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Quantitative and risk analysis of heavy metals in selected leafy vegetables

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ABSTRACT

Heavy metals are important environmental pollutants and their accumulation in green edibles poses significant impact on human health, when they cross admissible limits. The present study investigated the levels of five different heavy metals (Cadmium, Chromium, Lead, Nickel and Zinc) in six commonly used leafy vegetables (Bladder dock, Drumstick, Fenugreek, Kenaf, Mint and Spinach) collected in and around Pulivendula town of YSR Kadapa District, Andhra Pradesh. The results revealed that the all the leafy vegetables are contaminated by accumulating heavy metals and the concentrations of Cadmium and Lead has been crossed the safe limit and hence found hazardous to human health in the study area.

Key words: Heavy metals, leafy vegetables, toxicity, human health.

INTRODUCTION

Nutritious food is very important in well being of humanity. Malnutrition is the major problem for much of the humanity in developing world. Regular nutrient rich 'vegetable' food stuff is required to avoid nutrient deficient syndromes. Vegetables take a vital role in the nutrient food stuff as they are rich in carbohydrates, proteins, vitamins, minerals and trace elements. Leafy greens among them occupy the major portion in balanced diet (116 g/day) as they are rich in minerals and vitamins [1]. Random cultivation or unscientific wild collection of leafy vegetables in contaminated habitats is resulting in accumulation of heavy metals which poses a great danger for human health especially in developing countries.

Metal are ubiquitous in the environment and some of them are important in nutrition context, either for their essential nature or because of their toxicity. Dietary exposure to heavy metals like Cadmium (Cd), Lead (Pb), Zink (Zn) and Copper (Cu) may poses a risk to human health through regular vegetable usage [2]. Intake of heavy metal contaminated food has serious impact on reducing some essential nutrients in the body causing intrauterine growth retardation, impaired psycho-social behaviour, disabilities associated with malnutrition and a high prevalence of upper gastrointestinal cancer [3]. Heavy metals are not easily biodegradable and consequently can be accumulated in human vital organs leading to unwanted side effect [4&5].

Heavy metal accumulation in vegetables may be because of deposition of the metals on aerial parts by polluted air, or may be by up taking of water from the contaminated soil as per [6]. The main source of heavy metals is sewage water disposed in the soil which is not only rich source of organic matter and other nutrients but also rich in the elevate the level of heavy metals like Cu, Pb, Cr and Cd [7]. Utilization of waste water for the irrigation takes major part in the accumulation of heavy metals in agriculture soils and plants [8&9]. According to [10] irrigation with

untreated sewage water significantly increases the accumulation of heavy metals in soil and crops; also increases individual metal in soil by 2% to 80% and in crops by 14% to 90% [11].

Among the heavy metals elevated levels of Cadmium causes diseases such as tubular growth, excessive salivation, cancer, gastrointestinal irritation, kidney damage, diarrhoea and vomiting; exceeded levels of Lead can affect bones, teeth, liver, pancreases, and also causes weakness in the wrist and figure and blood diseases [12]. Chromium is a vital metal in all physiological processes in both animals and human beings; Cr (III) is an essential element in glucose, lipid and protein metabolism; Cr (VI) has adverse impact on living organisms due to its quick and easy penetrating capacity and have noxious effect on cells causing various cancers [13], it causes kidney and liver damage, stomach upset and ulcers, skin rashes, lung cancer, respiratory problems, and weakens the immune systems [14]. Zinc is an essential element in regular dietary supplement as it is involved in cellular metabolism, catalytic activity of enzymes, and also required in immune function, protein synthesis and wound healing. Supplement of Zinc reduces the incidence and duration of acute and chronic diarrhoea, acute lower respiratory tract infections in infants and children [15&16]. Uptake of Nickel beyond the permissible levels may cause adverse effects on human being in two stages; the immediate symptoms of headache, vertigo, nausea, vomiting, insomnia, irritability and later symptoms are tightness of the chest, non productive cough, dyspnoea, cyanosis, tachycardia, palpitations, vertigo; sweating and visual disturbances [17].

In the present study, the concentrations of heavy metals in selected cultivated leafy greens have been quantified which are widely used in Pulivendula area of YSR Kadapa district, Andhra Pradesh state, India. It is significant to note that Pulivendula area is located in the Buffer zone-II of Tummalapalle Uranium mining area [18]. The contamination levels were evaluated with respect to the safe limits of different heavy metals prescribed by national and international norms.

MATERIALS AND METHODS

Six leafy vegetables selected for the present study are Kenaf (*Hibiscus cannabinus* L.), Bladder dock (*Rumex vesicarius* L.), Mint (*Mentha spicata* (L.) Hudson), Drumstick (*Moringa oleifera* Lam.), Fenugreek (*Trigonella foenum-graecum* L.) and Spinach (*Spinacia oleracea* L.). The fresh samples of these species were collected randomly from local markets of Pulivendula town for analysis. These leafy vegetables were prioritized owing the higher consumption in the local area and are known potentially nutrient (Table-1). The levels of Cadmium (Cd), Chromium (Cr) and Lead (Pb) in different plant samples were compared with the permissible levels recommended by [19] viz., Cd 0.3 mg/kg, Pb 0.2 mg/kg and Cr 2.3mg/kg; in case of Zinc the upper safe limits is 50 mg/kg [20&21]. Regarding Nickel, the prescribed safe limit is 3 to 7 mg/kg for humans as advocated by ATDSR [22]. Milli Q water was used throughout the experimentation.

Table1: Nutritional constituents present in the sampled leafy greens

S. No	Name of sample	Carbo-hydrates(g)	Energy (K.Cal)	Moisture (g)	Protein (g)	Fat (g)	Mineral (g)	Fibre (g)	Ca (mg)	K (mg)	Fe (mg)
01	KE	10	56	86	2	1	1	-	172	40	1
02	BD	1.4	15	95.2	1.6	0.3	-	0.6	63	17	0.8
03	MN	6	48	85	5	1	2	2	200	62	16
04	DS	12	92	76	7	2	2	1	440	70	1
05	FG	6	49	86	4	1	1	1	39	51	2
06	SP	3	26	92	2	1	2	1	73	21	1

Source: Gopalan et al. (2004) [30].

All chemicals used were of analytical grade. All the glass ware and containers required for experimentation were first soaked in 10% Nitric acid (AR Grade) for few hours and then washed with Milli Q water. Surface contaminants of the leafy samples collected were removed by washing with deionised water twice and then with deionised double distilled water. The leaves were air dried in a clean drying chamber and then at 80°C for overnight in an oven following [23]. The samples were powdered in agate mortar and passed through -80 mesh sieve to get fine powders of the leafy greens. Sampling was done from this powder. The 0.5g of homogenized sample was weighed into a clean 100 ml beaker and 5ml of Nitric acid was added and heated on a hot plate at 80oC until the solution reduced to 2.5 ml and then add 2.5 ml Nitric acid and heated to 80oC until it is reduced to 2.5 ml. Now 2ml of Hydrogen peroxide is added drop by drop and heated at 80oC, until it reduced to 2ml. This solution was filtered and makes up with 100 ml Milli Q water. This was treated as stock and from this stock 5 ml of the solution was taken and make up to 100ml. Blank solutions were prepared using the same procedure described for the samples.

Elemental analysis of both sample and blank solutions were determined using an Inductively Coupled Plasma Mass Spectrometer (ICP-MS) ELAN DRC II, Perkin-Elmer Sciex Instrument [24]. All the results were tabulated with the help of Microsoft Excel software.

RESULTS

Results obtained in the present study are presented in Table-2 and Fig.1-5. The results revealed that Cd and Pb levels of the sampled vegetables have crossed the safe limits according to WHO/FAO standards.

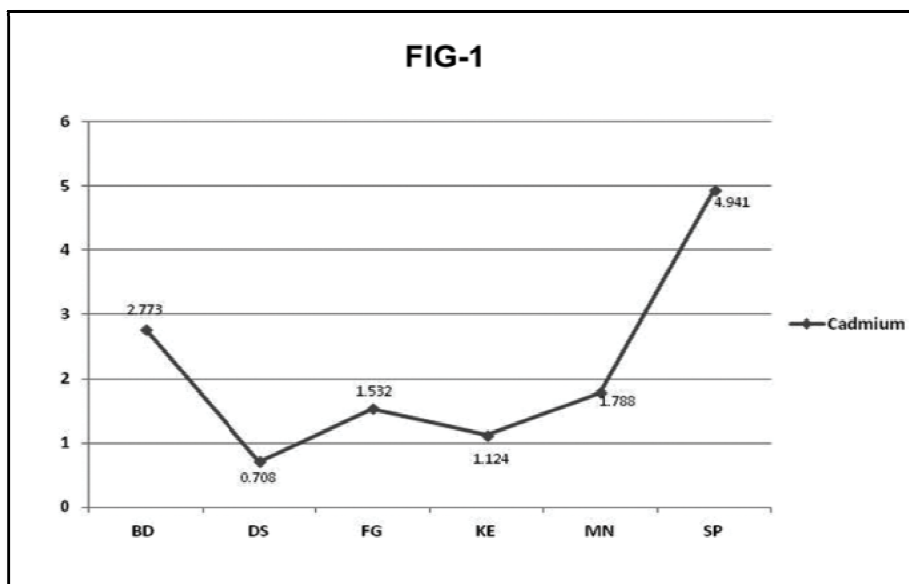
Bladder dock has the heavy metal concentrations which are as follows, Cd (2.773 mg/kg), Pb (0.922 mg/kg) (Table-2; Fig.1&Fig.3), Chromium (1.898), Zinc (12.61) and Nickel at 1.734 mg/kg (Fig.2, Fig-5 & Fig.4). Cd and Pb are present beyond the safe limit and other metals are present within the permissible limits.

Drumstick was found with Cr levels at 1.885 mg/kg (Fig.2), Ni at 1.21 mg/kg (Fig.4) and Zn at 10.029 mg/kg (Fig.5) which are present below the upper safe limits; whereas Cd (0.708 mg/kg) and Pb (0.8 mg/kg) has crossed the safe limits (Table-2, Fig.1& Fig.3). Fenugreek has shown high concentrations of Cd and Pb and was found 1.532 and 1.172 mg/kg (Table-2; Fig.1 &Fig.3) concentrations which are above the standard limits. Cr, Ni and Zn are found within the safe limits (Fig.2, 4 and 5; Table-2).

Kenaf is contaminated by accumulating high concentration of heavy metals. The concentrations of different metals in Kenaf are as follows; Cadmium 1.124 mg/kg (Fig.1), Chromium 1.83 mg/kg (Fig.2), Nickel 1.734 mg/kg (Fig.4), Lead 1.155 mg/kg (Fig.3) and Zinc 12.341 mg/kg (Fig.5) (Table-2). The concentrations of Cadmium (Cd), Lead (Pb) has been crossed the safe limit (0.3, 0.2 mg/kg). The concentration of Chromium, Nickel and Zinc are found within the safe limits (Table-2).

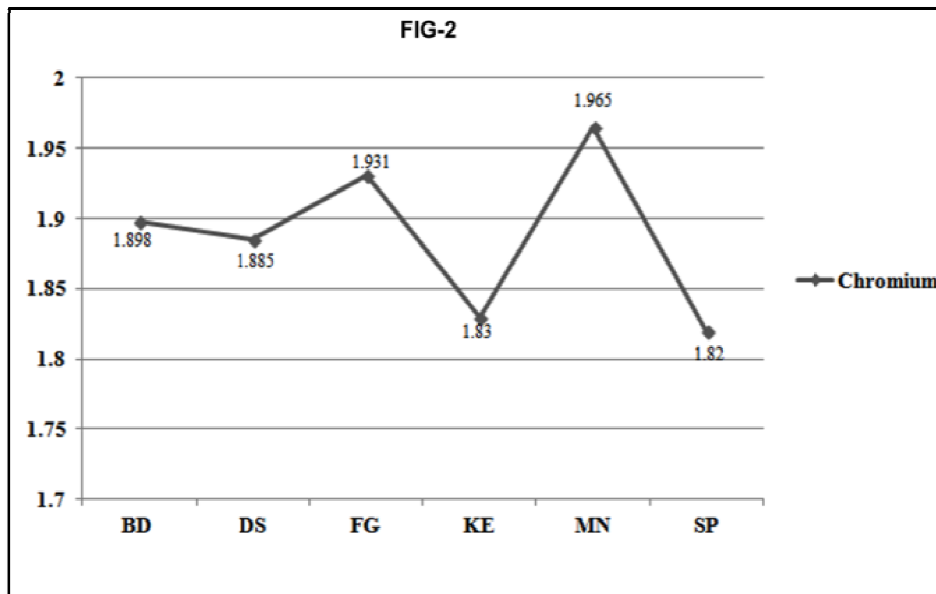
Mint exhibited high accumulation of Cd and Pb above the safe limit and are quantified 1.788 and 0.942 mg/kg respectively (Table-2; Fig.1 & Fig.3). The remaining metals are accumulated below the safe limits; Cr 1.965, Ni 1.69, Zn 12.072 mg /kg (Table-2; Fig.2, 4&5). Spinach is contaminated due to accumulation of heavy metals. The concentration of Cr at 1.82 mg/kg, Ni at 1.374 mg/kg and Zn at 14.524 mg/kg and all are below the safe limits (Fig.2, 4 &5; Table-2); whereas the Cadmium and Lead are determined above the safe limits (Table-2)

Fig.1 Cadmium concentration in the leafy vegetables



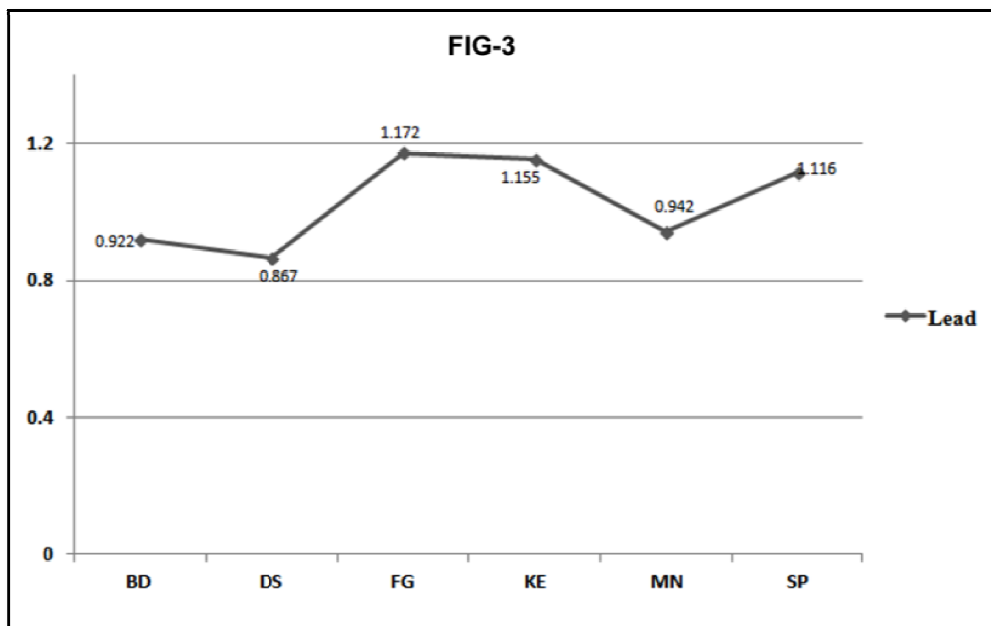
BD- Bladder Dock; DS-Drumstick; FG- Fenugreek KE- Kenaf; MN-Mint; SP-Spinach

Fig.2 Chromium concentration in the leafy vegetables



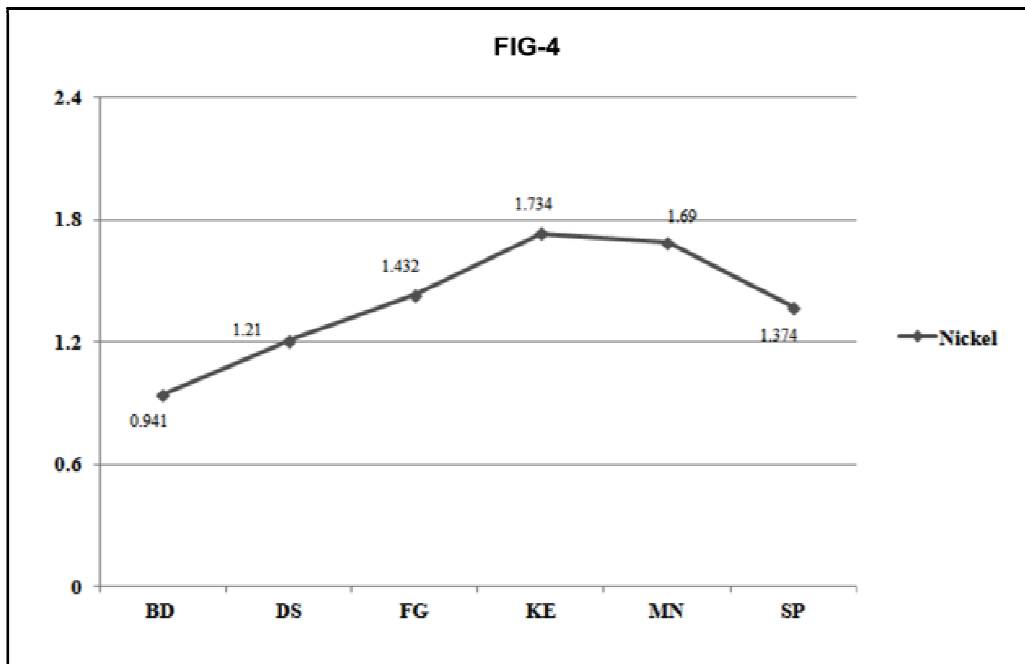
BD- Bladder Dock; DS-Drumstick; FG- Fenugreek KE- Kenaf; MN-Mint; SP-Spinach

Fig.3 Lead concentration in the sampled leafy vegetables



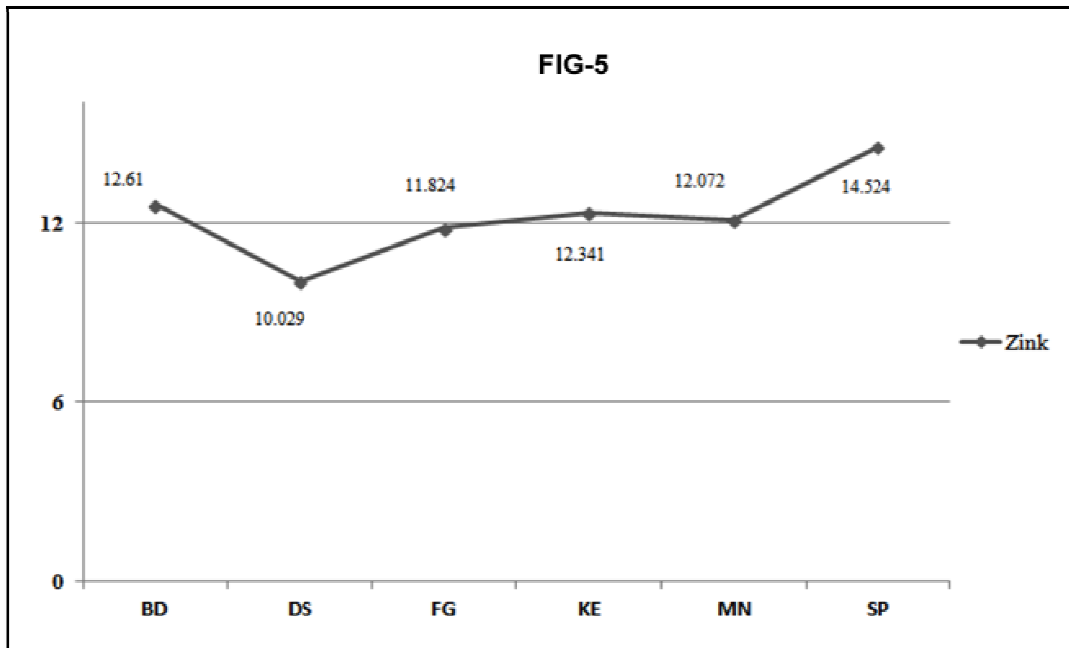
BD- Bladder Dock; DS-Drumstick; FG- Fenugreek KE- Kenaf; MN-Mint; SP-Spinach

Fig.4 Nickel concentration in the sampled leafy vegetables



BD- Bladder Dock; DS-Drumstick; FG- Fenugreek KE- Kenaf; MN-Mint; SP-Spinach

Fig.5 Zinc concentration in the leafy vegetables



BD- Bladder Dock; DS-Drumstick; FG- Fenugreek KE- Kenaf; MN-Mint; SP-Spinach

Table 2: Accumulation of heavy metals in leafy vegetables

S. No	Scientific name	Common & Vernacular name	Family	Cadmium mg/kg	Chromium mg/kg	Nickel mg/kg	Lead mg/kg	Zinc mg/kg
01	<i>Hibiscus cannabinus</i> L.	Kenaf (Gongura)	Malvaceae	1.124	1.83	1.734	1.155	12.341
02	<i>Rumex vesicarius</i> L.	Bladder dock (Chukkakura)	Poloygonaceae	2.773	1.898	0.941	0.922	12.61
03	<i>Mentha spicata</i> (L.) Hudson	Mint(Pudina)	Lamiaceae	1.788	1.965	1.69	0.942	12.072
04	<i>Moringa oleifera</i> Lam.	Drumstick (Munagaku)	Moringaceae	0.708	1.885	1.21	0.867	10.029
05	<i>Trigonella foenum-graecum</i> L.	Fenugreek (Menthikura)	Fabaceae	1.532	1.931	1.432	1.172	11.824
06	<i>Spinacia oleracea</i> L.	Spinach (Palakura)	Chenopodiaceae	4.941	1.82	1.374	1.116	14.524

DISCUSSION

Results revealed heavy metal accumulation in all the sampled leafy greens. In all the samples Cd and Pb are present beyond the safe limits. Maximum concentration of Cd is found in Spinach with 4.941 mg/kg followed by Bladder dock with 2.773 mg/kg (Table-2), drumstick leaves has lesser amount of Cd than the other leafy vegetables. It is found that the lowest concentration among all the heavy metals accumulated in all the leafy vegetables of the study was found with Cd. In Spinach, 4.62 mg/kg of Cd was quantified by [9] from Varanasi area. This result is almost same to the results obtained in the present study. Actual safe limit of this heavy metal is 0.3 mg/kg according WHO/FAO standards, but in all sampled leafy vegetables the concentration is found beyond the safe limits. According to [25] even low level of Cd is also toxic and may cause health disorders. In all the samples of the present study has exhibited higher concentrations Lead. Fenugreek has the highest concentration with 1.172 mg/kg followed by Kenaf 1.155 mg/kg and the least concentration was found in Drumstick leaves with 0.867 mg/kg (Table-2; Fig.3.).

In the present study, Cr is found with higher amounts in Mint (1.965 mg/kg) followed by Fenugreek (1.931 mg/kg). Spinach shows the lowest Cr accumulation with 1.82 mg/kg (Table-2 & Fig.2). Nickel concentration is found maximum in Kenaf with 1.73 mg/kg followed by Mint with 1.69 mg/kg and the minimum concentration in Bladder dock with 0.941 mg/kg (Table-2 & Fig.4). Ni, in a range of 0.2- 3.0 mg g⁻¹ in Palak (Spinach) from waste water irrigated areas of Hyderabad was reported by [26]. The prescribed safe limit of Ni is 3-7 mg/kg to humanity [22]. All the leafy vegetables of the present study exhibits Nickel accumulation below the upper safe limit.

Pertaining to Zinc, the safe limit is 50mg/kg (PFA standard) according to [21]. In the present investigation it was found that it is present in maximum concentration than the other heavy metals in all sampled leafy vegetables and is under the safe limit. Spinach is found to have more Zn accumulation (14.524 mg/kg) followed by Bladder dock (12.61 mg/kg) and very low concentration in Drumstick with (10.029 mg/kg) (Table-2 & Fig.5). Highest concentration of Zn in all the vegetables irrigated with waste water in Varanasi area was reported [9]. Among all the metals Zn is least toxic and has positive correlation than other inventoried heavy metals. According to [27&28] about 20% of the world's population could be at risk of Zn deficiency and the average daily intake of Zn; in case of men, 15 mg/day and for women 12 mg/day [29]. In the present study the average concentration of Zn in leafy greens is 10-14.5 mg/kg and thus correlates with average intake value of Zinc.

CONCLUSION

Present study revealed heavy metal accumulation in all the sampled leafy greens used by the people of Pulivendula and it is found that all the samples have Cd and Pb beyond the safe limits. It was observed that many factors such as inputs to the soil and water through over use of fertilizers, pesticides and usage of sewage waters as well industrial effluents mixed waters used for irrigation are resulting in high levels of heavy metal accumulation in leafy greens. As the study area is located in the buffer zone-II of Tummalapalle Uranium Mining area; the risk of accumulation of heavy metals in the leafy vegetables may reach highest levels due to contamination with the industrial waste generated from the mining activities in future.

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