

The influence of parity on bone alkaline phosphatase during the transition period in Saanen goats

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ABSTRACT

There is still much vagueness about changes in bone metabolism during pregnancy and lactation in small ruminants. The purpose of this study was to assess changes in BALP activity during pregnancy and lactation, as well as the effect of parity on bone formation in goats. The research was conducted on 35 goats of Saanen breed which were divided into 3 groups based on the number of previous deliveries (primiparous, biparous and multiparous goats). Blood samples were obtained 15 days before and 15 days after delivery. Differences between the study groups were tested by Student's t-test, and the correlation by Pearson's Rank Order test. In primiparous goats BALP activity was significantly lower ($P < 0.001$) 15 days after delivery compared to the activity measured 15 days before delivery. In biparous and multiparous goats there were no statistically significant differences in BALP activity pre- and post-delivery. There was also only a statistically significant difference in BALP activity between the primiparous group and bi- and multiparous groups of goats. There was a statistically significant negative correlation ($r = -0.88$; $P < 0.001$) between BALP and age in all goats. Results demonstrated active bone formation in late pregnancy and early lactation, especially in primiparous goats, indicating an increased bone turnover during the transition period while parity did not have an impact on bone formation.

Key words: bone alkaline phosphatase, goats, bone turnover, pregnancy, lactation

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Introduction

The reproductive cycle of dairy animals consists of mating, pregnancy, dry period, delivery and lactation. During pregnancy and lactation, animals are exposed to high metabolic strain which means a greater need for certain nutrients, vitamins and minerals, especially calcium and phosphorus which are essential for foetal growth, as well as for increased milk production (FILIPOVIĆ et al., 2010).

Bone turnover consists of bone formation and bone resorption, which are, in healthy subjects, balanced, so there is no net change in bone mass (ALLEN, 2003). However, in body conditions such as pregnancy and lactation, when there is a greater need for calcium and other minerals due to the growth of the foetus, a shift in bone turnover occurs in favour of bone resorption, which can lead to decreased bone mass (GIESEMANN et al., 1998; TOJO et al., 1998; KALKWARF and SPECKER, 2002; OLIVERI et al., 2004). Data about the effect of parity on bone turnover in animals are still scarce. To the authors' knowledge there has been only one study (LIESEGANG et al., 2007) that investigated changes in bone metabolism during the first and second pregnancy and lactation, but it did not include the influence of a higher number of pregnancies on bone turnover.

The most frequently used marker of bone formation is bone alkaline phosphatase (BALP), which is synthesised by osteoblasts during bone formation (MILLAN, 2006; MASROUR and MAHJoub, 2012). Since BALP reflects the exact osteoblast activity it is a useful parameter to monitor changes in bone formation (Van STRAALen et al., 1991; LEUNG et al., 1993).

Different responses of the bone metabolism to increased need for calcium during pregnancy and lactation have been noticed in small ruminants, and these responses are different than those in some other animal species (LIESEGANG and RISTELI, 2005). Therefore, knowledge about bone metabolism in humans, cows, rodents and dogs cannot completely be applied to small ruminants. Since there is still much vagueness about the changes in bone metabolism during pregnancy and lactation in small ruminants, the purpose of this research was the assessment of bone formation in goats of different parity status during late pregnancy and early lactation, by the evaluation of BALP changes during these metabolic states.

Materials and methods

The study was approved by the Ethics Committee of the Faculty of Veterinary Medicine, University of Zagreb.

Animal groups and blood collection. The research was conducted on 35 Saanen breed goats, aged between 1 and 8 years. The animals were accommodated on a farm in cotes, with free range. The average body mass of the animals (\pm SE) was 55 (\pm 0.73) kg. The goats were fed twice a day and always ate the whole ration. The rations consisted of hay,

straw and concentrate. Mean daily dry matter intake was 650 g with 5% of crude fat, 23% of crude fibre and 15% of crude protein. The Ca content of the diet was 18.1 g per day.

The animals were divided into 3 groups based on the number of previous deliveries. The first group was formed of primiparous goats ($n = 13$), the second of biparous goats ($n = 13$), and the third of multiparous goats (3-7 deliveries; $n = 9$). The average age of the primiparous goats at the moment of blood collection was one year, of the biparous goats two years, while the multiparous goats were approximately 5.2 years old. Blood samples were obtained twice, 15 days before the estimated date of delivery and 15 days after delivery.

The blood samples (2 mL) were collected in the morning before feeding from the v. jugularis in Vacutainer® tubes, without additives (Aichele Medico AG, Basle, Switzerland), centrifuged at 1500 pm for 15 minutes and then stored at $-70\text{ }^{\circ}\text{C}$ until further processing.

Assay for the BALP activity. The activity of BALP was measured with a commercial human enzyme immunoassay kit (Metra™ BAP EIA kit, Quidel Corporation, San Diego, USA). The assay is based on the ELISA method which uses anti-BALP antibodies for determination of BALP activity in serum and plasma. Enzyme activity is detected by the substrate pNPP (p-nitrophenil phosphate). Although the assay was developed to determine BALP activity in humans, a significant cross-reactivity with sheep and goat serum has been found (LIESEGANG et al., 2006).

Statistical analysis. Differences between the study groups were tested by the Student's *t*-test. To evaluate the correlation between BALP activity and age, Pearson's Rank Order correlation was used. The statistically significant difference between values was set at $P < 0.05$. SigmaStat 3.0 for Windows (Jandel Corporation, San Rafael, CA, USA) was used for statistical analysis.

Results

The mean values (\pm SE) of BALP activity in three studied groups before and after delivery are shown in Fig. 1. A statistically significant difference ($P < 0.001$) in BALP activity was found in the group of primiparous goats between the first and second blood collection, i.e. BALP activity was significantly lower 15 days after delivery compared to the activity measured 15 days before delivery. In biparous and multiparous goats there was no statistically significant difference between the first and second blood collection (Fig. 1).

When comparing BALP activity between the groups of different parity status, a statistically significant difference was found between primiparous and biparous goats, as well as between primiparous and multiparous goats ($P < 0.001$). There was no statistically significant difference in BALP activity between biparous and multiparous goats (Fig. 2).

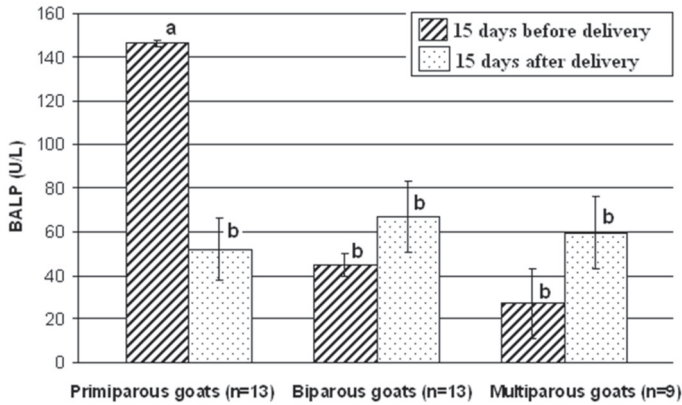


Fig. 1. Mean values (\pm SE) of BALP activity (U/L) measured 15 days before and 15 days after delivery in three groups of goats. A statistically significant difference ($P < 0.001$) is marked with different letters (a,b).

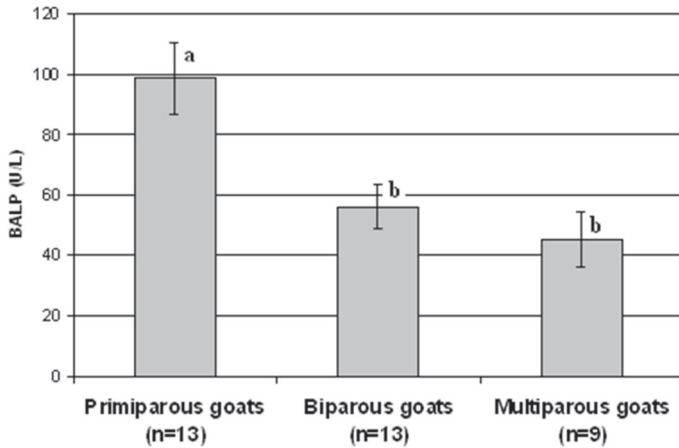


Fig. 2. Mean values (\pm SE) of BALP activity (U/L) in three groups of different parity status. A statistically significant difference ($P < 0.001$) is marked with different letters (a,b).

There was a statistically significant negative correlation ($r = -0.88$; $P < 0.001$) between BALP and age in all goats (Fig. 3).

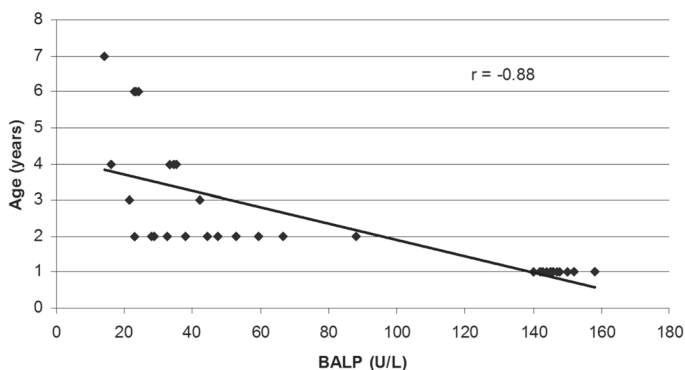


Fig. 3. Correlation between BALP activity (U/L) and age (years) in all goats before delivery; $n = 35$, $r = -0.88$, $P < 0.001$.

Discussion

According to the results of our study, late pregnancy and early lactation influence bone turnover in goats. In the group of primiparous goats, the mean BALP activity before delivery was significantly higher than after delivery. Moreover, the mean BALP activity before delivery was the highest and statistically significantly different compared to the other groups of goats. We believe that this result is due to the age of the animals. Particularly, the primiparous goats in this investigation were seven months old during their first mating, which is the time when the goats become sexually mature, but still have not reached their full body mass, i.e. their body growth is still not completely finished (MIOČ and PAVIĆ, 2002). Therefore, we believe the increased BALP activity in these primiparous goats is a physiological finding due to the growth of the animals resulting in intense bone turnover. LIESEGANG et al. (2006) also found elevated BALP activity in primiparous goats, but the reported values were not as high as in our study, possibly because the goats in their investigation were a year old at the time of the first mating. After delivery, in early lactation of primiparous goats, there was a sudden and significant decrease in BALP activity. The same was noticed by LIESEGANG et al. (2006) in their investigation, where the lowest BALP activity was recorded 7 days after delivery. In that period, especially in the first reproductive cycle, a misbalance between bone formation and resorption occurs. Greater loss in bone mass during first reproductive cycle than in later cycles was found in rats (BOWMAN and MILLER, 1999). LIESEGANG and RISTELI (2005) found that bone turnover in goats is extremely elevated during the first reproductive cycle, while in later cycles it is still elevated, but not nearly as high as the first time. We believe the decrease in BALP activity during early lactation in primiparous goats is a result of the sudden physiological change in the organism, since the animals are experiencing that kind of

change for the first time, i.e. their organism's strong need for calcium during foetal growth and milk production. In subsequent pregnancies, the animals adapt to calcium loss, and the calcium from their food is sufficient for their organisms' needs in lactation (WILKENS et al., 2012). Moreover, it is documented that goats increase intestinal Ca absorption when there is a lack of this mineral in their diet (WILKENS et al., 2012).

In biparous and multiparous goats the mean values of BALP activity during late pregnancy were significantly lower than in primiparous goats. This fact can be explained by the completion of animal growth and faster organism adjustment to a new physiological need for calcium. There were no statistically significant differences between BALP activity 15 days before and 15 days after delivery in biparous and multiparous groups of goats. We believe that the animals adapt to the new circumstances more quickly in the second or later pregnancies than in the first pregnancy.

The mean BALP activities in multiparous goats were lower than in other study groups. This result was expected since the animals in that group had the highest average age, and previous studies showed decreased bone turnover with age (SANECKI et al., 1993; ALLEN, 2003).

Parity did not seem to have any influence on bone turnover, since there was no significant difference in BALP activity in relation to the number of deliveries between all three groups of goats, except the statistically higher BALP activity in primiparous goats. However, as we explained before, we believe this elevation was due to the physiological growth of young animals and the first major change in their metabolism and mineral balance due to their first pregnancy.

The statistically significant ($P < 0.001$) negative correlation between BALP and age in all goats confirms previous investigations about decreased BALP activity with aging, since in older animals bone turnover is physiologically reduced (SANECKI et al., 1993; PRICE et al., 1995).

BALP values in this investigation show that there is active bone formation, which in general reflects as increased bone turnover, during late pregnancy and early lactation in primiparous goats. On the basis of our results we can conclude that the parity does not significantly influence bone formation in mature, healthy animals.

Further investigations using markers of both bone formation and bone resorption are necessary in order to gain a more specific and detailed insight into bone turnover during reproduction in goats.

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SAŽETAK

Promjene u koštanom metabolizmu malih preživača tijekom gravidnosti i laktacije nisu do kraja razjašnjene i istražene. Cilj ovog istraživanja bio je procijeniti promjene u aktivnosti BALP-a tijekom gravidnosti i laktacije, te utjecaj pariteta na koštanu izgradnju koza. Istraživanje je provedeno na 35 koza sanske pasmine koje su bile podijeljene na 3 skupine, s obzirom na broj prethodnih jarenja (prvojarke, drugojarke i višejarke). Uzorci krvi prikupljeni su 15 dana prije i 15 dana poslije jarenja. Razlike među ispitivanim skupinama testirane su Studentovim *t*-testom, a korelacija Pearson's Rank Order testom. U prvojarci aktivnost BALP-a bila je statistički značajno niska ($P < 0,001$) 15 dana nakon jarenja u usporedbi sa aktivnosti mjerenom 15 dana prije jarenja. U drugojarci i višejarci nije bilo statistički značajne razlike u aktivnosti BALP-a prije i poslije jarenja. Ustanovljena je statistički značajna negativna korelacija ($r = -0,88$; $P < 0,001$) između BALP-a i dobi u svih koza. Rezultati ukazuju na aktivnu koštanu izgradnju tijekom kasne gravidnosti i rane laktacije posebno u prvojarci, što upućuje na povećanu koštanu pregradnju za vrijeme prelaznog perioda, dok paritet nije imao utjecaj na pregradnju kostiju.

Ključne riječi: koštana alkalna fosfataza, koze, koštana pregradnja, gravidnost, laktacija
