International Journal of Dairy Science, 2011
ISSN 1811-9743 / DOI: 10.3923/ijds.2011.

A Survey of Selected Essential and Toxic Metals in Milk in Different Regions of Egypt using ICP-AES

[1]Elham M. Elsayed, [1]Ahmed M. Hamed, [1]Sanna M. Badran and [2]Amr A. Mostafa
[1]Department of Dairy Science,
[2]Department of Biochemistry, Faculty of Agriculture, Cairo University, Egypt

Corresponding Author: Elham M. Elsayed, Department of Dairy Science, Faculty of Agriculture, Cairo University, Egypt

ABSTARCT

The aim of this study was to assess the content of selected essential metals (Fe, Zn, Cu and Cr) and some toxic metals (Pb and Cd) in raw cow's milk collected from different Egyptian areas and during different periods (Jan.-Feb., May-Jun. and Sep.-Oct.) of one year 2009. Seventy two samples of raw milk were analyzed using ICP-AES for the quantitative determination of metals in this matrix. As regards essential metals, the highest average concentrations are those of Fe (3.860 mg kg^{-1}) and Zn (1.209 mg kg^{-1}), while the lowest average concentrations are (0.945 mg kg^{-1}) and (0.238 mg kg) for Cu and Cr, respectively. With regard to toxic metals the average concentrations for Pb and Cd were 0.327 mg kg^{-1} and 0.007, respectively. The highest trace metals accumulation levels for Cd, Cr, Fe and Cu are found in milk samples collected from Shubra while samples from Menofia and Tanash are high in Pb and Zn, respectively. According to the classification on the basis of their metal contents, the highest trace metal accumulation levels were during Jan.-Feb. for Cd, Cr and Zn. However, the period of May-June the content of Pb, Cu and Fe were the highest. Whereas samples during Sept.-Oct. is characterized by their lowest metal content. The obtained results showed significantly differences at $p \le 0.05$ in levels of trace elements determined in milk samples except Cd and Cr which may be attributed to seasonal variation or geographical station.

Key words: Essential and toxis metals, ICP-AES, accumulation levels, trace metal

INTRODUCTION

Milk is a primary source of nutrients in diets all around the world (Buldini *et al.*, 2002). Attention has been focused on milk as it is nearly almost the perfect single foodstuff specially for children, however lactating cattle may exposed to high quantities of toxic metals in the environment by air, water and ingestion of polluted feeds. Fortunately, these animals act as a very efficient biological filter against heavy metal contamination. Where, it is valid when the animals are grazing near motor ways and road with heavy car traffic (Carl, 1991).

In the last few years, the contamination of milk is considered as one of tha main dangerous aspect (Abou Ayana *et al.*, 2011).

Duruibe *et al.* (2007) suggested that biotoxic effects of heavy metals depend upon the concentrations and oxidation states of heavy metals, kind of sources and mode of deposition. Severe exposure of Cd may result in pulmonary effects such as emphysema, bronchiolitis and alveolitis. Renal effects may also result due to subchronic inhalation of Cd (European Union, 2002;

Young, 2005). Pb toxicity causes reduction in the haemoglobin synthesis, disturbance in the functioning of kidney, joints, reproductive and cardiovascular systems and chronic damage to the central and peripheral nervous systems (Ogwuegbu and Muhanga, 2005). Higher concentration of Zn can cause impairment of growth and reproduction (Nolan, 1983).

Metal toxicity creates multisystem dysfunction, which appears to be mediated primarily through mitochondrial damage from glutathione depletion (Neustacdt *et al.*, 2007).

Typically, a food product can be correlated with its geographical localisation, the quality of raw material and the production techniques (Brescia *et al.*, 2005). By their spread speed in biosphere and increasing concentrations heavy metals are considered to be among the most hazardous pollutants (Staniskiene and Garaleviciene, 2004).

Inorganic or aggregated forms of chemical substances (metalloids, heavy metals etc.) in feed and food represent a severe risk for their long term toxicological effects. Heavy metals are widely dispersed in the environment. The toxicity induced by excessive levels of some of these elements, such as chromium (Cr), cadmium (Cd), lead (Pb) and mercury (Hg), are well known (Llobet *et al.*, 2003).

The metals, namely Cu and Zn, are essential micro nutrients and have a variety of biochemical functions in all living organisms. While Cu and Zn are essential, they can be toxic when taken in excess; both toxicity and necessity vary from elementto-element (Tripathi *et al.*, 1999).

The concentrations of dangerous elements in the environment grow with the increase of urban, agricultural and industrial emissions. The almost ubiquitous presence of some metal pollutants, especially Cd and Pb, facilitates their entry into the food chain and thus increases the possibility of them having toxic effects on humans and animals (Licata *et al.*, 2004). The present study was aimed to elucidate any possible relationship among areas and time of year of samples collection and levels of essential and toxic metals in raw cow's milk.

MATERIALS AND METHODS

Sample collection: Seventy two 500 mL samples of raw milk were collected in different periods of one year (2009) (Jan.-Feb., May-June and Sep.-Oct.) from farms the 7 areas named in Table 1. These areas are characterized by different elevations and ecological features.The collected samples were homogenized and packed in polyethylene bags and stored below -20°C prior to analysis.

Reagents and solutions: All reagents were of analytical reagent grade unless otherwise stated. Double-deionized water was used for all dilutions. HNO_3 and H_2O_2 were of suprapure quality. Samples (0.5 g) were digested with 7 mL of HNO_3 (65%) and 1 mL of H_2O_2 (30%) in Advanced Microwave Digestion for 31 min and diluted to 100 mL with deionized water (Birghila *et al.*, 2008).

Apparatus and condition: A blank digest was carried out in the same way (digestion conditions for microwave system were: 2 min for 250 W, 2 min for 0 W, 6 min for 250 W,5 min for 400 W, 8 min for 550 W, vent: 8 min, respectively). This procedure was preferred because of its higher accuracy with respect to both time and recovery values. Analysis of trace elements in milk samples was by inductively coupled plasma atomic emission spectrometry (ICP-AES) using iCAP 6000 Series; Thermo Scientific.

Statistical analysis: A randomize complete block design with two factors was used for analysis all data with three replications. The treatment means were compared by least significant differences (L S D) was used for mean separation at $p \leq 0.05$ test as given by Snedecor and Cochran (1976).

RESULTS AND DISCUSSION

The concentration levels of the essential metals (Fe, Zn, Cu and Cr) and the toxic metals (Cd and Pb) were measured in the seven different areas during different period in the same year are given in Table 1. The results obtained show considerable differences among the levels of trace elements in raw milk samples collected from different areas of the same seasons. Even the areas and seasons have an influence on the levels of heavy metals pollution in raw milk. Statistically significant higher differences were detected in the samples of Fe, Zn,Cu and Pb values while no statistically significant differences were observed in the samples of Cr and Cd. The highest trace metals accumulation levels for Cd, Cr and Fe are found in milk samples collected from Shubra, during the period of Jan.-Feb. for Cd and Cr, May-June for Fe while samples from Menofia and Tanash had high levels in Pb, Cu and Zn, respectively during the period of May-Jun. but Zn for Pb and Cu were the highest during Jan-Feb. The large variance and some high levels of contamination could be attributed to the environmental condition as they are chosen for as indicated regions seem to be more industrialized than the other regions.

Figure 1 shows the changes in essential metals (Fe, Zn, Cu and Cr) (mg kg^{-1}) in milk samples during different periods of the same year. The iron concentration of the samples varied from 3.130 to 5.093 mg kg^{-1} as shown in Table 1. The highest average values were found during May-June. Shubra displayed the highest average values of areas (Fig. 1a) that agree with El-Gendy *et al.* (2007), who reported that sample collected during warm or hot months contained higher level of Fe than that of cold months. While, these values were higher than Florea *et al.* (2006), who recorded that the concentration level of Iron content in milk samples were 38.21 µg 100 g. Iron can represent a problem in dairy technology because of its catalytic effect on oxidation of lipids with development of unpleasant smell, bounding preferably proteins and membrane lipoproteins of milk fatty globule (Lant *et al.*, 2006).

The average zinc concentrations ranged from 0.695 to 1.826 mg kg (Table 1). Even areas and seasons had an influence and the levels of Zn values with significant differences. The highest polluted levels was found in Jan.-Feb. period in Tanash and Helwan, respectively. Also it is noticeable that the highest level was found during winter months and lowest one was for spring sample (Fig. 1b). These values were lower than those reported by Abou-Arab (1991), who recorded the variation in Zinc content in milk samples collected from great Cairo over one year ranged from 4.1 to 5.45 ppm with an average 4.25 ppm. Also it is noticeable that the highest level was found

Table 1: Content of essential and toxic metals (mg kg^{-1}) in milk samples from different areas

	Essential metals				Toxic metals	
Location	Fe	Zn	Cu	Cr	Pb	Cd
Helwan†	4.021±1.516^{ab}	1.600±0.545^{abc}	0.908±0.047^{ab}	0.295±0.169^{a}	0.278±0.009^{bc}	0.006±0.003^{a}
Tanash†	4.029±1.121^{ab}	1.826±0.425^{a}	1.234±0.165^{a}	0.178±0.020^{a}	0.412±0.020^{ab}	0.005±0.001^{a}
Shubra†	5.093±1.297^{a}	1.677±0.313^{ab}	1.194±0.333^{a}	0.350±0.107^{a}	0.577±0.028^{a}	0.018±0.002^{a}
Menofia	3.613±2.286^{b}	0.824±0.145^{cd}	0.614±0.355^{b}	0.259±0.132^{a}	0.614±0.202^{bc}	0.004±0.002^{a}
Mansoura	3.130±0.620^{b}	0.695±0.323^{d}	0.674±0.476^{b}	0.123±0.069^{a}	0.135±0.069^{c}	0.004±0.002^{a}
Awseem	3.415±1.317^{b}	0.807±0.259^{d}	0.821±0.278^{ab}	0.130±0.017^{a}	0.138±0.070^{c}	0.005±0.001^{a}
Gharbia	3.722±1.437^{b}	1.037±0.735^{bcd}	1.173 ±0.376^{a}	0.334 ±0.045^{a}	0.141±0.071^{c}	0.013±0.002^{a}
Mean	3.860±1.370	1.209±0.392	0.945±0.332	0.238±0.121	0.327± 0.124	0.007±0.004
MRL*	0.037	0.328	0.01		0.02	0.0026

*According to IDF Standard (1979). †Industrial areas

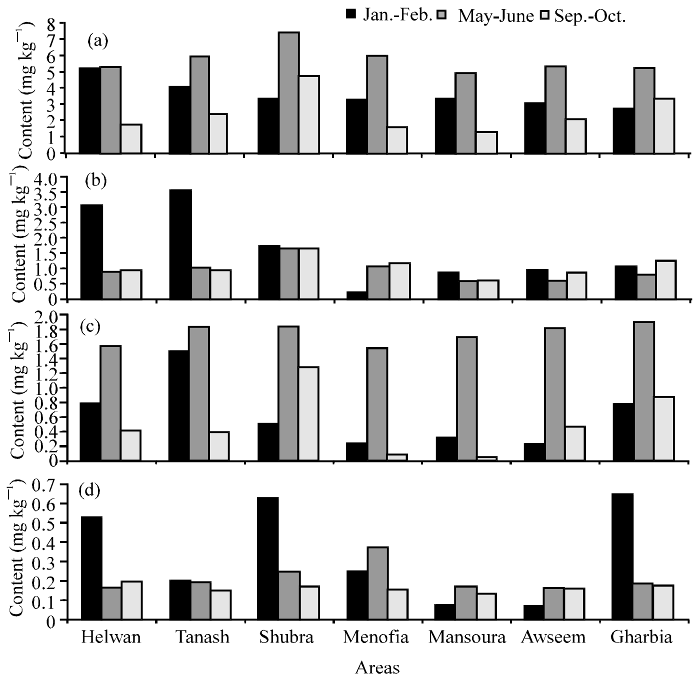

Fig. 1: Content of essential metals (a) Fe, (b) Zn, (c) Cu, and (d)Cr (mg kg^{-1}) in raw milk samples from different areas collected during Jan-.Feb., May-June and Sept.-Oct.

during winter months and lowest one was for spring sample. However, this disagree with Crrek *et al.* (1994), who found that zinc concentration in investigated samples of cows milk were slightly higher during summer than winter. On the other hand, these values were lower than the values recorded by Morsy (1991) for cow's milk (5.4 ppm). The maximum value were higher than Birghila *et al.* (2008) who reported that Zinc values were 0.98 ppm. However, these values were higher than IDF Standard (1979) which recommended that MRL for Zinc in milk is 0.328 mg kg^{-1}. A possible source of contamination of zinc in milk is the use of metal cans and processing equipment (Jarrett, 1979). Depending on the period of the year and the region collected.

The average copper concentration varied between 0.614-1.234 mg kg^{-1} in the samples (Table 1). It noticeable that the season had most effect than areas on polluted level by Cu hence the highest concentration was found during May-June period and the highest areas was Tanash follwed by Shubra and Gharbia (Fig.1 c). Both areas and seasons had statistically significant differences in the samples of Cu values levels. these values were higher than IDF standard (1979) which recommended that MRL for Copper in milk is 0.01 mg kg^{-1}. Possible contamination of milk with copper can occur from animal feed, high copper content of water also from copper bearing and copper alloys used in equipment (Mitchell, 1981).

Table 1 shows the average chromium content in the samples was 0.123-0.350 mg kg^{-1}. The results obtained show that even areas and seasons have no significant differences effect on Cr values. The lowest and highest Chromium concentrations were found in Mansoura and Shubra, respectively and the highest concentration was found during Jan.-Feb. period (Fig. 1d). These values were higher than recorded by (Coni *et al.*, 1996), who found that the average content of chromium in raw milk was (0.014 μg g^{-1} dry weight). And also were higher than Qin *et al.* (2009), who reported that Cr content were significantly higher in Chinese commercial milks

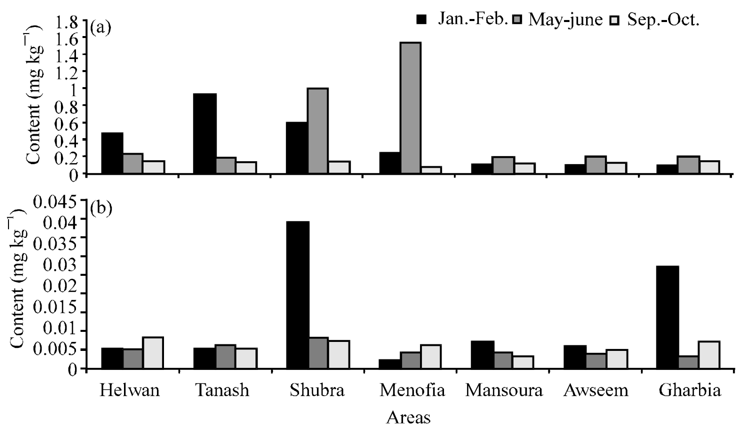

Fig. 2: Content of Toxic metals (a) Pb and (b) Cd (mg kg^{-1}) in raw milk samples from different areas collected during Jan.-Feb., May-June and Sept-Oct.

(0.17±0.05 mg kg^{-1}), than in Japanese commercial milks (0.09±0.03 mg kg^{-1}). The major possible source of chromium in dairy products is the contamination from used refrigeration brines and detergents containing compounds of chromium (Jarrett, 1979).

Regarding the toxic elements, the average lead content of samples was between 0.135-0.614 mg kg^{-1} (Table 1). Even the areas and seasons had significant differences effect on Pb concentration. The lowest and highest lead concentrations were found in Mansoura and Menofia, respectively and the highest concentration of it was found in May-June period. And the lowest was during Sep.-Oct. (Fig. 2a). These results agree with Gorska and Litwinczuk (1996), who found that the highest concentration of Pb in milk samples from Podlasie region Poland, was in March and the lowest was in December. In contrast, El-Awamry (1994) found that the milk samples taken during the winter months contained higher levels of Pb than that collected during summer months. The high level of lead in milk may result from industrial air pollution in these regions. These results were higher than Tajkarimi *et al.* (2008), who recorded that the mean level of lead content was 7.9 ng mL with a range from1to 46 ng mL^{-1}. These results were higher than MRL recommended by the CE Regulation no. 2001/466(The European Communities, 2001) establishes a limit for Pb in milk (MRL = 0.02 mg kg^{-1} w.w).

For cadmium the average varied between 0.004-0.018 mg kg in the samples (Table 1). Both the season and areas had no significant differences on Cd levels. The lowest and highest cadmium concentrations were found in Menofia and Shubra, respectively. And the highest concentration was found during Jan.-Feb. period (Fig. 2 b). These results were higher than Lant *et al.* (2006), who found that cadmium was absent in milk. The IDF Standard (1977) recommended that the level of 0.006-mg kg^{-1} cadmium is regarded as normal for unpolluted milk while the level of 0.015 mg kg^{-1} is considered as polluted. The contamination of milk with cadmium might be du to use of cadmium in the manufacture of pesticides and in some fertilizers and referring of zinc and other metals. Generally the most likely sources of contamination of milk by the buffalo and cow ingesting cadmium from fodder and water.

For an average adult (60 kg b.wt.), the Provisional Tolerable Daily Intakes (PTDI) for lead, iron, copper and zinc are 214 µg, 48 mg, 3 mg and 60 mg, respectively (Yuzbasi *et al.*, 2003).

CONCLUSION

According to the result obtained it could be observed that three sets of samples from Shubra, Menofia and Tanash seemed to show a higher level of contamination. This may be attributed to seasonal variation and or geographical station. Both seasons and areas have significant higher differences at $p \leq 0.05$ influence on the levels of heavy metals (Fe, Zn, Cu and Pb) pollution in raw milk samples except Cr and Cd.

REFERENCES

Abou-Arab, A.A.K., 1991. Microbiological and compositional quality of dairy products in relation to some pollutants. M.Sc. Thesis, Ain-Shams University.

Abou-Ayana, I.A.A., A.A. Gama-El-Deen and M.A. E-Metwally, 2011. Behaviour of certain Lactic Acid bacteria in the presence of pesticides residues. Int. J. Dairy Sci., 6: 44-57.

Birghila, S., S. Dobrinas, G. Stanciu and A. Soceanu, 2008. Determination of major and minor elements in milk through ICP-AES. Environ. Eng. Manage. J., 7: 805-808.

Brescia, M.A., M. Monfreda, A. Buccolieri and C. Carrino, 2005. Characterisation of the geographical origin of buffalo milk and mozzarella cheese by means of analytical and spectroscopic determinations. Food Chem., 89: 139-147.

Buldini, P.L., R. Ricci and J.L. Sharma, 2002. Recent applications of sample preparation techniques in food analysis. J. Chromatogr. A, 975: 47-70.

Carl, M., 1991. Heavy metals and other trace elements. Monograph on residues and contaminants in milk and milk products. International Dairy Federation Belgium, Special Issue 9101, pp: 112-119.

Coni, E., A. Bocca, P. Coppolelli, S. Caroli, C. Cavallucci and M.T. Marinucci, 1996. Minor and trace element content in sheep and goat milk and dairy products. Food Chem., 57: 253-260.

Crrek, J., V. Rehout, F. Hajic, K. Kosvanec and M. Soch, 1994. Seasonal variation in levels of microelements in cow milk. Zootechmicka Kada, 11: 49-60.

Duruibe, J.O., M.O.C. Ogwuegbu and J.N. Egwurugwu, 2007. Heavy metal pollution and human biotoxic effects. Int. J. Phys. Sci., 2: 112-118.

El-Awamry, Z.K., 1994. Reduction of the adverse effects of some chemical pollutant of milk in Egypt. M.Sc. Thesis, Ain Shams University, Cairo, Egypt.

El-Gendy, S.M., M.A. Mohran, N. Hanafy and T.H. Mohamed, 2007. Studies on chemical pollutants of milk produced in Assiut vicinity. Proceeding of the 10th Egyptian Conference for Dairy Science and Technology, Nov. 19-21, Cairo, Egypt, pp: 121-133.

European Union, 2002. Heavy metals in wastes. Final Report, February 2002. http://ec.europa.eu/environment/waste/studies/pdf/heavy_metalsreport.pdf.

Florea, T., S.O. Huszti and G.M. Costin, 2006. Heavy metal contaminants in milk and cheese. The annals of the university Dunarea de jose of galati Fascicl VI- food technology. pp: 62-32. http://www.ann.ugal.ro/tpa/Annals%2006%20papers/05%20Full%20paper%20Florea.pdf

IDF Standard, 1977. Metal contaminations in milk and milk products. International dairy federation Bulletin. Document No. A.Doe 37.

Jarrett, W.D., 1979. A review of the important trace elements in dairy review products. Aust. J. Dairy Technol., 34: 28-34.

Lant, A., G. Lomolino, M. Cagnin and P. Spettoli, 2006. Content and chrarchterisation of minerals in milk and in Crescenza and Squaacquerne italian fresh cheese by ICP-OES. Food Control., 17: 229-233.

Licata, P., D. Trombetta, M. Cristani, F. Giofre, D. Martino, M. Calo and F. Naccari, 2004. Levels of toxic and essential metals in samples of bovine milk from various dairy farms in Calabria, Italy. Environ. Int., 30: 1-6.

Llobet, J.M., J.L. Domingo, A. Bocio, A. Casas, A. Teixido and L. Muller, 2003. Human exposure to dioxins through the diet in Catalonia, Spain: Carcinogenic and non-carcinogenic risks. Chemosphere, 50: 1193-1200.

Mitchell, G.E., 1981. Trace metal level in Queensland dairy products. Aust. J. Dairy Technol., 6: 70-73.

Morsy, A.S., 1991. Studies on pesticides and heavy metals residue in milk. M.Sc. Thesis, Cairo University.

Neustadt, J. and S. Pieczenik, 2007. Heavy-metal toxicity-with emphasis on mercury. Integrative Med., 6: 26-32.

Nolan, K.R., 1983. Copper toxicity syndrome. J. Orthomol. Psychiatry, 12: 270-282.

Ogwuegbu, M.O.C. and W. Muhanga, 2005. Investigation of lead concentration in the blood of people in the copper belt province of Zambia. J. Environ., 1: 66-75.

Qin, L.Q., X.P. Wang, W. Li, X. Tong and W.J. Tong, 2009. The minerals and heavy metals in cow's milk from China and Japan. J. Health Sci., 55: 300-305.

Snedecor, G.A. and W.G. Cochran, 1976. Statistical Method. Iowa State University Press, Ames.

Staniskiene, B. and D. Garaleviciene, 2004. The amount of heavy metals in fish meat and bones. Vet. Zootech., 26: 46-52.

Tajkarimi, M., M.A. Faghih, H. Poursoltani, A.S. Nejad, A.A. Motallebi and H. Mahdavi, 2008. Lead residue levels in raw milk from different regions of Iran. Food Chem., 19: 495-498.

The European Communities, 2001. Commission regulation No. 2001/466 March 8, 2001: Setting maximum levels for certain contaminants in foodstuffs. Official J. Eur. Commun., L77: 1-13.

Tripathi, R.M., R. Raghunath, V.N. Sastry and T.M. Krishnamoorthy, 1999. Daily intake of heavy metals by infants through milk and milk products. Sci. Total Environ., 227: 229-235.

Young, R.A., 2005. Toxicity profiles: Toxicity summary for cadmium. Risk Assessment Information System. University of Tennessee.

Yuzbasi, N., E. Sezgin, M. Yildirim and Z. Yildirim, 2003. Survey of lead, cadmium, iron, copper and zinc in Kasar cheese. Food Chem., 20: 464-469.