

## HORMONAL AND METABOLIC PROFILE IN SIMMENTAL DAIRY COWS DURING DRY PERIOD, EARLY AND MID LACTATION\*

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*SUMMARY: The aim of this study was to investigate endocrine and metabolic characteristic of Simmental cows in dry period, early and mid lactation. Experiment was included 45 Simmental cows: 15 in dry period (2 weeks before calving, period 1), 15 in early lactation (in first week after calving, period 2) and 15 in mid lactation (4-5 month after calving, period 3). Frequency distribution analysis shows that most of the parameters takes a normal distribution of distribution, unless the value of the total lipids (early lactation), urea (early lactation), T3 (dry period and mid lactation), T4 (early lactation) and insulin (dry period). The moment of blood collection has a substantial impact on the value of hormones and metabolites, except for total protein, T4 and insulin. Significant F value and posthoc LSD were significant for: glucose (F=25.44; LSD 1:2, 1:3, significantly higher concentrations of glucose the dry period), cholesterol (F=15.62; LSD 1:3, 2:3, significantly higher concentrations of cholesterol in the middle of lactation), triglycerides (F= 50.42 ; LSD 1:2, 1:3, significantly higher concentrations in the dry period), total lipids (F =27.89; LSD 1:3 , 2:3, significantly higher concentration in the middle of lactation), NEFA (F=10.74; LSD 1:2, 1:3, significantly higher concentrations in early lactation ), BHB (F= 24.39; LSD 1:2, 1:3, 2:3, significantly higher concentrations in early lactation), albumin (F=9.2, LSD 1:2, 1:3, significantly higher concentrations in the dry period ), urea (F=11.61; LSD 1:2, 2:3, significantly reduced concentration in early lactation), T3 (F=3.31; LSD 1:3, 2:3, significantly lower concentrations in dry period and early lactation), growth hormone (F=4.17; LSD 1:2, 2:3, higher concentrations in early lactation). Concentrations of hormones and metabolites in Simmental cows depend on whether the blood was taken during the dry period, early lactation and mid lactation. Differences in metabolite values indicate the existence of reduced food intake, a negative energy balance, the mobilization of lipids and altered functional status of hepatocytes*

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*in early lactation. Most of the studied parameters showed a normal frequency distribution. The absence of normal frequency distribution for some parameters, particularly during early lactation, may indicate high heterogeneity of cows in the process of adaptation to the periparturient period.*

**Key words:** *simental cows, endocrine status, metabolic profile, lactation, dry period.*

## INTRODUCTION

Parturition and lactogenesis are accompanied by many physiological changes that facilitate the maintenance of homeostasis (Bauman and Currie, 1980). Adaptation of the endocrine system during the transitional period is the key factor in maintaining metabolic balance (Aceves et al, 1985). Homeorhesis induces intense lipid mobilization and ketogenesis, and the liver is adapted to metabolic changes in dairy cows. Intensive postpartal lipid mobilization and ketogenesis are sufficient for a series of compensatory metabolic processes with changes in blood metabolic profile during lactation in healthy cows (Bell, 1995; Drackley, 1999; Rempis et al, 2011; Cincović et al., 2012). In addition to these, the period around calving is characterized by numerous metabolic and hematological changes such as decreased glycemia, decreased cholesterol concentrations, low concentrations of urea, elevated bilirubin concentration, reduced calcemia (Cincović et al., 2011; Đoković et al., 2013). Interdependent changes occur in the GH/insulin/IGF-I/glucose signaling pathway in early lactation (Lucy, 2001). Insulin plays a role in the adaptation of organic matter metabolism in dairy cows during the transitional period and during lactation, particularly in terms of nutrient redistribution and partitioning towards the mammary gland (insulin resistance) (Butler et al., 2003; Balogh et al., 2008). A decrease in thyroid hormone levels (hypothyroidism) occurs in the blood of peripartal cows, particularly during early lactation, when body reserves are mobilized for the production of high amounts of milk (Tiirats, 1997; Huszenicza et al., 2002).

The aim of this study was to investigate endocrine and metabolic characteristics of Simmental cows in dry period, early and mid lactation.

## MATERIAL AND METHODS

Experiment was included 45 Simental cows: 15 in dry period (2 weeks before calving, period 1), 15 in early lactation (in first week after calving, period 2) and 15 in mid lactation (4-5 month after calving, period 3). Blood samples were obtained by venepuncture of v.jugularis, 3-4 hours after morning feeding. Serum epruvettes with vacutainer system were used. Samples were transported to the laboratory immediately after venepuncture. Concentration of T3, T4, insulin and growth hormone were determined using a standard ELISA kit (Endocrine technologies inc., USA) and measured by Humareader device. Biochemistry parameters (glucose, NEFA, cholesterol, total lipids, triglycerides, total proteins, albumin and urea) were determined using a Biosistem, Elitech or Randox kit and measured by CobasMira+ spectrophotometer.

Statistics: Characteristics of frequency distribution were analysed by calculation of standard skewness standard kurtosis and Shapiro-Wilk test of

normality. Difference in concentration of parameters in three periods was analysed using a ANOVA analysis with posthoc LSD test.

## RESULTS AND DISCUSSION

Frequency distribution analysis shows that most of the parameters takes a normal distribution of distribution, unless the value of the total lipids (early lactation), urea (early lactation), T3 (dry period and mid lactation), T4 (early lactation) and insulin (dry period). When you look at the symmetry of the distribution (skewness), it is for most parameters identical for all three test periods. However, there are some disparities when it comes to the distribution of NEFA, BHB and urea. For values of NEFA and BHB in early lactation was found negative skewness value (left turn), indicating the dominance above-average values of NEFA and BHB. In contrast, the frequency distribution of urea in early lactation had a high positive value of skewness, which indicates the dominance of below-average values. All these results are shown in Table 1.

The moment of blood collection has a substantial impact on the value of hormones and metabolites, except for total protein, T4 and insulin (Table 2, Figure 1-13). Significant F value and posthoc LSD were significant for: glucose (F = 25.44 ; LSD 1:2, 1:3 , significantly higher concentrations of glucose the dry period), cholesterol (F= 15.62; LSD 1:3, 2:3, significantly higher concentrations of cholesterol in the middle of lactation), triglycerides (F= 50.42 ; LSD 1:2, 1:3, significantly higher concentrations in the dry period), total lipids (F = 27.89 ; LSD 1:3 , 2:3, significantly higher concentration in the middle of lactation), NEFA (F= 10.74; LSD 1:2, 1:3, significantly higher concentrations in early lactation), BHB (F = 24.39; LSD 1:2, 1:3, 2:3, significantly higher concentrations in early lactation), albumin (F= 9.2 ; LSD 1:2, 1:3, significantly higher concentrations in the dry period ), urea (F= 11.61; LSD 1:2, 2:3, significantly reduced concentration in early lactation), T3 (F= 3.31; LSD 1:3, 2:3, significantly lower concentrations in dry period and early lactation), growth hormone (F = 4,17; LSD 1:2, 2:3, higher concentrations in early lactation).

It is found that concentration of many metabolites are depend of energy balance (Đoković et al, 2013). The concentration of glucose is significantly lower in the first week postpartum, because of the reduced food intake and increased utilization of glucose for milk production (Doepel et al., 2002). NEFA concentration increases in the week after calving as a result of energy deficit and changes in hormonal status of cows (Drackley et al., 2005, Bertoni et al., 1998). NEFA is used for energy purposes and is a precursor for the synthesis of BHB in hepatocytes (Guo et al., 2008). The concentration of BHB is a significant indicator of the health and productivity of cows. Cows with high values of BHB have lower reproductive capacity, significantly lose body condition, produce a small amount of milk and suffer from extreme metabolic changes (Kessel et al., 2008, Huszenicza et al., 2006). Serum levels of glucose, triglycerides, total cholesterol, total protein, albumin and urea are indicators of hepatic function and decreases in their concentration may imply fat infiltration in the liver (Sevinc et al., 2003; Wathes et al., 2007; Đoković et al., 2007; Đoković et al., 2011). The concentration of urea depends on the intake of food, and energy balance (Rastani et al., 2006). Reduced concentration of proteins may occur as a result of increased use of amino acids in processes of gluconeogenesis in the period around calving. In favor of this assumption there is an

elevated concentration of AST, which is a significant indicator of catabolism of proteins and their use for gluconeogenesis (Seal and Reynolds, 1993). Lower insulin concentration in periparturient period compared to mid lactation is consequence of insulin resistance when decreased insulin concentration provide higher lipolysis and glucose utilisation in udder (DeKoster and Opsomer, 2013). The hormonal activity of thyroid gland has an important role in the transitional period for determining the cell metabolism intensity, metabolism of lipids and carbohydrates and lactation course itself by its thyroid hormones. Under the conditions of a negative energy balance and of high lipomobilization, the concentrations of thyroidea hormone in the blood were significantly lower in transitional period, with a markedly declined triiodothyronine in the blood shortly before and after calving (Đoković et al., 2010).

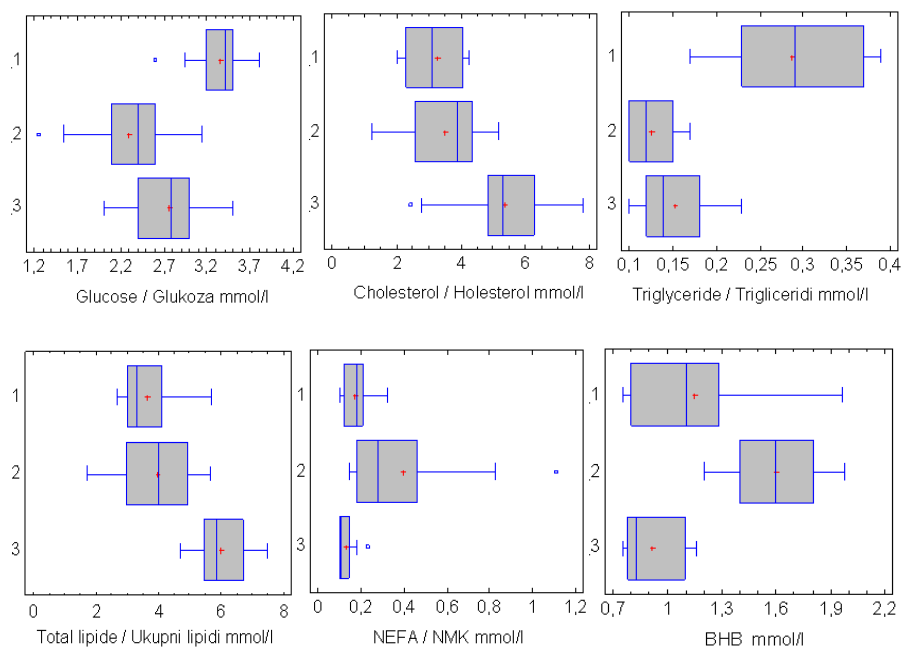
Table 1. Frequency distribution characteristics of endocrine and metabolic parameters in three different periods

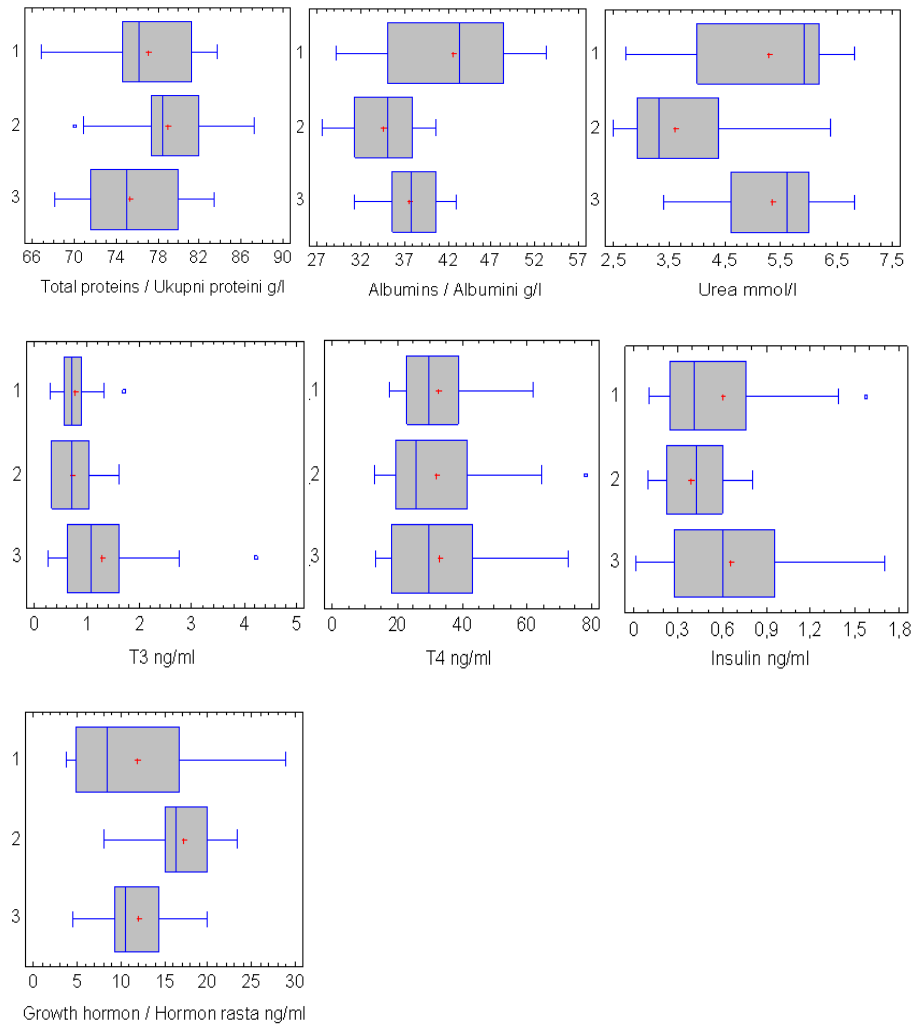
Parametar	Dry period			Early lactation			Mid lactation		
	Skew.	Kurt.	Normal. distrib.	Skew.	Kurt.	Normal. distrib.	Skew.	Kurt.	Normal. distrib.
Glucose mmol/l	-1,44	1,16	Yes	-0,9	0,42	Yes	-0,35	-0,15	Yes
Cholesterol mmol/l	-0,38	-1,11	Yes	-0,62	-0,33	Yes	-0,7	0,57	Yes
Triglycerides mmol/l	0,33	-0,57	Yes	0,88	-0,63	Yes	1,2	0,03	Yes
Total lipide mmol/l	1,7	1,7	Yes	3,1	2,6	No	0,65	-0,76	Yes
NEFA mmol/l	1,5	0,68	Yes	-1,1	0,79	Yes	1,84	1,55	Yes
BHB mmol/l	1,35	0,36	Yes	-0,1	-0,97	Yes	0,77	-1,3	Yes
Total proteins g/l	-1,01	0,44	Yes	-0,1	0,04	Yes	0,28	-0,54	Yes
Albumin g/l	-0,55	-0,51	Yes	-0,46	-0,27	Yes	-0,38	-0,22	Yes
Urea mmol/l	-1,21	-0,61	Yes	2,23	1,68	No	-0,75	-0,29	Yes
T3 ng/ml	2,01	1,5	No	1,17	0,33	Yes	3,32	3,84	No
T4 ng/ml	1,91	0,8	Yes	2,5	1,65	No	1,47	0,41	Yes
Insulin ng/ml	2,36	1,34	No	0,97	0,15	Yes	0,97	0,15	Yes
Growth hormon ng/ml	1,31	-0,26	Yes	0,55	-0,21	Yes	0,55	-0,21	Yes

Table 2. ANOVA and LSD test – Influence of blood sampling period to hormone and metabolite concentration

Parametar	F	LSD
Glucose mmol/l	25,44**	1:2, 1:3
Cholesterol mmol/l	15,62**	1:3, 2:3
Triglycerides mmol/l	50,42**	1:2, 1:3
Total lipide mmol/l	27,89**	1:3, 2:3
NEFA mmol/l	10,74**	1:2, 1:3
BHB mmol/l	24,39**	1:2, 1:3, 2:3
Total proteins g/l	2,26	/
Albumin g/l	9,2**	1:2, 1:3
Urea mmol/l	11,61**	1:2, 2:3
T3 ng/ml	3,31 *	1:3, 2:3
T4 ng/ml	0,04	/
Insulin ng/ml	1,17	/
Growth hormon ng/ml	4,17**	1:2, 2:3

\*p<0,05; \*\*p<0,01





Graph 1-13. Hormone and metabolite concentration in dry period (1), early (2) and mid (3) lactation.

## CONCLUSION

Concentrations of hormones and metabolites in Simmental cows depend on whether the blood was taken during the dry period, early lactation and mid lactation. Differences in metabolite values indicate the existence of reduced food intake, a negative energy balance, the mobilization of lipids and altered functional status of hepatocytes in early lactation. Most of the studied parameters showed a normal frequency distribution. The absence of normal frequency distribution for some parameters, particularly during early lactation, may indicate high heterogeneity of cows in the process of adaptation to the periparturient period.

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# **HORMONALNI STATUS I METABOLIČKI PROFIL KRAVA SIMENTALSKE RASE U PERIODU ZASUŠENJA, TOKOM RANE LAKTACIJE I SREDINE LAKTACIJE**

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## **Izvod**

Cilj ovog rada je da se ispituju endokrine i metaboličke karakteristike krava simentalске rase u različitim periodima laktacije i zasušenju. U ogled je uključeno 45 krava: 15 krava u zasušenju (2 nedelje pre teljenja, period 1), 15 krava u ranoj laktaciji (prva nedelja posle teljenja, period 2) i 15 krava u sredini laktacije (4-5 meseci posle teljenja, period 3). Analiza distribucije frekvencije pokazuje da većina parametara zauzima normalnu raspodelu distribucije, osim vrednosti ukupnih lipida (rana laktacija), uree (rana laktacija), T3 (period zasušenje i sredina laktacije), T4 (rana laktacija) i insulin (zasušenje). Ispitivanjem koncentracije hormona i metabolita u različitim periodima laktacije i zasušenju, zaključujemo da je uticaj momenta uzimanja krvi na vrednost metabolita značajna za sve ispitivane parametre, osim za ukupne proteine, T4 i insulin (Tabela 2, Grafik 1-13). Statistički značajna vrednost F testa i posthock LSD razlike iznose: glukoza (F=25,44; LSD 1:2, 1:3, značajno viša koncentracija glukoze u zasušenom periodu); holesterol (F=15,62, LSD 1:3, 2:3, značajno viša koncentracija holesterola u sredini laktacije); trigliceridi (F=50,42; LSD 1:2, 1:3, značajno viša koncentracija u periodu zasušenja); total lipide (F=27,89; LSD 1:3, 2:3, značajno viša koncentracija u sredini laktacije); NEFA (F=10,74; LSD 1:2, 1:3, značajno viša koncentracija u ranoj laktaciji); BHB (F=24,39; LSD 1:2, 1:3, 2:3, značajno viša koncentracija u ranoj laktaciji); albumini (F=9,2; LSD 1:2, 1:3, značajno viša koncentracija u periodu zasušenja); urea (F=11,61; LSD 1:2, 2:3, snižena koncentracija u ranoj laktaciji); T3 (F=3,31; LSD 1:3, 2:3, niža koncentracija u periodu zasušenja i ranoj laktaciji); growth hormon (F=4,17; LSD 1:2, 2:3, viša koncentracija u ranoj laktaciji). Koncentracija hormona i metabolita kod krava Simentalske rase zavisi od toga da li je krv uzeta u periodu zasušenja, rane laktacije ili sredine laktacije. Razlike u vrednosti metabolita ukazuju na postojanje smanjenog unosa hrane, negativnog energetskog bilansa, sa mobilizacijom lipida i izmenjenim funkcionalnim statusom hepatocita u ranoj laktaciji. Većina ispitivanih parametara pokazuje normalnu distribuciju frekvencije. Izostajanje normalne distribucije za pojedine parametre, posebno u periodu rane laktacije ukazuje na veliku heterogenost krava u procesu adaptacije na peripartalni period.

**Ključne reči:** simentalске krave, endokrini status, metabolički profil, laktacija, zasušenje.

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