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## Determination of Heavy Metals Content (Cadmium and Lead) in Raw Cow's and Buffalo's Milk

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#### Abstract

The collected bovine milk samples (n= 30) were analyzed for determination of Cadmium and Lead contents. Cadmium and Lead levels were determined using Atomic Absorption Spectrophotometer after wet digestion. The levels of Cadmium in all analyzed samples 30 (100%) were found to be over the permissible limit for Cadmium in liquid milk. The levels of Lead exceeded the permissible limit in 13(72.22%) and 4(33.33%) of analyzed Cow's milk and Buffalo's milk samples, respectively. The mean concentrations of Cadmium in examined cow's milk samples (n=18) were  $0.3067 \pm 0.0086$  (mg/L) and  $0.3084 \pm 0.0087$  (mg/L) for the examined buffalo's milk samples (n=12), while the mean concentrations of Lead were  $0.0934 \pm 0.0188$  (mg/L) for the examined cow's milk and  $0.05878 \pm 0.0178$ (mg/L) for examined buffaloe's milk. Estimated daily intake of Cadmium was found above the permissible limits in all examined milk. Also, the estimated daily intake of Lead was found above the permissible limits in examined cow's milk, while estimated daily intake of Lead in examined buffalo's milk was found under permissible limit. Public health significance and hazardous of these heavy metals were discussed.

#### Article Info

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#### Keywords

Cadmium
Heavy metals
Lead
Milk contamination

#### Introduction

Milk is a primary source of nutrients in diets all around the world (Buldini et al., 2001). Lactating cow's may possibly be exposed to high quantity of toxic elements such as Lead and Cadmium (Jeng et al., 1994). The cattle feed and environment might be the source of heavy metals like Lead and Cadmium in milk and capable of causing serious health problems among the people consuming milk with such contaminants (Monkiewicz et al., 1999). Heavy metals are non-biodegradable in nature and become accumulated in the food chains via bio-transformation, bio-accumulation

and bio-magnifications (Aslam et al., 2011). The presence of heavy metals in milk and dairy products may be attributed to the contamination of the milk during lactation, through feeding stuff, pesticides and water (Carl, 1991). Lead and Cadmium heavy metals are non-essential and have no biological role (Khan et al., 2013). However, at high concentrations, they can cause toxicity living organisms (Li to 2005). Accumulation of lead produces damaging effects in the hematopoetical, hematic, renal, gastrointestinal systems (Correia et al., 2000). Due to the growing environmental pollution it is necessary to determine and monitor the levels of heavy metals in milk, because they

can significantly influence human health (Licata et al., 2004). The aim of this investigation was to determine the levels of toxic heavy metals Cadmium and Lead in raw cow's and buffalo's milk in El-Behera region, Egypt.

#### Materials and methods

#### Milk samples

A total of 30 raw milk samples (18 cow's milk and 12 buffalo's milk samples) were collected from dairy shops at El-Behera Governorate, Egypt.

Storch's test for detection of heat treated milk samples (Lampert, 1975) was applied for all samples and proofed to be raw milk samples. Atomic absorption spectrophotometer (Licata et al., 2004) was used to determine the lead and cadmium content.

#### Reagents

All dissolutions were prepared using analytically pure reagents: Nitric acid and Perchloric acids (Merck, Germany). The detailed procedure followed were from Lajunen (1980) and Haswell (1991)

#### **Digestion procedures**

Prior to digestion, each sample was carefully shaken in its own packing at 40°C for 30 minutes on the Rotaterm agitation block.

Wet digestion: One milliliter of homogeneous sample was placed in a 20 mL tube, mineralized by addition of 3 mL HNO3-HClO4 (4: 1) mixture and heated to 120°C for 65 minutes in a thermostat-controlled digestion block. After cooling, the resulting solution was diluted to 25 mL with demineralized water. The apparatus has

also digital absorbance and concentration read out capable of operating at the following wave length (nm): 283.3 and 228.9, for Lead and Cadmium, respectively (Agemian et al., 1980) and Perkin–Elmer, 1980).

#### **Data analysis**

The concentrations were expressed as mean  $\pm$ SEM and minimum/ maximum values. The data was analyzed using SPSS version 16.

### Estimation of daily intake of Cadmium and Lead in examined milk samples

To evaluate the daily intake, mean concentration of selected heavy metals (Cadmium and Lead) in bovine milk category were multiplied by the milk consumption rate and divided by the body weight (75 kg) of the adult resident consumer.

#### **Results and discussion**

Cadmium level (Table 1) showed that 18 (100%) and 12 (100%) of examined cow's milk and Buffaloe's milk were contaminated with respectively. The mean concentration of Cadmium in raw cow's milk sample was  $0.3067 \pm 0.0086$  mg/L with a maximum concentration of 0.3135 mg/L and minimum concentration of 0.2993 mg/L (Table 1), while the mean concentration of Cadmium in raw milk samples from buffaloes was  $0.3084 \pm 0.0087$  mg/L with a maximum value of 0.3118 mg/L and a minimum value of 0.3022 mg/L. Results presented in Table 4 showed that the levels of Cadmium in all analyzed raw cow's milk samples 18(100%) and Buffaloe's milk samples 12(100%) were found over the permissible limit of (0.05 mg/kg) Cadmium in liquid milk determined by Egyptian standards (2012).

Table 1. Statistical analytical results of concentration of Cadmium (mg/L) in examined milk samples (n=30).

Mills comple	Number	Positi	ve samples	— Minimum	Maximum	Mean ± SEM	
Milk sample	Number	No.	%	Willimum	Maximum	Mean ± SEM	
Cow's milk	18	18	100	0.2993	0.3135	$0.3067 \pm 0.0086$	
Buffaloes milk	12	12	100	0.3022	0.3118	$0.3084 \pm 0.0087$	

These results were higher than permissible limits recommended by the Egyptian standards (2012) set it at 0.05mg/g for Cadmium in milk. Also, the obtained Cadmium level in this study was higher than the permissible limit of (0.010 mg/L) for Cadmium in cow milk determined by Commission regulation (EC)

No.1881/2006. Lower results for Cadmium were recorded by Enb et al. (2009), Elsayed et al. (2011), El-Bassiony et al. (2016), Gidikova et al. (2016) and Tunegova et al. (2016). Higher results were obtained by Abdulkhalequ et al. (2012) and Gasmalla et al. (2013). Milk and dairy products become contaminated with

heavy metals either through food stuff and water or through manufacturing and packaging processes (Ayar et al., 2009).

Table 2 showed the level of Lead in examined raw cow milk (n=18) was ranged from 0.0073 to 0.3407 mg/L with mean value of  $0.0934 \pm 0.0188$  mg/L, while the mean value of Lead in examined raw buffaloes milk

(n=12) was  $0.05878 \pm 0.01784$  mg/L with a maximum concentration of 0.2358 mg/L and minimum concentration of 0.0145 mg/L. Results presented in Table 4 showed that there were 4 (33.33%) of examined buffaloe's milk samples and 13 (72.22%) of examined cow's milk samples exceeded the permissible level (0.10 mg/kg) for Lead in milk, determined by Egyptian standards (2012).

**Table 2.** Statistical analytical results of concentration of Lead (mg/L) in examined milk samples (n=30).

Mills comple	Number	Positive samples		— Minimum	Maximum	Mean ± SEM	
Milk sample		No.	%	Willillium	Maximum	Mean ± SEM	
Cow's milk	18	18	100	0.0073	0.3407	$0.0934 \pm 0.0188$	
Buffaloes milk	12	12	100	0.0145	0.2358	$0.05878 \pm 0.0178$	

This value is above the permissible limits of (0.020 mg/L) determined by Commission regulation (EC) No.1881/2006. According to Codex Alimentarius (2012) the permissible value of Lead in milk and secondary milk products are 0.02 mg/kg. Higher results were recorded by Enb et al., (2009) for Lead in buffaloe's milk and cow's milk in Giza from Egypt. Higher results for lead in raw cow's milk were reported by Gasmalla et al. (2013) and El-Bassiony et al. (2016). Lower results for Cadmium (1.24  $\mu$ g/kg) and Lead (3.43  $\mu$ g/kg) in cow's milk were

reported by Elatrash and Atoweir (2014) in Libya. Lower results for Lead in cow's milk were recorded by Gidikova et al. (2016) and Muhib et al. (2016). In this study higher levels of heavy metals in cow's and buffalo's milk may be attributed to the high contamination of animal feed and water by such pollutants and could be excreted into milk at various levels. One of the most important sources of Lead contamination in milk is water, especially in more contaminated areas (Codex Alimentarius Commission, 2003).

**Table 3.** Interspecies comparative mean  $\pm$  SEM of Cadmium and Lead (mg/L) in the examined milk samples of cow and buffaloes.

Residue	Cow's milk	Buffalo's milk
Cadmium	$0.3067 \pm 0.0086$	$0.3084 \pm 0.0087$
Lead	$0.0934 \pm 0.0188$	$0.05878 \pm 0.0178$

**Table 4.** Frequency distribution of Cadmium and Lead heavy metals in examined raw cow's and raw Buffalo's milk samples (n=30).

Heavy	Permissible limit Egyptian	Samples within permissible limit			Samples exceeded permissible limit		
metal	Standards 2012	Milk sample	No.	%	Milk sample	No.	%
Cadmium	0.05 mg/kg	Buffalo	0	0	Buffalo	12	100
		Cow	0	0	Cow	18	100
Lead	0.10 mg/kg	Buffalo	8	66.66	Buffalo	4	33.33
		Cow	5	27.77	Cow	13	72.22

The presence of heavy metals as Lead and Cadmium even in low concentrations leads to metabolic disorders with extremely serious consequences and causing serious problems as it causes many health problems such as weakness, cancer and also affects the kidneys (Licata et al., 2004).

The heavy metal contents vary widely due to many factors such as differences between species, characteristics of the manufacturing practices and possible contamination coming from the equipment during the process (Caggiano et al., 2005).

The interspecies difference in the residual concentration (mg/L) of heavy metals (Cadmium and Lead) in the milk of cow and buffalo collected from local markets has been given in Table 3. The results showed that in the raw milk of cow mean ± SEM (mg/L) of Lead 0.0934  $\pm$  0.0188 was higher than mean  $\pm$ SEM values (mg/L) of Lead 0.05878  $\pm$  0.0178 in raw buffaloes milk, while the mean ±SEM values (mg/L) of Cadmium  $0.3084 \pm 0.0087$  obtained in the raw milk of buffaloes was higher than the mean ± SEM values for Cadmium  $0.3067 \pm 0.0086$  in raw cow's milk.

Assuming that a value of 200 (ml/day) of raw bovine milk consumption in Egypt. The estimated daily intake of Cadmium and Lead heavy metals were determined and recorded in Table 5, the estimated daily intake of Cadmium in examined raw cow's and Buffaloes milk was 0.8179 (mg/day) and 0.8224 (mg/day) respectively, which was higher than the permissible limits 0.046

(mg/day) for Cadmium (JECFA, 2003), while the estimated daily intake of Lead was 0.2491 (mg/day) for cow's milk which exceeded the permissible limits 0.21 (mg/day) for Lead (JECFA, 2003), while the estimated daily intake for buffalo's milk was 0.1567 (mg/day) which is below the permissible limits 0.21(mg/day) for Lead in raw bovine milk.

**Table 5.** The estimated daily intakes of (Cadmium and Lead) metals from consumption of 200 ml milk of the examined raw milk of cow's and Buffalo's.

Heavy metal	Sample	Estimated daily intake (mg/day)	Recommended permissible values (mg/day)	Reference
Cadmium	Cow's milk	0.8179	0.046 (mg/day)	JECFA (2003)
	Buffaloe milk	0.8224		
Lead	Cow's milk	0.2491	0.21(mg/day)	
	Buffaloe milk	0.1567		

Thus the daily intake rate of heavy metal hazard quotients and carcinogenic risk might be considered as exponentially increasing trend with the increasing rate of milk production. It can be recommended that proper monitoring of cattle feed quality as well as the techniques of milk processing should be carefully considered for the public health safety in Egypt.

#### **Conclusion**

From the obtained results of this study it could be concluded that all examined raw milk samples contained high concentrations of Cadmium exceeded the permissible limit. Lead levels in a high percent of examined raw cow's milk samples in El-Behera region, are a cause of concern. Risk of heavy metal contamination was higher in raw cow's milk. However, the number of analyzed heavy metals and sample size were limited in this study and further studies are necessary to evaluate of "toxic" heavy metals on a greater number of milk samples from various regions of Egypt.

#### **Conflict of interest statement**

Authors declare that they have no conflict of interest.

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