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Summary

About 22 mineral elements considered to be essential to the human diet are present in milk. They are divided into two main groups: macro minerals and micro minerals. Seven macro minerals (calcium, phosphorus, magnesium, potassium, sodium, chloride and sulphur) are present in milk. Copper, iron, zinc, manganese, selenium, cobalt, iodine, bromine, silicon and rubidium are the most common trace elements incorporated in the living tissues and present in blood, milk and other body fluids in little quantities. Lactating animals may be exposed to high quantities of toxic metals such as lead and cadmium in the environment. These heavy metals reach the body through air, water or ingestion of polluted food, excreted in milk and constitute a deleterious effect on human health. **This study aimed to;** 1- investigate the levels of some macro and micro mineral elements in normal and mastitis milk of dairy cows, buffaloes, sheep and goats. 2- Studying; the relationship between mineral elements in milk and mastitis and comparing the level some macro and micro mineral elements in milk samples from different species. 3- Investigation of lead and cadmium contamination of milk at Assiut Governorate. A total number of 294 cases of dairy cows, buffaloes, sheep and goats of different breeds, ages, weights and stage of production were used in this study (100 lactating cows (60 were healthy and 40 were mastitic), 71 lactating buffalo-cows (40 were healthy and 31 were mastitic), 63 lactating ewes (33 were healthy and 30 were mastitic) and 60 lactating goats (30 were healthy and 30 were mastitic). 100 ml milk sample was collected in clean sterile plastic bottles directly from the udder of each examined animal.

Clinical examination of each studied case especially the mammary gland was carried out. Physical and chemical changes in the milk samples as well as pathological changes in the glandular tissue were recorded. There is swelling, heat, pain and indurations in the mammary gland of 37 cows, 26 buffalo-cows, 28 ewes and 27 goats. Milk samples of these animals showed variant degrees of discoloration and presence of clots. These cases were classified as clinical mastitis.

California mastitis test was carried out on 175 milk samples collected from healthy animal with clinically healthy udder (63 cows, 44 buffalo cows, 35 ewes and 33 goats). Positive California mastitis test was detected in 3 (4.8 %), 4 (9.1 %), 2 (5.7 %) and 3 (9.1 %) of milk samples from cows, buffaloes, sheep and goats respectively. These cases suffered sub-clinical mastitis and their milk samples were classified as mastitis milk samples.

Collected milk samples were subjected to wet digestion. Copper, iron, zinc, manganese, selenium, lead and cadmium in ppm were estimated in the digested milk samples by means of atomic absorption spectrophotometer. Calcium and phosphorus in mg % were estimated colorimetrically by using test kits. The levels of Na and K in mg % were carried out by means of Flame photometer. Statistical analysis of the obtained data was done by means of soft ware computer program (SPSSWIN). Findings of the investigation of milk revealed the following:

1- Sodium: 66.67, 97.5, 12.12 and 3.33 of normal milk samples and 30, 64.52, 3.33 and 0 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain sodium in levels less than normal. However 25, 0, 66.67 and 46.67 % of normal milk samples and 70, 35.48, 90 and 63.33 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain sodium in levels higher than normal. The average \pm sd values of sodium in normal and mastitis milk were 52.5 ± 18.75 and 66.3 ± 14.9 mg/100 ml for milk from cows, 39.7 ± 7.9 and 57.5 ± 18.1 mg / 100 ml for milk from buffaloes, 71.76 ± 12.76 and 77.6 ± 11.5 mg / 100 ml for milk from sheep and 56.8 ± 16.5 and 64.4 ± 16.06 mg / 100 ml for milk from goats respectively. Sodium content in milk from animals with mastitis was significantly higher ($p < 0.05$) compared with that in healthy animals. Determination of sodium in milk was accurate method for predicting the infection status of the udder. Comparison of sodium concentration in milk from the studied four species revealed that there were a high level of sodium in sheep milk in comparison with that of cows, buffaloes and goats.

2- Potassium: 23.33, 27.5, 0 and 16.67 % of normal milk samples and 50, 64.52, 0 and 36.67 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain potassium in levels less than normal. However 78.33, 72.5, 100 and 73.33 % of normal milk samples and 50, 35.48, 73.33 and 33.3 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain potassium in levels higher than normal. The mean \pm sd values of potassium in normal and mastitis milk were 160 ± 37.5 and 130 ± 26.5 mg / 100 ml for milk from cows, 154 ± 30.77 and 122.5 ± 33.4 mg / 100 ml for milk from buffaloes, 205.7 ± 39.18 and 169.6 ± 56.5 mg / 100 ml for milk from sheep and 214 ± 67 and 160 ± 40.08 mg / 100 ml for milk from goats respectively. There was a significant decrease ($p < 0.05$) in potassium concentration in mastitis milk of the four species. Potassium concentration was significantly higher in milk from goats and sheep compared with that from cows and buffaloes.

3- Calcium: 40, 77.5, 18.18 and 76.67 % of normal milk samples and 42.5, 96.77, 50 and 80 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain calcium in levels less than normal, however 41.67, 5, 66.67 and 13.13 % of normal milk samples and 12.5, 0, 26.67 and 0 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain calcium in levels higher than normal. The mean \pm sd values of calcium in normal and mastitis milk were 110.8 ± 45.57 and 106.3 ± 24.54 mg % for milk from cows , 84.3 ± 28.63 and 70.03 ± 21.66 mg % for milk from buffaloes, 141.5 ± 26.49 and 112 ± 26.4 mg % for milk from sheep and 108.3 ± 21.95 and 99.97 ± 15.25 mg % for milk from goats respectively. Concerning mastitis, there were no significant variations in calcium concentration in normal and mastitis milk samples of cows and goat, however significant decrease was observed in milk from mastitic buffaloes and ewes. Comparing calcium levels in milk of the studied species revealed that there was a significant elevation in calcium concentration in normal milk of sheep and a significant reduction in normal and mastitis milk of buffaloes.

4- Phosphorus: 40, 7.5, 15.15 and 33.3 % of normal milk samples and 70, 45.16, 66.67 and 20 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain phosphorus in levels less than normal, however 51.67, 85, 84.85 and 33.3 % of normal milk samples and 22.5, 48.39, 30 and 60 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain phosphorus in levels higher than normal. The concentration levels of phosphorus in the studied normal and mastitis milk samples were, 97.97 ± 20.5 and 82.9 ± 18.68 mg % for milk from cows, 112.95 ± 16.5 and 95.42 ± 19.8 mg % for milk from buffaloes, 110.2 ± 17.59 and 87.3 ± 17.45 mg % for milk from sheep and 113.67 ± 22.56 and 101.4 ± 27.52 mg % for milk from goats, respectively. There was a significant increase ($p < 0.05$) in phosphorus levels in normal milk from cows, buffaloes, sheep and goats in comparison to its level in mastitis milk. Comparison of phosphorus concentration in milk samples of cows, buffaloes, sheep and goats revealed no significant variations among the studied species.

5- Magnesium: 48.3, 72.5, 24.24 and 0 % of normal milk samples and 90, 93.55, 43.3 and 10 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain magnesium in levels less than normal, however 40, 20, 75.76 and 100 % of normal milk samples and 2.5, 3.23, 43.3 and 76.67 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain magnesium in levels higher than normal. The mean \pm sd values of magnesium concentration in normal and mastitis milk samples were 12.4 ± 2.56 and 9.36 ± 1.89 for milk from cows, 10.6 ± 2.02 and 8.76 ± 2.14 for milk from buffaloes, 14.95 ± 3.15 and 12.11 ± 2.92 for milk from sheep and 24.3 ± 5.17 and 16.3 ± 3.8 for milk from goats respectively. Comparison of magnesium concentration in milk from healthy animal with that in milk from mastitis cases, revealed a highly significant increase of magnesium in normal milk of healthy species. Comparison of milk from each of the studied four species revealed a significant increase in magnesium content in milk of goats compared with that of cows, buffaloes and

sheep. Also milk from sheep showed a non significant high level of magnesium compared with that of cows and buffaloes.

6- Copper: Investigation of copper level in the studied milk samples revealed that: 46.67, 55, 0 and 23.33 % of normal milk samples and 45, 38.71, 3.33 and 60 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain copper in levels less than normal. However 51.76, 42.5, 96.97 and 46.67 % of normal milk samples and 35, 48.39, 80 and 16.67 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain copper in levels higher than normal. The concentration levels of copper in the studied normal and mastitis milk samples were, 0.41 ± 0.28 and 0.34 ± 0.25 ppm for milk from cows, 0.33 ± 0.27 and 0.34 ± 0.17 ppm for milk from buffaloes, 0.68 ± 0.28 and 0.59 ± 0.29 ppm for milk from sheep and 0.368 ± 0.14 and 0.25 ± 0.13 ppm for milk from goats, respectively. Regarding mastitis, there was a slight decrease (insignificant) in the copper level in milk from mastitic cows and sheep, however significant decrease was found in mastitis milk from goats. Comparison of copper concentration in milk samples from cows, buffaloes, sheep and goats, revealed a high significant increase of copper level in milk from sheep.

7- Iron: Investigation of iron level in milk samples of the present study revealed that; 50 % of mastitis milk samples from cows contain iron in levels less than normal. However 21.67, 93.94 and 30 % of normal milk samples and 7.5, 73.33 and 20 % of mastitis milk samples from cows, sheep and goats respectively; contain iron in levels higher than normal. All samples of milk from buffaloes contain normal iron levels. The mean \pm sd values of iron in the studied normal and mastitis milk were 2.1 ± 1.06 and 1.5 ± 0.8 ppm for cow's milk, 1.4 ± 0.65 and 1.12 ± 0.41 ppm for buffalo's milk, 4.47 ± 1.36 and 3.61 ± 1.26 ppm for sheep milk and 2.48 ± 1.05 and 1.96 ± 0.97 ppm for goat's milk respectively. Comparison of iron concentration in normal and mastitis milk revealed significant higher levels of iron in normal milk of cows, buffaloes, sheep and goats. Comparison of milk from different species revealed significant higher levels of

iron in milk from sheep compared with that of cows, buffaloes and goats. Iron in milk from goats some what higher than that of cows.

8- Zinc: 3.33, 17.5, 0 and 3.33 % of normal milk samples and 17.5, 12.9, 0 and 20 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively contain zinc in levels less than normal. However 95, 87.5, 100 and 96.97 % of normal milk samples and 80, 74.19, 100 and 80 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain zinc in levels higher than normal. The mean \pm sd values of zinc in the studied normal and mastitis milk were, 5.4 ± 2.05 and 4.4 ± 1.53 ppm for cow's milk, 5.7 ± 2.3 and 4.05 ± 0.45 ppm for buffalo's milk, 9.77 ± 3.17 and 8.077 ± 2.56 ppm for sheep's milk and 5.99 ± 1.52 and 4.22 ± 1.36 ppm for goat's milk respectively. Concerning mastitis there were a significant reduction ($p < 0.05$) in zinc level in mastitis milk of cows, sheep and goats and high significant reduction ($p < 0.01$) in mastitis milk of buffaloes in comparison to its level in normal milk. Regarding species differences, zinc level in milk from sheep was significantly higher ($p < 0.05$) than that in milk from cows, buffaloes and goats.

9-Manganese: 1.67, 12.5 and 45.45 % of normal milk samples and 2.5, 6.45 and 23.33 % of mastitis milk samples from cows, buffaloes and sheep respectively; contain manganese in levels higher than normal. Manganese level in all samples of milk from goats was in the normal range. There isn't any examined sample contain manganese in level less than normal. The estimated mean \pm sd values of manganese in the investigated normal and mastitis milk were; 0.3 ± 0.09 and 0.283 ± 0.08 ppm for milk from cows, 0.33 ± 0.11 and 0.23 ± 0.12 ppm for milk from buffaloes, 0.48 ± 0.29 and 0.35 ± 0.24 ppm for milk from sheep and 0.226 ± 0.068 and 0.19 ± 0.07 ppm for milk from goats respectively. With regard to differences in manganese level in normal and mastitis milk, there were slight decreases in manganese concentration in mastitis milk. Comparison of manganese level in milk of cows, buffaloes, sheep and goats revealed insignificant increase of manganese in milk of sheep and insignificant decrease of it in goat's milk.

10- Selenium: 25, 37.5, 36.36 and 53.33 % of normal milk samples and 7.5, 16.13, 13.33 and 16.67 % of mastitis milk samples from cows, buffaloes, sheep and goats respectively; contain selenium in levels higher than normal. There isn't any examined sample contain selenium in level less than normal. Selenium concentrations in normal and mastitis milk were 0.97 ± 0.45 and 0.65 ± 0.35 ppm for milk from cows, 1.26 ± 0.91 and 0.93 ± 0.49 ppm for milk from buffaloes, 1.2 ± 0.45 and 0.9 ± 0.3 ppm for milk from sheep and 1.37 ± 0.34 and 0.84 ± 0.39 ppm for milk from goats respectively. Concerning mastitis, there was a significant decrease ($p < 0.5$) in selenium concentration in mastitis milk of examined cases. With regard to species difference, only slight increase was observed in selenium level in milk of buffaloes, sheep and goats in comparison to that of cow's milk.

11 – Heavy metals (Lead and cadmium): Lead and cadmium were detected in all examined samples in levels higher than the permissible limit stated by WHO (1980) as well as that allowed in several countries such as; Germany, Nether land, Sweden and Australian.

The mean \pm sd concentrations of lead in normal and mastitis milk were, 0.7 ± 0.26 and 0.73 ± 0.22 ppm for milk from cows, 0.55 ± 0.15 and 0.58 ± 0.18 ppm for milk from buffaloes, 0.677 ± 0.26 and 0.84 ± 0.31 ppm for milk from sheep and 0.5 ± 0.135 and 0.36 ± 0.16 ppm for milk from goats respectively.

The estimated mean \pm sd values of cadmium in normal and mastitis milk in the present study were, 0.24 ± 0.078 and 0.3 ± 0.096 ppm for milk from cows, 0.18 ± 0.067 and 0.24 ± 0.099 ppm for milk from buffaloes, 0.1997 ± 0.108 and 0.26 ± 0.14 ppm for milk from sheep and 0.169 ± 0.07 and 0.16 ± 0.1 ppm for milk from goats respectively.

Concerning mastitis, there were no significant variations between Lead and cadmium levels in mastitis and normal milk. Comparison of milk samples from various species revealed insignificant difference among lead and cadmium levels in milk from cows, buffaloes, sheep and goats.