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Effect of selected factors on mineral parameters in plasma of cows

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Abstract

Objective of this paper was to evaluate the effects of herd, exploitation, management system, and season on blood macro and micro-minerals in cows kept in a mountainous region. The cows were kept in four herds, two exploitation manners, and two systems. Concentrations of calcium (Ca), phosphorus (P) were significantly differed in the factors of herd and exploitation only, levels of copper (Cu) and zinc (Zn) showed differences in all factors. Beef animals had higher level of P than dairy exploitation (1.59 mg.L^{-1} vs. 1.42 mg.L^{-1}; $P<0.01$) and lower level of Ca (2.10 mg.L^{-1} vs. 2.33 mg.L^{-1}; $P<0.001$). Higher concentrations of Cu and Zn were recorded in dairy exploitation (0.91 mg.L^{-1}, 1.17 mg.L^{-1}) versus beef exploitation (0.67 mg.L^{-1}, 0.86 mg.L^{-1}) ($P<0.001$). We found higher concentrations in Cu (0.84 mg.L^{-1} vs. 0.62 mg.L^{-1} , $P<0.001$) and Zn (1.06 mg.L^{-1} vs. 0.96 mg.L^{-1}, $P<0.05$) in non-ecological system. There were higher values of Cu and Zn in fall than spring (0.90 mg.L^{-1} and 1.10 mg.L^{-1} vs. 0.67 mg.L^{-1} and 0.93 mg.L^{-1}) ($P<0.001$).

Keywords: cow, macroelements, calcium, phosphorus, copper, zinc

Zusammenfassung

Einfluss ausgewählter Faktoren auf Mineralgehalte im Blutplasma von Kühen

Untersucht wurden die Einflüsse von Herde, Nutzungstyp, Haltungssystem und Jahreszeit auf Makro- und Mikromineralgehalte im Blutplasma von Kühen in Gebirgsstandorten. Die Untersuchungen fanden an Milch- und Fleischrindrassen in vier Herden mit zwei unterschiedlichen Haltungssystemen statt. Signifikante Unterschiede fanden sich bei den Konzentrationen von Kalzium (Ca) und Phosphor (P) für die Einflussfaktoren Herde und Nutzungstyp während bei den Werten für Kupfer (Cu) und Zink (Zn) sich alle vier Einflussgrößen signifikant unterschieden. Fleischrinder wiesen höhere P-Werte als Milchrinder (1.59 mg.L^{-1} gg. 1.42 mg.L^{-1}; $P<0.01$) und niedrigere Ca-Werte (2.10 mg.L^{-1} bzw. 2.33 mg.L^{-1}) auf. Gegenüber den Fleischrindern (0.67 mg.L^{-1} und 0.86 mg.L^{-1}) fanden sich bei Milchrindern höhere Cu- und Zn-Konzentrationen (0.91 mg.L^{-1} und 1.17 mg.L^{-1}). In den nichtökologischen Haltungssystemen wurden höhere Cu- (0.84 mg.L^{-1} bzw. 0.62 mg.L^{-1}) und Zn-Konzentrationen (1.06 mg.L^{-1} bzw. 0.96 mg.L^{-1}) nachgewiesen. Im Herbst fanden sich gegenüber dem Frühling höhere Cu- (1.10 mg.L^{-1} bzw. 0.90 mg.L^{-1}) und Zn-Werte (0.93 mg.L^{-1} und 0.67 mg.L^{-1}). Für sämtliche genannten Werte wurde Signifikanz nachgewiesen.

Schlüsselwörter: Kuh, Mineralgehalt im Blutplasma, Kalzium, Phosphor, Kupfer, Zink

Introduction

Assessment of macro- and micro- element status identifies whether current mineral supplementation of cattle feed is adequate and whether improved productivity is likely to occur with changes in supplementation. While some elements such as copper (Cu), zinc (Zn) are necessary for animal life, they may be toxic and harmful at high levels of exposure and toxic when ingested in excess, of course (GALLO *et al.* 1996, MASSANYI *et al.* 2001, SOLAIMAN *et al.* 2006). Dangerously can be their accumulating in the body through food chain, water and air. Industrial development is largely responsible for pollution of the environment with toxic metals, although some contamination is derived from natural geological sources (RAJCAKOVA *et al.* 2003, CERMAK *et al.* 2006).

Agricultural activity may also increase environmental concentrations of trace metals, such as Cu and Zn, in certain areas. The cattle are mostly fed by the locally produced bulky fodder and this can be exposed to metal contamination. In animal body the concentration of essential metal elements is regulated in the metabolic pathways in contrast to the toxic elements, which accumulate from the polluted nutrients. The amount of an element which accumulates in the organs depends on the interval of exposure, the quantity ingested, the production and reproduction phases of the animals, as well as their age and breed (KOTTFEROVA and KORENEKOVA 1997, HERNANDEZ *et al.* 1997, NRC 2001, McDOWELL and ARTHINGTON 2005, KEBREAB and VITTI 2005). More information is needed on the mineral requirements in relation to environment factors of the cattle.

Of the major minerals the most important are calcium (Ca) and phosphorus (P), they play vital roles in most body tissues with structural roles in intracellular communication, DNA synthesis, and maintain homeostasis (UNDERWOOD and SUTTLE 2001, McDOWELL and ARTHINGTON 2005, KEBREAB and VITTI 2005). Decreased growth and milk production, poor conception and depraved appetite are the general symptoms of Ca and P deficiency (VITTI *et al.* 2005).

Cu is an essential component of several enzymes that are required to maintain homeostasis and has a basical role in the iron absorption, haemopoiesis and various enzyme activities, and in the oxidation-reduction process (REECE 2004, AHOLA *et al.* 2005). It may perform several functions in the immune system (STANTON *et al.* 2000, UNDERWOOD and SUTTLE 2001).

Zn is required for normal growth, development, and function in all animal species and it´s also important for the synthesis of proteins and nucleic acids, a number of catalytic, structural and regulatory functions, formation of skin structures and skeleton metabolism (HAP and ZEMAN 1995). It plays an important role in rumen fermentation processes, mainly cellulose digestion and formation of volatile fatty acids. The movement of Zn into and within the body is precisely regulated at levels of intake within the requirement range and also by the absorption (MASSANYI *et al.* 2001, HAGEMEISTER *et al.* 1998).

Feeding cattle specific amino acid complexes of Zn and Cu reduced somatic cell counts, and increased milk production (NOCEK *et al.* 2000). Increasing Zn and Cu intake of intensively grazed dairy cattle improved lactation and reproductive performance, also increased feed utilization efficiency (STANTON *et al.* 2000, AHOLA *et al.* 2004, SOLAIMAN *et al.* 2006).

The objective of the present study was to evaluate the the hypotheses that Ca, P, Cu and Zn contents in blood of cattle are impacted of herd, exploitation manner, management system, and season.

Material and methods

In the present study we investigated the minerals in beef and dairy cows in selected herds reared in a mountainous region.

Management of the experiment

The cattle were kept in four herds. Blood samples were divided according to factors herd (H1: n=120, Holstein breed and Czech Spotted breed; H2: n=60, Czech Spotted breed; H3: n=82, Angus and Simmental breeds; H4: n=40, Czech Spotted breed and crosses with beef breeds), manner of exploitation (dairy: n=180, beef: n=122), systems (ecological: n=40, non-ecological: n=262), and season (spring: n=139), fall: n=163).

Feed intake was monitored daily in each observation for 3 days. Feed offered and refusals were recorded daily prior to the morning feeding. Samples of each feed were collected daily and composites were made each month. Feed samples were dried and ground in a mill to pass through a 1 mm mesh screen. Consumption of feeds and the amount of microelements in daily rations are presented in Tables 1 and 2. Animals were fed to meet daily requirements of cattle according to nutrient requirements. Minerals were remained available for *ad libitum* consumption at a single location in each pasture and outside yard in free-choice mineral feeders. All cows received the same respective mineral supplement. *Ad libitum* access to water was maintained throughout the study. The cattle were in good condition during the whole experiment and clinically, they were healthy.

The cows received mineral mixture. Used mineral fodder supplement (Turmix S3, Tekro, s.r.o.) contained - Ca 12 %, P 8 %, Na 8.5 %, Mg 8 %, vitamin A, D3, E, Cu 630 mg, Zn 3 050 mg, Mn 2 800 mg, I 54 mg, Se 16 mg, Co 16 mg. The levels of Zn and Cu calculated per 1 kg of received dry matter did not exceed the levels allowed for the daily requirements of cattle.

Table 1
Daily consumption of feeds during observations
Täglicher Futterverzehr im Untersuchungszeitraum

Herd	Season	Pasture, kg	Hay, kg	Haylage, kg	Straw, kg	TMR, kg
1	Spring 2005					16.9
	Fall 2005					17.5
	Spring 2006					16.4
	Fall 2005					15.7
2	Spring 2005					16.2
	Fall 2005					15.8
	Spring 2006		4.0	11.8	1.5	
	Fall 2005		1.8			14.0
3	Spring 2005	13.0				
	Fall 2005	10.4				
	Spring 2006	9.5		8.0		
	Fall 2005	10.2	2.8			
4	Spring 2005	13.5				
	Fall 2005	13.5				
	Spring 2006	10.7	2.8			
	Fall 2005	13.2				

TMR total mixed ration

Table 2
The content of trace minerals in the dry matter of feed ration in observations (mg.kg^{-1})
Gehalt an Spurenelementen in der Trockensubstanz der Futterrationen (mg.kg^{-1})

Season	Herd							
	1		2		3		4	
	Cu	Zn	Cu	Zn	Cu	Zn	Cu	Zn
Spring 2005	10.7	65.0	28.1	99.4	10.4	45.1	5.1	75.1
Fall 2005	10.1	51.7	21.8	141.2	24.9	26.8	13.4	76.9
Spring 2006	12.6	88.3	23.2	114.3	30.4	11.8	11.2	33.5
Fall 2006	9.8	86.4	18.7	78.1	9.5	30.4	17.9	61.6
Average	10.8	72.8	22.9	108.25	18.8	28.5	11.9	61.8

General health parameters were obtained in the each three days observation. Vital body signs recorded were respiration rate, heart rate and rectal temperature. Blood samples were taken in the third day of observation by jugular venipuncture into heparinized tubes from apparently healthy adult female animals. Samples were placed on ice immediately after collection and transported to the laboratory. The plasma was separated and stored at –24 °C until processing.

The concentration of Cu and Zn in blood plasma and in dry matter of a diet was analysed by flame atomic absorption method using an AA Spectrometer Unicam 969. Bio-LA tests Lachema Brno, CZ were used on analysing of Ca and P contens.

Herd No. 1 (non-ecological system) is localized 400 m above sea level in south-west Czechia with the majority of Holstein breed (65 %) and Czech Spotted breed (35 %). There are 470 dairy cows with the average milk production 5 700 kg per year (non-ecological system). Cows were kept in free-stall housing and tie-stall housing and artificially inseminated. The cows were milked twice a day. Animals were fed *ad libitum* twice a day to meet daily requirements.

Conventional dairy herd No. 2 (non-ecological system) is placed 550 m above sea level. There were 350 of dairy cows with the majority of Czech Spotted breed (60 %) and Holstein breed (40 %) with the annual production of milk 4 600 kg. Cows are kept in free-stall housing with straw bedding. The cows were milked twice a day to meet daily requirements and artificially inseminated throughout the year.

Non-ecological beef herd (No. 3) was localized in the hilly region (675 m above sea level). During the grazing season, the cows and calves (90 heads of beef cows majority of Aberdeen Angus breed (70 %) and Simmental breed (30 %) were grazed in the rotational grazing system outdoors on pasture throughout the year without any shelter and were supplementary fed by locally produced hay and haylage. Mineral treatments were provided at a single location in each pasture in free-choice mineral feeders. During winter, animals had access to a stone cowshed with clay floor and straw bedding and there were fed locally produced hay. Breeding period was beginning by artificially insemination at April and continued from May to June by the naturally mating.

A suckler ecological herd (no. 4) of 210 beef cows (Czech Pied cattle and their crosses with beef breeds Hereford, Charolais, and Galloway) was kept on pasture (910 m above sea level). Cows were together with bulls. It was paddock grazing system used. Cattle were grazed outdoors on pasture throughout the year without any shelter. Mineral treatments were provided at a single location in each pasture in free-choice mineral feeders. During the winter, the cattle had an access in the cowshed and there were fed locally produced hay. Farm had been certified for 5 years under the guidelines for organic farming.

The Descriptive Statistics procedure of statistical package STATISTIX Version 9.0 (ANONYMOUS 2008) was used for computing of mean and standard error of the mean. Values are expressed as means ± S.E., because the sample sizes are not the same for all groups.

The normality of data was evaluated by Wilk-Shapiro/Rankin Plot procedure. This method examines whether a variable conforms to a normal distribution. A rankit plot of each variable was produced, and an approximate Wilk-Shapiro normality statistic was calculated. The tested variables that showed values of W>0.80 were considered normal and therefore were then submitted to an ANOVA. We found that all variables conformed to a normal distribution.

The homogenity of variance of the observed variables in groups, whose average values are being compared, was calculated by preliminary variance tests which determined whether the variabilities are equal. Bartlett's test for equality of variance tests was applied with an unequal size of samples. The ratio of the largest within-group variance over the smallest was also tested (Pearson and Hartley test).

Among-group comparisons were evaluated by four-factorial analysis of variance using a General linear model ANOVA (General AOV/AOCV).

Results

Mean concentrations of Ca (Table 3) were significantly differed in the factor of herd ($P<0.001$), specialization ($P<0.001$), and system ($P<0.01$). The highest values were found in herd 1 (400 m above sea level) (2.42±0.05 mg.L^{-1}). There were higher levels in dairy specialization (2.33±0.04 mg.L-1) and non-ecological system (2.27±0.04 mg.L^{-1}).

Values of P were highly significantly differed in factor herd ($P<0.01$). The highest values were found at the herd 3 (675 m above sea level) (1.64±0.06 mg.l^{-1}). The lowest concentrations were recorded in the herd 2 (550 m above sea level) (1.37±0.05 mg.L^{-1}). Difference between dairy and beef specialization was significant. Beef animals had higher level of P than dairy specialization (1.59±0.04 mg.L^{-1} vs. 1.42±0.04 mg.L^{-1}; $P<0.01$) and lower level of Ca (2.10±0.05 mg.L^{-1} vs. 2.33±0.04 mg.L^{-1}; $P<0.001$). We found interaction in P and Ca between Season×Herd ($P<0.001$) (Table 3).

Factor herd was differed in the both microminerals ($P<0.001$) (Table 4). The highest values of Cu and Zn were measured in the herd 2 (550 m above sea level) (0.97±0.03 mg.L^{-1}, 1.24±0.03 mg.L^{-1}) and the lowest concentration of Cu in the herd 4 (910 m above sea level (0.62±0.06 mg.L^{-1}) and Zn in the herd 3 (675 m above sea level) (0.81±0.02 mg.L^{-1}).

Higher concentrations were recorded in dairy specialization (0.91±0.02 mg.L^{-1}, 1.17±0.02 mg.L^{-1}) versus beef specialization (0.67±0.02 mg.L^{-1}, 0.86±0.02 mg.L^{-1}) ($P<0.001$). We found higher concentrations in non-ecological system in both parameters Cu and Zn (0.84±0.01 mg.L^{-1} and 1.06±0.02 mg.L^{-1} vs. 0.62±0.04 mg.L^{-1} and 0.96±0.05 mg.L^{-1}). In the both microminerals Cu and Zn, there were higher values in fall than spring (0.90±0.02 mg.L^{-1} and 1.10±0.02 mg.L^{-1} vs. 0.67±0.02 mg.L^{-1} and 0.93±0.02 mg.L^{-1}) ($P<0.001$). We found interactions in Cu an Zn between Season×Herd ($P<0.001$).

Table 3
Concentrations of calcium and phosphorus in blood plasma
Kalzium- und Phosphorkonzentration im Blutplasma

Factor		Calcium, mg.L^{-1}				Phosphorus, mg.L^{-1}			
		N	$\bar{x} \pm SE$	*P*	Significance	N	$\bar{x} \pm SE$	*P*	Significance
Herd	- 1	99	2.42 ± 0.05	0.0000***	1:4***	99	1.44 ± 0.04	0.0041**	3:1,2*
	- 2	36	2.09 ± 0.08		1:3.2**	36	1.37 ± 0.05		
	- 3	76	2.16 ± 0.05			77	1.64 ± 0.06		
	- 4	40	1.98 ± 0.12			40	1.49 ± 0.08		
Exploit	- 1	135	2.33 ± 0.04	0.0006***		135	1.42 ± 0.04	0.0023**	
	- 2	116	2.10 ± 0.05			117	1.59 ± 0.04		
System	- 1	40	1.98 ± 0.08	0.0014**		40	1.49 ± 0.07	0.8752	
	- 2	211	2.27 ± 0.04			212	1.50 ± 0.03		
Season	- 1	139	2.22 ± 0.04	0.2675		139	1.50 ± 0.04	0.5779	
	- 2	112	2.14 ± 0.05			113	1.54 ± 0.04		

*$P<0.05$, **$P<0.01$, ***$P<0.001$, Exploit exploitation, Interactions: Ca - Season×Herd 0.0000***, P - Season×Herd 0.0000***, Exploitation: 1 dairy, 2 beef, System: 1 ecological, 2 non-ecological, Season: 1 spring, 2 fall

Table 4
Concentrations of copper and zinc in blood plasma
Kupfer- und Zinkkonzentration im Blutplasma

Factor		Copper, mg.L^{-1}				Zinc, mg.L^{-1}			
		N	$\bar{x} \pm SE$	*P*	Significance	N	$\bar{x} \pm SE$	*P*	Significance
Herd	- 1	120	0.88 ± 0.01	0.0000***	2,1:3,4***	120	1.14 ± 0.02	0.0000***	2,1:3,4***
	- 2	60	0.97 ± 0.03			60	1.24 ± 0.03		
	- 3	82	0.69 ± 0.02			82	0.81 ± 0.02		
	- 4	40	0.62 ± 0.06			40	0.95 ± 0.04		
Exploit	- 1	180	0.91 ± 0.02	0.0000***		180	1.17 ± 0.02	0.0000***	
	- 2	122	0.67 ± 0.02			122	0.86 ± 0.02		
Syst	- 1	40	0.62 ± 0.04	0.0000***		40	0.96 ± 0.05	0.0431*	
	- 2	262	0.84 ± 0.01			262	1.06 ± 0.02		
Season	- 1	139	0.67 ± 0.02	0.0000***		139	0.93 ± 0.02	0.0000***	
	- 2	163	0.90 ± 0.02			163	1.10 ± 0.02		

*$P<0.05$, **$P<0.01$, ***$P<0.001$, Exploit exploitation, Syst system, Interactions: Cu - Season×Herd 0.0000***, Zn - Season×Herd 0.0002***, Exploitation: 1 dairy, 2 beef, System: 1 ecological, 2 non-ecological, Season: 1 spring, 2 fall

Discussion

At the present study, Ca, P, Cu, and Zn levels were significantly influenced by herd of cow. Probably different cattle breeds can metabolize these elements differently. WARD *et al.* (1995) suggested that Simmental and Charolais steers have a greater Cu requirement than Angus steers, because of lesser apparent absorption and retention of Cu. In a comparison among dairy cattle, Jersey cows were found to accumulate liver Cu faster and to a greater extent than Holstein cows fed a similar diet (McDOWELL and ARTHINGTON 2005). However, most of effects which influenced these herds can have a multifactorial origin (HERNANDEZ *et al.* 1997, BÖMKES *et al.* 2004, ENJALBERT *et al.* 2006). Normal physiological values of different blood parameters of animals are influenced by a number of factors such as age, sex, breed, season, altitude, climatic conditions, nutrition and life habits (KIRK and DAVIS 1970, WARD *et* *al.* 1995, BARANOWSKI 2002, AHOLA *et al.* 2004, REECE 2004, PATKOWSKI *et al.* 2006, MORRIS *et al.* 2006). Insufficiencies are strongly associated with low production, poor health, and impaired locomotion (KEBREAB *et al.* 2005, ENJALBERT *et al.* 2006).

At the present work were obtained higher levels Ca in dairy specialization and non-ecological system. Contrary, higher level of P had beef animals. The absorption, accumulation and toxicity of each element may be affected by diverse factors, including interactions with others (GALLO *et al.* 1996, LAVEN *et al.* 2007). There have been many reports of interactions between Cu and other elements in cattle. These interactions probably indicate that mineral balance in the body is regulated by important homeostatic mechanisms in which macro-elements compete with the essential metals (AHOLA *et al.* 2005). An important factor for absorption and utilization of trace elements is their desirable chemical form (KINAL *et al.* 2007). Recommendations for performance increasing and health betterment have been showed in several other studies when complexes trace minerals have replaced inorganic trace minerals (GRIFFITHS *et al.* 2007). Moreover, the trace mineral status of animals depends not only on dietary allowance, but also on the efficiency of digestion and storage, which both can be affected by interactions with other food constituents.

Higher concentrations of Cu and Zn were recorded in dairy specialization versus beef specialization and in non-ecological system at the present study. ENJALBERT *et al.* (2006) suggests that Cu and Zn deficiencies are risk factors for impaired production, reproduction and health in both beef and dairy herds. Cu and Zn content in the cows under study was not marginal, we did not find the deficiency in herds with possible exception of herd 4 during spring 2005 observation (5.1 $mg.kg^{-1}$ of Cu in dry matter) (Table 2).

The lowest Zn content in blood plasma was in the herd 3 during the spring 2006, where dry matter feed really contained the lowest amount of Zn (the spring 2006 11.8 $mg.kg^{-1}$, average for all observations of 28.5 $mg.kg^{-1}$) (Table 2). A comparison of the content of Zn between dairy and beef herds has found a 50 % lower concentration in beef herds (45.1 $mg.kg^{-1}$ vs. 90.5 $mg.kg^{-1}$) vs. dairy herds. The mineral supplementation of diets is fully compensated in dairy herds, but is much less inspecting in beef herds.

At the present study, there were significant season effects in the both microminerals Cu and Zn. Higher values were recorded in autumn than spring. Individual blood samples showed variation at different seasons. According to MTUI *et al.* (2007), plasma concentrations of *P*, Cu, and Zn increased in dry season, while that of Ca did not vary with seasons. Cu and Zn liver levels increased progressively with soil levels, and the pattern was especially marked for copper (SREJBEROVA *et al.* 2008). Because Zn and Cu compete for binding sites on enzymes and metalloproteins, it may be reasonable to speculate that these minerals are antagonistic to some degree (HATFIELD *et al.* 2001). Additionally, seasonal spring calving in beef herds may make it particularly difficult for cows to meet their Cu and Zn requirements, as Cu and Zn contents of pasture tend to be lowest in spring (SOCHA *et al.* 2002, citated GRIFFITHS *et al.* 2007, CERMAK *et al.* 2006). Another explanation for the season differences might be related to breed of cattle.

At the present study was found interaction in Ca and P between Season×Herd. These interactions probably indicate that mineral balance is influenced of breeds and especially by the feeding during year. So far the feed rations in dairy herds have been mostly balanced, while rations in beef herds were unstable and frequently fluctuated.

There have been many reports of interactions between Cu and other elements in cattle and other livestock species. Three-way interactions between Cu, molybdenum and sulphur have also been reported (UNDERWOOD and SUTTLE 2001), so that the daily Cu requirements of cattle are strongly dependent on molybdenum and sulphur levels in the diet. Also sulfur interferes with normal Cu absorption in cattle (McDOWELL and ARTHINGTON 2005).

The toxic effects of observed elements on cows may become evident in health disturbances and decreased performance and reproductive indices (KOTTFEROVA and KORENEKOVA 1997). However, not record any health problems in the observed animals.

In conclusion although exposure of toxic elements in cattle in this study was very low and macrominerals and trace essential metals were within the adequate ranges, there were significant associations between observed elements and environmental factors. Results showed that different in plant species and seasonal changes contribute highly to the variation in mineral content of forage grazed beef cows.

It was confirmed that the level of metabolic processes, as demonstrated using blood indicators, may affect the values of macro and microminerals in cattle.

The present results of variable impact of seasonal changes on the concentration of minerals in the blood and feed resources suggests the need for supplementation of deficient minerals like Cu and Zn in the forms, which are available for optimal cows' productivity.

In beef cows nutrition, the precisely balance of daily rations, particularly on minerals, is necessary. The content of minerals in different plant fodders given to cattle can be namely much diversified.

It can be conclude that micromineral and trace mineral contents may be impacted of herd, exploitation, management system, and season in cows.

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