ORIGINAL ARTICLE

Seasonal variations of some serum electrolyte concentrations in sheep and goats

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Abstract The aim of this study was to evaluate the serum concentration of calcium, chloride, phosphorus, magnesium, sodium, and potassium, in sheep and goats, during 12 months. The experimental subjects were six female sheep (Valle del Belice breed; aged 4 ± 1 years; mean weight, 68±6 kg) and six female goats (Maltese breed; aged 3 ± 1 years; mean weight, 45 ± 3 kg). All subjects were clinically healthy and not pregnant nor lactating before and during the study. Blood samples were collected every 30 days at the same time (09:00) for 1 year (12 months). On the obtained sera, calcium, chloride, phosphorus, and magnesium were assessed by means of UV spectrophotometry; sodium and potassium were assessed by means of semiautomatic flame photometry. The application of the periodic model and statistical analysis of the cosinor procedure demonstrated seasonal rhythms of calcium, chloride, and sodium only in goats. Calcium showed the acrophase during the spring season, whereas the sodium and chloride serum values showed a seasonal rhythm with the acrophase in early summer. Our study sheds light on the seasonal difference existing between sheep and goats and provides useful information in livestock productions.

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I. Vazzana · S. Dara Experimental Zooprophylactic Institute of Sicily, Palermo, Italy Keywords Seasonal variations \cdot Sheep \cdot Goats \cdot Serum electrolyte

Introduction

The significance of determining physiological indices in domestic animal production has been well documented (Daramola et al. 2005). Small ruminant species have instead been poorly studied, despite their importance in livestock productions in the Mediterranean area (Piccione et al. 2009). The most important reason to assess the mineral status of these animals is usually regarding the lowering of the productivity performance (Kincaid 1999). Few information, however, has been provided during the last years about the macro-mineral status in sheep and goat serum. Electrolytes are a critical element in cellular metabolism, muscle contraction, nerve transmission, and enzyme reactions (Piccione et al. 2007b). Numerous studies have shown that mineral deficiencies lead to impaired growth and reproduction and to an increase in disease incidence, and it seems that these are strictly related to the diet mineral concentrations (Xin et al. 2010). In fact, the most frequent disease is related to some physiological status, during which the body requests a major amount of some electrolytes, such as pregnancy and lactation (Liesegang and Risteli 2005). The liver is the organ that often represents the status of several mineral elements in animals (Kincaid 1999). As demonstrated by several studies on serum and physiological parameters, a seasonal variation occurs in different animal species, and this is due to the hypothalamic suprachiasmatic nuclei activity. It is the principal generator of the rhythms which follow the life of an animal (Alila-Johansson et al. 2004). It is known to participate in all the conditions of light and dark and seems to regulate some metabolic and endocrine functions (Alila-Johansson et al. 2004). The seasonal rhythms reflect the endogenous adaptive mechanism to react in advance to the regular environmental changes associated with the seasons (Piccione et al. 2009). Limited information is provided about the seasonal rhythms of small ruminants (Piccione et al. 2009; Piccione et al. 2007a; Alila-Johansson et al. 2001, 2004). As of now, no studies are available regarding the seasonal mineral variations in small ruminants, not related to pregnancy or lactation.

For such consideration, the aim of this study was to evaluate the serum content of electrolytes both in goats and sheep, during 12 months of study.

Materials and methods

Subjects

The experimental subjects were six female sheep (Valle del Belice breed; aged 4 ± 1 years; mean weight, 68 ± 3 kg) and six female goats (Maltese breed; aged 3±1 years; mean weight, 45 ± 3 kg). All subjects were clinically healthy and not pregnant nor lactating before and during the study. They were free from internal and external parasites. No medication was administered from 1 month before the study. Their health status was evaluated daily based on behavior, rectal temperature, heart rate, respiratory profile, cough, nasal discharge, ocular discharge, appetite, fecal consistency, and hematological profile. Animals were housed in Sicily, Italy (latitude, 38° 6' N; longitude, 13° 20' E; altitude, 50 m above sea level) in two pens different for each species. Photoperiod in this region varies from 15 h and 53 min of light at the summer solstice to 10 h and 31 min of light at the winter solstice. The area is characterized by an annual rainfall of 51.5 mm (range, 5-98), generally occurring in autumn and winter. Mean annual maximum and minimum temperatures are 33°C in August and 10°C between January and February, respectively, with relative humidity between 69% and 73% through the year. Ambient temperature and relative humidity were continuously recorded during all the experimental period with a data logger (Gemini, Chichester, West Sussex, UK). Successively, temperature-humidity index (THI) was calculated using the following equation:

 $\text{THI}_{[^{\circ}\text{C}]} = t_{\text{bs}} - (0.55 - 0.55\varphi/100) \ (t_{\text{bs}} - 14.4)$

Where t_{bs} =dry-bulb temperature (degrees centigrade) and φ =relative humidity (percent) (Fig. 1).

All the animals had free access to water and to goodquality alfalfa hay. Concentrate (oats, 23%; corn, 36%; barley, 38%; and mineral and vitamin supplements, 3%) was provided once daily (200 g per animal per day).



Fig. 1 Ambient temperature (°C), relative humidity (%), **a** and THI (temperature–humidity index, °C), **b** recorded during the experimental period (12 months)

Protocols of animal husbandry and experimentation followed the applicable regulation in Italy.

Procedures

Blood samples were collected through an external jugular venipuncture using Vacutainer tubes (Terumo Corporation, Japan) with no additive every 30 days at the same time (09:00) for 1 year (12 months). After collection, blood samples were centrifuged at 3,000 rpm for 10 min. On the obtained sera, stored at -20° C pending analysis, calcium, chloride, phosphorus, and magnesium were assessed by means of UV spectrophotometry; sodium and potassium were assessed by means of semiautomatic flame photometry (FP 20, SEAC, Italy).

Statistical analyses

All the results were expressed as mean \pm SD. Data were normally distributed (P < 0.05, Kolmogorov–Smirnov test). One-way ANOVA for repeated measures was applied to evaluate a significant effect of time on the parameters studied. P values <0.05 were considered statistically significant. Data were analyzed using the software STATISTICA 5.5 (StatSoft Inc., USA). Using cosinor rhythmometry, four rhythmic parameters were determined: mesor (mean level), amplitude (half the range of oscillation), and acrophase (Φ , time of peak). _

Table 1 Mean values ± standard deviation of the serum minerals of six sheep and six goats, during the experimental period (12 months), expressed in their units of measurement

Months	Parameters									
	Ca ²⁺ (mmol/L)	P ⁻ (mmol/L)	K ⁺ (mmol/L)	Cl ⁻ (mmol/L)	Na ⁺ (mmol/L)					
	Sheep									
May	2.20±0.39	1.55 ± 0.30	4.55 ± 0.56	113.17±7.25	145.83 ± 6.86					
Jun	2.37±0.34	2.63 ± 0.33	$4.50 {\pm} 0.55$	128.83 ± 16.41	159.50±18.36					
Jul	$1.71 {\pm} 0.20$	$2.72 {\pm} 0.46$	4.12 ± 0.15	111.33 ± 2.88	$145.83 {\pm} 9.19$					
Aug	$2.36 {\pm} 0.22$	$2.34 {\pm} 0.33$	4.23 ± 0.20	$109.17 {\pm} 2.40$	$142.33 {\pm} 9.19$					
Sept	$1.85 {\pm} 0.26$	$3.08 {\pm} 0.65$	$4.57 {\pm} 0.27$	112.83 ± 6.37	$146.83 {\pm} 9.87$					
Oct	$1.59 {\pm} 0.22$	$2.15 {\pm} 0.58$	$4.50 {\pm} 0.40$	110.83 ± 6.47	$144.50 {\pm} 9.78$					
Nov	1.95 ± 0.35	1.65 ± 0.10	$4.38 {\pm} 0.44$	113.50 ± 7.37	$147.33 {\pm} 6.86$					
Dec	1.42 ± 0.24	$2.36 {\pm} 0.58$	$4.28 {\pm} 0.84$	111.33 ± 7.40	146.17 ± 6.11					
Jan	$2.50 {\pm} 0.60$	$2.14{\pm}0.56$	4.62 ± 0.23	110.40 ± 3.61	142.80 ± 8.79					
Feb	2.25 ± 0.25	1.41 ± 0.02	4.65 ± 0.51	113.67±7.09	$145.83 {\pm} 6.86$					
Mar	$2.36 {\pm} 0.47$	$2.72 {\pm} 0.33$	$4.50 {\pm} 0.40$	$109.17 {\pm} 9.40$	$144.50 {\pm} 7.78$					
Apr	$1.85 {\pm} 0.40$	$2.34 {\pm} 0.46$	$4.38{\pm}0.44$	112.83 ± 9.37	$147.33 {\pm} 6.86$					
	Goats									
May	$2.24 {\pm} 0.14$	$1.60 {\pm} 0.58$	$4.25{\pm}0.49$	116.00 ± 7.02	$145.00{\pm}13.92$					
Jun	$2.41 {\pm} 0.09$	$2.33 {\pm} 0.59$	$4.88 {\pm} 0.23$	118.00 ± 9.71	152.20 ± 8.56					
Jul	2.12 ± 0.20	$1.88 {\pm} 0.57$	$4.70 {\pm} 0.91$	126.50 ± 6.29	$151.70{\pm}13.88$					
Aug	$2.41 {\pm} 0.22$	$1.71 {\pm} 0.36$	4.43 ± 0.22	110.70 ± 7.23	$146.80 {\pm} 6.82$					
Sept	$1.01 {\pm} 0.19$	$2.09{\pm}0.40$	$4.40 {\pm} 0.35$	104.70 ± 5.39	$141.70 {\pm} 8.50$					
Oct	2.02 ± 0.22	$2.16 {\pm} 0.68$	$5.01 {\pm} 0.24$	$113.30{\pm}6.28$	$138.50 {\pm} 5.99$					
Nov	$1.68 {\pm} 0.35$	$1.55 {\pm} 0.41$	$4.65 {\pm} 0.30$	$105.50 {\pm} 8.21$	$137.20 {\pm} 9.78$					
Dec	$1.71 {\pm} 0.06$	$2.04 {\pm} 0.42$	$4.45 {\pm} 0.28$	106.70 ± 5.92	$140.20 {\pm} 5.60$					
Jan	$1.97 {\pm} 0.10$	$2.40 {\pm} 0.25$	$4.81 {\pm} 0.35$	$109.80{\pm}2.31$	$144.70 {\pm} 2.06$					
Feb	$2.19 {\pm} 0.25$	$2.08 {\pm} 0.44$	$4.83\!\pm\!0.30$	106.00 ± 4.94	$137.30 {\pm} 5.20$					
Mar	2.21 ± 0.22	$1.92 {\pm} 0.61$	$4.91 {\pm} 0.74$	104.70 ± 3.01	$135.30{\pm}2.42$					
Apr	$2.59 {\pm} 0.19$	$1.82 {\pm} 0.24$	$4.76 {\pm} 0.37$	110.20 ± 4.70	$142.70 {\pm} 5.88$					

Results

Table 1 shows the mean values of the parameters studied, together with the standard deviation of the means. One-way ANOVA for repeated measures showed a statistically significant effect of time on calcium and phosphorus in both the species. The application of the periodic model and statistical analysis of the cosinor procedure demonstrated seasonal rhythms of calcium, chloride, and sodium only in goats. Table 2 shows the acrophases of the parameters showing a seasonal rhythmicity. Figure 2 represents graphically the trend of the serum electrolytes which showed a seasonal rhythmicity.

Discussion

In our study, calcium, sodium, chloride, potassium, phosphorus, and magnesium values were within the physiological range for sheep and goats. A seasonal rhythmicity was

Table 2	Mesor (M) with 9	95% confidence ir	terval (CI),	amplitude	(A), and a	acrophase (Φ)	, express	ed in mon	ths, with	95% CI an	d F and	P values,
resulting	from ANOVA ap	plication, of the	parameters ($(Ca^{2+}, Cl^{-},$	and Na ⁺) showing a s	easonal r	periodicity	during	12 months	of study	in goats

Parameters	Goats	Goats									
	Mesor	CI 95%	A	Φ	CI 95%	F _(11; 5)	P<				
Ca ²⁺	2.63	2.89-2.35	1.62	16 Apr	(29 Jun-3 Feb)	2.89	0.005				
Cl ⁻	111.09	114.39-107.79	6.17	16 Jun	(20 Sep-26 Mar)	Ns	Ns				
Na ⁺	142.77	140.17-145.37	5.57	20 Jun	(19-Apr-22 Aug)	Ns	Ns				

Ns not significant



Fig. 2 Mean (\pm SD) serum concentrations of calcium, sodium, and chloride, expressed in their conventional units of measurement, obtained in six sheep and six goats, with the acrophase (Φ), during 12 months of study

demonstrated for calcium, sodium, and chloride in the serum of goats. Calcium is required for the normal functioning of a wide variety of tissues and physiologic processes (Horst et al. 1994). The most amount of calcium is absorbed from the lumen of the intestine, in tracts different in sheep and in goats (Huber et al. 2002). Calcium is present in plasma in three forms: bound to proteins, complexed, and ionized (Piccione et al. 2007b). Serum calcium is reported as total calcium (Piccione et al. 2007b). More studies support the conception that the feeding schedule acts as a regulator of many endogenous peacemakers, more than light-dark cycles, but since all the animals had the same kind of feed and husbandry, we can hypothesize that a substantial difference occurs between these species (Piccione et al. 2007b; Milhaud et al. 1972). Calcium showed the acrophase during the spring season, and it is rarely limiting in forage diets; however, it was observed that, during winter, its serum value is lower, probably because of a lower vitamin D concentration, known to regulate calcium homeostasis, and to the relative ability of the animal to absorb and utilize it (Underwood and Suttle 1999). Another important aspect is represented by the dietary content of phosphorus which can influence calcium absorption and utilization (Huber et al. 2002).

Sodium is the primary ion involved in every metabolic process, from glucose transport to neural transmission, and its change influences the chloride concentration, as an increase or a decrease (Piccione et al. 2007b). In our research, the sodium and chloride serum values showed a seasonal rhythm with the acrophase in early summer, so it is possible that a thermal stress occurred in the subjects of the study. The general homeostatic responses to thermal stress in mammals include reduction in fecal and urinary water losses and an increase in respiratory rates (Alamer 2006: Silanikove 2000). This could take to a minimal hemoconcentration and, consequently, to an increase of sodium and chloride in the serum of goats. Moreover, the seasonal variation in sodium and chloride content is consistent with that occurring in forages and in soil mineral content, that just in spring-summer seasons are richer in minerals than during the rest of the year (Xin et al. 2010). It seems that, in special environmental conditions, ruminants develop a special regulatory mechanism in the kidney in terms of glomerular filtration rate (Xin et al. 2010). As stated before, there is a different absorption of minerals in goats and sheep. In goats, there exists a phosphorusdependent transport of sodium in the jejunum, whereas in sheep, there is a sodium-independent transport in the duodenum (Huber et al. 2002). In our experiment, a significant effect of time was observed in phosphorus. The higher phosphorus serum values were observed concomitantly with the acrophase of sodium, in June.

Our study sheds light on the seasonal difference existing between sheep and goats in serum electrolyte content and demonstrated that a seasonal fluctuation of some electrolytes exists. This can be helpful to support the lowering of livestock productions due to a lack of proper integration of the electrolyte where it is needed.

Further studies should be conducted to better investigate how different environmental conditions influence the serum values of electrolyte concentrations.

References

Alamer M (2006) Physiological responses of Saudi Arabia indigenous goats to water deprivation. Small Rum Res 63:100–109

Alila-Johansson A, Eriksson L, Soveri T, Laakso M (2001) Seasonal variation in endogenous serum melatonin profiles in goats:

a difference between spring and fall? J Biol Rhythms 16:254-263

- Alila-Johansson A, Eriksson L, Soveri T, Laakso M (2004) Daily and annual variations of free fatty acid, glycerol and leptin plasma concentrations in goats (*Capra hircus*) under different photoperiods. Comp Biochem Physiol A 13:119–131
- Daramola JO, Adeloye AA, Fatoba TA, Soladoye AO (2005) Haematological and biochemical parameters of West African Dwarf goats. Livest Res Rural Dev, 17
- Horst LR, Goff JP, Reinhardt TA (1994) Symposium: calcium metabolism and utilization. Calcium and Vitamin D in the dairy cow. J Dairy Sci 77:1936–1951
- Huber K, Walter C, Bernd S, Breves G (2002) Phosphate transport in the duodenum and jejuni of goats and its adaptation by dietary phosphate and calcium. Am J Physiol Regul Integr Comp Physiol 283:R296–R302
- Kincaid RL (1999) Assessment of trace mineral status of ruminants: a review. Proceedings of the American Society of Animal sciences.
- Liesegang A, Risteli J (2005) Influence of different calcium concentration in the diet on bone metabolism in growing dairy goats and sheep. J Anim Physiol Anim Nutr 89:113–119

- Milhaud G, Perault Staub AM, Staub JF (1972) Diurnal variations of plasma calcium and calcitonin function in the rat. J Physiol 222:559–567
- Piccione G, Caola G, Refinetti R (2007a) Annual rhythmicity and maturation of physiological parameters in goats. Res Vet Sci 83:239–243
- Piccione G, Casella S, Giannetto C, Fazio F, Caola G (2007b) Effects of short time fed deprivation on some blood electrolytes in horses. Folia Vet 51:137–141
- Piccione G, Giannetto C, Casella S, Caola G (2009) Annual rhythms of some physiological parameters in *Ovis aries* and *Capra hircus*. Biol Rhythm Res 40:455–464
- Silanikove N (2000) Effects of heat stress on the welfare of extensively managed domestic ruminants. Livest Prod Sci 67:1–18
- Underwood EJ, Suttle NF (1999) The mineral nutrition of livestock, 3rd edn. CABI Publishing, New York, pp 112–149
- Xin GS, Long RJ, Guo XS, Irvine J, Ding LM, Ding LL, Shang ZH (2010) Blood mineral status of grazing Tibetan sheep in the Northeast of the Qinghai-Tibetan Plateau. Livest Sci. doi:10.1016/j.livsci.2010.08.007