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Heavy Metal Accumulation in Soybeans Cultivated in Iran, 2015-2016

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ABSTRACT

Background: Due to environmental contamination in recent years, contamination of food chain by heavy metals is not far-fetched. The purpose of this study is to determine heavy metals in soybeans cultivated in Iran to monitor the food chemical contaminants. **Methods**: To assess metal contamination, four varieties (Sahar, Katool, Williams, and M_7) of soybean cultivated in Iran, were collected. Metal concentration of samples was analyzed by inductively coupled plasma-optical emission spectrophotometer (ICP-OES). **Results**: The concentrations of Arsenic (As), Cadmium (Cd), Copper (Cu), Nickel (Ni), Lead (Pb), and Zinc (Zn) in soybean samples were ranged from 0.008 to 0.21, 0.008 to 0.16, 9.51 to 87.71, 4.08 to 22.37, 0.015 to 1.18, and 35.53 to 65.02 mg/kg, respectively. Taken together, findings of this study showed that heavy metal content of all taken samples, except Pb in M_7 variety, were below the maximum limits. So, there is no need to concern about the presence of heavy metal contents in soybeans cultivated in Iran. **Conclusion**: Since soybeans are used in production of other soy-based products (such as soy milk, soy cheese, soy sauce), regular monitoring of soybean in terms of heavy metals is necessary.

Key words: *Soybean; Heavy metals; ICP-OES.*

Introduction

In recent years, excessive use of fertilizers, sewage sludge, and pesticides in agriculture (Shute and Macfie, 2006), environmental pollution (in air, water, and soil) (Corguinha *et al.*, 2015), industrialization, and urbanization (Gill, 2014) have drawn public attention to contamination of food chain by heavy metals (Salazar *et al.*, 2012). Metallic elements which have relatively high density and are poisonous at low concentrations are called heavy metals (Gill, 2014). Agricultural crops can uptake these elements and accumulate them in their edible parts (Corguinha *et al.*, 2012). Accumulation and translocation of these elements in edible and harvested parts of plants depend on soil, climate, plant genotypes, and agronomic management (McLaughlin *et al.*, 1999). Consumption of cereals is considered as the most important daily intake of non-essential trace elements by human (Corguinha *et al.*, 2015). Consumption of crops containing heavy metals can

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put human health into danger and cause severe toxicity(Corguinha *et al.*, 2012). Soybean (Glycine max) has high protein content with beneficial effects on health and nutrition. It lowers plasma cholesterol, prevents from cancer, diabetes, osteoporosis, obesity, and is importance in food industry (soy milk, soy cheese, soy sauce) (Li *et al.*, 2013). According to Bojinova's report, beans such as soybeans can accumulate heavy metals more than others (Bojinova *et al.*, 1994). Therefore, it is necessary to determine the presence and accumulation rate of heavy metals by such crops (Reddy and Dunn, 1984).

Due to lack of information about heavy metal contents of soybeans cultivated in Iran, concentrations of Arsenic (As), Cadmium (Cd), Copper (Cu), Nickel (Ni), Lead (Pb), and Zinc (Zn) in soybeans were investigated and then compared with the standard limits. So, four varieties of soybeans cultivated in Iran were analyzed to calculate the metal accumulation.

Materials and Methods

Sampling: Four varieties of soybean (Glycine max L.) samples were collected from research center of Iran, then the samples were cleaned by hand so that any additional physical pollutions such as stones were picked by hand. In order to remove any particles, samples were washed and rinsed three times with distilled water. Dried samples were ground using a stainless steel grinder for heavy metal analysis (Zhung *et al.*, 2013).

Sample analysis: To decrease the risk of contamination, all glass wares were soaked in diluted nitric acid and rinsed with distilled water before use. One gram of dried and ground bean was digested with HNO_3 (Merck, Germany, 65%), HCl (Merck, Germany, 37%), and H_2O_2 (Merck, Germany, 30%) in a 5:2:1 ratio, the mixtures were then put on the hotplate (50-60 ^{0}C) until a transparent solution was obtained. The sample solution was filtered through 0.45 µm membrane filter and then it was filled to exact volume of 25ml with distilled water. All chemical reagents were of analytical grade including the standard stock solutions of different heavy metals used for the preparation of working calibration solutions. All analyses were done in triplicate. Metal contents of final solution were determined by inductively coupled plasma-optical emission spectrophotometer (SPECTRO GENESIS model). Heavy metal concentrations of soybean were calculated on the basis of dry weight. The details of the instrumental operating conditions are listed in **Table 1**.

Data analysis: Statistical Analysis Software (SAS) version 9.1 was used for the statistical analyses. The treatment effects were determined by a one way ANOVA followed by LSD test to investigate the statistically significant differences at P-value of < 0.05.

Results

For As, the observed ranges in soybean grain samples were from 0.008 to 0.21 mg/kg. The As concentration of Katool was higher than those of the other three samples. The As level in Sahar was similar to the average arsenic concentration of M_7 variety. As recorded in **Table 2**, the maximum (22.37 mg/kg) and minimum (4.08mg/kg) Ni content of soybean was observed in the Williams and Katool varieties, respectively. The mean concentration of Cu in seeds of Williams was 9 times higher than that of Katool. The Cu concentration in Katool was evaluated as the lowest among all varieties. Therefore, significant differences in soybean Cu contents were found among four varieties. In the current study, significant differences in Zn content of seeds were reported among soybean varieties. Zn level in all samples vary from 35.53 mg/kg (Katool) to 65.02 mg/kg (Williams) (**Table 2**). Among the four soybean varieties investigated in the present study, Pb content of M_7 variety (1.18 mg/kg) was estimated higher than those of others. However, the Pb concentration of Katool and Williams were approximately similar. The Cd contents in the soybean were significantly different among different varieties, ranging from 0.008 to 0.16 mg/kg DW. Means followed by the different letter in each row are significantly different at the 95% level.

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Table 1. ICP-OES instrumental operating conditions

Parameters	Characteristics
RF generator power (W)	1400
Frequency of RF generator (MHz)	27.12
Plasma gas flow rate (l/min)	13
Auxiliary gas flow rate (l/min)	0.8
Nebulization gas flow rate (l/min)	0.9
Optic temperature	29.65
Type of Nebulizer	Modified LichteSpectro
Type of spray chamber	Cyclonic
Elements (λ/nm)	Zn:213.856; Pb:220.353; Cd:214.438; As:228.812; Cu:327.394; Ni:221.648;

As: Arsenic, Cd: Cadmium, Cu: Copper, Ni: Nickel, Pb: Lead (Pb), Zn: Zinc

Table 2. Measured heavy metal concentration in soy bean samples (mg/kg DW)

Soybean strain	Sahar (Mean ± SD)	Williams (Mean ± SD)	Katool (Mean ± SD)	M_7 (Mean ± SD)
As	0.008 ± 0.002[c]	0.021 ± 0.01[b]	0.21 ± 0.09[a]	0.008 ± 0.003[c]
Cd	0.15 ± 0.05[b]	0.12 ± 0.02[c]	0.008 ± 0.002[d]	0.16 ± 0.07[a]
Cu	81.53 ± 14.8[b]	87.71 ± 12.93[a]	9.51 ± 3.84[d]	20.62 ± 6.52[c]
Ni	10.79 ± 4.87[c]	22.37 ± 6.24[a]	4.08 ± 1.58[d]	11.44 ± 4.98[b]
Pb	0.081 ± 0.08[b]	0.015 ± 0.005[c]	0.015 ± 0.008[c]	1.18 ± 0.4[a]
Zn	64.79 ± 9.25[b]	65.02 ± 10.91[a]	35.53 ± 6.22[d]	41.91 ± 8.98[c]

The different letters in each row are significant at the $p \leq 0.05$ level. As: Arsenic, Cd: Cadmium, Cu: Copper, Ni: Nickel, Pb: Lead (Pb), Zn: Zinc

Discussion

Heavy metals are found in most soils which can damage human health. Accumulation of metals in soybean is a great concern because this can raise the possibility of metal entrance into food chain. In the present study, the metal concentrations (Cd, As, Cu, Ni, Pb, and Zn) in soybean crops grown in Iran were investigated. Several studies indicated the accumulation of heavy metals in soybean plants. However, to the best of our knowledge, there has been no comprehensive study on the level of heavy metals in soybeans cultivated in Iran. As it is shown in **Table 2**, significant differences are observed among the studied four soybean samples in accumulation of different heavy metals. According to other studies (Arao *et al.*, 2003, Zhao *et al.*, 2014), these differences can be attributed to genetic differences and metal content of the soil.

As is a metalloid that disperses into soil and water through human activities (smelting, mining, and agricultural practices) and natural occurrences (disintegration of rocks). According to **Table 2**, As levels of all soybean samples in the present study were below the maximum allowable level (0.5 mg/kg) (Zhao *et al.*, 2014). But, As concentration of Katool Variety was significantly higher than others. The present results are not compatible with those reported in a study conducted in southern China by Zhao, who reported 0.53 to 3.83 mg/kg concentration of As in soybean (Zhao *et al.*, 2014). These differences depend on several factors such as the As concentration in soils, soy bean cultivar, and country of origin (Peralta-Videa *et al.*, 2009).

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The major source of Cd pollution is fertilizers produced from phosphate ores (Peralta-Videa *et al.*, 2009). The average Cd concentration was reported as 200 µg/kg DW (0.2 mg/kg) by Codex Alimentarius Commission (Corguinha *et al.*, 2012), taking into account none of the samples in this study exceeded this maximum concentration. The results showed that Cd content in M_7 variety (0.16) was significantly higher than the others. Accumulation of Cd in seed tissues depends on transfer mechanisms in root tissue that allow retention of the greatest portion of Cd. Furthermore, according to Sugiyama study, soy bean cultivars are different in accumulation of Cd (Sugiyama *et al.*, 2007). This fact is compatible with the present findings. Cd concentration in the current study was lower than those of Salazar (0.2 mg/kg), Zhou (2.95 mg/kg), Zhung (0.23 mg/kg), Zhao (2.16 mg/kg), and Liu (0.24 mg/kg) studies (Liu *et al.*, 2005, Salazar *et al.*, 2012, Zhao *et al.*, 2014, Zhou *et al.*, 2013, Zhung *et al.*, 2013). This can be attributed to differences in the ability of cadmium accumulation in Iranian varieties and also less contamination of soils by metals. The average Cd concentration in Corguinha (Corguinha *et al.*, 2012) study was estimated as 23 µg/kg, which was lower than the maximum level in the present study.

Copper is a cofactor of certain enzymes or a component of other proteins but its excess amount can be toxic to human (Gonçalves Jr *et al.*, 2014). In the current study, Cu concentration ranged from 9.51 mg/kg (Katool variety) to 87.71 mg/kg (Williams variety), which were higher than those reported by Zhung (23.10 mg/kg) and Liu (17.77 mg/kg)(Liu *et al.*, 2005, Zhung *et al.*, 2013). According to the research conducted by Reddy, in comparison to other soybean tissues, seed accumulates greater amount of Cu concentration (Reddy *et al.*, 1989).

Williams' variety had significantly higher Ni concentration in comparison to others. The maximum level of Ni concentration in Zhung research (11.90 mg/kg) (Zhung *et al.*, 2013) was estimated lower than that of Williams' variety.

Lead contamination of agricultural soils occurs by combustion of fuels containing Pb in areas close to urban centers and by addition of Pb fertilizers to soils (McLaughlin *et al.*, 1999). It was found that concentration of Pb in M_7 variety exceeded the allowed maximum levels (0.2 mg/ kg). In contrast, other soybean grain samples had a low Pb accumulation. This fact could be attributed to genetic differences among grains of soybean varieties. Lead concentration in M_7 variety was higher than those reported by Lavado et al. (0.85 mg/kg), Zhung et al. (0.34 mg/kg), and Liu et al. (0.2 mg/kg), (Lavado *et al.*, 2001, Liu *et al.*, 2005, Zhung *et al.*, 2013).

The Zn concentration (41.91 to 65.02 mg/kg) in all samples was below the allowable maximum levels of contaminants in food (100 mg/kg)(Zhao *et al.*, 2014). The maximum concentration of Zn was reported in Williams (65.02 mg/kg) which was lower than the maximum levels of Zn in Zhou et al. (76.1 mg/kg), Zhung et al. (85.40 mg/ kg), Zhao et al. (88 mg/ kg), and Liu et al. (70.35 mg/kg) studies, and higher than the one reported by Salazar et al. (38.36 mg/kg) (Liu *et al.*, 2005, Salazar *et al.*, 2012, Zhao *et al.*, 2014, Zhou *et al.*, 2013, Zhung *et al.*, 2013). According to the previous studies, uptake and translocation of Cd and Zn in plants are related to each other. In general, plant nutritional status can affect metal uptake and translocation. In other words, addition of Cd to soils with Zn-deficiency can accumulate Cd in plant (Shute and Macfie, 2006).

Conclusions

According to the achieved results, except for Pb content of M_7 variety and copper in some varieties, soybeans produced in Iran are not polluted by heavy metals. This fact shows that soils in which the soybeans were cultivated had low accumulation of heavy metals because if there was a metal contamination, these metals were transferred into plants (Zhou *et al.*, 2013). However, since this crop is used in production of soy milk, cheese, yoghurt, and other products, heavy metals can condense and

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increase in it, which can pose human beings to serious potential health risks. Therefore, regular monitoring of agricultural products in terms of heavy metals is necessary.

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Authors' contributions

Moshtaghi Boroujeni, H and Khalili Sadrabad, E designed the study. Khalili Sadrabad, E and Heydari, A collected the samples and carried out the experiments. All authors read and approved the final manuscript.

Conflict of interest

There is no conflict of interest.

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