

## **Influence of Gender and Smoking Habit on the Trace Elements Levels of Washed Scalp Hair of a Control Population from Karbala, Iraq**

Joda A. Baker\* and Neil I. Ward\*\*

\*Chemistry of Department, College of Science, University of Karbala, \*\* ICP-MS Facility, Chemical Sciences, Faculty of Health and Medical Sciences, Guildford, Surrey, GU2 7XH  
(\* author for correspondence n.ward@surrey.ac.uk)

### **Abstract**

Hair samples (n=236) (control or healthy) individuals were collected from Karbala, a city in south-western Iraq. The study population consisted of males (n=196) and females (n=40). All cases were subdivided according to smoking habits (non and active) so as to compare the levels of trace elements in scalp hair in relation to smoking habits. V, Mn, Co, Cu, Zn, Sr and Cd levels in washed scalp hair were measured by inductively coupled plasma mass spectrometry (ICP-MS). The validity and accuracy of the methodology were evaluated by using a certified reference material GBW 09101 Human Scalp Hair with an acceptable range for elemental recoveries ranging from 90 to 107 %. The results obtained showed significantly higher mean level ( $\mu\text{g/g}$  dry. weight) of Sr (11.58) in the scalp hair when compared with the reference range values for control or healthy individuals reported in different countries (0.06 – 6.31  $\mu\text{g/g}$ ). It was found that the mean values of Sr and Co were significantly higher in females than males, whilst the levels of V, Mn, Cu, Zn and Cd were similar (at a probability level  $p = 0.05$ ). Hair of smokers contained significantly more V, Sr and Cd than the hair of non-smokers (at  $P = 0.05$ ). The levels of Mn, Co, Cu and Zn were similar in both sub-groups of smoking activity (at  $P = 0.05$ ).

**Keywords:** Scalp hair analysis. Trace elements. Karbala, Iraq. ICP-MS. Smoking habits

### **الخلاصة**

نماذج من الشعر (n = 236) تم جمعها من أفراد أصحاء من مدينة كربلاء التي تقع جنوب غرب العراق. الأفراد الذين شاركوا في هذه الدراسة كانوا ذكور (n = 196) وإناث (n = 40). هذه الحالات قُسمت إلى مجموعتين مدخنين وغير مدخنين وذلك لمقارنة مستويات العناصر النزرة في شعر فروة الرأس فيما يتعلق بالتدخين. تراكيز كل من Cd, Sr, Zn, Cu, Co, Mn, V في نماذج الشعر المغسول تم قياسها باستخدام تقنية البلازما المقترنة بالحث – طيف الكتلة (ICP-MS). جرى تقييم دقة الطرق المستخدمة باستعمال مادة شعر رأس الإنسان المرجعية – القياسية GBW 09101 حيث كان مدى الأسترداد المئوي يتراوح بين 90 و 110%. تشير النتائج إلى ارتفاع تركيز عنصر Sr في نماذج الشعر المقاسة عند مقارنتها بالمديات القياسية في أقطار أخرى (0.06 – 6.31  $\mu\text{g/g}$ ). لقد وجد أن قيم المتوسط الحسابي لعناصر Co, Sr كانت أعلى بكثير في الإناث إذا ما قورنت بالذكور، بينما كانت هذه القيم متشابهة في كلا الجنسين بالنسبة لعناصر V, Mn, Cu, Zn, Cd عند  $P = 0.05$ . إن نماذج الشعر للأفراد المدخنين تميزت بتراكيز مرتفعة لعناصر Cd, Sr, V إذا ما قورنت بالأفراد غير المدخنين عند  $P = 0.05$ . إن تراكيز كل من Zn, Cu, Co, Mn كانت متشابهة إلى حد ما في كل من الأفراد المدخنين وغير المدخنين.

### **Introduction :**

In the last century, human scalp hair has widely been used as a good biomarker in the assessment of exposure to various pollutants in an occupational and/or environmental setting and in terms of assessing the metabolic state of humans, for essential and toxic trace elements [1 - 6]. Trace elements analysis of human scalp hair has several advantages over blood and urine, including easily sampled, potentially represents a long-term growth material so may provide useful data in determining the health status of an individual for long periods, and several trace elements may be accumulated in hair fibers [2, 7]. On the other hand, the analysis of human fluids such as blood and

## The First Scientific Conference the Collage of Sciences 2013

urine is accompanied by several problems, including analysis the composition at the time of sampling and may trace elements levels are regulated by homeostatic [8, 9]. Many authors have reported that hair is a good indicator to assess the levels of essential and non-essential elements in terms of human health [1, 3]. In terms of this advantage, the concentration of these elements can be used to investigate: (1) the dietary intake of trace elements, especially for non-essential or 'toxic' elements; (2) environmental exposure from anthropogenic sources including chemical pollutants that are released into the environment [10]; (3) the relationship with smoking activity (non, passive and active); and (4) any possible link between specific trace elements and diseases, such as diabetes (type 2) [11, 12]. To date, no studies have been published on the use of human hair material in Iraq; therefore, this study will be valuable in establishing a database of elemental levels for comparison with other countries and for evaluating for future environmental pollution or health disease studies in Iraq. The aim of this study is to investigate the composition of scalp hair of a population living in Karbala and to evaluate any variation in hair trace elements content based on gender and smoking habit.

### Materials and methods :

#### Sample Population

Human scalp hair samples were collected from healthy individuals living in Karbala, a city in Iraq located about 100 Km southwest of Baghdad (Fig. 1). The population statistic are reported for (control or healthy) individuals. The study was approved by the Ethics Committee of the University of Surrey, Guildford (EC/2009/15/FHMS) (United Kingdom).

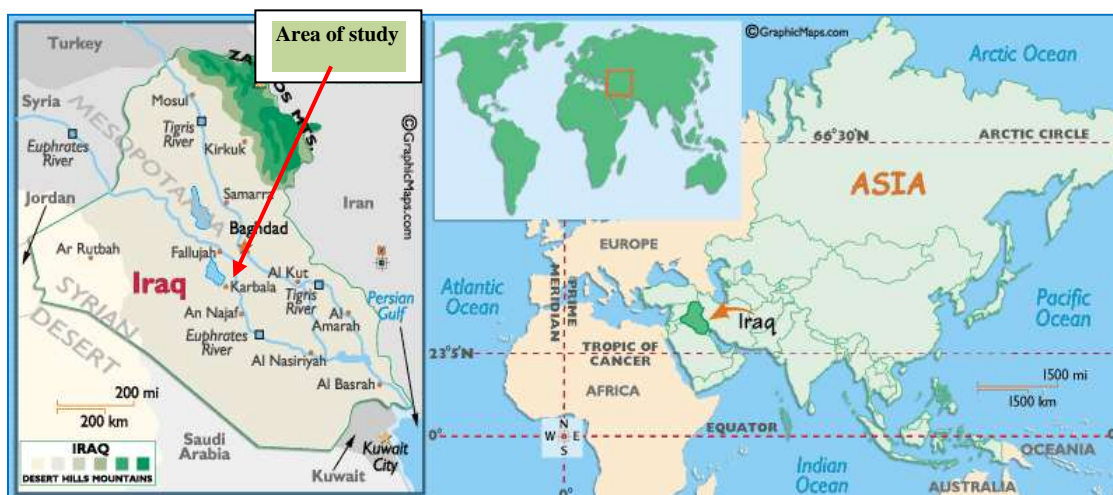


Figure 1: Map of Iraq showing the sampling city (Karbala)

#### Sample collection

Hair samples were collected from the same site of the head in all individuals, namely, from the back of the head, less than 1 cm from the scalp using acetone/distilled deionised water washed scissors. Generally, a sample (mass > 0.5 g) was collected and stored in polyethylene bags at room temperature until the time of analysis [11, 13, 14, 15].

## Instrumentation

A Finnigan MAT SOLA ICP-MS with argon plasma (Finnigan MAT, Hemel Hempstead, UK) was used for multi-elements analysis. The ICP-MS operating conditions are shown in (Table 1). Trace element levels were determined using  $^{51}\text{V}^+$ ,  $^{55}\text{Mn}^+$ ,  $^{59}\text{Co}^+$ ,  $^{63+65}\text{Cu}^+$ ,  $^{66+68}\text{Zn}^+$ ,  $^{88}\text{Sr}^+$  and  $^{112}\text{Cd}^+$ . Internal standard solutions were prepared to monitor isomer stability, using 100  $\mu\text{g/L}$   $^9\text{Be}^+$ ,  $^{76}\text{Ge}^+$ ,  $^{115}\text{In}^+$ ,  $^{209}\text{Bi}^+$ .

**Table 1:** ICP-MS Instrument Operating Parameters.

ICP-MS Parameters	Sitting used
Plasma argon flow rate	16 L/min
Auxiliary argon flow rate	1.2 L/min
Nebuliser argon flow rate	0.8 L/min
Incident power	1400 W
Reflected power	5W
Nebuliser pressure	2.0 bar
Sample orifice (nickel)	1.1 mm
Skimmer orifice (nickel)	0.7 mm
Spray chamber temp.	2 °C
Cooling water temp.	16 °C
Pump Speed	10.2 rpm
Spray chamber temp.	2 °C
Resolution	32
Sample cone	Nickel
Skimmer cone	Nickel

## Sample preparation

Hair samples were cut into small pieces and placed in labeled polypropylene beakers. Acetone was added to each beaker to cover the samples (~ 25 mL). After 5 minutes of sonication in an ultrasonic bath, the hair samples were also washed using the same procedure with distilled deionised water (DDW) (three times) and once again with acetone. The washed samples were dried in an oven at 60 °C for 2-3 hours. The dried samples were weighed (0.15 to 0.50 g) depending on available material, then transferred into a Kjeldhal™ Tube. Concentrated nitric acid, 1 mL (Aristar 65%) (Fisher Scientific UK Limited, Bishop Meadow Road, Loughborough, Leicestershire, UK) was added to each tube. Then the digestion tubes were placed on a digestion block for heating at 165 °C ( $\pm 10$  °C) until the hair sample is digested ~ half hour. All digested samples were diluted using polyethylene volumetric flasks (15 to 50 ml) depending on the sample mass and the volume made up with DDW, resulting in a dilution factor of 100 fold. Sample solutions were centrifuged for 10 minutes at 3000 rpm (MSE Mistral 2000 Thermo Life Sciences) and filtered through Millex filter units with MF-Millipore (0.45  $\mu\text{m}$ ) (Millipore, Carrigtwohill, Co. Cork, Ireland). The digested hair solutions were stored at 4°C until analysis by ICP-MS. Along with the scalp hair samples, a reference human hair material (GBW09101 Human Hair) provided by the National Research Centre for Certified Reference Materials, China and an experimental blank were treated in the same manner in order to check the analytical accuracy and precision of the digestion procedure.

## Validation of analytical methods

The accuracy of the methodology was tested by using certified reference material GBW 09101 Human Scalp Hair (Table 2) [2]. Recovery (% R) was calculated as  $100 \times \text{measured concentration} / \text{certified concentration}$  and an acceptance limit between 90 and 110% was considered as the desired range according to the criteria described by the Commission Decision 2002/657/EC [6]. In general, there is a good agreement between the measured and certified values and the recovery values were between 90-107 %. The precision of the digestion method, based on triplicate analysis of the GBW 09101 material produced acceptable levels of relative standard deviation (RSD) between 0.4 – 7.8 % (Table 2). Precision levels were also checked for matrix effects of the typical human scalp hair samples under investigation by replicate analysis (n=10). Mean, standard deviation (SD) and relative standard deviation (RSD) values were summarized in Table 3. In generally, good levels of precision were obtained with acceptable range of 3 – 6.4 % RSD. The limit of quantification (LOQ) was calculated as  $10s/m$ , where  $s$  is the standard deviation of 10 successive analyses of the blank (1% HNO<sub>3</sub> solution), and  $m$  is the slope of the calibration curve (Table 3) [16].

**Table 2:** Accuracy and Precision Assessment Using Human Scalp Hair GBW 09101 Certified Reference Material with Wet Digestion (Kjeldahl™ Tube) and a 0.5 g Mass with Constant Dilution Factor, (n=3)

Element	Elemental Level (µg/g)			Precision % RSD
	Measured (Mean)	Accuracy Certified	Percentage Recovery	
V	0.062	0.069	90	4.1
Mn	2.69	2.94	92	2.9
Co	0.125	0.135	93	3.6
Cu	22.5	23	98	2.7
Zn	177	189	94	0.4
Sr	4.54	4.19	108	1.5
Cd	0.089	0.095	94	7.8

## Statistical analysis

The results obtained for the washed scalp hair samples were evaluated using a t-test in order to assess whether any significant differences exist between the mean values for trace elements as a function of gender and smoking activity [17]. The statistical significance was determined at a probability level of  $P = 0.05$ .

**Table 3:** Precision Levels of Washed "Pooled" Human Scalp Hair Samples (n=10) Using ICP-MS.

Element	Elemental Levels ( $\mu\text{g/g}$ )			
	Measured (Mean)	$\pm\text{SD}$	% RSD	*LOQ ( $\mu\text{g/L}$ )
V	0.08	0.01	6	0.3
Mn	3.74	0.24	6.4	0.4
Co	0.17	0.01	5.8	0.07
Cu	14.6	0.7	4.8	2
Zn	122	4	3	202
Sr	3.74	0.18	4.7	0.2
Cd	0.21	0.01	5.8	0.2

\* LOQ is limit of quantification

## Results and Discussion :

### *Elemental levels of washed human scalp hair*

Multi-elemental analysis of washed scalp hair samples from Karbala, Iraq was performed by using inductively coupled plasma mass spectrometry (ICP-MS). The elemental levels of washed scalp hair are compared with reported normal concentration ranges from other literature values of washed hair for control or healthy population from different countries [18, 19, 20, 21]. The findings confirmed that the concentration of most of the elements in literature values have some degree of variation with those reported in one country from another. These differences arise because the trace element levels in biological samples may be affected by different factors, such as; age, gender, smoking habit, hair color, ethnical and geographic origin, dietary program, etc. [3, 13, 18].

The mean and standard deviation values of population of Karbala are reported in Table 4. Elemental values of hair are compared with reference range values of control or healthy population living in Sweden, Italy, Rio de Janeiro, England and the USA [18, 19, 22, 21, 22]. It was found that the mean levels ( $\mu\text{g/g}$  dry weight) of V (0.33), Mn (1.76), Co (0.131), Cu (23.53), Zn (201), and Sr (11.58) of the scalp hair from individuals living in Karbala were higher than the normal ranges in the scalp hair ( $\mu\text{g/g}$  d.w.) of healthy population living in Sweden in terms of V (0.005 – 0.134), Co (0.002 – 0.063), Zn (68 – 198) and Sr (0.14 – 5.54) [18]; Rio de Janeiro in terms of Mn (0.26 – 0.75), Zn (125 – 165) and Sr (1 – 7.6) [19, 21]; and Italy in terms of Sr (0.06 – 6.31) [20]. On the other hand, the mean values in the present study were completely in agreement with reported ranges ( $\mu\text{g/g}$  dry weight) in England for V (0.05 – 0.5), Mn (1 – 3), Co (0.1 – 0.8), Cu (15 – 35), Zn (160 – 220) and Cd (0.2 – 2.0) [22]. In the light of these results, it can be seen that some of these elements were in agreement with those reported in other countries while other elements were in disagreement. A possible explanation is that the main problem in Iraq is the environmental contamination. One of the most important sources of pollution in Iraq is from military-weapons that were used in the wars, along with oil spills and scrap metal from destroyed military vehicles [20]. As a result, air, water and soil environments were contaminated. Eventually, these chemicals pass into vegetables, fruit, plants and livestock, then into human body through food chain and environmental exposure. The range values for most of the elements in this study overlap with published ranges reported by Rodushkin and Axelsson [18], the only exception is for maximum value of Zn (884  $\mu\text{g/g}$  d.w.). The wide concentration ranges reported in the literature may be due to the variety of specific local conditions and the effect of the development of analytical techniques

## **The First Scientific Conference the Collage of Sciences 2013**

[18]. In addition, these results are typical for the elemental levels of scalp hair as there is no homeostatic regulation of this tissue and some samples contain "outline". In the light of these results, the concentration ranges for trace elements need to pay more attention in terms of the high levels of one (or possibly more) value/s that appears to differ from other values in the study data set such as maximum values. To closely follow the levels of Sr in this study, it can speculate that the higher levels of Sr should be followed-up in further studies to establish whether a new link can be found through the analysis of soils and main food in this region.

**Table 4:** Element Mean, Standard Deviation and Range Values ( $\mu\text{g/g}$ ) in Washed "Control or Healthy" Scalp Hair from Population Living in South-West Iraq (Karbala) (236 Subjects)

Element	*Mean	Standard Deviation	Range
V	0.33	0.41	< LOQ – 3.77
Mn	1.76	2.38	0.06 – 16.65
Co	0.13	0.34	0.01 – 3.74
Cu	23.53	23.22	0.2 – 247.8
Zn	201	159	45 – 884
Sr	11.58	13.93	0.67 – 82.95
Cd	0.33	0.41	< LOQ – 3.77

\*mean value for 236 subjects, the only exception for Zn (n=217) and Sr (n=231).

### **Influence of Gender**

The population was divided into two subgroups, male (n=40), and female (n=40), both groups were non-smokers and at the same range of ages. The results obtained by this study were compared with those reported for males and female population living in others countries (Table 5). It was found that the high mean of level ( $\mu\text{g/g d.w.}$ ) of V (0.37), Mn (2.21), Co (0.22), Cu (25.2), Zn (330), Sr (31) and lower level ( $\mu\text{g/g d.w.}$ ) of Cd (0.36) are observed in the scalp hair of females of healthy individuals from Karbala than their levels V (0.35), Mn (1.81), Co (0.07), Cu (24.8), Zn (232), Sr (8.8) and Cd (0.37) in males. The high mean of level of Mn and lower levels of Cd in the scalp hair of females than males were also reported for individuals from Italy, India and Poland [20, 23, 24]. Moreover, the results obtained by this study for Co, Cu, Zn and Sr were in the same comparison with those reported by other authors for individuals from India (regarding Co), Poland (regarding Cu) and Italy (regarding Zn and Sr) (Table 5). In contrast, the results of V, Co, Cu, and Zn levels in males and female hairs that reported by the present work are in different with those reported for males and females from Italy (in terms of V, Co and Zn), India (in terms of Cu and Zn) and Poland (in terms of Co and Zn) (Table 5).

**Table 5:** Mean and Standard Deviation ( $\pm$ SD) Values for the Washed Scalp Hair of Control or Healthy Male (M) and Female (F) Individuals from Different Countries ( $\mu\text{g/g}$ )

Element	Karbala, Iraq (This study)		Italy [20]		India [23]		Poland [24]	
	M	F	M	F	M	F	M	F
V	0.35 ( $\pm 0.19$ )	0.37 ( $\pm 0.21$ )	1.41 ( $\pm 1.59$ )	0.99 ( $\pm 1.09$ )	-	-	-	-
Mn	1.81 ( $\pm 1.10$ )	2.21 ( $\pm 2.09$ )	0.34 ( $\pm 0.51$ )	0.36 ( $\pm 0.67$ )	3.98	4.72	1.71 ( $\pm 1.49$ )	3.08 ( $\pm 2.61$ )
Co	0.07 ( $\pm 0.05$ )	0.22 ( $\pm 0.09$ )	0.75 ( $\pm 0.97$ )	0.66 ( $\pm 1$ )	0.25	0.36	0.50 ( $\pm 0.79$ )	0.39 ( $\pm 0.65$ )
Cu	24.8 ( $\pm 8.96$ )	25.2 ( $\pm 12.7$ )	25 ( $\pm 69.7$ )	18.3 ( $\pm 22$ )	12.5	9.23	7.88 ( $\pm 8.7$ )	8.04 ( $\pm 9.54$ )
Zn	232 ( $\pm 144$ )	330 ( $\pm 214$ )	146 ( $\pm 46$ )	155 ( $\pm 58$ )	95	86	131.58 ( $\pm 56.52$ )	126.42 ( $\pm 63.7$ )
Sr	8.8 ( $\pm 6.9$ )	31 ( $\pm 12$ )	0.9 ( $\pm 0.73$ )	1.59 ( $\pm 1.29$ )	-	-	-	-
Cd	0.37 ( $\pm 0.21$ )	0.36 ( $\pm 0.20$ )	0.24 ( $\pm 0.39$ )	0.22 ( $\pm 0.31$ )	0.36	0.24	0.79 ( $\pm 1.51$ )	0.44 ( $\pm 0.5$ )

Table 6 shows the findings as the mean, standard deviation (SD) and range of trace elements ( $\mu\text{g/g}$  d.w.) for each study group. Although the levels of V, Mn, Cu and Zn are slightly higher in females than males, there are no significant differences at the probability of 0.05. In contrast, based on t-test results there are significant differences reported between the Co and Sr levels ( $\mu\text{g/g}$  d.w.) in females ( $0.22 \pm 0.09$ ); ( $31 \pm 12$ ) than males ( $0.07 \pm 0.05$ ); ( $8.8 \pm 6.9$ ), respectively at this level of significance (Table 6). On the other hand, the level of Cd was also smaller higher in male hair than in female hair. The reason for this difference may be due to the differences in outdoor activities, health behaviors, and difference in urine excretion or kidney activities [25]. The high levels of Co and Sr in female hair than male hair may be probably due to the majority of women living in the study area have long term exposure to exogenous contamination; namely permanent solutions, dyes and bleaches [13]. The results were also compared with literature findings reported by other authors. Similar findings have been reported: higher level ( $\mu\text{g/g}$ ) of Cu ( $8.71 \pm 4.53$ ) in females than males ( $6.53 \pm 2.02$ ) [26]; higher levels ( $\mu\text{g/g}$ ) of Sr (3.5), Mn (1.5) in female's hair than males [13]; lower levels ( $\mu\text{g/g}$ ) of Cd (0.56) in female hair than male (0.46) [3]; higher levels ( $\mu\text{g/g}$ ) of Cu (18.6), Zn (152.9) and lower levels ( $\mu\text{g/g}$ ) of Cd (83.3) were reported in females hair than males Cu (15.8), Zn (139.9) and Cd (89.3) [27]; higher level ( $\mu\text{g/g}$ ) of Mn (0.94) in females than males Mn (0.65) [8]; and higher levels ( $\mu\text{g/g}$ ) of Co ( $0.016 \pm 0.014$ ) and Sr ( $1.64 \pm 1.32$ ) in females hair than males ( $0.01 \pm 0.007$ ) and Sr ( $0.891 \pm 0.731$ ) [18].

**Table 6:** Trace Element Levels (Mean and Range) and SD in the Washed Scalp Hair ( $\mu\text{g/g}$ ) of Females (n=40) and Males (n=40) from Karbala, Iraq along with t-test

Element	Elemental Concentration ( $\mu\text{g/g}$ )				t-test Calculated Value <sup>+</sup>
	Male		Female		
	Mean ( $\pm$ SD)	Range	Mean ( $\pm$ SD)	Range	
V	0.35 ( $\pm$ 0.19)	0.12 – 1.41	0.37 ( $\pm$ 0.21)	0.04 – 0.95	0.45
Mn	1.81 ( $\pm$ 1.10)	0.33 – 5.94	2.21 ( $\pm$ 2.09)	0.16 – 16.65	1.08
Co	0.07 ( $\pm$ 0.05)	0.01 – 0.38	0.22 ( $\pm$ 0.09)	0.01 – 0.49	9.56 <sup>b</sup>
Cu	24.8 ( $\pm$ 8.96)	8.2 – 49.6	25.2 ( $\pm$ 12.7)	0.2 – 94.6	0.17
Zn	232 ( $\pm$ 144)	61 - 647	330 ( $\pm$ 214)	60 – 884	2.42
Sr	8.8 ( $\pm$ 6.9)	0.7 – 35.6	32 ( $\pm$ 12)	4 – 71	10.36 <sup>b</sup>
Cd	0.37 ( $\pm$ 0.21)	0.12 – 1.41	0.36 ( $\pm$ 0.20)	< LOQ – 0.95	0.12

SD = standard deviation, n = number of samples, <sup>+</sup> t-test, calculated values, t critical value = 2.44, degree of freedom (df) = 78, <sup>b</sup> indicate significant difference level at  $P = 0.05$ .

### Influence of Smoking

The study population 50 % males smokers and 50 % males non-smokers are used in order to evaluate whether the effects of a smoking habit influence the trace element levels of the population living in Karbala. The passive smoking group (n=6) was removed from any statistical evaluation. Table 7 reports the mean, standard deviation and range of V, Mn, Co, Cu, Zn, Sr and Cd for smokers and non-smokers groups. It was found that the higher levels of V, Zn, Sr, Mn and Cd and lower levels of Co and Cu are observed in the hair samples of smokers than non-smokers. The effect of a smoking habit on the elemental levels for scalp hair samples were examined by using a two tailed t-test (Table 7). In spite of the mean levels ( $\mu\text{g/g d.w.}$ ) of Mn and Zn are slightly higher in smokers ( $170 \pm 77$ ) and ( $1.35 \pm 0.89$ ) than non-smokers ( $147 \pm 55$ ) and ( $1.24 \pm 0.91$ ), respectively, there are no significant differences at the probability of 0.05. In contrast, there are significant differences between the V, Sr and Cd mean levels ( $\mu\text{g/g d.w.}$ ) in smokers ( $0.30 \pm 0.1$ ), ( $9.27 \pm 5.92$ ), and ( $0.31 \pm 0.23$ ) and non-smokers ( $0.2 \pm 0.13$ ), ( $5.46 \pm 2.75$ ) and ( $0.18 \pm 0.12$ ), respectively, at probability level of  $P = 0.05$ . Cadmium was found in commercial cigarette tobacco at ranges of ( $0.24 - 2.03 \mu\text{g/g d.w.}$ ). This represents a higher Cd levels than that found in normal plant material (typically  $< 0.5 \mu\text{g/g Cd}$ ) [28]. In addition Cd mean level in smokers was ( $0.31 \pm 0.23 \mu\text{g/g d.w.}$ ) which is higher than the typical level in human scalp hair ( $0.2 \mu\text{g/g d.w.}$ ). It was found previously that 30 % of the total Cd is recovered in the ash and filter whilst 70 % passed with the smoke fraction. Studies using unwashed hair sample was collected from pregnant women smokers and pregnant women non-smokers.



**Table 7:** Effect of smoking activity on trace elements levels ( $\mu\text{g/g}$ ) in human scalp hair (n=102) samples from Karbala, Iraq.

Elements	Elemental concentration ( $\mu\text{g/g}$ )				<sup>+</sup> t-test Value
	Smokers		Non-Smokers		
	Mean ( $\pm$ SD)	Range	Mean ( $\pm$ SD)	Range	
<sup>b</sup> V	0.30 ( $\pm$ 0.26)	< LOQ – 3.77	0.20 ( $\pm$ 0.13)	< LOQ – 1.01	2.44
Mn	1.35 ( $\pm$ 0.89)	0.14 – 5.88	1.24 ( $\pm$ 0.91)	0.06 – 4.7	0.58
Co	0.065 ( $\pm$ 0.059)	0.01 – 0.892	0.098 ( $\pm$ 0.099)	0.01 – 1.155	2.04
Cu	19.53 ( $\pm$ 8.81)	7.11 – 50.48	20.78 ( $\pm$ 9.25)	6.95 – 65.81	0.71
Zn	170 ( $\pm$ 77)	52 – 618	147 ( $\pm$ 55)	47 – 549	1.79
<sup>b</sup> Sr	9.27 ( $\pm$ 5.92)	1.56 – 36.51	5.46 ( $\pm$ 2.75)	1.07 – 18.95	4.21
<sup>b</sup> Cd	0.31 ( $\pm$ 0.23)	0.02 – 2.69	0.18 ( $\pm$ 0.12)	< LOQ – 1.01	3.58

SD = standard deviation, n = number of samples, <sup>+</sup> t critical value = 2.428, <sup>b</sup> indicate to significant difference level at P=0.05, df (degree of freedom) = 102.

This study showed significantly elevated Cd levels in smokers group compared with non-smokers group;  $1.43 \pm 0.32$  and  $0.53 \pm 0.27$   $\mu\text{g/g}$  Cd d.w., respectively. However, these levels of Cd can reduce the birth weight and gestational age in pregnant women (smokers) when compared with pregnant women (non-smokers) [28]. On the other hand, the pregnancy may involve increased toxicity (kidney and bone) and increased accumulation of cadmium in the kidney [29]. Similar results were found [18, 12, 30]. They reported that smokers have increase cadmium levels in the scalp hair when compared with non-smokers. Strontium mean level was found in 16 imported commercial cigarettes from Karbala (75.29  $\mu\text{g/g}$  d.w) and the mean level of Sr in smoker's hairs was ( $9.27 \pm 5.92$ ). This represents a higher Sr levels than that found in typical levels of Sr in human scalp hair (0.1  $\mu\text{g/g}$  d.w) [28]. Previous study found that Sr has been associated with the prevalence of dental carries [31]. In addition, the levels of Sr in cairns teeth were significantly higher than those in sound teeth [32]. Moreover, one study used young rats has observed that the ingestion of high doses of Sr can cause skeletal disorders [33]. Inspection of the above data showed that some non-smokers also reflected high levels of Sr in scalp hair. The high Sr levels in non-smokers may be due to drinking water (1.82  $\mu\text{g/ml}$ ) from Karbala [34]. V mean level in smoker hairs was ( $0.3 \pm 0.26$   $\mu\text{g/g}$  d.w) which is higher than typical value in human scalp hair (0.05  $\mu\text{g/g}$  d.w) [28]. The high levels of V may be because the effects of smoking activity or the high levels of V in tap water ( $7.9 \pm 5.7$   $\mu\text{g/ml}$ ) in Karbala comparison with typical levels in fresh water (0.5  $\mu\text{g/ml}$ ) [28]. In the light of these results, the levels of trace elements under investigation based on the smoking habit of individuals, but these effects vary from element to another.

## **Conclusion :**

The scalp hair is a good indicator to assess the levels of essential and non-essential elements in monitoring of environmental exposure from anthropogenic sources including chemical pollutants that are released into the environment, the dietary intake of trace elements, especially for non-essential or 'toxic' elements, the relationship with smoking activity and any possible connection between specific trace elements and diseases. The results in this study might be useful in establishing a reference values for the level of elements in hair with respect of gender and smoking

## **The First Scientific Conference the Collage of Sciences 2013**

habit. These results can also use for comparison with other countries and for evaluating for future environmental pollution or health disease studies in Iraq. In general, the elemental levels of scalp hair varied with gender and smoking habit. It was found that the levels of Sr, Cd and V were significantly different between smokers and non-smokers and the higher levels of V, Mn, Co, Cu, Zn and Sr and lower levels of Cd were obtained in the hair samples of females than males. The results was also compared with the data reported by other authors, therefore some results were confirmed. In the light of these findings, it can speculate that in order to establish reference levels of trace elements in human scalp hair it is necessary to study the effect of gender and smoking activity on large population group living in different regions. This way will make the scalp hair analysis to be more reliable.

### **Acknowledgements :**

The authors wish to thank the Iraqi government for its financial support, in particular the Ministry of Higher Education and Scientific Research. The authors also wish to acknowledge the sampling of the human scalp hair by Muneer Al-Daami, Mohammad Al-Kaabi, University of Karbala, Iraq, Allaa Mohammad, University of Baghdad, Chemistry department for his literature review and Adnan Hashim, Al-Hussein hospital, Karbala, Iraq. The authors would like to thank Judith Peters, University of Surrey, Division of Chemical Sciences for her help in the samples preparation laboratory. The authors thank Hannah Farnfied for help in tobacco analysis by ICP-AES.

### **References :**

- 1- Ashraf W, Jafaar M, Anwer K, Ehsan U (1995) Age and sex based comparative distribution of selected metals in the scalp hair of an urban population from two cities in Pakistan, *Environmental Pollution* 87: 61-4.
- 2- Bass DA (2001) Trace elements analysis in hair: Factors determining accuracy, precision, and reliability. *Alternative Medicine Review* 6(5): 472-474.
- 3- Ozden TA, Gokcay G, Erten HV, Suoglu OD, Kilic A, Sokucu S, Saner G (2007) Elevated hair levels of cadmium and lead in school children exposed to smoking and in highways near schools. *Clin Biomech* 40: 52-56.
- 4- Amaral AFS, Arruda M, Cabral S, Rodrigues AS (2008) Essential and non-essential trace metals in scalp hair of men chronically exposed to volcanogenic metals in the Azores, Portugal. *Environ Int* 34: 1104-1108.
- 5- Gault AG, Rowland HAL, Charnock JM, Wogelius RA, Gomez-Morilla I, Vong S, Leng M, Samreth S, Sampson ML, Polya DA (2008) Arsenic in hair and nails of individua exposed to arsenic-rich groundwater in Kandal province, Cambodia. *Sci Total Environ* 393: 168-176.
- 6- Olmedo P, Pla A, Hernandez AF, Lopez-Guarido O, Rodrigo L, Gil F (2010) Validation of a method to quantify chromium, cadmium, manganese, nickel and lead in human whole blood, urine, saliva and hair samples by electrothermal atomic absorption spectrometry. *Anal Chim Acta* 659: 60-67.
- 7- Wang T, Fu J, Wang Y, Liao C, Tao Y, Jiang G (2009) Use of scalp hair as indicator of human exposure to heavy metals in an electronic waste recycling area. *Environ Pollut* 157: 2445-2451.
- 8- Dong X, Nakaguchi Y, Hiraki K (1998) Determination of chromium, copper, Iran, manganese and lead in human hair by graphit furnace atomic absorption spectrometry. *Analytical Sciences* 14: 785-789.
- 9- Hannigan RE, Harden WL (2005) Elemental analysis of human hair by LA-ICP-MS. *Arkansas State University Department of Chemistry* 870: 680-4360.

## **The First Scientific Conference the Collage of Sciences 2013**

- 10- Esteban M, Castano A (2009) Non-invasive matrices in human biomonitoring: A review. *Environ Int* 35: 438-449.
- 11- Senofonte O, Violante N, D'Ilio S, Caimi S, Peri A, Caroli S (2001) Hair analysis and the early detection of imbalances in trace elements for members of expeditions in Antarctica. *Microchem J* 69: 231-238.
- 12- Sukumar A, Subramanian R (2007) Relative element levels in the paired samples of scalp hair and fingernails of patients from New Delhi. *Sci Total Environ* 372: 474-479.
- 13- Chojnacka K, Gorecka H, Gorecki H (2006) The effect of age, sex, smoking habit and hair color on the composition of hair. *Environ Toxicol Pharmacol* 22: 52-57.
- 14- Hartman JE (2006) Neurology in operation Iraqi freedom: Risk factors for referral, clinical presentations and incidence of disease. *J Neurol Sci* 241: 83-90.
- 15- Rodrigues JL, Batista BL, Nunes JA, Passos CJS, Jr FB (2008) Evaluation of the use of human hair for biomonitoring the deficiency of essential and exposure to toxic elements. *Sci Total Environ* 405: 370-376.
- 16- Harris DC. (2007) *Quantitative Chemical Analysis*, Seventh Edition. New York, W. H. Freeman and Company 83-90.
- 17- Miller JN, Miller JC (2000) *Statistics and Chemometrics for Analytical Chemistry*. Fourth Edition, Pearson Education Limited, England 42-283.
- 18- Rodushkin I, Axelsson MD (2000) Application of double focusing sector field ICP-MS for Multielemental characterization of human hair and nails: Part II A study of the inhabitants of northern Sweden. *The Science of the Total Environment* 262: 21-36.
- 19- Chojnacka K, Gorecka H, Chojnacki A, Gorecki H (2005) Inter-element interactions in human hair. *Environ Toxicol Pharmacol* 20: 368-374.
- 20- Senofonte O, Violante N, Caroli S (2000) Assessment of reference values for elements in human hair of urban schoolboys. *J. Trace Elements Med. Biol* 14: 6-13.
- 21- Ward NI (2000) Trace Elements, Chapter 15, in *Environmental Analytical Chemistry*, Fifield FW, Haines PJ. (eds), Second Edition, Blackwell Science, Oxford. 360-392.
- 22- Stovell AG (1999) *Trace Elements and Human Fertility*. Department of Chemistry, Guildford, University of Surrey. PhD.
- 23- Rao KS, Balaji T, Rao TP, Babu Y, Naidu GRK (2002) Determination of iron, cobalt, nickel, manganese, zinc, copper, cadmium and lead in human hair by inductively coupled plasma atomic emission spectrometry. *Spectrochimica Acta Part B* 57: 1333-1338.
- 24- Nowak B (1998) Contents and relationship of elements in human hair for a non-industrialised population in Poland. *The Science of the Total Environment* 209: 59-68
- 25- Mortada WI, Sobh MA, El-Defrawy MM, Farahat SE (2002) Reference intervals of cadmium, lead, and mercury in blood, urine, hair, and nails among residents in Mansoura city, Nile Delta, Egypt. *Environmental Research Section A* 90: 104-110.
- 26- Forte G, Alimón ti A, Violante N, Di Gregorio M, Senofonte O, Petrucci F, Sancesario G, Bocca B (2005) Calcium, copper, iron, magnesium, Silicon and zinc content of hair in Parkinson's disease. *J Trace Elem Med Biol* 19: 195-201.
- 27- Miekeley N, Dias MTW, Silveira CL (1998) How reliable are human hair reference intervals for trace elements. *The Science of the Total Environment* 218: 9-17.
- 28- Ward NI (2000) Trace Elements, Chapter 15, in *Environmental Analytical Chemistry*, Fifield FW, Haines PJ. (eds), Second Edition, Blackwell Science, Oxford. 360-392.
- 29- Vahter M, Berglund M, Akesson A, Liden C (2002) Metals and women's health. *Environmental Research Section A* 88: 145-155.
- 30- Kazi TG, Memon A, Afridi H, Jamali M, Aria M, Jalbani N, Sarfraz R (2008) Determination of cadmium in whole blood and scalp hair samples of Pakistan male lung cancer patients by electrothermal atomic absorption spectrometer. *Sci Total Environ* 389: 270-276.

## **The First Scientific Conference the Collage of Sciences 2013**

- 31- Muynck DD (2008) Isotopic analysis of archaeological artefacts by means of single-collector and multi-collector ICP-mass spectrometry. PhD, Department of Analytical Chemistry, University of Kent, UK.
- 32- Shigemi T, Tanaka T, Hayashida Y, Maki K (2008) Study of salivary strontium and silver concentrations in primary school children related to dental caries. *Biol Trace Elem Res* 123:80-90
- 33- Greve K, Nielsen E, Ladfoged O (2007) Evaluation of health hazards in drinking water. *Toxicol Lett* 172S: 1-240
- 34- Joda BA (2010) The Impact of Minor and Trace Elements on the Health Status of Iraqi Individuals and Relationship with Smoking Activity, Age and Gender. Transfer report from MPhil to PhD. University of Surrey 1-30