lower by 10 % in women<sup>[66]</sup>.

Contrary to Cd, the Pb content is higher in both women hair and blood. Several explanations are possible: in addition to the direct contamination by the food and contact with wastewater and the grounds dust, women of this zone have particular habits to treat diseases or to make up themselves. They use mineral, vegetable and animals products which are sometimes very rich in certain trace elements like henna and especially kohl (cosmetic eyepiece). This last, is made of at least 65 % of lead<sup>[36,42,43,66]</sup> and its preparation utilizes other minerals, vegetables or animal substances which make its use dangerous (antimony, arsenic, mercury, silver, ginger, musk, camphor, crushed cores of olive, opium, birds bile, viper extract ...). The list of the products using in the preparation of kohl is very long and increasingly complex according to the product finality.

Nevertheless, this cosmetic has a great reputation within the population thanks to its supposed therapeutic virtues. Moreover, the use of this product is dangerous because in addition to the cutaneous contamination, women are accustomed to soaking Miroued (instrument of make-up) by saliva before the application of Kohl, method which permits to the metal to pass in the body by oral way. This particular risk is significant because women use this cosmetic at least three times per week and as of their youth.

It should be stressed that the quantity of cadmium and lead in children hair especially those of less than ten

years (child not yet in school) appears as high as in the other age groups. This could be explained by defective hygiene, children behaviour particularly during their activities inside the habitats notably consumption of fragments of wall linings, fallen on the ground or snatches from the walls. Nevertheless, for this last assumption, the investigation revealed that the majority of the houses (in the two villages) are built with soil and are bleached inside with lime, which could limit this risk. The behaviour outside the habitat also has a significant role in this contamination. Children spend a long period in the day to play in neighbourhoods of the two villages, which puts them in direct contact with wastewater and dust of the grounds charged in metal trace elements. Moreover, they have a behaviour of hand-mouth 'Pica syndrome' which possibly worsens this pollution<sup>[43]</sup>.

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