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CONTENT OF SODIUM AND POTASSIUM IN TISSUES AND ORGANS OF FREE-RANGING EUROPEAN BISONS

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Abstract

The study aimed at assessing the sodium and potassium content in chosen tissues and organs of free-ranging European bisons in Białowieża Primeval Forest depending on the gender and age of animals. In order to determine the content of elements in parenchymal tissues, ribs and hair, the ICP-OES method was used. In the hooves, sodium and potassium were determined with the help of *ICP-MS*. The sodium content in organs and skin appendages varied from 0.30 in hair to 4.77 mg g^{-1} in ribs. In the analysis of the age effect, some significant differences were observed between the investigated groups in the sodium content of the hoof wall, namely, a higher mean value was noted in young individuals. The potassium content in the examined samples was within the range of 0.96 in the hoof wall to 3.63 mg g^{-1} in kidneys. Significant age dependent differences were noted only in the ribs. Sodium and potassium concentrations in the hoof wall were correlated in a highly significant way. Similar dependences also appeared between the content of sodium and potassium in kidneys and liver and kidneys and muscles. On the basis of the results it can be concluded that the status of sodium and potassium supply in the European bison from Białowieża Forest is adequate.

Key words: sodium, potassium, European bison, tissues, organs.

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INTRODUCTION

Sodium and potassium, which belong to the group of alkaline metals, are the main cations which condition the proper functioning of human and animal organisms. About 95% of potassium in the organism is inside cells, while sodium is mainly in the extracellular fluid (97%). The concentrations of those two elements inside and outside cellular membranes is regulated by the sodium-potassium pump, which is essential for animal and human cells (Willard 1989, Munteanu, Iliuță 2011). 60% of potassium in the body is in muscles, while 10% is in the skeleton (Georgiewskii et al. 1982). The amount of potassium is regulated by the kidneys, which respond to changeable potassium tissue concentrations nu increasing or decreasing the discharge of that cation (Willard 1989). About 90% of potassium and 95% of sodium in the body are absorbed from food (Serfass, Manatt 1985).

Animals fed on feeds with an insufficient amount of sodium can be observed to manifest symptoms of sodium deficit, which are similar in different species. Ruminants, for example, respond to sodium deficit by a decrease of milk yield with an accompanying decrease of fat in milk, by licking walls and other objects, by muscular contractions, neurological disorders, polydipsia and a decrease of the immunological response (Groppel 1995, Anke 2004, Anke et al. 2006). Symptoms of sodium deficit may appear in dairy cows fed feeds with a high potassium content. The following are possible manifestations: vaginitis, irregular ovarian cycles and ovarian cysts (Groppel 1995).

As a rule, potassium deficit does not occur in animals. Research results show that green fodder supplies adequate amounts of that element. Occasionally, potassium deficit may occur only in milk-fed calves. There are also few reports on potassium deficit in green fodder from sandy soils without proper potassium fertilization (Groppel 1995). Decidedly more often there is an excessive amount of potassium in feed. It may occur on the soil fertilized with large doses of liquid manure. However, its excess consumed with feed is removed through homeostatic mechanisms with urine (Willard 1989). In plants cultivated on soils rich in potassium, the content of that component reaches 2.5 to 6.0% d.m. (Grzebisz 2004). Plants originating from mineral soils contain more of that element than plants grown on organic soils (Grzegorczyk et al. 2013). Sodium concentrations in green parts of plants may not cover the requirements of ruminants (Anke et al. 2006). Therefore, it is important to monitor the mineral supply of domestic as well as free-ranging ruminants by analyses of the tissue content of particular macro- and microelements. Until now, regarding wild ruminants, examinations of the mineral composition of tissues and organs have been performed in animals repressenting the *Cervidae* family. In the recent years, the investigations have encompassed the European bison, which is a unique species in Europe, reintroducted to natural habitats (Dębska 2005, Dymnicka et al. 2009, Skibniewski et al. 2010, Kośla et al. 2011, 2012).

The aim of the study was to of sodium and potassium contents in the chosen tissues and organs of the biggest free-ranging ruminant in Poland inhabiting Białowieża Forest in relation to the gender and age of investigated animals.

MATERIAL AND METHODS

The experimental material comprised segments of the liver, kidneys, muscles, ribs, hair and hooves of 20 European bisons representing the lowland subspecies (*Bison bonasus bonasus*), obtained during a winter culling in the Polish part of Białowieża Forest (52°35′-52°55′ N, 23°30′-24°00′ E). They were animals of both genders (6 males and 14 females) which were divided into two age groups (calves up to 1 year of age – 15 animals, older than 2 years – 5 animals). Samples of soft tissues and bones were collected to aseptic polyethylene containers and then stored at the temperature of -20°C. Hair was collected into paper bags. Soft tissues were homogenized, then each sample was divided, 0.5 g collected and then mineralized in nitric acid (67% Merck, Germany) in a microwave apparatus (CEM-81 D) under pressure. Ribs were mineralized according to the above procedure, while hair samples were degreased using 70% ethyl alcohol in an apparatus SER 148 Velp Scientifica (Italy) and then washed, first with hot distilled water and then three times with redistilled water. Hair mineralization was performed as described above, having first cut it into fragments and weighing 0.5 g samples. Samples of the hoof wall were dry burnt. Mineralization was performed in a muffle furnace (LM 212.11 VEB Elektro Bad Frankenhausen/Deutschland) at the temperature 450°C and the resultant ash was quantitatively transferred to measuring flasks, to which spectrally pure HCl was added up to 2.5%. Sodium and potassium in the parenchymatous organs, ribs and hair were determined using the method of optical emission spectrometry with inductively coupled plasma (ICP-OES) versus the reference material (BCR 184, 185, *CRM 397*). In the hoof samples, the elements were determined by inductively coupled plasma mass spectrometry (*ICP-MS) versus the reference material CRM 397*. The results are presented in mg g^{-1} of the tissue fresh matter in the case of parenchymatous organs and ribs, in mg g^{-1} of the air dried sample of hair and in mg g^{1} of dry matter of the hoof sample. Statistical analysis of the results was performed with Statistica 10 ™ software (StatSoft, Inc.). Due to the nonparametric distribution of data, the differences between the investigated groups were determined with the the *U* Mann-Whitney test at $p \leq 0.05$ and $p \leq 0.01$. Analysis of correlations between the given parameters was performed using the Speraman's correlation.

Table 1

Sodium and potassium content in the tissues of European bisons depending on their gender (mg g^{-1} of fresh tissue)

Tissue /organ		Male			Female			All animals		
		mean ± SD	median	min.-max.	mean ± SD	median	min.-max.	mean ± SD	median	min.-max.
Liver	Na	1.460 ± 0.410	1.360	1.065-2.078	1.700±0.670	1.570	0.994-3.610	1.630±0.600	1.510	0.994-3.610
	K	3.300 ± 0.240	3.240	3.053-3.634	3.160±0.350	3.160	2.604-3.798	3.200±0.320	3.180	2.604-3.798
Kidneys	Na	3.300 ± 0.730	3.320	2.489-4.242	3.140±0.650	3.300	0.930-4.200	3.900±0.660	3.300	0.930-4.242
	K	3.610 ± 0.270	3.590	3.262-3.909	3.640±0.480	3.560	2.839-4.423	3.630±0.420	3.560	2.839-4.423
Muscles	Na	0.700 ± 0.250	0.630	0.522-1.194	1.070±0.480	1.250	0.423-2.035	0.500±0.450	0.680	0.423-2.035
	K	1.340 ± 0.090	1.320	1.269-1.499	1.280±0.170	1.260	0.925-1.584	1.290±0.150	1.290	0.925-1.584
Ribs	Na	5.060 ± 0.520	5.110	4.190-5.800	4.640±0.360	4.730	1.980-5.330	4.770±0.450	4.760	1.980-5.800
	K	2.290 ± 1.240	1.940	1.248-4.640	1.760±0.540	1.860	0.850-2.570	1.930±0.840	1.870	0.850-4.640
Hair	Na	0.180 ± 0.100	0.190	0.118-0.435	0.340±0.260	0.310	0.084-1.191	0.300±0.240	0.250	0.084-1.191
Hoof d.m.	Na	1.220 ± 0.240	1.110	0.972-1.518	1.110±0.270	1.100	0.780-1.474	1.150±0.260	1.100	0.780-1.518
	K	1.110 ± 0.260	1.060	0.808-1.550	0.870±0.240	0.900	0.484-1.180	0.960±0.270	0.930	0.484-1.550

d.m. – the content in dry matter in the hoof samples

RESULTS AND DISCUSSIONS

It was found that the sodium content in tissues and organs of European bisons were within the range of 0.30 in hair to 4.77mg g^{-1} in the ribs. The results are presented in Tables 1 and 2. The mean sodium content in the liver and kidneys of European bisons was higher than reported by others. Dębska (2005) observed the following values in animals of that species: 1.10 (mg g^{-1} of fresh tissue) in the calf liver, 1.02 in juveniles, 1.09 in adult animals. The content of sodium in a calf's kidneys was 2.37 mg g^{-1}, in juveniles – 2.27 mg g^{-1}, and in adult European bisons – 2.64 mg g^{-1}. The sodium content in the liver of European bisons observed in our own investigations was within the reference values reported by Puls (1998), i.e. from 0.53 to 3.45 mg g^{-1}.

Regarding the kidneys, the mean sodium concentrations were higher than those reported by Puls (1998), which were within the limit from 1.45 to 1.70 mg g^{-1}. Publications about the mineral composition of the European bison's tissues are few and it is impossible to submit them to a comprehensive discussion, nor to compare reliably our results with those reported by other authors. While juxtaposing our results with the data about cattle it is evident, the former were higher. The content of sodium in animal tissues bought as food products, according to Anke et al. (2006), could be described in the following way: the bovine liver contained 648 mg Na kg^{-1} of fresh tissue and kidneys had 1865 mg Na kg^{-1} of fresh tissue.

Compared with the data about monogastric herbivorous animals such as horses, the current results are also higher. Kośla and Anke (1988) stated that the mean sodium content in the horse liver was about 3.3 mg g^{-1} d.m. and in kidneys – 9.7 mg g^{-1} d.m. Considering the degree of the organ's hydration in accordance to the conversion factor, Anke et al. (2006) noted that the content of that metal was about 1.0 mg g^{-1} of fresh tissue in the liver and about 2.35 mg g^{-1} of fresh tissue in kidneys. The data obtained from the cited research are similar to those observed by Dębska (2005) in her analysis of the European bison's issues.

The sodium content in hair of the European bison was lower than the values reported by Puls (1998) but close to the results by Dębska (2005). While analysing the effect of the health of individuals on the content of sodium in their coat, Dymnicka et al. (2009) noted 0.15 mg of per sodium g^{-1} dry matter in healthy animals and 0.11 – in animals with *balanoposthitis*. Those values were lower than ours.. In another investigation on the mineral composition of the European bison's hair, Kośla (1993) reported that hair contained on average 0.62 mg g^{-1} d.m. of sodium. A significant correlation was demonstrated between the sodium content in animal hair and the colour of hair. Kośla et al. (1985) noted that the mean content of sodium in the cattle hair equalled 0.88 mg g^{-1} d.m., but depending on the colour those values were 0.30 mg g^{-1} in brown hair and 0.52 mg g^{-1} d.m. in black hair. Anke et al. (2006) observed that in black-and-white cows the content of sodium in black hair averaged 579 mg kg^{-1} and in white hair – 325 mg kg^{-1} d.m. Brown hair contained

478 mg kg^{-1} d.m. of sodium, on average. These results justify the assumption that sodium plays an important role in the process of pigment accumulation in hair keratin (Anke et al. 2006). The authors mentioned above calculated correlations between the sodium content in hair and in particular tissues, noting that hair sodium is a good indicator of the content of sodium in the organism.

The absence of reports on the sodium content in bones and skeletal muscles of the European bison does not allow us to compare our results with those of other authors. However, the values obtained in our own investigations correspond with the research results on the sodium content in the equine osseous tissue performed by Kośla and Anke (1988). The mean value observed by those authors was 8.5 mg g^{-1} d.m. Considering the degree of bone hydration according to Fernandez-Seara et al. (2004), the result obtained in our own investigations was about 4.25 mg g^{-1} of fresh tissue. The sodium content in muscles of the European bison is similar to the results of analyses on equine muscles, where the reported level was 3.0 mg g^{-1} d.m., i.e. about 0.81 mg g^{-1} of fresh tissue (Kośla, Anke 1988). This value is definitely higher than the data obtained from analysis of skeletal muscles of cattle, where Anke et al. (2006) reported the mean value of sodium at 776 mg kg^{-1} d.m.

It was only in the hoof tissue, in which the sodium content could not be compared with other references due to the lack of relevant literature data, a statistically significant difference between the age groups was observed. Hooves of calves contained more sodium than the hooves of mature animals. The potassium content in tissues of the European bison is presented in Table 2.

Table 2

Sodium and potassium content in tissues of European bisons depending on the age of animals (mg g^{-1} of fresh tissue)

Tissue /organ		Calves up to 1 year		Animals older than 2 years		All animals	
		mean ±SD	median	mean± SD	median	mean± SD	median
Liver	Na	1.530±0.410	1.440	1.920±1.000	1.510	1.630±0.600	1.51
	K	3.160±0.310	3.100	3.340±0.360	3.310	3.200±0.320	3.180
Kidneys	Na	3.180±0.710	3.360	3.240±0.520	3.230	3.900±0.660	3.30
	K	3.570±0.340	3.550	3.820±0.610	4,030	3.630±0.420	3.560
Muscles	Na	0.870±0.350	0.650	1.160±0.650	1.350	0.500±0.450	0.68
	K	1.290±0.160	1.280	1.320±0.110	1.360	1.290±0.150	1.290
Ribs	Na	4.760±0.500	4.760	4.820±0.290	4.730	4.77±0.450	4.76
	K	2.180±0.740*	1.900	1.060±0.190*	1.090	1.930±0.840	1.870
Hair	Na	0.320±0.280	0.210	0.240±0.180	0.290	0.300±0.240	0.25
Hoof (d.m.)	Na	1.240*±0.220	1.220	0.960*±0.250	0.980	1.150±0.260	1.10
	K	1.050±0.240	1.060	0.780±0.230	0.810	0.960±0.270	0.930

d.m. – the content in dry matter of the hoof samples

* differences statistically significant depending on age at $p \leq 0{,}05$

The potassium content in the liver of European bisons reported herein was higher than that observed by Dębska (2005). In animals representing three age groups (calves, young and adult animals), the liver potassium content was 2.66, 2.90 and 2.88 mg g^{-1} of fresh tissue, respectively. Analogously to our results, Dębska (2005) noted a lower amount of potassium in calves, although the difference was not statistically significant.

While comparing the potassium content in the liver of European bisons, with a focus on animals with *balanoposthitis*, Dymnicka et al. (2009) did not observe any statistically significant differences between the examined groups. In healthy males, the average potassium content was 3.07 while in diseased animals, it was to 3.05 mg g^{-1} of fresh tissue. The reference values of the liver potassium content in cattle are from 1.4 to 3.95 mg g^{-1} of fresh tissue (Puls 1998). The current data concerning individuals of both genders and various age groups fell within that range. While analysing the effect of breed, gender, age and management environment on the potassium content in the equine liver, Kośla (1988) noted the mean values from 8.0 to 9.2 g kg^{-1} d.m. of potassium, which submitted to the conversion method according to Anke et al. (2006) yielded from 2.4 to 2.76 g kg^{-1} of fresh tissue. The investigated groups did not differ in a statistically significant way.

While assessing the potassium content in kidneys of European bisons, it can be noticed that – similarly to the liver tissue – the authors of the present report observed values that were higher than those demonstrated by Dębska (2005), who in the three age groups determined 2.57, 2.58 and 2.51 mg of potassium 1 g^{-1} of fresh matter, respectively. Similarly as in our investigations, Dębska (2005) did not observe any statistically significant differences depending on the age of individuals. Comparing the potassium content in kidneys and including the effect of the health status, Dymnicka et al. (2009) did not observe any statistically significant differences between the groups. The mean potassium content in kidneys of healthy individuals was 2.64 mg g^{-1} and in those suffering from *balanoposthitis* it equalled 2.44 mg g^{-1} of fresh tissue. The reference data concerning bovine kidneys are from 1.80 to 2.60 mg g^{-1} of fresh tissue according to Puls (1998), thus being lower than the values generated by the analysis of tissue material from European bisons. The potassium content in muscles of European bisons may be compared to the data by Kośla (1988) about horses. The author observed significant differences in potassium levels, depending on the place of animal housing. Mean values for particular groups were within the limit from 12 to 14 mg g^{-1} d.m. pr, according to Anke et al. (2006), from 3.26 to 3.81 mg g^{-1} of fresh tissue. The potassium content in a horse's muscles, depending on breed, gender and age, ranged from 11 to 13 mg g^{-1} d.m., i.e. 2.99 to 3.54 mg g^{-1} of fresh tissue. Significant differences in the potassium content in muscles of European bisons and horses result from the species-specific differences, different diets and different analytical methods.

The potassium content in the ribs of the investigated animals differed significantly depending on their age. Similar results were obtained by Kośla

(1988) in investigations on equine tissues. The youngest individuals had 1.7 mg g^{-1} d.m. in their bones, while older horses aged 11 to 15 years had 1.1 mg g^{-1} d.m., on average. In line with the conversion factor by FERNANDEZ-SEARA et al. (2004), the potassium content in the youngest individuals amounted to 1.28 mg g^{-1} of fresh tissue and in the group of older animals it was 0.83 mg g^{-1} of fresh tissue. The potassium content in the hooves does not differ in a significant way between the age groups, although, similarly to sodium, the group of calves was characterized by a higher content of that element in the hoof wall.

A highly significantly correlated dependence was observed between the sodium and potassium content in kidneys and in the liver (r = 0.82, $p \leq 0.01$). A similar relationship between those elements also occurred in the hoof wall. The correlations between the concentrations of sodium and potassium in particular organs are presented in Table 3.

Table 3

Correlation coefficients between potassium and sodium in the examined organs

Tissues/organs		Potassium					Sodium			
		liver	kidneys	muscles	ribs	hoofs	liver	kidneys	muscles	ribs
Potassium	kidneys	0.110								
	muscles	-0.060	0.140							
	ribs	0.070	0.070	-0.030						
	hoofs	-0.030	0.160	0.390	0.240					
Sodium	liver	-0.280	0.370	0.210	-0.290	0.050				
	kidneys	-0.590**	0.110	0.180	-0.120	0.110	0.320			
	muscles	0.170	0.820**	0.350	-0.010	0.310	0.290	-0.010		
	ribs	0.250	0.110	-0.090	0.190	0.140	-0.280	-0.290	0.120	
	hoofs	0.120	0.380	0.500*	0.200	0.820**	0.230	-0.080	0.480*	0.020

* differences statistically significant at $p \leq 0{,}05$

** differences highly significant statistically at $p \leq 0{,}01$

CONCLUSIONS

1. The mean sodium content in the tissues and organs of European bisons were high in comparison with the investigation results of other authors. The sodium level in the liver fell within the reference range reported in literature, while in kidneys it surpassed that range.

2. In young animals, a higher sodium content was observed in the hoof wall and a higher potassium content in ribs as compared with the group of adult animals.

3. A highly significant correlation was observed between the sodium and potassium content in the hoof wall.

4. On the basis of the results, it can be concluded that the sodium and potassium supply in European bisons from Białowieża Primeval Forest is adequate.

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