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Biomonitoring of 33 Trace Elements in Blood Samples from Inhabitants of Southern India by ICP-MS

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Abstract: ***Trace elemental levels have a profound effect on human health. However, only limited information is available on biomonitoring of trace elements in India. But due to rapid industrial development and urbanization, pollution levels have increased and therefore there is a need to monitor trace elemental concentrations in human beings. In this work 33 trace elements (Li, Be, Rb, Sr, Cs, Ba, V, Mn, Co, Ni, Cu, Zn, Se, La, Ce, Pr, Nd, Sm, Eu, Gd, Dy, Ho, Er, Tm, Yb, Lu, Th, Cd, Tl, Pb, U, As and Ag) in 98 human blood samples, collected from occupationally non-exposed volunteers, living in Greater Visakhapatnam in southern part of India, were monitored. The samples were analysed by inductively coupled plasma mass spectrometry (ICP–MS). The samples for analysis were prepared by treating with supra pure nitric acid and digesting in a microwave digester. The method of validation is described for all 33 elements and results about internal and external quality assurance are discussed. The information about the sample donors was collected by questionnaire-based interviews. Statistical data has been presented (mean values, geometric mean values, ranges and selected percentiles) for all elemental concentrations in blood.***

Keywords: Biomonitoring, Whole blood, Trace elements and ICP-MS

1. Introduction

Trace elements play an important role in biological processes and have profound effect on human health. Deficiency of essential trace elements or accumulation of potential toxic elements in the human body causes various diseases [1], [2] and [3]. But very little information is available on the concentration of the ranges of the trace elements, which can vary with age, sex, habits, geographical conditions, pollution levels and various other factors [4] Elements like Pb, Cd, Se, Zn, Hg and few others are measured frequently and reference values are well established, but elements like Be, Li, V, Rb, Sr, Co etc. along with others have to be studied and there is a need to establish the reference levels. Several studies on trace elements in India such as Fe, Co, Ni, Zn and Se were carried out by different researchers [5], [6] in serum samples. Fourteen minor and trace elements (Na, K, P, Fe, Br, Co, Cr, Cs, Hg, Rb, Sb, Se, Sr and Zn) have been determined in pre and postoperative blood samples of breast cancer affected subjects [7]. But none of them studied the other trace elements such as Li, Rb, Ce, V, etc.

Biomonitoring of trace elements in human body fluids is used as a diagnostic and toxicological indicator in human beings. The available media for Biomonitoring of trace elements in human beings are blood, plasma, serum, urine, saliva, nail and hair. In some cases breast milk was also used for the biomonitring studies in humans. According to Vivek Singh and Garg (1998), blood is the most reproducibly accessible body fluid analyzed for monitoring the trace elemental status in humans [7]. However few of the studies have been carried out using blood as a medium. Rodushkin et al. (1999) have determined 50 elements in blood samples of 31 human subjects from Sweden [8]. White and Sabbioni (1998) have determined seven elements in blood of unexposed British subjects [9]. Minoia et al. (1998) have determined 35 elements in blood of the Italian population [10]. Trace element reference values for blood were presented by Cornelis et al. (1994) for the Belgian population [11], by Poulsen et al. (1994) for the Danish population [12], by Kucera et al. (1995) for the Czech and Slowak population [13] and by Michael et al. (2004) for the German population [14], by Ivanenko et al.(2013) for the Russian population [4], by Xiaobing Liu et al.(2014) for the Chinese population [15]. The above studies concluded that reference values of the trace elements differ among different population groups.

According to Alessandro et. al the concentration ranges of the trace elements, which can vary with age, sex, habits, geographical conditions, pollution levels and various other factors [4]. The main aim of the study is to assess the trace elements levels in inhabitants of Greater Visakhapatnam city. The present study area have been surrounded by so many industries such as steel, casting industries, thermal power plant and so many number of industries has been established, Visakhapatnam is a coastal city, inhabitants of the city highly depends on sea food. Advancement in the multi elemental analysis instrumentation ICPMS and ICP-OES we are capable to determined trace elements simultaneously. Introduction of the reaction/ collision gas cell technique allows us to remove polyatomic interferences more efficiently and determination of trace elements simultaneously with good precision and accuracy in whole blood samples. In the present study we monitored 33 trace elements in 98 whole blood samples collected from different subjects living in Visakhapatnam city in Southern India. Samples we were digested using microwave accelerate reaction system. The digested blood samples were analysed using ICPMS with collision/reaction gas cell and the complete validation of the method along with internal and external quality assurance has been described. Statistical data about 33 trace elements are provided, which will be useful for the researchers working in the fields of clinical and toxicological studies.

2. Materials and Methods

Instrumentation:

The blood samples were analysed with an Agilent 7700s Inductively Coupled Plasma Mass Spectrometer with a 27.12MHz solid state generator. The instrument has an octapole - based collision / reaction cell with helium as collision and hydrogen as reaction cell gases. A flow of 4.4 mL/min helium (Indian gases, India) with a purity of 99.999% (V/V) was introduced into the cell. ^{9}Be, ^{51}V, ^{55}Mn, ^{59}Co, ^{60}Ni, ^{63}Cu, ^{66}Zn, ^{75}As, ^{82}Se, ^{85}Rb and ^{88}Sr were analysed in He gas mode and the remaining isotopes were analysed in no gas mode. Here Pt sampler and skimmer cones were used with an orifice diameter of 1.0 and 0.7mm, respectively and the sample introduction system was equipped with PFA micro flow concentric nebulizer combined with a Scott double pass spray chamber. The spray chamber was Peltier cooled at 2^{O}C to ensure temperature stability and to reduce the water vapour present in the nebulizer gas flow. The ICP torch consists of an injector tube with a large inner diameter of 2.5mm to reduce the risk of particle deposition or clogging. The instrument was optimized to get highest signal to background ratio with a solution containing ^{7}Li, ^{59}Co, ^{89}Y and ^{205}Tl along with the ratios $^{140}Ce^{16}O^{+}/^{140}Ce^{+} < 1.5\%$ and $^{140}Ce^{2+}/^{140}Ce^{+} < 5\%$ for a solution with 1µg L^{-1}. ICPMS instrument operating conditions were as follows RF power 1500w, outer gas flow 15 L min^{-1}, carrier gas flow 0.75 L min^{-1}, nebulizer gas flow 1.0 L min^{-1}, Nebulizer pump 0.1 rps, integration time 0.1 sec.

Sample collection and sample preparation:

Blood samples of 98 occupationally non-exposed human subjects were collected. The subjects were living in and around greater Visakhapatnam, a coastal city in southern India. This area has a population density of 384 inhabitant's km^{-2} and is close to many industrial establishments such as steel plant, casting industries, fertilizer plant, thermal power plant, rare earth extraction industry, pharmaceutical and drugs, and food industry. All the volunteers gave their consent for the use of their blood samples for the present survey. Information on exposure conditions were collected by questionnaire-based interviews and the following data about age, gender, place of residence, occupation, smoking habits, and fish consumption prior to sample collection are available. The age group of the subjects ranges from 18 to 70 years. Sixty three subjects of this group are female and thirty five are male. Twenty subjects are smokers and 78 subjects are non-smokers. Nineteen subjects are alcohol consumers and eighty one subjects never consumed alcohol. Seventeen subjects have consumed seafood within 48 h prior to sample collection. The bar diagram depicted below in Fig. 1 represent the characteristics of the population under study. A sample of 5mL blood was drawn with stainless steel needles into BD vacutainer tubes containing K3EDTA as the anti-coagulant. Samples were collected at Andhra university medical lab and Visakha diabetic center, Visakhapatnam. Samples were stored at -20□ C and digested within 48 hours using microwave digester (CEM MARS). The Microwave digestion system was equipped with 100mL Teflon PFA vessel and a turntable. In each vessel about 0.5mL of blood sample was taken and 1mL of supra-pure nitric acid was added and then made up to 10mL with deionized water. Routinely, 12 vessels were subjected to the following digestion procedure. Samples were allowed to predigest for a minimum of 15 minutes and then the vessel is closed by using a Capping station (CEM Corp). The sample digestion process was done in the following steps - in the first step temperature was gradually increased from room temperature to 150^{O}C with a ramp time of 20 min. The temperature was maintained for 5 min and then the vessels allowed to cool down to room temperature. After cooling, the digested samples were diluted to a final volume of 25mL with deionized water and analyzed. For each set of 10 samples two blanks were prepared in the similar way (without samples) to check for possible contamination. The samples were stored in the refrigerator at 4^{O}C until the analysis was carried out. During this study ultrapure water (18.2 mV) obtained from the water purification system (Millipore synergy) and suprapure nitric acidwere used. Each sample was spiked with internal standard containing Bi, Ge, In, ^{6}Li Sc, Tb, Y (SPEX CertiPrep). All the plastic ware and pipette tips used for blood, acid, water, standards and internal standard solution were rinsed with dilute nitric acid and then deionized water in that sequence.

Standard solutions and control samples:

The multi-element standard solution containing Ag, Al, As, Ba, Be, Ca, Cd, Ce, Co, Cr, Cu, Dy, Er, Eu, Fe, Gd, Ga, Ho, La, Lu, K, Li, Mg, Mn, Na, Nd, Ni, Pb, Pr, Rb, Sc, Sm, Se, Sr, Tb, Th, Ti, Tm, U, V, Y, Yb and Zn each at a concentration of 10 mg L^{-1} (SPEX CertiPrep) was used. 33 elements including Li, Be, Rb, Sr, Cs, Ba, V, Mn, Co, Ni, Cu, Zn, Se, La, Ce, Pr, Nd, Sm, Eu, Gd, Dy, Ho, Er, Tm, Yb, Lu, Th, Cd, Tl, Pb, U, As and Ag)were detected. The standard curve was prepared with 0, 0.25, 0.5, 1, 5, 10, 25, 50, 100 µg L^{-1}, concentrations. Accuracy and validity of the method has been assessed with the human whole blood (Seronorm level 3) certified for the elements As, Cd, Co, Cu, Mn, Ni, Se, Tl, Pb and Zn. For the uncertified elements the method was validated with spiked pooled blood.

3. Results and Discussion

Analytical results:

Before carrying out the sample analysis the method was validated. This included the relevant parameters such as determination of linear calibration ranges and limit of detection, the short term and long term stability of the instrument, the analysis of reference materials and the discussion of precision and accuracy in internal and external quality assurance [16]. For the elements As, Cd, Co, Cu, Mn, Ni, Se, Tl, Pb and Zn in human blood the data were compared with reference material seronorm (Table 1).

Table 1: Comparison of measured and certified concentrations

Element	Certified Range (µg/L)	Measured (µg/L)
As	24.9 – 36.5	32.55 ± 0.21
Cd	11.1 – 13.5	11.1 ± 0.04
Co	9.3 – 13.3	11.34 ± 0.092
Cu	2145 – 2469	2297.69 ± 5.35
Mn	44.7 – 54.7	45.81 ± 0.35
Ni	25.4 – 35.8	28.39 ± 1.13
Se	160 – 360	298.09 ± 6.67
Tl	27.7 – 34.1	29.70 ± 0.01
Zn	8165 – 9392	8426.63 ± 61.99
Pb	594 – 682	627.57 ± 2.91

For the elements where certified materials were not available, the recovery percentage was calculated through spike recovery by preparing a pooled blood sample by mixing of 10 randomly selected samples. Then pooled blood sample was spiked with 10 µg L^{-1} multi element standard solution and the spiked samples were assayed. Recoveries were estimated using the formula: recovery (%) = ((amount of original sample + Spike) – (amount found in original sample)/ (amount spiked)) X 100 [15]. For the 33 elements the procedure was repeated and relevant analytical figures of merit (recoveries for control materials, inter-day and intraday variation coefficients, spike recoveries, etc.) (Table 2). Measured concentrations in the control materials are in good agreement with certified values. Intra-day variation coefficients are in the range 1.48 to9.43%, whereas inter-day variation coefficients range from 3.84 to13.86%. Spike recoveries are in the range 90 to 110%. Limit of detection (LOD) has been calculated from LOD = 3RSDbc / SBR [16] where RSDb is the relative standard deviation of the background intensity of 10 measurements, c the concentration of the element in solution and SBR the signal-to-background ratio. Limit of detection values varied from 0.03 – 0.087 µg L-1 for the elements Ni, Se, Mn, Cu and Zn and for remaining elements it was in the range 0.0005 – 0.006 µg L-1.

The analytical results of the 98 subjects for the 33 elements are summarized in Table 3 which describes the mean concentrations of the element and standard deviation, concentration ranges and selected percentiles. Table 3 provides statistical data that is useful for the researchers working in the fields of clinical chemistry, epidemiology or toxicology. Elements determined in this study are discussed.

Trace elements play a vital role in the management of human health. Deficiency of these trace elements leads to adverse conditions that can be prevented or reversed by their supplementation [1]. Essential trace elements viz., V, Mn, Co, Ni, Cu, Zn and Se were studied in this work and the geometric means of the these elements were found to be 0.85, 18.3, 0.9, 39.3, 745, 4347 and 12.9 µg L^{-1}, respectively. Alkaline and alkaline earth metals Li, Be, Rb, Sr, Cs and Ba we were analysed. The geometric means of Li, Be, Ba, Cs, Sr and Rb 0.91, 0.01, 61.7, 0.85, 34.9 and 3375 µg L^{-1} respectively. In this study the toxic elements Ag, Cd, Pb, Tl, U and As were also studied. The geometric means of Ag, Pb and U are 0.32, 36.2 and 0.0223 µg L^{-1}, respectively. Tl was not detected in these samples. As levels in the present study ranged from 0.12 to 5.55 µg L^{-1} with the geometric mean of 0.97 µg L-1, Cd concentrations ranged from 0.05 to 5.69 µg L-1 with the geometric mean of 0.58 µg L^{-1}.

For some elements we were found significant differences between male and female subjects. The mean concentrations of Zn (4996 µg L^{-1}), Ni (51.4 µg L^{-1}) Rb (3645 µg L^{-1}), Se (19.6 µg L^{-1}), V (2.61 µg L^{-1}), Pb (49.4 µg L^{-1}) and Cs (1.26 µg L^{-1}) are showing significantly higher than the female subjects. The mean concentrations of Zn, Ni, Rb, Se, V, Pb and Cs in female subjects are 4299, 29.3, 3094, 16.1, 42.8 and 0.86 µg L^{-1} respectively. Whereas elements Sr (41.8 µg L^{-1}), Cu (807 µg L^{-1}) and Ba (62.3 µg L^{-1}) showing higher levels in female subjects than the male subjects, remaining elements not showing any significant relation between male and female subjects. The mean values of Sr, Cu and Ba in male subjects are 33.9, 723 and 47 µg L^{-1} respectively. Some elements showing higher concentrations in smokers compared with non-smokers. Elements Ni, Sr, Cd, Ba and Pb showing significantly higher concentrations in smokers compared to the non-smokers. We were observed statistically significant correlation between age and some blood trace elements Mn, Co, Cu, Zn, Rb (β = -0.41, p = 0.001; β = -0.36, p = 0.004; β = 0.62, p = 0.000; β = -0.53, p = 0.000; β = -0.54, p = 0.000). We were observed correlations between essential trace elements in whole blood Mn – Co (β = 0.75, p = 0.000), Mn – Cu (β = 0.58, p = 0.000), Mn – Zn (β = 0.38, p = 0.001), Mn – Se (β = 0.26, p = 0.034), Cu – Co (β = -0.57, p = 0.000), Co – Zn (β = 0.47, p = 0.000) and Cu – Se (β = 0.27, p = 0.007). Statistical correlation was also found between non-essential As – Pb (β = 0.48, p = 0.000), As – Cd (β = 0.44, p = 0.000) Cd – Pb (β = 0.43, p = 0.000).

Table 2: Selected isotopes, limits of detection (LOD), intra-day and inter-day variation coefficients (VCs) for the measured concentrations and spike recoveries

Isotope	Limit of detection (LOD)	VC (%)		% of recovery
		Inter day	Intra day	
^{7}Li	0.0015	10.83	4.92	104.49
^{9}Be	0.0005	11.68	7.47	95.3
^{51}V	0.006	12.97	4.20	90.04
^{55}Mn	0.030	10.66	4.34	94.17
^{59}Co	0.006	13.06	3.94	95.79
^{60}Ni	0.064	10.51	3.34	110.17
^{63}Cu	0.064	11.54	3.42	98.44
^{66}Zn	0.087	7.97	3.40	99.77
^{75}As	0.005	10.00	3.20	92.44
^{82}Se	0.064	13.86	6.09	97.41
^{85}Rb	0.005	6.95	5.23	97.67
^{88}Sr	0.021	7.14	9.43	97.26
^{107}Ag	0.005	5.42	8.11	98.6
^{111}Cd	0.005	4.67	4.23	96.9
^{133}Cs	0.006	4.06	5.27	93.13
^{137}Ba	0.047	9.28	5.35	107.4
^{139}La	0.002	3.92	5.06	93.46
^{140}Ce	0.003	3.84	4.60	94.17
^{141}Pr	0.001	6.21	4.01	93.55
^{146}Nd	0.001	3.95	3.28	95.25
^{147}Sm	0.002	4.14	2.96	95.55
^{153}Eu	0.001	5.48	2.50	94.01
^{157}Gd	0.001	5.47	2.43	92.65
^{163}Dy	0.001	7.57	1.56	93.22
^{165}Ho	0.001	7.77	1.78	93.63
^{166}Er	0.001	7.22	1.68	93.7
^{169}Tm	0.001	8.27	1.48	94.17
^{172}Yb	0.001	7.79	1.67	95.87
^{175}Lu	0.001	9.39	2.03	96.04
^{205}Tl	0.003	11.95	3.88	92.17
^{208}Pb	0.023	12.74	3.66	91.07
^{232}Th	0.031	11.99	5.72	93.12
^{238}U	0.003	13.71	6.00	96.86

Data regarding the 'f' block elements in human blood are very limited. Few of the researchers studied the concentrations of these elements in human blood Rodushkin et al. (1999) and Inagaki and Haraguchi (2000) studied the concentration of these elements in blood serum. In this work few of these elements were analyzed and an attempt was made to establish the base line concentrations [8], [17]. Here La, Ce, Pr, Nd, Sm, Eu, Gd, Dy, Ho, Er, Tm, Yb, Lu and Th are studied. Amongst these elements La, Ce, Pr, Nd, Sm, Eu, Gd, Dy and Th showed high concentrations. The ranges of the elements La, Ce, Pr, Nd, Sm, Eu, Gd, Dy and Th are 0.0035 – 0.87 (0.15), 0.0185 – 1.82 (0.38), 0.014 – 0.62 (0.12), 0.018 – 0.94 (0.20), 0.002 – 0.47 (0.073), 0.003 – 0.49 (0.045), 0.0046 – 0.49 (0.065), 0.0045 – 0.31 (0.058) and 0.0035 – 0.38 (0.079) µg L^{-1}, respectively. Here the values in the brackets indicate the geometric mean value of the respective elements. For the elements Ho, Er, Tm, Yb, and Lu the ranges are 0.00038 – 0.089 (0.014), 0.0035 – 0.099 (0.028), 0.002 – 0.098 (0.022), and 0.0025 – 0.08 (0.02) µg L^{-1}, respectively.

Zn was decreasing with age where as another essential element Cu increasing with age. Essential trace element concentrations are in a very small range for Co (0.11–4.14 µg L^{-1}), Cu (416 - 1217 µg L^{-1}) and Zn (1790 - 8920 µg L^{-1}). Other essential trace elements concentration ranges are 0.1 – 7.6 µg L^{-1} for V, 3.7 – 34.9 µg $L^{-1}$for Mn and 2.24 – 36.5 µg L^{-1} for Se. In these present study levels of manganese varied from 3.7 to 34.9 µg L^{-1} with the average value of 17.3 µg L^{-1}. These values are higher than the values published by Helitland (8.6 µg L^{-1}) [16] and Rodoskin (7.4 µg L^{-1}) [8] and lower than the values reported by Afridi et al. (2009) [18]. Cu concentrations are ranged between 416 and 1217 µg L^{-1} with a geometric mean of 745 µgL^{-1}, and these ranges are lower than the ranges published by Heitland (2006, 20), Rodushkin et al (1999) [16], [8]. Zn range varies between 1798 to 8928 µg/L with the mean value of 4498 µg/L. The concentrations of Zn and Cu presented here are lower than those reported by Viveksingh and Garg (1998) [7].

Table 3: Statistical data about elemental concentrations in blood for 98 human subjects

Isotope	% < LOD	MEAN	Range	GEOMEAN	Percentile			
					5th	50th	75th	95th
^{7}Li	0	1.17	0.13 – 3.19	0.91	0.20	1.08	1.73	2.36
^{51}V	6	1.43	0.1 - 7.6	0.85	0.22	0.81	1.72	5.01
^{55}Mn	0	17.3	3.74 - 34.9	15.3	5.58	15.7	23.0	30.8
^{59}Co	0	1.14	0.11 - 4.14	0.72	0.17	0.55	1.80	3.48
^{60}Ni	0	39.3	10.0 – 95.3	33.3	11.6	37.0	49.9	82.0
^{63}Cu	0	763	416 - 1271	745	525	753	878	1041
^{66}Zn	0	4498	1798 - 8920	4347	2872	4339	5011	6259
^{82}Se	5	15.4	2.24 - 36.5	12.9	3.75	15.0	21.3	29.2
^{85}Rb	0	3442	1949 - 5071	3375	2237	3478	3825	4511
^{88}Sr	0	39.0	9.39 - 80.1	34.9	14.7	38.5	48.3	73.5
^{133}Cs	74	0.94	0.17 - 1.91	0.85	0.33	0.95	1.20	1.60
^{137}Ba	0	87.3	10.5- 435	61.7	18.5	57.2	107	282
^{75}As	11	1.35	0.13 - 5.55	0.97	0.19	1.19	1.76	3.38
^{107}Ag	33	0.41	0.02 - 0.98	0.32	0.11	0.33	0.53	0.94
^{111}Cd	4	0.91	0.06 - 5.68	0.58	0.01	0.56	0.94	3.61
^{139}La	49	0.27	0.003 - 0.87	0.15	0.02	0.16	0.39	0.75
^{140}Ce	47	0.58	0.02 - 1.83	0.38	0.07	0.48	0.81	1.55
^{141}Pr	61	0.18	0.01 - 0.62	0.12	0.03	0.13	0.31	0.51
^{146}Nd	55	0.30	0.02 - 0.94	0.20	0.03	0.18	0.42	0.85
^{147}Sm	64	0.13	0.002 - 0.48	0.07	0.01	0.1	0.19	0.41
^{153}Eu	69	0.08	0.003 - 0.49	0.05	0.01	0.06	0.09	0.22
^{157}Gd	62	0.11	0.01 - 0.49	0.07	0.01	0.06	0.17	0.35
^{163}Dy	65	0.09	0.01 - 0.31	0.06	0.01	0.06	0.13	0.26
^{165}Ho	77	0.03	0.01 - 0.09	0.03	0.001	0.017	0.039	0.059
^{166}Er	66	0.038	0.003 - 0.099	0.028	0.005	0.029	0.048	0.094
^{169}Tm	67	0.016	0.002 - 0.087	0.009	0.001	0.012	0.022	0.043
^{172}Yb	69	0.034	0.002 - 0.098	0.022	0.004	0.031	0.045	0.074
^{175}Lu	78	0.029	0.002 - 0.080	0.020	0.005	0.022	0.035	0.079
^{208}Pb	0	40.6	6.26 - 100	36.2	16.4	36.8	50.9	79.0
^{232}Th	42	0.13	0.003 - 0.38	0.08	0.007	0.1	0.18	0.37
^{238}U	70	0.03	0.01 - 0.07	0.02	0.003	0.027	0.043	0.071

High values of the Ni were observed in the present study than those reported by Heitland and Koster (2006), Rodushkin et al. (1999), and Iyengar et al. (1998) [16], [8] and[19] and Ni values were ranging from 10 to 95 µg L^{-1} with the geometric mean of 33.33 µg L^{-1}. These values are coinciding with the observations of Danadevi et al. (2003) who have worked on blood and semen quality of Indian welders occupationally exposed to nickel and chromium [20]. Se concentration ranged from 2 to 36 µg L^{-1} with the mean of 12.88 µg L^{-1}. In this study area low Se concentrations were observed than the values published by Heitland and Koster (2006, 20), Rodushkin et al. (1999), in Germany and Safaralizadeh et al. (2005) in Tehran and Kłapcińska (2006) in upper Silesia [16], [8], [21] and [22]. Among all the essential elements low concentrations were observed for the elements V and Co. V ranged from 0.1 to 7.65 µgL^{-1} with a mean value of 1.42 µg L^{-1} and Co varied from 0.1 to 4.42 µgL^{-1} with the mean value of 1.13 µg/L. Except Se all the remaining elements were observed to be in higher concentrations in this present study.

Essential elements Ni, Mn, Co and V have been observed in high concentrations than those reported by other researchers. This might be due to contamination from the steel and casting industries located in the surroundings and of the geographic conditions. Other essential elements such as Zn and Cu were observed in normal levels where as concentrations of the Se were observed in lower levels. Subjects who consumed sea food had high As concentrations and also the subjects who were having smoking habit showed higher Cd concentrations. Elements such as Ag, Tl, La, Pr, Nd, Sm, Eu, Gd, Dy, Ho, Er, Tm, Yb, Lu and Th were detected in few subjects only. For some elements in the present investigation the limit of the concentrations and their influence on health are not known till now. It suggests that more biomonitoring surveys are needed to define the limits of these trace elements and their influence on health and disease. This is the first attempt at a biomonitoring survey in this area and these reference levels may be useful for further clinical and toxicological studies.

4. Conclusions

ICPMS with collision/ reaction cell technique enabled us to analyse elements in µg L^{-1} level in blood samples with high accuracy and repeatability. The major goal of this study is to present the ranges of trace elements in blood of normal subjects occupationally unexposed to trace elements in greater Visakhapatnam in southern India.

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