

## EPIDEMIOLOGICAL ASSESSMENT OF PER CAPITA ABSORPTION OF HEAVY METALS IN FOUR NATIVE SPECIES OF RICE, YASUJ, IRAN- 2016

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### ABSTRACT

**Aim:** Rice with scientific name of *Oryza sativa* is one of the most important cereals and food items in the world that is the world's second most important cereal after wheat. It has been estimated that in 2010, about 603 million tons of rice were produced around the world, that most of which (about 583 million tons) was cultivated in Asia. Most studies have mentioned the most common cases of rice contamination with heavy metals to be of infection with elements arsenic, copper, cadmium, mercury and lead. Although several studies have been conducted and reported in other areas of the country on native species of each region, but no report has been found about the native species of Kohgiluyeh and Boyer-Ahmad province, therefore researchers have designed an independent study on the measurement of heavy metals pollutants in local varieties of consumed rice in order to fill existing information vacuum.

**Material and Method:** In order to run tests, samples of rice were prepared from two species as 5g, and then they were burned in a temperature of 450°C for 4 to 5 hours and after adding 0.5cc concentrated nitric acid, the samples were dried. After this time, then a solution of 7: 1000 nitric acid was prepared. Samples were entered into atomic absorption for measurement of heavy metals. VARIAN atomic absorption spectrophotometer was used with AA240FS model which had been made in America with flame system accuracy of ppm. In order to estimate the average weekly absorption of studied consumed rice, element daily intake (EDI) using the following equation and the results were used to calculate weekly and monthly per capita consumption. Weekly absorption of the heavy metals including cadmium, lead and arsenic was calculated by multiplying the amount of these metals in daily and weekly rice consumption amount. The calculated mean values were compared PTWI (Provisional Tolerable weekly intake : PTWI) and EDI index was evaluated. PTWI is the reference value which has been determined by the Jib committee of experts of the UN FAO and the WHO on food additives (JECFA).

**Results:** The concentration of cadmium, lead and chromium lower than the national and international standards but this did not conform to results and the reason may be how to plant and water pollution and soil of rice fields are the other factors. The average concentration of 290 micrograms for cadmium accumulated during a week was 290 µg/kg for Champa, 156.4 µg/kg for cadmium species, 102.8 for Shamim, and for cadmium it was 80.4 µg/kg for rice species of Gerdeh. The average cumulative concentration of lead per week was obtained 33.2 µg/kg for Champa, 27.74 µg/kg for Shamim, for lead 13.02 µg/kg for Fajr, and 25.83 µg/kg for Gerdeh species. This is due to the cumulative effects of heavy metals and adverse effects on the human population. As well as a significant threat to food security in particular.

**Discussion:** The comparison of the level of exposure to the estimated densities of heavy metals in metals in different species PTWI levels indicates that the level of exposure for all of the metals mentioned is lower than the standard level. Though it indicates an appropriate situation about the cases investigated, it is necessary to do a constant and exact examination of such foods. It shows the necessity of making a proper program to recognize the possible sources of contamination and to make the contaminations less dense in the local kinds of rice.

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### Introduction

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Rice with scientific name of *oryza sativa* is one of the most important cereals and food items in the world that is the world's second most important cereal after wheat [1]. "Ten thousands types of rice" in the world make half the main dish of people [2]. It has been estimated that in 2010, about 603 million tons of rice were produced around the world, that most of which (about 583 million tons) was cultivated in Asia [3]. Rice as a worldwide product is also one of the major sources of carbohydrates in the diet of Asian nations including Iranians. Per capita consumption of rice in Iran is estimated as 42.2 kg, therefore the average daily consumption of rice in the country is estimated at 116.5 grams. Dependence of rice cultivation on rich water resources and higher local demand than production beside the recent drought, have caused that Iran has been considered one of the main importers of this product [4].

The probability of contamination of consumed rice at all stages of cultivation, harvest, processing, packing, and offering until use, has always been a concern for exporters and consumers. Among all the above steps, contamination in the cultivation and harvest stages is more important than other stages due to the influence of contaminants in the internal structure of the product. Extreme weather and climate changes in recent decades have reduced water consumption in the industrial and agriculture sectors in many countries, especially in the Middle East. The lack of coherent national and regional programs in developing countries for achieving better efficiency from existing water resources has led to tendency of people and farmers to benefit from non-specialized approaches and also the use of waste of industrial plants for irrigation of rice fields. Increasing global demand due to population growth along with the decrease in production due to regional dehydration crises contributes to the problem and has strengthened the possibility of using industrial waste on farms. This has led researchers to investigate the contamination of consumed rice which is produced interior as one of their priorities in research.

Among the possible multiple contaminants, since wastewater often contains heavy metals, then valuable sources of foods such as rice may be contaminated with heavy metals and these substances enter the food chain of consumers, so that it has been shown in some reports. In some reports, closeness of rice farms to the industrial centers and contamination of water and soil with wastewater without clearing system has been known as the cause for accumulation of heavy metals, especially Lead, Cadmium, and Arsenic [5-7], [8-10]. The researchers believe that absorption of heavy metals from contaminated lands by plants and especially by agricultural products is one of the most important ways for entering these elements into the food chain [11].

The use of heavy metals can lead to chronic complications arising from the accumulation of other resources and sometimes cause acute poisoning. Heavy metal poisoning such as lead and cadmium, and copper can lead to complications such as neurological disorders, cancer, nutrient deficiency, hormone imbalance, miscarriage, respiratory disorders, cardiovascular disease, damage to the liver, kidneys, brain, allergies, anorexia, premature aging, memory loss, hair loss, osteoporosis, insomnia, weakened immune system, anemia, genetic damage and even death [12, 13].

The most damaging effect of heavy metals is due to increase in the oxidation of free radicals. Free radical is a free and energetic molecule that has an unpaired electron and absorbs another electron from other molecules to reach equilibrium and that can be followed by the possibility of entering multiple biological reactions resulting in the disruption of these mechanisms [14, 15]. Most studies have mentioned the most common cases of rice contamination with heavy metals to of infection with elements arsenic, copper, cadmium, mercury and lead. Among the main agricultural products, rice is a particular product with high absorption and accumulation of heavy metals including cadmium, lead and arsenic [16]. Since rice is considered as a staple in various countries and regions and evaluation of the amount of heavy metals is a priority for standard and monitoring programs of the country, in the case of imported rice, it is expected that the necessary evaluations be carried out by the government and relevant institutions, but in the case of interior produced rice and also due to a variety of sources, regional-scale production and local consumption appears to have no regular monitoring processes on such products by national and international standards. Therefore, it seems logical and necessary to the health of such widely used products, and especially rice be assessed for the presence of toxic elements such as heavy metals by researchers and under the periodic projects and independent and foreign assessments of organizations directly involved in monitoring and evaluation. This issue is more important about the local production, especially in areas that have been centers of local culture, and on the other hand taste of people also toward the local products is higher than similar imported products with the foreign origin. According to epidemiological approaches, evaluation of exposure can be considered in programming in order to make the necessary sensitivity in human populations. Meanwhile, Kohgiluyeh and Boyer-Ahmad Province is also one of the provinces has areas of cultivation of rice with known local species including Gerdeh, Shamim, and Champa which have enthusiasts due to its special geographical position and benefiting from the numerous rivers and water resources, especially in semi-cold regions that is because this provinces of country on the southern edge of the Zagros Mountains. Although several studies have been

conducted and reported in other areas of the country on native species of each region, but no report has been found about the native species of Kohgiluyeh and Boyer-Ahmad province, therefore researchers have designed an independent study on the measurement of heavy metals pollutants in local varieties of consumed rice in order to fill existing information vacuum.

#### **Materials and Methods:**

In the present study, four types of common consumed rice in the province including Champa, Shamim, Fajr, and Gerdeh species were used to measure the concentrations of heavy metals including lead, cadmium and copper on the country based standard approaches. Then the researchers estimated the average exposure concentration to these elements based on their initial findings.

In order to run tests, samples of rice were prepared from two species as 5g, and then they were burned in a Chinese plant and furnace at a temperature of 450°C for 4 to 5 hours. The samples are cooled and after adding 0.5cc concentrated nitric acid, the samples were dried. For removing acid from the sample containing plant, plant was placed in an oven at 100°C, and after ensuring the withdrawal of acid for 1 to 2 hours; the plant was placed in the furnace at a temperature of 450°C. After this time, the plant was exited and was allowed to cool (sample preparation). Then a solution of 7: 1000 nitric acid was prepared. In order to prepare nitric acid, 7cc concentrated nitric acid was poured into the balloon and was brought to volume in 1000 using deionized water. The prepared tower sample was moved from the plant into the 10cc balloon using the funnel and filter paper and was brought to volume using nitric acid 7: 1000 production (based Institute of Standards and Industrial Research of Iran). Samples were entered into atomic absorption for measurement of heavy metals. VARIAN atomic absorption spectrophotometer was used with AA240FS model which had been made in America with flame system accuracy of ppm (part per million) and furnace system with accuracy of ppb (Part per billion). Researchers in this research used flame system, compliance with standards outlined in the brochure guide to measure heavy metals.

In order to estimate the average weekly absorption of studied consumed rice, element daily intake (EDI) was calculated based on the average daily consumption of rice in Asian countries (equivalent to 165 grams per day for a 60 kg person) using the following equation and the results were used to calculate weekly and monthly per capita consumption (Zhong et al, 2006). Weekly absorption of the heavy metals including cadmium, lead and arsenic was calculated by multiplying the amount of these metals in daily and weekly rice consumption amount. This relationship represents the following formula:

$DI = (\text{Concentration} \times \text{Consumption}) \div \text{Body weight}$

DI :symbol studied daily absorption of heavy metals: cadmium, lead, arsenic

Symbol C indicates the concentration of heavy metals in consumed rice as microgram per kg.

Consumption parameter is the symbol of the average daily consumption of rice in the study area (130 grams per day)

BW parameter represents the median estimations for a person weighing 60 in this study.

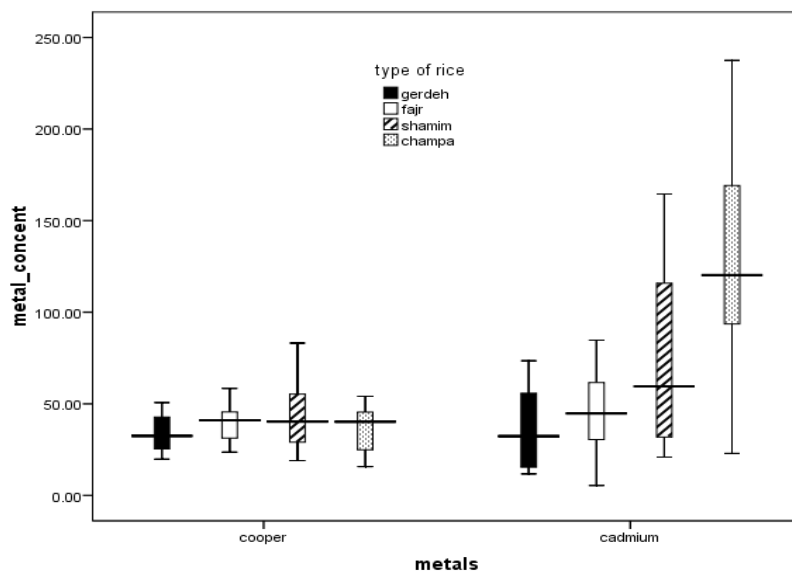
The calculated mean values were compared PTWI(Provisional Tolerable weekly intake : PTWI) and EDI index was evaluated. PTWI (indicator represents the tolerable weekly intake, and is the reference value which has been determined by the Jib committee of experts of the UN Food and Agriculture Organization and the World Health Organization on food additives (JECFA) and represents a safe weekly intake of pollutants. The PTWI is determined based on the weekly intake of pollutants such as heavy metals, which shouldn't be cumulated lifetime in the human body [17].

#### **Results:**

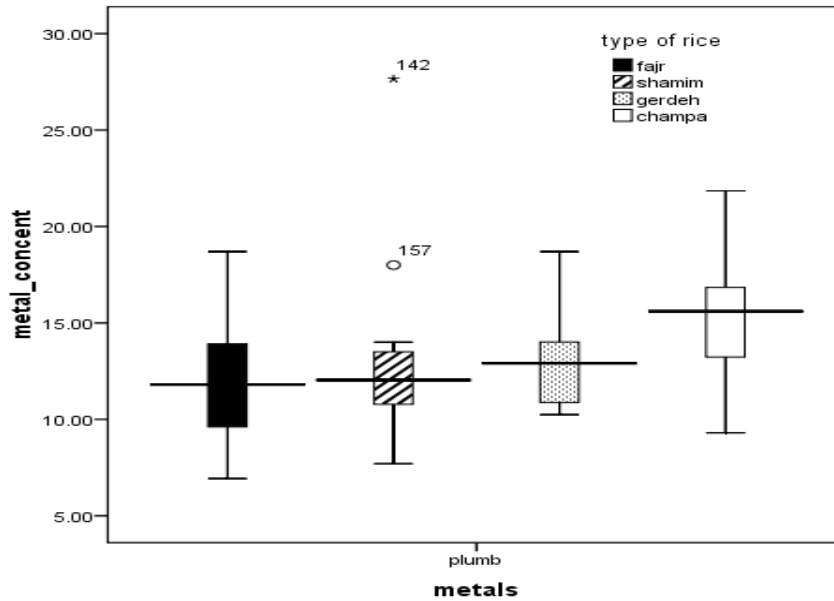
Table A shows the distribution of mean and standard deviation of heavy metals in different studied rice varieties.

**Table 1:** The mean and 95% confidence interval of heavy metals concentration (ppm) by type of rice

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
copper (ppm)	champa	15	36.4053	11.98881	3.09550	29.7662	43.0445	15.70	54.08
	shamim	16	44.6200	19.19484	4.79871	34.3918	54.8482	19.00	83.16
	gerdeh	16	33.9112	10.52746	2.63186	28.3016	39.5209	19.80	50.71
	fajr	16	39.9544	9.86715	2.46679	34.6965	45.2122	23.60	58.38
	Total	63	38.7595	13.73322	1.73022	35.3009	42.2182	15.70	83.16
cadmium (ppb)	champa	15	133.93	62.96685	16.25797	99.0568	168.7965	22.90	237.50
	shamim	16	72.1597	46.19736	11.54934	47.5429	96.7765	20.90	164.50
	gerdeh	16	37.0762	21.19222	5.29805	25.7837	48.3688	11.75	73.50
	fajr	16	47.4400	21.75844	5.43961	35.8457	59.0343	5.42	84.75
	Total	63	71.6780	55.07208	6.93843	57.8083	85.5477	5.42	237.50
plumb (ppb)	champa	15	15.2367	3.23115	.83428	13.4473	17.0260	9.30	21.85
	shamim	16	12.7656	4.75470	1.18867	10.2320	15.2992	7.70	27.65
	gerdeh	16	13.0256	2.37311	.59328	11.7611	14.2902	10.24	18.70
	fajr	16	11.9200	3.03379	.75845	10.3034	13.5366	6.93	18.70
	Total	63	13.2052	3.59153	.45249	12.3007	14.1098	6.93	27.65



**Figure 1:** The box plot of metal concentration (Cooper and Cadmium) by type of rice



**Figure 2:** The box plot of plumb concentration (ppm) by type of rice

**Table 2:** The result of one sample t test and analysis of variance analysis for compare of mean concentration of heavy metal by type of rice

type of rice	Heavy metal	N	Standard value	One sample T test	P value	F test	Sig.	Post hoc significant pairs
CHAMPA	cadmium (ppb)	15	48	5.285	.000	16.819	.000	CHAMPA : SHAMIM
SHAMIM	cadmium (ppb)	16	48	2.092	.054			CHAMPA : GERDEH
GERDEH	cadmium (ppb)	16	48	-2.062	.057			CHAMPA :FAJR
FAJR	cadmium (ppb)	16	48	-.103	.919			
CHAMPA	plumb (ppb)	15	120	-125.574	.000	2.555	.064	FAJR : CHAMPA 0.06
SHAMIM	plumb (ppb)	16	120	-90.213	.000			Ns
GERDEH	plumb (ppb)	16	120	-180.311	.000			Ns
FAJR	plumb (ppb)	16	120	-142.502	.000			Ns
CHAMPA	copper (ppm)	15	20	5.300	.000	1.903	.139	Ns
SHAMIM	copper (ppm)	16	20	5.131	.000			Ns
GERDEH	copper (ppm)	16	20	5.286	.000			Ns
FAJR	copper (ppm)	16	20	8.089	.000			Ns

**Table 3:** The DI and WI calculation for mean body weight 60 kg and mean consumption of 116.5 per day

Heavy metals	Type of rice	Concentration (PPM)	DI	WI	PTDI(17)	PTWI
Cadmium (PPb)	CHAMPA	133.93	0.00026005	0.0001820	0.001 mg/kg	0.007
	SHAMIM	72.16	0.00014011	0.000980	0.001 mg/kg	0.007
	FAJR	37.08	0.00007199	0.000503	0.001 mg/kg	0.007
	GERDEH	47.44	0.00009211	0.000644	0.001 mg/kg	0.007
Cooper (PPM)	CHAMPA	36.41	0.0007069	0.000494	0.003 µg/kg	0.021
	SHAMIM	44.62	0.0008664	0.000606	0.003 µg/kg	0.021
	FAJR	33.91	0.0006584	0.000460	0.003 µg/kg	0.021
	GERDEH	39.95	0.0007757	0.000542	0.003 µg/kg	0.025
Plumb (PPb)	CHAMPA	15.24	0.00002959	0.00207	0.0036 mg/kg	0.025
	SHAMIM	12.77	0.00002479	0.00173	0.0036 mg/kg	0.025
	FAJR	13.026	0.00002529	0.00177	0.0036 mg/kg	0.025
	GERDEH	11.92	0.00002314	0.00161	0.0036 mg/kg	0.025

**Table 4:** The decision making matric for ranking of type of rice by heavy metal level

Type of heavy metal	Type of rice			
	CHMPA	SHAMIM	FAJR	GERDEH
CADMIUM	d=1	c=2	b=3	a=4
PLUMB	d=1	b=3	a=4	c=2
COOPER	b=3	d=1	c=2	a=4
Final Ranking	D=5	C=6	B=9	A=10

*a:best , b:acceptable, c:poor and d:worse rank*

**Table 5:** The matrices of correlation coefficient between heavy metals by type of rice

Type of rice	Metal name	R. square	Assessment	R. square	Type of association
		Cubic		Linear	
CHAMPA	Copper-cadmium	0.667	Mild association	0.563	Direct
	Copper-plumb	0.162	Pure association	0.063	Direct
	Plumb-Cadmium	0.516	Mild association	0.140	Direct
SHAMIM	Copper-cadmium	0.280	Pure association	0.138	Direct
	Copper-plumb	0.115	Pure association	0.009	Direct
	Plumb-Cadmium	0.120	Pure association	0.076	Direct
FAJR	Copper-cadmium	0.074	Pure association	0.045	Direct
	Copper-plumb	0.330	Pure association	0.0005	Direct
	Plumb-Cadmium	0.390	Pure association	0.353	Indirect
GERDEH	Copper-cadmium	0.225	Pure association	0.062	Indirect
	Copper-plumb	0.185	Pure association	0.112	Indirect
	Plumb-Cadmium	0.401	Pure association	0.397	Direct

Strong non-linear correlation (determination coefficient higher than 0.64) between concentrations pair of metal: zinc-cadmium and moderate correlation (determination coefficient of between 0.49 and 0.64) between the pair of metals: zinc - lead in Champa rice species were observed. Only moderate linear relationship was observed between pairs of zinc-cadmium pair in Champa rice species.

**Conclusion:**

In this study, heavy metals cadmium, copper, lead, four species of Iranian rice contains Champa, Shamim, Fajr and pollen that are used in Yasouj, were studied. In the case of cadmium, the lowest pollution related to rice pollen and Fajr that were allowed in the standard range. Most of the contamination was related to species Champa and Shamim. In the lead-lowest, respectively Fajr species, Shamim, pollen and Champa was. In each of four local rice, the amount of pollution it has been standardized and less. In the case of copper according to the standard range was very wide, from the lowest to the highest amount of copper in the form of pollen, Champa, Fajr and Shamim were reported. In all four cases the average concentration of copper, more than the bottom threshold but less than the upper threshold has been set for this element. In other words, all four rice were in the standard range.

The final decision matrix, each four local rice has relatively acceptable concentration of the three elements cadmium, lead and copper, but from the perspective of the health of the species, in accordance with the matrix [4] The species of heavy elements discussion respectively include pollen, Fajr, has been Shamim and Champa. In other words, any pollen best about standards in the four species studied rice was allocated.

Rice is a global product and main source of carbohydrates in the diet of Asian countries, including Iran. 42.2 kg per capita rice consumption is estimated in Iran that in the second crop after the wheat. This has caused Iran is now considered one of the main importers of the product [18,19]. Malakootian and colleagues in 1389 a study to determine the amount of lead, cadmium, nickel and chromium in Hindi rice imported into the country have done. Cadmium concentrations in both studies have shown similar results. The study also found a higher level of cadmium and lead study Hedayatifar R. et al. In 1389 to measure the amount of lead and cadmium less than the limit (the limit set [20], as well as in M. Pirzada et al. [21] In 1391 to assess the "state of zinc and cadmium in rice paddy soils and Isfahan, Fars and Khuzestan. And their impact on food security "in 54 percent of the rice grains is less than the critical concentrations in rice grain (20 mg per kg), respectively. Measured cadmium concentrations in edible rice paddy soils and showed that in 12% of samples of Isfahan province, the concentration of cadmium more than recommended by the World Health Organization, was also higher [20]. In addition, the study Mohammad Shokrzadeh et al (1391) [22] in the central cities of Mazandaran province "in water samples in the study areas was more than suggested amount of cadmium in all areas and it was similar in case of cadmium but in relation to the lead in Champa rice and Shamim varieties it was higher in study but at the dawn of rice and pollen that were less than the standard rate was consistent.

In a study Mahmoud N. et al in 2014, "heavy metals concentration in Shiraz rice in rice and rice Khmrh Kamfiruz highest amount was lowest. The average cadmium significantly exceeds the maximum recommended standard Iranian national. Average lead and cadmium in rice samples from internal and external significantly more than determined amount and that had consistency with increasing [23].

The study also Takao Watanabe et al. [24] In 1996, a study on the "level of cadmium in rice samples from different regions of the world, the highest and lowest levels of cadmium in rice samples of Asian rice outside Asia. Japanese rice is very similar average value measured cadmium levels in the 1980s. The results showed significant differences in levels of cadmium in rice for local consumption depends on different parts of the world. Japanese rice also showed cadmium levels were not significantly changed in over a decade, but the differences compared to the rice has been less studied than any other rice species that is probably less use of pollution sources that mentioned above, however, it can be used in hi maple leave different effects. By studying Rahman Azizur et al. [25] In 2014, research on cadmium concentrations of elements in imported rice that is consistent Australian, but the lead did not match, although the concentrations of elements such as cadmium, chromium, copper and nickel in Hindi rice imported rice Australian above and concentrations of elements such as copper, nickel and cadmium concentrations in Vietnamese rice, chromium, copper, nickel and lead in Thai rice imports is higher than the Australian rice. The concentration of heavy elements in Pakistani rice is substantially lower than that of Australian rice in the rice situation reviewed in comparison with rice Hindi, Pakistani and Thai was different.

The current study is consistent with the study conducted by Mehrdad Cheraghi on weekly absorption of cadmium in samples of imported rice in the rice cultivated in cities of Borujerd and Isfahan in 2011, Indian and Iranian rice supply in the cities of Tehran and Hamedan, comparing the average concentration of cadmium in imported rice and Iran with the standard World Health Organization and the UN Food and Agriculture, pollution concentrations lower than the limit showed that rice had studied the differences [26]. The study also proof Mansouri and colleagues [27] in 1394 to investigate the concentrations of lead, cadmium and chromium in Hindi and Pakistani rice imports in Sanandaj, and Synadvbradarn studies in Bushehr[28] The concentration of cadmium, lead and chromium lower than the national and international standards but this did not conform with results and the reason may be how to plant and water pollution and soil of rice fields are the other factors. The average concentration of 290 micrograms for cadmium accumulated during a week was 290µg/kg for Champa, 156.4 µg/kg for cadmium species, 102.8 for Shamim, and for cadmium it was 80.4 µg/kg for rice species of Gerdeh. The average cumulative concentration of lead per week was obtained 33.2 µg/kg for Champa, 27.74 µg/kg for Shamim, for lead 13.02 µg/kg for Fajr, and 25.83 µg/kg for Gerdeh species. This is due to the cumulative effects of heavy metals and adverse effects on the human population. As well as a significant threat to food security in particular.

The comparison of the level of exposure to the estimated densities of heavy metals in metals in different species PTWI levels indicates that the level of exposure for all of the metals mentioned is lower than the standard level. Though it indicates an appropriate situation about the cases investigated, it is necessary to do a constant and exact examination of such foods. These results are in according to the results of Falco and colleagues [29]" the level of Arsenic, Plumb ,Cadmium caused by consuming sea foods in different groups of consumers in Spania" their results showed that level of all metals except Mercury ,in all groups except boys was lower that level determined. It was also in accordance with the results of Suhaimi and Colleagues [30] the level of heavy metals Arsenic ,Mercury ,Plumb, Cadmium in some kinds of find and shell in Singopour.

It shows the necessity of making a proper program to recognize the possible sources of contamination and to make the contaminations less dense in the local kinds of rice.

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