The concentration of biologically active molecules in colostrum is strongly related to the metabolic and endocrine status of the cow during the early postpartum period. The aim of this study was to determine concentrations of insulin like growth factor-I (IGF-I), triiodothyronine (T₃) and thyroxine (T₄) in mammary gland secretion during the late dry and colostral period and its possible association with the energy status of the cows as represented by blood glucose concentration. Ten healthy Holstein-Friesian dairy cows were involved in this study. Two samples of colostrum were taken from each animal: one between 19 and 53 hours (mean ± sd 32.10 ± 9.47 hours) before calving, and the second 2 hours after calving. Concentrations of IGF-I, T₃ and T₄ tended to decrease at the start of lactation but the changes were not statistically significant. Linear regression of colostral IGF-I and protein concentration before calving revealed a positive relationship (R² = 0.668, p<0.01) before calving but not post partum (R² = 0.348, p>0.05). Blood glucose concentration was 2.88 ± 0.32 mmol/L before and 2.28 ± 0.29 mmol/L after calving and the decrease was statistically significant (p<0.001). Blood glucose levels after calving were positively correlated with colostral IGF-I concentration after calving (R² = 0.695, p<0.01), but not before parturition (R² = 0.383, p>0.05).

Key words: cows, colostrum, IGF-I, thyroid hormones

INTRODUCTION

Colostrum contains not only nutrients but also biologically active molecules such as hormones, growth factors and cytokines (Grosvenor et al., 1993; Schams et al., 1994; Blum et al., 2008). Some colostral components are enriched in the mammary gland during the late dry period and then decrease rapidly during the early phase of lactation (Bojkovski et al., 2005). It seems that prepartal mammary secretion of bioactive molecules alters mammary gland function (Vega et al.,...
(Grosvenor et al., 1993).

The concentration of biologically active molecules in colostrum is strongly related to the metabolic and endocrine status of the cow during the early postpartum period. Peripheral blood concentrations of some hormones and metabolites, such as insulin-like growth factor-I (IGF-I) and glucose, decrease immediately after parturition markedly more so in ketotic than in healthy cows (Nikolić et al., 2001). Blood IGF-I and glucose concentrations were correlated ($R^2 = 0.669$) in ketotic cows after calving (Šamanc et al., 1998). However, no general association between circulating IGF-I or glucose and thyroid hormones concentrations was found during the peripartal period in dairy cows (Nikolić et al., 1997), even though peripheral circulating IGF-I concentrations appeared to be related to thyroid hormone status in ketotic cows. Namely, ketotic cows with low thyroid hormone levels maintained IGF-I concentrations within normal physiological limits, while those with high thyroid hormone levels had extremely low blood IGF-I concentrations.

The aim of this study was to determine concentrations of IGF-I, triiodothyronine ($T_3$) and thyroxine ($T_4$) in mammary gland secretion during the late dry and colostral period and its possible association with the energy status of the cows as represented by blood glucose concentration.

**MATERIALS AND METHODS**

**Animals**

Ten healthy Holstein-Friesian dairy cows that had yielded 7000 liters of milk per annum in the previous lactation were involved. Cows were at their 2nd to 5th calving.

**Colostral sampling**

Two samples of colostrum were taken from each animal: one between 19 and 53 hours (mean [sd] 32.10 [9.47] hours) before calving, and the second 2 hours after calving. These samples were stored in sterile bottles and frozen at $-20^\circ$C until analysis.

**Blood sampling**

Blood samples, taken by jugular venipuncture at the same time as colostrum samples were obtained with a sterilized needle into tubes containing sodium-fluoride and put immediately on ice. Within 30 to 60 min the tubes were centrifuged at 1000 x g for 20 min at 4°C.

**Analyses**

Concentrations of IGF-I, $T_3$ and $T_4$ in colostrum were measured by radioimmunoassay using commercial kits (INEP-Zemun, Serbia). Plasma concentrations of glucose were measured using commercial kits (GOD/PAP test, Dialab Austria).
Statistical analysis

The results are expressed as mean ± standard deviation [sd] for each group of calves. Probability and the statistical significance of differences between mean values were calculated using Student’s t-test. Simple correlation coefficient and regression of colostral IGF-I concentrations in relation to colostral protein and blood glucose, respectively were also calculated and analyzed.

RESULTS AND DISCUSSION

The mean concentrations of IGF-I, T3 and T4 in colostrum before and after calving are shown in Figure 1.

Figure 1. Mean (se) concentrations of insulin-like growth factor-I (IGF-I), triiodothyronine (T3) and thyroxine (T4) in cow colostrum sample taken before and after calving

The results show that, unlike the situation in the blood circulation, the levels of T3 and T4 were similar. Concentrations of IGF-I, T3 and T4 tended to decrease at the start of lactation but the changes were not statistically significant. Linear regression of colostral IGF-I and protein concentration before calving (Bojkovski et al., 2005) revealed a positive relationship ($R^2 = 0.668$, $p<0.01$; Figure 2) before calving but not post partum ($R^2 = 0.348$, $p>0.05$).

Blood glucose concentration (mean [sd]) was 2.88 [0.32] mmol/L before and 2.28 [0.29] mmol/L after calving and the decrease was statistically significant ($p<0.001$). Blood glucose levels after calving were positively correlated with colostral IGF-I concentration after calving ($R^2 = 0.695$, $p<0.01$; Figure 3), but not before parturition ($R^2 = 0.383$, $p>0.05$).
High levels of IGF-I in mammary gland secretion before and after parturition are in agreement with previously published data (Malven et al., 1987; Schams et al., 1991; Kirovski et al., 2002). The content of IGF-I in bovine colostrum does not reflect the level of IGF-I in cow plasma (Oda et al., 1989). The close positive correlation between IGF-I and protein concentration in mammary secretion before

Figure 2. Linear regression of colostral IGF-I and protein concentrations before calving ($y = 0.37x + 107.81, R^2 = 0.688, p<0.01$)

Figure 3. Linear regression of colostral insulin-like growth factor- I (IGF-I) and blood glucose concentrations after calving ($y = 0.012x + 1.54, R^2 = 0.695, p<0.01$)
calving indicates that IGF-I secretion may follow overall protein secretion. On the other hand, the significant positive correlation between blood glucose and colostral IGF-I concentration after calving could indicate that the energy status of the cow has a significant impact either on active transport of IGF-I from blood to colostrum or on its synthesis within the mammary gland and/or liver. It was found that glucose, in the form of glucose 6-phosphate, stimulated IGF-I synthesis and secretion in cultured fetal hepatocytes (Goya et al., 1999). It has been reported that the blood glucose and IGF-I concentration are in strong correlation around parturition (Šamanc et al., 1998), but there were no data about the relationship between blood glucose and colostral IGF-I concentrations.

The mean levels of T3 and T4 obtained in our study are higher than those found earlier (Slebodzinski et al., 1986), which might be explained by differences in the time of sampling in relation to parturition. Our results show no significant differences between T3 and T4 concentrations in mammary gland secretions before and after calving. T3 concentrations in colostrum were higher and T4 concentrations were lower than those found in blood during the peripartal period (Stojić et al., 2001). The difference between T4 and T3 concentrations in mammary secretion is due in part to a difference in the permeability of the blood-mammary barrier towards T4 and T3 (Sato and Suzuki, 1979), different transport mechanisms for T4 and T3, and to enzymatic generation of T3 via T4 to T3 conversion within the mammary gland (Slebodzinski and Brzezinska-Slebodzinska, 1991). Moreover, colostral thyroid hormone concentrations were not significantly correlated with blood glucose during that period.

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KONCENTRACIJA INSULINU SLIČNOG FAKTORA RASTA-I (IGF-I) I TIROEIDNIH HORMONA U KOLOSTRUMU KRAVA HOLŠTAJN-FRIZIJSKE RASE PRE I POSLE TELJENJA

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SADRŽAJ

Konzentracija biološki aktivnih molekula u kolostrumu je u visokoj korelaciji sa metaboličkim i endokrinim statusom krava tokom peripartalnog perioda. Cilj ovog rada je bio da se odrede koncentracije insulinu sličnog faktora rasta-I (IGF-I), trijodtironina (T3) i tiroksina (T4) u sekretu mlečne žlede tokom perioda kasnog zasušenja i ranog kolostralnog perioda, kao i njihova moguća korelacija sa energetskim statusom krava izraženim kroz koncentraciju glukoze u krvi. Ispitivanje je izvedeno na deset zdravih krava holštajn-frizijske rase. Koncentracije IGF-I, T3 i T4 su imale tendenciju pada na početku laktacije, ali promene nisu bile statistički značajne. Između koncentracije IGF-I i proteina u kolostrumu postojala je pozitivna korelacija (R² = 0,668, p<0,01) pre teljenja, ali ne i posle teljenja (R² = 0,348, p>0,05). Koncentracija glukoze u krvi je bila 2,88 ± 0,32 mmol/l pre i 2,28 ± 0,29 mmol/l posle teljenja, a smanjenje koncentracije je bilo statistički značajno (p<0,001). Koncentracija glukoze u krvi posle teljenja je bila u pozitivnoj korelaciji sa koncentracijom IGF-I u kolostrumu posle teljenja (R² = 0,695, p<0,01), ali ne i pre teljenja (R² = 0,383, p>0,05).