Effects of seasons on the concentration of selected trace elements in horse hair

Wpływ pory roku na stężenie wybranych pierwiastków śladowych w sierści koni

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Abstract

The studies were conducted on a group of 20 mares of the Hucul breed kept in free range system. The horses made use of pastures in summer periods while being fed with hay or silage during the winter. Mares that bore offspring regularly, in good conditions and healthy were involved in the analysis. Hair samples of about 500 mg were taken from areas around the neck under the mane of each horse four times a year. The concentration of 6 elements, i.e., B, Ni, Si, Mo, I and Cr were measured in each sample. Much higher concentrations of molybdenum and nickel were found in hair samples collected in spring, but more of boron in autumn while those from winter contained more of chromium, silicon and iodine. The concentration of all the analyzed minerals differed significantly in relation to the season. A significant positive relationship between levels of nickel and molybdenum on the one hand as well as between silicon and chromium on the other was demonstrated. A negative correlation was, however, observed between the levels of iodine and molybdenum as well as between iodine and nickel.

Keywords: trace elements, horses, hair, season

Streszczenie

Badania przeprowadzono na grupie 20 klaczy rasy huculskiej utrzymywanych systemem bezstajennym. Konie latem korzystały z pastwisk, zimą otrzymywały siano lub sianokiszonkę. W analizie uwzględniono klacze regularnie rodzące potomstwo, zdrowe, w dobrej kondycji. Próbki sierści pobrano z okolic karku spod grzywy w ilości około 500 mg od każdego zwierzęcia (czterokrotnie w ciągu roku). Określono poziom 6 pierwiastków (B, Ni, Si, Mo, I, Cr) w każdej z prób. W sierści z pobrania wiosennego stwierdzono zdecydowanie wyższe stężenia molibdenu i niklu, z jesiennego boru, zaś z zimowego chromu, krzemu i jodu. Stężenia wszystkich analizowanych minerałów różniły się istotnie w zależności od pory roku. Wykazano istotną dodatnią interakcję między poziomem niklu i molibdenu oraz krzemu i chromu. Ujemną zależność wykazano natomiast między poziomem jodu i molibdenu oraz jodu i niklu.

Słowa kluczowe: pierwiastki śladowe, konie, sezony

Streszczenie szczegółowe

Badaniom poddano 20 sztuk koni rasy huculskiej. Do analiz wykorzystano klacze klinicznie zdrowe i w dobrej kondycji. Konie utrzymywane były systemem bezstajennym. Przez cały rok przebywały na górskich pastwiskach. Latem korzystały z pastwiska, zimą otrzymywały siano lub sianokiszonkę. Jako dodatek otrzymywały sól w postaci lizawki.

Pobrań materiału do badań dokonano uwzględniając pory roku, tj. wiosna (czerwiec), lato (wrzesień), jesień (grudzień) i zima (marzec). Próbki sierści pobrano z okolic karku spod grzywy w ilości około 500 mg od każdego zwierzęcia. Określono poziom 6 pierwiastków (B, Ni, Si, Mo, I, Cr) w każdej z prób. Analiza pierwiastkowa została wykonana z wykorzystaniem spektrometru emisji atomowej z plazmą wzbudzoną indukcyjnie (ICP-OES) serii Optima 5300 DV firmy Perkin Elmer. Podstawą metody był pomiar emisji atomowej z zastosowaniem optycznej techniki spektroskopowej.

Wyniki opracowano statystycznie wykorzystując pakiet statystyczny Statistica 9.0.W sierści ocenianych koni najwyższe stężenie boru występowało jesienią – 1.205±1.094mg/kg s.m., najniższe zaś w okresie zimowym – 0.220±0.091mg/kg s.m. Najwyższą koncentrację molibdenu i niklu stwierdzono w sierści z pobrania wiosennego, najniższą chromu i jodu. Natomiast w sierści z pobrania zimowego występowało wysokie stężenie chromu, krzemu i jodu, najniższe zaś boru, molibdenu i niklu. Wykazano istotne różnice między stężeniem analizowanych pierwiastków śladowych w sierści w zależności od pory roku ($P \leq 0.01$). Wykazano istotne dodatnie zależności między poziomem chromu i krzemu oraz molibdenu i niklu a ujemne między stężeniem jodu a molibdenu i niklu.

Introduction

Metabolic disorders of trace elements are commonly diagnosed in livestock (Humann-Zehank et al. 2008). This significantly impacts on their health and productivity. Likewise, supplementation of selected bio-elements poses problems as mineral compounds occurring in living organisms are interrelated and cannot be treated separately. Trace elements serve numerous building and regulatory functions thus ensuring body homeostasis (Asano et al. 2005a, Jackson 1998, Soetan et al. 2010). They are, in the case of young and developing horses, necessary for proper bone formation processes, among other things (Madejon et al. 2009). Their concentration in animal organisms is typically within narrow limits, and both deficiency and excess could be harmful. It is interesting to note that mineral deficits might persist for relatively long periods without noticeable clinical symptoms. Signals of existing disorders could, among others, decrease immunity or poor productive results of animals (Humann-Zehank et al. 2008). Subclinical nature of disorders or variation in reactions hinder the diagnosis of improper mineral supply in animal bodies (Rieker et al. 2000, Soetan et al. 2010). Hence, very often, additive formulations containing trace elements are supplied as protective measures against any potential deficiencies in feeds, especially in cases of increased demands such as during pregnancy or lactation (Kavazis et al. 2002).

The legitimacy of ration supplements as well as mineral and vitamin formulations hitherto commonly used in animal breeding are often being negated, especially in ecological systems of husbandry. The lack of benefits from increased efficiency on the one hand and excessive mineral supply on the other can lead to toxicity in animals.

Interests in the possibility for a quick and accurate diagnosis of the body's mineral supply has increased in recent years, which offers the chance for adequate mineral supplementation.

Having in mind the well-being of animals, efforts are being made to explore non-invasive diagnostic methods. Studies on the hair and fur of animals seems to be in this direction. The research material is obtainable in a simple and safe way. It is easy to transport and store, which ensures the reliability of the results (Asano et al. 2005b, Combs 1987, Gabryszczuk et al. 2010, Hawkins and Ragnarsdottir 2009). The composition of hair reflects the course of metabolic processes over a significant time period (Dunnett and Lees 2003). It is therefore an excellent source of information on such areas as exposure to heavy metals (Chyla and Zyrnicki 2000, Dunnett 2005), or nutritional state (Combs 1987, Hintz 2000). An advantage of hair and fur is the lack of impact of current consumption on the concentration levels of several minerals (Wichert et al. 2002). In addition, the concentration of most trace elements is higher in hair than in blood (Hawkins and Ragnarsdottir 2009). Hair is a stable biological material from which, on the basis of its analysis, we can obtain information on the concentration of macro- and micro-nutrients, trace as well as toxic elements or even residues of veterinary medications even after a long period of time (Dunnett and Lees 2003).

The aim of the research study was to assess changes in mineral content level in the hair of Hucul horses depending on the seasons while being fed on bulky feed.

Materials and methods

The research involved 20 horses of the Hucul breed. Mares that regularly gave birth to foals and which were both clinically healthy and in good condition were accepted for analysis. The horses were reared in free range management system. They were kept on mountain pastures all year round. They were grazed on pastures in summer while being fed in winter on hay or silage obtained from meadows belonging to the studs. The horses were ensured total access to feed. They also, in addition, got salt in the form of salt blocks. Chemical analysis of feeds was, however, not undertaken. In free range management system, ad libitum feed uptake, soil not excluded, biting-off tree and shrub barks, chewing of herbs that often accumulate limited elements makes the factual assessment of feed and mineral in-take difficult. Consequently, attempting to estimate minerals content in a horse' organism based on their concentrations in feeds would be misleading.

The hair of 20 adult mares, aged 4-17 (7 on the average) with mouse-coloured and bay-coloured hair, was collected for analysis. Collection of test materials was done having regard to the season i.e., June in spring, September in summer, December in autumn and March in winter. The hair samples of about 500 mg each were taken from the neck area under the mane of each animal. The level of 6 elements, B, Ni, Si, Mo, I, and Cr was ascertained in each sample. The analysis of elements was performed using the atomic emission spectrometer with inductively coupled plasma (ICP-OES) Optima 5300 DV series from Perkin Elmer company. The basis of the method was the measurement of atomic emission spectroscopy using an optical technique. Indications were made in the Laboratory of Trace Elements in Łódź.

The results were analyzed statistically giving the extreme values (minimum - maximum), the mean and standard deviation. The significance of the differences between the means were determined using the student's t-Test. The level of significance was set as P≤0.01. In order to estimate the relationship between the various trace elements the co-efficient of correlation were calculated. The calculations were performed using the statistical package Statistica 9.0.

Results

The levels of selected trace elements in horses' hair are summarized in Table 1. The highest concentration of boron in the hair of horses studied occurred in autumn i.e., 1.205±1.094mg/kg dry mass while the lowest was in winter - 0.220±0.091mg/kg dry mass. The highest concentration of molybdenum and nickel were found in hair collected in spring with the lowest values being those of chromium and iodine. The hair collected in winter, however, showed high concentrations of chromium, silicon and iodine while those of boron, molybdenum and nickel were lowest (Table 1). The existence of significant differences between the concentrations of the trace elements analyzed in hair depending on the season, (P≤0.01), was confirmed.

Table 1. Concentrations of trace elements in horses' hair depending on the season

Elements		Season				
		Total	Spring	Summer	Autumn	Winter
B	$\bar{x}$	0.678	0.978 ^{A}	0.728 ^{B}	1.205 ^{C}	0.220 ^{ABC}
	SD	0.734	0.330	0.768	1.094	0.091
	Mn	0.043	0.490	0.080	0.043	0.090
	Mx	3.366	1.690	3.366	3.312	0.370
Cr	$\bar{x}$	0.324	0.133 ^{BC}	0.357 ^{aCD}	0.158 ^{AD}	0.478 ^{aAB}
	SD	0.221	0.074	0.226	0.065	0.191
	Mn	0.029	0.029	0.086	0.064	0.249
	Mx	1.037	0.271	0.857	0.292	1.037
Mo	$\bar{x}$	0.050	0.252 ^{BCD}	0.010 ^{bAC}	0.010 ^{abD}	0.009 ^{aAB}
	SD	0.093	0.046	0.001	0.0009	0.001
	Mn	0.006	0.200	0.009	0.008	0.006
	Mx	0.343	0.343	0.013	0.011	0.011
Ni	$\bar{x}$	0.0378	1.125 ^{CDE}	0.271 ^{aAD}	0.340 ^{aBE}	0.126 ^{ABC}
	SD	0.373	0.296	0.116	0.058	0.023
	Mn	0.084	0.509	0.108	0.275	0.084
	Mx	1.800	1.800	0.526	0.451	0.176
Si	$\bar{x}$	16.377	7.657 ^{BC}	21.446 ^{CD}	5.841 ^{AD}	22.050 ^{AB}
	SD	15.697	2.052	19.750	3.494	15.169
	Mn	1.491	3.370	1.491	1.594	6.820
	Mx	64.630	12.230	64.630	12.795	53.300
I	$\bar{x}$	8.146	1.431 ^{ABC}	9.310 ^{aB}	9.908 ^{aC}	9.453 ^{A}
	SD	3.361	0.227	0.883	0.814	2.217
	Mn	1.070	1.070	7.480	7.949	3.210
	Mx	13.890	1.910	10.691	11.159	13.890

Values with the same letters differ significantly – small letters P≤ 0.05; capitals P≤ 0.01

The coefficients of correlation of the analyzed elements are provided in Table 2.

Table 2. Levels of coefficients of correlation for trace elements

	B	Cr	Mo	Ni	Si	I
B	x					
Cr	-0.4428 ** p=0.000	x				
Mo	ns	-0.3997 ** p=0.000	x			
Ni	0.2363 * p=0.022	-0.4885 ** p=0.000	0.8813 ** p=0.000	x		
Si	-0.3960 ** p=0.000	0.8024 ** p=0.000	-0.2502* p=0.015	-0.3497 ** p=0.001	x	
I	ns	0.3648 ** p=0.000	-0.8822 ** p=0.000	-0.7968 ** p=0.000	ns	x

Differ significantly – * P≤ 0.05; ** P≤ 0.01

While there were significant positive correlation between the levels of chromium and silicon, including molybdenum and nickel a negative correlation was found between concentrations of iodine and molybdenum / nickel. Additionally, a significant correlation was established between the levels of silicon and nickel as well as between molybdenum and boron / iodine in the hair collected in spring (Table 3).

Table 3. Levels of coefficients of correlation for trace elements in spring hair

	B	Cr	Mo	Ni	Si	I
B	x					
Cr	ns	x				
Mo	0.5736* p=0.020	ns	x			
Ni	ns	ns	ns	x		
Si	ns	ns	ns	0.9972 ** p=0.000	x	
I	ns	ns	0.5233* p=0.038	ns	ns	x

Differ significantly – * P≤ 0.05; ** P≤ 0.01

While the level of minerals in the hair from summer (Table 4) and autumn (Table 5) collections showed relationship between silicon and chromium (P≤0.01) in that of autumn (Table 5) and winter (Table 6) the relationship was between nickel and molybdenum (P≤0.01).

Table 4. Levels of coefficients of correlation for trace elements in summer hair

	B	Cr	Mo	Ni	Si	I
B	x					
Cr	-0.4510* p=0.016	x				
Mo	ns	ns	x			
Ni	0.4317* p=0.022	-0.6427 ** p=0.000	0.4544* p=0.015	x		
Si	-0.4744* p=0.011	0.9158 ** p=0.000	ns	-0.7299 ** p=0.000	x	
I	ns	-0.4138* p=0.029	ns	0.6257 ** p=0.000	-0.4611* p=0.014	x

Differ significantly – * P≤ 0.05; ** P≤ 0.01

Table 5. Levels of coefficients of correlation for trace elements in autumn hair

	B	Cr	Mo	Ni	Si	I
B	x					
Cr	ns	x				
Mo	ns	0.6470 ** p=0.005	x			
Ni	ns	ns	0.8765 ** p=0.000	x		
Si	ns	0.7608 ** p=0.000	0.5394* p=0.025	ns	x	
I	ns	ns	ns	ns	ns	x

Differ significantly – * P≤ 0.05; ** P≤ 0.01

Table 6. Levels of coefficients of correlation for trace elements in winter hair

	B	Cr	Mo	Ni	Si	I
B	x					
Cr	ns	x				
Mo	ns	ns	x			
Ni	ns	ns	0.8269 ** p=0.000	x		
Si	ns	0.6057 ** p=0.000	ns	ns	x	
I	ns	ns	ns	ns	ns	x

Differ significantly – ** P≤ 0.01

Discussion

Grasslands serve important roles in the nutrition of herbivorous animals. The quality of the feed is dependent on, inter alia, the mineral composition with particular regards to trace elements. They influence the effective uptake of nitrogen, phosphorous and other macro-elements by plants (Baran et al. 2011). The mineral composition of plants depends on the plant species, season and prevailing weather conditions. Madejon et al. (2009) demonstrated that there are higher mineral concentration in green biomass in autumn. Higher concentrations of trace elements were also confirmed in grasses. The herd of horses covered by the study were fed on bulky feed obtained from mountain meadows that were extensively exploited, which could affect the availability of bio-elements.

Dobrzanski et al. (2005) obtained results of chromium concentration similar to ours (0.324±0.221mg/kg dry mass) from Silesian mares in the Herd of Książ Stallion. In the afore-mentioned studies, higher concentrations were noted in hair of half-bred noble mares, i.e., 0.55±0.49mg/kg dry mass. The lowest concentration of chromium in our studies of hair of Hucul horses was observed in spring. Gabryszczuk et al. (2010) had demonstrated in the hair of cows from organic farms a much lower concentration of chromium, which might have been due to the specificity of their use. Chromium helps to increase the resistance of animals, minimize the effects of stress, and also prevents the loss of trace elements from the body (Jackson 1998, Harper 2006). Ott and Kivipelto (1999) suggest that with intensive nourishment of young or sporting horses, the impact of chromium addition to feeds resulting in lowering plasma glucose and increasing the rate at which glucose is expelled from circulation may be beneficial.

Boron, which is found in all tissues of organisms affects the metabolism of many minerals. A slightly lower level of the mineral was observed in our studies of the hair of Hucul horses (table 1) when compared to that obtained from hair of cattle in organic farms (Gabryszczuk et al. 2010).

Unlike ruminants, horses did not reveal any alarming high levels of molybdenum. The excess is usually excreted in urine and thus does not interfere with copper metabolism (Rieker et al. 2000, Strickland et al. 1987). The highest concentrations of molybdenum, in our studies, were noted in hair collected in spring, i.e., 0.252±0.046mg/kg dry mass. In analyzing the impact of sex Asano et al. (2002) found higher levels of this element in hair of stallions.

Lower levels of nickel, (0.0378±0.373mg/kg dry mass), were found in the hair of Hucul horses when compared to results reported by Dobrzański et al. (2005). The said authors provided 1.03± 0.20mg/kg dry mass and 0.91± 0.22mg/kg dry mass for Silesian and half-bred mares respectively. The highest concentrations, in our studies, were obtained from spring collection of hair (Table 1). Gabryszczuk et al. (2010) also obtained lower concentrations from the hair of cattle from organic farms. Asano et al. (2005a) in their analysis, that included half-bred horses noted their highest concentrations of nickel in hair geldings. High concentrations of nickel were also confirmed among horses with chestnut-coloured hair (Asano et al. 2005b).

Iodine is an essential component of thyroid hormone and its deficiency has been found to be widespread among animals benefitting from pastures. The level of iodine in plants depends on the species, type of soil, fertilization and climate (Soetan et al. 2010). It regulates oxygen uptake hence the appropriate concentration of iodine is crucial for horses engaged in intensive training (Jackson 1998). Budzyńska et al. (2006) had obtained, in the hair of pure-bred Arabian horses, lower iodine concentrations of 5.34±0.76mg/kg dry mass when compared to 8.146±3.361mg/kg dry mass obtained in our studies. They also obtained slightly lower concentrations of silicon. Results of our studies showed lower concentrations of silicon in spring and autumn but higher levels during summer and winter. Similar levels of this element were in hair collected in autumn from cattle kept on organic farms (Dunnett and Lees 2003). Asano et al. (2005a), in analyzing mineral concentrations in various breeds of horses noted the highest concentration of silicon in pure-breed horses as well as in geldings. While higher concentrations of the mineral was observed in males (Asano et al. 2002), its level also increased with the age of the horses (Asano et al. 2002).

Similarly, Biricik et al. (2005) indicated a change in the level of minerals depending on the season. With the exception of copper, higher concentrations of minerals were noted in all hair samples collected in winter with changes in diet having no significant impact on the level of the elements covered by the research (Biricik et al. 2005).

In concluding, it should be emphasized that in the studies undertaken, the season was noted to have had significant impact on the concentration of selected trace elements in the hair of Hucul horses. Moreover, while a positive correlation was demonstrated between levels of chromium and silicon as well as molybdenum and nickel, that of between iodine concentration and molybdenum / nickel was negative. In addition, the good health, fitness and reproductive performance of mares covered by the studies suggest that meeting the demand for trace elements in animals kept in conditions similar to what obtains in nature is proper.

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