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Determination of Lead and Cadmium in Human Milk and Measure Some of its Composition

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Abstract: Infants are particularly sensitive to the toxic effects of lead. It is very important to know the contribution of milk to lead intake. The purpose of this study is to determine lead and cadmium present in samples of human milk by using graphite furnace atomic absorption spectrometry (GFAAS).Thirty human milk samples from three different localities were collected from healthy lactating women on the end of fifth month. The mean ± standard deviation of lead and cadmium concentrations in human milk were (1.7±0.085, 5.923 ±0.296, 5.11±0.25) ppb and (0.638 ± 0.0319, 1.842 ± 0.092, 2.65 ± 0.132) ppb respectively. In fact the determination of trace elements requires the use of sensitive analytical methods with high precision at low detection limits. These concentration ranges found were in a good agreement with those reported in the literature. Also, FTIR technique was used to determine fats and protein. The percentage range of fat was (0.65- 2.52) % and protein was (1.25-2.31) %.

Key wards: Human milk-cadmium and lead determination- graphite furnace fat and protein measurements.

INTRODUCTION

Milk provides the primary source of nutrition for young mammals before they are able to digest other type of foods .The early lactation milk is known as colostrums, for that it is usually the only source of food for infants during the five months of their lives. The exact components of raw milk vary by species, but it contains significant amount of saturated fats, protein and calcium as well as vitamin C.

Lead has often been called the leading environmental health threat to children (Hondaet al 2003, Gueu et al 2007 and Samarghandi at al 2007). It is toxic to the developing brain, and at high level results in numerous poisoning symptoms. Where in many areas of the world, a significant level of Pb turn up in human milk was (5-20 ppb) (Rabinwitz et al. 1985). That said, lead does not concentrate in breast milk because it does not attach to fat, indeed, levels of lead are generally higher in a mother's blood than in her milk (Tellez- et al. 2002). Also, cadmium is a toxic to the male reproductive system, the kidneys, and the brain. All of these contaminants are more likely to affect bottle- fed infants, because they are water contaminants or contaminants in infant formula (Karbassi et al, 2008 and Ursiyova and Masanova 2005). Because of the cadmium and the lead in milk are better absorbed into the baby than other dietary components, therefore high cadmium and lead concentration in breast milk is the first source of poisoning with these heavy metals in neonates (Frkovic et al.,1997) . Lead and cadmium level in breast milk correlate closely with area where lead is still used in gasoline, lead smelters and cigarette smoking respectively (Tellez et al 2002and Namhira et al., 1993) . (Saleh et al 1996) determined lead in human milk in three different areas from Egypt; he found that the lead level was higher duo to the heavy automobile traffic using leaded gasoline in addition to the use of lead water pipelines in these areas.

By far most advanced and widely used high sensitivity sampling technique for atomic absorption is the graphite furnace (Szkoda and Zanudzki 2005). In this technique, a tube of graphite is located in the sample compartment of the atomic absorption spectrometer, with the light path passing through it. A small volume of sample solution is quantitavely placed into the tube, normally through a sample injection hole located in the center of the tube wall. The tube is heated through a programmed temperature sequence unit; finally the analyte present in the sample is dissociated into atoms and atomic absorption occurs. Fourier transforms infrared (FTIR) spectrometry used for the determination of fats, protein and lactose in milk (Dror at al., 2005).

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(FTIR) is routinely used by laboratories specializing in milk analysis because it is a fast, nondestructive, and easy procedure that enables simultaneous measurement of several components in a complex natural media

The aim of this study is to develop a direct spectrometric technique using graphite furnace atomic absorption spectrometric for the determination of Pb and Cd in human milk samples of healthy lactating women were living in an industrial area (Nasr city, Helwan and El Khanka). Also, fat and protein can be determined in these samples by using Infrared Spectrophotometer.

METHODS AND MATERIALS

Instrumentation:

Perkin Elmer model 3100 spectrometer with auto sampler 60 equipped with a HGA – graphite furnace atomizer was used .Table (1) illustrates the operating conditions considered in this work.

FTIR Berkin Elmer 4100 type a ranged (400 – 4000) cm^{-1} was used for determined fat and protein.

Table 1: The operating Conditions for elements measured by GFAAS.

Element	Wave length nm	Slit Width nm	Vev	Chemical modifier	Drying Temp. (c°)	Ashing Temp. (c°)	Atomization Temp. (c°)	Cleaning
Cd	228.8	0.7	220	0.015mg pb+0.01 mgMg $(NO_3)_2$	120	300	1650	2400°C
Pb	283.3	0.7	220	0.01mg $Mg(NO_3)_2$	120	500	180	2500° C

Sample Collection:

Thirty lactating mothers, aged 25-35 (mean 30 years), living in three different cities of Egypt. The sampling centers were located in the following area (1) Nasr city (2) Helwan (3) El Khanka. Samples of ~10 g of milk at the end of the fives month of lactation were collected. Special care was taken to avoid any contamination, loss or alteration phenomena that would seriously affect the reliability of the data.

The mother's nipple was cleaned with high purity de ionized water and milk was collected directly into decontaminated polyethylene vials by mean of a polyethylene manual breast pump.

Vials were indelibly marked with a code number and immediately frozen at -20° C.

Sample Preparation:

All milk samples vials ware were soaked in nitric acid for 10 min and rinsed with distilled water before used. After that it was dried at hot plate. To eliminate the organic part of milk, 0.50 g of milk sample was treated with 6 ml of 65 % HNO_3, 1 ml 30 % H_2O_2 and 0.1 of EDTA then it heated gently on a hot plate at 50° C to concentrate, then diluted to 25 ml with distilled water (El Rahimi et al 2005)

The additive grams of EDTA to milk sample were tested in different concentrations as (0.1, 0.2, 0.3 and 0.4). The variation of these concentrations is shown in fig (1). Illustrate an example of this study on sample no (1). It was shown from the result that as a concentrate 0.1, the absorbance is the best one and it was added to all samples.

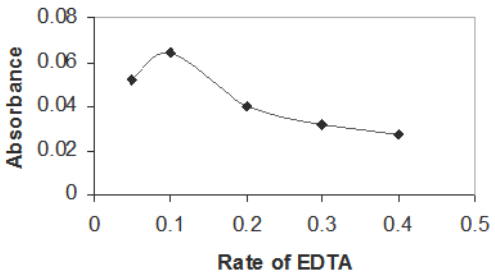


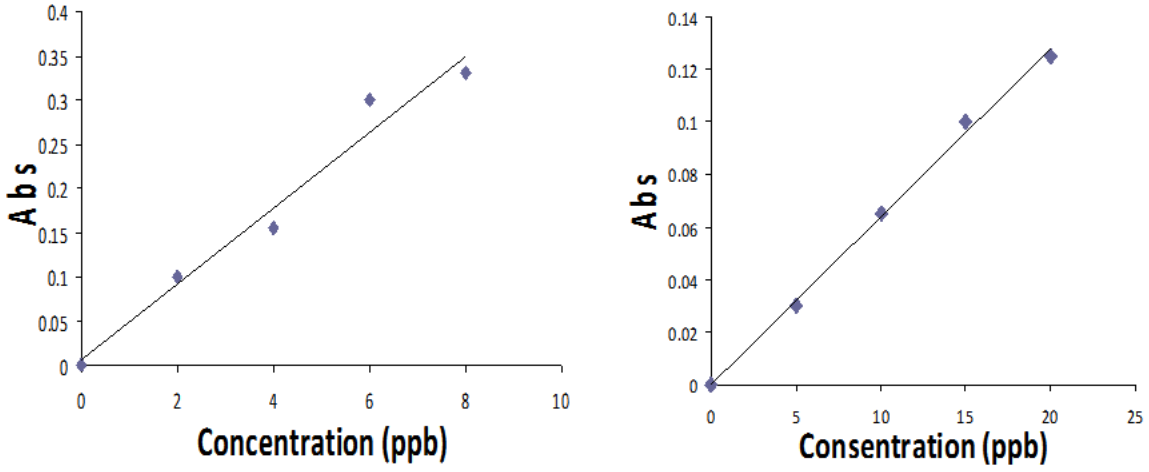
Fig. 1: The relation between Rate of EDTA and Absorbance.

Preparation of Standard Solution:

Standard solutions were prepared from stock solution containing 1000 µg/ml, for each studied element. This stock solution was diluted with bi-distilled water to give different concentrations were ranging from 0.05 to 5 µg / ml for different elements. The standard solutions were prepared fresh daily.

Analytical Calibration Curve:

20 µl of standard solution in increasing concentration were injected into the graphite tube under the operating conditions illustrated in table (1). Fig (2) shows the analytical calibration curve for (a) cadmium and (b) lead.



(A) (B)

Fig. 2: Analytical calibration curve of (a) Cd and (b)Pb

RESULTS AND DISCUSSION

Determination of Cadmium and Lead:

A sample solution of 20 µl was dispensing into the furnace under the same operating condition. The obtained results for Cd and Pb for tested samples were shown in Table (2).Also, this table is shown the range of metals detected in breast milk around the world (WHO).

Table 2: Pb and Cd concentration (µg/L) in human milk in different three areas.

samples	Cd	Pb
1	.638 ±0.032 (.485-.865)	1.7±.085 (.26-3.33)
2	1.84 ±0.092(1.02-2.54)	5.92±.296 (4.2-7.74)
3	2.56 ±0.12 (1.25-3.86)	5.11±.25 (3.41-6.88)
WHO	1.1 (0.1-3.8)	5 (0.0-41)

WHO: World health organization (Somogyi, A., 1993)
1- Samples of Nasr city 2 - samples of Helwan 3- samples of El Khanka

Determination of Fats and Protein:

For fat measurements, the most important absorption band appear at 1730 cm^{-1} and it can be assigned to C=O stretching variation and stretching vibration of C–O.

The band of aimed compound is the most important absorption band of the secondary structure of protein and it appear in the range of (1600 – 1700 cm^{-1}). The absorption band of protein appear at 1653 cm^{-1} and it can be assigned to 80 % stretching vibration of C=O. The variation of fat and protein in different samples is shown in table (3). Fig (3) was shown an example spectrum of a human milk sample.

Table 3: the percentage of fat and protein in human milk.

samples	Fat %	Protein %
1	0.828 (0.64-1.06)	2.2 (1.25-3.0)
2	1.11 (0.82-1.3)	1.98 (1.05-2.82)
3	1.05 (0.58-1.1)	2.06 (1.15-2.56)
4	2.2	3.1

1, 2, 3: Samples of the three different area.
4: WHO: World health organization 1993

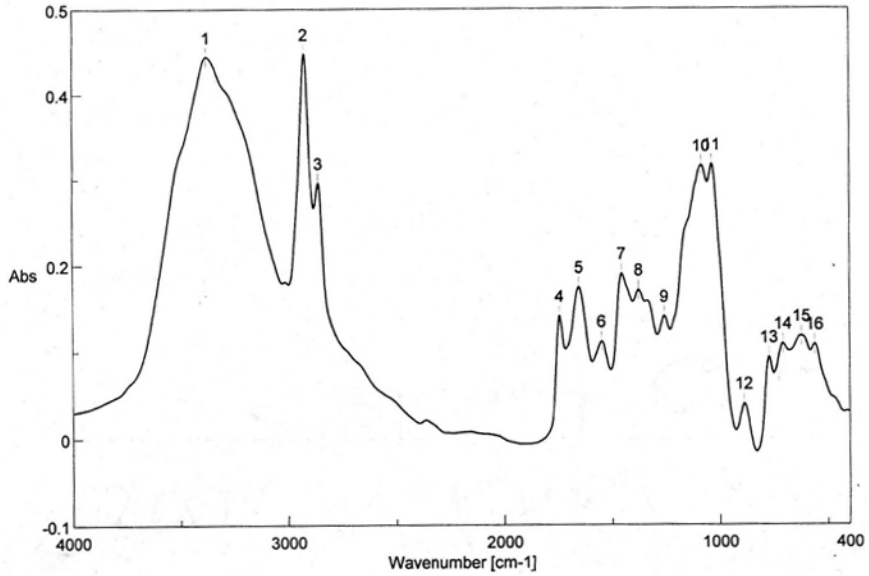


Fig. 3: An example spectrum of human milk.

Comparison of Results:

Comparison Between Human Milk and Cow Milk:

To find the difference between breast milk and cow's milk in terms of use for the infants as a form of nutrition set of fat in it to see if it can be used or not.

At the same procedure of measuring both (Cd, Pb) and (fat, protein), 10 samples of canted cow's milk were tested.

Fig (4) shows the relationship between fat and protein in mother's milk in the three different areas and cow's milk. This figure shows that the concentration of fat in cow's milk often higher than of mother's milk, which makes it harder to digest.

Also, Pb and Cd level in cow milk was higher than the breast milk, as shown in Fig (5), due to the animals feed on agricultural remaining also the agriculture supplied with the sewage and herbicide increases the concentration of toxic elements in the plants which transfer to animals.

Conclusion:

In the present work, a graphite furnace atomic absorption spectroscopy was used to develop a method for a direct determination of trace elements concentration (Cd and Pb) present in breast milk samples. The main advantages of the direct determination are a reduced risk of losses and also of contamination. This technique has high sensitivity (analyte amounts 10^{-8} -10^{11} g absolute), the ability to handle micro samples (5-100 ml) and a low noise level from the furnace, the detection limit (0.005µl /g) for Cd and (0.05µl /g) for Pb. The precision is typically (5-10) % using GF-AAS. Lead and cadmium were determined using this technique due to their lower concentration level in the samples.

According to the previous work, the concentrations of both Pb and Cd were found higher than results in the present work ,this is duo to the area which samples was taken. From the present results the concentration of both Pb and Cd were (1.7and 0.6 ppb) respectively- in a quiet and modernized area (Nasr City) but, in industrial and densely populated area (Helwan and ElKhanka) which found (5.92 and 2.65ppb) respectively in human milk. The World Health Organization (WHO) has set a daily permissible intake (DPI) level of 5 micrograms per kilogram per day of lead for children, and the DPI for cadmium is 1 microgram per kilogram per day.

Another technique was used FTIR for determination fat and protein which are present in both different breast milk and cow milk samples. From the results it appears that, the concentration of fat in cow's milk was higher than in human milk so the cow's milk is not commensurate with the child's age in this period. Although, the concentration of protein in cow's milk (3.2 %) was found higher than the human milk (2.1%) but, the human milk is the besting digest and most complete nutrition source for young infants; breast feeding should be encouraged because the absolute values of the effects are small within this range of lead concentration.

REFERENCES

Dror, M., *et al*., 2005. Pediatrics, 116(3): e432-e435.
Frkovic, A., M. Kras, A. Alebic Juretic, 1997. Bull.Environ.Contam.Toxicol., 58(1): 16-21.
Gueu, S., *et al*., 2007. Int. J.Environ. Sci.Tech., 4(1): 11-17.
Honda, R., *et al*., 2003. Toxicology, 180(3): 255-259.
Karbassi, A.R., *et al*., 2008. Environ.Geo., 53(8): 1811-1816.
Namihira, D., *et al*., 1993. J.Toxical.Environ. Health, 38(3): 225-232.
Rahimi, E., *et al*., 2009. Int. J.Environ. Sci. Tech., 6(4): 671-676.
Rabinowitz, M., A. Leviton and H. Needleman, 1985. Archives of Environmental Health, 40(5): 283-286.
Saleh, M.A., A.A. Ragab, A. Kamel, J. Jones and A.K. El-Sebae, 1996. Chemosphere, 32: 1859-1867.
Samarghandi, M.R., *et al*., 2007. Int.J.Environ. Sci.T ech., 4(1): 19-25.
Somogyi, A., 1993. 101(suppl 2): 45-52.
Szkoda, J. and Zmudzki, 2005. J., Bulletin of the veterinary institute in puawy, 49(1): 89-92.
Tellez-Rojo, *et al*., 2002. Am J. Epidem, 155: 420-428.
Ursiyova, M., V. Masanova, 2005. Food Addit.Contam., 22(6): 579-589.