ISSN 2320-5407 International Journal of Advanced Research (2015), Volume 3, Issue 3, 982-993

ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

Comparative Study of Zinc, Manganese and Calcium in Commercially Available Milk Samples

Humaira Bano[1*], Azhar Mahmood[2] and Noor Ul Ain[1]

1.Department of Chemistry, University of Karachi, Karachi-75270, Pakistan

2.National University of Sciences and Technology, Islamabad, Pakistan

Manuscript Info

Manuscript History:

Received: 25 January 2015
Final Accepted: 22 February 2015
Published Online: March 2015

Key words:

Microwave digestion, atomic absorption spectrometry

Corresponding Author

Humaira Bano

Abstract

Aim of the present study is to determine the concentration of metals, namely Zn, Mn, and Ca in different dry and liquid milk samples. All samples were collected in October 2014 from different markets of Karachi, Pakistan. Microwave digestion system was used for mineralization of samples. Zn and Mn were analyzed by flame atomic absorption spectrometry and Ca was analyzed by volumetric method of complexometric titration. The concentration of Zn, Mn and Ca in liquid milk samples were found to be (1.96 − 3 .640), (0.37− 0.52) and (195 − 1528) ppm respectively while in dry milk samples (23.22 − 30.5137), (7.67 − 11.6) and (328 − 336) ppm respectively. In liquid milk samples, the highest concentration of Zn, Mn and Ca were found in WM2 (3.64 ppm), WM6 (0.52 ppm) and WM2 (1528 ppm) respectively. In dry milk samples, the highest concentration of Zn, Mn and Ca were found in DM3 (30.5137 ± 0.0057) ppm, DM4 (11.6 ppm) and DM2 (336 ppm) respectively. The results of milk samples were compared to the limits, set by International Dairy Federation. Quality of liquid and dry milk samples according to International Dairy Federation was also determined. Concentration of Zn in most of the liquid milk sample was in agreement and some were below to the value set by International Dairy Federation. However, in dry milk, all samples exceeded the limit. Value of Mn in liquid as well as in dry milk samples exceeded the recommended value. Furthermore, value of Ca in dry milk samples was below to the limit, on contrary value was within the limit in some of the liquid milk samples while in others, value exceeded the recommended limit.

The study showed that there is wide variation in the concentration of Zn, Mn and Ca between liquid milk samples and dry milk samples. Some of milk samples exceeded the permissible limits. So it is suggested that the concerned authorities should check and improve the level of these essential metals in milk available in markets.

INTRODUCTION

Milk and its products are very common in our food list due to its nutrient value [1]. Milk in various forms is the main or only source of nutrition in infants. In addition, growing children as well as mature adults consume varying quantities of milk [2].

Cow's milk comprise of about 88 percent water, 5 percent carbohydrate and 3.5 percent fat. Nutrient contents of milk vary from species to species [3]. It is known as an excellent source of Ca, it can supply moderate amounts of Mg, smaller amounts of Zn and very small amounts of Fe and Cu [4]. Quality of milk has been studied in various milk samples, including infant formulas, dry milk, raw milk, processed milk, human milk and animal milk. The samples have been selected from different countries including Egypt, Spain, Poland, Saudi Arabia, Bangladesh, Nigeria, Ethiopia, Italy, Brazil, Germany, China and Japan for studies (A. Enb. et al [5], E. M. R. Rodríguez, et al [6], Z. Dobrzañski, et al [7], A. S. Al Khalifa, et al [3], S. M. Farid, et al [1], P. C. Onianwa, et al [8], A. G, Dawd, et al [9], R. Caggiano et al [10], V. A. Soares, et al [11], P. Ostapczuk, et al [12] and L. Qin, et al [13]).

R. M. Tripathi, et al [14] reported the daily intake of heavy metals by infants through milk and its products in India. They found that the daily intake levels of essential elements were significantly lower than the recommended desirable levels. The content of 38 microelements and trace elements in raw milk of cows from Lower Silesia (LS) and Upper Silesia (US) were analyzed by Z. Dobrzañski, et al [7]. Their results showed that the location of cows has significant impact on the content of many microelements and trace elements in milk. E. M. R. Rodríguez, et al [6] determined the mineral concentrations in cow's milk from the Canary Island. They found that the cow's milk sampled in spring and winter presented higher concentrations of Se, Zn and Mg than cow's milk sampled in autumn. Moreover, G. Zurera-Cosano, et al [15] investigated the effect of processing on contents and relationships of mineral contents of milk. They found that, the processes like pasteurization, sterilization and drying, affect the mineral concentration of milk. P. Licata, et al [16] determined the Levels of “toxic” and “essential” metals in samples of bovine milk from various dairy farms in Italy. Furthermore, S. Saracoglu, et al [17] analyzed the trace elements present in Turkey’s baby foods while T. G. Kazi, et al [18] described the assessment of toxic metals (Al, Cd, Ni and Pb) in raw and processed milk samples.
Aim of the present study is to determine the concentration of metals, namely Zn, Mn, and Ca in different dry and liquid milk samples available in Karachi, Pakistan.

Materials and Methods

Washing of Glassware

All glasswares were washed with detergent and water. After being rinsed with de-ionized water three times, the glasswares were soaked in 10 % HNO_3 (v/v) for 24 hours. This solution was discarded and the glasswares were rinsed again three times with de-ionized water and then dried.

Collection of Samples

For the analysis of Zn, Mn and Ca in milk, samples were collected from different markets of Karachi, Pakistan.

Codes of Milk Samples

Codes of dry and liquid milk samples are given in Table 1 and 2 respectively.

Table 1: Detail of Dry Milk Samples

Sample Codes	Types of Milk	Weight Taken (g)
DM1	Branded	0.5287
DM2	Branded	0.5603
DM3	Unbranded	0.5178
DM4	Unbranded	0.5512

Table 2: Detail of Liquid Milk Samples

Sample Codes	Types of Milk	Volume Taken (ml)
WM1	Branded	10.0
WM2	Branded	10.0
WM3	Branded	10.0
WM4	Branded	10.0
WM5	Branded	10.0
WM6	Branded	10.0

Digestion of Milk Samples

Microwave Digestion system (MARS Express. CEM Corporation, USA) was used to eliminate the organic part of milk. All digestion vessels were washed and rinsed with de-ionized water. 0.1 M HNO_3 was filled in the digestion vessels for two hours. After two hours the HNO_3 was discarded and vessels were kept in oven for drying.

0.5 to 0.6 g of dry milk samples /10.0 ml of liquid milk samples were transferred to dried digestion vessels. With the help of pipette 15.0 ml of concentrated HNO_3 and 4.0 ml of concentrated $HClO_4$ were added in the digestion vessels. The digestion vessels were placed in a microwave digestion system. A blank solution was prepared by digesting 15.0 ml of concentrated HNO_3 and 4.0 ml of concentrated $HClO_4$ using the same digestion procedure. Parameters of the microwave digestion system are given in Table 3. All vessels were automatically cooled following the completion of the digestion program. Then the vessels were placed in fuming hood and vessel covers were removed. Transparent solution obtained after digestion was quantitatively transferred to 100.0 ml volumetric flask and volume was made up to the mark with de-ionized water. All the samples were digested in the same manner. Blank consisting of de-ionized water and reagents was subjected to a similar sample preparation and analytical procedure.

Table 3: Parameters of the Microwave Digestion System

Stage	Power		Ramp Time	Pressure	Temperature	Stirring	Hold Time
	Level	%	mm:ss	psi-limit	°C		mm:ss
1	400 W	100	15:00	800	200	off	15:00

Determination of Zn and Mn in Milk Samples by Atomic Absorption Spectrometry

Working standard solutions for Zn and Mn were prepared by appropriate dilution of their respective stock standard solution of 1000 ppm purchased from Fisher Scientific. Concentrations of working standard solutions were 20, 40 and 60 ppm.

Zn and Mn in milk samples were analyzed by using Analyst 700 (Perkin Elmer) atomic absorption spectrometer equipped with hollow-cathode lamp. Standard instrumental conditions are given in Table 4.

Table 4: Standard Instrumental Condition for Atomic Absorption Spectrometer

Element	Wavelength	Slit Width	Lamp Current	Flame Gas	Slit Height
	nm	nm	mA		
Zn	213.9	0.7	25	Air-Acetylene	High
Mn	279.5	0.2	30	Air-Acetylene	High

Determination of Ca in Milk Samples by Volumetric Method of Complexometric Titration

Weight of disodium salt of EDTA was calculated to prepare 0.05 M standard solution in 500.0 ml volumetric flask. 9.3666 g of disodium salt of EDTA was accurately weighed in a vial, transferred to a 500.0 ml volumetric flask and volume of the solution was made up to the mark with distilled water. About 500 ml of about 0.05 M $CaCO_3$ solution was prepared in a beaker. For this purpose, about 1.2-1.4 g of $CaCO_3$ was weighed on a watch glass transferred to a 500 ml beaker, dissolved in about 2 ml of 6 M HCl solution and volume of the solution was made up to mark with distilled water. Burette was washed and rinsed with distilled water then with EDTA solution. EDTA solution was filled in the burette and air bubbles were removed from the jet and initial burette reading was recorded.

25.0 ml of $CaCO_3$ solution was transferred to a conical flask with pipette, then about 15 ml of 1 M NaOH solution and a pinch of calcon indicator were added to it and the content of the conical flask was titrated against EDTA solution until pink solution present in conical flask turned blue. Final burette reading was recorded and difference of final and initial burette reading was calculated. The whole titration was repeated until concordant reading was obtained.

0.5-0.6 g of dry milk samples/10.0 ml of liquid milk samples was weighed accurately and transferred to a conical flask. 20.0 ml of standard 0.05 M EDTA solution was transferred with pipette then about 15 ml of 1 M NaOH solution and pinch of calcon indicator were added to it and the content of the conical flask was titrated against standard $CaCO_3$ solution until pink solution present in the conical flask turned blue. Final burette reading was recorded and difference of final and initial burette reading was calculated. The whole titration was repeated until concordant reading was obtained.

Results and Discussion

The results of the concentration of Zn, Mn and Ca for Dry and Liquid milk samples are presented in Figures 1-6. The mean concentration (or ranges) in dry milk samples were: Zn, (23.22 ± 0.007) to (30.5137 ± 0.0057) ppm; Mn, (7.67 ± 0.0017) to (11.6 ± 0.0058) ppm and Ca, (328 to 336) ppm (Figure 1-3). In dry milk samples, the highest concentration of Zn was found in sample DM3 (30.5137 ± 0.0057) ppm as shown by Figure 1, Mn in DM4 (11.6 ± 0.0058) as shown by Figure 2 and Ca in DM2 (336 ppm) as shown by Figure 3.

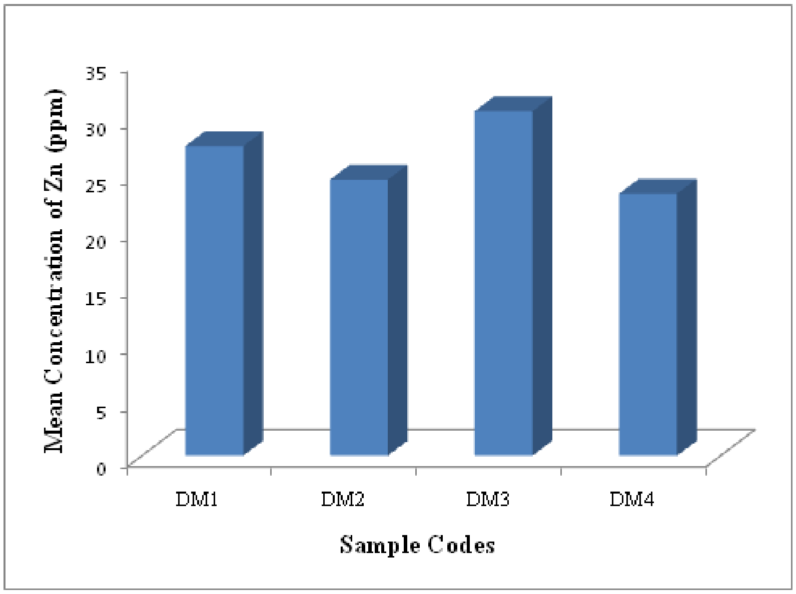

Figure 1: Comparison of Mean Concentration of Zn in Dry Milk Samples

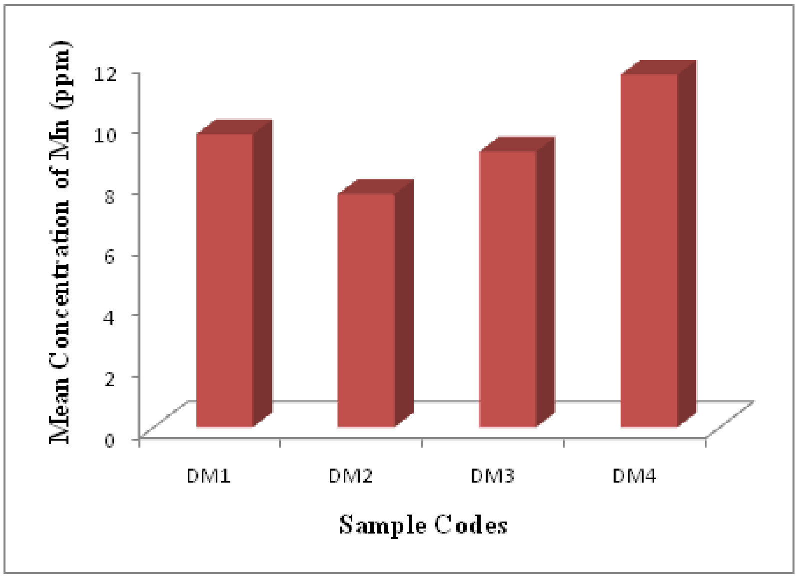

Figure 2: Comparison of Mean Concentration of Mn in Dry Milk Samples

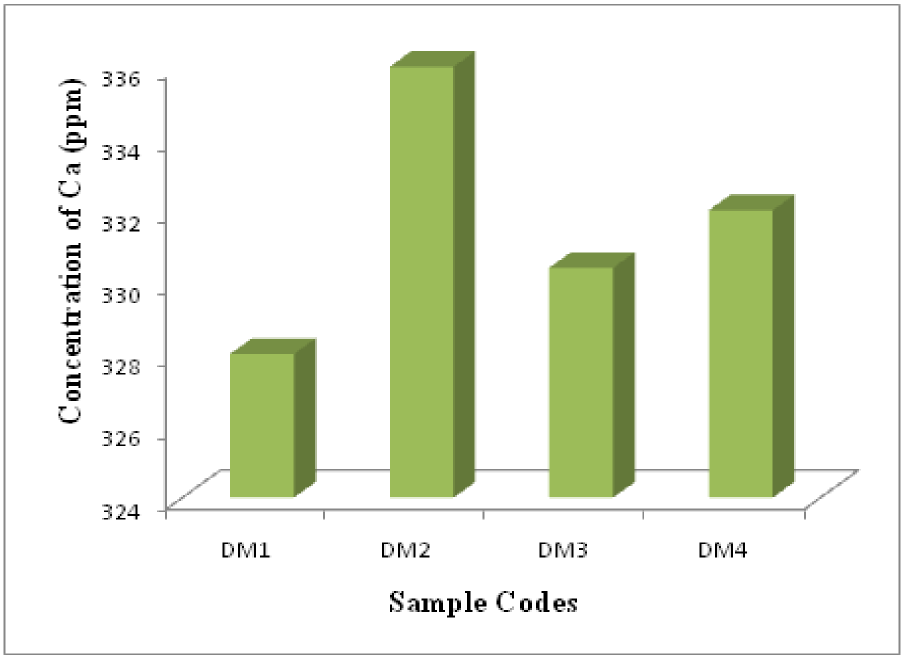

Figure 3: Comparison of Concentration of Ca in Dry Milk Samples

The mean concentration (or ranges) in liquid milk samples were: Zn, (1.96 ± 0.0022) to (3.640 ± 0.0086) ppm; Mn, (0.37 ± 0.0051) to (0.52 ± 0.0112) ppm and Ca (552.4 to 1528) ppm. In liquid milk samples, the highest concentration of Zn was found in sample WM2 (3.64 ± 0.0086) as shown by Figure 4, Mn in WM6 (0.52 ± 0.0112) ppm as shown by Figure 5 and Ca in WM2 1528 ppm as shown by Figure 6. The concentration of metals (Zn, Mn and Ca) in liquid milk samples is associated to water contents of liquid samples [9].

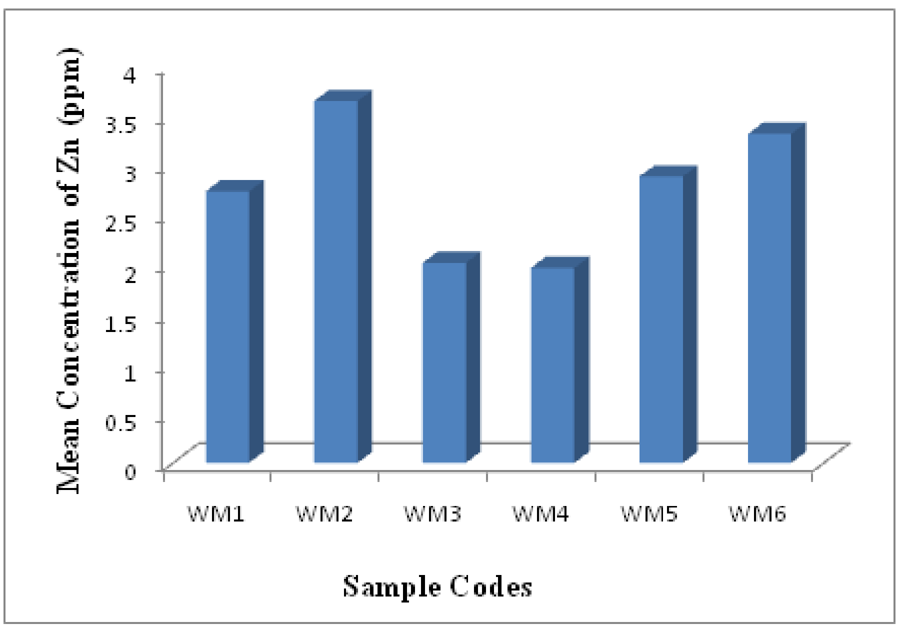

Figure 4: Comparison of Mean Concentration of Zn in Liquid Milk Samples

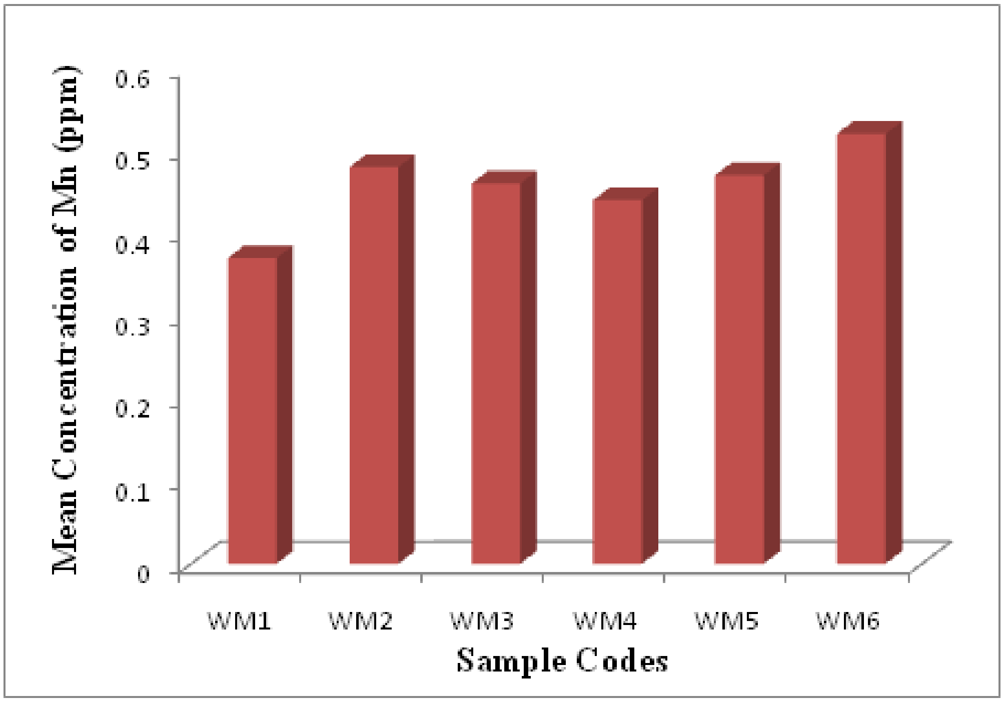

Figure 5: Comparison of Mean Concentration of Mn in Liquid Milk Samples

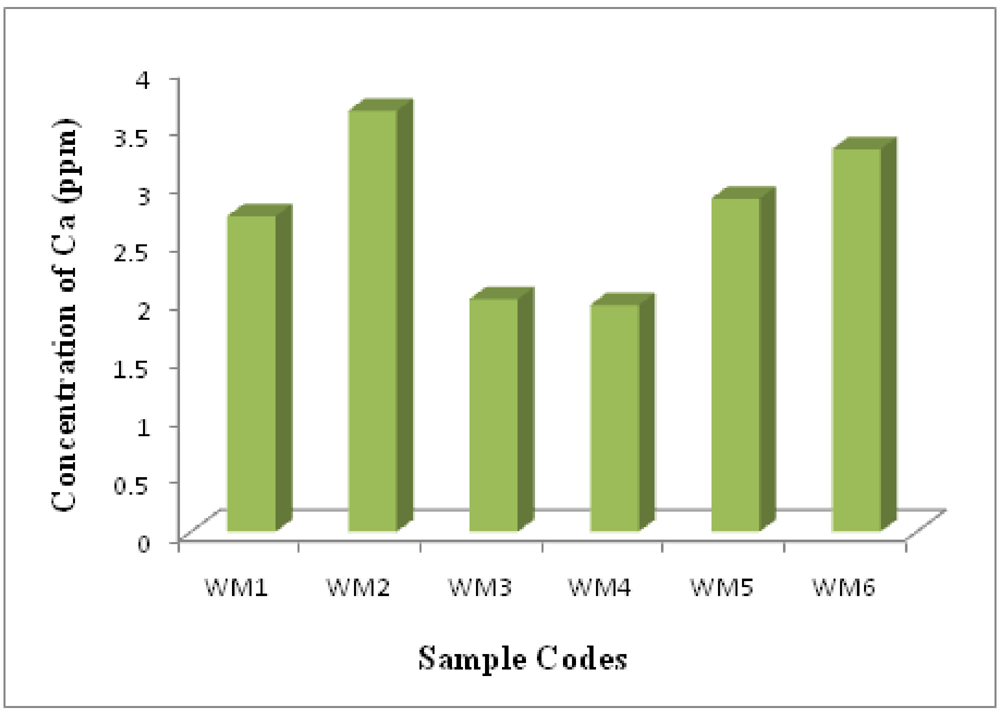

Figure 6: Comparison of Concentration of Ca in Liquid Milk Samples

The concentration of metals observed for both dry and liquid milk samples were in following order: Ca > Zn > Mn.

Data obtained from the chemical analysis of the dry and liquid milk samples was evaluated statistically and is given in Table 5 and Table 6 respectively. These Tables show that in both dry and liquid milk samples, Ca has more standard deviation than Zn and Mn.

Table 5: Descriptive Statistics for Dry Milk Samples

Metals	N	Minimum	Maximum	Mean	Median	Standard Deviation
Zn	4	23.22	30.51	26.4	25.94	± 3.26
Mn	4	7.67	11.6	9.496	9.357	± 1.629
Ca	4	328	336	331.6	331.2	± 3.36

Table 6: Descriptive Statistics for Liquid Milk Samples

Metals	N	Minimum	Maximum	Mean	Median	Standard Deviation
Zn	6	1.96	3.64	2.755	2.805	± 0.677
Mn	6	0.37	0.52	0.4567	0.465	± 0.0501
Ca	6	195	1528	888	903	± 484

The results of milk samples were also compared to the limits, set by International Dairy Federation. The recorded limit of International Dairy Federation for Zn, Mn and Ca were 3.28 ppm, 0.025 ppm [5] and 1200 ppm [19] respectively. Concentration of Zn in most of the liquid milk sample was in agreement and some were below to the value set by International Dairy Federation. However, in dry milk, all samples exceeded the limit. Value of Mn in liquid as well as in dry milk samples exceeded the recommended value. Furthermore, value of Ca in dry milk samples was below to the limit, on contrary value was within the limit in some of the liquid milk samples while in others, value exceeded the recommended limit.

The concentration of Zn, Mn and Ca in tested milk samples were also compared with previous studies reported by other researchers worldwide. Table 7 represents the comparison of Zn, Mn and Ca concentration of liquid milk in Pakistan with the published data. E. M. R. Rodríguez, et al [6] reported the concentration of Zn (2.30 – 6.60) ppm in cow's milk of Spain, which is the highest concentration reported among all published data in Table 7. Milk samples analyzed in present study contain less concentration of Zn (1.96−3 .64) ppm as compared to the concentrations reported in different countries by various authors, such as: V. A. Soares et al, [11] (4.59 ppm)—Brazil, A.G. Dawd, et al [9] (4.923 ± 0.277) ppm—Ethiopia, P. Ostapczuk, et al [12] (3.73 ppm)—Germany, F. Mahmood, et al [20] —(Multan) Pakistan, H. Benemariya, et al [21] (4.1ppm)—South Africa, R. Moreno-Rojas, et al [22] (4.2 –0.41) ppm—Spain. On the other hand, higher values of Zn concentration were reported as compared to the present study by S. M. Farid, et al [1] (0.5992 ± 0.0052) ppm, and A. A. Kinsara, et al [23] (0.944 ± 0.0024) ppm in Saudi Arabia. In addition, the results presented by H. A. Khan, et al [1] from Bangladesh (1.2145 ppm) also shows higher values in comparison to the present study (Table 7).

The results of present study are in agreement with the results reported in Italy, India, Poland, Lower Silesia Region, Upper Silesia Region, Japan and China reported by M. Sikirić, et al [4] (3.82 ppm), R.M. Tripathi, et al [14] (3.1 ppm), R. M. Tripathi (3.177 ppm) [24], Z. Dobrzañski, et al [7] (3.163 ± 0.71) ppm, (3.085 ± 0.589) ppm, L. Qin, et al [13] (2.72 ± 0.61) ppm, and L. Qin, et al [13] (2.38 ± 0.50) ppm respectively.

Some of the results of present study are within the range and some exceeded the range, reported in Nigeria by P. C. Onianwa, et al [8] (0.39 –2.75) ppm and in Egypt by A. Enb, et al [5] (3.001−3 .940) ppm. In addition, E. M. Rodríguez, et al [6] from Spain reported (2.30 –6.60) ppm in raw cow's milk and (2.81 –3.19) ppm in sterilized cow's milk.

Concentration of Mn present in liquid milk samples reported by present study (Table 7) is highest among all data reported by other researchers. As indicated in Table 7, concentration of Ca in liquid milk samples is in agreement with the concentration reported in Croatia (1403.94 ± 322.33) ppm [4], China (1041 ± 53) ppm and Japan (1053 ± 72) ppm (14329 ± 249) ppm [13].

Table 7: Comparison of Zn, Mn and Ca Concentration of Liquid Milk in Pakistan with the Published Data

Country	Sites	Types of Milk or Brand	Concentration of Metals (ppm)			References
			Zn	Mn	Ca	
Egypt	Giza	Raw cow's milk	3.001– 3.940	0.048 – 0.084	–	[5]
China	Inner Mangolia	Raw cow's milk	2.38 ± 0.50	0.20 ± 0.01	1041 ± 53	[13]
Spain	Canary Island	Sterilized cow's milk	2.81 – 3.19	–	1235 –1395	[6]
		Raw cow's milk	2.30 – 6.60	–	1196 – 2236	[6]
Spain	Córdoba	Cow's milk	4.21 ± 0.41	0.026 ± 0.006	–	[22]
Japan	Yamanashi Prefecture	Raw cow's milk	3.04 ± 0.46	0.19 ± 0.02	1053 ± 72	[13]
Poland	Lower Silesia Region	Raw cow's milk	3.163 ± 0.71	0.051 ± 0.014	–	[7]
	Upper Silesia Region	Raw cow's milk	3.085 ± 0.589	0.101 ± 0.025	–	[7]
India	Bombay	Not mentioned	3.1	–	–	[14]
India	Mumbai	Cow's milk	3.177	–	–	[24]
South Africa	Burundi	Cow's milk	4.1	–	–	[21]
Germany	Jülich	Raw cow's milk	4.1	–	–	[12]
Bangladesh	–	Cow's milk	1.214	–	–	[2]
Nigeria	Ibadan	peak	0.39 – 2.75	–	–	[8]
Saudi Arabia	Jeddah	Fresh cow's milk	0.944 ± 0.0024	0.017 ± 0.0004	–	[23]
Saudi Arabia	Makkah, Madina, Jeddah and Gassim city	Al-Shafi	0.5992 ± 0.0052	–	–	[1]
		Al-Marai	0.3523 ± 0.0052	–	–	[1]
		Al-Hanah	1.115 ± 0.0086	–	–	[1]
		KAAU	1.144 ± 0.0089	–	–	[1]
		KSU	1.180 ± 00.0092	–	–	[1]

Table 7: Comparison of Zn, Mn and Ca Concentration of Liquid Milk in Pakistan with the Published Data (Continued)

Country	Sites	Types of Milk or Brand	Concentration of Metals (ppm)			References
			Zn	Mn	Ca	
Saudi Arabia		Gassim (Gessim area)	0.9902 ± 0.0073	–	–	[1]
		Gassim (Burayda area)	1.2352 ± 0.0093	–	–	[1]
		Najayah	1.0463 ± 0.0085	–	–	[1]
		Nadec	0.6232 ± 0.0058	–	–	[1]
		Tabiah	0.9084 ± 0.0061	–	–	[1]
		Assafwa	1.2001 ± 0.0098	–	–	[1]
Croatia	Zagreb Region	Raw cow's milk	0.51 ± 0.16	–	1403.94 ± 322.33	[4]
Italy	Marche Region	Raw cow's milk	3.82	–	–	[4]
Ethiopia	Addis Ababa	Cow's milk	4.923 ± 0.277	–	–	[9]
Brazil	Vale do Paraíba region	Cow's milk	4.59	–	–	[8]
Pakistan	Multan	Tetra-packs	2.88 –7.31	–	777.50 –1178.76	[20]
Pakistan	Karachi	WM1 (Branded milk)	2.73 ± 0.0094	0.37 ± 0.0051	1044	Present Study
		WM2 (Branded milk)	3.64 ± 0.0086	0.48 ± 0.0039	1528	Present Study
		WM3 (Branded milk)	2.01 ± 0.0044	0.46 ± 0.0077	762.8	Present Study
		WM4 (Branded milk)	1.96 ± 0.0022	0.44 ± 0.0097	552.4	Present Study
		WM5 (Branded milk)	2.88 ± 0.0057	0.47 ± 0.0044	194.64	Present Study
		WM6 (Branded milk)	3.31 ± 0.0139	0.52 ± 0.0112	1244	Present Study

Table 8 represents the comparison of Zn, Mn and Ca concentration of dry milk of different countries with the present study. Dry milk samples analyzed in present study varied widely in comparison to the published data. The concentration of Zn in different dry milks obtained in the present study was lower than the value reported by G. Zurera-Cosano, et al, [15] (44.5 ± 0.6) ppm. Comparasion of the results also indicated that the concentration of Zn in different dry milks obtained in the present study was higher than the values reported in milks of Germany, Nigeria and Saudi Arabia. Concentration of Mn present in dry milk samples reported by present study is highest among all data presented in Table 8. Concentration of Ca in dry milk samples reported in Spain (14329 ± 249) ppm [15] was higher than present study as shown by Table 8.
Mineral content of milk may be affected by many factors including mainly the content of a given element in soil, water, air, veterinary drugs such as antimicrobials, hormones, antihelmentic drugs and pesticides, containers, processing and packaging materials, as well as phenomena of inter element interactions [9, 15]. Mineral content may also vary by other factors like breed of animal and season of the year [4].

Table 8: Comparison of Zn, Mn and Ca Concentration of Dry Milk in Pakistan with the Published Data

Country	Sites	Types of Milk or Brand	Concentration of Metals (ppm)			References
			Zn	Mn	Ca	
Germany	Jülich	Instant powder milk	11.8	–	–	[12]
Nigeria	Ibadan	Cow Bell	15.1 – 21.5	–	–	[8]
Spain	Granada	Powdered milk	44.5 ± 0.6	0.308 ± 0.024	14329 ± 249	[15]
Saudi Arabia	Makkah, Madina, Jeddah and Gassim city	Al-Rabie	0.978 ± 0.0074	–	–	[1]
		Saudia	0.891 ± 0.0071	–	–	[1]
		Cortina	0.482 ± 0.0061	–	–	[1]
		Jamjoom	1.078 ± 0.0087	–	–	[1]
		Danya	1.111 ± 0.0089	–	–	[1]
		Noor	1.200 ± 0.0032	–	–	[1]
Pakistan	Karachi	DM1 (Branded milk)	27.4258 ± 0.0071	9.644 ± 0.0091	328	Present Study
		DM2 (Branded milk)	24.4512 ± 0.0012	7.67 ± 0.0017	336	Present Study
		DM3 (Unbranded milk)	30.5137 ± 0.0057	9.07 ± 0.0021	330.4	Present Study
		DM4 (Unbranded milk)	23.2221 ± 0.007	11.6 ± 0.0058	332	Present Study

Conclusions

Milk contains macroelements as well as microelements and is an important part of our daily diet. In present study, Zn, Mn and Ca were analyzed in commercially available dry and liquid milk samples. The concentration of Zn, Mn and Ca in liquid milk samples was found to be (1.96 –3.640), (0.37–0.52) and (195 –1528) ppm respectively while in dry milk samples it was found (23.22 –30.5137), (7.67 –11.6) and (328–336) ppm respectively.
The results of milk samples were compared to the limits, set by International Dairy Federation. The recorded limit of International Dairy Federation for Zn, Mn and Ca were 3.28 ppm, 0.025 ppm and 1200 ppm respectively [5]. Quality of liquid and dry milk samples according to International Dairy Federation was also determined (Table 9 and 10). Concentration of Zn in most of the liquid milk sample was in agreement and some were below to the value set by International Dairy Federation. However, in dry milk, all samples exceeded the limit. Value of Mn in liquid as well as in dry milk samples exceeded the recommended value. Furthermore, value of Ca in dry milk samples was

below to the limit, on contrary value was within the limit in some of the liquid milk samples while in others, value exceeded the recommended limit. The concentration of Zn, Mn and Ca in tested milk samples were also compared with the published data of other countries.

The study showed that there is wide variation in the concentration of Zn, Mn and Ca between liquid milk samples and dry milk samples. Some of milk samples exceeded the permissible limits. So it is suggested that the concerned authorities should check and improve the level of these essential metals in milk available in markets.

Table 9: Quality of Liquid Milk Samples

Sample Codes	Types of Milk /Brand	Zn	Mn	Ca
WM1	Branded	**	**	***
WM2	Branded	***	*	**
WM3	Branded	*	*	**
WM4	Branded	*	*	*
WM5	Branded	***	*	*
WM6	Branded	****	*	****

**** Best, ***Good, **Fair and*poor

Table 10: Quality of Dry Milk Samples

Sample Codes	Types of Milk /Brand	Zn	Mn	Ca
DM1	Branded	*	*	*
DM2	Branded	*	*	*
DM3	Unbranded	*	*	*
DM4	Unbranded	*	*	*

*poor

Acknowledgment

Thanks to the Dean, Faculty of Science, University of Karachi, Pakistan for providing financial assistance. Authors acknowledge the support of Centralized Science Laboratories, University of Karachi, Pakistan for providing facility of Atomic absorption spectrometer. Thanks to M. Yousuf for his help. Authors declare that there is no conflict of interest.

References

[1] S. M. Farid, M. A. Enani and S. A. Wajid, Determination of Trace Elements in Cow's Milk in Saudi Arabia, *Journal of King Abdulaziz University*, **15** (2) (2004) 131–140.

[2] P. E. Johnson and G. W. Evans, Relative Zinc Availability in Human Breast Milk, Infant Formulas and Cow's Milk, *The American Journal of Clinical Nutrition*, **31** (1978) 416–421.

[3] A. S. Al Khalifa and D. Ahmad, Determination of the Key Elements by ICP-OES in Commercially Available Infant Formulae and Baby Foods in Saudi Arabia, *American Journal of Food Science*, **4** (7) (2010) 464–468.

[4] M. Sikirić, N. Brajenović, I. Pavlovic, J. L. Havranek and N. Plavljanić, Determination of Metals in Cow's Milk by Flame Atomic Absorption Spectrometry, *Czech Journal of Animal Science*, **48** (11) (2003) 481–486.

[5] A. Enb, M. A. Abou-Donia, N. S. Abd-Rabou, A. A. K. Abou-Arab and M. H. El-Senaity, Chemical Composition of Raw Milk and Heavy Metals Behavior During Processing of Milk Products, *Global Veterinaria*, **3** (3) (2009) 268–275.

[6] E. M. R. Rodríguez, M. S. Alaejos and C. D. Romero, Mineral Concentrations in Cow's Milk from the Canary Island, *Journal of Food Composition and Analysis*, **14** (2001) 419–430.
[7] Z. Dobrzañski, R. Kołacz, H. Górecka, K. Chojnacka and A. Bartkowiak, The Content of Microelements and Trace Elements in Raw Milk from Cows in the Silesian Region, *Polish Journal of Environmental Studies*, **14** (5) (2005) 685–689.
[8] P. C. Onianwa, I. G. Adetola, C. M. A. Iwegbue, M. F. Ojo And O. O. Tella, Trace Heavy Metals Composition of Some Nigerian Beverages and Food Drinks, *Food Chemistry*, **66** (1999) 275–279.
[9] A. G. Dawd, T. B. Gezmu and G. D. Haki, Essential and Toxic Metals in Cow's Whole Milk from Selected Sub-Cities in Addis Ababa, Ethiopia, *Online International Journal of Food Science*, **1** (1) (2012) 12–19.
[10] R. Caggiano, S. Sabia, M. D. Emilio, M. Macchiato, A. Anastasio, M. Ragosta and S. Paino, Metal Levels in Fodder, Milk, Dairy Products, and Tissues Sampled in Ovine Farms of Southern Italy, Environmental Research, 99 (2005) 48–57.
[11] V. A. Soares, M. M. Kus, A. L. C. Peixoto, J. S. Carrocci, R. F. S. Salasar and H. J. I. Filho, Determination of Nutritional and Toxic Elements in Pasteurized Bovine Milk from Vale Do Paraiba Region (Brazil), *Food Control*, **21** (1) (2010) 45–49.
[12] P. Ostapczuk, P. Valenta, H. Rutzel and H. W. Nurnberg, Application of Differential Pulse Anodic Stripping Voltammetry to the Determination of Heavy Metals in Environmental Samples, *The Science of the Total Environment*, **60** (1987) 1–16.
[13] L. Qin, X. Wang, W. Li, X. Tong and W. Tong, The Mineral and Heavy Metals in Cow's Milk from China and Japan, *Journal of Health Science*, **55** (2) (2009) 303–305.
[14] R. M. Tripathi, R. Raghunath, V. N. Sastry and T. M. Krishnamoorthy, Daily Intake of Heavy Metals in Bombay City, *The Science of the Total Environment*, **208** (1997) 149–159.
[15] G. Zurera-Cosano, R. Moreno-Rojas and M. Amaro-Lopez, Effect of Processing on Contents and Relationships of Mineral Elements of Milk, *Food Chemistry*, **51** (1994) 75–78.
[16] P. Licata, D. Trombetta, M. Cristani, F. Giofrè, D. Martino, M. Calò and F. Naccari, Levels of Toxic and Essential Metals in Samples of Bovine Milk From Various Dairy Farms in Calabria, Italy, *Environment International*, **30** (2004) 1–6.
[17] S. Saracoglu, K. O. Saygi, O. D. Uluozlu, M. Tuzen and M. Soylak, Determination of Trace Element Contents of Baby Foods from Turkey, *Food Chemistry*, **105** (2007) 280–285.
[18] T. G. Kazi, N. Jalbani, J. A. Baig, G. A. Kandhro, H. I. Afridi, M. B. Arain, M. K. Jamali and A.Q. Shah, Assessment of Toxic Metals in Raw and Processed Milk Samples Using Electrothermal Atomic Absorption Spectrophotometey, *Food and Chemical Toxicology*, **47** (2009) 2163–2169.
[20] F. Mahmood, S. B. Niazi, A. Raza and M. Y. Khokhar, Physico-analytical Studies on Commercial Milk Samples, *Journal of Chemical Society of Pakistan*, **26** (3) (2004) 212–216.
[21] H. Benemariya, H. Robberecht and H. Deelstra, Zinc, copper, and selenium in milk and organs of cow and goat from Burundi, Africa, *The Science of the Total Environment*, **128** (1) (1993) 83–98.
[22] R. Moreno-Rojas, M. Amaro-Lopez and G. Zurera-Cosano, Micronutrients in Natural Cow, Ewe and Goat Milk, *International Journal of Food Science and Nutrition*, **44** (1993) 37–46.
[23] A. A. Kinsara and S. M. Farid, Concentration of Trace Elements in Human and Animal Milk in Jeddah, Saudi Arabia, *Medical Journal of Islamic World Academy of Sciences*, **16** (4) (2008) 181–188.
[24] R. M. Tripathi, R. Raghunath, V. N. Sastry and T. M. Krishnamoorthy, Daily Intake of Heavy Metals by Infants through Milk and Milk products, *The Science of the Total Environment*, **227** (1999) 229–235.