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INFLUENCE OF COMPOSITION OF FEED AND LACTATION PERIOD ON MINERAL COMPOSITION OF MARE'S

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ABSTRACT

Effects of lactation period and feed on essential minerals composition of mare's milk were studied. Average Ca, P, Na and Mg concentrations in feed DM were 0.66, 4.30, 0.13 and 2.21 $g.kg^{-1}$ of DM (dry matter), respectively. In regard to milk all elements concentrations were not similar to each other due to the changes of the lactation day differences. Average Ca, P, Na and Mg concentrations (in DM) caused by breed differences and lactation days were 1.95, 1.08, 0.53 and 0.22 $g.kg^{-1}$, respectively. During the milk period, a high-quality feed were maintaining the major mineral composition of mare's milk in 1.5 – 2 times higher amount than milk of mare fed with pasture and a low-quality feed. The colostrum stage of mare was much shorter than other animals, the mare's milk on the 2nd day of lactation showed properties of initial milk in which its composition, particularly in minerals, were two times higher when compare to that on milk periods that was started from 5th day after parturition. It seemed that some factors, except well physiological conditions, such as mare's age, number of foaling, sex of foal etc. did not affect milk nutrient composition.

Keywords: Mare's milk; nutrients; minerals; lactation periods; feed; lactating mare

INTRODUCTION

Milk is a fluid secreted by the female of all species of mammals or opaque white liquid produced by the mammary glands of mammals. Milk provides the complete nutritional requirements of the neonate of the species, as well as some immunological protection and other physiological requirements. The milk samples of all species are basically similar but there are very significant species-specific differences. Interspecies differences in the quantitative composition of milk probably reflect differences in the metabolic processes of the lactating mothers and in the nutritive requirements (amino acids, minerals, fatty acid, vitamins etc.) of the suckling. In regard with horse, now there are approximately 260 of different breeds of *Equus ferus caballus* all over world. The mare's milk is one of the basic foodstuffs for human populations in Kazakhstan, Kirghizia, Tadzhikistan, and Uzbekistan and mainly in Mongolia. Milk-alcoholic beverages, called Koumiss, Airag and Kumis, are traditionally produced through fermentation (**Montanari et al., 1996, Malacarne et al., 2002, Raffaela et al., 2004, Pikul and Wojtowski, 2008**). To the lesser extent, horses have been used as dairy herds (**Doreau et al., 2007**) in Eastern Europe (Belarus, Ukraine and Bulgaria) and Central Europe (especially Hungary).

The composition of mare's milk fat, in addition to the properties of its protein fractions and amino acid composition, suggests that this milk is more similar to human milk than cow's milk. For this reason, and because of the low cross-reactivity between cow's and mare's milk proteins, a clinical study has suggested that mare's milk could be used as a valid replacement for cow's milk in children with severe IgE-mediated cow's milk protein allergy (**Barello et al., 2008**). More recently, the consumption of mare's milk and other dairy foods, by virtue of their mineral, bioactive lipids, and protein components, have been shown to help reduce the risk of chronic disease disorders including osteoporosis, hypertension, excess body weight and body fat, dental caries, and some cancers (**Businco et al., 2000, Guradi et al., 2001, Official Methods, 1990**). Important nutrients are secreted by the mare to supply her foal with energy, protein, fat, carbohydrates, vitamins and minerals for optimal development and growth. To correct these nutrient losses and at the same time support maintenance requirements, lactating mares must consume adequate amounts of quality feeds. It is known that intake of minerals and amino acids are particularly important for the growth foals.

In the last decade, many studies have been carried out on equine milk and colostrums composition and, especially on proteins, lipids and minerals for above-mentioned reasons. Essential nutrients, such as minerals and amino acid composition, and vitamins of mare's milk have been reported, however, there is no complete study regarding mare's milk properties. Moreover, there are a few studies of relationship between changes of mare's milk main components and affecting factors such as lactation stage and other factors components.

Scientific hypothesis

This paper is focused on changes of contents of essential nutrients of mare's milk depending on lactation stages, feeding system and some breeds. Aims were to analyze the content of significant nutrient elements in mare's milk and feed for the lactating mare, as well as to characterize effects of several factors that affect milk properties such as lactation periods, breed and mare's diet. More specifically, the aims were i) to analyze changes in major minerals composition of mare's milk caused by differences of broodmares and periods of the first month after parturition, ii) to analyze the major mineral and dry matter of the feed used to feed the lactating broodmares on the lactation days after parturition, and iii) to study the relations between mineral composition of milk and feed. The final goal was to improve on knowledge of the relationship between minerals in mare's milk and their feed, and, if possible, to expand the findings on equine dairy science.

MATERIAL AND METHODOLOGY

Reagents and solutions:

Nitric acid (≥ 65%) and water (both Trace Select Ultra, Ultra Trace ACS, for trace analysis, Fluka, Buchs, Switzerland) were used. Standards of elements Ca, Na, Mg and P (Astasol-Ca, Astasol-Na, Astasol-Mg and Astasol-P were purchased from Analytika (Prague, Czech Republic). The working standard solutions were prepared by diluting the Ca, Na, Mg stock solutions 1.000 ±0.002 $g.L^{-1}$ in 2% nitric acid and P stock solution in 0.005% sulphuric acid. All working standard solutions were stored in polypropylene bottles. Deionised water was used for the preparation of all solutions.

All glassware was initially washed with detergent and water, and then the glassware was rinsed several times with deionised water and dried. Argon (purity 4.8) and nitrogen (purity 4.6) was from Messer (Prague, Czech Republic). All other reagents and solutions were of analytical grade purity (Sigma-Aldrich, St. Louis, MO, USA).

Equipments:

Microwave oven MarsXpress (CEM Corporation, Matthews, NC, USA) equipped with 20 mL PTFE high pressure vessels was used for microwave digestion (power 800 W). A Prodigy ICP-OES (Teledyne Leeman Labs, Hudson, NH, USA) was used for determination of calcium, phosphorus, sodium and magnesium contents in milk and feed. A vacuum lyophiliser Christ Alpha 1–4 (Martin Christ GmbH, Osterode am Harz, Germany) was used for sample vacuum lyophilisation.

Selection of broodmares and broodmare's milk samples
In order to collect mare's milk and colostrum samples, eight different broodmares were selected. All the mares of age from seven to sixteen years (10 ±3 parities) were mature and well-developed live weight between 500 and 600 kg. They were kept indoor and outdoor individually, and fed with 1.3 kg wheat bran and 2.1 kg homily, 5.0 kg oats in every day. Water was available months.

The milk samples (1P – 8P, approximately 40 – 50 mL each) were taken in plastic containers at days 2, 5, 10, and 28 days postpartum (Table 1, and in some cases also at 56th day for 1F and 2P). Before milking, foals were separated from their mothers for approximately 2 hours to prevent from suckling. First and second milking of mares was undertaken with hand milking as deep as possible. Some mares had not previously been subjected to any milking procedures. Collected milk samples were taken directly to the laboratory and frozen at -80 °C for 4 hours, and then lyophilised at 12 Pa and -40 °C using vacuum lyophilisation. All lyophilised samples were stored in fridge at -34°C until use for analysis.

The feed samples (in amount of approximately 500 g) were collected in polyethylene bags from feed mixture for the lactating mares at same days as milk sample collecting (Table 1). Then all samples were taken to the laboratory and stored at room temperature. All feed samples were homogenized and grounded. The feed samples were put into plastic containers and stored at room temperature until use for analysis.

Procedures

Analysis of dry matter

Dry matter (DM) was determined according to the standard procedure of AOAC. Grounded feed samples were dried in oven at 105 ±1 °C to constant weight for 5 h **(Official Methods, 1990, Nat. Res. Council, 1989).**

Element analysis

The minerals contents of all milk and feed samples were determined by inductively coupled argon plasma optical emission spectrometry (ICP-OES). The lyophilized milk (20 mg) and feed samples (20 mg) were accurately weighted and put into a 20 mL high pressure PTFE containers and 4 mL digestion mixture (2 mL deionised water and 2 mL 65% nitric acid) were added and placed in the microwave oven for mineralization. Program of decomposition consisted of three steps i) temperature increase to180 °C in 25 minutes, ii) maintaining the temperature at 180 °C for 10 minutes and iii) cooling at room temperature. Afterwards, mineralized samples were put into 10 mL volumetric flasks and diluted with deionised water up to 10 mL for mineral analysis.

Calcium, phosphorus, sodium and magnesium contents (on DM basis) of milk and feed were determined by using ICP-OES. After scanning a blank, a standard solution and a sample solution in the programmed wavelength range, the background correction wavelengths were selected manually at appropriate background positions for each analyte signal. The detection limits of the method at the pre-selected wavelengths (Ca – 317.933 nm, P – 214.914 nm, Na – 588.995 nm and Mg – 280.271 nm) were high enough and permitted the determination. For each sample three determinations were performed. Instrument configuration and general experimental conditions are summarized in Table 2.

Statisic analysis

All data on the content of four minerals in DM of all samples are expressed as arithmetic mean accompanied by a standard deviation (mean ± SD), with outlying results excluded by performing a Dean-Dixon test (Q-test). To calculate arithmetic average and standard deviation was used by Office Excel® Microsoft. Data were evaluated by producing summary statistics and analyzing the variance using an ANOVA. Single Factor Scattering and Scale Comparisonthe Scheffé's method was implemented using the statistical program QC Expert 3.3 (Trilobite Statistical Software, Pardubice, CZ). Level of significance single factor analysis of scattering and pair comparison by Scheffé's method was set to 5% (p <0.05). In regard to lyophilized milk samples, all data revealed by ICP-OES were converted to the mean on fresh milk.

RESULTS AND DISCUSSION

Dry matter of the feed

The dry matter (DM) content of all feed samples for the lactating mares is showed in Table 3. The percentage of DM content of every feed sample was a little different from each other due to the preparation conditions of feed mixture and the ratio of forages in feed mixture. The average DM content of the feed was 89.2 ±0.7 % w/w (p <0.05) or in the range from 88.6 to 90.6%.

According to some authors **(Berg, 2009)** the DM content of some high quality feed types for the lactating mare such as grains, hay and chaff ranged from 87 to 91% (w/w). These results of DM content of feed samples also were in agreement with National Research Council (NRC) values **(Nat. Res. Council, 1989)**. DM content of all feeds, therefore, had the standard level of DM of feed and forage for the lactating mare.

Mineral composition of feed

The concentration ranges of major four elements (Ca, P, Na and Mg) determined in the 29 feed samples for lactating broodmares on the 2nd, 5th, 10th, 28th and 56th days after parturition are summarized in Tables 4 – 7. These data were expressed as mean ± SD (p <0.05). There were significant differences among the feeds with regard to four elements content. For all feed samples, significant changes of contents of minerals were observed causing by days after parturition. Ca, P, Na and Mg concentrations (on a DM basis) were respectively 0.66 – 0.78, 4.95 – 5.05, 0.13 – 0.15 and 2.59 – 2.61 $g.kg^{-1}$ in feed. Also these tables show the average content of these four elements in feed samples related to same lactation days.

As shown in Table 4, generally, the average concentrations of Ca in feeds for the lactating mares on 5th day after parturition was higher 0.72 ±0.18 $g.kg^{-1}$ than other days, and on 28th, was the lowest 0.57 ±0.05 $g.kg^{-1}$. Calcium average concentration of feeds caused by kinds of forage and feed for horse ranges from 0.4 to 17.1 $g.kg^{-1}$ and the feed type of grains such as oats and wheat, in, particular, ranges from 0.5 to 0.9 $g.kg^{-1}$ **(Berg, 2009)**. Ca concentrations of 1F and 3F on 5th and, the on 10th days after parturition, were increased gradually and then decreased. In regard to 2F, 5F, 7F and 8F, slight decrease, from the 2nd to 28th, was observed. Concentrations of 4F and 6F minerals were increased on the 5th day, but afterwards, decreased dramatically. The average concentration of Ca in the feed ranged from 0.70 ±0.14 to 0.57 ±0.05 $g.kg^{-1}$. Generally, Ca concentration increased gradually at interval between the 2nd and 5th days 0.70 ±0.14 and 0.72 ±0.18 $g.kg^{-1}$, and on 10th and 56th days, then decreased slightly 0.61 ±0.23 and 0.57 ±0.05 $g.kg^{-1}$. The average Ca concentration of all feed samples depending on days was 0.66 ±0.09 $g.kg^{-1}$ and these all results of Ca concentration in feed samples were in agreement with NRC values to feed the lactating mare **(Berg, 2009, Saastamoinen and Koskinen, 1993)**.

Table 5 presents P concentrations of feeds. As it results from the table, feeds for mares on the 2nd day were characterized by the highest P concentrations (4.54 ±0.69) $g.kg^{-1}$ when compared with the 5th, 10th, 28th and 56th days, 4.03 ±0.32, 4.05 ±0.51, 4.41 ±0.52 and 4.44 ±0.01 $g.kg^{-1}$, respectively. According to NRC **(Nat. Res. Council, 1989)**, daily P requirement of the lactating mare is 31 – 51 $g.kg^{-1}$ and average P concentration of feeds caused by kinds of forage and feed for horse ranges from 1.0 to 12.7 $g.kg^{-1}$ and the feed type of grains such as oats and wheat, pasture and hay/chaff, particularly, ranges from 2.0 to 4.1 $g.kg^{-1}$. P concentrations of 1F, 2F, 4F and 6F on the 5th day decreased, although on the 10th day, increased slightly again. In regard to 3F, 5F, 7F and 8F, P concentrations decreased continuously and slightly. The average P concentration of all feed samples depending on experiment days was 4.30 ±0.26 $g.kg^{-1}$, and these all results of P concentration in feed samples were in agreement with NRC to feed the lactating mare **(Berg, 2009, Saastamoinen and Koskinen, 1993)**.

Na concentrations of feeds are shown in Table 6. It was observed that the highest Na concentration was in 8F on 10th day as 0.21 ±0.01 $g.kg^{-1}$, and it was 0.12 ±0.01 $g.kg^{-1}$, on the 2nd day. The Na concentration of all feed samples, except 5F, 6F and 7F, increased gradually during lactation days. The average Na concentrations of all feeds on lactation days were 0.12 ±0.01, 0.13 ±0.02, 0.13 ±0.03 and 0.14 ±0.02 $g.kg^{-1}$, respectively. On the other hand, Na content rose slightly till 56th day and approximately constant on the 5th and 10th days. The average Na content was 0.13 ±0.01 $g.kg^{-1}$. Mg concentrations of feeds were not similar to each others, which are shown in Table 7. On the other hand, an increase and decrease of Mg content of all feeds were observed in different ranges caused by lactating days, and the average Mg concentration was 2.21 ±0.12 $g.kg^{-1}$.

According to NRC **(Nat. Res. Council, 1989)**, daily Mg requirement of the lactating mare is 12.0 – 15.2 $g.kg^{-1}$. Mg contents of 1F and 6F on the 10th day, 2F, 3F, 4F, 7F an 8F on the 2nd day and 5F on the 5th day were the highest compare to that on other days, 2.59 ±0.01, 2.59 ±0.01, 2.44 ±0.01, 2.78 ±0.01, 2.45 ±0.03, 2.16 ±0.03 and 2.26 ±0.01 $g.kg^{-1}$, respectively. The average Mg concentrations of feeds for lactating mares demonstrated that Mg contents on the 5th an 10th days were constant and, further on the 28th, increased gradually again. Overall, the feed mineral composition is influenced by many factors such as storage condition, contamination, and ratio of feed mixtures **(Csapó-Kiss et al., 1995)**.

Table 1 Milk and feed samples characteristics.

Milk	Days postpartum				Feed	Days postpartum			
	2	5	10	28		2	5	10	28
1P[a]	+	+	+	+	1F	+	+	+	+
2P[b]	+	+	+	+	2F	+	+	+	+
3P	+	+	+	+	3F	+	+	+	+
4P	+	+	+	+	4F	+	+	+	+
5P	+	+	+	+	5F	+	+	+	-
6P	+	+	+	-	6F	+	+	+	-
7P	+	+	+	-	7F	+	+	+	-
8P	+	+	+	+	8F	+	+	+	-

Note: +, sample was taken; -, sample was not taken, a a sample on 56th day was also collected for 1P, b a sample on 56th day was also collected for 2P.

Table 2 ICP-OES operating conditions.

Operating conditions		Operating conditions	
Radio frequency	27.12 Hz	Outer gas flow rate	Ar 17 L/min
Radio frequency power	2.5kW	Intermediate gas flow rate	Ar 1 L/min
Plasma's temperature	8000 – 9000 K	Carrier gas flow rate	Ar 1 L/min

Table 3 The dry matter content of feed (%, w/w).

Feed	Dry matter (mean value ±SD)			
	2[nd]	5[th]	10[th]	28[th]
1F[a]	89.38 ±0.19	89.32 ±0.15	89.04 ±0.51	88.96 ±0.66
2F	90.56 ±0.67	88.64 ±0.34	89.54 ±0.37	89.36 ±0.55
3F	90.32 ±0.54	89.55 ±0.86	88.69 ±0.98	89.04 ±0.22
4F	88.89 ±0.42	89.14 ±0.51	89.06 ±0.20	88.90 ±0.08
5F	89.64 ±0.40	88.99 ±0.70	88.65 ±0.68	n.a.
6F	90.12 ±0.39	88.80 ±0.57	88.81 ±0.58	n.a.
7F	90.47 ±0.31	89.37 ±0.37	88.81 ±0.53	n.a.
8F	90.00 ±0.63	88.80 ±0.39	88.7 ±0.51	n.a.

Note: F – feed samples; SD – standard deviation; n.a. – not analyzed, a DM = (89.49 ±0.51) on 56th day.

Mineral composition of milk

The nutritive minerals found in mare's milk as mainly ionized form are essential for nutritional and metabolic functions of newborn neonate. The data on major essential mineral composition of milk from different eight broodmares fed with supplement feed mixture are given in Tables 3 – 7. All minerals concentrations were described as mean ±SD (g.kg^{-1}). The results of mineral concentration of milk samples demonstrated that Ca, P, Na and Mg contents of broodmares' milk were not similar to each other due to breed differences and lactation days. As shown in Table 4, Ca concentrations of 5P 2.82 ±0.01 g.kg^{-1} and 8P 2.72 ±0.01 g.kg^{-1} on the 2nd lactation day were the highest in that of others, which were two times higher than that reported by Csapo-Kiss (**Csapó-Kiss et al., 1995**), while the lowest Ca concentration 0.66 ±0.01 g.kg^{-1} in 1P on the 56th day which was comparable to the mean of literature values. As reported in the literature, the mineral level of mare's milk caused by lactation stage differs greatly, which ranges 0.8 – 1.3 g.kg^{-1} on 4 – 180 lactation days. In many papers, Ca level is higher in colostral stage, in which milk is rich in proteins, than milk stage because of Ca ions play an important role in the structure and stability of casein micelles **(Csapó et al., 2009)**. From dependence on lactation days, Ca concentrations of 1P and 5P decreased gradually from the 2nd to 5th and 10th days and then increased on the 28th day. In regard to others, a decrease on the 5th and 28th days and an increase on the 10th day were observed. The observations were in agreement with major papers **(Csapó-Kiss et al., 1995, Schryver et al., 1986, Summer et al., 2004, Cisla et al., 2009, Nascimento et al., 2010)**.

With regard to phosphorus concentration of studied broodmares milk, the highest amounts were observed in all milk on the 2nd day during the lactation periods, which ranged from 1.93 g.kg^{-1} to 0.94 g.kg^{-1} caused by breed of broodmares. P concentration of 5P was the highest on the 2nd day while the lowest in 1P, but it was declined continuously on other days. Some authors reported that P content ranged from 0.7 to 0.9 g.kg^{-1} on 0 – 45 days of the lactation **(Schryver et al., 1986)**. Although, mineral content is caused significantly by type of breed and feed which are the most powerful affecting factors on it **(Summer et al., 2004)**.

Table 4 Ca content in feed and milk (g.kg^{-1}).

Feed	ADM (%)	Ca content (mean value ±SD)				AC[2]	Milk	Ca content (mean value ±SD)				AC[2]
		2nd	5th	10th	28th	2-56th days		2nd	5th	10th	28th	2-56th days
1F[3]	89.24 ±0.23	0.83 ±0.02	1.03 ±0.01	1.13 ±0.00	0.64 ±0.00	0.86 ±0.22	1P	1.43 ±0.03	0.77 ±0.01	0.66 ±0.00	0.97 ±0.01	0.99 ±0.31
2F	89.53 ±0.79	0.95 ±0.00	0.80 ±0.00	0.55 ±0.01	0.59 ±0.00	0.72 ±0.19	2P	1.81 ±0.01	2.06 ±0.00	2.75 ±0.02	1.26 ±0.01	1.97 ±0.62
3F	89.40 ±0.71	0.68 ±0.00	0.69 ±0.00	0.73 ±0.00	0.52 ±0.00	0.65 ±0.09	3P	2.38 ±0.00	2.35 ±0.00	2.67 ±0.00	n.a.	2.47 ±0.18
4F	88.99 ±0.12	0.62 ±0.01	0.89 ±0.00	0.61 ±0.00	0.55 ±0.01	0.67 ±0.15	4P	2.44 ±0.01	2.06 ±0.01	2.15 ±0.02	1.97 ±0.01	2.16 ±0.20
5F	89.09 ±0.50	0.68 ±0.01	0.62 ±0.00	0.45 ±0.00	n.a.	0.58 ±0.12	5P	2.82 ±0.01	2.35 ±0.02	1.43 ±0.01	2.57 ±0.02	2.29 ±0.60
6F	89.24 ±0.76	0.56 ±0.00	0.75 ±0.00	0.47 ±0.00	n.a.	0.59 ±0.14	6P	n.a	1.91 ±0.01	n.a.	n.a.	-
7F	89.55 ±0.84	0.53 ±0.01	0.51 ±0.00	0.49 ±0.00	n.a.	0.50 ±0.03	7P	2.35 ±0.02	n.a.	1.82 ±0.03	n.a.	2.09 ±0.37
8F	89.17 ±0.72	0.76 ±0.01	0.51 ±0.00	0.48 ±0.00	n.a.	0.58 ±0.15	8P	2.72 ±0.00	n.a.	n.a.	n.a.	-
AC[1]	**89.28 ±0.20**	**0.70 ±0.14**	**0.72 ±0.18**	**0.61 ±0.23**	**0.57 ±0.05**	**0.66 ±0.10[a]**	**AC[1]**	**2.28 ±0.50**	**1.92 ±0.59**	**1.91 ± 0.79**	**1.70 ±0.72**	**1.95 ±0.24[a]**

Note: ADM – average dry matter of feed; [3] (0.66 ±.01) and (0.661 ± 0.002) for 1F and 2P on 56th day; AC[1] – average content in feeds at same lactation days; AC[2] – average content in feeds at different lactation days; [a] – average content of AC[1]; n.a. – not analyzed.

Table 5 P content in feed and milk (g.kg^{-1}).

Feed	ADM (%)	P content (mean value ±SD)				AC[2]	Milk	P content (mean value ±SD)				AC[2]
		2nd	5th	10th	28th	2-56th days		2nd	5th	10th	28th	2-56th days
1F[3]	89.24 ±0.23	4.39 ±0.01	4.29 ±0.01	4.93 ±0.03	4.52 ±0.01	4.51 ±0.25	1P	0.94 ±0.01	0.39 ±0.00	0.30 ±0.00	0.39 ±0.00	0.50 ±0.26
2F	89.53 ±0.79	5.12 ±0.02	3.99 ±0.01	4.30 ±0.02	3.76 ±0.00	4.29 ±0.59	2P	1.10 ±0.01	1.00 ±0.00	1.30 ±0.02	0.52 ±0.00	0.98 ±0.33
3F	89.40 ±0.71	4.59 ±0.01	3.40 ±0.01	3.83 ±0.02	4.35 ±0.01	4.19 ±0.34	3P	1.41 ±0.00	0.96 ±0.01	1.46 ±0.01	n.a.	1.28 ±0.28
4F	88.99 ±0.12	5.61 ±0.04	3.68 ±0.01	4.10 ± .00	5.02 ±0.02	4.60 ±0.88	4P	1.85 ±0.01	1.14 ±0.01	1.00 ±0.00	0.83 ±0.00	1.20 ±0.45
5F	89.09 ±0.50	3.27 ±0.01	4.46 ±0.01	3.43 ±0.01	n.a.	3.72 ±0.65	5P	1.93 ±0.00	1.31 ±0.00	0.69 ±0.00	1.70 ±0.00	1.41 ±0.54
6F	89.24 ±0.76	4.30 ±0.01	4.20 ±0.00	4.42 ±0.03	n.a.	4.31 ±0.11	6P	n.a	1.66 ±0.00	n.a.	n.a.	-
7F	89.55 ±0.84	4.84 ±0.03	4.13 ±0.01	3.98 ±0.03	n.a.	4.32 ±0.46	7P	1.56 ±0.00	n.a.	0.63 ±0.01	n.a.	1.09 ±0.54
8F	89.17 ±0.72	4.22 ±0.03	3.49 ±0.03	3.44 ±0.02	n.a.	3.72 ±0.44	8P	1.69 ±0.00	n.a.	n.a.	n.a.	-
AC[1]	**89.28 ±0.20**	**4.54 ±0.69**	**4.03 ±0.32**	**4.05 ±0.51**	**4.41 ±0.52**	**4.30 ±0.26**	**AC[1]**	**1.50 ±0.37**	**1.08 ±0.42**	**0.90 ±0.44**	**0.86 ±0.59**	**1.08 ±0.29[a]**

Note: ADM – average dry matter of feed; [3] (4.44 ±0.01) and (0.481 ±0.001) for 1F and 1P on 56th day; AC[1] – average content in feeds at same lactation days; AC[2] – average content in feeds at different lactation days; [a] – average content of AC[1]; n.a. – not analyzed.

P content of 1P was lowest on all days when compared to others which ranged from 0.39 to 0.94 g.kg^{-1}, but it was in agreement with data on previous studies **(Summer et al., 2004, Solaroli et al., 1993)**.

Generally, it is reported that P content is declined after colostral period, and in the case of present study, P concentration of all milk, except 3P and 5P, decreased gradually after the 2nd day which is shown in Table 5.

The concentration of Na in milk exists in lower amount than that of Ca and P, because Ca and P are associated in form of the colloid calcium phosphate, which are a large class of milk proteins **(Solaroli et al., 1993)**.

Table 6 Na content in feed and milk (g.kg^{-1}).

Feed	ADM (%)	Na content (mean value ±SD)				AC[2]	Milk	Na content (mean value ±SD)					AC[2]
		2nd	5th	10th	28th	2-56th days		2nd	5th	10th	28th	2nd	2-56th days
1F[3]	89.24 ±0.23	0.12 ±0.00	0.13 ±0.00	0.13 ±0.00	0.13 ±0.00	0.13 ±0.01	1P	0.60 ±0.01	0.18 ±0.00	0.15 ±0.00	0.32 0.00	0.31 ±0.18	0.60 ±0.01
2F	89.53 ±0.79	0.13 ±0.00	0.14 ±0.00	0.12 ±0.00	0.17 ±0.00	0.14 0.02	2P	0.68 ±0.01	0.34 ±0.00	0.36 ±0.01	0.21 ±0.00	0.40 ±0.20	0.68 ±0.01
3F	89.40 ±0.71	0.13 ±0.00	0.12 ±0.00	0.14 ±0.00	0.14 ±0.00	0.13 ±0.01	3P	0.81 ±0.00	0.64 ±0.01	0.82 ±0.00	n.a.	0.76 ±0.11	0.81 ±0.00
4F	88.99 ±0.12	0.13 ±0.00	0.11 ±0.00	0.11 ±0.00	0.12 ±0.00	0.12 0.01	4P	0.97 ±0.00	0.49 ±0.01	0.33 ±0.01	0.55 ±0.00	0.58 ±0.28	0.97 ±0.00
5F	89.09 ±0.50	0.11 ±0.00	0.14 ±0.00	0.11 ±0.00	n.a.	0.12 ±0.02	5P	0.87 0.00	0.41 ±0.00	0.33 ±0.00	0.73 ±0.01	0.59 ±0.26	0.87 ±0.00
6F	89.24 ±0.76	0.13 ±0.00	0.15 ±0.00	0.11 ±0.00	n.a.	0.13 ±0.02	6P	n.a.	0.88 ±0.01	n.a.	n.a.	-	n.a.
7F	89.55 ±0.84	0.11 ±0.00	0.14 ±0.00	0.11 ±0.00	n.a.	0.12 ±0.02	7P	0.79 ±0.01	n.a.	0.34 ±0.00	n.a.	0.57 ±0.32	0.79 ±0.01
8F	89.17 ±0.72	0.12 ±0.00	0.10 ±0.00	0.21 ±0.00	n.a.	0.14 ±0.06	8P	0.87 ±0.01	n.a.	n.a.	n.a.	-	0.87 ±0.01
AC[1]	**89.28 ±0.20**	**0.12 ± .01**	**0.129 ±0.02**	**0.13 ±0.03**	**0.14 ±0.02**	**0.13 ± .01[a]**	**AC[1]**	**0.80 ±0.13**	**0.49 ±0.24**	**0.39 ±0.23**	**0.45 ±0.23**	**0.53 ±0.18[a]**	**0.80 ±0.13**

Note: ADM – average dry matter of feed; [3] (0.13 ±0.01) and (0.28 ±0.00) for 1F and 2P on 56th day; AC[1] – average content in feeds at same lactation days; AC[2] – average content in feeds at different lactation days; **[a]** – average content of AC[1]; n.a. – not analyzed.

Table 7 Mg contents in feed (g.kg^{-1}).

Feed	ADM (%)	Mg contents (mean value ±SD)				AC[2]	Milk	Mg contents (mean value ±SD)				AC[2]
		2nd	5th	10th	28th	2-56th days		2nd	5th	10th	28th	2-56th days
1F[3]	89.24 ±0.23	2.36 ±0.04	2.26 ±0.02	2.59 ±0.01	2.46 ±0.00	2.39 ±0.14	1P	0.16 ±0.00	0.08 ±0.00	0.05 ±0.00	0.08 ±0.00	0.09 ±0.04
2F	89.53 ±0.79	2.59 ±0.01	2.06 ±0.00	2.16 ±0.02	1.95 ±0.01	2.19 ±0.28	2P	0.24 ±0.00	0.22 ±0.00	0.28 ±0.00	0.11 ±0.00	0.21 ±0.07
3F	89.40 ±0.71	2.44 ±0.01	2.04 ±0.01	1.89 ±0.01	2.31 0.01	2.19 ±0.25	3P	0.27 ±0.00	0.22 ±0.00	0.34 ±0.00	n.a.	0.28 ±0.06
4F	88.99 ±0.12	2.78 ±0.01	2.00 ±0.01	2.03 ±0.01	2.50 ±0.01	2.33 ±0.38	4P	0.33 ±0.00	0.23 ±0.00	0.19 ±0.00	0.179 ±0.00	0.23 ±0.07
5F	89.09 ±0.50	1.73 ±0.01	2.26 ±0.01	1.76 ±0.01	n.a.	1.91 ±0.30	5P	0.33 ±0.00	0.26 ±0.00	0.14 ±0.00	0.30 ±0.00	0.26 ±0.08
6F	89.24 ±0.76	2.26 ±0.00	2.18 ±0.00	2.26 ±0.02	n.a.	2.23 ±0.04	6P	n.a.	0.35 ±0.00	n.a.	n.a.	-
7F	89.55 ±0.84	2.45 ±0.03	2.10 ±0.00	1.96 ±0.01	n.a.	2.17 ±0.26	7P	0.31 ±0.00	n.a.	0.17 ±0.00	n.a.	0.24 ±0.10
8F	89.17 ±0.72	2.16 ±0.03	1.76 ±0.00	1.81 ±0.01	n.a.	1.91 ±0.22	8P	0.33 ±0.01	n.a.	n.a.	n.a.	-
AC[1]	**89.28 ±0.20**	**2.34 ±0.31**	**2.08 ±0.16**	**2.06 ±0.28**	**2.30 ±0.25**	**2.210 ±0.122[a]**	**AC[1]**	**0.28 ±0.06**	**0.22 ±0.09**	**0.20 ±0.10**	**0.16 ±0.10**	**0.22 ±0.05[a]**

Note: ADM - average dry matter of feed; [3] (2.26 ±0.01) and (0.09 ±0.00) for 1F and 2P on 56th day; AC[1] – average content in feeds at same lactation days; AC[2] – average content in feeds at different lactation days; **[a]** – average content of AC[1]; n.a. – not analyzed.

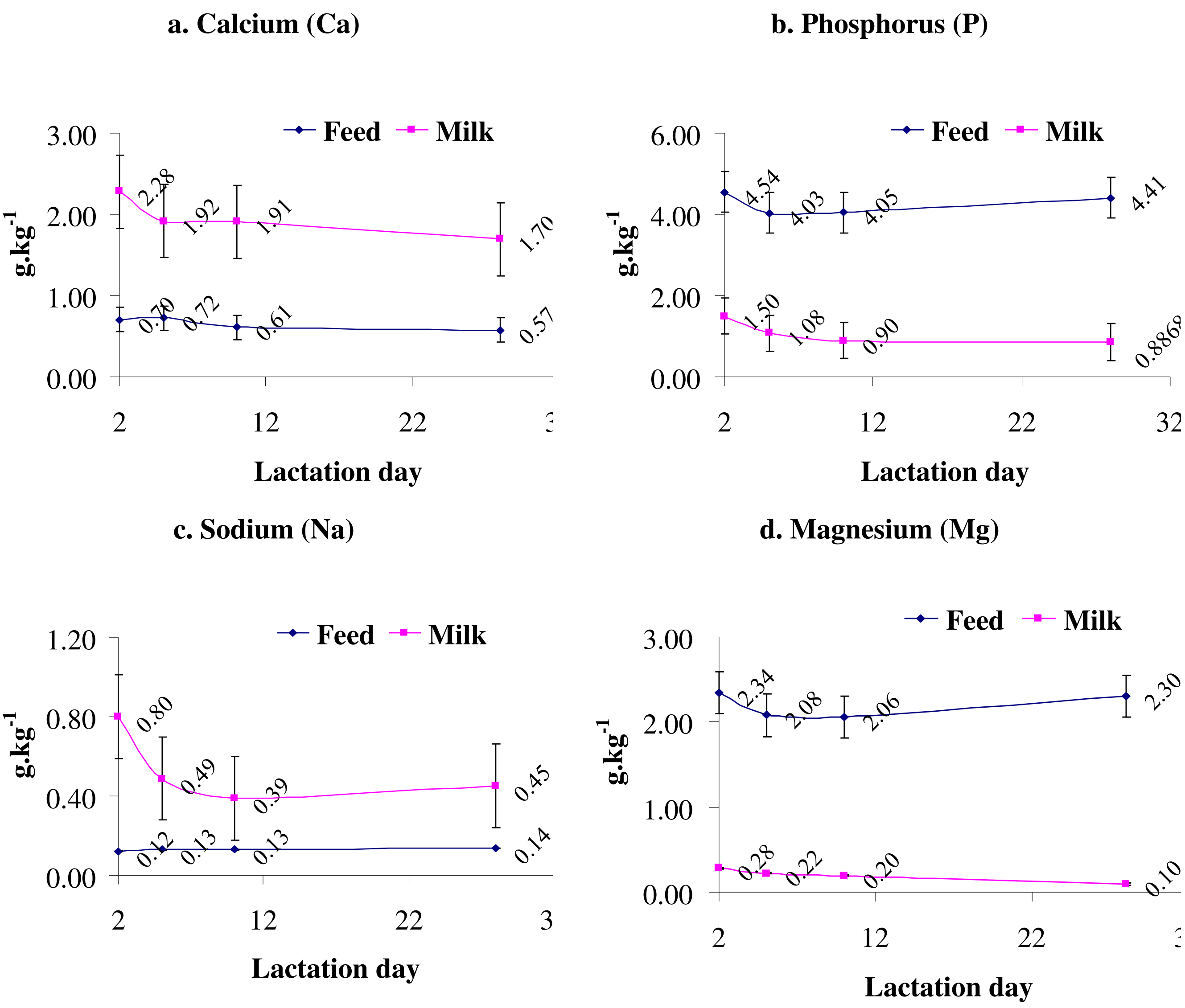

Figure 1a-d. Relation between average Ca, P, Na, Mg contents in milk and feed.

Na content of mare's milk is lower than other farm animals **(Sarwar et al., 1998)**. According to the data shown in Table 6, significantly differences among Na contents of milk during lactation days were observed. Na concentration of all milk on the 2nd day was the highest and the lowest on the10th day. The highest Na amount 0.97 $g.kg^{-1}$ was in 4P which was three times higher than the value reported in literature and the lowest 0.21 $g.kg^{-1}$ in 2P, which was in agreement with some authors **(Sarwar et al., 1998)**. However, many papers have shown different values of Na content of equine milk caused by breed and feed **(Schryver et al., 1986, Gálik et al., 2012)**. Na concentration of 1P per each day of lactation was lower than others and of 2P, 4P and 5P was almost similar to each other during lactation days. In regard to 3P, the highest concentration 0.82 $g.kg^{-1}$ was on the 10th day. According to some reports in the literature that Na concentration is high at the beginning of colostrum and then declines gradually, 1P, 2P and 7P were comparable to it, but of 3P, 4P and 5P decreased till the 10th and then increased on the 28th day of lactation.

Table 7 shows Mg concentration of all mares' milk during lactation periods. The only Mg concentration of 1P on all
days, which ranged from 0.16 $g.kg^{-1}$ to 0.05 $g.kg^{-1}$, was in agreement with earlier studies 50 – 180 $mg.L^{-1}$ of fresh milk **(Summer et al., 2004, Nascimento et al., 2010)**, but also Mg concentrations of 2P and 3P on the 2nd day were less higher, 0.24 $g.kg^{-1}$ and 0.27 $g.kg^{-1}$ respectively. It was two times higher than the mean of the literature values in the case of others on the 2nd, 5th and 28th days. The highest Mg concentrations were observed in all milk on the 2nd day of the lactation, after that it was decreased dramatically.

The average mineral concentrations depended on breed during the lactation periods is also shown in Tables 4 – 7. Average Ca concentration in all broodmare's milk caused by breed difference was much higher (ranged from 1.70 to 2.28 $g.kg^{-1}$) than other three major elements P, Na and Mg in milk and ratios of Ca to the others were 1.8:1, 3.7:1 and 9:1, respectively which was comparable to the some literature values **(Schryver et al., 1986, Summer et al., 2004)**.

Figure 2 Mare with foal (source: https://zlinsky.denik.cz/galerie/).

P was the second major element in mare's milk in regard to its amount and the average P concentration ranged from 0.86 to 1.50 g.kg^{-1} during the 1st month during lactation. With regard to Na, the highest amount was observed on 2nd day as 0.80 g.kg^{-1} and then two times dropped on the 5th day until 0.49 g.kg^{-1}. The average Mg decreased gradually after the 2nd day of lactation until 0.16 g.kg^{-1} on the 28th day and it was 1.5 times lower than the 2nd day of lactation.

Overall, Ca, P, Na and Mg concentrations in mare's milk were high in the beginning of the lactation periods and then declined as approximately 1.5 times on the 5th day of lactation and continuously till the 28th day. These results on four minerals were in agreement with and comparable to results of previous researches **(Csapó-Kiss et al., 1995, Schryver et al., 1986, Summer et al., 2004, Nascimento et al., 2010, Solaroli et al., 1993, Sarwar et al., 1998, Gálik et al., 2012, Martin et al., 1992)**.

Relation between the mineral composition of feed and milk

The changes in average content of major four elements in different eight broodmares' milk and feed for them depended on the 2nd, 5th, 10th and 28th days of the lactation and the relations between them are given in Figure 1. All data were expressed as mean ± SD, which indicated significant differences between changes in mineral content of milk and feed. Generally, lactation period is the most important factor influencing on mare's milk composition. The results conducted by changes in mineral content of milk showed that except Na, other three mineral Ca, P and Mg contents decreased gradually from 2.28, 4.54 and 0.28 to 1.92, 4.03 and 2.08 g.kg^{-1} during lactation periods, in particular on the 5th day of lactation, all minerals contents were declined significantly and further, decreased slightly. Some author reported that the ash content did not vary or decreased only slightly and the effect of the lactation period on mare's milk composition was similar for different breed **(Martin and Doreau, 2006)**. However, the results of present study on milk mineral were not in agreement with above report. Moreover, it was in agreement with many results demonstrated in the literature that with regard to ash composition of mare's milk, the highest ash content is observed during the 1st week of lactation period. Afterward, total ash content regularly decreased, due to a decline of all minerals, the ash content in the later stages of lactation wais about 39% lower than that of the earlier stages **(Summer et al., 2004, Martuzzi et al., 1997)**. In regard to Na concentration of milk decreased till the 10th day of lactation and then, on 28th day, a slight increase was observed. Also it was similar to results reported by some authors **(Summer et al., 2004, Martuzzi et al., 1997)**. The changes in mineral concentration of feeds for the lactating broodmares during lactation were not similar to each other. Ca and Mg concentration decreased during lactation period while P and Na increased gradually, except a slight decrease on the 5th day of lactation. In fact, the mineral composition of these feeds depended greatly on preparation of feed mixture.

As shown in Figure 1a-d, there was no significant relation between changes in average mineral content of milk and feed. Although, P and Mg concentrations in milk and feed on the 5th day of lactation decreased dramatically, perhaps it was due to, in the case of milk, ending of colostral period. In contrast, Ca and Na concentration of milk decreased considerably while Ca and Na in feed increased. Further, Ca, P, Na and Mg concentration in milk on the 2nd, 5th, 10th and 28th days after parturition to feed ratios were found in amounts of Ca - 3.2:1, 2.6:1, 3:1 and 3:1, P - 1:3, 1:3.7, 1:4.2 and 1:5.1, Na - 6.6:1, 3.8:1, 3:1 and 3.2:1, Mg - 1:8.4, 9.5:1, 10.3:1 and 23:1, respectively. Overall, changes in milk mineral content caused by lactation periods were not correlated with that of feed, it was in agreement with some authors **(Martuzzi et al., 1997, Hidiroglou and Proulx, 1982)**, perhaps, due to milk production metabolic functions of the lactating mare and mineral transfer on it **(Kavazik et al., 2002)**.

CONCLUSION

All results revealed indicated significant relations and changes in major minerals composition of different lactating mares' milk depending on the lactation periods and feed used to feed the mares. The average DM content of the feed was 89.2 ±0.72 % w/w (p <0.05). The highest Ca, P, Na and Mg concentration of feed were 1.13 g.kg^{-1} in 1F on the 10th day, 5.02 g.kg^{-1} in 4F on the 28th, 0.21 g.kg^{-1} in 8F 10th and 2.59 g.kg^{-1} in 1F 10th, respectively while the lowest concentration were 0.47 g.kg^{-1} in 5F on 10th, 3.27 g.kg^{-1} in 5F on 2nd, 0.11 g.kg^{-1} in 5F on 10th and 1.73 g.kg^{-1} in 5F on 2nd day, respectively. Average Ca, P, Na and Mg concentrations of all feeds caused by all days were 0.66, 4.30, 0.13 and 2.21 g.kg^{-1}, respectively. In regard to milk minerals, the highest Ca, P, Na and Mg concentration were observed in amounts of 2.82 g.kg^{-1} in 5P on 2nd, 1.93 g.kg^{-1} in 5P on 2nd, 0.97 g.kg^{-1} in 4P on 2nd and 0.32 g.kg^{-1} in 5P and 8P on 2nd day, respectively, when compared to the lowest were 0.66 g.kg^{-1}, 0.30 g.kg^{-1}, 0.05 g.kg^{-1} and 0.15 g.kg^{-1} in 1P on 10th day, respectively. All minerals concentrations were not similar to each other due to changes of the lactation days and breed differences. Average Ca, P, Na and Mg concentrations caused by breed differences and lactation days ranged from 1.95, 1.08, 0.53 to 0.22 g.kg^{-1}, respectively. Generally Ca and Na concentrations of feed were much lower and P and Mg concentration were higher than that of milk. The ratios of the average Ca, P, Na and Mg concentrations of milk caused by lactation days and breed difference to those of all feeds were 3:1, 1:4, 4:1 and 1:10, respectively.

These ratios, except Mg, were kept during all experienced lactation days, without exception of the 2nd day.

REFERENCES

Barello, C., Garoffo, L. P., Montorfano, G., Zava, S., Berra, B., Conti, A., Giuffrida, M. G. 2008. Analysis of major proteins and fat fractions associated with mare's milk fat globules. *Mol. Nutr. Food Res.*, vol. 52, no. 12, p. 1448-1456. https://doi.org/10.1002/mnfr.200700311 PMid:18727006

Berg van den, B.M. 2009. Feeding the lactating mare. *Horses People Magaz.*, p. 12-14.

Businco, L., Giampietro, P. G., Lucenti, P., Lucaroni, F., Pini, C., Di Felice, G., Iacovacci, P., Guradi, M. C., Orlandi, M. J. 2000, Allergenicity of mare's milk in children with cow's milk allergy. *Allergy Clin. Immunol.,* vol. 105, no. 5, p. 1031-1034. https://doi.org/10.1067/mai.2000.106377

Caroprese, M., Albenzlo, M., Marino, R., Musclo, A., Zezza, T., Sevi, A. 2007. Behavior, milk yield, and milk composition of machine- and hand-milked. Murgese mares. *J. Dairy Sci.,* vol. 90, no. 6, p. 2773-2777. https://doi.org/10.3168/jds.2006-603 PMid:17517717

Cisla, A., Palacz, R., Janiszewska, J., Skorka, D. 2009. Total protein, selected protein fractions and chemical elements in the colostrum and milk of mares. Archiv Tierzucht, 1, 1-6. ISSN: 000-9438.

Csapó, J., Salamon, S., Loki, K., Csapó-Kiss, Z. 2009. Composition of mare's milk colostrum and milk II. Protein content, amino acid composition and contents of macro- and micro-elements. *Acta Univ. Sapientlie, Alim.,* vol. 2, no. 1, 133-148.

Csapó-Kiss, Z., Stefler, J., Martin, T. G., Makray, S., Csapó, J. 1995.Composition of mare's colostrum and milk. Fat content, fatty acid composition and vitamin content. *Inter. Dairy J.,* vol. 5, no. 4, p. 403-415. https://doi.org/10.1016/0958-6946(94)00014-G

Doreau, M., Martin-Rosset, W. 2002. Horse. In *Encyclopedia of Dairy Sci*., New York, USA : Academic Press, p. 630-637. ISBN: 978-0-12-374402-9 https://doi.org/10.1016/B0-12-227235-8/00115-2

Guradi, M. C., Giampietro, P. G., Lucenti, P., Orlandi, M. 2001. Use of mare milk in pediatric allergology. *Proc. Assoc. Sci. Prod. Anim.. XIV Cong.,* Firenze, June 12-15, vol. 14, p. 647-649.

Gálik, B., Šimko, M., Juráček, M., Bíro, D., Buňka, F., Misař, D., Kráčmar, S. 2012. Content changes of selected mineral nutrients in mare´s colostrums in the first 72 hours after foaling. *Acta univ. agric. et silvic. Mendel. Brun.,* vol. 60, no. 1, p. 45-48. https://doi.org/10.11118/actaun201260010045

Hidiroglou, M., Proulx, J. G. 1982. Factors affecting the calcium, magnesium and phosphorus content of beef cow milk. *Canad. J. Comparat. Med.,* vol. 46, no. 2, p. 212-214. PMid:7093815

Kavazik, A. N., Kivipelto, J., Ott, E. A. 2002. Factors affecting the calcium, magnesium and phosphorus content of beef cow milk. *J. Equine Vet. Sci.,* vol. 22, p. 460-464.

Malacarne, M., Martuzzi, F., Summer, A., Mariani, P. 2002. Protein and fat composition of mare's milk: some nutritional remarks with reference to human and cow's milk. *Inter. Dairy J.,* vol. 12, no. 11, p. 869-877. https://doi.org/10.1016/S0958-6946(02)00120-6

Martin, R. G., Mcmeniman, N. P., Dowsett, K. F. 1992. Milk and water intakes of foals sucking grazing mares. *Equine Vet. J.,* vol. 24, no. 4, p. 295-299. https://doi.org/10.1111/j.2042-3306.1992.tb02839.x PMid:1499538

Martuzzi, F., Catalano, A. L., Summer, A., Mariani, P. 1997. Calcium, phosphorus and magnesium in the milk of nursing mares from Italian Saddle Horse breed and their variations during lactation. *Ann. Fac. Med. Vet., Univ. Parma,* vol. 27, p. 205-212.

Martuzzi, F., Doreau, M. 2006. Mare milk composition: Recent findings about protein fraction and mineral content. In *Nutr. Feed. Broodmare* (eds. N. Miraglia and W. Martin-Rosset EAAP Publication No. 120, Wageningen, The Netherlands : Wageningen Academic Publishers, p. 65-76.

Montanari, G., Zambonelli, C., Grazia, L., Kamesheva, G. K., Shigaeva, M. K. 1996. *Saccharomyces unisporus* as the principal alcoholic fermentation microorganism of traditional koumiss. *J. Dairy Res.,* vol. 63, no. 2, p. 327-331. https://doi.org/10.1017/S0022029900031836

Nascimento, I. R., Jesus, R. M., Santos, W. N. L., Souza, W. D. F., Reis, P.S. 2010. Determination of the minerals composition of fresh bovine milk from milk-producing areas located in the state of Sergipe in Brazil and evaluation employing exploratory analysis. *Microchem. J.,* vol. 96, no. 1, p. 37-41. https://doi.org/10.1016/j.microc.2010.01.010

Nat. Res. Council. 1989. Nutrient Requirements of Horses, 5th Ed. 294-304. National Academy Press, Washington DC.

Official Methods of Analysis AOAC. 1990. 15th Ed,, Washington DC.

Pikul J., Wojtowski J. 2008. Fat and cholesterol content and fatty acid composition of mares' colostrums and milk during five lactation months. *Livestock Sci.,* vol. 113, no. 2-3, p. 128-290. https://doi.org/10.1016/j.livsci.2007.06.005

Raffaela, D. C., Antonia, T. G. G., Cosimo, L. M. D. A., Michele, F., Paolo, A., Marco, G. 2004. Uses of mares' milk in manufacture of fermented milks. Inter. *Dairy J.,* vol. 14, no. 9, p.767-775. https://doi.org/10.1016/j.idairyj.2004.02.005

Saastamoinen, M. T., Koskinen, E. 1993. Influence of quality of dietary protein supplement and anabolic steroids on muscular and skeletal growth of foals. *Anim. Prod.,* vol. 56, no. 1, p. 135-144. https://doi.org/10.1017/S0003356100006255

Sarwar, G., Botting, H. C., Davis, T. A., Darling, P., Pencharz, P. B. 1998. Free amino acids in milks of human objects, other primates and non-primates. *Brit. J. Nutr.,* vol. 79, no. 2, p. 129-131. https://doi.org/10.1079/BJN19980023 PMid:9536856

Schryver, H. F., Oftedal, O. T., Williams, J., Soderholm, L. V., Hintz, H. F. 1986. Lactation in the horse: the mineral composition of mare milk. *J. Nutr.,* vol. 116, no. 11, p. 2142-2147. https://doi.org/10.1093/jn/116.11.2142 PMid:3794822

Solaroli, G., Pagliarini, E., Peri, C. 1993. Composition and nutritional quality of mare's milk. *Ital. J. Food Sci.,* vol. 5, no. 1, p. 3-10.

Summer, A., Sabbioni, A., Formaggioni, P., Mariani, P. 2004. Trend in ash and mineral element content of milk from Haflinger nursing mares throughout six lactation months. Livestock *Prod, Sci.,* vol. 88, no. 1-2, p. 55-62. https://doi.org/10.1016/j.livprodsci.2003.11.002

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