ESTIMATION OF HEAVY METALS TOXICITY IN VEGETABLES & MEDICINAL PLANTS COLLECTED FROM ROAD SIDE MARKET SITES OF DIFFERENT AREAS OF RANCHI CITY

UGC APPROVAL LETTER NO. AND DATE

F. No. 41-1373/2012 (SR) dated 30 Jul 2012

FINAL REPORT

Submitted by

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Introduction

Heavy metals such as Arsenic (As), Cadmium (Cd), Chromium (Cr), Copper (Cu), Nickel (Ni) and Lead (Pb) are distributed in the environments naturally or anthropogenically, released from industrial as well as agricultural use. Most of the heavy metals, with long half lives can be translocated and accumulated into agricultural products through absorption from contaminated soil (Huang et al., 2006; Römkens et al., 2009). Particularly, Cd and Pb are easily absorbed from the soil to roots of agricultural plants (Qian et al., 2010). High contents of heavy metals in soil have become one of the most important sources to contaminate the agricultural products, and thus can be easily exposed to human beings through various pathways, such as ingestion of food, dermal contact, and inadvertent ingestion of soil (Chatterjee et al., 2010). The food consumption has been suggested to be a major exposure of heavy metals to humans. Hence, the excessive heavy metals exposure through intake of agricultural products like vegetables may be of great concern in causing health risks to consumers (Singh et al., 2010).The health risks of consumers exposed to heavy metals through intake of vegetables were assessed by probabilistic risk assessment to reduce the uncertainty and variability of input parameters (Mondal et al., 2008). Therefore, the present study was to investigate the heavy metals contents in various vegetables collected from different market sites and to determine the health risks for consumers based on Metal Pollution Index, Daily Intakes of Heavy metals and Health Risk Index. Heavy metals contents in vegetables were also measured by safety limit of PFA limit, ATSDR guidelines and PTDI value by Joint FAO/WHO Expert Committee on Food Additives.

In the present study concentrations of Arsenic (As), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Cupper (Cu), Nickel (Ni) and Lead (Pb) were estimated by using an Inductively Coupled Plasma - Optical Emission Spectrometer (ICP-OES). Concentrations of heavy metals were calculated on a dry weight basis. All analyses were replicated six times.

Study Areas

The study was conducted around Ranchi (23°21’ N latitude 85°20’ E longitude and 729 m (2,392 ft) above the sea level) city in Jharkhand eastern plains of India during July 2010 to February 2012. Various small scale industries situated in this city. A large area around industries have no access to clean water resources, so farmers use treated and untreated wastewater for irrigation. The hypothesis behind the present study is that the irrigation with waste water, transportation and marketing site of vegetables in contaminated environment may elevate the levels of heavy metals in vegetables through surface deposition. Eight Road side Markets viz. Site-1 (Lalpur Market), Market Site-2 (BIT More Market), Site-3 (Daily Market), Site-4 (Kanke Road Market), Site-5 (Booti More Market), Site-6 (RIMS Market), Site-7 (Morabadi Stadium Market), Site-8 (Bahu Bazar Market) and two organised Markets i.e. Site-9 (Reliance Fresh/ Reliance Mart) & Site-10 (Big Bazar) were demarcated for vegetable purchasing.

Sample and Sampling

The edible portions of Vegetables were collected from different markets during Sep 2012 to May 2014. Samples were brought back to the laboratory and washed with clean tap water to remove the soil particles and dusts of the vegetables. After removing the extra water from the surface of vegetables with blotting paper, samples were cut into pieces, packed into separate bags, and kept in an oven until a constant weight was achieved. The dried samples were grinded and passed through a sieve of 2 mm size and then kept at room temperature for further analysis.

Name of the different vegetables are as follows:

1. **Leafy Vegetables:** Spinach, Corriander leaves, Cabbage
2. **Roots & Tubers:** Beet root, Carrot, Radish, Ginger, Potato
3. **Leguminous vegetables:** Bean, Pea
4. **Other Vegetables:** Cucumber, Lady’s finger, Tomato

Table 3.1: Botanical Name and Family of the different vegetables

S.No	Common Name	Botanical Name	Family
1	Spinach	*Spinacia oleracea*	Chenopodiaceae
2	Corriender leaves	*Coriandrum sativum*	Apiaceae
3	Cabbage	*Brasssica oleracea*	Brassicaceae
4	Beet root	*Beta vulgaris*	Amaranthaceae
5	Carrot	*Ductus carrotus*	Apiaceae
6	Radish	*Raphanus sativus*	Brassicaceae
7	Ginger	*Gingiber officinalis*	Zingiberaceae
8	Potato	*Solanum tuberosum*	Solanaceae
9	Bean	*Phaseolus vulgaris*	Fabaceae
10	Pea	*Pysum sativum*	Fabaceae
11	Cucumber	*Cucumis sativus*	Cucurbitaceae
12	Lady’s finger	*Abelmoschus esculentus*	Malvaceae
13	Tomato	*Lycopersicum esculentum*	Solanaceae

Analysis of Heavy Metals

In the present study concentrations of Arsenic (As), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Cupper (Cu), Nickel (Ni) and Lead (Pb) were estimated by using an Inductively Coupled Plasma - Optical Emission Spectrometer (ICP-OES). Concentrations of heavy metals were calculated on a dry weight basis. All analyses were replicated six times. The concentration value for Arsenic (As) was found zero in each vegetables since none of the vegetables were contaminated with As.

Among thirteen vegetables, Beet, Cucumber, Pea, Beans, Lady's finger, Corriender leaves and Tomato showed high levels of Lead (Pb) in vegetables collected from all sites. Within the selected vegetables, the highest concentrations of Lead (Pb) were noticed in Peas collected from Site-5 followed by Site-7 and Site-8. Cadmium (Cd) was found in fair amount in Cucumber collected from Site-6 and Site-8 and Spinach from Site-6. Nickel (Ni) was found to be in higher concentrations in Pea and Beans collected from all sites. Lady's finger also contains fair amount of Nickel. **The concentration levels of these three heavy metals i.e. Lead (Pb), Cadmium (Cd) and Nickel (Ni) were found to be beyond the PFA permissible limit in vegetables.** The high contamination found in vegetables might be closely related to the pollutants in irrigation water, farm soil or due to pollution from the highways traffic (Igwegbe et al., 1992; Qiu et al., 2000).

Results of two way ANOVA test showed that variations in the heavy metal concentrations were significant for some heavy metals due to site, vegetable and site-plant interaction. The results of the ANOVA (Two-Factor without Replication) between concentration of heavy metals and PFA permissible limits suggest that in case of Lead, the P value (1.53E-07) is less than the significance level (0.05) and F (68.37) is more than F crit (4.41). The amount of Lead was found to be significantly higher than the PFA limit. The variations in heavy metal concentrations in vegetables of the same site may be ascribed to the differences in their morphology and physiology for heavy metal uptake, exclusion, accumulation and retention. The use of contaminated irrigation water may also increase the uptake and accumulation of the heavy metals in the vegetables.

Concentration of Heavy metals of individual market sites:

Site-1 (Lalpur Market) - The range of Cadmium was found to be 0.100 ± 0.045 ppm to 0.933 ± 0.295 ppm and the range of Cobalt concentration was from 0.200 ± 0.103 ppm to

1.133 ± 0.818 ppm. The ranges of Chromium and Copper were found to be 0.100 ± 0.045 ppm to 4.500 ± 0.888 ppm and 2.267 ± 0.048 ppm to 20.467 ± 1.982 ppm respectively. Highest Concentration of Nickel (4.833 ± 0.600 ppm) and Lead (13.733 ± 2.733 ppm) were found in Pea. The lowest concentrations of Nickel in Ginger (0.150 ± 0.022 ppm) and Lead in Potato (0.300 ± 0.086 ppm) were observed in Site-1.

Site-2 (BIT More Market) - The range of Cadmium was found to be 1.067 ± 0.276 ppm to 0.167 ± 0.021 ppm. The range of Cobalt concentration was from 0.267 ± 0.080 ppm to 1.317 ± 0.793 ppm. The ranges of Chromium and Copper were found to be 0.217 ± 0.083 ppm to 4.233 ± 0.953 ppm and 2.300 ± 0.169 ppm to 21.033 ± 0.953 ppm respectively. The range of Nickel concentration was found 1.33 ± 0.021ppm to 4.667 ± 0.422 ppm and range of Lead was observed 0.700 ± 0.145 ppm to12.667 ± 1.520 ppm.

Site-3 (Daily Market) - The range of Cadmium was found to be 0.200 ± 0.063 ppm to 1.317 ± 0.793 ppm and range of concentration of Cobalt was found 0.433 ± 0.076 ppm to 1.633 ± 0.716 ppm.The ranges of Chromium and Copper were found to be 0.467 ± 0.062 ppm to 5.100 ± 0.943 ppm and 2.183 ± 0.319 ppm to 20.667 ± 1.838 ppm respectively. The range of Nickel was found 0.150 ± 0.022 ppm to 4.333 ± 0.760 ppm and the range of Lead was observed 1.133 ± 0.191 ppm to 9.667 ± 0.803 ppm.

Site-4 (Kanke Road Market) - The highest concentration of Cadmium (1.267 ± 0.299 ppm) and Cobalt (1.567 ± 0.167 ppm) was found in Spinach, Chromium (4.000 ± 0.683 ppm) in Pea, Copper (17.000 ± 2.864 ppm) in Tomato, Nickel (4.333 ± 0.667 ppm) and Lead (11.000 ± 0.447 ppm) in Pea. The lowest concentration of Cadmium in Potato (0.267 ± 0.080 ppm), Cobalt in Carrot (0.450 ± 0.150 ppm), Chromium in Cabbage (0.600± 0.146 ppm), Copper in Potato (3.533 ± 0.657 ppm). Nickel in Beet (0.200 ± 0.045 ppm) and Lead in Carrot (1.033 ± 0.216 ppm) ware observed.

Site-5 (Booty More Market) - The highest concentration of Cadmium (0.667 ± 0.095 ppm) in Corriender leaves, Cobalt (0.717 ± 0.054 ppm) in Pea, Chromium (6.167 ± 2.429 ppm) and Copper (17.133 ± 2.999 ppm) in Tomato, Nickel (5.833 ± 0.792 ppm) and Lead (12.067 ± 2.467 ppm) in Pea. The lowest concentration of Cadmium in Pea (0.117 ± 0.031 ppm), Cobalt in Carrot (0.300 ± 0.081 ppm), Chromium in Cabbage (1.767 ± 1.647 ppm), Copper in Potato (3.933 ± 1.539 ppm), Nickel in Ginger (0.200 ± 0.045 ppm) and Lead in Spinach (0.500 ± 0.304 ppm) were observed.

Site-6 (RIMS Market) - The range of Cadmium was found to be 0.233 ± 0.033 ppm to 1.983 ± 0.681 ppm and the range of Cobalt concentration was from 0.433 ± 0.084 ppm to 1.033 ± 0.279 ppm. The range of Chromium was found to be 2.067 ± 1.244 ppm to 7.833 ±

2.483 ppm and Copper 6.400 ± 1.215 ppm to 15.633 ± 3.125 ppm. The range of Nickel was found 0.283 ± 0.054 ppm to 1.833 ± 0.307 ppm and the range of Lead was observed 0.683 ± 0.101ppm to 9.067 ± 2.356 ppm.

Site-7 (Morabadi Stadium Market-The highest concentration of Cadmium (0.900 ± 0.446 ppm) in Cabbage, Cobalt (1.467 ± 0.797 ppm) in Beet, Chromium (3.667 ± 0.992 ppm) and Copper (17.133 ± 3.605 ppm) in Tomato, Nickel (4.833 ± 0.792 ppm) in Beans and Lead (10.733 ± 2.992 ppm) in Pea. The lowest concentration of Cadmium and was found in Potato (0.400 ± 0.137 ppm), Cobalt in Carrot (0.367 ± 0.196 ppm), Chromium in Radish (0.267 ± 0.152 ppm), Copper in Potato (3.933 ± 1.539 ppm), Nickel in Beet (0.267 ± 0.182 ppm) and Lead in Spinach (0.500 ± 0.304 ppm).

***Site-8 (Bahu Bazar Market)* –** The range of Cadmium was found to be 0.400 ± 0.163 ppm to 2.150 ± 0.638 ppm and the range of Cobalt concentration was from 0.350 ± 0.115 ppm to 0.967 ± 0.315 ppm. The range of Chromium was found to be 0.733 ± 0.402 ppm to 6.167 ± 2.429 ppm and Copper 5.600 ± 1.930 ppm to 22.300 ± 0.621 ppm. The range of Nickel was found 0.333 ± 0.183 ppm to 5.000 ± 1.291 ppm and the range of Lead was observed 0.800 ± 0.288 ppm to 10.733 ± 1.971 ppm.

***Site-9 (Reliance Fresh/ Reliance Mart)* –** The range of Cadmium was found to be 0.283 ± 0.166 ppm to 0.833 ± 0.198 ppm and the range of Cobalt concentration was from 0.467 ± 0.145 ppm to 0.883 ± 0.207 ppm. The range of Chromium was found to be 0.383 ± 0.306 ppm to 4.833 ± 1.364 ppm and Copper 4.700 ± 1.268 ppm to 15.633 ± 2.539 ppm. The range of Nickel was found 0.367 ± 0.173 ppm to 4.507 ± 0.885 ppm and the range of Lead was observed 0.500 ± 0.151ppm to 10.400 ± 2.626 ppm.

Site-10 (Big Bazar)-The highest concentration of Cadmium (0.717 ± 0.232 ppm) and Cobalt (1.017 ± 0.229 ppm) in Tomato, Chromium (4.500 ± 1.506 ppm) and Lead (7.400 ± 2.091 ppm) in Pea, Copper (17.067 ± 1.535 ppm) and Nickel (3.833 ± 1.046 ppm) in Beans.The lowest concentration of Cadmium was found in Carrot (0.350 ± 0.178 ppm), Cobalt in Ginger (0.467 ± 0.184 ppm) and Radish (0.467 ± 0.161 ppm). Lowest concentration of Chromium was found in Radish (0.550 ± 0.330 ppm), Copper in Ginger (6.367 ± 2.766 ppm), Nickel in Ginger (0.483 ± 0.189 ppm) and Lead in Spinach (0.467 ± 0.313 ppm).

In context to the hypothesis taken regarding heavy metals concentration in various vegetables collected from road side market of Ranchi city, it was observed from above results that the null hypothesis (i.e. there is no significant difference between the heavy metal concentrations in vegetables collected from road side market of Ranchi city), was found to be wrong and the alternate hypothesis (i.e. there is a significant difference between the heavy metal concentrations in vegetables collected from road side market of Ranchi city) was found to be correct and significant upto $P<0.001$.

Regarding the hypothesis related to heavy metals concentration in various vegetables collected from road side markets and organized markets of Ranchi city, again the alternate hypothesis i.e. there is a significant difference between the heavy metal concentrations in vegetables collected from road side market and organized markets of Ranchi city was found to be correct significant upto $P<0.001$.

Metal Pollution Index

Metal Pollution Index (MPI) is suggested to be a reliable and precise method for metal pollution monitoring. Among different vegetables pea showed highest value of MPI followed by Cucumber. Seven vegetables out of thirteen showed higher MPI i.e. more than two. These were pea, cucumber, tomato, beans, spinach, lady finger and cabbage. Higher MPI suggests that these vegetables may cause more human health risk due to higher accumulation of heavy metals in the edible portion. Site-6 and Site-8 can be classified as high risk sites as the MPI of all vegetables were higher than one. Among the vegetables Pea, Beans. Beet and Cucumber were found to be highly contaminated with the heavy metals. The Metal Pollution Index of the vegetables of different sites (Site-1 to Site-10) were subjected to two way analysis of variance (ANOVA) test for assessing the significance of differences in heavy metal concentrations due to different sites, irrigation practices, environmental pollutants, etc. The results of the ANOVA (Two-Factor without Replication) suggests that in case of vegetables, the P value (1.12E-38) was found to be less than the significance level (0.05), and F (50.513) was more than F crit (1.843) i.e. there is significant difference between MPI among the vegetables. Similarly, in case of Sites, the P value (4.65E-10) was less than the significance level (0.05), so we can reject the null hypothesis that the means are equivalent. F (9.017) was observed more than F crit (1.968) so we can reject the null hypothesis i.e. there is a significant difference between MPI among the Sites.

Dietary Intake of Metal

Dietary intake of heavy metals by the population through the consumption of the vegetables was calculated by dividing the daily consumption rates of the heavy metals with the values of Provisional Tolerable Daily Intake.

PTDI value for Cadmium was 60 µg/day. In present study of the daily intake of Cadmium was observed 98.52 µg/day in Site-6 and 142.58 µg/day in Site-8 through Cucumber. In Site-4, Site-6 and Site-8 the daily intake for Cadmium was 89.03 µg/day, 131.47 µg/day and 129.00 µg/day respectively through Spinach. The consumption of these vegetables may pose health risk for the consumer as the DIM values for Cadmium were beyond the PTDI limit. In case of Cobalt the DIM value in all vegetables were within the safe limit of ATSDR, 1994. So, there is no health risk for Cobalt consumption. According to Food and Nutrition Board of the NAS/NRC the Safe and Adequate Intake of Chromium for an adult was 50–200 µg/day. In this study daily intake of Chromium was above the Safe and Adequate Intake limit according to Food and Nutrition Board of the NAS/NRC in all vegetables except Ginger and Corriender Leaves. Provisional Tolerable Daily Intake of Copper was 3 mg/day. In this study DIM for Copper was within the permissible limit. According to ATSDR, 1999 Safety limit for human consumption of Nickel- 3 to 7 mg/day. In present study, all values of DIM for Nickel were within the ATSDR permissible limit. Daily intake for Lead was found to be higher than PTDI value in all sites through consumption of Cucumber, Pea, Lady’s finger and Tomato. PTDI value for Lead was 214 µg/day. Thus, the consumption of these vegetables may pose a human health risk.

Health Risk Index

The health risk index is the ratio of estimated exposure of test vegetables and oral reference dose. Estimated exposure is obtained by dividing daily intake of heavy metals by their safe limits. An index more than one is considered as not safe for human health.

High HRI for Cadmium i.e., more than two was observed in Cucumber of Site-8 i.e. 2.39 and in Spinach of Site-6 and Site-8 i.e. 2.19 and 2.15 respectively and more than one in Spinach i.e. 1.48 of Site-4. HRI was observed more than one for Cobalt in Cabbage of Site-6, Site-7, Site-8, Site-9 and Site-10; Tomato of Site-6, Site-8 and Site-10; Potato of Site-4, Site-9 and Site-10; Spinach of Site-4, Site-7 and Site-10 and Radish of Site-7. HRI for Lead was found more than one in Cucumber in all sites i.e. Site 1 to Site-10; Pea of all sites except Site-6 and Site-10; Beans of all sites, except Site-1 and Site-3; Lady’s finger of all sites, except

Site-2 and Site-3 and in Tomato it was found to be more than one in all sites except Site-1, Site-2, Site-4 and Site-10. High HRI for Lead i.e., more than two was found in Cucumber of Site-8 and Site-9 i.e. 3.55 and 2.66 respectively. Thus from these result it is concluded that consumption of these vegetables are not safe for human consumption which may pose toxicity to human health.

In present study the concentration of Cadmium ranges from 0.100 ppm to 2.150 ppm in various vegetables. The maximum concentration of Cadmium 2.150 ppm was found in spinach collected from Site-8, while minimum concentration 0.100 ppm was found in Cucumber, Pea and Ladies finger collected from Site-1. Cadmium concentration was found to be significantly high in Cucumber and Spinach in both Site-6 and Site-8 at different significant level in comparison to all other sites. HRI was found more than one i.e. 1.64 and 2.38 in Cucumber from Site-6 and Site-8 respectively. In Spinach the HRI was 1.484, 2.19 and 2.15 respectively for Site-4, Site-6 and Site-8. HRI more than one is considered as not safe for human health (USEPA, 2002). Acute doses of Cadmium can cause severe gastrointestinal irritation, vomiting, diarrhea, and excessive salivation, and doses of 25 mg of Cd/kg body weight can cause death. Low-level chronic exposure to Cadmium can cause adverse health effects including gastrointestinal, hematological, musculoskeletal, renal, neurological, and reproductive effects. The main target organ for Cadmium following chronic oral exposure is the kidney (ATSDR 1999a).

The Cobalt content varies from 0.200 ppm to 1.633 ppm. The lowest concentration 0.200 ppm of Cobalt was observed in Carrot collected from Site-1. On the other hand, Beet from Site-3 showed highest concentration of Cobalt i.e. 1.633 ppm. HRI was found more than 1 in Cabbage (Site-6, Site-7, Site-8, Site-9 and Site-10), Tomato (Site-6, Site-8 and Site-10), Potato (Site-4, Site-9 and Site-10), Spinach (Site-4, Site-7 and Site-10) and Radish (Site-7). Higher concentrations of Cobalt were observed in Beet of Site-3 and Spinach of Site-4 which were significantly higher ($P<0.05$) than all other sites (Ghosh et al, 2011). Overdose of Cobalt may lead to angina, asthma, cardiomyopathy, polycythemia and dermatitis. The safety limit for human consumption of Cobalt is 0.05 to 1 mg/day in humans (ATSDR 1994).

Chromium concentration varies from 0.100 ppm to 7.833 ppm. The highest concentration of Chromium was found in Tomato collected from Site-6 i.e. 7.833 ppm, while lowest concentration 0.100 ppm was recorded in Radish and Cabbage collected from Site-1. Its long-term effect is Lung cancer. Apart from the carcinogenic potential, prolonged exposure can result in bronchitis, rhinitis or sinusitis or the formation of nasal mucosal polyps. Besides the lungs and intestinal tract, the liver and kidney are often target organs for chromate toxicity (Rom 2007).

Present investigation reveals that Copper varies from 2.183 ppm to 22.300 ppm. The highest concentration of Copper was found in Tomato collected from Site-8 i.e. 22.300 ppm, while lowest concentration 2.183 ppm was recorded in Potato collected from Site-3. Excess intake of Copper leads to anorexia, arthritis, asthma, autism, male infertility, migraine headaches, hypothyroidism, insomnia and neuralgia.

The presence of Nickel ranges from 0.133 ppm to 5.833 ppm in various vegetables. Peas from Site-5 and Beans from Site-6 showed high content of Nickel 5.833 ppm, while Potato from Site-2 contains low value of Ni 0.133 ppm. Nickel concentration was found high in Cucumber of Site-3, Cabbage of Site-4 and it was found to be significantly higher in Pea and Beans of Site-5 and Site-6 respectively in comparison to all other sites at different significant level ($P<0.001$, $P<0.01$ and $P<0.05$).Excess intake of Nickel leads to hypoglycemia, asthma, nausea, headache, and epidemiological symptoms like cancer of nasal cavity and lungs.

During the present study, the concentration of Lead content varies from 0.300 ppm to 13.733 ppm. High concentration of Lead was found in Peas collected from Site-1 i.e. 13.733 ppm. Lead concentration was found to be significantly higher in Pea of Site-1, Cucumber and Beans of Site-2, Corriender leaves and Tomato of Site-3, Spinach and Lady's finger of Site-4, and Cabbage of Site-6 in comparison to all other sites at different significant levels ($P<0.001$, $P<0.01$ or $P<0.05$). Potato collected from Site-1 showed low concentration of Lead 0.300 ppm. Health risk index for Lead was found more than 1 in Cucumber (All sites), Pea (All sites except Site-6 and Site-10), Beans (All sites; except Site-1 and Site-3), Lady's finger (All sites; except Site-2 and Site-3), and Tomato (All sites except Site-1, Site-2, Site-4 and Site-10). High HRI was found in Cucumber for Lead in Site-8 and Site-9 i.e. 3.545, 2.658 respectively. Todd (1996) emphasized that most of the accumulated Lead is sequestered in the bones and teeth. This causes brittle bones and weakness in the wrists and fingers. Lead that is stored in bones can reenter the blood stream during periods of increased bone mineral recycling (i.e., pregnancy, lactation, menopause, advancing age etc.). Mobilized lead can be redeposited in the soft tissues of the body and can cause musculoskeletal, renal, ocular, immunological, neurological, reproductive and developmental effects (ATSDR 1999b).

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Annexure – IV

UNIVERSITY GRANTS COMMISSION
BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002

Utilization certificate

Certified that the grant of Rs. 45,000/- (Rupees Forty-five thousand only) received from the University Grants Commission under the scheme of support for Minor Research Project entitled "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city." vide UGC letter No. F. No. 41-1373/2012 (SR) dated 30 Jul 2012 has been fully utilized for the purpose for which it was sanctioned and in accordance with the terms and conditions laid down by the University Grants Commission.

SIGNATURE OF THE PRINCIPAL INVESTIGATOR

26/03/21
REGISTRAR/PRINCIPAL
(Seal)
Registrar
Birla Institute of Technology
Mesra; Ranchi

CHIEF ACCOUNTANT /STATUTORY AUDITOR
(Seal)
432532
VIKASH RANJAN & CO.
RANCHI
CHARTERED ACCOUNTANTS

Annexure -III

UNIVERSITY GRANTS COMMISSION
BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002.

Annual/Final Report of the work done on the Minor Research Project.
(Report to be submitted within 6 weeks after completion of each year)

1. Project report: Final

2. UGC Reference No. F. No. 41-1373/2012 (SR) dated 30 Jul 2012

3. Period of report: From Nov 2014 to May 2018

4. Title of research project: "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city."

5. (a) Name of the Principal Investigator: Dr. Manik
   (b) Deptt.: Department of Pharmaceutical Sciences & Technology
   (c) College where work has progressed: Birla Institute of Technology, Mesra, Ranchi

6. Effective date of starting of the project: Nov 2014

7. Grant approved and expenditure incurred during the period of the report:
   a. Total amount approved: Rs. 80,000/- (Received Rs. 45,000/-)
   b. Total expenditure: Rs. 46,350/-
   c. Report of the work done: (Please attach a separate sheet) –
   i. Brief objective of the project:
      1. To monitor the heavy metals contaminations in various vegetables from the road side market and organized market in different areas of Ranchi.
      2. To estimate the concentrations of heavy metal in collected raw vegetables.
      3. To observe whether the concentrations of Heavy metals are in accordance with Prevention of Food Adulteration Act (PFA) and WHO / FAO permissible limits or not.
      4. To assess Health risk and daily intake of heavy metals through vegetables.

   ii. Work done so far and results achieved and publications, if any, resulting from the work (Give details of the papers and names of the journals in which it has been published or accepted for publication
      1. Ratna Ghosh, Reshma Xalxo and Manik Ghosh. Estimation of Heavy Metal in Vegetables from different Market Sites of Tribal based Ranchi City through ICP-OES and to assess Health Risk. Current World Environment 2013, 8(3): 435-444. ISSN: 0973-4929, Online ISSN: 2320-8031 (http://dx.doi.org/10.12944/CWE.8.3.13)
      2. Ratna Ghosh, Reshma Xalxo, Mukul Chandra Gope, Sougata Mishra, Bindu Kumari, Manik Ghosh. Estimation of Heavy Metals in locally available Vegetables collected from Road Side Market Sites (1-4) of different areas of Ranchi City. PHARMBIT, Jan – Dec 2011; XXIII & XXIV (1& 2): 68-73. ISSN: 0973-6204 (Published in 2012)

iii. Has the progress been according to original plan of work and towards achieving the objective? if not, state reasons – Progress was according to original plan of work

iv. Please enclose a summary of the findings of the study. One bound copy of the final report of work done may also be sent to the concerned Regional Office of the UGC. – FINAL REPORT ATTACHED

v. Any other information: Second Installment of Rs. 35,000/- not received

SIGNATURE OF THE PRINCIPAL INVESTIGATOR

21/03/18

REGISTRAR/PRINCIPAL

Registrar

Birla Institute of Technology

Mesra, Ranchi

BIRLA INSTITUTE OF TECHNOLOGY
A Deemed University u/s 3 of UGC Act, 1956
MESRA : RANCHI - 835 215 (INDIA)

Phone : (EPBX) 0651-2275444/2275896, 2276002/2276006 FAX : 0651-2275401/2275351 Website: www.bitmesra.ac.in

UNIVERSITY GRANTS COMMISSION

UTILIZATION CERTIFICATE

It is to certify that an amount of **Rs. 45,000/- (Rupees forty-five thousand only)** has been utilized as on 31st March 2018 from the total amount of **Rs. 45,000/-** released by **University Grants Commission** vide **Sanction No.: F. No. 41-1373/2012 (SR) dated 30th Jul 2012** towards Minor Research Project (MRP) entitled "**Estimation of heavy metals toxicity in vegetables and medicinal plants collected from roadside market sites of different areas of Ranchi city**" to Birla Institute of Technology, Mesra, Ranchi-835215, Jharkhand.

27/07/18

Registrar
Birla Institute of Technology
Mesra, Ranchi

Annexure – V

UNIVERSITY GRANTS COMMISSION
BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002

STATEMENT OF EXPENDITURE IN RESPECT OF MINOR RESEARCH PROJECT

1. Name of Principal Investigator: Dr. Manik
2. Deptt. of PI: Department of Pharmaceutical Sciences & Technology
i. Name of College: Birla Institute of Technology, Mesra, Ranchi
3. UGC approval Letter No. and Date: F. No. 41-1373/2012 (SR) dated 30 Jul 2012
4. Title of the Research Project: "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city."
5. Effective date of starting the project: Nov 2014
6. a. Period of Expenditure: From Nov 2014 to May 2018
   b. Details of Expenditure:

S. No.	Item	Amount Approved (Rs.)	Amount Released (Rs.)	Expenditure Incurred (Rs.)
i.	Books & Journals	10,000/-	10,000/-	10,250/-
ii.	Equipment	nil		
iii.	Contingency	10,000/-	35,000/-	7,842/-
iv.	Field Work/Travel	30,000/-		12,835/-
v.	Hiring Services	nil		
vi.	Chemicals & Glassware	30,000/-		15,423/-
	Total	**80,000/-**	**45,000/-**	**46,350/-**

7. If as a result of check or audit objection some irregularly is noticed at later date, action will be taken to refund, adjust or regularize the objected amounts.
8. It is certified that the grant of Rs. 45,000/- (Rupees Forty-five thousand only) received from the University Grants Commission under the scheme of support for Minor Research Project entitled "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city." vide UGC letter No. F. No. 41-1373/2012 (SR) dated 30 Jul 2012 has been fully utilized for the purpose for which it was sanctioned and in accordance with the terms and conditions laid down by the University Grants Commission.

Signature of Principal Investigator

2.7.9/18

Signature of Registrar
(Seal)

Registrar
Birla Institute of Technology
Mesra, Ranchi

Annexure - VI

UNIVERSITY GRANTS
COMMISSIONBAHADUR SHAH
ZAFAR MARG NEW DELHI – 110 002

STATEMENT OF EXPENDITURE INCURRED ON FIELD WORK

Name of the Principal Investigator: Dr. Manik

Name of the Place visited	Duration of the Visit		Mode of Journey	Expenditure Incurred (Rs.)
	From	To		
Indian Council of Agricultural Research, New Delhi	18.12.2016 Ranchi 20.12.2016 New Delhi	18.12.2016 New Delhi 20.12.2016 Ranchi	Air	12,835/-

Certified that the above expenditure is in accordance with the UGC norms for Major Research Projects

SIGNATURE OF PRINCIPAL INVESTIGATOR

REGISTRAR/PRINCIPAL
Registrar
Birla Institute of Technology
Mesra, Ranchi

Annexure – VIII

UNIVERSITY GRANTS COMMISSION
BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002

PROFORMA FOR SUBMISSION OF INFORMATION AT THE TIME OF SENDING THE FINAL REPORT OF THE WORK DONE ON THE PROJECT

1. Title of the Project: "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city."

2. NAME AND ADDRESS OF THE PRINCIPAL INVESTIGATOR:
Dr. Manik
Assistant Professor
Department of Pharmaceutical Sciences & Technology, ,
Birla Institute of Technology, Mesra, Ranchi – 835215
E-Mail: manik@bitmesra.ac.in; Phone: +919430360991; Fax: +916512275290

3. NAME AND ADDRESS OF THE INSTITUTION:
BIRLA INSTITUTE OF TECHNOLOGY, Mesra, Ranchi – 835215
E-Mail: manik@bitmesra.ac.in; Phone: +919430360991; Fax: +916512275290

4. UGC APPROVAL LETTER NO. AND DATE:
F. No. 41-1373/2012 (SR) dated 30 Jul 2012

5. DATE OF IMPLEMENTATION: Nov 2014

6. TENURE OF THE PROJECT: 4 Years

7. TOTAL GRANT ALLOCATED: Rs. 80,000/-

8. TOTAL GRANT RECEIVED: Rs. 45,000/-

9. FINAL EXPENDITURE: Rs. 46,350/-

10. TITLE OF THE PROJECT: "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city."

11. OBJECTIVES OF THE PROJECT:
Heavy metal contamination may occur due to contaminated water, fertilizers, metal-based pesticides, industrial emissions, transportation, harvesting process, storage and/or sale. In case of road side markets the chances of contaminations are more than that of organized markets. So, our ultimate objectives are:
    1. To monitor the heavy metals contaminations in various vegetables from the road side market and organized market in different areas of Ranchi.
    2. To estimate the concentrations of heavy metal in collected raw vegetables.
    3. To observe whether the concentrations of Heavy metals are in accordance with Prevention of Food Adulteration Act (PFA) and WHO / FAO permissible limits or not.
    4. To assess Health risk and daily intake of heavy metals through vegetables.

12. WHETHER OBJECTIVES WERE ACHIEVED: Yes

1. The concentration levels of Pb, Cd and Ni in vegetables were found to be beyond the PFA permissible limit. The high contamination found in vegetables might be closely related to the pollutants in irrigation water, farm soil or due to pollution from the highways traffic.
2. The concentration levels of these three heavy metals i.e. Lead (Pb), Cadmium (Cd) and Nickel (Ni) were found to be beyond the PFA permissible limit in vegetables.
3. Health Risk Index (HRI) more than 1 is considered to be not safe for human health. In present study, HRI indicates considerable risk and negative impact on human health.

13. ACHIEVEMENTS FROM THE PROJECT

- Two publications in International Journals of repute.
- One B. Pharm. Project
- It is suggested that regular survey of heavy metals should be done on all food commodities in order to evaluate whether any health risks from heavy metal exposure do exist, to assure food safety and to protect the end user from food that might injure their health.

14. SUMMARY OF THE FINDINGS:

In the present study concentrations of Arsenic (As), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Cupper (Cu), Nickel (Ni) and Lead (Pb) were estimated by using an Inductively Coupled Plasma - Optical Emission Spectrometer (ICP-OES). Concentrations of heavy metals were calculated on a dry weight basis. All analyses were replicated six times. The concentration value for Arsenic (As) was found zero in each vegetables since none of the vegetables were contaminated with As.

Among thirteen vegetables, Beet, Cucumber, Pea, Beans, Lady's finger, Corriender leaves and Tomato showed high levels of Lead (Pb) in vegetables collected from all sites. Within the selected vegetables, the highest concentrations of Lead (Pb) were noticed in Peas collected from Site-5 followed by Site-7 and Site-8. Cadmium (Cd) was found in fair amount in Cucumber collected from Site-6 and Site-8 and Spinach from Site-6. Nickel (Ni) was found to be in higher concentrations in Pea and Beans collected from all sites. Lady's finger also contains fair amount of Nickel. **The concentration levels of these three heavy metals i.e. Lead (Pb), Cadmium (Cd) and Nickel (Ni) were found to be beyond the PFA permissible limit in vegetables.** The high contamination found in vegetables might be closely related to the pollutants in irrigation water, farm soil or due to pollution from the highways traffic (Igwegbe et al., 1992; Qiu et al., 2000).

Results of two way ANOVA test showed that variations in the heavy metal concentrations were significant for some heavy metals due to site, vegetable and site-plant interaction. The results of the ANOVA (Two-Factor without Replication) between concentration of heavy metals and PFA permissible limits suggest that in case of Lead, the P value (1.53E-07) is less than the significance level (0.05) and F (68.37) is more than F crit (4.41). The amount of Lead was found to be significantly higher than the PFA limit. The variations in heavy metal concentrations in vegetables of the same site may be ascribed to the differences in their morphology and physiology for heavy metal uptake, exclusion, accumulation and retention. The use of contaminated irrigation water may also increase the uptake and accumulation of the heavy metals in the vegetables.

In context to the hypothesis taken regarding heavy metals concentration in various vegetables collected from road side market of Ranchi city, it was observed from above results that the null hypothesis (i.e. there is no significant difference between the heavy metal concentrations in vegetables collected from road side market of Ranchi city), was found to be wrong and the alternate hypothesis (i.e. there is a significant difference between the heavy metal concentrations in vegetables collected from road side market of Ranchi city) was found to be correct and significant upto $P<0.001$.

Regarding the hypothesis related to heavy metals concentration in various vegetables collected from road side markets and organized markets of Ranchi city, again the alternate hypothesis i.e. there is a significant difference between the heavy metal concentrations in vegetables collected from road side market and organized markets of Ranchi city was found to be correct significant upto $P<0.001$.

15. CONTRIBUTION TO THE SOCIETY

The project is interdisciplinary and is directly linked to the society. Lead and cadmium were among the most abundant heavy metals in the selected vegetables. The excessive content of these heavy metals in food may causes number of diseases. Health Risk Index (HRI) more than 1 (ONE) is considered to be not safe for human health. In present study, HRI indicates considerable risk and negative impact on human health. The present study provides additional data on heavy metals contamination in Ranchi, Jharkhand. It is suggested that regular survey of heavy metals should be done on all food commodities in order to evaluate whether any health risks from heavy metal exposure do exist, to assure food safety and to protect the end user from food that might injure their health.

16. WHETHER ANY PH.D. ENROLLED/PRODUCED OUT OF THE PROJECT: - NA -

17. NO. OF PUBLICATIONS OUT OF THE PROJECT

1. Ratna Ghosh, Reshma Xalxo and Manik Ghosh. Estimation of Heavy Metal in Vegetables from different Market Sites of Tribal based Ranchi City through ICP-OES and to assess Health Risk. Current World Environment 2013, 8(3): 435-444. ISSN: 0973-4929, Online ISSN: 2320-8031 (http://dx.doi.org/10.12944/CWE.8.3.13)

2. Ratna Ghosh, Reshma Xalxo, Mukul Chandra Gope, Sougata Mishra, Bindu Kumari, Manik Ghosh. Estimation of Heavy Metals in locally available Vegetables collected from Road Side Market Sites (1-4) of different areas of Ranchi City. PHARMBIT, Jan – Dec 2011; XXIII & XXIV (1& 2): 68-73. ISSN: 0973-6204 (Published in 2012)

(PRINCIPAL INVESTIGATOR)

27/02/18

(REGISTRAR/PRINCIPAL)

Registrar
Birla Institute of Technology
Mesra, Ranchi

Annexure -VI

UNIVERSITY GRANTS COMMISSION
BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002.

Annual Report of the work done on the Minor Research Project.
(Report to be submitted within 6 weeks after completion of each year)

1. Project report: ANNUAL REPORT

2. UGC Reference No.F. No. 41-1373/2012 (SR) dated 30 Jul 2012

3. Period of report: From Aug 2012 to Oct 2014

4. Title of research project: "Estimation of heavy metals toxicity in vegetables & medicinal plants collected from road side market sites of different areas of Ranchi city."

5. (a) Name of the Principal Investigator: Dr. Manik
   (b) Deptt.: Department of Pharmaceutical Sciences & Technology
   (c) College where work has progressed: Birla Institute of Technology, Mesra, Ranchi

6. Effective date of starting of the project: Aug 2012

7. Grant approved and expenditure incurred during the period of the report:
   a. Total amount approved: Rs. 80,000/- (Received Rs. 45,000/-)
   b. Total expenditure: NIL
   c. Report of the work done: (Please attach a separate sheet) –
   i. Brief objective of the project:
      1. To monitor the heavy metals contaminations in various vegetables from the road side market and organized market in different areas of Ranchi.
      2. To estimate the concentrations of heavy metal in collected raw vegetables.
      3. To observe whether the concentrations of Heavy metals are in accordance with Prevention of Food Adulteration Act (PFA) and WHO / FAO permissible limits or not.
      4. To assess Health risk and daily intake of heavy metals through vegetables.

   ii. Work done so far and results achieved and publications, if any, resulting from the work (Give details of the papers and names of the journals in which it has been published or accepted for publication
      1. Ratna Ghosh, Reshma Xalxo and Manik Ghosh. Estimation of Heavy Metal in Vegetables from different Market Sites of Tribal based Ranchi City through ICP-OES and to assess

Health Risk. Current World Environment 2013, 8(3): 435-444. ISSN: 0973-4929, Online ISSN: 2320-8031 (http://dx.doi.org/10.12944/CWE.8.3.13)

2. Ratna Ghosh, Reshma Xalxo, Mukul Chandra Gope, Sougata Mishra, Bindu Kumari, Manik Ghosh. Estimation of Heavy Metals in locally available Vegetables collected from Road Side Market Sites (1-4) of different areas of Ranchi City. PHARMBIT, Jan – Dec 2011; XXIII & XXIV (1& 2): 68-73. ISSN: 0973-6204 (Published in 2013)

iii. Has the progress been according to original plan of work and towards achieving the objective? if not, state reasons – Progress was according to original plan of work

iv. please enclose a summary of the findings of the study. One bound copy of the final report of work done may also be sent to the concerned Regional Office of the UGC. – SUMMARY ATTACHED

v. Any other information: Second Installment of Rs. 35,000/- not received

Dr. MANIK

Name and Signature of Principal Investigator

30/11/14

Registrar/Principal
(Signature with Seal)

Registrar
Birla Institute of Technology
Mesra; Ranchi

ISSN : 0973-6204

PHARMBIT

INTERNATIONAL JOURNAL

INDEXING IN "CHEMICAL ABSTRACT"

Vol. XXIII & XXIV. No. 1 & 2, Jan - Dec, 2011

The Official publication of

PHARMACEUTICAL SOCIETY

DEPARTMENT OF PHARMACEUTICAL SCIENCES

BIRLA INSTITUTE OF TECHNOLOGY

MESRA, RANCHI, INDIA

www.bitmesra.ac.in

Contents

Estimation of Heavy Metals in Locally Available Vegetables Collected from Road Side Market Sites (1-4) of Different Areas of Ranchi City

Ratna Ghosh[1*], Reshma Xalxo[1], Mukul Chandra Gope[2], Sougata Mishra[3], Bindu Kumari[3], Manik Ghosh[3]
[1*] Department of Home Science, Ranchi University, Ranchi, India
[2] Department of Physiology, Rajendra Institute of Medical Sciences, Ranchi, India
[3] Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra, Ranchi, India

ABSTRACT
Heavy metals are among the major contaminants of food supply and may considered the most important problem to our environment. The hypothesis behind the present study is that the irrigation with waste water, transportation and marketing site of vegetables in contaminated environment may elevate the levels of heavy metals in vegetables through surface deposition. Eight Road side Markets and two organised Markets were demarcated for vegetable purchasing. The present study was focused on Site-1 to Site-4 only. Six vegetables out of thirteen showed higher Metal Pollution Index in Site-3 and Site-4. All sites showed several fold higher concentrations of Lead (Pb), than the permissible PFA limit. Site-4 contains significantly higher concentration of Pb ($P<0.001$) than all other sites. The present study has generated data on heavy metal pollution in and around Ranchi City, Capital of Jharkhand and associated risk assessment for consumer's exposure to the heavy metals.

Key words : *Heavy metals, vegetables, PFA, MPI, ICP-OES*

INTRODUCTION

Food safety is a major public concern worldwide and food consumption has been identified as the major pathway for human exposure to certain environmental contaminants, accounting for >90% of intake compared to inhalation or dermal routes of exposure. About 30% of human cancers are caused by low exposure to initiating carcinogenic contaminants in the diet[1]. During the last decades, the increasing demand of food safety has stimulated research regarding the risk associated with consumption of foods contaminated by pesticides, heavy metals and/or toxins[2].

Heavy metals (Lead, Arsenic, Cadmium, Copper, Chromium and Nickel) contamination of vegetables cannot be underestimated as these foodstuffs are important components of human diet. Vegetables are rich sources of vitamins, minerals, and fibers, and also have beneficial antioxidative effects. However, intake of heavy metal-contaminated vegetables may pose a risk to the human health. Heavy metal contamination of the food items is one of the most important aspects of food quality assurance[3-6]. International and national regulations on food quality have lowered the maximum permissible levels of toxic metals in food items due to an increased awareness of the risk these metals pose to food chain contamination[4].

Heavy metals are among the major contaminants of food supply and may considered the most important problem to our environment[7]. Heavy metals, in general, are not biodegradable, have long biological half-lives and have the potential for accumulation in the different body organs leading to unwanted side effects[8-9]. Lead and cadmium are among the most abundant heavy metals and are particularly toxic. The excessive content of these metals in food is associated with etiology of a number of diseases, especially with cardiovascular, kidney, nervous as well as bone diseases[8, 10-12]. In addition, they are also implicated in causing carcinogenesis, mutagenesis and teratogenesis[13-14].

Heavy metal contamination may occur due to irrigation with contaminated water, addition of fertilizers and metal-based pesticides, industrial emissions, transportation, harvesting process, storage and/or sale. Emissions of heavy metals from the industries and vehicles may be deposited on the vegetable surfaces during their production, transport and marketing.

It is well known that plants take up metals by absorbing them from contaminated soils as well as from deposits on parts of the plants exposed to the air from polluted environments[15-16].

Heavy metals are non-biodegradable and persistent environmental contaminants, which may be deposited on the surfaces and then absorbed into the tissues of vegetables. Plants take up heavy metals by absorbing them from deposits on the parts of the plants exposed to the air from polluted environments as well as from contaminated soils. A number of studies have shown heavy metals as important contaminants of the vegetables. Heavy metal contamination of vegetables may also occur due to irrigation with contaminated water. The potential toxicity, persistent nature and cumulative behavior as well as the consumption of vegetables and fruits, there is necessary to test and analyze these food items to ensure that the levels of these contaminants meet the agreed international requirements.

Regular survey and monitoring programmes of heavy metal contents in foodstuffs have been carried out for decades in most developed countries. But, in developing countries limited data are available on heavy metals. Therefore this study will present data on the level of heavy metals in selected vegetables and this study will be also dealing with the daily intake of these metals through consumption of vegetables.

MATERIALS AND METHODS

Study Areas

The study was conducted around Ranchi city (23°21′ N latitude 85°20′ E longitude and 729 m (2,392 ft) above the sea level) in Jharkhand eastern plains of India during July 2010 to February 2012. Various small scale industries situated in this city. A large area around industries have no access to clean water resources, so farmers use treated and untreated wastewater for irrigation. The hypothesis behind the present study is that the irrigation with waste water, transportation and marketing site of vegetables in contaminated environment may elevate the levels of heavy metals in vegetables through surface deposition. Eight Road side Markets viz. Site-1 (Lalpur Market), Market Site-2 (BIT More Market), Site-3 (Daily Market), Site-4 (Kanke Road Market), Site-5 (Booti More Market), Site-6 (RIMS Market), Site-7 (Morabadi Stadium Market), Site-8 (Bahu Bazar Market) and two organised Markets i.e. Site-9 (Reliance Fresh) & Site-10 (Big Bazar) were demarcated for vegetable purchasing. The present study was focused on Site-1 to Site-4 only.

Sampling

The edible portions of Vegetables were collected from different markets during July 2010 to June 2011. Samples were brought back to the laboratory and washed with clean tap water to remove the soil particles and dusts of the vegetables. After removing the extra water from the surface of vegetables with blotting paper, samples were cut into pieces, packed into separate bags, and kept in an oven until a constant weight was achieved. The dried samples were grinded and passed through a sieve of 2 mm size and then kept at room temperature for further analysis.

Digestion of Plant Samples

0.5 gm of the dried powdered sample was digested in Microwave Digester 3000 SOLV at 1400 watt for 3 hours in the solvent system of HNO_3 : H_2O_2, in 7:0.5 ratio until a transparent solution was obtained. After cooling, the digested sample was filtered using Whatman Grade No. 44 (Quantitative Filter Paper, Ashless) and the filtrate was finally maintained to 100 ml with distilled water.

Analysis of Heavy Metals

Concentrations of As, Cd, Co, Cr, Cu, Mn, Ni and Pb in the filtrate of digested plant samples were estimated by using an Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) (Model Optical 2100DV ICP-OES, Perkin Elmer, USA) with argon laser. The Spectral range was of 160 nm to 900 nm and resolutuion of 0.009 nm at 200 nm. The instrument was fitted with UV sensitive dual backside – illuminated CCD array detector.

DATA ANALYSIS

Metal Pollution Index (MPI)

To examine the overall heavy metal concentrations in all vegetables, metal pollution index (MPI) was computed. This index was obtained by calculating the geometrical mean of concentrations of all the metals in the vegetables[17].

$$\text{MPI } (\mu g/g) = (Cf_1 \times Cf_2 \times \ldots \times Cf_n)^{1/n}$$

Where, Cf_n = concentration of metal n in the sample.

Statistical Analysis

The data of heavy metal concentrations in the vegetables of different sites (Site-1 to Site-4) were subjected to two way analysis of variance (ANOVA) test for assessing the significance of differences in heavy metal concentrations due to different irrigation practices, environmental pollutants, etc followed by Bonferroni's multiple comparison test. All the statistical tests were performed using GraphPad Prism.

RESULTS

Metal Pollution Index

Metal Pollution Index (MPI) is suggested to be a reliable and precise method for metal pollution monitoring of wastewater irrigated areas[17]. Among different vegetables in Site-1, pea showed highest value of MPI followed by spinach. In Site-2 pea, spinach and beans showed higher MPI. Six vegetables out of thirteen showed higher MPI i.e. more than 2, in Site-3 and Site-4. These were pea, spinach, tomato, cucumber, lady finger and beans. Higher MPI suggests that these vegetables may cause more human health risk due to higher accumulation of heavy metals in the edible portion.

Concentration of Heavy Metals

Heavy metal concentrations showed variations among different vegetables collected from different market sites (Figure 1 to Figure 6). All sites showed several fold higher concentrations of Lead (Pb), than the permissible PFA limit, in cucumber (range, 7.10 ± 1.52 ppm to 12.67 ± 1.52 ppm), pea (range, 6.17 ± 1.51 ppm to 20.67 ± 3.29 ppm) and tomato (range, 5.00 ± 1.97 ppm to 9.67 ± 0.33 ppm). Site-1 and Site-4 also showed higher concentration of Pb in lady finger (6.40 ± 2.89 ppm and 7.83 ± 1.51 ppm, respectively). Site-2 and Site-4 showed higher concentration of Pb in beet, beans and spinach. Apart from these Site-3 showed higher concentration of Pb in cabbage and coriander. Nickel (Ni) was found to be higher in pea (4.33 ± 0.56 ppm to 4.83 ± 0.60 ppm) and beans (3.83 ± 0.48 ppm to 4.67 ± 0.42 ppm) than PFA permissible limit in all sites. Cucumber (2.93 ± 0.31 ppm) of Site-3 and tomato

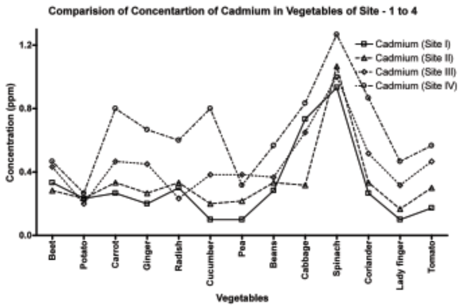

Figure 1 : *Concentration of Cadmium (Cd) in vegetables of all four sites*

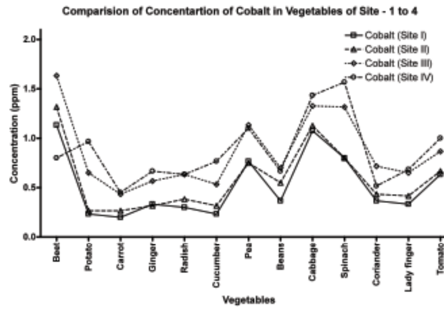

Figure 2 : *Concentration of Cobalt (Co) in vegetables of all four sites*

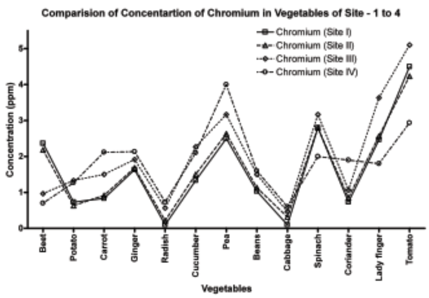

Figure 3 : *Concentration of Chromium (Cr) in vegetables of all four sites*

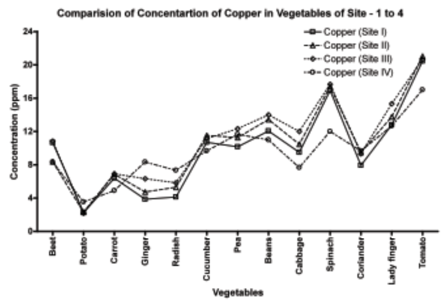

Figure 4 : *Concentration of Copper (Cu) in vegetables of all four sites*

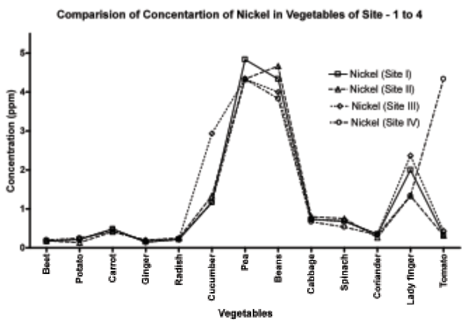

Figure 5 : *Concentration of Nickel (Ni) in vegetables of all four sites*

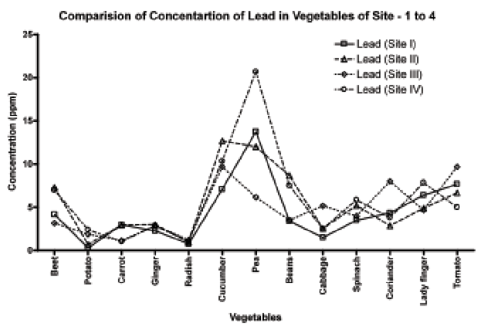

Figure 6 : *Concentration of Lead (Pb) in vegetables of all four sites*

(4.33 ± 0.67 ppm) of Site-4 were found to contain high amount of Ni than the permissible PFA limit. Site-4 contains significantly higher concentration of Pb (*P<0.001*) than all other sites. Concentration of Nickel in cucumber of Site-3 and tomato of Site-4 were significantly higher (*P<0.001*) in comparison to all other sites. Cadmium was found to be significantly higher Cucumber (*P<0.01*) and Coriander (*P<0.05*) of Site-4 in comparison of Site-1. Rest all heavy metals were found to be below the PFA permissible limit in all vegetables collected from different sites. Results of two way ANOVA test showed that variations in the heavy metal concentrations were significant for some heavy metals due to site, vegetable and site plant interaction (Table 1). The variations in heavy metal concentrations in vegetables of the same site may be ascribed to the differences in their morphology and physiology for heavy metal uptake, exclusion, accumulation and retention[18-19]. The use of contaminated irrigation water may also increase the uptake and accumulation of the heavy metals in the vegetables.

CONCLUSION

The variations in the concentrations of the heavy metals in vegetables observed during the present study may be ascribed to the physical and chemical nature of the soil of the production sites, absorption capacities of heavy metals by vegetables, atmospheric deposition of heavy metals, which may be influenced by innumerable environmental factors such as temperature, moisture and wind velocity, and the nature of the vegetables, i.e. leafy, root, fruit, exposed surface area, hairy or smoothness of the exposed parts[20]. The variations in the concentrations of heavy metals in the vegetables tested may also be ascribed to the variations in the anthropogenic activities such as heavy traffic, addition of phosphate fertilizers or use of metal-based pesticides around production sites and urban industrial activities at market sites.

Table 1. Variation in heavy metal concentration vegetables of different sites

S. No.	Vegetable	Heavy metal	Site	Concentration (ppm)	Level of Significance
1	Pea	Lead	Site-1	13.733 ± 2.733**	Site-4 vs All
			Site-2	12.000 ± 1.932***	**P<0.01
			Site-3	06.167 ± 1.515***	***P<0.001
			Site-4	20.667 ± 3.293	
2	Tomato	Nickel	Site-1	0.333± 0.076***	Site-4 vs All
			Site-2	0.317± 0.054***	***P<0.001
			Site-3	0.433± 0.076***	
			Site-4	4.333± 0.667	
3	Cucumber	Nickel	Site-1	1.167± 0.167**	Site-3 vs All
			Site-2	1.333± 0.211***	**P<0.01
			Site-3	2.933± 0.981	***P<0.001
			Site-4	1.167± 0.211***	
		Cadmium	Site-1	0.100 ± 0.068**	Site-4 vs All
			Site-2	0.200± 0.045*	**P<0.01
			Site-3	0.383± 0.083ns	*P<0.05
			Site-4	0.800± 0.073	ns: Non significant
4	Coriander	Cadmium	Site-1	0.267± 0.123*	Site-4 vs All
			Site-2	0.333± 0.120ns	*P<0.05
			Site-3	0.517± 0.122ns	ns: Non
			Site-4	0.867± 0.152	significant

Results represented as Mean ± SEM (n=6). Two way analysis of variance (ANOVA) followed by Bonferroni's multiple comparison test.

The present study has generated data on heavy metal pollution in and around Ranchi City, Capital of Jharkhand and associated risk assessment for consumer's exposure to the heavy metals. The proposed hypothesis that the transportation and marketing of vegetables in contaminated environment may elevate the levels of heavy metals in vegetables through surface deposition has been proved through this study. Appropriate precautions should also be taken at the time of transportation and marketing of vegetables.

Heavy metals have a toxic impact, but detrimental impacts become apparent only when long-term consumption of contaminated vegetables occurs. It is therefore suggested that regular monitoring of heavy metals in vegetables and other food items should be performed in order to prevent excessive build-up of these heavy metals in the human food chain.

ACKNOWLEDGEMENT

The authors are thankful to the Head, Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra for providing the facilities to carry out this work and Mr. Sanjay Swain (Technician), Central Instrumentation Facility, Birla Institute of Technology, Mesra Ranchi for his constant help during the analytical work.

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PHARMBIT

ISSN : 0973-6204

Scientific Journal of Pharmaceutical Society

Vol. XXIII & XXIV. No. 1 & 2, Jan - Dec, 2011

Indexing in "Chemical Abstract & Natural Science Database"

CONTENTS

Single copy INR 250

Printed & Published by :
Dr. R.N. Gupta, *Editor*
on behalf of Pharmaceutical Society
Department of Pharmaceutical Sciences
BIRLA INSTITUTE OF TECHNOLOGY
Mesra, Ranchi, India
Printed at Kailash Paper Conversion Pvt. Ltd.
2, Bharatpuri, Purulia Road, Ranchi-834001
Tel.: 0651-2532251, Fax : 0651-2532285

1788/08/12 • KAILASH PAPER, TEL.: 0651-2532251

Current World Environment Vol. **8**(3), 435-444 (2013)

Estimation of Heavy Metal in Vegetables From Different Market Sites of Tribal Based Ranchi City Through ICP-OES and to Assess Health Risk

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http://dx.doi.org/10.12944/CWE.8.3.13

(Received: November 03, 2013; Accepted: November 28, 2013)

ABSTRACT

Inductively Coupled Plasma - Optical Emission Spectrometry (ICP-OES) was used to estimate and evaluate the levels of heavy metals in vegetables collected from various sites of Ranchi city (tribal dominated population) followed by health risk assessment by determining Metal Pollution Index (MPI), Daily intake of metal (DIM) and Health Risk Index (HRI). The concentration levels of Pb, Cd and Ni in vegetables were found to contain beyond than the permissible PFA limit. All sites showed quite a few higher concentrations of Lead (Pb), than the permissible PFA limit. Among thirteen vegetables, Beet, Cucumber, Pea, Beans, Lady's finger, Corriender leaves and Tomato showed high levels of Pb in vegetables collected from all sites. Health Risk Index was also found > 1 for Cd, Co and Pb. Health Risk Index for Cadmium was 1.64 and 2.38 in Cucumber from Site-6 and Site-8 respectively. In Spinach it was 2.19 and 2.15 respectively for Site-6 and Site-8. Health Risk Index for Pb was > 1 in Cucumber (All sites; 3.54 in Site-8), Pea (All sites except Site-10; 2.45 in Site-7), Beans (All sites; 1.38 in Site-9), Lady's finger (All sites; 2.03 in Site-7), and Tomato (All sites except Site-10; 2.79 in Site-8). Lead and cadmium were among the most abundant heavy metals in the selected vegetables. The excessive content of these heavy metals in food may causes number of diseases. HRI more than 1 is considered to be not safe for human health. In present study, HRI indicates considerable risk and negative impact on human health.

Key words: Vegetables, ICP-OES, Health Risk Index (HRI), Metal Pollution Index (MPI), Ranchi, Tribal.

INTRODUCTION

Heavy metals are important environmental pollutants, particularly in areas with high anthropogenic pressure. Their presence in the plants, atmosphere, soil and water, even in traces, can cause serious problems to all organisms. The presence of heavy metals in sewage sludge, used as agricultural fertilizer is a major problem for crop and environmental qualities and their impact on human health, because most of the heavy metals are persistent due to their immobile nature (Devkota *et al*., 2000; Itanna, 2002; Keller *et al*., 2002; McBride, 2003). The transfers of metals from bio solids to soil and subsequently to plants pose potential health risks because they can enter the food chain and the environment (Ghaedi *et al*., 2008). Plant uptake is one of the major pathways by which sludge-borne heavy metals enter the food chain (Chaney, 1990). Inputs of heavy metals to agricultural soils generate negative impact on soil fertility and accumulate in the human food chain (McLaughlin *et al*., 1999). Food contamination by heavy metals depends both on their mobility in the soil and their bioavailability. Though some of the mobility and bioavailability factors are easy to measure, determination of the food risk contamination is tricky. Regulation, handling and bioremediation of hazardous materials require an assessment of the risk to some living species other than human being, or assessment of hazard to the entire ecosystem. Heavy metal accumulation in soils is of concern in agricultural production due to the adverse effects on food quality (safety and marketability), crop growth (due to phytotoxicity) (Ma *et al*., 1994; Msaky and Calvert, 1990; Fergusson, 1990) and environmental health (soil flora/fauna and terrestrial animals). The mobilization of heavy metals into the biosphere by human activity has become an important process in the geochemical cycling of these metals. This is acutely evident in urban areas where various stationary and mobile sources release large quantities of heavy metals into the atmosphere and soil, exceeding the natural emission rates (Nriagu, 1989; Bilos *et al*., 2001) and it is often caused by accidental releases of chemicals or the improper disposal of hazardous waste. Increased inputs of metals and synthetic chemicals in the terrestrial environment due to rapid industrialization coupled with inadequate environmental management in the developing country like India, has led to large-scale pollution of the environment. These chemicals in the terrestrial environment clearly pose a significant risk to the quality of soils, plants, natural waters and human health. Heavy metal content of soil is of major significance in relation to their fertility and nutrient status (Gowd *et al*., 2010).

However, high concentrations of these metals become toxic. Other metals, which are not included in the group of essential elements, such as Pb or Cr, may be tolerated by the ecosystem in low concentration, but become harmful in higher concentrations. Soluble metal compounds and metals held in exchange complexes are considered to be available to vegetation uptake. Prolonged exposure to heavy metals such as cadmium, copper, lead, nickel can cause deleterious health effects in humans (Reilly 1991).

In this study, we estimated the concentrations of Cd, Co, Cr, Cu, Pb, Ni and As in vegetables collected from different market sites of Ranchi city (tribal dominated population). The study was conducted around Ranchi (23°21' N latitude 85°20' E longitude and 729 m (2,392 ft) above the sea level) city in Jharkhand eastern plains of India. Various small scale industries situated in this city. A large area around industries have no access to clean water resources, so farmers use treated and untreated wastewater for irrigation. The long term uses of treated and untreated wastewater for irrigation may also increase the uptake and accumulation of the heavy metals in the vegetables. From the cultivated sites these vegetables are supplied to the wholesale vegetable market and the rest enter the local markets. The levels of contamination were compared with the PFA and ASTDR guidelines to assess the potential hazards of heavy metals to public health. The hypothesis behind the present study is that the irrigation with waste water, transportation and marketing site of vegetables in contaminated environment may elevate the levels of heavy metals in vegetables through surface deposition.

MATERIALS AND METHODS

Study Area

Thirteen vegetables were collected randomly from the different market sites of urban and suburban area of Ranchi, Jharkhand to estimate the total heavy metal content in these samples. Ranchi City (23°21' N latitude 85°20' E longitude and 729 m (2,392 ft) above the sea level) is Capital of Jharkhand, India. Eight Road side Markets viz. Site-1 (Lalpur Market), Market Site-2 (BIT More Market), Site-3 (Daily Market), Site-4 (Kanke Road Market), Site-5 (Booti More Market), Site-6 (RIMS Market), Site-7 (Morabadi Stadium Market), Site-8 (Bahu Bazar Market) and two organized Markets i.e. Site-9 (Reliance Mart and Site-10 (Big Bazar) were demarcated for vegetable purchasing. A report related to Site-1 to Site-4 has already been discussed in one of the previous study (Ghosh *et al*., 2011). The present study was focused on Site-5 to Site-10 only.

Sampling

The freshly mature vegetables were brought to the laboratory and washed primarily with running clean tap water to remove the soil particles. After removing the extra water from the surface of vegetables with blotting paper, samples were cut into pieces, packed into separate bags, and kept in an oven until a constant weight was achieved. Sample were dried in oven at 70 °C for 48 h and then ground to powder. The grinded samples were passed through a sieve of 2 mm size and then kept at room temperature for further analysis.

Analytical Procedure

Digestion

Dried and sliced vegetable samples were grounded into powder with an electric blender and stored in polythene bags until analysis. Approximately 0.5 grams of samples were digested in replicate (along with a blank) with 7 ml HNO_3 and 0.5 ml H_2O_2 in a MultiwaveTM 3000 Microwave digestion system (Anton Paar). This is an industrial type microwave oven which can be equipped with various accessories to optimize the sample digestion. In this case, pre cleaned HF-100 vessels were used in an 8-position rotor. A pressure / Temperature (P/T) Sensor Accessory, which simultaneously measures temperature and pressure for one vessel, was also used. All vessels temperature were monitored with the IR Temperature Sensor Accessory. This device gives thermal protection to the reactions in all of the vessels by measuring the temperature remotely on the bottom surface of each vessel liner during the digestion process. The digestion program consisted of 30 minutes of heating and 15 minutes of cooling as shown in Table 2.

The samples were completely dissolved, resulting in clear solution. After cooling, the digested sample was filtered using Whatman® Quantitative Filter paper, Ashless Grade No. 44 and the filtrate was finally maintained to 100 ml with Millipore water.

All reagents used were Merck, analytical grade (AR) including Standard Stock Solutions of known concentrations of different heavy metals. Heavy metal concentrations of vegetable samples were estimated by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) (Model Optical 2100DV ICP-OES, Perkin Elmer, USA) with argon laser. The Spectral range was of 160 nm to 900 nm and resolution of 0.009 nm at 200 nm. The instrument was fitted with UV sensitive dual backside - illuminated CCD array detector.

Concentrations of heavy metals were calculated on a dry weight basis. All analyses were replicated six times. To assess the contamination level of heavy metals, mean, median, minimum, maximum and standard deviation of vegetable samples were performed by using Microsoft Excel (Version 2007).

Data Analyses

Metal Pollution Index (MPI)

To examine the overall heavy metal concentrations in all vegetables, metal pollution index (MPI) was computed. This index was obtained by calculating the geometrical mean of concentrations of all the metals in the vegetables (Usero *et al*, 1997).

MPI (µg/g) = (Cf1 x Cf2 x . . . x Cfn)1/n

Where, Cfn = concentration of metal n in the sample.

Table 1: Common Name and Botanical Name of vegetables used in study

S.No.	Common Name	Botanical Name	Family	Veg. Category
1	Spinach	Spinacia oleracea	Chenopodiaceae	Leafy
2	Corriander	Coriandrum sativum	Apiaceae	Leafy
3	Cabbage	Brasssica oleracea	Brassicaceae	Leafy
4	Beet root	Beta vulgaris	Amaranthaceae	Roots and Tubers
5	Carrot	Ductus carrotus	Apiaceae	Roots and Tubers
6	Radish	Raphanus sativus	Brassicaceae	Roots and Tubers
7	Ginger	Gingiber officinalis	Zingiberaceae	Roots and Tubers
8	Potato	Solanum tuberosum	Solanaceae	Roots and Tubers
9	Bean	Phaseolus vulgaris	Fabaceae	Legumes
10	Pea	Pysum sativum	Fabaceae	Legumes
11	Cucumber	Cucumis sativus	Cucurbitaceae	Others
12	Lady's finger	Abelmoschus esculentus	Malvaceae	Others
13	Tomato	Lycopersicum esculentum	Solanaceae	Others

Estimate Daily Intake of Heavy Metals

Data of the average diet per person per day were collected from a survey .The average daily vegetable intake was calculated by conducting a survey where 50 people having average body weight of 60 kg and age group 18 years to 70 years were asked for their daily intake of particular vegetable from the each experimental area in every sampling.

Daily intake was calculated by the following equation:

Daily intake of metal (DIM) = $C_{metal} \times D_{food\ intake} / B_{average\ weight}$

Where, C_{metal}, $D_{food\ intake}$ and $B_{average\ weight}$ represent the heavy metal concentrations in vegetables (µg g^{-1}), daily intake of vegetables and average body weight respectively (Singh *et al*., 2010).

Health Risk Index (HRI)

The health risk index was calculated as the ratio of estimated exposure of test vegetables and oral reference dose (Cui *et al*., 2004). Oral reference doses (RfDo) were 4 x 10^{-2} and 1 x 10^{-3} mg kg^{-1} day^{-1} for Cu and Cd, respectively (USEPA, 2002) and 0.004, 0.02 and 1.5 mg kg^{-1} day^{-1} for Pb, Ni and Cr, respectively (USEPA, 1997). Estimated exposure is obtained by dividing daily intake of heavy metals by their safe limits. An index more than 1 is considered as not safe for human health (USEPA, 2002).

Therefore, Health Risk Index = DIM/RfDo,

Here, RfDo (Oral Reference Dose) represents safe levels of exposure by oral for life time

Statistical Analysis

Concentrations of heavy metals were calculated on a dry weight basis. All analyses were replicated six times. To assess the contamination level of heavy metals, mean, median, minimum, maximum and standard deviation of vegetable samples. The Metal Pollution Index of the vegetables of different sites (Site-1 to Site-10) were subjected to two way analysis of variance (ANOVA) test for assessing the significance of differences in heavy metal concentrations due to different sites, irrigation practices, environmental pollutants, etc. All analysis was performed by using Microsoft Excel (Version 2007) and GraphPrism 5.

Table 2: Microwave digestion program for the sample preparation

Step	Power (W)	Ramp (min)	Hold (min)	Fan
1	1000	15	15	1
2	0	0	15	3

RESULTS AND DISCUSSION

Concentration levels of heavy metals

In present study, the concentration range of various heavy metals such as Cadmium (Cd), Cobalt (Co), Copper (Cu), Chromium (Cr), Nickel (Ni) and Lead (Pb) in different vegetables collected from road side market and organized market were estimated. The mean concentrations of Cd, Ni and Pb found in vegetables collected from local markets were summarized in graphical form (Fig 1 to 3). All sites showed quite a few higher concentrations of Lead (Pb), than the permissible PFA limit. Among thirteen vegetables, Beet, Cucumber, Pea, Beans, Lady's finger, Corriender and Tomato showed high levels of Pb in vegetables collected from all sites. Within the selected vegetables, the highest concentrations of Pb were noticed in Peas collected from Site-5 followed by Site-7 and Site-8. Cadmium (Cd) was found in fair amount in Cucumber collected from Site-6 and Site-8 and Spinach from Site-6. Nickel (Ni) was found to be in higher concentrations in Pea and Beans collected from all sites. Lady's finger also contains fair amount of Ni. The concentration levels of these three heavy metals (Pb, Cd and Ni) in vegetables were found to contain beyond than the permissible PFA limit. The high contamination found in vegetables might be closely related to the pollutants in irrigation water, farm soil or due to pollution from the highways traffic (Igwegbe *et al*., 1992; Qiu *et al*., 2000).

The concentration of Cadmium (Cd) ranges from 0.116 ppm to 2.150 ppm in various vegetables. The maximum concentration of Cd

2.150 ppm was found in spinach collected from Site-8, while minimum concentration 0.116 ppm was found in Pea collected from Site-5. Cd concentration was found to be significantly higher (P<0.001) in Cucumber and Spinach in both Site-6 and Site-8 in comparison to all other sites. Health Risk Index (HRI) was found more than 1.64 and 2.38 in Cucumber from Site-6 and Site-8 respectively. In Spinach the HRI was 2.19 and 2.15 respectively for Site-6 and Site-8 (Fig 4). HRI more than 1 is considered as not safe for human health (USEPA, 2002). Acute doses of Cadmium can cause severe gastrointestinal irritation, vomiting, diarrhea, and excessive salivation, and doses of 25 mg of Cd/kg body weight can cause death. Low-level chronic exposure to Cd can cause adverse health effects including gastrointestinal, hematological, musculoskeletal, renal, neurological, and reproductive effects. The main target organ for Cd following chronic oral exposure is the kidney (ATSDR 1999a).

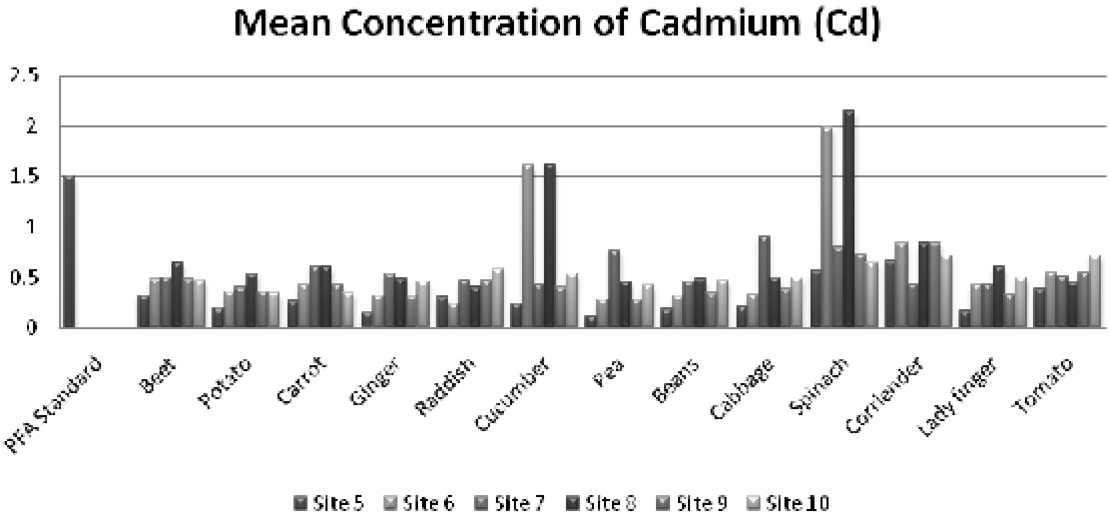

Fig. 1: Mean concentration (n=6) of Cadmium (Cd) in all vegetables collected from Site-5 to Site-10 in comparison to PFA Standard Limit. Cd concentration was found to be significantly higher (P<0.001) in Cucumber and Spinach in both Site-6 and Site-8 in comparison to all other sites.

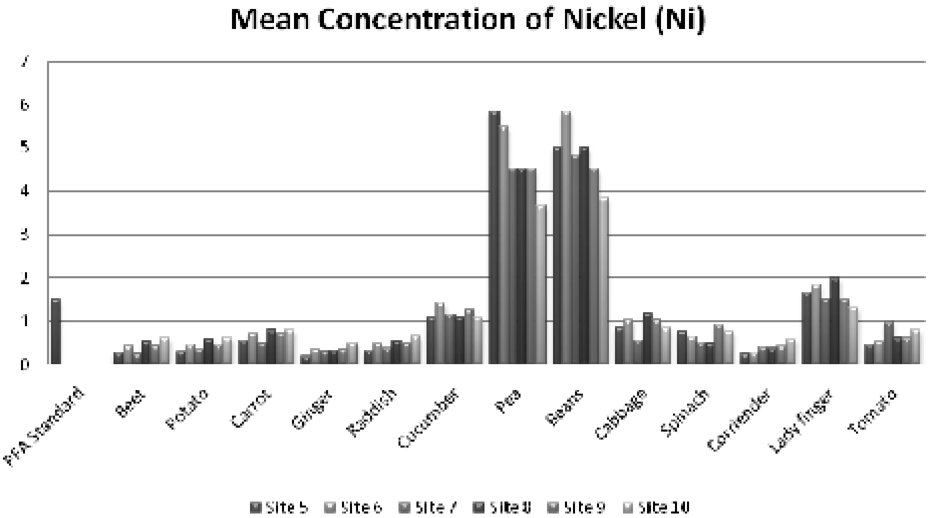

Fig. 2: Mean concentration (n=6) of Nickel (Ni) in all vegetables collected from Site-5 to Site-10 in comparison to PFA Standard Limit. Nickel was found to be significantly higher (P<0.001) in Pea and Beans of Site-5 and Site-6 respectively in comparison to Site-10

The Cobalt (Co) content varies from 0.300 ppm to 1.466 ppm. The lowest concentration 0.300 ppm of Co was observed in Carrot collected from Site-5. On the other hand, Beet from Site-7 showed highest concentration of Co i.e. 1.466 ppm but was less than what observed in Site-3 (1.633 ppm) (Ghosh *et al*, 2011). Health risk index was found more than 1 in Cabbage (All sites except Site-5; 1.46 in Site-6), Tomato (Site-6, 8 and 10), Potato (Site-9 and Site-10; 1.57 in Site-10) and Spinach (Site-7 and Site-10) (Fig 5). Higher concentrations of Co were observed in Beet of Site-3 and Spinach of Site-4 which were significantly higher (P<0.05) than all other sites (Ghosh *et al*, 2011). Overdose of Co may lead to angina, asthma, cardiomyopathy, polycythemia and dermatitis. The safety limit for human consumption of Co is 0.05 to 1 mg/day in humans (ATSDR 1994).

Present investigation reveals that Copper (Cu) varies from 3.933 ppm to 22.300 ppm. The highest concentration of Cu was found in Tomato collected from Site-8 (22.300 ppm), while lowest concentration 3.93 ppm was recorded in Potato

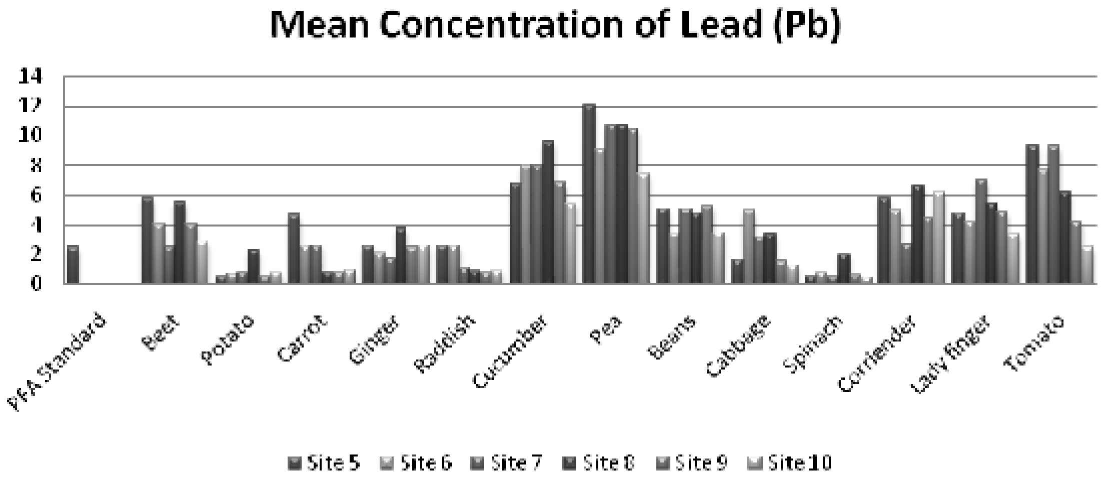

Fig. 3: Mean concentration (n=6) of Lead (Pb) in all vegetables collected from Site-5 to Site-10 in comparison to PFA Standard Limit. Pb concentration was found to be significantly higher (P<0.01) in Pea of Site-5 in comparison to all other sites.

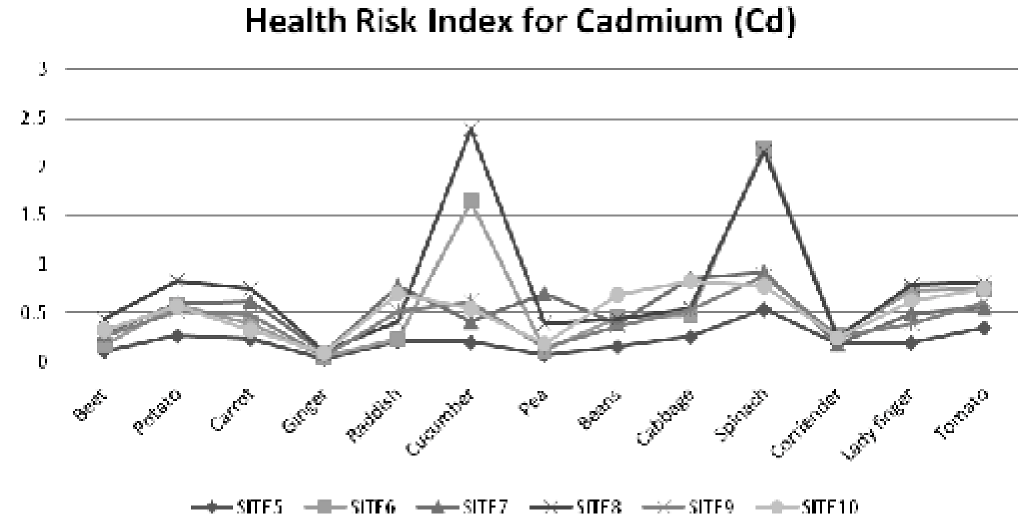

Fig . 4: Health Risk Index (HRI) for Cadmium (Cd) in all vegetables collected from Site-5 to Site-10. (HRI Cd > 1 in Cucumber of Site-6 & Site-8; Spinach of Site-6)

collected from Site-7. Chromium (Cr) concentration varies from 0.266 to 7.833 ppm. The highest concentration of Cr was found in Tomato collected from Site-6 (7.833 ppm), while lowest concentration 7.833 ppm was recorded in Radish and Cabbage collected from Site-7. The presence of Nickel (Ni) ranges from 0.200 to 5.833 ppm in various vegetables. Peas from Site-5 and Beans from Site-6 showed high content of Nickel 5.833 ppm, while Ginger from Site-5 contains low value of Ni 0.200 ppm. Nickel was found to be significantly higher (P<0.001) in Pea and Beans of Site-5 and Site-6 respectively in comparison to Site-10. Excess intake of Ni leads to hypoglycemia, asthma, nausea, headache, and epidemiological symptoms like cancer of nasal cavity and lungs.

During the present study, the concentration of Lead (Pb) content varies from 0.466 ppm to 12.066 ppm. High concentration of Pb was found in Peas collected from Site-5 (12.066 ppm) but less than what observed in Site-1 (13.733

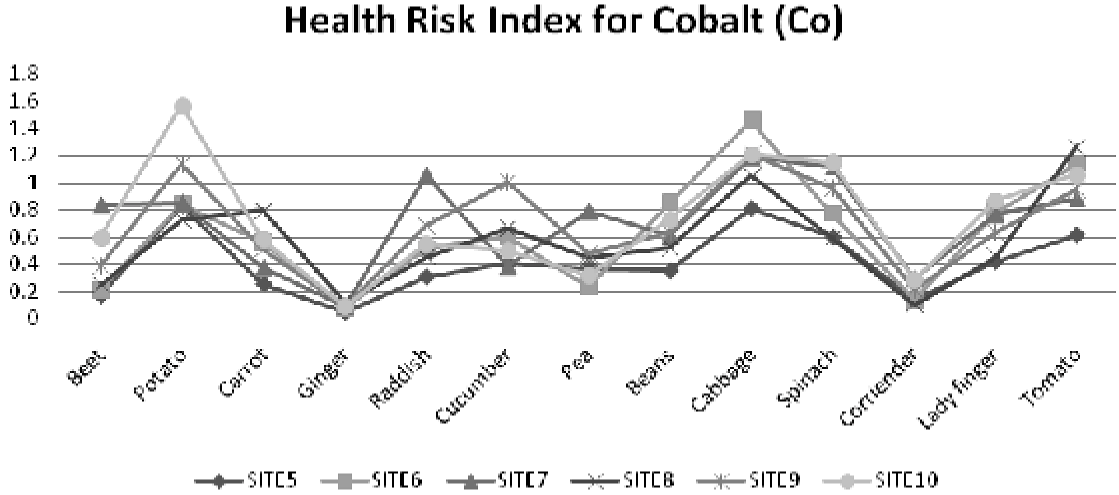

Fig. 5: Health Risk Index (HRI) for Cobalt (Co) in all vegetables collected from Site-5 to Site-10. (HRI Co > 1 in Potato and Cucumber of Site-9; Potato of Site-10; Raddish, Spinach, Cabbage of Site-7; Tomato of Site-6, Site-8 & Site-10)

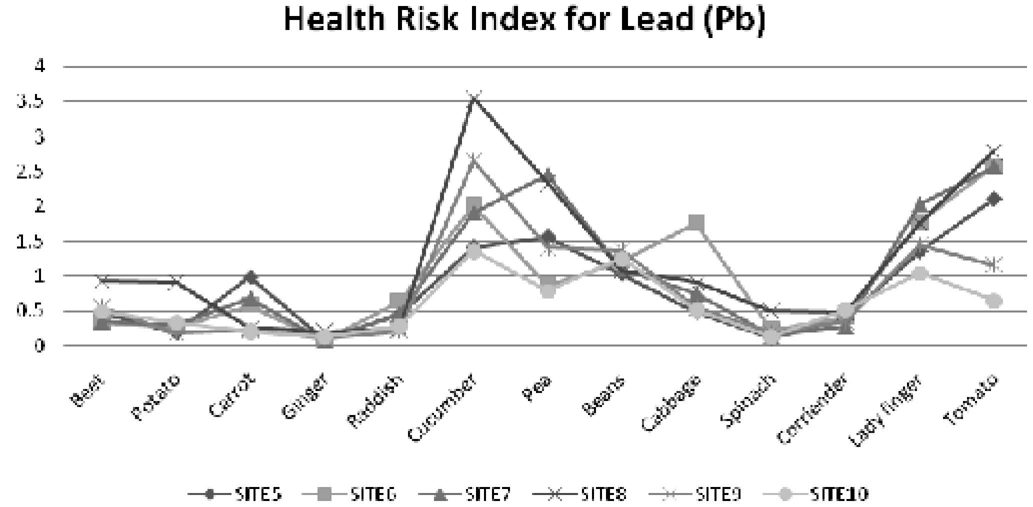

Fig. 6: Health Risk Index (HRI) for Lead (Pb) in all vegetables collected from Site-5 to Site-10. (HRI Pb > 1 in Cucumber, Beans and Lady finger of all Sites; Pea of Site-5, Site-7, Site-8 & Site-9; Cabbage of Site-6; Tomato of all Sites except Site-10)

ppm) (Ghosh *et al*, 2011). The concentration of Lead was found to be significantly higher ($P<0.01$) in Tomato, Pea and Cucumber of Site-5 in comparison to Site-2, Site-3, Site-9 and Site-10. Spinach collected from Site-10 showed low concentration of Pb (0.466 ppm). Health risk index for Pb was found more than 1 in Cucumber (All sites; 3.54 in Site-8), Pea (All sites except Site-10; 2.45 in Site-7), Beans (All sites; 1.38 in Site-9), Lady's finger (All sites; 2.03 in Site-7), and Tomato (All sites except Site-10; 2.79 in Site-8) (Figure 6). Todd (1996) emphasized that most of the accumulated Lead is sequestered in the bones and teeth. This causes brittle bones and weakness in the wrists and fingers. Lead that is stored in bones can reenter the blood stream during periods of increased bone mineral recycling (i.e., pregnancy, lactation, menopause, advancing age, etc.). Mobilized lead can be redeposited in the soft tissues of the body and can cause musculoskeletal, renal, ocular, immunological, neurological, reproductive, and developmental effects (ATSDR 1999b).

Metal Pollution Index (MPI) is suggested to be a reliable and precise method for metal pollution monitoring. Among different vegetables pea showed highest value of MPI followed by Cucumber. Seven vegetables out of thirteen showed higher MPI i.e. more than 2. These were pea, cucumber, tomato, beans, spinach, lady finger and cabbage. Higher MPI suggests that these vegetables may cause more human health risk due to higher accumulation of heavy metals in the edible portion. Metal Pollution Index of various vegetables from all sites (Site-1 to Site-10) was reported in Figure 7. Site-6 and Site-8 can be classified as high risk sites as the MPI of all vegetables were higher than 1. Among the vegetables Pea, Beans. Beet and Cucumber were found to be highly contaminated with the heavy metals. The results of the Anova (Two-Factor without Replication) suggests that in case of vegetables, the P value (1.12E-38) was found to be less than the significance level (0.05), and F (50.513) was more than F crit (1.843) i.e. there is significant difference between MPI among the vegetables. Similarly, in case of Sites, the P value (4.65E-10) was less than the significance level (0.05), so we can reject the null hypothesis that the means are equivalent. F (9.017) was observed more than F crit (1.968) so we can reject the null hypothesis i.e. there is a significant difference between MPI among the Sites.

CONCLUSION

The heavy metals not only affect the nutritive values of vegetables but also have deleterious effect on human beings using these food items. The value of HRI more than 1 indicates considerable risk of negative impact on human health. National and International regulations on food quality have lowered the maximum permissible levels of toxic metals in human food; hence, an increasingly important aspect of food quality should be to control the concentrations of trace metals in food (Radwan *et al*., 2006). The residues of the

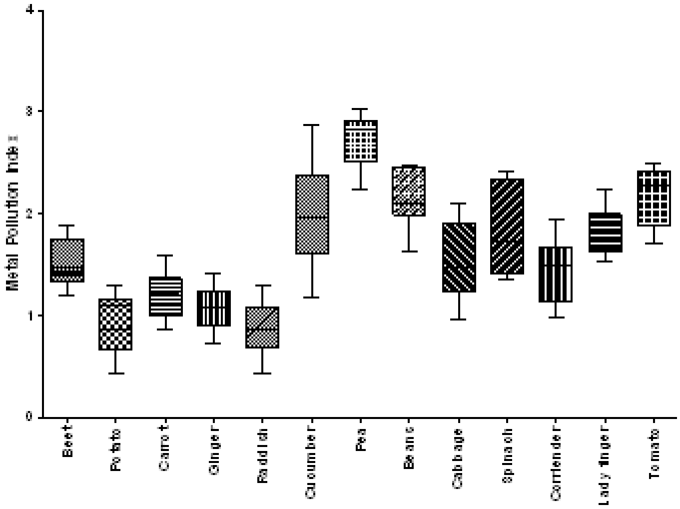

Fig. 7: Metal Pollution Index of various vegetables for all sites (Site-1 to Site-10)

heavy metals still appear as pollutants in vegetables as well as in the environment. Their occurrence and long-range transport at local, regional and global scales has been recently investigated. In developing countries heavy metal contamination is receiving increasing attention from the public as well as governmental organization. There are few major pathways for human exposure to heavy metal contamination in vegetables. Data here demonstrates that Pb, Cd and Co in vegetables may pose health risk to consumer.

The present study provides additional data on heavy metals contamination in Ranchi, Jharkhand. It is suggested that regular survey of heavy metals should be done on all food commodities in order to evaluate whether any health risks from heavy metal exposure do exist, to assure food safety and to protect the end user from food that might injure their health.

ACKNOWLEDGEMENT

The authors are thankful to the Head, Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra for providing the facilities to carry out this work and Dr. Sanjay Swain, Central Instrumentation Facility, Birla Institute of Technology, Mesra Ranchi for his constant help during the analytical work.

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