



ORIGINAL ARTICLE

Measurement of Zinc, Copper, Lead, and Cadmium in the Variety of Packaging Milk and Raw Milk in Tehran Markets by Anodic Stripping Voltammetry

Naficeh Sadeghi^{*1,2,3}, Masoomeh Behzad¹, Shervin Homay Razavi¹, Behrooz Jannat², Mohammad Reza Oveisi¹, Mannan Hajimahmoodi¹

¹Department of Drug and Food Control, School of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran

²Halal Research Center, Ministry of Health and Medical Education, Tehran, Iran

³Water Health Research Center, Ministry of Health and Medical Education, Tehran, Iran

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KEYWORDS

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ABSTRACT: Considering the importance of milk in our daily diet and the increased environmental pollutants, it is important to assess heavy metals in milk. This study seeks the idea of concentration of some heavy metals in packaging milks collected from 22 districts in Tehran. Moreover, the focuses on heavy metals are checked with the recommended permissible amounts. The samples are analyzed by pulsed ultrasonography and anodic pulse techniques. The polarography apparatus is used for this investigation. This device comprises of three workups of estimating, solubilizing, and clear qualities. Each time a cradle, test, and standard is included, these three stages finish. Each time including the above gauge, which is a similar bend underneath the chart, is plotted by the gadget. According to the program given to the device, the device repeats its work three times every 3 steps and adds buffer, sample, and standard. The ranges obtained from the mean Cd, Cu, Zn, and Pb correspondingly are in raw milk 0.099 ± 0.116 , 2.424 ± 4.017 , 4.990 ± 6.244 , 0.271 ± 0.640 $\mu\text{g}\cdot\text{ml}^{-1}$, in packaging milk (pasteurized and sterilized) 0.049 ± 0.037 , 0.228 ± 0.188 , 0.999 ± 0.873 , 0.048 ± 0.033 $\mu\text{g}\cdot\text{ml}^{-1}$ respectively. In almost all cases, concentrations of sub-metals allowed limitations and health concerns for milk and dairy consumption. The amount of all four heavy metals in raw milk was greater than that in pasteurized milk. This alteration was significant for Zinc, Lead, and Copper, but not significant for Cadmium.

INTRODUCTION

Environmental pollution brought about by heavy metals is a significant concern on the worldwide scale and the risk of exposure to lead and cadmium in food products is considered as a genuine threat to human health [1].

Animal utilization of lead and cadmium makes heavy metals aggregate in their meat and milk. More significant levels of substantial metals have likewise been found in animal feed because of bioaccumulation due to animal feeding in contaminated soils. [2]. Consumption of

modern technologies in food production has augmented the chances of contamination of food with various environmental pollutants, particularly lead and cadmium. Raw and packaging milk are considered as highly valuable foods since they contain considerable amounts of nutritional constituents such as proteins, minerals, and vitamins. Unfortunately, in industrial regions, heavy metals can be readily transferred through milk and milk-based products and they have adverse effects on human

health [3]. In this manner, exposure to the distinctive overwhelming metals through the consumption of milk as a significant piece of the everyday diet is perceptible. As indicated by the Environmental Protection Agency (EPA), lead, cadmium, copper, and zinc are probably the most widely recognized overwhelming metals instigating contamination [1]. It is well known that the Cd and Pb have the greatest detrimental effects on kidney and nervous system, respectively [4]. Cu and Zn are essential metals for normal function of the human body, however, from the toxicological point of view; their intake in amounts above recommended safe levels may cause a threat to the human health [2]. A few diagnostic strategies have been accounted for the assurance of these metals in milk and dairy items including inductively coupled plasma-mass (ICP-MS) spectrometry [5], inductively coupled plasma-optical discharge spectrometry (ICP-AES) [2], nuclear ingestion spectroscopy (AAS) [6], potentiometric stripping [7], and anodic stripping voltammetry (ASV) techniques [8]. Contrasted with these systematic procedures, the AAS method has been utilized to decide the vast majority of the components present in milk and related items, it is dull and uneconomical at the same time to investigate a few components in numerous examples. Additionally, the anodic stripping voltammetry (ASV) strategy is practical, profoundly delicate, and particular for synchronous assurance of a few metals. It is entrenched that for the assurance of Zn, Cu, Pb, and Cd in numerous frameworks the ASV is a reasonable technique while CSV has end up being progressively delicate for Se [9]. In this manner, these two methods can be utilized for judgments of these metal particles. Recently, the screen printed anodes were utilized for assurance of overwhelming metals [8, 10] in milk. The researchers announced another scientific strategy to decide lead particles in milk by utilizing a dispensable screen printed terminal adjusted with Nafion; and an "in situ" bismuth film with anodic stripping voltammetry [10]. A research study conducted revealed an electrochemical detecting stage dependent on electrochemically diminished graphene oxide film adjusted screen-printed terminal for synchronous assurance of cadmium and lead in milk [8]. The

significant point in the synchronous assurance of metal particles is the chance of intermetallic impedances that may happen during the examination [11]. As possible, by the end of this impact, the simultaneous utilization of the two methods for the assurance of various components in a solitary example arrangement would be conceivable. From the viewpoint of general wellbeing, it is essential to decide the sum and assimilation of substantial metals in milk and its items and to contrast their salts possible for general wellbeing. The points of this review were to: (I) create ASV-CVS strategies, to decide the substantial metals in milk and dairy items, and (ii) contrast the outcomes and worldwide measures as far as possible.

MATERIALS AND METHODS

Reagents

Nitric Acid 65%, Applichem Co. (with 8N003706 Code), Acetic Glycolic Acid, Merck (Code K35377356546), Tartaric acid, Merck (code 819A135004), Sodium acetate 3 Abe, Merck (code K37186665742), Copper Nitrate 3 Abe, Merck (code 6397182), Nitrate on 6 Abe, Merck (code 8912071), Lead nitrate, Merck (code 7397), Cadmium Nitrate 4 Abe, Merck (Code 211B981619).

Sampling

To improve the probability of finding ideal components, arbitrary examining was utilized to choose crude milk and other dairy items (purified and sanitized milk) in 22 areas of Tehran. In all areas, 30 examples of crude milk and 26 examples of other dairy items (purified and sanitized milk) were gathered from mass milk tanks during early daytime draining in summer 2016. Milk tests were moved to the lab and put away, lastly dissect at -20°C for metal particles.

Apparatus

The polarography apparatus consists of three stir-up, measuring, solubilizing and sweep values. Each time the gadget is plotted by including the above base, which is the curve underneath the chart. Each time the gadget is plotted by including the above base, which is the bend underneath the diagram. According to the program given to the gadget, each of the 3 stages of the gadget are

rehashed multiple times while including the support, test and standard, and different conditions applied to the gadget are as per the following: A 10 ml sodium acetate buffer is added to the tuberculosis machine, stirred at the rate of 2000 rpm for 60 seconds and nitrogen gas deoxidization. Then the measurement step commences. At this stage, the baseline is drawn. The electrode used in this measurement is Mercury hinge droplet electrode or HMDE. The droplet size is 5 μ l and the potential use is as the following: the pulse gain is 50 mV and the pulse time is 40 msec. The current measurement time is 20 milliseconds and the total time of the voltage was applied in each step. In the dissolution step and the measurement, the voltages vary from 200 mV to 1500 mV at 60 mV.s⁻¹ and 6 mV steps. Then, 500 μ l of the sample prepared for the machine is added and the nitrogen is stirred and deoxygenated for 100 seconds. Then, as described above, measurements are made and the voltammogram is obtained. Then, 100 μ l of the standard 4 doses of the specified concentration is added to the tubes in three separate steps. The measurement operation is repeated 3 times per addition of the standard from the first stage (concentration). The device takes a mean of 3 times measurement. These are called standard ads. Each sample is measured three times [12].

RESULTS AND DISCUSSION

The base measures of Zn, Cu, Pb, and Cd that could be estimated by means of the Differential Pulse Anodic Stripping Voltammetry (DPASV) technique applied in this review were determined to be 0.05mg.kg⁻¹, 0.01mg.kg⁻¹, 0.005mg.kg⁻¹ and 0.005mg.kg⁻¹, respectively. The exactness of estimations was determined depending on the three estimations for every component utilizing standard mistakes and was seen as 5.6%, 3.2%, 3.5% and 4.5% for the referenced components individually. The researchers demonstrated the grouping of iron in four brands of powder milk and baby food (in light of rice and wheat) was broke down by DPASV and voltammeter gadget. All out Means \pm SE of Iron in child nourishments and powder milk (n = 40) were 8.55 \pm 1.18 mg.100g⁻¹, 5.21 \pm 0.66, respectively. Iron level in child food type I and II was lower than

named esteem (p < 0.05). Moreover, concentration of Iron in powder milk type I and II was lower than the value of label (p < 0.05) [13]. In a study in 2016, the levels of trace elements Zinc, Copper, and heavy metals Lead and Cadmium were measured by Differential Pulse Anodic Stripping Voltammetry in 19 barley grain cultivars and their malts in Iran. The mean degrees of Zn, Cd, Pb, and Cu were estimated to be 18.813 \pm 8.575, 0.212 \pm 0.116, 0.278 \pm 0.163, and 3.746 \pm 1.118 mg.100g⁻¹ in the grain tests and 14.364 \pm 6.391, 0.153 \pm 0.098, 0.179 \pm 0.082, and 3.033 \pm 1.392 mg.100g⁻¹ in the malt tests, individually [14]. In an investigation in 2014, convergence of zinc, copper, lead, and cadmium in four brands of infant food (rice and wheat based) and powder milk was broke down with DPASV and polarography set. Complete Mean \pm SE of zinc, copper, lead, and cadmium in child nourishments (n = 240) were 11.86 \pm 1.474 mg.100g⁻¹, 508.197 \pm 83.154 μ g.100g⁻¹, 0.445 \pm 0.006, 0.050 \pm 0.005 mg.Kg⁻¹ separately. Additionally these sum in powder milk (n = 240) were 3.621 \pm 0.529 mg.100g⁻¹, 403.822 \pm 133.953 μ g.100g⁻¹, 0.007 \pm 0.003, and 0.060 \pm 0.040 mg.Kg⁻¹, respectively [15]. In a study in 2015 Ca, Mg, Zn, and P bioavailability from two experimental ultra-filtered fermented goats' milk (one of them with the probiotic *Lactobacillus plantarum* and another one without it), and fermented goats' milk samples available in the market were evaluated. Solubility, daily stability, and a model combining simulated gastrointestinal digestion and mineral retention, transport and uptake by Caco-2 cells were used to assess bioavailability. The most elevated Ca, Mg, Zn, and P bioavailability esteems consistently related to the aged milk created by our exploration gathering, which could be clarified by the impact of milk ultrafiltration. The fermented milk with *L. planetarium* showed higher Ca retention than the ones without the microorganism, and major Ca uptake when compared with commercial products. This could be attributed to a positive effect exerted by the probiotic strain [16]. Polarogram bends of the four components drawn by the gadget for bundling milk and crude milk tests are portrayed in Figures 1 and 2, respectively.

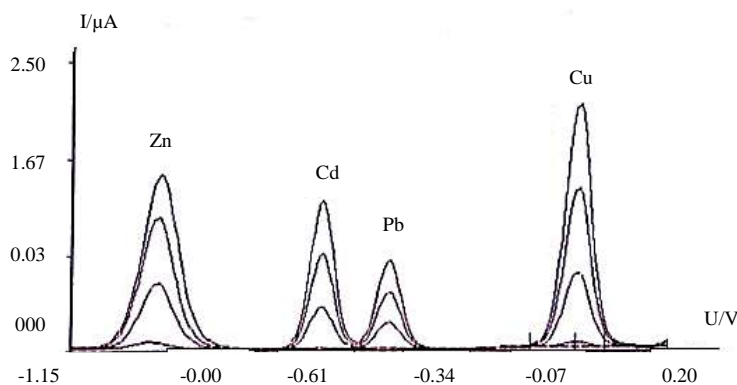


Figure 1. Polarogram Curves of the four element in packaging milk samples.

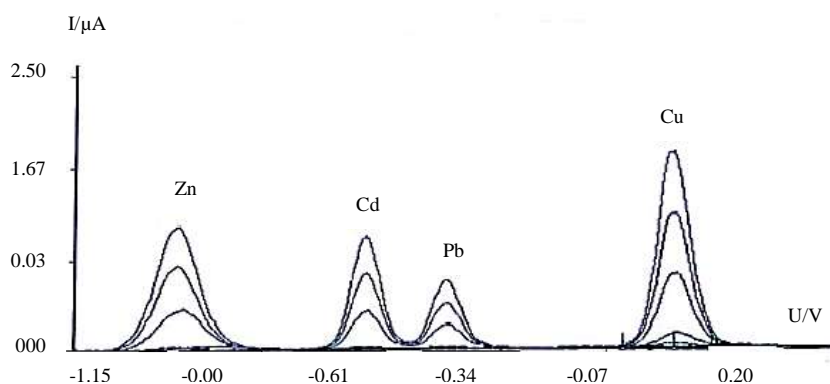


Figure 2. Polarogram Curves of the four element in crude milk samples.

Occurrence of Zn, Cd, Pb, and Cu in crude milk and packaging (pasteurized and sterilized milk)

The mean contents of Zn, Cd, Pb, and Cu in the crude milk and other dairy items are listed in Table 1. The request for metal fixations in all crude milk are as Zn>Pb>Cu>Cd and in bundling milk (purified and disinfected milk) Zn>Cu>Cd>Pb. The focus of Zn, Cd, Pb, and Cu can be positioned as follows: crude milk > purified and sanitized milk. The methods grouping of Zn, Cd, Pb, and Cu in crude milk tests of 22 areas of Tehran are shown in Table 2. According to Table 2 and the T-Test results, no statistically significant differences ($P > 0.05$) was found between the Cd content of raw milk and packaging milk (pasteurized and sterilized milk). The possible reason for this could be that a significant

reduction cannot occur in the Pb and Cd by thermal processing used in dairy industry, i.e. pasteurization and ultra-high temperature (UHT) treatment [17]. The concentration variation of Cu and Zn, in each sample is very remarkable and reported to be $2.195 \pm 0.519 \text{ mg.kg}^{-1}$ for Cu and $4.818 \pm 1.168 \text{ mg.kg}^{-1}$ for Zn. The methods convergence of Zn, Cd, Pb, and Cu in no flavor bundling milk tests of Tehran are indicated in Table 3. Tables 4 to 7 show the measure of every component as per milk flavor. Table 8 shows the correlations between the measures of zinc, copper, lead, and cadmium in crude and pressed milk tests, which are additionally appeared in Figure 3.

Table 1. The mean contents of Zn, Cd, Pb, and Cu in the raw milk and other dairy products.

	Zn (Mean±SD)	Cd (Mean±SD)	Pb (Mean±SD)	Cu (Mean±SD)
Raw milk	4.989±6.244	0.099±0.116	0.271±0.640	2.424±4.017
Pasteurized and sterilized milk	0.999±0.873	0.049±0.037	0.048±0.033	0.228±0.188

Table 2. The means concentration of Zn, Cd, Pb, and Cu in raw milk samples of 22 districts of Tehran.

Regions	Mean Zn± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Pb ± SD (mg.kg ⁻¹)	Mean Cu ± SD (mg.kg ⁻¹)
Region 1	0.92±2.21	0.05±0.34	0.01±0.01	0.14±0.10
Region 2	3.15±1.64	0.07±0.02	0.06±0.039	0.73±0.93
Region 3	0.67±0.53	0.04±0.03	0.05±0.07	0.58±0.30
Region 4	1.12±0.21	0.03±0.02	0.03±0.00	0.14±0.10
Region 5	2.35±2.01	0.14±0.06	0.12±0.02	0.66±0.86
Region 6	11.12±9.25	0.04±0.04	0.16±0.02	2.28±1.16
Region 7	1.97±0.06	0.05±0.03	0.03±0.02	0.50±0.34
Region 8	2.90±0.89	0.17±0.07	0.31±0.10	2.00±0.92
Region 9	4.80±2.67	0.07±0.06	0.11±0.11	0.59±0.61
Region 10	11.70±11.40	0.09±0.05	0.28±0.14	6.58±3.15
Region 11	8.20±1.96	0.22±0.10	0.41±0.22	2.80±3.36
Region 12	7.70±1.84	0.08±0.06	0.25±0.17	3.16±1.96
Region 13	1.00±0.33	0.04±0.04	0.04±0.05	0.48±0.17
Region 14	1.50±0.66	0.09±0.01	0.65±0.77	2.08±1.76
Region 15	4.35±0.69	0.06±0.02	0.28±0.00	5.67±7.65
Region 16	15.02±9.94	0.11±0.12	0.10±0.09	5.54±6.73
Region 17	12.89±10.68	0.20±0.06	0.66±0.65	3.15±2.44
Region 18	3.21±2.03	0.16±0.23	0.31±0.19	7.16±10.81
Region 19	0.57±0.67	0.11±0.04	0.24±0.26	0.28±0.13
Region 20	2.94±0.21	0.16±0.08	0.38±0.47	1.69±0.35
Region 21	0.67±0.19	1.31±2.34	0.04±0.02	3.97±2.87
Region 22	0.15±0.05	0.09±0.13	0.07±0.01	0.654±0.34

Table 3. The means concentration of Zn, Cd, Pb, and Cu in no flavor packaging milk samples of Tehran.

Milk Brands	Mean Cu ± SD (mg.kg ⁻¹)	Mean Pb± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Zn ± SD (mg.kg ⁻¹)
low fat Sahar	0.41±0.30	0.03±0.02	0.05±0.06	1.48±0.28
semi fat Sahar	0.19±0.25	0.05±0.08	0.02±0.05	1.62±0.27
full fat Sahar	0.64±0.14	0.04±0.14	0.09±0.12	1.33±0.17
low fat Mihan	0.25±0.12	0.02±0.02	0.04±0.02	1.12±0.95
semi fat Mihan	0.08±0.26	0.03±0.05	0.03±0.15	1.68±0.65
full fat Mihan	0.18±0.58	0.04±0.14	0.03±0.14	0.25±0.69
low fat Domino	0.08±0.04	0.05±0.04	0.06±0.03	0.74±0.17
semi fat Domino	0.05±0.03	0.04±0.03	0.08±0.06	0.97±0.26
full fat Domino	0.08±0.03	0.05±0.05	0.02±0.02	1.66±0.15
low fat Roozanh	0.23±0.07	0.05±0.02	0.02±0.00	1.75±0.67
semi fat Roozanh	0.26±0.04	0.04±0.03	0.01±0.00	2.12±0.48
full fat Roozanh	0.29±0.03	0.05±0.02	0.00±0.00	1.74±0.69
low fat Choopan	0.19±0.11	0.05±0.03	0.01±0.01	1.91±0.48
semi fat Choopan	0.17±0.09	0.06±0.02	0.02±0.02	2.02±0.11
full fat Choopan	0.21±0.06	0.02±0.01	0.01±0.01	2.08±0.21

Table 3. Continued.

low fat Damdaran	0.10±0.09	0.06±0.05	0.05±0.05	0.71±1.51
semi fat Damdaran	0.23±0.06	0.10±0.04	0.11±0.02	0.19±0.89
full fat Damdaran	0.10±0.04	0.03±0.02	0.03±0.03	2.24±0.27
low fat Manga	0.13±0.19	0.05±0.02	0.05±0.02	0.83±0.45
semi fat Manga	0.17±0.12	0.07±0.01	0.09±0.02	0.93±0.43
full fat Manga	0.19±0.12	0.04±0.02	0.03±0.01	1.05±0.27
low fat Pazhan	0.24±0.21	0.03±0.01	0.05±0.01	0.67±0.53
semi fat Pazhan	0.21±0.18	0.04±0.02	0.06±0.02	0.54±0.47
full fat Pazhan	0.25±0.16	0.03±0.02	0.05±0.01	0.84±0.18
low fat Pegah	0.07±0.00	0.02±0.01	0.02±0.03	0.15±0.33
semi fat Pegah	0.09±0.01	0.03±0.01	0.03±0.02	0.25±0.26
full fat Pegah	0.06±0.00	0.04±0.03	0.01±0.01	0.33±0.42
low fat Pak	0.51±0.30	0.03±0.05	0.06±0.03	1.30±0.90
semi fat Pak	0.44±0.24	0.04±0.03	0.03±0.02	1.46±0.56
full fat Pak	0.23±0.33	0.07±0.06	0.03±0.03	0.51±0.24
low fat Liona	0.21±0.29	0.02±0.01	0.12±0.05	1.41±1.47
semi fat Liona	0.12±0.25	0.02±0.02	0.08±0.03	1.22±1.02
full fat Liona	0.00±0.00	0.01±0.01	0.10±0.01	1.24±0.97
low fat Kaleh	0.17±0.12	0.08±0.03	0.06±0.03	0.52±0.74
semi fat Kaleh	0.18±0.09	0.05±0.02	0.04±0.02	0.69±0.54
full fat Kaleh	0.21±0.11	0.04±0.01	0.03±0.01	1.56±0.21
low fat Mimas	0.43±0.18	0.03±0.02	0.02±0.01	0.89±0.14
semi fat Mimas	0.35±0.12	0.02±0.03	0.03±0.02	1.04±0.29
full fat Mimas	0.19±0.10	0.03±0.03	0.02±0.01	0.69±0.28
low fat Haraz	0.17±0.13	0.05±0.04	0.06±0.05	0.37±0.16
semi fat Haraz	0.16±0.08	0.07±0.03	0.08±0.02	0.28±0.24
full fat Haraz	0.17±0.06	0.07±0.02	0.03±0.01	0.46±0.19
low fat Pakban	0.25±0.25	0.05±0.05	0.04±0.03	0.61±0.56
semi fat Pakban	0.32±0.21	0.03±0.03	0.03±0.04	0.63±0.61
full fat Pakban	0.41±0.15	0.04±0.01	0.07±0.02	0.67±0.55

Table 4. The means concentration of Zn, Cd, Pb, and Cu in chocolate milk packaging of Tehran.

Milk Brands	Mean Cu ± SD (mg.kg ⁻¹)	Mean Pb± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Zn ± SD (mg.kg ⁻¹)
Pak	0.19±0.10	0.05±0.05	0.02±0.00	1.68±0.12
Damdaran	0.07±0.40	0.03±0.01	0.07±0.01	1.85±0.15
Pegah	0.07±0.00	0.05±0.01	0.05±0.02	1.09±0.25
Choopan	0.02±0.14	0.04±0.00	0.00±0.00	1.58±0.19
Roozaneh	0.18±0.16	0.04±0.01	0.01±0.02	1.82±0.50
Mihan	0.24±0.16	0.10±0.26	0.13±0.25	0.18±0.16
Pajan	0.17±0.10	0.08±0.05	0.08±0.01	0.95±0.00
Pakban	0.25±0.00	0.02±0.05	0.08±0.18	0.68±0.10
Domino	0.09±0.08	0.02±0.06	0.06±0.72	0.34±0.40
Sahar	0.53±0.10	0.03±0.48	0.06±0.18	1.28±0.45
Kaleh	0.19±0.18	0.05±0.28	0.02±0.16	1.68±0.28
Mimas	0.07±0.01	0.03±0.04	0.07±0.48	1.85±0.16

Table 5. The means concentration of Zn, Cd, Pb, and Cu in honey packaging milks of Tehran.

Milk Brands	Mean Cu ± SD (mg.kg ⁻¹)	Mean Pb± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Zn ± SD (mg.kg ⁻¹)
Pegah	0.29±0.98	0.06±0.00	0.03±0.28	1.98±0.10
Roozaneh	0.08±0.00	0.05±0.38	0.07±0.91	1.80±0.28
Mihan	0.07±0.19	0.07±0.04	0.07±0.00	1.03±0.02
Domino	0.02±0.18	0.03±0.19	0.00±0.00	1.09±0.38
Kaleh	0.19±0.38	0.05±0.15	0.01±0.06	1.34±0.15
Damdaran	0.58±0.76	0.18±0.49	0.12±0.08	0.68±0.28
Sahar	0.21±0.12	0.08±0.37	0.05±0.00	0.87±0.46
Pak	0.33±0.39	0.02±0.00	0.07±0.01	0.68±0.12

Table 6. The means concentration of Zn, Cd, Pb, and Cu in coffee packaging milks of Tehran.

Milk Brands	Mean Cu ± SD (mg.kg ⁻¹)	Mean Pb± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Zn ± SD (mg.kg ⁻¹)
Roozaneh	0.29±0.45	0.06±0.07	0.03±0.00	1.98±0.98
Mihan	0.08±0.12	0.05±0.05	0.07±0.00	1.80±0.49
Kaleh	0.07±0.00	0.07±0.97	0.07±0.31	1.03±0.56
Pak	0.02±0.07	0.03±0.08	0.00±0.00	1.09±0.13
Domino	0.19±0.37	0.05±0.44	0.01±0.11	1.34±0.57
Damdaran	0.58±0.55	0.18±0.81	0.12±0.00	0.67±0.51
Pegah	0.21±0.51	0.08±0.00	0.05±0.01	0.87±0.32
Sahar	0.33±0.64	0.02±0.00	0.07±0.37	0.68±0.15

Table 7. The means concentration of Zn, Cd, Pb, and Cu in banana packaging milks of Tehran.

Milk Brands	Mean Cu ± SD (mg.kg ⁻¹)	Mean Pb± SD (mg.kg ⁻¹)	Mean Cd ± SD (mg.kg ⁻¹)	Mean Zn ± SD (mg.kg ⁻¹)
Pegah	0.35±0.19	0.06±0.72	0.02±0.00	1.65±0.19
Damdaran	0.09±0.37	0.05±0.64	0.08±0.00	1.58±0.46
Mihan	0.01±0.01	0.07±0.01	0.05±0.00	1.65±0.19
Roozaneh	0.03±0.05	0.03±0.08	0.00±0.00	1.08±0.94
Kaleh	0.16±0.18	0.05±0.00	0.01±0.19	1.36±0.19
Domino	0.58±0.76	0.25±0.16	0.15±0.00	0.69±0.05
Pak	0.35±0.25	0.03±0.02	0.05±0.00	0.88±0.33
Sahar	0.33±0.75	0.03±0.43	0.08±0.00	0.59±0.13

Table 8. Comparison of Zn, Cd, Pb and Cu between raw milk and packaging milk (pasteurized and sterilized milk).

	T-Test	Sig	Result
Comparison of Zn between raw milk and packaging	4.819	0.000	(P<0.05)There are significant differences
Comparison of Cd between raw milk and packaging	0.131	0.113	(P>0.05)There are not significant differences
Comparison of Pb between raw milk and packaging	0.222	0.009	(P<0.05)There are significant differences
Comparison of Cu between raw milk and packaging	2.195	0.000	(P<0.05)There are significant differences

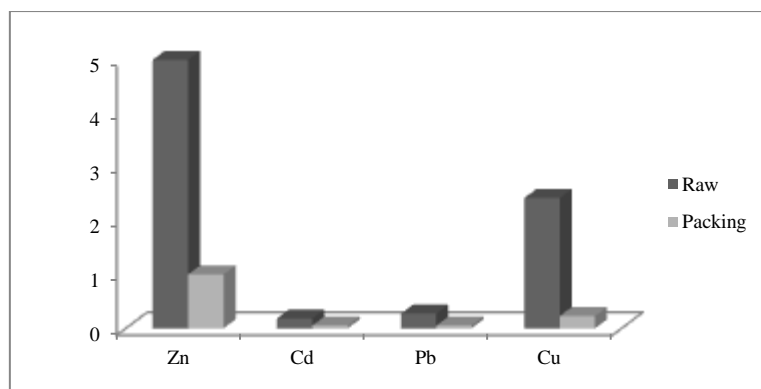


Figure 3. Comparison of heavy metal content between raw and packaging milk.

An examination of heavy metal substance with standard cutoff points

Maximum permitted level (MPL) for Pb was $20\mu\text{g.kg}^{-1}$ by wet weight. The Pb content levels in all of samples were below the permissible limit. Based on the type of food and Cd level in the food-producing environment, the concentration of Cd in milk varies largely. A recent study revealed that the main inputs of Cd to animal feed in domestic animals are crops, trace element premixes, fishmeal, and minerals such as limestone and phosphate [18]. The extreme allowable degree of Cd, Zn, and Cu in dairy items has been accounted to be 10, 50, and $30\mu\text{g.kg}^{-1}$ of wet load in FAO/WHO limits [19]. By contrasting our information results and as far as possible on Cd, Zn, and Cu, which is the most careful official, all examples appear to be considered as protected.

CONCLUSIONS

The admission amounts of lead, copper, zinc, and cadmium in Iran indicated that they are lower than the suggested passable sums. The minimal proportion of overwhelming metal pollutions (Pb and Cd) is in locale 1 and 4 for crude milk and Roozaneh, Domino, and Choopan for packaged milk.

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Conflict of interests

The authors declare that there is no conflict of interest.

REFERENCES

1. Saei-Dehkordi S.S., Fallah A.A., 2011. Determination of copper, lead, cadmium and zinc content in commercially valuable fish species from the Persian Gulf using derivative potentiometric stripping analysis. *Microchem J.* 98(1), 156–162.
2. Licata P., Di Bella G., Potorti A.G., Lo Turco V., Salvo A., Dugo G.mo., 2012. Determination of trace elements in goat and ovine milk from Calabria (Italy) by ICP-AES. *Food Addit Contam Part B.* 5(4), 268–271.
3. Pavlovic I., Sikiric M., Havranek L., Plavljanic N., Brajenovic N., 2004. Lead and cadmium levels in raw cow's milk from an industrialised Croatian region determined by electrothermal atomic absorption spectrometry. *Czech J Anim Sci.* 49 (4), 164–168.
4. Neal A.P., Guilarte T.R., 2013. Mechanisms of lead and manganese neurotoxicity. *Toxic Res.* 2(2), 99–114.
5. Khan N., Jeong I. S., Hwang I. M., Kim J.S., Choi S.H., Nho E.Y., Choi J.Y., Park K.S., Kim K.S., 2014. Analysis of minor and trace elements in milk and yogurts by inductively coupled plasma-mass spectrometry (ICP-MS). *Food Chem.* 147, 220–224.
6. Freschi G.P.G., Fortunato F.M., Freschi C.D., Neto J.A.G., 2012. Simultaneous and direct determination of As, Bi, Pb, Sb, and Se and Co, Cr, Cu, Fe, and Mn in milk by electrothermal atomic absorption spectrometry. *Food Anal Methods.* 5(4), 861–866.
7. Munoz E., Palmero S., 2004. Determination of heavy metals in milk by potentiometric stripping analysis using a home-made flow cell. *Food Control.* 15(8), 635–641.
8. Ping J., Wang Y., Wu J., Ying Y., 2014. Development of an electrochemically reduced graphene oxide modified disposable bismuth film electrode and its application for

- stripping analysis of heavy metals in milk. *Food Chem.* 151, 65–71.
9. Mahesar S., Sherazi S., Niaz A., Bhangar M., Rauf A., 2010. Simultaneous assessment of zinc, cadmium, lead and copper in poultry feeds by differential pulse anodic stripping voltammetry. *Food Chem Toxicol.* 48(8), 2357–2360.
10. Quintana J.C., Arduini F., Amine A., Van Velzen K., Palleschi G., Moscone D., 2012. Part two: Analytical optimisation of a procedure for lead detection in milk by means of bismuth-modified screen-printed electrodes. *Anal Chim Acta.* 736, 92–99.
11. Adeloju S., Bond A., Hughes H., 1983. Determination of selenium, copper, lead and cadmium in biological materials by differential pulse stripping voltammetry. *Anal Chim Acta.* 148, 59–69.
12. Sadeghi N., Jodakhanlou M., Oveisi M.R., Jannat B., Behzad M., Hajimahmoodi M., 2017. Determination of Zinc and Copper micronutrients and Lead and Cadmium contaminants in non-alcoholic malt beverages by anodic stripping voltammetry. *JFSH.* 3, 54-58.
13. Jannat B., Sadeghi N., Oveisi M.R., Behzad M., Hajimahmoodi M., Aghazadeh F., 2017. Determination of Iron in baby weaning food and powder milk. *J.B.M.* 5, 1- 6.
14. Sadeghi N., Oveisi M.R., Jannat B., Hjimahmoodi M., Malayeri N., Behzad M., 2016. Assessment of some heavy metals concentration and antioxidant activity in Barely Grain Cultivars and Their Malts from Iran. *JACEN.* 5, 121-131.
15. Sadeghi N., Oveisi M.R., Jannat B., Hajimahmoodi M., Behfar A., Behzad M., Norouzi N., Oveisi M., Jannat B., 2014. Simultaneous Measurement of Zinc, Copper, Lead and Cadmium in Baby Weaning Food and Powder Milk by DPASV. *Iran J Pharm Res.* 13(1), 345-349.
16. Bergillos-Meca T., Cabrera-Vique C., Artacho R., Moreno-Montoro M., Navarro-Alarcón M., Olalla M., Giménez R., Seiquer I., Ruiz-López M.D., 2015. Does *Lactobacillus plantarum* or ultrafiltration process improve Ca, Mg, Zn and P bioavailability from fermented goats' milk? *Food Chem.* 15,187,314-321.
17. Moreno J., Hernandez T., Garcia C., 1999. Effects of a cadmium-contaminated sewage sludge compost on dynamics of organic matter and microbial activity in an arid soil. *Biol Fertil Soils.* 28(3), 230–237.
18. Bilandz'ic' N., Dokic' M., Sedak M., Solomun B., Varenina I., Knez'evic' Z., Beni'c' M., 2011. Trace element levels in raw milk from northern and southern regions of Croatia. *Food Chem.* 127(1), 63–66.
19. Codex Alimentarius Commission, 1999. Discussion paper on maximum level for Pb in milk and secondary milk products. Joint FAO/WHO food standards programme, twenty-third session.

