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*Full Length Research Paper*

# Effect of metal poisoning and the implications of gender and age on the elemental composition in patients with mental behavioural disorders

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**The objective of this work was to investigate the possible correlation between the exposure to selected toxic metals and the behavioural disorder of mentally ill patients. The study also sought to establish if gender and age of the patient had an effect on the pattern of the elemental distribution in their head hair and blood samples. To achieve this, the concentrations of a number of selected toxic metal elements were determined in 60 mentally ill patients and 43 healthy individuals (control) in Ile-Ife area, in Nigeria, using inductively coupled plasma spectrophotometer-optical emission spectrometer (ICP-OES). The behavioural disorder cases investigated were 8 bipolar, 2 post partum psychosis, 43 schizophrenia and 7 non-specific cases. The concentration ranges of Cu, Zn, Ca, Li, V, Be (for both males and females), Cd and Sr (for females only) as analyzed from the patients' head hair with behavioural disorders, were found to be similar with those of the controls. However, the concentration ranges of Al, Ba, Mg, Cr and Cd, Sr (for males only) were higher in patients than in the controls, while those for K and Fe were found to be higher in the controls than in the patients for both males and females. Blood samples analysis showed that, nearly all the elements were higher in the female (patients and control) than in the males; a possible indication that women may be at greater risk than men. It was also shown that, age may have an influence on the accumulation of some specific elements. The accuracy of the analytical results was experimentally demonstrated by NCS DC 73347 certified reference material that was analyzed along the standards while the significance of the data obtained was tested statistically at both p = 0.01 and 0.05.**

**Key words:** Toxic metals, behavioural disorder, gender, age, inductively coupled plasma-optical emission spectrometer.

## INTRODUCTION

Exposure to toxic metals is known to have adverse effects on the central nervous system (CNS) functions and on an individual's mood which may translate into depresssion, insomnia and other neurodegenerative disorders such as autism and bipolar disorder (Watts, 1990; Ibrahim et al., 2006; Eck and Wilson, 1989; Pfeiffer 1975; Bowler et al., 2006). Aluminum and lead exposure for example, has been reported to be responsible for behavioral problems and learning deficiency in children which persists for a long time after the initial exposure (Yuan et al., 2006). Other disorders such as delays and/or lack in intellectual ability, academic achievement and psychomotor development have all been associated with the exposure of children to these toxic metals (Yuan et al., 2006). Other toxic metals such as antimony, uranium, arsenic, beryllium, mercury, cadmium and aluminum are also known to cause autism and other behavioural problems

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(Adams et al., 2006). On the other hand, the bioaccumulation tendency of these toxic metals tend to aggravate the situation further because even at trace concentrations, they can gradually build up in the body and reach levels where they can cause serious health problems and behavioural disorders.

Once in the body, the biological fluids such as blood or plasma tend to shape the distribution pattern of the toxic metal elements. Therefore, considerable variations in terms of elemental distribution patterns can occur between specific population subgroups which may be factored in gender and/or age (Christensen, 1995; Cornelis et al., 1994; Gil et al., 2006; Goullé et al., 2005; Kucera et al., 1995; Minoia et al., 1990). Besides investigating the link between psychiatric disorders and the presence of some toxic metals in the blood and head hair samples of patients in this study, the effect of gender and age on the elemental distribution pattern among the patients, was determined.

Elemental analysis typically utilizes the atomic absorption spectrometry (both flame FAAS; and graphite furnace, GFAAS or electrothermal, ETAAS) (Parsons and Barbosa, 2007; Barbosa et al., 2004; Loureiro et al., 2007; Barbosa et al., 2001; Parsons and Slavin, 1993; Zanão et al., 2002). However, nowadays many laboratories are switching towards methods based on inductively coupled plasma interfaced to either optical emission (ICP-OES) or mass spectrometry (ICP-MS) (Parsons and Barbosa, 2007; Batista et al., 2008; Rodrigues et al., 2008; Palmer et al., 2006).

The increasing preference to these techniques is based on the fact that, ICP methods have a number of advantages over AAS based techniques which include the ability for simultaneous multi-element measurement capability, much lower detection limits and a wider linear dynamic range which allow the determination of major and trace elements in the same sample aliquot (Parsons and Barbosa, 2007).

In this work, the results of the analysis of toxic metal elements from blood and head hair samples of mentally ill population selected from Ile-Ife Nigeria was reported. The technique used was ICP-OES, after a microwave-digestion sample preparation step. For convenience, the patients with the behavioural disorders investigated (8 bipolar, 2 post partum psychosis, 43 schizophrenia and 7 non-specific cases) will be referred to as 'patients' throughout this paper.

## MATERIALS AND METHODS

### Sample collection

The sampling was conducted from 18/06/ 2007 to 30/08/2007. Samples were collected from the out-patients of the Department of Mental Health of the Obafemi Awolowo University Teaching Hospital, Ile-Ife, Nigeria, with the permission of the institution's Ethical Committee Review and Certification (The Consent and Ethiics Certificate Number given is - IRB/IEC 00005422).

### Design and setting

The design for sample collection and analysis was carried out on a prospective clinical and laboratory study of mentally-ill patients and was conducted on patients at the Department of mental health, Obafemi Awolowo University Teaching Hospital, Ile- Ife; which included in-patients and out patients.

The Obafemi Awolowo University Teaching Hospital is an affiliate of Obafemi Awolowo University with major hospital at Ile-Ife and Ilesa. Catchments area for patients included Osun, Ekiti, Ondo and parts of Kwara and Oyo States.

### Inclusion and exclusion criteria

Patients with confirmed cranio-cerebral/head injury and organic psychosis were excluded.

### Hair collection criteria

Hair sample was collected at the nape of the head with scissors to avoid contamination with facial secretions and cosmetics after permission from the patients or their responsible relatives, having endorsed the consent form.

### Location and general features of the study area

Ile-Ife is the locus of the research work and is situated in Osun State, south western Nigeria. It lies approximately between latitudes 7°26'N to 7°33'N of the equator and longitude 4°30'E to 4°35'E of the prime meridian on a general elevation of about 350 m above the mean sea level. It is about 30 km to Ilesa and about 35 km south of Osogbo, the Osun State capital and 610 km south west of Abuja, the Nigerian capital.

The catchments areas for patients included Osun, Ekiti, Ondo and parts of Oyo States. All were located within the south western Nigeria and there are no large industrial concentrations but mostly agrarian communities with increasing tendencies towards the use of pesticides and fertilizers.

### Reagents, chemicals and materials

The Analar grade $HNO_3$ (70%) used for digestion was purchased from Merck (Darmstadt, Germany). The standard solutions were prepared using ultra pure water (18 MΩcm) purified using a Mill-Q® reagent water system (Millipore, Molsheim, France). The accuracy of the method was ascertained by examining the certified reference material (CRM) - NCS DC 73347 supplied by Industrial Analytical (Pty) Ltd, South Africa. For quantitative analysis, the multi-elements standard solutions used for calibration purposes were prepared by diluting the stock solution of 10 mg/l of the given elements and these were stored in polyethylene bottles, supplied by PolyChem, Congella, Durban 4013, South Africa. Polyethylene sample bottles and Teflon beakers were soaked in 10% $HNO_3$ for 48 h and later rinsed with distilled de-ionized water prior to metals analyses.

### Instrumentation and apparatus

#### Inductively coupled plasma- optical emission (ICP-OES)

The ICP-OES used was a Perkin Elmer Optima 5000 DV model with Echelle optical system. The following features and settings were utilized; axial viewing configuration with RF power 1300 W, plasma argon gas flow of 15 L/min, auxiliary gas flow of 0.5 L/min,

**Table 1.** Analysis of CRM (NCS DC 73347) with microwave digestion (n = 3).

| Element | Microwave digestion ( µg/g) (X) | Expected (µg/g) (Y) | Difference |
|---|---|---|---|
| Ba | 16.830 ± 1.860 | 17.00 ± 0.200 | 0.17 |
| Be | 0.055 ± 0.001 | 0.063 ± 0.002 | 0.08 |
| Cr | 0.430 ± 0.001 | 0.370 ± 0.060 | 0.06 |
| Cu | 13.60 ± 1.400 | 10.600 ± 1.200 | 3.00 |
| Fe | 49.80 ± 6.200 | 54.00 ± 0.100 | 0.20 |
| Li | 1.90 ± 0.0200 | 2.00 ± 0.010 | 0.20 |
| Sr | 25.80 ± 0.200 | 24.00 ± 1.00 | 1.80 |
| Zn | 189.90 ± 3.00 | 190.00 ± 2.900 | 0.10 |

nebulizer flow of 8 L/min, view distance of 15 mm, sample uptake flow rate of 15 ml/min, wash rate of 1.5 ml/min and wash time of 30s.

#### Microwave digester

The microwave digester used for the sample preparation was Auto Paar multiwave 300 with pressure/temperature sensor supplied from Austria. An analytical Mettler AE240 balance was used for all weights measurements. Digestion was carried out under the following settings; power = 400 W at 170°C for 25 min and at a holding time of 10 min.

#### Sample preparation

#### Certified reference material (CRM)

CRM weights of 10, 20, 30, 40 and 50 mg were weighed in clean PTFE crucibles and digested in a closed-vessel high-pressure microwave digestion unit to minimize sample contamination and analyte losses. To avoid incomplete sample digestion, an oxidizing acid mixture was used which composed 6 ml nitric acid, 3 ml hydrogen peroxide and 2.4 ml hydrogen fluoride.

#### Head hair samples

The hair samples (50 mg) were cut into smaller pieces approximately 0.3 cm and were thoroughly mixed to allow a representative sub-sampling of the head hair specimen and approximately 50 mg of the head hair samples were weighed into 25 ml Teflon vessels and then, concentrated $HNO_3$ (70% w/v, 6 ml), hydrogen peroxide (30% w/v, 3 ml) and hydrogen fluoride (2.4 ml) were added to each vessel and digested using microwave digester. The cleaning time was set at 20 min, after which it was made up to 15 ml with ultra pure water (18 MΩcm), filtered and was analyzed on ICP-OES.

#### Blood samples

The blood samples were drawn from the cubital vein using 5 ml syringe and polyethylene needle and were stored in pre-treated heparinized bottle and later freeze dried. The hair pieces were collected at the nape of the head to prevent contamination from cosmetics and facial secretions. After cutting, each sample was washed four times with 1:200 v/v dilution of Triton X-100. The samples were rinsed with acetone and allowed to drain. This was followed by three rinses with de-ionized water and two rinses with acetone. The samples were then, dried in an oven at 75 ± 5°C and kept in a clean standard envelopes or plastic papers, for the extraction step.

#### Statistical analysis

For the statistical analysis, data were analysed using the SPSS for windows software, version 15.0.1 (SPSS Inc., Chicago, USA). Statistically significant differences between groups were compared using analysis of independent t-test. The data are displayed as means ± standard deviation (SD) and p values of 0.01 or 0.05 were considered significant.

## RESULTS AND DISCUSSION

### Analytical validation

In order to ensure validity of results in any quantitative analytical work, the following aspects must be addressed; the correctness in the preparation of the sample and avoidance of errors due to sample loss or contamination (Becker and Dietze, 1999). In this work the use of certified reference material (NCS DC 73347) was employed, so as to provide a measure of accuracy and precision for the experimental data obtained in the analytical measurements. For quantitative analysis, calibration curves of different elements (prepared from a multi-element standard solution of 10 mg/l) were found to be linear with correlation coefficients above 0.99 as shown in Table 1 (curves not included). From Table 1, it is evident that by comparison with the CRM observations, the concentration values for Fe were lower than expected while for Cu, the values were higher than expected. The higher values for Cu may be due to a considerable contamination during sample preparation because of the small sample size used. Moreover, the lower results for Fe could either be as a result of losses in the washing step of sample preparation or inadequate digestion of the sample.

### Choice of sample specimens for analysis

Human hair has been extensively studied in clinical prac-

**Table** 2. Gender based elemental distribution pattern in patients' blood samples.

| Element | Patient (µg/g) | | | Control (µg/g) | | |
|---|---|---|---|---|---|---|
| | **Male** | **Female** | **Difference** | **Male** | **Female** | **Difference** |
| Al | 5.05 ± 0.072 | 4.85 ± 0.084 | 0.100 | 3.25 ± 0.040 | 2.76 ± 0.02 | 49.00 |
| Ba | 2.22 ± 0.089 | 2.09 ± 0.038 | 0.13 | 1.67 ± 0.002 | 1.09 ± 0.02 | 0.58 |
| Be | 0.28 ± 0.007 | 0.29 ± 0.008 | 0.010 | 0.22 ± 0.005 | 0.19 ± 0.001 | 0.03 |
| Cd | 0.39 ± 0.005 | 0.11 ± 0.004 | 0.28 | 0.15 ± 0.005 | 0.105 ± 0.005 | 0.045 |
| Cr | 1.24 ± 0.009 | 1.24 ± 0.009 | 0.00 | 0.36 ± 0.008 | 0.21 ± 0.006 | 0.15 |
| Li | 0.04 ± 0.007 | 0.037 ± 0.002 | 0.3 | 0.04 ± 0.001 | 0.037 ± 0.001 | 0.003 |
| Sr | 0.324 ± 0.01 | 0.330 ± 0.001 | 0.0006 | 0.28 ± 0.008 | 0.24 ± 0.001 | 0.004 |
| Mg | 140.13 ± 2.75 | 131.05 ± 3.51 | 9.08 | 86.13 ± 1.19 | 74.83 ± 3.27 | 11.30 |
| K | 5606.02 ± 16.35 | 4781.20 ± 14.08 | 824.82 | 7127.23 ± 15.42 | 6443.29 ± 16.56 | 683.94 |
| Fe | 2025.11 ± 12.25 | 1847.00 ± 15.02 | 178.11 | 1640.71 ± 13.50 | 1342.18 ± 15.57 | 298.53 |
| V | 3.42 ± 0.07 | 3.73 ± 0.89 | 0.31 | 2.63 ± 0.07 | 2.22 ± 0.02 | 0.41 |
| Zn | 19.11 ± 0.40 | 26.09 ± 1.08 | 6.98 | 10.73 ± 1.12 | 15.29 ± 1.05 | 4.56 |

tices because it serves as a good non-invasive medium for the determination of metal elements in the body as it gives an average concentration of trace elements over cumulative longer periods of time. In the case of body fluids, blood is usually preferred as the most appropriate specimen for elemental determination. In this study, head hair and blood samples were used for the elemental analysis.

## Elemental patterns in patients versus healthy persons' head hair and blood samples

The elemental composition of blood, plasma and head hair samples is known to be altered by some diseases (Dombova´ri et al., 1999). The main pattern changes as far as the elemental distribution was concerned were some elements that exhibited an increase in levels and others that showed decreased levels. In this study, it was observed that, the concentration ranges of Cu, Zn, Ca, Li, V, Be (for both males and females), Cd and Sr (for females only) as analyzed from hair samples from patients with behavioural disorders, were found to be similar with those of the healthy persons (controls). However, the concentration ranges of Al, Ba, Mg, Cr and Cd, Sr (for males only) were higher in patients than in the controls, while those for K and Fe were found to be higher in the controls than in both male and female patients. The increased elemental composition in patients may be due to the fact that some proteins expressed in the body due to natural defensive mechanisms provided additional binding sites for these metal elements. The trend shown for K and Fe is in line with what was reported in a clinical practice (Fischbach, 1988); their concentrations in the patient's body deviated from the normal range.

## Influence of gender on the elemental composition in the patients' blood and head hair

In order to study the effect of gender in elemental distribution among the group of people with behavioural disorder, a group of male and female was selected randomly from both patients as well as the healthy group (control). For the blood samples analyzed, the concentrations of most elements were found to be higher in males than in females with the exception of Zn, Ca and V (Table 2). Tables 3 shows that, majority of the elements had higher levels in female hair samples than in male hair samples, an indication that women may be at greater health risk than men because hair specimens give a measure of long term toxic effect. (Schuhmacher , 1996) This trend observed with hair specimen may be due to increased gastrointestinal uptake of metals such as lead and cadmium at low iron stores; a common scenario with women at childbearing age (Chen et al., 2006; Moon et al., 2007; Ashraf et al., 1994; Barany et al., 2002; Vahter et al., 2004; Sharma et al., 2004). The observed variation in the metal concentration patterns occurred mainly among the patients, pointing to the possibility of the involvement of these metals in the sickness.

## Influence of age on the metal elemental composition in the patients' blood and hair

The results pertaining to the influence of age on the concentration pattern of the elements in the hair are depicted in Table 4. The studied population (for patients and controls) was divided into two age groups; (under 30 years old and 31 to 60 years old). The results in Table 4 reveals that, for most of the elements analyzed, higher levels were recorded in the 31 to 60 years old group (n =

**Table 3.** Gender based elemental distribution pattern in patients' head hair samples.

| Element | Patients (µg/g) | | | Control (µg/g) | | |
|---|---|---|---|---|---|---|
| | Male | Female | Difference | Male | Female | Difference |
| Al | 12.47 ± 6.00 | 17.70 ± 0.70 | 5.23 | 14.31 ± 1.89 | 14.58 ± 3.13 | 0.27 |
| Ba | 41.60 ± 4.00 | 62.29 ± 2.12 | 20.69 | 14.13±1.90 | 15.17 ± 1.08 | 1.04 |
| Be | 0.12 ± 0.05 | 0.12 ± 0.06 | 0 | 0.09 ± 0.07 | 0.06 ± 0.06 | 0.03 |
| Cd | 0.17 ± 0.01 | 0.36 ± 0.07 | 0.19 | 0.38 ± 0.06 | 0.16 ± 0.04 | 0.22 |
| Cr | 4.34 ± 4.91 | 8.32 ± 0.20 | 3.98 | 1.85 ± 0.30 | 1.11 ± 0.06 | 0.74 |
| Li | 0.03 ± 0.002 | 0.04 ± 0.001 | 0.01 | 0.02 ± 0.01 | 0.014 ± 0.001 | 0.006 |
| Sr | 5.10 ± 0.78 | 6.09 ± 0.37 | 0.99 | 1.35 ± 0.04 | 2.48 ± 0.79 | 1.13 |
| Zn | 132.16 ± 4.40 | 123.09 ± 6.58 | 9.07 | 95.73 ± 1.52 | 78.29 ± 3.65 | 17.44 |
| Mg | 156.57 ± 5.33 | 253.75 ± 3.83 | 97.18 | 40.31 ± 2.63 | 199.04 ± 2.17 | 158.73 |
| K | 77.73 ± 4.56 | 149.46 ± 3.18 | 71.73 | 25.43 ± 1.46 | 117.02 ± 3.65 | 91.53 |
| Fe | 49.46 ± 3.90 | 62.37 ± 0.54 | 12.91 | 49.45 ± 0.95 | 19.51 ± 0.89 | 19.94 |
| Na | 318.99 ± 4.36 | 646.12 ± 4.24 | 327.13 | 136.39 ± 1.21 | 282.15 ± 4.41 | 0.24 |

**Table 4.** Age based elemental distribution pattern in the patients' head hair samples.

| Element | Patients head hair (µg/g) | | Control head hair (µg/g) | |
|---|---|---|---|---|
| | Below 30 years | 31 to 60 years | Below 30 years | 31 to 60 years |
| Al | 17.20 ± 1.70 | 15.46 ± 1.40 | 17.09 ± 2.20 | 12.88 ± 1.28 |
| Ba | 45.25 ± 4.70 | 57.66 ± 5.05 | 11.98 ± 1.20 | 16.15 ± 1.27 |
| Be | 0.11 ± 0.005 | 0.12 ± 0.006 | 0.59 ± 0.006 | 0.09 ± 0.005 |
| Cd | 0.24 ± 0.018 | 0.33 ± 0.02 | 0.14 ± 0.01 | 0.44 ± 0.01 |
| Cr | 4.01 ± 0.61 | 8.80 ± 0.32 | 1.34 ± 0.04 | 1.87 ± 0.07 |
| Fe | 56.95 ± 4.50 | 59.04 ± 2..91 | 41.52 ± 1.01 | 45.53 ± 2.50 |
| Li | 0.03 ± 0.002 | 0.04 ± 0.002 | 0.13 ± 0.02 | 0.16 ± 0.017 |
| Sr | 6.10 ± 0.90 | 5.59 ± 1.11 | 1.80 ± 0.08 | 1.66 ± 0.070 |
| Zn | 156.57 ± 4.38 | 112.68 ± 5.01 | 77.63 ± 5.49 | 98.45 ± 3.80 |
| Mg | 220.78 ± 2.17 | 223.44 ± 2.05 | 99.80 ± 1.00 | 77.16 ± 1.29 |
| K | 100.34 ± 1.99 | 141.83 ± 1.53 | 36.23 ± 3.60 | 62.14 ± 5.28 |
| Na | 420.36 ± 3.29 | 612.36 ± 5.15 | 41.52 ± 1.01 | 182.94 ± 2.29 |

24) than under 30 years interval (n = 14) with the exception of Al, Be Sr, Ca and Mg. Similar results were previously reported by other researchers (Schuhmacher et al., 1996; Nowak, 1998). The low levels of Ca and Mg is attributed to these elements being mostly responsible for bone formation which normally takes place in individuals who are still growing (under 30 years old). For the older generation, there is almost no active bone formation, hence, no accumulation of Ca and Mg.

Aluminum was found to be higher in the hair samples within the 31 to 60 age group (Table 4) while levels of Al in blood samples were higher in the younger generation (under 30 years old) than in the older one (Table 5) and the correlation of its accumulation to the normal neurological function is supported by the work of Capel et al. (1981). The concentration levels of some elements such as Sr in the blood seemed to be independent of age bracket, since the concentration levels assessed in the patients and in the control (of all age groups) were almost similar.

In general, the younger age group (below 30 years old) represents a group that is known to be more susceptible to the effects of some toxic metals because they accumulate several times the percent ingested compared with adults (Pamphlett et al., 1997; Kristiansen et al., 1997). Tables 6 and 7 give the comparison of the data obtained in this work with the reported literature values. There are some differences in terms of the reported levels of metals accumulated that are obvious in the comparison. This may be due to the fact that, elemental composition of head hair samples or body tissue may vary as a result of natural variance of head hair composition such as hair colour and texture. With respect to other tissues of body fluid such as blood, the variations may emanate from dietary related factors.

The data obtained in this work were also validated statistically at confidence levels of $p = 0.01$ and at $p = 0.05$ and then, based on the data obtained the paired t-

**Table 5.** Age elemental distribution pattern in patients' blood samples.

| Element | Patients blood (µg/g) | | Control blood (µg/g) | |
|---|---|---|---|---|
| | Below 30 years | 31 to 60 years | Below 30 years | 31 to 60 years |
| Al | 4.83 ± 0.35 | 4.97 ± 0.10 | 2.57 ± 0.64 | 3.35 ± 0.60 |
| Ba | 1.16 ± 0.06 | 2.69 ± 0.10 | 1.04 ± 0.07 | 1.72 ± 0.10 |
| Be | 0.27 ± 0.01 | 0.29 ± 0.07 | 0.19 ± 0.02 | 0.2 ± 0.06 |
| Cd | 0.13 ± 0.02 | 0.127 ± 0.03 | 0.20 ± 0.03 | 0.10 ± 0.05 |
| Cr | 0.59 ± 0.06 | 1.61 ± 0.10 | 0.21 ± 0.03 | 0.36 ± 0.03 |
| Fe | 1913.15 ± 16.96 | 1898.02 ± 15.62 | 1456.64 ± 16.58 | 1595.70 ± 13.21 |
| K | 5714.65 ± 15.49 | 4656.87 ± 22.94 | 6181.38 ± 26.89 | 7277.24 ± 22.66 |
| Li | 0.03±0.01 | 0.04 ± 0.01 | 0.038 ± 0.002 | 0.043 ± 0.005 |
| Mg | 135.52 ± 6.54 | 133.02 ± 3.26 | 74.00 ± 5.31 | 86.92 ± 8.62 |
| Sr | 0.33 ± 0.01 | 0.33 ± 0.10 | 0.23 ± 0.01 | 0.29 ± 0.02 |
| V | 3.46 ± 0.15 | 3.73 ± 0.84 | 2.24 ± 0.15 | 2.62 ± 0.08 |

**Table 6.** A comparison of some of the elemental concentration ranges in whole blood (n = 3) obtained in this work and from the literature for similar elements.

| Element | This Work ( µg/g) | Literature ( µg/g ) | Reference |
|---|---|---|---|
| Al | 2 - 5 | 1.23 | El-Amri et al. (1994) |
| Be | 0 - 0.3 | - | - |
| Cr | 0.1 – 1.6 | 0.36 | El-Amri et al. (1994) |
| | | 0.028 | Iyengar et al. (1984) |
| Fe | 1400 - 1900 | 536 | Teresa et al. (1997) |
| | | 32.70 | El-Amri et al. (1994) |
| Zn | 10 - 26 | 20.70 | Zhuk et al. (1997) |
| | | 17.0 | Iyengar et al. (1984) |

**Table 7.** A comparison of some of the elemental concentration ranges in head hair samples (n = 3) from this work and from the literature.

| Element | This work ( µg/g) | Literature ( µg/g) | Reference |
|---|---|---|---|
| Al | 12 - 17 | 14.94 | Chojnacka et al. (2005) |
| | | 9.48 | Oluwole et al. (1994) |
| Cd | 0.1 – 0.4 | 0.114 | Chojnacka et al. (2005) |
| | | 12.10 | Popko et al. (2003) |
| | | 2.81 | Nowak (1995) |
| | | 0.61 | Teresa et al. (1997) |
| Zn | 77 - 78 | 156.48 | Chojnacka et al. (2005) |
| | | 128.04 | Nowak (1995) |
| | | 159.00 | Teresa et al. (1997) |
| Cr | 1- 9 | 0.568 | Chojnacka et al. (2005) |
| | | 0.60 | Oluwole et al. (1994) |
| | | 0.12 | Teresa et al. (1997) |
| Fe | 19 - 62 | 45.70 | Nowak (1995) |
| | | 29-84 | Teresa et al. (1997) |
| Na | 41 - 641 | 217.33 | Chojnacka et al. (2005) |
| | | 242.16 | Oluwole et al. (1994) |
| | | 165.9 | Nowak (1995) |
| K | 25 - 149 | 71.77 | Nowak (1995) |

**Table 8.** Statistical correlation among elements in patients and control samples at p = 0.01 and 0.05.

| Element | Correlating elements: Statistically significant ( p = 0.01) | | | | Correlating elements: Statistically significant (p = 0.05 | | | |
|---|---|---|---|---|---|---|---|---|
| | Hair (control), r ≥ 0.4 | Blood (control), r ≥ 0.4 | Hair (patients), r ≥ 0.38 | Blood (patients), r ≥ 0.38 | Hair (control) r < 0.35 ≥ 0.314 | Blood (control), r < 0.4 ≥ 314 | Hair (patients), r < 0.37 ≥272 | Blood (patients), r < 0.35 ≥ 0.276 |
| Al | Ba,Be,Cr,K,Sr,Fe,Li, V, Mg, Zn | Ba,Be,Cr,Fe,K,Li,Mg,, Sr,Zn,V | Be, Cd , Cr, Fe, Li, | Ba,Be, Cr, Fe ,Li, Sr,V | NC | NC | NC | NC |
| Ba | Al ,Fe,Be,K, Mg Sr,Zn,V | Al,Be,Fe,K,Mg,Sr,Zn, V | ND | Al, Ba.Cr, Fe , Ca ,Li, Sr,V | Cr,Li | Cr, Li | ND | NC |
| Be | Al Fe,Ba,K,Li,Mg,Sr, Zn,V | Al,Ba,Fe,K,Li,Mg , Sr, Zn,V | Al , Fe, Li | Al,Ba,Cr,Fe,Li,, Sr,V,Ca | NC | NC | ND | NC |
| Cd | NC | NC | Al,Cr,Fe,Li, K, Na | NC | NC | NC | Sr,Mg | Fe, |
| Cr | Al | Al, | Al, Cd,Fe,Li | Al , Ba,Be, Li, Sr,V | Ba , Zn | Ba ,Zn | NC | Ca |
| Cu | NC | NC | ND | ND | NC | NC | ND | ND |
| Fe | Al,Ba,Be,K,Li,Mg, ,Sr,Zn,V | AL,Ba,Be,K,Li,Mg,Sr, Zn,V | Al, Be , Cd,Cr,Li | Al Ba ,Be, Li , Sr ,V,Ca | NC | NC | NC | Cd |
| K | Al, Ba ,Be, Fe, Li , Mg, Sr, Zn,V | Al, Ba ,Be,,Fe,Li, Mg, Sr, Zn, V | Cd, Li, Na, Mg | Ca | NC | NC | NC | Mg |
| Li | Al,Be,Fe,K,Mg,Sr, Zn,V | Al,Be,Fe,K,Mg,Sr,Zn, V | Al,Be,Cd,Cr,Fe, Sr, K,Na | Al,Be,Ba,Cr,Fe, Sr,V,Ca | Ba | Ba | Mg | NC |
| Mg | Al,Ba,Be,Fe,K,Li,Sr, Zn,V | Al,Ba,Be,Fe,k,Li,Sr,Zn ,V | ,Sr,Na | NC | NC | NC | Li | Fe , Ca , K |
| Sr | Al,Ba,Be,Fe,K,Mg, Zn, V | Al,Ba,Be,Fe,K,Li,Mg, Zn,V | Mg,Na, Li | Al,Ba,Be,Cr,Fe,Li,V, Ca | NC | NC | Cd | NC |
| Zn | Al,Ba,Be,Fe,K,Li, Mg,Sr,V | Al,Ba,Be,Fe,K,Li,Mg, Sr ,V | ND | ND | Cr | Cr | ND | ND |
| V | Al,Ba,Be,Fe,K,Li, Sr ,Mg ,Zn | Al,Ba,Be,Fe,K,Li,Mg ,Sr,Zn | ND | Al,Ba,Be,Cr,Fe,Li, Sr,Ca | NC | NC | ND | NC |
| Na | ND | ND | Cd, Li, Sr, Mg ,K | ND | ND | ND | NC | ND |
| Ca | ND | ND | ND | Ba ,Be ,Fe ,Li , Sr, V,K | ND | ND | ND | Cr , Mg |

ND = not determined; NC = not correlating.

test and analysis of variance (ANOVA) statistical analyses using SPSS was utilized as data reduction techniques to generate a visual data for qualitative evaluation on the link between metal presence and the behavioural disorder as well as the influence of gender and age for the tested the samples. The results in Table 8 indicate that the levels of a number of elements were significant at the probability levels tested. However, although, the differences of the content between the samples from different groups (gender and age) for some metals were obvious, it may be difficult still to directly link their toxicity to the mental behavioural disorder, as more other factors such as metal analysis of the diet taken by the individuals, environmental (air and water) analysis of metals in areas they live are not included in this work. Never-theless, one may still theorize that the link may exist based on the data obtained for the patients versus the healthy individuals.

## Conclusions

The study revealed that, some toxic metals such as cadmium and aluminum may be linked to some forms of behavioural disorder as the elemental distribution of these toxic metals was shown to vary between the patients and the healthy individuals from the same locality. Gender and age appeared to have a strong influence on the distribution pattern for some elements and for the

type of specimen; blood and hair.

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### REFERENCES

Adams JB, Holloway CE, George F, Quig D (2006). Analyses of toxic metals and essential minerals in the head hair of arizona children with autism and associated conditions, and their mothers. Biol. Trace Element Res. 110: 193-209.

Ashraf M, Jaffar M, Tariq J (1994). Copper, iron, lead and zinc concentration distribution and correlation in fish from fresh-water lakes, Pakistan. Toxicol. Environ. Chem. 42: 93-98.

Barany E, Bergdahl IA, Bratteby LE, Lundh T, Samuel-son G, Schutz A, Skerfving S, Oskarsson A (2002). Trace element levels in whole blood and serum from Swedish adolescents, Sci. Total Environ. 286: 129-141.

Barbosa F, Lima EC, Zanão RA, Krug FJ (2001). The use of a W-Rh permanent modifier for direct determination of bismuth in urine and whole blood by electrothermal atomic absorption spectrometry. J. Anal. Atomic Spectrom. 16: 842-846.

Barbosa F, Palmer CD, Krug FJ, Parsons PJ (2004). Determination of total mercury in whole blood by flow injection cold vapor atomic absorption spectrometry with room temperature digestion using tetramethylammonium hydroxide. J. Anal. At. Spectrom. 19: 1000 - 1005.

Batista BL, Rodrigues JL, Nunes JA, Tormen L, Curtius AJ, Barbosa F (2008). Simultaneous determination of Cd, Cu, Ni, Pb and Zn in nail samples by inductively coupled plasma mass spectrometry (ICP-MS) after tetramethylammonium hydroxide solubilization at room temperature: Comparison with ETAAS. Talanta, 76: 575-579.

Becker JS, Dietze H-J (1999). Application of double-focusing sector field ICP mass spectrometry with shielded torch using different nebulizers for ultratrace and precise isotope analysis of long-lived radionuclides - Invited lecture. J. Anal. Atomic Spectrom. 14: 1493-1500.

Bowler RM, Gysens S, Diamond E, Nakagawa S, Drezgic M, Roels, HA (2006). Manganese exposure: Neuropsychological and neurological symptoms and effects in welders. Neurotoxicology, 27: 315-326.

Capel ID, Pinnock MH, Dorrell HM, Williams DC, Grant ECG (1981). Comparison of concentrations of some trace, bulk, and toxic metals in the hair of normal and dyslexic-children. Clinical Chem. 27: 879-881.

Chen C, Lin D, Wei Y (2006). Serum Sample levels of bromine, Iron , Scandium and Zinc in preschool children of Atalal and Bunum aborigines living in central Taiwan. J. Radioanal. Nucl. Chem. 268: 83-90.

Chojnacka K, Górecka H, Chojnacki A, Górecki H (2005). Inter-element interactions in human hair. Environ. Toxicol. Pharmacol. 20:368–374.

Christensen JM (1995). Human exposure to toxic metals: factors influencing interpretation of biomonitoring results. Sci. Total Environ. 166: 89-135.

Cornelis R, Sabbioni E, Van der Venne MT (1994). Trace element reference values in tissues from inhabitants of the European Community. VII. Reviez of traces elements in blood, serum, and urine of the Belgian population and critical evaluation of their possible use as reference values. Sci. Total Environ. 158: 191-226.

Dombova´ri J, Pappa L, Uzonyi I, Borbe´ly-Kiss I, Elekes Z, Varga Z, Ma´tyus J, Kakuk G (1999). Study of cross-sectional and longitudinal distribution of some major and minor elements in the hair samples of haemodialysed patients with micro-PIXE. J. Anal. Atomic Spectrom. 14: 553-557.

Eck P, Wilson L (1989). Toxic metals in human health and disease, Eck Institute of Applied Nutrition and Bioenergetics, Ltd., Phoenix, AZ.

El-Amri FA (1994). Determination of trace-elements in normal and diabetic whole-blood by neutron-activation analysis. Biol. Trace Element Res. 43(5): 383-388.

Fischbach F (1988). A Manual of Laboratory Diagnostic Tests, 3rd edn, ed. D. Inteno, J.B. Lippincott Co., Philadelphia pp: 41, 46, 66, 97, 159.

Gil F, Capitan-Vallvey LF, de Santiago E, Ballesta J, Pla A, Hernadez AF, Gutierrez-Bedmar M, Fernadez-Crehuet J, Gómez J, Lopez-Guarnido O, Rodrigo L, Villanueva E (2006). Heavy metal concentrations in the general population of Andalusia, South of Spain: a comparison with the population within the area of influence of Aznalcóllar mine spill (SW Spain). Sci. Total Environ. 372: 49-57.

Goullé JPM, Mahieu L, Castermant J, Neveu N, Bonneau L, Bouige D, Lacroix C (2005). Metal and metalloid multi-elementary ICP-MS validation in whole blood, plasma, urine and hair: Reference values. Forens. Sci. Intern. 153: 39-44.

Ibrahim D, Froberg B, Wolf A, Rusyniak DE (2006). Heavy metal poisoning: Clinical presentations and pathophysiology. Clin. Lab. Med. 26: 67–97.

Iyengar GV (1984). Reference values for elemental concentrations in some human samples of clinical interest - A preliminary evaluation. Sci. Total Environ. 38: 125-131.

Kristiansen J, Christensen JM, Iversen BS, Sabbioni E (1997). Toxic trace element reference levels in blood and urine: influence of gender and lifestyle factors**.** Sci. Total Environ. 204: 147-160.

Kucera J, Bencko V, Sabbioni E, Van der Venne MT (1995). Review of trace elements in blood, serum and urine for the Czech and Slovak populations and critical evaluation of their possible use as reference values. Sci. Total Environ. 166: 211-234.

Loureiro VR, Saleh MAD, Moraes PM, Neves RCF, Silva FA, Padilha CCF, Padilha PM (2007). Manganese determination by GFAAS in Feces and fish feed slurries. J. Braz. Chem. Soc. 18: 1235-1241.

Minoia C, Sabbioni E, Apostoli P, Pietra R, Pozzoli L, Gallorini M, Nicolaou G, Alessio L, Capodaglio E (1990). Trace element reference values in tissues from inhabitants of the European Community. I. A study of 46 elements in urine, blood and serum of Italian subjects. Sci. Total Environ. 95: 89-103.

Moon J, Chung Y, Park K, Lee O (2007). Serum analysis of coronary heart disease patients by Instrumental Neutron Activation Analysis. J. Radioanal. Nucl. Chem. 272: 409-412.

Nowak B (1995). Occurrence of heavy metals and sodium, potassium and calcium in human teeth. Analyst 120: 747-750.

Nowak B (1998). Contents and relationship of elements in human hair for a non- industrialized population in Poland. Sci. Total Environ. 209: 59-68.

Oluwole AF, Ojo JO, Durosimi MA, Asubiojo OI, Akanle OA, Spyrou NM, Filby RH (1994). Elemental composition of head hair and fingernails of some Nigerian subjects. Biol. Trace Element Res. Editor: G N Schrauzer .Humana Press Inc. pp: 443-451.

Palmer CD, Lewis ME, Geraghty CM, Barbosa F, Parsons PJ (2006). Determination of lead, cadmium and mercury in blood for assessment of environmental exposure: A comparison between inductively coupled plasma-mass spectrometry and atomic absorption spectrometry. Spectrochim. Acta Part B, 61: 980-990.

Pamphlett R, Ewan KBR, Mcquilty R, Waley P (1997). Gender Differences in the Uptake of Inorganic Mercury by Motor Neurons.

Parsons PJ, Barbosa F (2007). Atomic spectrometry and trends in clinical laboratory medicine. Spectrochim. Acta Part B, 62: 992-.1003.

Parsons PJ, Slavin W (1993). A rapid Zeeman graphite-furnace atomic-absorption spectrometric method for the determination of lead in blood. Spectrochim. Acta Part B, 48: 925-939.

Pfeiffer C (1975). Mental and elemental nutrients, Keats Publishing, New Canaan, CT.

Popko J, Olszewski S, Hukałowicz K, Markiewicz R, Borawska MH,

Szeparowicz P (2003). Lead, cadmium, copper and zinc concentrations in blood and hair of mothers of children with locomotor system malformations. Polish J. Environ. Studies 12: 375-379.
Rodrigues JL, Nunes JA, Batista BL, Souza SS, Barbosa F (2008). A fast method for the determination of 16 elements in hair samples by inductively coupled plasma mass spectrometry (ICP-MS) with tetramethylammonium hydroxide solubilization at room temperature. J. Anal. Atomic Spectrom. 23: 992-996.
Schuhmacher M, Belles M, Rico A, Domingo JL, Corbella J (1996). Impact of reduction of lead in gasoline on the blood and hair lead levels in the population of Tarragona Province, Spain 1990-1995. Sci. Total Environ. 184: 203-209.
Sharma R, Singh LC, Tanveer S, Verghese P, Kumar A (2004). Trace elements contents in human head hair of residents from Agra City, India. Bull. Environ. Contam. Toxicol. 72: 530-534.
Teresa M, Vasconcelos SD, Tavares HMF (1997). Trace elements concentration in blood and hair of young apprentices of a technical professional school. Sci. Total Environ. 205: 189-199.
Vahter M, Berglund M, Åkesson A (2004). Toxic metals and the menopause. Menopause Int. 10: 60-65.
Watts DL (1990). Trace elements and neuropsychological problems as reflected in Tissue Mineral Analysis (TMA) patterns. J. Orthomol. Med. 5: 159-166.
Yuan WH, Holland SK, Cecil KM, Dietrich KN, Wessel SD, Altaye M, Hornung RW, Ris MD, Egelhoff JC, Lanphear BP (2006). The impact of early childhood lead exposure on brain organization: a functional magnetic resonance imaging study of language function. Pediatrics. 118: 971-977.
Zanão RA, Barbosa F, Souza SS, Krug FJ, Abdalla AL (2002). Direct determination of selenium in whole blood by electrothermal atomic absorption spectrometry using W-Rh-coated platform and co-injection of Rh as thermal stabilizer. Spectrochim. Acta Part B, 57: 291-301.