ASSESSMENT OF HEAVY METAL CONTENT OF HEN EGGS IN THE SURROUNDINGS OF URANIUM MINING AREA, INDIA

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Abstract
The present study was conducted to investigate the quality of hen eggs in respect of mineral content and to elaborate the possibility of contamination of heavy metals and their distribution. The eggs were collected from surroundings of Tummalapalle Uranium mining site, India. The heavy metals (Al, Cr, Mn, Fe, Ni, Cu, Zn, Pb, V, Co, As, Cd and U) were estimated with the help of Inductively Coupled Plasma Mass Spectrometer (ICP – MS) by following open acid digestion technique. Concentration of essential elements like Fe, Al and Zn were in the range of 34.4 – 98.3 mg/kg, 2.5 – 71.1 mg/kg and 3.9 – 63.0 mg/kg, respectively. Elements like U and Cd were in the range of 1.3 – 12.6 µg/kg and 2.3 – 15.4 µg/kg, respectively. The mean concentration of hen egg samples for all the estimated heavy metals are found to be below the permissible limit and are in comparison with the literature reported values from various parts of the world. Further, the estimated heavy metal data were subjected to statistical analysis like correlation matrices to examine the inter relationship between the investigated trace elements and source identification for the heavy metal contamination in hen eggs.

Keywords: eggs, heavy metals, uranium mining, ICP–MS, correlation

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1. INTRODUCTION

Egg has been an important nutritional deity food for human since ancient times. It is one of the highly nutritious foods among the human food stuff cycle and can be an effective delivery system for health – regulating nutrients, vitamins, proteins, fat etc. (Hashish et al., 2012). Eggs are readily digested and can provide a significant portion of the nutrients required daily for the proper growth and maintenance of body tissues. However, hen's egg might contain elevated levels of heavy metals that originate from food and water feed, which are mainly influenced with the surrounding environment. Knowledge and assessment of the mineral content of eggs is very important for many reasons that are related to health and nutritional value of eggs. Therefore, monitoring and estimation of heavy metals is of great importance for nutritional, toxicological and environmental aspect (Abdulkhaliq et al., 2012). The heavy metals like Cr, Co, Cu, Fe, Mn and Zn are essential, which are so called micronutrients and are toxic when taken in above threshold levels. Whereas, the heavy metals especially Pb, Cd, Hg and As are non essential elements and even toxic in trace levels (Siddiqui et al., 2011). The studies on heavy metal content in hen eggs have been widely reported in the literature (Abduljaleel et al., 2011, Abdulkhaliq et al., 2012, Azza et al., 2011, Hashish et al., 2012, Nisianakis et al., 2009, Siddiqui et al., 2011, Uluozlu et al., 2009, Vincent et al., 2008). However, the data on heavy metals in hen egg samples particularly in regions of mining activity is very limited and will be useful to the authorities and environmentalists for better assessment and understanding of the essential and non – essential elemental content present in the egg samples in and around these sites. In the present study, the content of Al, Cr, Mn, Fe, Ni, Cu, Zn, Pb, V, Co, As, Cd and U were determined using Inductively Coupled Plasma Mass Spectrometer (ICP – MS) in hen eggs, collected from in and around the villages of Tummalapalle Uranium mining site.

2. MATERIAL AND METHODS

2.1. Study Area

The study area, Tummalapalle uranium mine is located between latitudes 14°18'36" N & 14°20'20" N and longitudes 78°15'16" E & 78°18' 03.3" E as per India Toposheet Nos. 57 J/3 and 57 J/7.

2.2. Materials

All reagent used for the analysis were of analytical grade and Milli – Q water is used for all dilutions. Supra pure grade reagent (Merck, Germany) of nitric acid is used for the open digestion of samples. All the glass wares were cleaned by soaking in dilute nitric acid and was rinsed with distilled water prior to use.

2.3. Sample Collection

The study area is divided into two parts for the sample collection based on the radial distance from the mining site is as Core Zone – which is covering 10 km radial distance around the mining site and contains 3 sampling locations C_1, C_2 and C_3 and Buffer Zone – which is spreading over the radial distance from 10 – 25 km around the mining site and contains 2 sampling locations B_1 and B_2. The hen egg samples were collected in the study area and preserved in the refrigerator during year 2010 – 2011. The geographical location map indicating with two zones of sampling locations is shown in Fig. 1.

2.4. Sample Processing and Analysis

One gram of homogenated fluid egg sample was weighed using 0.01 mg sensitive weighing balance and transferred to 100 ml glass beaker. To the weighed sample, 15 ml of supra pure HNO_3 and 3 ml of $HClO_4$ were added and covered with the watch glass (Suseela et al., 2001, Tripathi et al., 1998, Tripathi et al., 2002). After one hour, the beaker was placed on hot plate to raise its temperature up to 140°C until the sample completely digested with reducing of its volume to 1 – 3 ml.

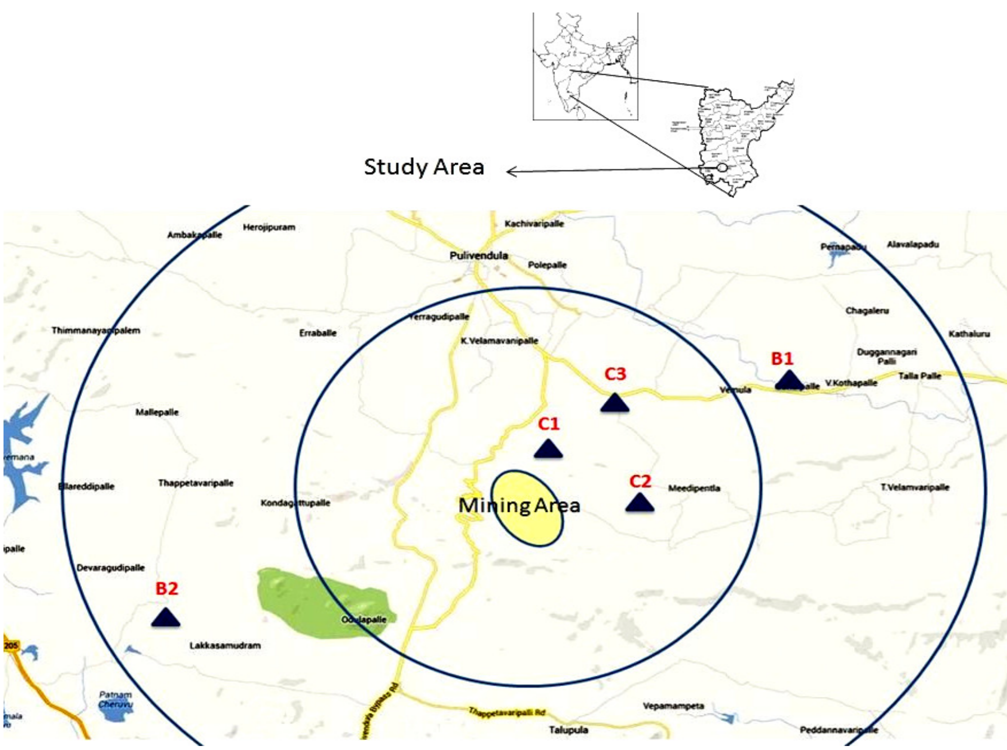

Fig. 1. The geographical representation of different sampling locations map

The digested sample was cooled, diluted with 2 N supra pure HNO_3 and made up to 50 ml in a standard flask. The processed samples have been analyzed for the estimation of trace metals using Inductively Coupled Plasma Mass Spectrometer (ICP – MS), make Agilent Technologies (Model No. 7700). ICP – MS can detect trace elements at the sub – ng/mg level (lower than 0.001 ppb) with simultaneous detection of more than 20 metals in short time.

2.5. Quality Assurance

The quality assurance of the procedure for the estimation of heavy metals in egg samples by ICP – MS has been assessed by calibrating the instrument using multi element calibration standard 2A (Lot No. 28 – 68 JB) obtained from Agilent Technologies. The validity of the method was further ascertained by spike recovery and replicate analysis.

3. RESULTS AND DISCUSSION

All the heavy metal concentrations are estimated in fresh wet weight basis. The estimated heavy metal content in hen eggs at all sampling locations in and around mining site along with the reported literature values from various parts of the world and calculated intake of heavy metal from consumption of one egg per day per person is summarized in Table 1. The obtained metal concentration in egg is divided into two categories namely macro (Al, Cr, Mn, Fe, Ni, Cu, Zn and Pb) and micro (V, Co, As, Cd and U) concentration, based on concentration levels. The heavy metal content in egg were determined based on the various factors taken into consideration particularly like the content of energy feed, the levels of organic components as fat, vitamins, proteins.

3.1. Macro Concentration Metals

The estimated macro concentration metals in hen egg were found in the range of 2.5 – 71.1 mg/kg for aluminium (Al), 1.2 – 12.4 mg/kg for chromium (Cr), 0.5 – 3.5 mg/kg for manganese (Mn), 34.4 – 98.3 mg/kg for iron (Fe), 0.1 – 0.8 mg/kg for nickel (Ni), 0.6 – 2.1 mg/kg for copper (Cu), 3.9 – 63.0 mg/kg for zinc (Zn), 0.1 – 0.9 mg/kg for lead (Pb). From the macro concentration metal data obtained, iron has the highest concentration followed by aluminium, zinc, chromium and manganese, where as nickel has the lowest concentration followed by lead and copper. The obtained mean macro concentration of metals is found to be comparable to the reported literature values from various parts of the world.

The average calculated intake of macro concentration metals based on consumption of one egg per day were determined 1.10 mg/day for Al, 0.19 mg/day for Cr, 0.10 mg/day for Mn, 2.99 mg/day for Fe, 0.02 mg/day for Ni, 0.07 mg/day for Cu, 1.50 mg/day for Zn and 0.02 mg/day for lead. The maximum permissible intake of an adult is 60 mg per day for aluminium, 3 – 32 µg per day for lead, 1 – 2.5 mg per day for copper, 100 µg per day for nickel (Nordberg et al., 2005, Uluozlu et al., 2009). Approximately 0.5 – 5% of each macro concentration metal contributing dietary intake through one egg consumption per day except Pb (contributing approximately 50%). The main sources of getting Al and Pb into the egg sample are environmental pollution like atmospheric Pb and Al particulates. The obtained macro concentrations of these metals in egg are below the permissible intake limits. The graphical representation of mean macro concentration of evaluated metals is shown in Fig. 2.

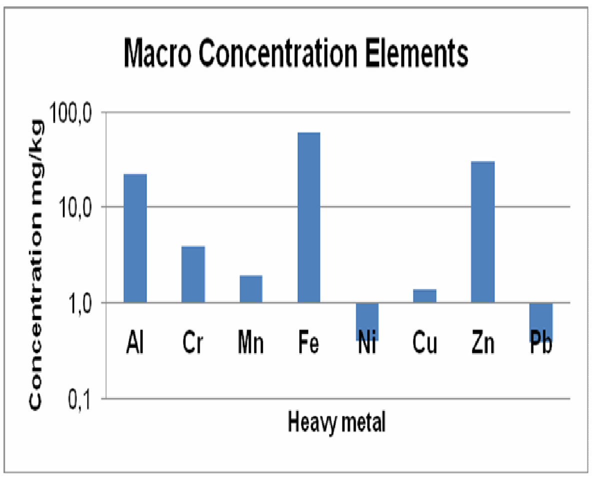

Figure 2: Graphical representation of mean macro concentration metals in egg sample

Table 1: Observed heavy metal concentrations in hen eggs with study region and the reported literature values from around the world and intake from consumption of one egg/day

Sampling Time	Sampling Zone	Sampling Location	Al	Cr	Mn	Fe	Ni	Cu	Zn	Pb	V	Co	As	Cd	U
			mg/kg (Macro concentration elements)								µg/kg (Micro concentration elements)				
2010	Core Zone	C_1	8.5	4.0	1.8	58.7	0.4	1.0	25.2	0.2	28.2	72.2	24.2	5.3	1.3
		C_2	23.6	3.1	3.1	78.7	0.3	1.4	37.1	0.3	98.5	23.0	26.2	7.2	3.1
		C_3	22.3	2.7	2.3	54.5	0.6	1.6	33.1	0.3	87.5	26.7	39.7	10.7	3.1
	Buffer Zone	B_1	20.7	2.7	3.5	56.0	0.4	1.6	33.2	0.4	76.2	22.1	27.0	8.9	3.9
		B_2	11.2	2.9	2.3	69.3	0.3	1.7	30.2	0.5	35.2	23.5	12.6	6.5	4.4
2011	Core Zone	C_1	36.0	4.4	2.0	98.3	0.5	2.1	63.0	0.9	97.4	29.5	94.4	2.3	10.5
		C_2	20.3	2.5	1.6	62.4	0.4	1.6	35.3	0.8	62.3	16.9	71.9	15.4	12.6
		C_3	71.1	2.7	0.9	54.7	0.2	1.2	38.1	0.3	30.9	14.7	29.3	8.2	3.1
	Buffer Zone	B_1	6.9	1.2	0.5	41.6	0.1	0.9	3.9	0.1	12.9	11.6	13.7	8.9	4.8
		B_2	2.5	12.4	1.7	34.4	0.8	0.6	6.7	0.1	18.5	18.3	3.5	6.1	5.2
Minimum			2.5	1.2	0.5	34.4	0.1	0.6	3.9	0.1	12.9	11.6	3.5	2.3	1.3
Mean - Present study in India			22.3	3.9	2.0	60.9	0.4	1.4	30.6	0.4	54.8	25.8	34.3	8.0	5.2
Maximum			71.1	12.4	3.5	98.3	0.8	2.1	63.0	0.9	98.5	72.2	94.4	15.4	12.6
Intake of Heavy metal from consumption of one egg/day (mg or µg / day)			1.10 mg	0.19 mg	0.10 mg	2.99 mg	0.02 mg	0.07 mg	1.50 mg	0.02 mg	2.69 µg	1.27 µg	1.68 µg	0.39 µg	0.26 µg
Literature Values from various parts of the world															
Palestine (mg/kg) (Hashish et al., 2012)			-	-	-	46.8	-	4.6	-	1.39	-	-	-	0.13	-
Egypt (mg/kg) (Abdulkhaliq et al., 2012)			-	-	4.4	117.3	-	15.5	129.5	0.31	-	-	0.033	0.015	-
Egypt (mg/kg) (Azza et al., 2011)			-	-	-	-	-	1.0	74.4	2.3	-	-	-	-	-
London, UK (mg/kg) (Siddiqui et al., 2011)			-	0.45	0.51	18.4	-	6.6	33.2	0.15	-	-	-	-	-
Malaysia (mg/kg) (Abduljaleel et al., 2011)			17.1	3.24	-	-	1.11	-	34.22	0.42	-	-	0.3	0.06	-
Turkey (mg/kg) (Uluozlu et al., 2009)			0.59	0.07	0.97	48.8	0.18	0.9	25.5	0.11	-	0.02	0.18	3.8	-
Bangladesh (mg/kg) (Nisianakis et al., 2009)			-	-	-	124.5	4.26	0.64	104.14	12.1	-	-	-	1.04	-

3.2. Micro Concentration Metals

On the basis of the evaluated concentration of metals in collected hen egg samples, vanadium (V), cobalt (Co), arsenic (As), cadmium (Cd) and uranium (U) are categorized as micro concentration metals. Among these five metals, V and Co are probably essential metals whereas, As, Cd and U are toxic metals. The calculated micro concentration metals in hen egg samples were found in the range of 12.9 – 98.5 µg/kg for V, 11.6 – 72.2 µg/kg for Co, 3.5 – 94.4 µg/kg for As, 2.3 – 15.4 µg/kg for Cd and 1.3 – 12.6 µg/kg for U. From the obtained micro concentration metal data, V is predominantly noticed with high concentration followed by As, Co, Cd and U. The evaluated micro concentration metals are showing good comparison with reported literature from various parts of the world (Table 1) which are within the limits.

The intake of micro concentration metals is calculated based on consumption of one egg per day per person. The intake of micro concentration metals is found to 2.69 µg/day for V, 1.27 µg/day for Co, 1.68 µg/day for As, 0.39 µg/day for Cd and 0.26 µg/day for U. The permissible limit and dietary intake range for various metals V: 6 – 30 µg/day; Co: 5 – 50 µg/day; Cd: 10 – 60 µg/day; As: 0.07 – 0.37 mg/day and for U: 0.9 - 1.5 µg/day (Nordberg et al., 2005, Uluozlu et al., 2009). 8.9% of V, 2.5% of Co, 0.5% of As, 0.7% of Cd and 17.3% of U levels are contributing dietary intake through the consumption of one egg per day per person.

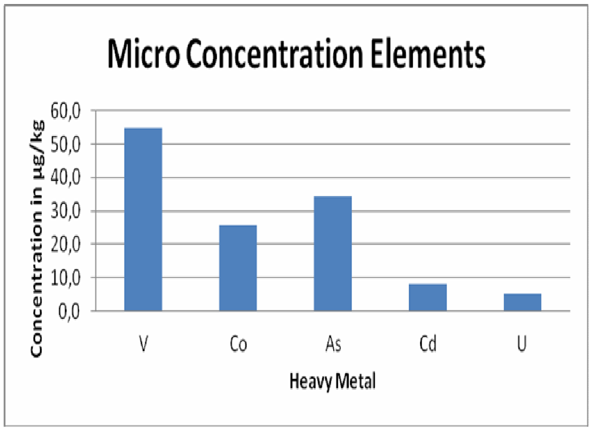

Fig 3: Graphical representation of mean micro concentration metals in egg samples

The reported literature values from various parts of the world are in the range of 0.0 – 0.02 mg/kg for Co, 0.03 – 0.18 mg/kg for As and 0.02 – 1.04 mg/kg for Cd (Abdulkhaliq et al., 2012, Abduljaleel et al., 2011, Azza et al., 2011, Hashish et al., 2012, Nisianakis et al., 2009, Siddiqui et al., 2011, Uluozlu et al., 2009). The main sources of getting the high levels of these micro concentration metals are feeding behavior of hens, anthropogenic activities, environmental contamination etc. The graphical representation of mean micro concentration metals is shown in Fig. 3.

3.3. Correlation – Analysis

A linear regression correlation test was performed to investigate the correlation between all the heavy metals under consideration and to determine the likely sources of contamination of the egg samples. The values of the correlation coefficients between metal concentrations are given in Table 2. Iron is showing strong correlation with Cu, Zn, Pb, V and As with corresponding regression coefficient (r) values of 0.82, 0.89, 0.76, 0.71 and 0.70, respectively. Copper is showing good correlation with Zn, Pb, V and As with corresponding r values of 0.86, 0.85, 0.76, 0.73 and 0.45 respectively. This correlation indicates that, the sources of these metals (Fe, Cu, Zn, Pb, V and As) have common origin and is expected from the earth's crust as Fe, Zn and Cu are more abundant crustal elements including anthropogenic activities for the metals like Pb, V and As. The transfer of metals in eggs is likely to be takes place in the sequential order of Soil → Local Feed → Hen → Eggs.

Table 2: Correlation between the metal concentrations of egg sample

	Al	Cr	Mn	Fe	Ni	Cu	Zn	Pb	V	Co	As	Cd	U
Al	1.00												
Cr	-0.30	1.00											
Mn	-0.16	0.00	1.00										
Fe	0.31	-0.31	0.38	1.00									
Ni	-0.30	**0.79**	0.29	-0.11	1.00								
Cu	0.31	-0.48	0.46	**0.82**	-0.06	1.00							
Zn	**0.60**	-0.30	0.37	**0.89**	0.00	**0.86**	1.00						
Pb	0.27	-0.24	0.19	**0.76**	0.04	**0.85**	**0.80**	1.00					
V	0.23	-0.25	**0.68**	**0.71**	0.19	**0.76**	**0.76**	**0.56**	1.00				
Co	-0.23	0.03	0.15	0.18	0.15	-0.05	0.11	-0.08	-0.03	1.00			
As	0.36	-0.24	0.03	**0.70**	0.10	**0.73**	**0.80**	**0.88**	**0.63**	0.02	1.00		
Cd	-0.01	-0.35	-0.09	-0.37	-0.14	-0.01	-0.21	0.07	0.01	-0.38	0.06	1.00	
U	0.04	0.02	-0.15	0.37	0.15	0.45	0.37	**0.81**	0.26	-0.34	**0.76**	0.27	1.00

4. CONCLUSIONS

The present Investigation is mainly focused on the evaluation of various heavy metals including essential and non-essential metals in hen egg samples collected from the various locations in and around Uranium mining site. Heavy metal concentrations obtained in the present study in hen egg samples were found to be acceptable and are within the permissible limits to human consumption in respect of nutritional and toxic levels as compared with the literature data reported from various parts of the world.

5. ACKNOWLEDGEMENT

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