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Determination of Trace Elements in Ashes of Milk Samples by Using XRF Technique

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Energy dispersive X-ray fluorescence system has been used to determine the heavy metal ions present in cow's milk. Samples have been collected from farm and markets within the border of Kahramanmaras province, Turkey. An Epsilon 5 PANalytical Almelo, the Netherlands model energy dispersive X-ray fluorescence spectrometer has been used to analyze the samples. As a result of analysis of the samples, the various concentrations of elements such as Na, Mg, Al, Si, P, S, Cl, K, Ca, Cr, Mn, Fe, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Zr, I, Yb, Hf and Pb were found.

Key Words: Milk, X-ray fluorescence, Trace element, X-ray.

INTRODUCTION

A nutrient is a chemical that an organism needs to live and grow or a substance used in an organism's metabolism which must be taken in from its environment. One of the most important of these nutrients is the milk. Milk is a truly important foodstuff necessary to every human being since the moment of birth. The composition of milk is largely affected by the type of animal feed and soil contamination[1]. Milk is a universal nutrient alone or in combination with other foods and contains water, milk fat, lactose, protein, lipids, carbohydrates and mineral matter[2,3]. The different elements presented in milk play an important role in human growth and well-being[4] but if drinking milk contains toxic metals, these toxic metals can be dangerous for human health. For example, nickel may also be an essential nutrient for human but a daily dose of 250 mg of soluble nickel may cause toxic symptoms in humans. It is highly improbable that the amounts found in milk consumed in a mixed diet could cause problems[3]. Similarly, arsenic is a well-known classic poison but has also been a component of many medicines. It may be an essential nutrient, but in limited quantities, 0.76 to 1.95 mg As/kg body wt of a 70-kg human, is lethal[5]. Since the amount in milk is small, it is unlikely to ever present a problem even with excess environmental contamination. The metals of concern are lead, cadmium and mercury which at sufficient levels in milk and dairy products could cause problems. Others of interest are arsenic, chromium and nickel[6]. Chromium may be an essential trace mineral for humans[7]. Therefore, the elemental analysis of the milk is very important. Broadly used methods for elemental analysis of milk, *e.g.* inductively coupled plasma optical emission and mass spectrometry, include either preliminary dry or wet ashing[8-11] or microwave-assisted digestion of organic matrix[9,12]. Such techniques of sample preparation are time-consuming and require large amounts of expensive reagents, which can produce hazardous waste and might contaminate samples with analytes[13]. Despite these problems in these techniques, energy dispersive X-ray fluorescence analysis systems are very fast and quick[14]. Therefore, energy dispersive X-ray fluorescence analysis of milk has become widespread in dairy industry. Therefore, the method has a great potential, as the dried samples can be analyzed directly without any chemical treatment and X-ray fluorescence equipment is rather accessible. Elemental analysis has been known as important process of accurately determining the composition of elements in materials. Elemental analysis can be qualitative (determining what elements are present) and it can be quantitative (determining how much of each are present). In addition, there is a lot of research done by using X-ray fluorescence technique[15-17].

The aim of this study is to do the heavy metal analysis of the sample that collected from dairy farms and markets in Kahramanmaras province by using energy dispersive X-ray fluorescence system.

EXPERIMENTAL

The samples used in research have been collected from farms and markets within the border of Kahramanmaras province in Turkey. All samples were dried in an oven at 240 ºC

TABLE-1
ELEMENTAL ANALYSIS VALUES OF THE SAMPLES USING EDXRF SYSTEM

	Sample 1		Sample 2		Sample 3		Sample 4		Sample 5		Sample 6		Sample 7		Sample 8	
Na	2.66 ± 0.133	%	0.38 ± 0.02	%	6.35 ± 0.317	%	0.34 ± 0.02	%	2.66 ± 0.13	%	0.38 ± 0.02	%	6.35 ± 0.32	%	0.60 ± 0.03	%
Mg	-	-	-	-	-	-	0.210.01	%	392.55 ± 19.63	ppm	-	-	-	-	813.51 ± 40.68	ppm
Al	392.55 ± 19.63	ppm	-	-	-	-	-	-	0.45 ± 0.03	%	-	-	-	-	0.25 ± 0.01	%
Si	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
P	0.45 ± 0.03	%	1.29 ± 0.06	%	1.26 ± 0.06	%	1.27 ± 0.06	%	1.41 ± 0.07	%	1.29 ± 0.06	%	1.26 ± 0.06	%	1.17 ± 0.58	%
S	1.41 ± 0.07	%	0.66 ± 0.03	%	0.67 ± 0.03	%	0.67 ± 0.03	%	1.24 ± 0.06	%	0.66 ± 0.03	%	0.67 ± 0.03	%	0.64 ± 0.08	%
Cl	1.24 ± 0.06	%	0.92 ± 0.05	%	7.16 ± 0.36	%	0.8 ± 20.04	%	4.92 ± 0.25	%	0.92 ± 0.05	%	7.16 ± 0.36	%	0.88 ± 0.04	%
K	4.92 ± 0.25	%	1.90 ± 0.09	%	1378.81 ± 68.94	ppm	1.63 ± 0.08	%	0.48 ± 0.02	%	1.90 ± 0.09	%	1378.81 ± 68.94	ppm	1.55 ± 0.08	%
Ca	0.48 ± 0.02	%	2.59 ± 0.13	%	2.21 ± 0.11	%	2.54 ± 0.18	%	1.89 ± 0.09	%	2.59 ± 0.13	%	2.21 ± 0.11	%	2.182 ± 0.11	%
Cr	-	-	-	-	34.45 ± 1.72	ppm	62.06 ± 3.10	ppm	-	-	-	-	34.45 ± 1.72	ppm	54.89 ± 2.74	ppm
Mn	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Fe	1.89 ± 0.09	%	63.39 ± 3.17	ppm	88.33 ± 4.42	ppm	160.71 ± 8.04	ppm	44.84 ± 2.24	ppm	63.40 ± 3.17	ppm	88.33 ± 4.42	ppm	149.68 ± 7.484	ppm
Ni	44.84 ± 2.24	ppm	16.52 ± 0.83	ppm	13.21 ± 0.66	ppm	162.97 ± 8.15	ppm	39.19 ± 1.96	ppm	16.52 ± 0.83	ppm	13.21 ± 0.66	ppm	29.89 ± 1.49	ppm
Cu	39.17 ± 1.96	ppm	67.32 ± 3.37	ppm	30.79 ± 1.54	ppm	86.14 ± 4.31	ppm	66.88 ± 3.34	ppm	67.33 ± 3.37	ppm	30.79 ± 1.54	ppm	42.15 ± 2.11	ppm
Zn	66.88 ± 3.34	ppm	149.58 ± 7.48	ppm	152.66 ± 7.63	ppm	160.83 ± 8.04	ppm	141.74 ± 7.09	ppm	149.58 ± 7.48	ppm	152.66 ± 7.63	ppm	123.57 ± 6.18	ppm
As	-	-	0.764 ± 0.04	ppm	-	-					0.76 ± 0.04	ppm	-	-	-	-
Br	141.74 ± 7.09	ppm	58.44 ± 2.92	ppm	10.62 ± 0.53	ppm	39.73 ± 1.99	ppm	8.87 ± 0.4	ppm	58.44 ± 2.92	ppm	10.62 ± 0.53	ppm	30.19 ± 1.51	ppm
Rb	8.87 ± 0.44	ppm	27.90 ± 1.40	ppm	-	-	19.85 ± 0.99	ppm	-	-	27.90 ± 1.40	ppm	-	-	14.31 ± 0.72	ppm
Sr	17.58 ± 0.88	ppm	19.33 ± 0.97	ppm	18.16 ± 0.91	ppm	23.46 ± 1.17	ppm	17.58 ± 0.88	ppm	19.33 ± 0.97	ppm	18.16 ± 0.91	ppm	19.44 ± 0.97	ppm
Y	-	-	30.29 ± 1.51	ppm	-	-	-	-	-	-	30.29 ± 1.51	ppm	-	-	-	-
Zr	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
I	10.07 ± 0.50	ppm	35.01 ± 1.75	ppm	36.38 ± 1.82	ppm	-	-	-	-	35.01 ± 1.75	ppm	36.38 ± 1.82	ppm	20.75 ± 1.04	ppm
Yb	-	-	-	-	108.59 ± 5.43	ppm	-	-	-	-	-	-	108.59 ± 5.43	ppm	-	-
Hf	-	-	44.15 ± 2.21	ppm	23.49 ± 1.17	ppm	50.71 ± 2.54	ppm	-	-	44.19 ± 2.21	ppm	23.49 ± 1.17	ppm	51.12 ± 2.56	ppm
Pb	-	-	8.19 ± 0.41	ppm	-	-	-	-	10.07 ± 0.50	ppm	8.19 ± 0.41	ppm	-	-	-	-

for 24 h and grinded with mill so that has been turned into powder. To reduce particle size effects, the powder obtained was sieved using a 400 mesh sieve and then mixed for 20 min. Before the measurements of the samples, to evaporate the water in the samples, samples were dried at 105 ºC for 4 h again. Samples are compressed for 20 sec with 7 ton hydraulic press and reduced to 40 mm in diameter and the pellets at the mass 500 ± 3 mg were prepared. It is well known that the measurement of thin samples in energy dispersive X-ray fluorescence presents several advantages over the methods using thick samples. In the latter case, the increase of background by multiple scattering within the sample causes absorption and enhancement effects that dominate over the gain in characteristic line intensity resulting from the increase in sample thickness. However, the absorption and enhancement effects are less significant and can be easily corrected or neglected for thin samples[16]. An Epsilon 5 PANalytical Almelo the Netherlands model energy dispersive X-ray fluorescence spectrometer was used to analyse prepared milk samples. The obtained values are given in Tables 1 and 2.

RESULTS AND DISCUSSION

Samples were taken from four different dairy farms and from three different pasteurized milks. The energy dispersive X-ray fluorescence spectrometer (Epsilon 5, PANalytical, Almelo, the Netherlands) was used for the analysis of samples of 15 milks and the elements Na, Mg, Al, Si, P, S, Cl, K, Ca, Cr, Mn, Fe, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Zr, I, Yb, Hf and Pb found in different concentrations. However, some of the elements were not found in all samples. These elements are aluminium (in 9 samples), arsenic (in 3 samples) and lead (in 5 samples). If these elements exceed a certain limit, they may cause various diseases in humans. For example, if a certain limit of the amount of aluminium exceeds in human body, it is known that this may lead to Alzheimer's disease. In addition to these, iodine (in 9 samples) hafnium (in 11 samples), ytterbium (in 4 samples),

TABLE-2
ELEMENTAL ANALYSIS VALUES OF THE SAMPLES USING EDXRF SYSTEM

	Sample 9		Sample 10		Sample 11		Sample 12		Sample 13		Sample 14		Sample 15	
Na	6.97 ± 0.35	%	0.34 ± 0.02	%	4.18 ± 0.21	%	3.62 ± 0.181	%	0.40 ± 0.02	%	0.83 ± 0.42	%	0.64 ± 0.03	%
Mg	-	-	-	-	-	-	-	-	1135.30 ± 56.77	ppm	-	%		
Al	-	-	-	-	-	-	0.29 ± 0.21	%	0.28 ± 0.014	%	0.28 ± 0.01	%	0.27 ± 0.01	%
Si	-	-	-	-	-	-	-	-	-	-	-	-	-	-
P	1.58 ± 0.08	%	1.51 ± 0.08	%	5.84 ± 0.292	%	4.22 ± 0.21	%	1.80 ± 0.04	%	1.89 ± 0.09	%	1.74 ± 0.09	%
S	0.80 ± 0.04	%	0.75 ± 0.04	%	0.98 ± 0.05	%	0.96 ± 0.05	%	0.85 ± 0.43	%	0.99 ± 0.049	%	0.72 ± 0.04	%
Cl	7.64 ± 0.38	%	1.11 ± 0.06	%	2.70.14	%	2.33 ± 0.12	%	1.24 ± 0.061	%	1.20 ± 0.60	%	1.15 ± 0.06	%
K	1328.14 ± 66.41	ppm	2.09 ± 0.10	%	0.72 ± 0.06	%	0.46 ± 0.02	%	2.15 ± 0.17	%	2.08 ± 0.10	%	2.01 ± 0.10	%
Ca	2.82 ± 0.14	%	2.93 ± 0.15	%	2.15 ± 0.11	%	2.13 ± 0.11	%	2.70 ± 0.13	%	2.72 ± 0.14	%	2.65 ± 0.13	%
Cr	-	-	-	-	-	-	-	-	38.12 ± 1.91	ppm	50.22 ± 2.51	ppm	52.867 ± 2.64	ppm
Mn	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Fe	45.95 ± 2.30	ppm	75.52 ± 3.77	ppm	53.58 ± 2.68	ppm	40.48 ± 2.02	ppm	84.17 ± 4.21	ppm	95.97 ± 480	ppm	95.29 ± 4.76	ppm
Ni	7.94 ± 0.34	ppm	16.49 ± 0.82	ppm	8.72 ± 0.45	ppm	4.94 ± 0.23	ppm	16.35 ± 0.82	ppm	16.67 ± 0.83	ppm	11.66 ± 0.58	ppm
Cu	27.07 ± 1.35	ppm	31.11 ± 1.55	ppm	20.48 ± 1.02	ppm	21.50 ± 1.08	ppm	38.47 ± 1.92	ppm	42.28 ± 2.11	ppm	34.54 ± 1.73	ppm
Zn	119.82 ± 5.10	ppm	142.90 ± 7.15	ppm	126.93 ± 6.35	ppm	157.45 ± 7.87	ppm	108.77 ± 5.44	ppm	117.86 ± 5.85	ppm	135.12 ± 6.76	ppm
As	-	-	-		-		-	-	-	-	4.47 ± 0.22	ppm	-	-
Br	12.18 ± 0.68	ppm	41.93 ± 2.01	ppm	11.15 ± 0.56	ppm	8.14 ± 0.41	ppm	66.80 ± 3.35	ppm	40.02 ± 2.01	ppm	43.55 ± 2.18	ppm
Rb	-	-	23.04 ± 1.15	ppm	10.72 ± 0.55	ppm	-	-	16.79 ± 0.84	ppm	11.39 ± 0.57	ppm	22.76 ± 1.14	ppm
Sr	30.92 ± 1.55	ppm	18.45 ± 0.07	ppm	-		13.84 ± 0.69	ppm	14.89 ± 0.74	ppm	16.07 ± 0.80	ppm	17.24 ± 0.86	-
Y	-	-	-	-	-		-	-	-	-	-	-	-	-
Zr	-	-	-	-	13.49 ± 0.67	ppm	-	-	-	-	-	-	-	-
I	-	-	38.981 ± 1.95	ppm	44.56 ± 2.23	ppm	28.84 ± 1.44	ppm	24.40 ± 1.22	ppm	34.86 ± 1.74	ppm	-	-
Yb	-	-	120.04 ± 6.00	ppm	-		-	-	-	-	114.60 ± 5.70	ppm	106.82 ± 5.34	ppm
Hf	-	-	42.53 ± 2.13	ppm	26.77 ± 1.34	ppm	22.47 ± 1.12	ppm	55.84 ± 2.79	ppm	59.67 ± 2.98	ppm	-	-
Pb	-	-	-	-	6.28 ± 0.31	ppm	5.99 ± 0.30	ppm	-	-	-	-	-	-

chromium (in 6 samples), magnesium (in 3 samples) and zirconium (in 1 samples) have been found. Variations of the concentrations of elements according to sample numbers such as Al, Mg, P, S, Cl, K, Ca, Fe, Cu, Zn, I and Pb are given in Figs. 1 and 2.

Important factors that influence the occurrence of many trace elements, including toxic ones, is environmental pollution, mainly of anthropogenic origin[18]; as well as phenomena of interelement interactions[19]. Therefore, different data are reported in the literature. Kiliç, *et al.*[20] measured the seasonal variations of trace elements and radioactivity concentrations in raw cow's milk samples collected from villages of Havsa, Hayrabolu, Malkara and Uzunköprü towns of west Thrace region in Turkey by using atomic absorption spectrophotometry. They reported that mean values of Cd, Co, Cr, Cu, Fe, Ni, Pb and Zn were found to be 10.0 ± 1.2, 51.5 ± 5.0, 5.7 ± 1.1, 51.2 ± 6.3, 257.0 ± 43.6, 43.3 ± 10.0, 120 ± 31 and 4187 ± 162 μg kg^{-1}, respectively. In addition, they obtained that average element concentrations in raw cow's milk samples can be arranged in a decreasing order as Zn > Fe > Pb > Co-Cu-Ni > Cd > Cr, respectively, in Thrace region in Turkey[20]. The content of micro elements and trace elements in raw milk from cows in the Silesian region were analyzed by Dobrzañski, *et al.*[21] by using ICP-MS system and they reported that no statistically significant differences were observed in the case of Zn, Ba, Cu, Cr, V, Ni, As, Mo, Pt, Sb, Au, Hf, Ce, U, Re, Tl, Ru, Rh, Ir, Ta, Be, but in the case of La, Nb, Ag, In and Y in the majority of samples the concentration was below detection limit. Flynn[22] reports the following elemental composition of milk: I (0.1-0.77), B (0.5-1.0), Fe (0.5), Si (3.0), Zn (3.5 mg/L) and Cr (2.0), Sc (10), F(20), Ni (26), Mn (30), As (20-60), Mo (50) and Cu (90

µg/L). It could also contain bismuth or lithium[23,24]. Trace element content of different tissues of dairy cows in Poland has been studied by Kosla *et al.*[25]. Relatively wide elemental composition of milk was reported by Hurleyw[26] B (0.27), F (0.15), Al (0.46), Si (1.43), V (0.09 µg/L), Cr (0.015), Mn (0.022), Fe (0.45), Ni (0.027), Co (0.6), Cu (0.13), Zn (3.9 mg/L), As (0.05), Se (0.04-1.27), Br (0.6), Sr (0.171), Mo (0.073), Ag (0.047) and I (0.043). Pashkova[8] has analyzed milk and dairy products by using X-ray fluorescence analysis (WDXRF) and he reported that the examined milk samples have been found in different concentrations minerals (Na, Mg, P, S, Cl, K, Ca) and trace elements (Mn, Fe, Ni, Cu, Zn, Rb, Sr, Br). Minor, major and trace elements in milks of some animal, such as cow, camel, goat, ewe and buffalo were analyzed by Rushed by using AAS1 and he reports that the elements Na, Mg, K, Ca, Cr, Mn, Fe, Co, Ni, Cu, Zn, Sr, Ag, Au and Pb *etc*. were found in milk samples. In addition, the contents of elements in a dry residue of cow milk were determined by Gunicheva by using X-ray fluorescence technique[27] and he said that the elements Na, Mg, Al, Si, P, S, Cl, K, Ca, Mn, Fe, Ni, Cu, Zn, Rb, Sr and Zr were found in dry residue of cow milk.

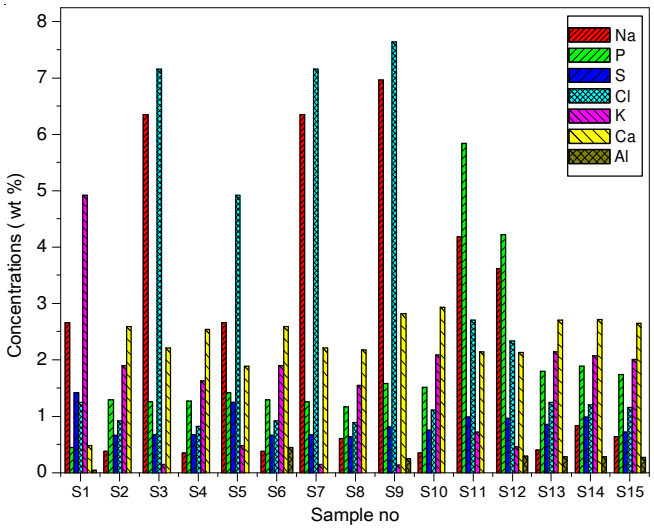

Fig. 1. Graph of the elemental concentrations *versus* sample numbers for Na, Al, P, S, Cl, K and Ca

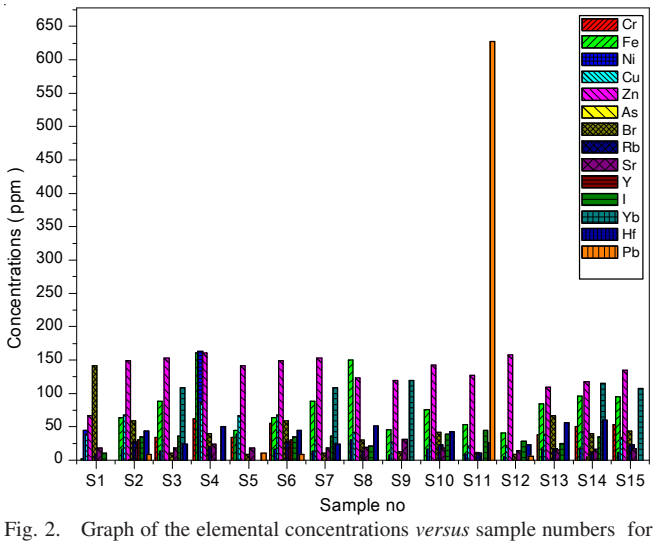

Fig. 2. Graph of the elemental concentrations *versus* sample numbers for Cr, Fe, Ni, Cu, Zn, As, Br, Rb, Sr, Y, I, Yb, Hf and Pb

Conclusion

In this study, the various milk samples were analyzed by using energy dispersive X-ray fluorescence system. The elements Na, Mg, Al, Si, P, S, Cl, K, Ca, Cr, Mn, Fe, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Zr, I, Yb, Hf and Pb found in different concentrations. However, as seen from Figs. 1 and 2, there were no indications of abnormal levels of Cr, Mn, Fe, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Zr, I, Yb, Hf and Pb in the milk samples collected from local markets and dairy farms in Kahramanmaras, Turkey. Therefore, it is suggested that the amount of the elements situated in the examined milk samples do not pose any risk for human health.

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