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Trace element profiles of human hair samples collected from a non-occupationally unexposed population in Nagbhid, Maharashtra (India)

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Abstract
The present study demonstrates; mean concentrations of trace elements in the hair samples collected amongst the occupationally and environmentally unexposed human population residing in Nagbhid taluka of Chandtapur district in Maharashtra (India) shows the following order for male subjects: Zn>Fe>Cu>Co>Ni>Mn>Cr>Cd>Pb>As>Hg; while in female subject, values of Arsenic (As) are slightly higher than lead (Pb). The elemental analysis of hair samples indicates zinc (Zn) as the most abundant trace element, while mercury (Hg) as least found trace element. Defining baseline levels of trace elements in hair and nail samples for the general unexposed human populations is challenging due to significant variability based on factors such as geographical locality, diet, age, and sex of individuals. Therefore, a single normal range does not exist, and multiple reports different mean concentrations for a non-occupationally exposed or control group populations can serve as a reference rage for the purpose of comparison with values from exposed population The values of trace elements in hair samples obtained by Atomic Absorption Spectroscopy (AAS) in present study will act as baseline reference values for further studies to assess the impact of pollution due to industries and mining activities in Chandrapur district.

Keywords: *Hair, trace elements, emission.*

Introduction

The health assessment of community with regards to the exposure to different kind of toxic trace element becomes realistic by the measure of the amount of the contaminant actually present in the human body, as it is realized by human biomonitoring, which is the direct measurement of individual's exposure to the environmental contaminants by measuring them in human specimens blood, serum, urine, hair, nail etc. [1-3]. Nutritionally, essential trace elements play a vital role in body functions, including growth, development and metabolism in the human body. They need to be consumed through diet, as they cannot be synthesized by the body. The essential elements include Zn, Cu, Fe, Mn, Ni and Co participates in a wide range of enzymatic processes involving many aspects of the intermediary metabolism and physiological functions of the organism. [4,5]. Although needed in tiny quantities, they also play vital roles in maintaining health, and deficiencies or excesses can lead to health problems. [6-7].

The biochemical function of trace elements, appears as co-enzyme, binds to the enzyme's active site and can act as carrier for electron. Alterations in trace element may results in the development of physiological disturbances which may leads to diseases. [8].

The only data obtained by the environmental monitoring not been able to provide a complete characterization of the human exposure and then, of human health risks. First because a comprehensive knowledge of different exposure ways of toxic trace elements (TTE), capacity of their absorption and individual susceptibilities to these toxic elements were not been possible by environmental monitoring. [2]. The use of biological samples to diagnose, detect, and monitor acute and chronic diseases has revolutionized medical practice as it provide necessary insight to treat patient precisely. [8-9].

Multiple studies on biomonitoring of trace elements in biological samples were undertaken to assess environmental or occupational exposure to metals like arsenic, lead, copper, and zinc by measuring their concentrations in accessible human samples such as blood, urine, hair, and nails. [4,6,10,11]. It serves as a diagnostic and toxicological tool to explore health risks, which were seen as guidelines to chalk out public health policies.

The trace elements are not fixed in each part, but are constantly being transferred from one to another as the parts are neither in equilibrium with each other nor with the temporary storage sites. Nonetheless, in the era of precision medicine, the preferred scientific use of hair analyses and testing as a tool for forensic toxicology and medico-legal purposes over other biomarkers like urine and blood is due to the its metabolically inert nature. [4,10,12,13]. Unlike blood, hair is homogeneous and metabolically inert, that is a mature hair shaft is homogeneous and metabolically inert because its cells are dead and fully keratinized by the time they emerge from the skin. Each section of hair contains chemical information from the time it was formed, providing a retrospective image of exposure to different contaminants in past life. [10,14].

In view of the above mentioned discussion on fluctuations in the concentrations of trace elements, and different influencing factors for such variations, hair emerges as important biomarker in the biomonitoring studies in India and abroad. The present study was undertaken with an aim to assess fluctuations of trace elements in Nagbhid taluka of Chandrapur district of Maharashtra in India which represents occupationally and environmentally unexposed human population, as it is lacking any industry or mining activity which otherwise contaminates air, soil and water.

Material and Methods

Study Site

Administratively, the Chandrapur district in Maharashtra is now having 15 talukas which includes, Chandrapur, Bhadravati, Warora, Chimur, Nagbhir, Bramhpuri, Sindhewahi, Mul, Gondpipri, Pomburna, Saoli, Rajura, Korpana, Jivati and Balharshah. Most of the industries and coal mines are located within the 50 km. periphery of Chandrapur city, while Nagbhid, Bramhapuri and Sindewahi taluka do not have any kind of industry and mining activities. Volunteers for the present study were selected from male and female subjects residing in less polluted Nagbhid Taluka, who were undergone routine check up like blood pressure and Hb count and were healthy with no lifetime history or current diagnosis of any disorder.

Collection of hair samples and preparation of sample for AAS spectroscopy

Prior to the collection of the sample from the subjects, survey of the study area was undertaken to assess the health status, socio-economic background and surrounding hygienic condition. For ethical and legal reasons and to get voluntary participation in the hair and nail sample, subjects underwent clinical interview and were provided questionnaires asking details regarding their personal and medical history. The information required included gender, age, BMI, place of residence, occupation, possible exposure to harmful trace elements, and medical treatment. Most of the data obtained through questionnaire as recommended by World Health Organization (WHO) was in the form of multiple choice questions. After getting requisite data as regards to their health, subjects were informed about the objective of the study and permission thus obtained for the submission of the hair and nail samples for the assessment of trace elements. Accordingly, male and female subjects from different developmental stages, pre-puberty (age 11 to 13 yrs.), adolescent (age 18 to 20 yrs.), young adulthood (age 23-25 yrs.), middle adulthood (age 33 to 35 yrs.), middle aged adults (age 42 to 45 yrs.) and Late adulthood (age 58 to 60 yrs.) were selected. [15]. Trace elements were determined in samples of human hair collected between October, 2022 to November, 2023 from 30 male and 30 female inhabitants.

Preparation of nail samples for AAS

While collecting of hair specimens, care was taken to see that the hair samples were around 500 mg to 1 gm, as sample loss occurs during transfer, washing, and the other procedures involved. Hair samples of male and female subjects of age range (11-20, 21-35, 35-60) were collected from the nape of the scalp by cutting 2mm from the scalp using a pair of sterilized stainless-steel scissors washed with ethanol, a neutral solvent to remove external contamination and packed in cellophane bags. In the course of present study, scalp hair samples from pooled specimens obtained from a barber's shop in and around Nagbhid town and surrounding villages. These samples were brought to the laboratory for further procedures. The steps in the laboratory includes the washing of the samples, digestion of the samples, prior to estimation of elemental content by Atomic Absorption Spectroscopy (AAS).

Atomic Absorption Spectrophotometer (AAS) analysis of hair samples

For the determination of trace elements in the present study, flame Atomic Absorption Spectrophotometer (AAS)has been used. The Atomic Absorption Spectrophotometer used for measurement of trace elements (Cu,Zn,Co,Mn,Fe,Cr,Cd, Ni,As, Pb and Hg) is Shimadzu makeAA-7000F, 1.04 ROM version and with serial number - A30925501246 at Central Research Facility of Manipal University, Jaipur. For the purpose of atomisation, air-acetylene flame has been used. The lamps used for the elemental determination were hollow cathode lamps filled with Neon (Ne). The AAS determination of cations was performed under the recommended condition for each heavy metal.

Results and Discussion

In the present study values as obtained by Atomic Absorption Spectroscopy (AAS) for the mean concentration of trace elements in hair samples of male and female subjects from control group of occupationally environmentally un-exposed human population residing in Nagbhid taluka of Chandrapur district of Maharashtra (India). The Chandrapur district of Maharashtra is industrially leading in state as the raw material for coal-fired thermal power plants (CSTPS) and cement industry located within the periphery of 50 km to Chandrapur city. In addition, baseline level of mean concentrations of trace elements in scalp hair and nail samples of volunteers from control group human subjects residing in 'less polluted' area of rural agricultural landscape of Nagbhid taluka. Defining baseline levels of trace elements in hair and nail samples for the general unexposed human populations is challenging due to significant variability based on factors such as geographical locality, diet, age, and sex of individuals. Therefore, a single normal range does not exist, and multiple reports stating different mean concentrations for a non-occupationally exposed or control group populations can serve as a reference rage for the purpose of comparison with values from exposed population. [9,16,17,18].

Hair represents a pathway of excretion of substances from the human body that reflects health status of an individual. Each geographical area has a typical profile of hair elemental composition of its inhabitants. The level of trace elements in hair samples of human population not exposed to occupational hazards do not reflect healthy condition, but can be considered 'normal' and thus values obtained may be considered as reference values for comparing and interpreting laboratory results from occupationally exposed group of populations. [17-19]. The advantages of hair as a study material for the determination of mean concentration of trace elements have already been elaborated in introduction chapter. The easy collection, transportation, handling, and inert nature of keratin matrix makes the work of researcher more convenient. [14,20].

During present investigation on the mean concentration of trace elements of nail samples in control group of population residing in Nagbhid, shows the following order for male subjects: Zn>Fe>Cu>Co>Ni>Mn>Cr>Cd>Pb>As>Hg; while in female subject, values of Arsenic (As) are slightly higher than lead (Pb). Several research studies committed for the cause of elucidating the role of trace elements reveals that, man depends upon at least nine trace elements, iron, zinc, copper, manganese, iodine, chromium, selenium, molybdenum and cobalt for performance variety of functions including catalytic, structural and regulatory activities in which they interacts with macromolecules such as enzymes, pro-hormones, pre-secretory granules and biological membranes. [7, 21]. Zinc is an essential trace element that functions as cofactor for nearly 300 different enzymes and involved in the metabolism of proteins, carbohydrates and lipids. [16,22]. In this context results in our present investigations on the trace elements find its parallel in many other studies on unexposed human populations in India and abroad [18].

Table.1. Table showing concentrations of trace elements (µg/gm) in hair samples of both the male and female subjects residing Nagbhid (Control). (Mean± SE)

Trace Elements	11-13 Yrs		17-20 Yrs		23-25 Yrs		33-35 Yrs		42-45 Yrs		58-60 Yrs	
	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
Cu	4.45880±3.78	4.51140±3.80	6.12160 ± 4.4	6.20440± 4.5	8.26600± 5.1	8.55320± 5.2	10.24240± 5.7	10.63920± 5.8	13.61280 ±6.6	14.02060± 6.7	17.80560 ±7.5	18.05600±7.6
Zn	54.4886±13.2	56.39412±13.4	66.0170±14.5	71.3292±15.1	82.1780±16.2	85.8488±16.5	88.5108±16.8	93.0516±17.3	77.8852± 15.7	79.8489±15.9	59.9660±13.8	63.6000±14.3
Co	1.35640 ± 2.1	1.59520± 2.26	2.28760 ± 2.7	2.50160± 2.83	2.69840± 2.94	3.07288± 3.14	3.39320± 3.3	3.49420± 3.34	3.53500± 3.36	3.62320± 3.41	4.13060± 3.64	4.29540± 3.71
Mn	2.24900 ±2.68	2.50000± 2.83	3.12440 ±3.16	3.40660± 3.30	3.56240± 3.38	3.86840± 3.52	4.12340± 3.6	4.52540± 3.8	5.04860± 4.02	5.24860± 4.10	5.59580± 4.23	5.84220± 4.32
Fe	25.07800±8.96	24.37300±8.83	26.64720 ±9.2	25.19580± 8.9	36.64440±10.8	33.3816± 10.3	38.0538±11.0	36.8130± 10.8	35.0692±10.6	34.4068±10.5	31.2996±10.0	28.51060± 9.5
Cr	1.04860± 1.83	0.91840± 1.71	1.58680± 2.25	1.30380± 2.04	2.04140± 2.56	1.78860± 2.39	2.82740± 3.01	2.41820± 2.78	3.25640± 3.23	2.86560± 3.03	3.50860± 3.35	3.41160± 3.30
Cd	0.00080± 0.05	0.00054± 0.04	0.00786± 0.16	0.00499± 0.13	0.00959± 0.18	0.00818± 0.16	0.01835± 0.24	0.01365± 0.21	0.02733± 0.30	0.02429± 0.28	0.04516± 0.38	0.03128± 0.32
Ni	0.15040± 0.69	0.17380± 0.75	0.21720± 0.83	0.27740± 0.94	0.31240± 1.00	0.33066± 1.03	0.42840± 1.17	0.47240± 1.23	0.55680± 1.33	0.70240± 1.50	0.71920± 1.52	0.86420± 1.66
As	0.00096± 0.06	0.00076± 0.05	0.00190± 0.08	0.00149± 0.07	0.00594± 0.14	0.00364± 0.11	0.01144± 0.19	0.00863± 0.17	0.01527± 0.22	0.01181± 0.19	0.02088± 0.26	0.01630± 0.23
Pb	0.00462± 0.12	0.00311± 0.10	0.00785± 0.16	0.00523± 0.13	0.00988± 0.18	0.00789± 0.16	0.01134± 0.19	0.00962± 0.18	0.0203 ± 0.26	0.01882± 0.25	0.04058 ± 0.4	0.03152± 0.32
Hg	0.00011± 0.02	0.00007± 0.02	0.00027± 0.03	0.00023± 0.03	0.00074± 0.05	0.00047± 0.04	0.00183 ± 0.1	0.00113± 0.06	0.00527± 0.13	0.00458± 0.12	0.00764± 0.16	0.00496± 0.13

The concentration of trace elements in hair and nail can vary significantly based on geographical location and local environment. The values of trace elements concentration in our study on occupationally unexposed control human population from Nagbhid taluka in Chandrapur district serves as base-line values for interpreting results from exposed group of population. Gender of individuals had a significant influence on the concentration of trace elements in hair, mean concentrations of potentially toxic trace elements (TTE) like As, Cd, Pb, and Hg are mostly higher in male than female while with exception of Fe all other essential trace elements are comparatively higher in female than male subjects, these findings of ours find their similarities in many other studies in India and abroad. (Carneirro et al., 2023; Randive and Godbole, 2025).

In present investigation on trace elements in control group of human population, values of Cu, Zn, Co, Cr, Pb, and As were found comparatively higher in hair samples than in nail samples, while values of mean concentrations for Mn, Fe, Cd, Ni and Hg were comparatively higher in nail than hair. Hair is often considered better for monitoring long-term, low-level exposure to certain elements significantly impacted variation in the concentration of trace elements than in nail. [4,8,12,24]. Nevertheless, nail has certain advantages over hair, former biomaterial found to be less affected by external contamination. [11,23]. The measurement of trace elements in human nails has recently generated considerable interest as a biomarker for assessing exposure toxic trace elements due to anthropogenic activities. [11,23]. There are multiple research studies reporting sex-dependant differences in cobalt (Co) levels in hair and nail samples, though those vary with age, geographic location and lifestyle factors. [21]. During present investigation, in order to assess an influence of environmental exposure on human subjects and age-dependant shift on the values of trace elements in hair and nail samples were compared, accordingly, our findings indicates, values of mean concentrations of cobalt (Co) in hair and nail samples reveals interesting sex-dependant pattern, with values of cobalt (Co) in hair samples of female subjects were comparatively higher than male, conversely, values of cobalt in nail samples of male were comparatively higher than female. These findings of ours are in line with similar studies in India and abroad.[21]. Multiple studies on amino acid composition of alpha-keratin of nails reported that, in human males has more leucine (Leu) and alanine (Ala) and less Cys, Arg and Ile than in female nails, that provide binding matrix for cobalt. [3].

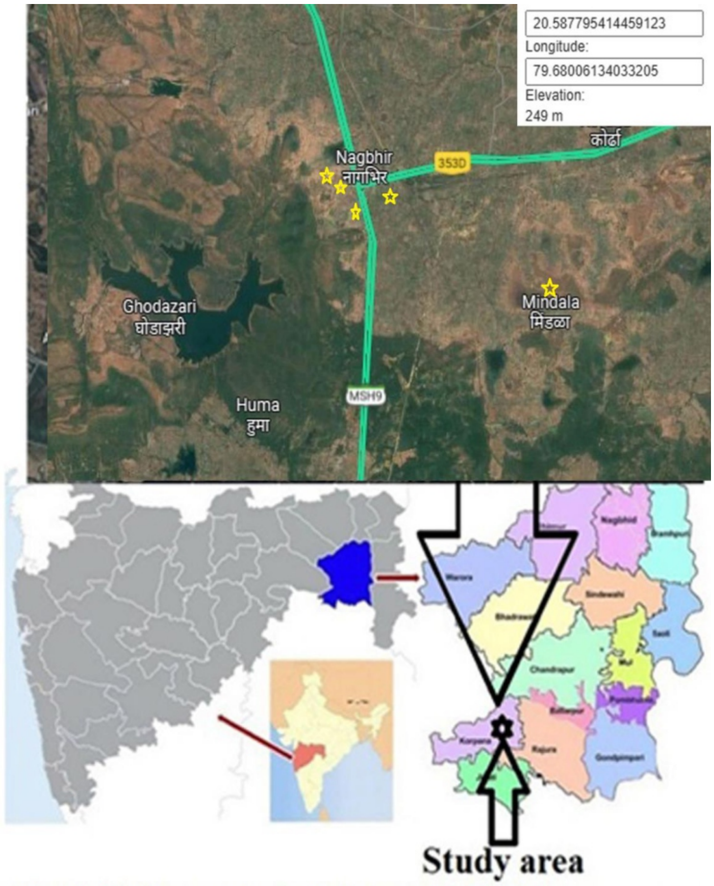

Fig.1. Study area in and around Nagbhid

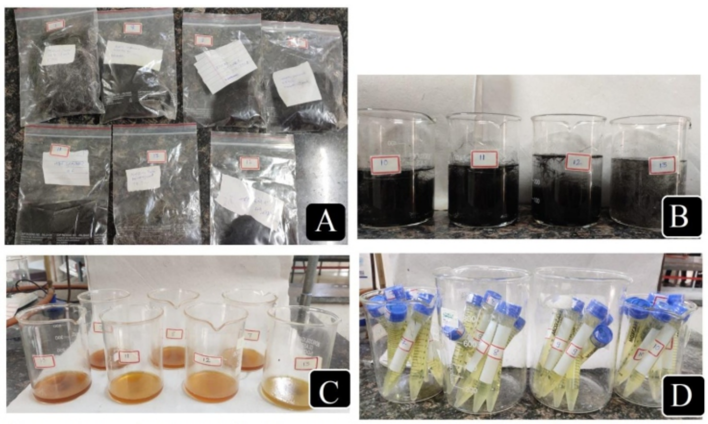

Fig. 2. Photos showing methodology used to prepare samples ready for elemental analysis
A. Collected hair samples from male and female subjects B. Washing of samples C. Digestion of hair samples D. Samples ready to be sent for AAS analysis of trace elements

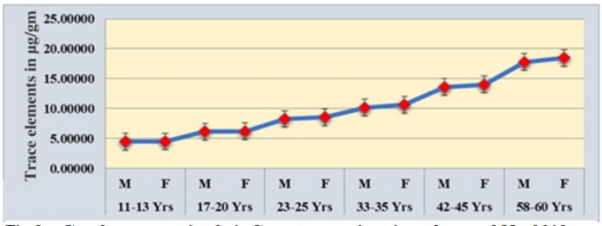

Fig.3a. Graph representing hair Cu concentrations in and around Nagbhid

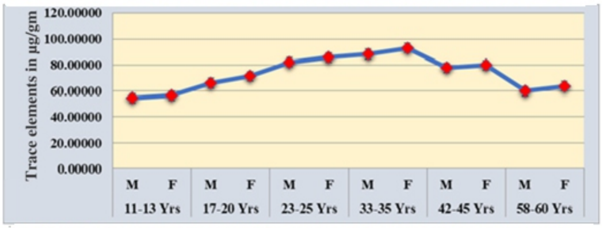

Fig.3b. Graph representing hair Zn concentrations in and around Nagbhid

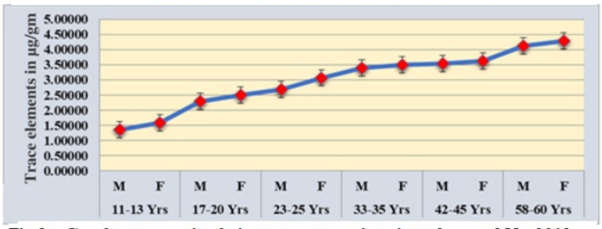

Fig.3c. Graph representing hair Co concentrations in and around Nagbhid

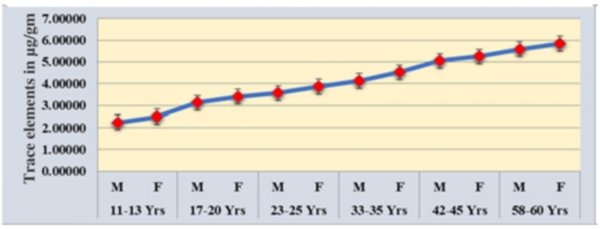

Fig.3d. Graph representing hair Mn concentrations in and around Nagbhid

Fig.3e. Graph representing hair Fe concentration in and around Nagbhid

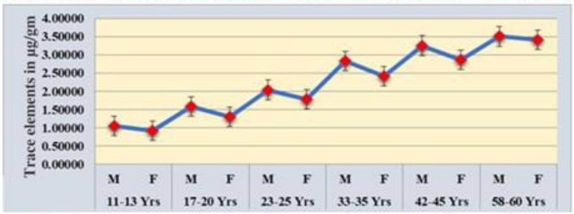

Fig.3f. Graph representing hair Cr concentration in and around Nagbhid

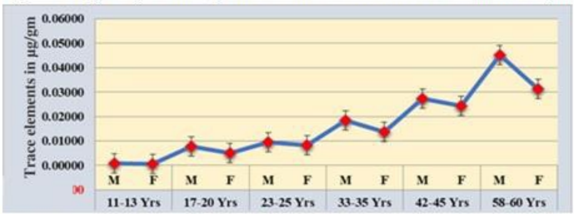

Fig.3e. Graph representing hair Cd concentration in and around Nagbhid

Fig.3h. Graph representing hair Ni concentration in and around Nagbhid

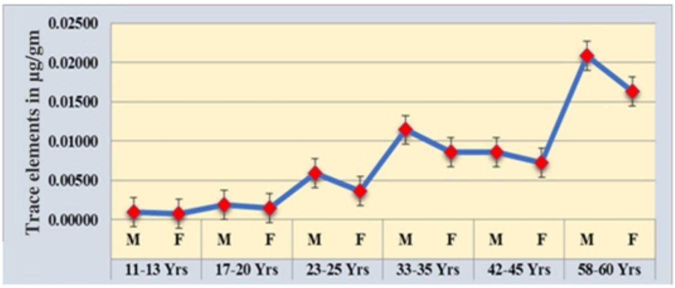

Fig.3i. Graph representing hair As concentrations in and around Nagbhid

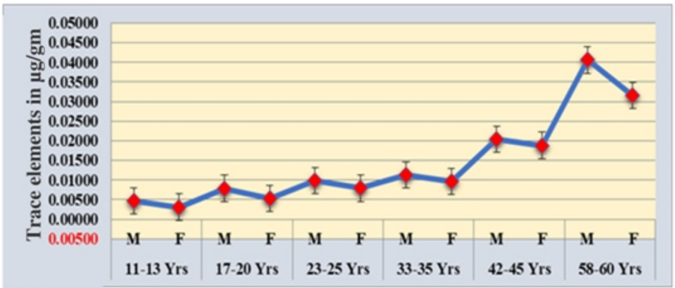

Fig.3j. Graph representing hair Pb concentrations in and around Nagbhid

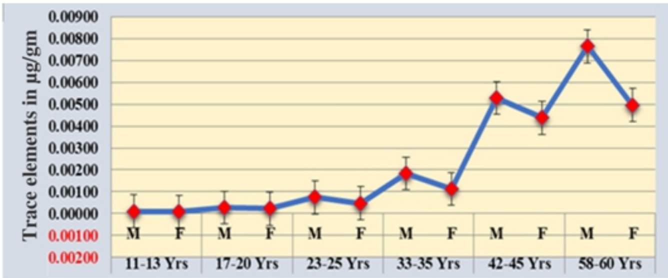

Fig3k. Graph representing hair Hg concentrations in and around Nagbhid

Conclusions

The present study demonstrates; mean concentrations of trace elements in the hair samples collected amongst the occupationally and environmentally unexposed human population residing in Nagbhid taluka of Chandtapur district in Maharashtra (India) shows the following order for male subjects: Zn>Fe>Cu>Co>Ni>Mn>Cr>Cd>Pb>As>Hg; while in female subject, values of Arsenic (As) are slightly higher than lead (Pb). The elemental analysis of hair samples indicates, zinc (Zn) as the most abundant trace element, while mercury (Hg) as least found trace element. The values of trace elements in hair samples obtained by Atomic Absorption Spectroscopy (AAS) in present study will act as baseline reference values for further studies to assess the impact of pollution due to industries and mining activities.

Abbreviations used in Figures: PP- Pre-puberty, AD- Adolescent, YA- Young Adulthood, MA – Middle Adulthood, MAA- Middle Aged Adults, LA-Late Adulthood

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