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DETECTION OF HEAVY METALS POLLUTION IN TYPES OF MILK SAMPLES IN BAGHDAD MARKETS

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ABSTRACT

The levels of lead (pb), copper (cu), cobalt (co) and cadmium (cd) were determined in different kinds of milk and the health risks were evaluated. The mean levels were 0.73 ± 0.21 , 0.06 ± 0.01 , 0.12 ± 0.01 and 0.14 ± 0.01 ppm for these metals respectively. The levels of pb and cu were found to be insignificant differences ($p < 0.05$), whereas the levels of co and cd, were no significant differences ($p > 0.05$). The dry and liquid kinds of milk were different significantly ($p < 0.05$), whereas the original, was no significant differences ($p > 0.05$). The values for all metals were more than one. The metals pb and cd were detected at highest concentrations in most dry and liquid milk samples.

Keywords: Heavy metal, dry and liquid milk, target hazard quotient.

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الكشف عن تلوث المعادن الثقيلة في انواع الحليب في اسواق بغداد

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الخلاصة

تم قياس مستويات الرصاص (Pb) والنحاس (Cu) والكوبالت (Co) والكاديوم (Cd) في أنواع مختلفة من الحليب وتقييم المخاطر الصحية، وكان معدل المستويات 0.73 ± 0.21 و 0.06 ± 0.01 و 0.12 ± 0.01 و 0.14 ± 0.01 جزء بالمليون لتلك العناصر بالترتيب، ووجد ان مستويات الرصاص والنحاس ذات فروقات معنوية ($P < 0.05$) بينما لم تكن مستويات الكوبالت والكاديوم ذات فروقات معنوية ($P > 0.05$)، وكان لنوع الحليب جاف او سائلا ذو اختلاف معنوي ($P < 0.05$) في جميع الانواع بينما لم يكن المنشأ ذو اختلاف معنوي ($P > 0.05$)، وكانت قيم معامل الخطورة المستهدف (THQ) لجميع المعادن اكثر من واحد، وأكتشفت تراكيز عالية لمعدني الرصاص والكوبالت في اغلب العينات السائلة والصلبة للحليب.

الكلمات المفتاحية: المعادن الثقيلة، حليب مجفف وسائل، معامل الخطر المستهدف



INTRODUCTION

Milk is considered as essential food in human nutrition, especially for children. Milk is a complete meal because it consists of active nutrients like proteins, fatty acids, lactose, vitamins, and metals. On the other hand, milk may be hazards for the health of the consumer if contain different contaminants including microbes (**Haug *et al.*, 2007**). Scientifically there are some elements as carbone, hydrogen, oxygen, sodium, potassium, nitrogen, phosphor, sulfide, magnesium, calcium, and chloride considered as main essentials for life and boron, silicon, vanadium, chromium, manganese, cobalt, nickel, copper, zinc, selenium, molybdenum and iodine considered as trace elements essential for life. For instance, cobalt (II) is a component of vitamin B12. A minimum level of these metals is necessary for body growth, but high levels convert to toxic substances. Some of metals like lead and cadmium are accumulated in the body because they are not biodegradable. Metals content of milk can be divided into essential elements (Cu and Co) at low doses and nonessential or toxic (Pb and Cd) (**Zhai *et al.*, 2015; Lata Bansal & Asthana, 2018**). Results of the previous study of risk assessments for breast milk contain metals designated that the consumption of food contaminated with metals as lead, mercury, and arsenic by children considered as worry situation for health (**Rebelo & Caldas, 2016**). Some industries are causing pollution or contamination to different environments as water, soils, nutrients, and vegetation with heavy metals reason absorption interested to the elements of food chain and carry out an abundant danger to the life of different organisms (**Bilandzic *et al.*, 2011**). According to the world health organization, the nutrients are the routine mode of heavy metals exposure to the human with percent more than ninety in non-smokers individuals (**WHO, 2007**). Additionally, intake of food polluted with Cd is considered as common way of exposure. A study was proved that the exposure to the contaminant cadmium was related with carcinogenesis effects to body tissues and organs as breast, stomach, intestines, prostate, testes, lungs and esophagus (**Carver & Gallicchio, 2017**).

Interns of Cd inside the cells cause many changes as proliferation and apoptosis. International Agency for Research on Cancer (IARC) was putted the Cd in a carcinogenic list substances. Although Cd consider as poor genotoxic, indirect effects may resulting from it ingesting to the body as reactive oxygen species (ROS) and DNA damage. Furthermore Cd is playing a role of reducing actions of proteins complex in antioxidant resistances also in gene expression and signal transduction. A study was documented that Cd is correlated with DNA repair inhibits (**Bertin & Averbec, 2006**). A local study was concluded that the Iraqi consumers preference the imported dairy products to local product according to its quality in all its elements (**Alkhafaji, 2018**). It is known that some of heavy metals are necessary to human health called micronutrient components like copper, zinc, and iron. Presence of these metals in the body with high concentrations will be dangerous to human health. In opposite deficiencies of these metals will lead to disease (**Kazi *et al.*, 2009**).

An extra understanding of heavy metal intake via milk consumption and possible hazards to human health are still wanted. Hence, this work was carried out to determine the concentrations of some important heavy metals in milk and evaluate the possible health risks resulting from drinking of milk contaminated with these metals.

MATERIAL AND METHODS

Collection of samples:

In this work, we collect 19samples of different trademarks of milk (15 dry and 4 liquid) from locally markets. The locally bovine milk was collected from Al-Rasheed area (South of Baghdad). Atomic Absorption Spectrophotometer (AAS) was used to determine the metals

concentrations. For dry milk samples 2g were digested into mixture of nitric and hydrochloric acid (4:1 volume/volume) to obtain a transparent solution, same step was applied to liquid milk samples 2mL (Patra *et al.*, 2008). Samples were filtered then diluted to a suitable volume later.

Daily Intake of Metals (EDI)

DI was calculated by modified equation below:

$$EDI = DMC \times MC \dots\dots\dots (\text{Ahmed } et \text{ al.}, 2015)$$

DMC = daily milk consumption 200mL

MC = mean metal concentration in milk

Target hazard quotient (THQ)

The THQ was evaluated according to the food chain and the reference oral dose (RFD_o) for each metal. If the THQ value is more than one, that means the exposed individuals is supposed to be safely (Zhuang *et al.*, 2009).

$$THQ = EDI/RFD_o$$

Statistical analysis

Concentrations of all metals were expressed as mean±standard error (SE), minimum and maximum values. All calculations were performed with the (SPSS v25). Non-parametric qi-square and Kruskal Wallis analysis were used to calculate the significant differences depending on (P= 0.05) probability level.

RESULTS AND DISCUSSION

The levels of heavy metals Pb, Cd, Co, and Cu of milk samples are refer it in (Table 1, 2). These metals were reported in most samples of milk, the arrangement of these metals was Pb>Cd > Co> Cu.

The levels of Pb, Cu, Co, and Cd in dry and liquid milk samples were found to be in the ranges of 0-2.6, 0.01-0.31, 0.01-0.25, and 0.02-0.3 ppm respectively. The means of these concentrations were found to be 0.73±0.21, 0.06±0.01, 0.12±0.01 and 0.14±0.01 ppm respectively (Table 1). There are many studies about heavy metal contamination in milk conducted in Iraq (Alani & Al-Azzawi, 2015; Al-Dabbagh, 2013).

Heavy metals in milk

Lead in milk

Reports on pollution of milk with lead heavy metal were occurred in many states by many researchers (Tajkarimi *et al.*, 2008; Rahimi, 2013) in Iran, (Kazi *et al.*, 2009) in Pakistan, (Bilandzic *et al.*, 2011) in Croatia, (Maas *et al.*, 2011) in France, and (Temiz & Soyly, 2012) in Turkey. They are calculated the levels of Pb in milk and they found it at ranges of 0.001-0.046, 0.00018-0.000611, 0.0418-0.0587, 0.0362-0.0587, 0.009-0.126, 0.028-0.068 and 0.015-0.061 mg/L respectively.

The contamination sources of Pb in milk are industrial and environmental (atmospheric deposition, waste disposal, vehicle exhausts, urban effluent, etc. (Standards Codex Alimentarius, 1995). In our work, the results refer that the levels of Pb in milk samples were in high level of a total 19 sample of milk, 11 samples were in over of the acceptable level of 0.02 ppm (Table 2).

According to WHO (2007), lead is found in the body in many organs as liver, brain, bones, and kidney. In addition, lead is stored in the skeletal system of human especially in teeth and other bones and it has ability to accumulate in these positions through the time.

In this work, the concentrations of Pb and Cu were significant differences ($P < 0.05$). But, the concentrations of Co and Cd were had no significant differences ($P > 0.05$). The dry and liquid kinds of milk were significantly differences ($P < 0.05$). But, the origin of milk or manufacture country were no different significantly ($P > 0.05$).

Copper in milk

Copper contents in dry and liquid milk samples were in the ranges of 0.01-0.31 ppm with mean concentrations of 0.06 ± 0.01 ppm (Table 1). Our findings were agreed with the levels got by (Abdou *et al.*, 2017), they found that the mean value of Cu in milk was (0.68 ± 0.17) . Also, it was agreed with the levels obtained by (Alani & Al-Azzawi, 2015), their results showed the highest content of Cu in milk were higher than standards of the International Dairy Federation (IDF, 1979). However, our results were noticed that Cu content in most dry types of milk samples was more than the permissible limit 0.05 ppm (WHO, 2007). But, all liquid types of milk were less than the permissible limit (Table 2).

A study was reported the necessity of Co for deferent activities in the body for growth, cardiovascular system, lungs elasticity, function of neuro endocrine, and metabolism of metals as iron considered as essential trace element of the human body (Sieber *et al.*, 2006).

Cobalt in milk

Cobalt contents in dry and liquid milk samples were in the ranges of 0.01-0.25 ppm with mean concentrations of 0.12 ± 0.01 ppm (Table 1).

Our findings were agreed with the levels got by (Huque *et al.*, 2018), they showed that cobalt content in pasteurized milk 1.48 ppm was significantly higher than raw milk 1.02 ppm. However, our results were noticed that Co content in all dry kinds of milk samples was more than the permissible limit 0.02 ppm (WHO & FAO, 2018). But, all liquid types of milk were less than the permissible limit (Table 2).

A study was proved that the cobalt metal is important component of vitamin B12 (cyanocobalamin) and considered as main substance for the human body. It is coenzyme in the cellular process like oxidation of fatty acids, synthesis of DNA, synthesis of amino acids with folic acid and production of red blood cells (Barceloux & Barceloux, 1999).

Cadmium in milk

Recently, the contamination of food by cadmium is considered as most important subject (Austin & Vincent, 2018). In the current study, cadmium levels in studied samples of many type of milk were found to be in ranges of 0.027-0.3 ppm. The mean levels of Cd were found to be 0.14 ± 0.01 ppm (Table 1 2). The occurrence of Cd in milk may be due to either natural or anthropogenic origins (fertilizers and atmospheric deposition in soils). Our findings were agreed with the funding and limits of levels documented by (Zhou *et al.*, 2019), they found that the mean value of Cd in milk was 0.05 ppm. However, our results were indicated that Cd levels in dry milk were higher than the acceptable level of 0.1 ppm established by (Abdou *et al.*, 2017). But, the liquid milk samples were saved (Table 2).

Cadmium is known as toxic metal to human health and considered as important contaminant in ecological studies. Exposure of human to Cd occurs through inhalation, smoking, food and water. Mechanism of carcinogenic effect of Cd poisoning include generation of ROS inside the body, preventing the processes of DNA repair and event some alterations of epigenetic traits (Bertin & Averbek, 2006).

Estimation of daily intake

EDI of Pb

To evaluate the health risk associated with heavy metal contamination of dry and liquid types of milk, we compared the obtained EDI with the tolerable daily intake (TDI) as a



standard. The tolerable weekly intakes (PTWI) of Pb was limit by FAO/WHO Expert Committee on Food Additives as 25 mg/kg bw (equal 3.6 mg/kg bw/day) for body of human depending on consider the lead as a cumulative poison (WHO & FAO, 2018). The mean EDI of Pb from consumption of milk was found to be 2.43 ± 3.1 mg/kg bw/day 68.2% of total daily intake (TDI) (Table 3). Although the mean EDI of Pb was less than the PTWI, we thought that the consumption of milk is not safe because the EDI was calculated for one source of food, so we expect the risk will be more if we calculate other sources of food with milk.

EDI of Cu

The mean EDI of Cu from ingesting of milk was recorded as 0.20 ± 0.2 mg/kg bw/day 5.8% of TDI (Table 3).

EDI of Co

The mean EDI of Co from consumption of milk was found to be 0.43 ± 0.2 mg/kg bw/day 12.1% of TDI (Table 3).

EDI of Cd

The PTMI of Cd was limited to 25 mg/kg bw (equal $0.83 \mu\text{g/kg}$ bw/day) (WHO & FAO, 2018). The mean EDI of Cd via drinking of milk was found to be 0.29 ± 0.2 mg/kg bw/day 13.9% of total EDI (Table 3). Although the mean EDI of Cd was less than the PTWI, we thought that the consumption of milk is not safe because the EDI was calculated for one source of food, so we expect the risk will be more if we calculate other sources of food with milk.

Daily intake of copper

The results of current work showed that the total daily intake (mg/day) for Cu from all kinds of milk was $2.37E+02$ mg/day, which contributes $2.63E+04\%$ of the RDA value (Table 4). Thus, all kinds of milk were considered a rich store of copper. Deficiency of Copper in human body is rare except in status of malnutrition severity.

Target hazard quotient (THQ)

The THQ has been standard as appropriate parameter to calculation of dangers linked to the ingesting of metal via polluted nutrients. It is known that the THQ is a rate of determined dose of a contaminant to a reference oral dose (RFD0) for any material (Zhuang *et al.*, 2009). The results of this work showed that the mean of THQ of all metals Pb, Cu, Co, and Cd via milk consumption were more than one (Table 5).

According to the results of THQ in this study, we suggested that the populations in Iraq will be in contact with possible health danger via consumption of dry milk in percent more than consumption of liquid milk. In addition, there are other sources of metal exposures such as dust via inhalation, skin, food and water, which may increase the possibility of health risks.

Table (1): Total concentrations (ppm) of Pb, Cu, Co and Cd in all types of milk with limits concentrations.

Metal	Concentrations (ppm)			Limits (ppm)
	Min	Max	Mean \pm SE	
lead	ND	2.6	0.73 ± 0.21	0.02 (FAO/WHO, 2009)
Cooper	0.01	0.31	0.06 ± 0.01	0.05 (WHO, 2008)
Cobalt	0.01	0.25	0.12 ± 0.01	0.02 (WHO, 1989)
Cadmium	0.02	0.3	0.14 ± 0.01	0.03 (IDF, 1979)
Total	0.04	3.46	0.26 ± 0.24	

**Table (2):** Concentrations of Pb, Cu, Co and Cd (ppm) in kinds of milk.

Type	Nature	Origin	Patch no.	Element (ppm)					
				Pb	Cu	Co	Cd	Mean	SE
Dielac	Dry	Newslan	07	0.4	0.07	0.1	0.16	0.182	0.07
Dano	Dry	Denmark	1218177051	ND§§	0.07	0.1	0.21	0.095	0.04
Altunsa	Dry	EAU	12PABA20	ND	0.07	0.2	0.21	0.120	0.05
Mahmood	Dry	Turkey	37CFA29	ND	0.07	0.2	0.16	0.107	0.04
Lancy	Dry	Jordan	DR75:3	ND	0.04	0.1	0.21	0.087	0.04
Silva	Dry	Jordan	DR778	2.6	0.04	0.15	0.1	0.722	0.62
Majan	Dry	EAU	NL§	ND	0.04	0.2	0.21	0.112	0.05
Fresh	Dry	Oman	NL	1.3	0.07	0.1	0.21	0.420	0.29
Nido	Dry	EAU	NL	2.6	0.07	0.15	0.1	0.730	0.62
Mudhish	Dry	Oman	1212180643	2.6	0.07	0.15	0.3	0.780	0.60
Redcaw	Dry	KSA	NL	ND	0.04	0.2	0.26	0.125	0.06
Aldiyar	Dry	EAU	2111181110	ND	0.04	0.2	0.16	0.100	0.04
Puck	Dry	Denmark	1218325051	1.3	0.04	0.15	0.1	0.397	0.30
Landoz	Dry	Jordan	NL	1.3	0.07	0.25	0.21	0.457	0.28
Powder	Dry	Iraq	NL	1	0.31	0.14	0.1	0.387	0.20
Kdd	Liquid	Kuwait	NL	0.2	0.023	0.027	0.04	0.072	0.04
Bovine milk	Liquid	Iraq	NL	0.2	0.02	0.014	0.04	0.068	0.04
Almaraaey	Liquid	KSA	NL	0.15	0.022	0.022	0.027	0.055	0.03
Alrafidin	Liquid	Iraq	-	0.23	0.01	0.01	0.033	0.070	0.05
Total				13.88	1.185	2.463	2.84	5.086	2.84

§NL, no liable; §§ND, non-detected

Table (3): EDI of metals (mg/kg bw/day) via consumption of milk by adult person (60 kg bw).

Kind of milk	EDI (mg/kg/day)			
	Pb	Cu	Co	Cd
Dielac	1.33	0.23	0.33	0.53
Dano	0.00	0.23	0.33	0.70
Altunsa	0.00	0.23	0.67	0.70
Mahmood	0.00	0.23	0.67	0.53
Lancy	0.00	0.13	0.33	0.70
Silva	8.67	0.13	0.50	0.33
Majan	0.00	0.13	0.67	0.70
Fresh	4.33	0.23	0.33	0.70
Nido	8.67	0.23	0.50	0.33
Mudhish	8.67	0.23	0.50	1.00
Redcaw	0.00	0.13	0.67	0.87
Aldiyar	0.00	0.13	0.67	0.53
Puck	4.33	0.13	0.50	0.33
Landoz	4.33	0.23	0.83	0.70
Powder	3.33	1.03	0.47	0.33
Kdd	0.67	0.08	0.09	0.13
Bovine milk	0.67	0.07	0.05	0.13
Almaraaey	0.50	0.07	0.07	0.09



Alrafidin	0.77	0.03	0.03	0.11
Mean \pm SD	2.43 \pm 3.1	0.20 \pm 0.2	0.43 \pm 0.2	0.29 \pm 0.2
Total	13.88 (68.2%)	1.185 (5.8%)	2.463 (12.1%)	2.84 (13.9%)

Table (4): Copper daily intake (mg/day) via drinking of milk and RDAs values (Recommended Dietary Allowances 0.9 mg/day).

Cu		
Type of milk	DI (mg/day)	Contribution of DI to RDA (%)
Dielac	1.40E+01	1.56E+03
Dano	1.40E+01	1.56E+03
Altunsa	1.40E+01	1.56E+03
Mahmood	1.40E+01	1.56E+03
Lancy	8.00E+00	8.89E+02
Silva	8.00E+00	8.89E+02
Majan	8.00E+00	8.89E+02
Fresh	1.40E+01	1.56E+03
Nido	1.40E+01	1.56E+03
Mudhish	1.40E+01	1.56E+03
Redcaw	8.00E+00	8.89E+02
Aldiyar	8.00E+00	8.89E+02
Puck	8.00E+00	8.89E+02
Landoz	1.40E+01	1.56E+03
Powder	6.20E+01	6.89E+03
Kdd	4.60E+00	5.11E+02
Bovine milk	4.00E+00	4.44E+02
Almaraaey	4.40E+00	4.89E+02
Alrafidin	2.00E+00	2.22E+02
Total	2.37E+02	2.63E+04

Table (5): THQ for daily exposure to metals through consumption of milk.

Type of milk	THQ (mg/kg-day/mg/kg-day)			
	Pb	Cu	Co	Cd
Dielac	3.81E+02	6.67E+01	9.52E+01	1.52E+02
Dano	0.00E+00	6.67E+01	9.52E+01	2.00E+02
Altunsa	0.00E+00	6.67E+01	1.90E+02	2.00E+02
Mahmood	0.00E+00	6.67E+01	1.90E+02	1.52E+02
Lancy	0.00E+00	3.81E+01	9.52E+01	2.00E+02
Silva	2.48E+03	3.81E+01	1.43E+02	9.52E+01
Majan	0.00E+00	3.81E+01	1.90E+02	2.00E+02
Fresh	1.24E+03	6.67E+01	9.52E+01	2.00E+02
Nido	2.48E+03	6.67E+01	1.43E+02	9.52E+01
Mudhish	2.48E+03	6.67E+01	1.43E+02	2.86E+02
Redcaw	0.00E+00	3.81E+01	1.90E+02	2.48E+02
Aldiyar	0.00E+00	3.81E+01	1.90E+02	1.52E+02
Puck	1.24E+03	3.81E+01	1.43E+02	9.52E+01
Landoz	1.24E+03	6.67E+01	2.38E+02	2.00E+02
Powder	9.52E+02	2.95E+02	1.33E+02	9.52E+01
Kdd	1.90E+02	2.19E+01	2.57E+01	3.81E+01
Bovine milk	1.90E+02	1.90E+01	1.33E+01	3.81E+01
Almaraaey	1.43E+02	2.10E+01	2.10E+01	2.57E+01
Alrafidin	2.19E+02	9.52E+00	9.52E+00	3.14E+01
Mean	6.97E+02	5.94E+01	1.23E+02	1.42E+02
Total	1.32E+04	1.13E+03	2.35E+03	2.70E+03



CONCLUSION

The recent study provides available key data on the concentrations of heavy metals (lead, cadmium, copper and cobalt) in dry and liquid milk included in this work. Lead and cadmium were the metals detected at highest levels in milk. Also, the funding of this work showed that drinking of liquid milk batter than drinking of dry milk. Consumption of milk is at risks, because the ability of lead and cadmium to bioaccumulation in human body.

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