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PERIPARTURIENT METABOLIC PROFILE, BLOOD PICTURE AND BODY CONDITION SCORE IN HEALTHY AND COWS WITH ASSOCIATED PERIPARTURIENT DISEASE*

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SUMMARY: Experiment included 39 cows, 15 healthy and 24 diseased. Ketosis was diagnosed in 13 out of 24 diseased cows, and it was combined with mastitis and metritis (in 10 cows) and displaced abomasum (in 3 cows). The combination of diseases consisting of placenta retention and metritis was present in 8 cows, whereas the combination of displaced abomasum and lameness was present in 3 cows. The cows with associated peripartal diseases have significantly higher concentration of NEFA, BHB, bilirubin, AST and the number of neutrophils in the four weeks postpartum. The concentration of glucose, albumin, urea, calcium, cholesterol was lower in the diseased cows. The hematologic profile of the sick cows was characterized by a decreased number of erythrocytes and lymphocytes with a decrease in the concentration of hemoglobin. A total number of lymphocytes in the diseased cows was lower a week after the calving, but subsequently it considerably increased, exceeding the number determined in the healthy cows. The body condition of the diseased cows significantly declines, exceeding 0.7 of measure unit in the first month whereas the decline of body condition of healthy cows is not so noticeable with the amount of 0.4 of measure unit. In the group of sick cows was found significantly higher proportion of cows that had values of metabolic and hematological profile outside the normal values and that had a significant loss of body condition in early lactation. Cows with associated periparturient disease showed worst adaptation capacity in early lactation which is reflected in metabolic profile, blood pictures and loss in body condition score.

Key words: *dairy cows, peripartal diseases, metabolic profile, haemogram, body condition score.*

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INTRODUCTION

Calving period represents the most important period in a production cycle of a cow when cows are moving from the dry period to the lactation period. Due to the increasing milk production, reduced feed intake and the domination of catabolic hormones, energy balance of cows is negative in this stage. Due to the negative energy balance the cows mobilize their own body reserves to meet their demand for energy which results in an increased lipid mobilization and the release of non-esterified fatty acids (NEFA) into the bloodstream as well as the incresed ketogenesis with the increase in the concetration of beta-hydroxybutyrate (BHB) followed bythe loss of physical condition (Ingvarsten, 2006). Apart from the above-mentioned, a lot of other adaptational changes in the organism which largely depend on the intensity of lipid mobilization and ketogenesis occur, such as: reduced glycaemia, decreased concentration of cholesterol, increased concentration of bilirubin, decreased calcaemia and other changes (Cincović et al., 2011); as well as the reduced number of erythrocytes and the concentration of hemoglobin, the reduced number of leucocytes, but the increased number of neutrophils etc (Belić et al., 2011).

The previously mentioned adaptational processes in the cow bodies result in the development of associated peripartal diseases, and the most frequent are the following: ketosis, metritis, mastitis, rennet-bag dislocation and lameness. Pathophysiological connection can be established between all these diseases and metabolic changes due to the negative energy balance (Drackley et al., 2005; Huzzey et al., 2011; Leblanc, 2010; Ospina et al., 2010).

Due to all the previously mentioned information we assumed that there will be differences in parameters values of the metabolic profile and the haemogram in the first few weeks postpartum with both healthy and diseased cows. This paper is aimed at examining the values of the metabolite, the haemogram and physical condition of the healthy cows and the cows suffering from peripartal diseases.

MATERIALS AND METHODS

This experiment included 39 Holstein-Friesian cows: 15 healthy cows, 24 cows suffering from associated peripartal diseases-having more than one peripartal disease in the first four weeks of lactation (subclinical ketosis, placenta retention, metritis, mastritis, abomasal dislocation, lameness). The cows come from a few farms in Vojvodina and were raised under the same conditions of nutrition and care (free stall system, the diet was based on silage that was given to the cattle piecemeal in TMR mixture).

The blood was taken from the jugular vein in the first, second, third and fourth week postpartum and analyzed the same day in the laboratory. The following biological parameters were examined: NEFA, BHB, glucose, cholesterol, bilirubin, AST, albumin, urea, and Ca. Standard types of kits were used manufactured by Randox (UK), and spectrophotometric measurements were performed on the semi-auto chemistry analyzer Rayto RT 1904c. In addition to all of the previously mentioned, the following haematologic parameters were recorded: the number of erythrocytes, the concentration of hemoglobin, the number of leucocytes, and differential white blood cells line. A hematologic analyzer Hemavet 950c was used.

The body condition score (BCS) was determined according to the 1-5 scoring

system on the recommendation of Elanco Animal Health.

The difference in the values of the metabolic profile, haemogram, and physical condition between the group of healthy cows and the group of cows suffering from various diseases during the four weeks of monitoring was performed using the ANOVA method and LSD test.

RESULTS

Peripartal diseases in dairy cows most frequently occur in combination with ketosis regardless of whether it is subclinical or clinical form. (Fig. 1). Ketosis was diagnosed in 13 out of 24 diseased cows ketosis was diagnosed, which was combined with mastitis and mestritis (in 10 cows) and displaced abomasum (in 3 cows). The combination of diseases consisting of placenta retention and metritis was present in 8 cows, whereas the combination of abomasum dislocation and lameness was present in 3 cows.

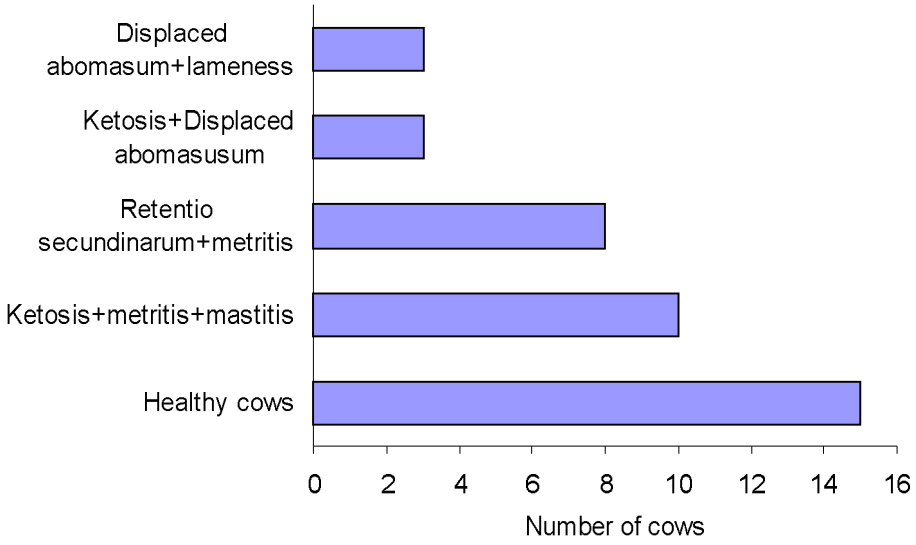

Fig. 1. Frequency distribution of the cows according to the diseases that were diagnosed

The concentration of NEFA I BHB was significantly higher (<0.001) in the diseased cows in all the weeks postpartum (Fig. 2 and 3), but for NEFA the difference disappears in the fourth week postpartum. The week had a significant effect as well (<0.01). The concentration of NEFA and BHB was decreasing in the groups of both healthy and sick cows, but this decline was more intensive in the healthy cows:-0.17:-0.09 mmol/l per week regarding NEFA, , -0.21:-0.17 mmol/l regarding BHB.

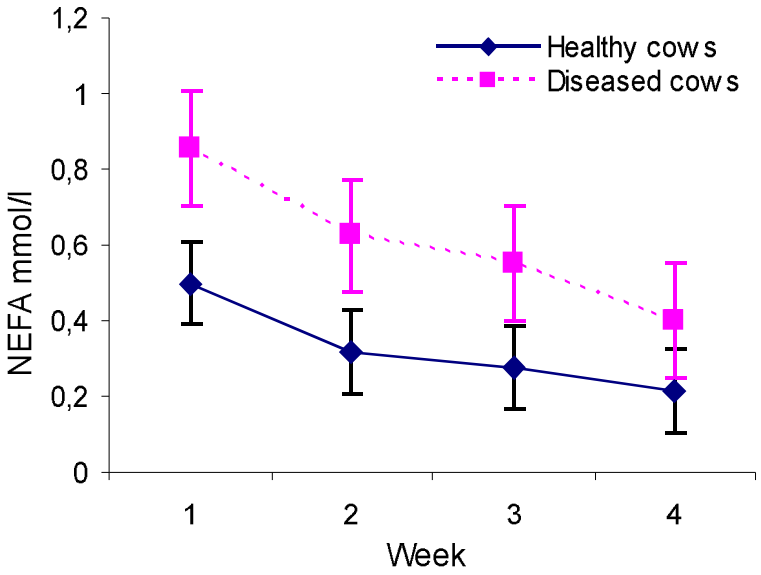

Fig. 2. The Concentration of NEFA in healthy and sick cows

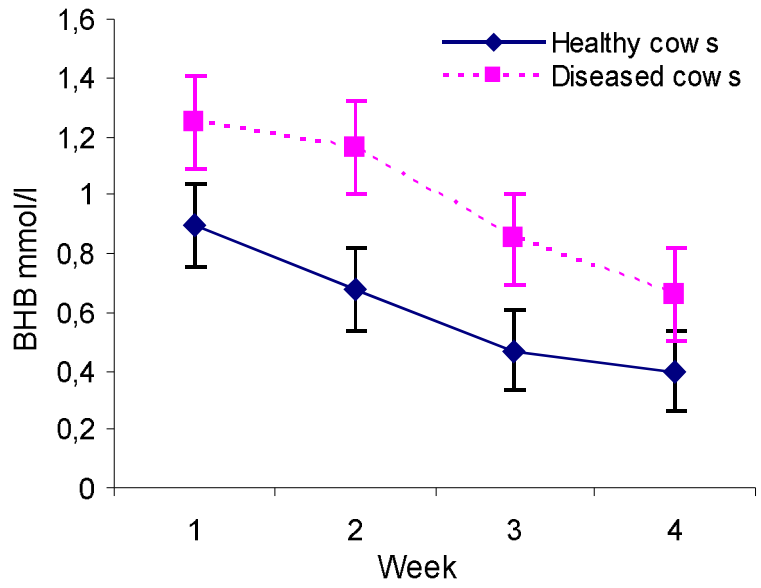

Fig. 3. The Concentration of BHB in healthy and sick cows

The concentration of glucose (Fig. 4) was significantly lower in the diseased cows (<0.001), and the cows from the same group had significantly lower concentration of cholesterol (<0.05) (Fig. 5). The effect that the time had on the development of the values of examined parameters was significant. The change dynamics of the concentration of glucose during the four weeks was approximately identical in both healthy and diseased cows +0.35 mmol/l per week, and the change dynamics of the cholesterol concentration +0.69 mmol/l per week was also identical.

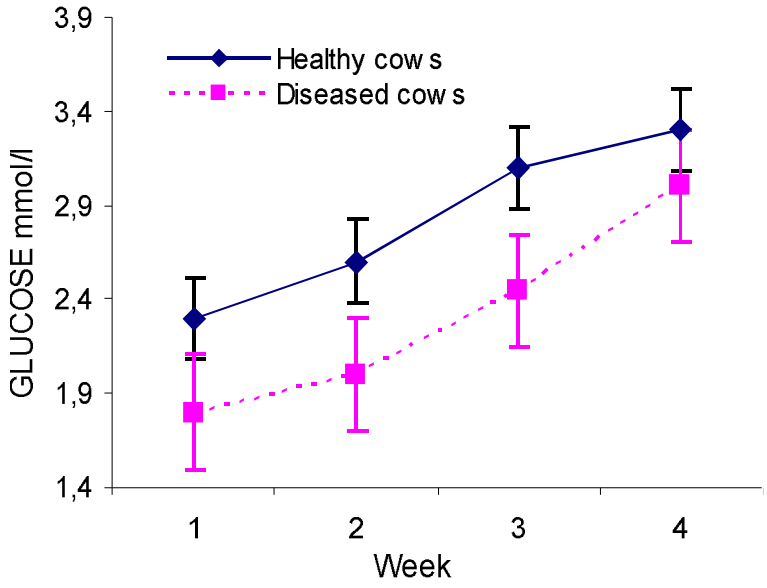

Fig. 4. The concetration of glucose in diseased and healty cows

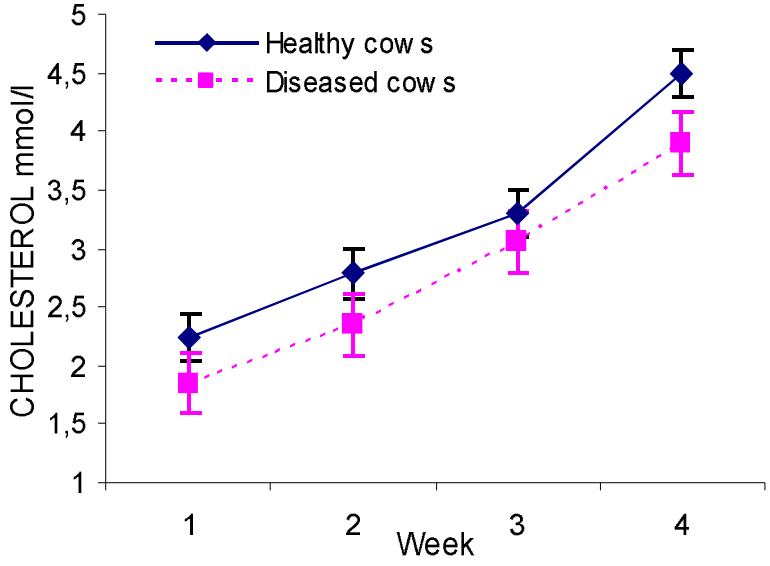

Fig. 5. The concetration of cholesterol in healthy and diseased cows

The concetration of albumin (Fig. 6) in healthy cows significantly increases during the first four weeks pospartum (<0.05), but until the third week the concetration disappeared and then in the fourth week it became significant again, because the albumin concetration in diseased cows declined. The concetration of urea (Fig. 7) was significantly lower in diseased cows in the first week postpartum (<0.01) followed by a gradual decline of that difference. The concentration of urea increased significantly in the first four weeks postpartum (<0.01), and this increase was more intensive in sick cows: +0.6:+0.4 mmol/l per week.

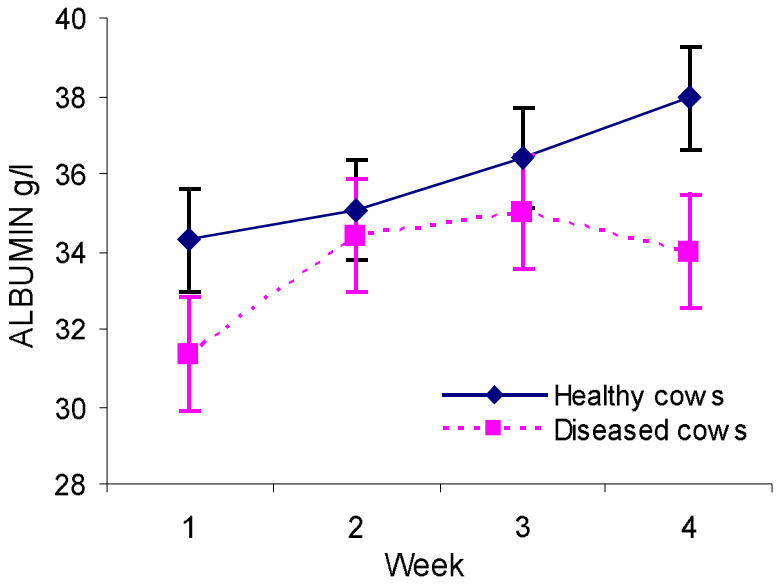

Fig. 6. The albumin concetration in healthy and diseased cows

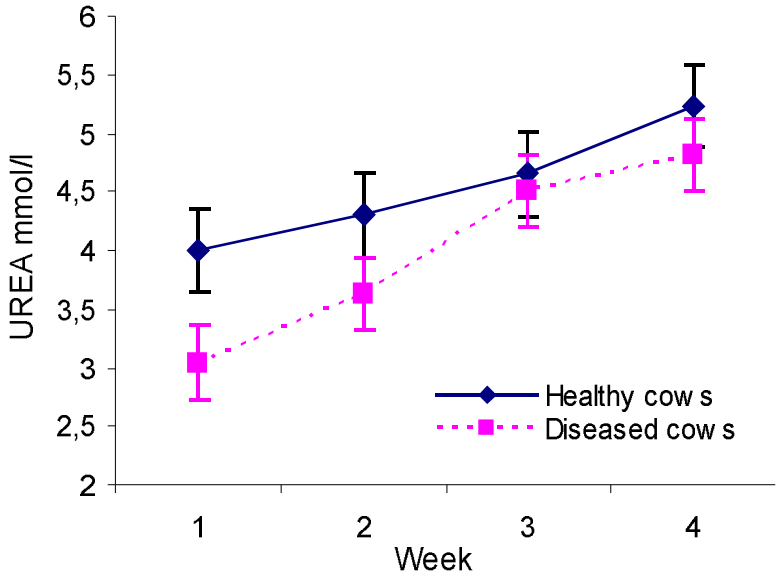

Fig. 7. The urea concetration in healthy and diseased cows

The bilirubin concetration (Fig. 8) was significantly higher in diseased cows during the whole four weeks postpartum (<0.01). Bilrubin concentration was declining significantly during the first four weeks postpartum (<0.01) in both of the groups of cows. The effect of the week on the concentration of AST was statistically insignificant, due to the high variation of values (Fig. 9). The diseased cows showed a significantly higher concentration of this enzyme in the first, second, third and fourth week postpartum.

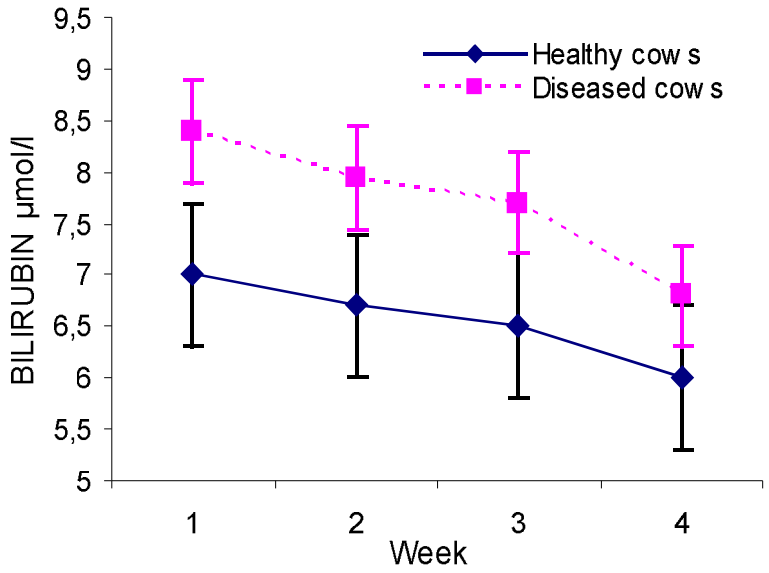

Fig. 8. Bilirubin concetration in healthy and diseased cows

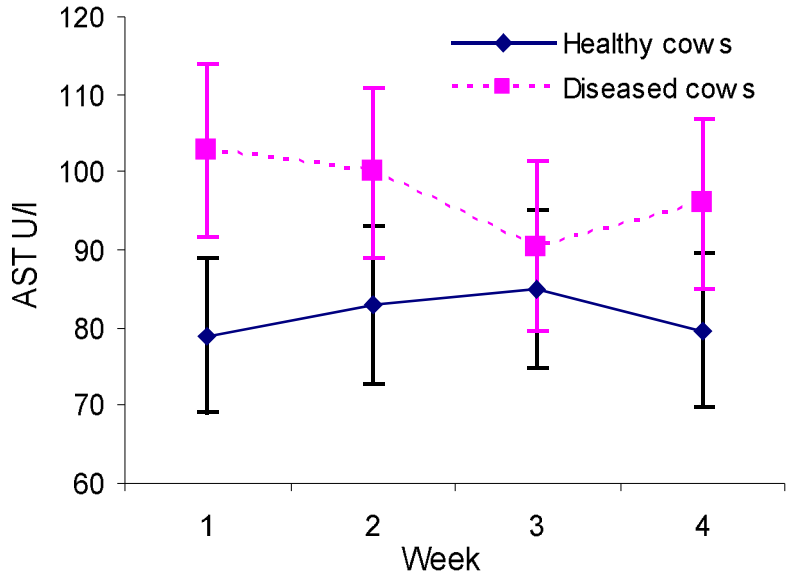

Fig. 9. AST concetration in healthy and diseased cows

Calcium concetration (Fig. 10) was significantly lower in diseased cows (<0.01) in the first and the third week postpartum. The effect of the week was not statistically significant, but the concentration of calcium showed a tendency to change under the influence of the week (<0.1). There was a constant and steady increase in calcaemia in healthy cows, whereas calcaemia in diseased cows increased and declined.

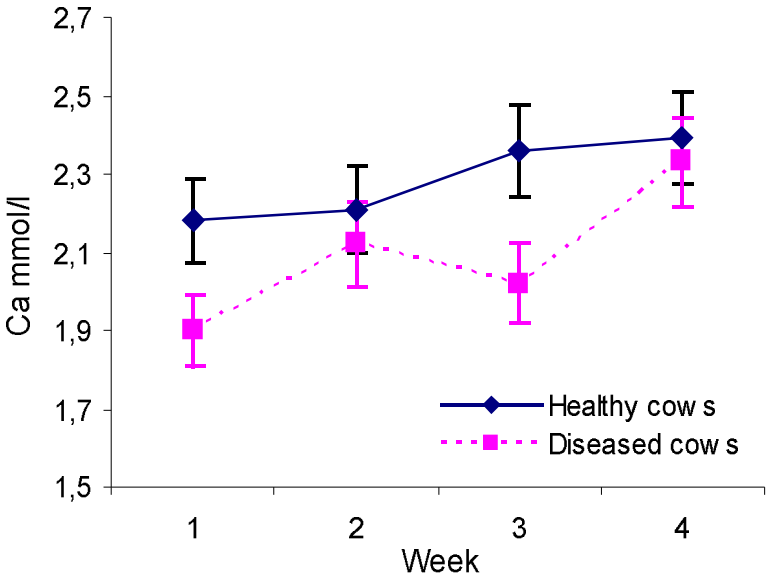

Fig. 10. Calcium concetration in healthy and diseased cows

The diseased cows in the first four weeks postpartum show a tendency towards anemia which is reflected in a notably lower number of erythrocytes and hemoglobin in this group of cows (Fig. 11 and 12). The difference in value of the selected parameters of the red blood cells line is the most noticeable in the first week of partus between healthy and diseased cows but it was observed that this difference gradually declined until the fourth week. The effect of the week on the number of erythrocytes was insignificant, whereas the week during which the blood was sampled had a significant impact on hemoglobin concentration.

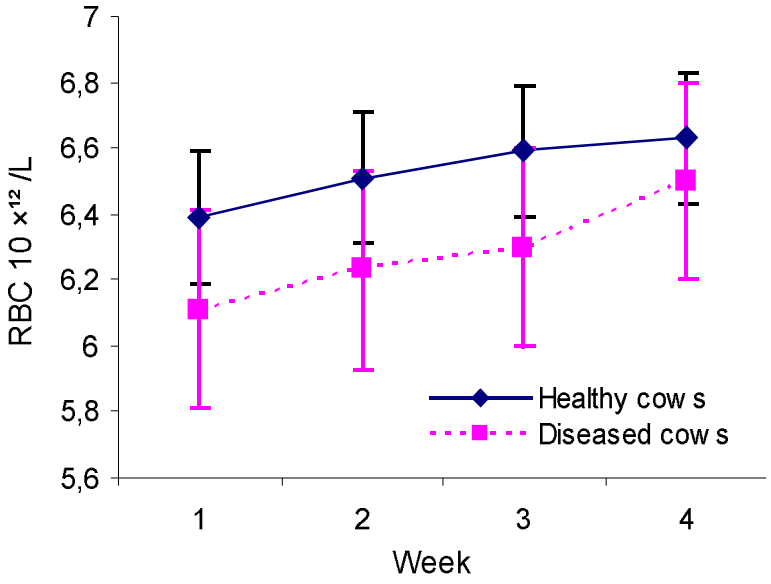

Fig. 11. The number of erythrocytes in healthy and diseased cows

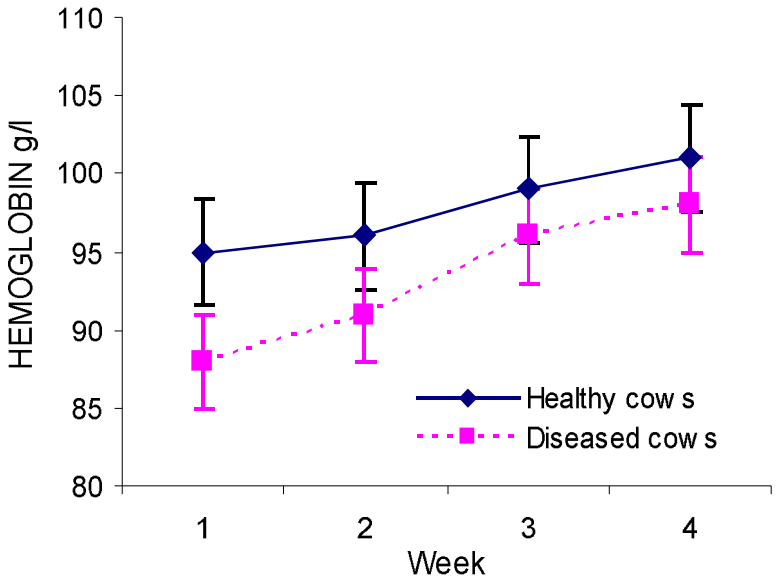

Fig. 12. Hemoglobin concetration in healthy and diseased cows

A total number of leucocytes (Fig. 13) was notably smaller in diseased cows in the first week postpartum. In the second, third and the fourth week an inversion occurs leading to the significant increase in the number of leucocytes in diseased cows. The number of neutrophils is notably higher, whilst the number of lymphocytes is significantly smaller in diseased cows (Fig. 14 and 15). The number of lymphocytes in diseased cows showed the tendency to be higher (<0.1) in cows in the fourth week postpartum.

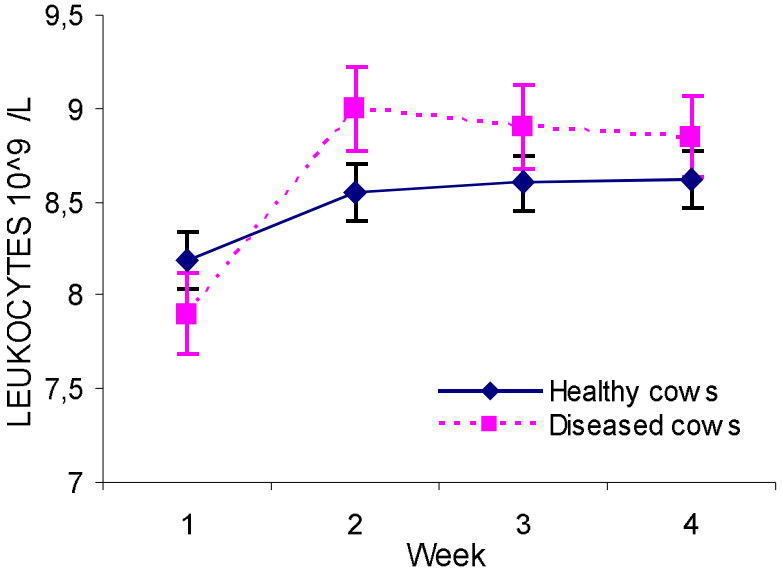

Fig. 13: The number of leukocytes in healthy and diseased cows

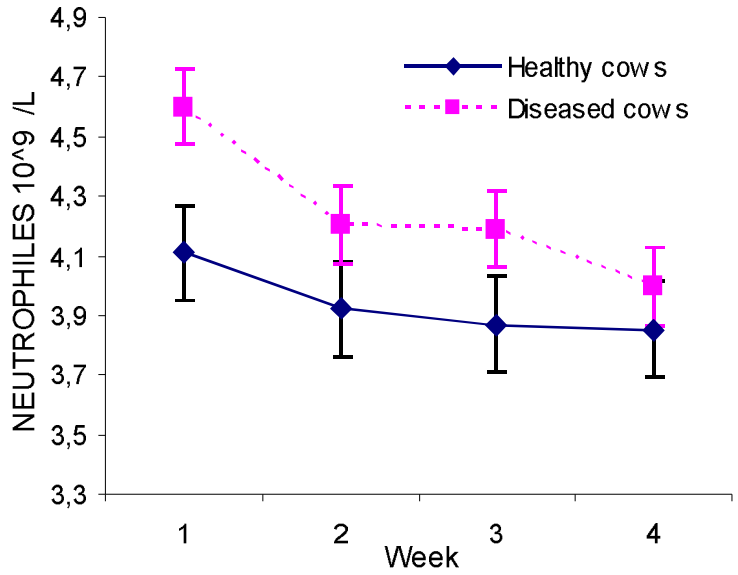

Fig. 14. The number of neutrophils in healthy and diseased cows

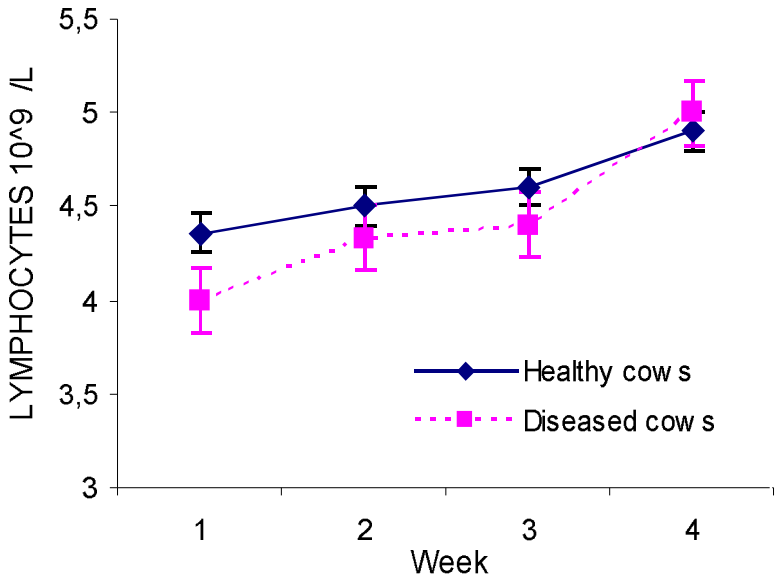

Fig. 15. The number of lymphocytes in healthy and diseased cows

The diseased cows showed significant loss of body condition (Fig. 16) in the first four weeks postpartum (0.7 of measure unit with diseased cows and 0.4 of measure unit with healthy cows) and so the body condition scoring of the cows is significantly lower in the second, third and fourth week postpartum (<0.01).

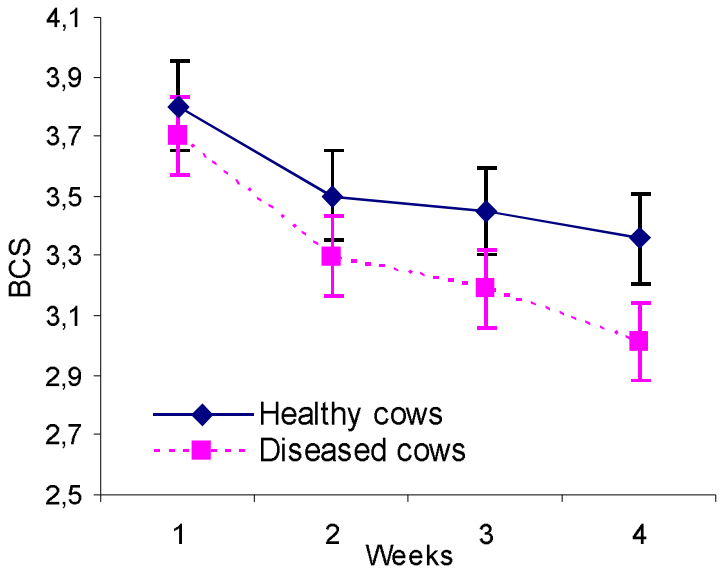

Fig. 16. Body condition of healthy and diseased cows

In the group of sick cows was found significantly higher proportion of cows that had values of metabolic and hematological profile outside the normal values and that had a significant loss of body condition in early lactation. Elevated serum NEFA and BHB, a significant loss of body condition and stress leukogram were the most common signs in diseased cows, while hypoglycemia, hyperbilirubinemia, hypocalcemia and anemia was lower. A certain number of cows in a healthy group had a parameters value outside the reference, but their presence was lower than in a group of sick cows. (See Table 1)

Table 1: Percent of cows with parameters out of reference range in healthy and diseased group during first week after calving

Parametar	Diseased (%)	Healthy (%)	p
NEFA	62,5	26,7	<0,01
BHB	54,2	20,0	<0,01
Glucose	45,83	20,0	<0,01
Bilirubine	45,83	20,0	<0,01
Ca	41,67	13,33	<0,05
RBC	37,5	20,0	<0,05
Hemoglobine	37,5	13,33	<0,05
N:L >1	62,5	26,7	<0,01
BCS loss >0,5	83,3	13,33	<0,01

DISCUSSION

The experiment included 24 diseased cows and 15 healthy cows. There were symptoms of ketosis combined with mastitis, metritis and displaced abomasum. It is well-known that a high concentration of NEFA during the period before calving and a

high concentration of BHB (subclinical ketosis) during the period after calving increases the chances of developing abomasum dislocation (Cameron et al., 1998; Geishauser et al., 2000; LeBlanc et al., 2005). The increased concentration of NEFA increases the risk of occurrence of placenta retention (LeBlanc et al., 2004). Subclinical ketosis increases duration and seriousness of the disease (Sordillo et al., 2009). These results are in agreement with our results showing that the diseased cows have a significantly higher concentration of NEFA and BHB in the first four weeks after calving. The average NEFA concentration in diseased cows was 0.9 mmol/l and BHB concentration was 1.3 mmol/l in the first week postpartum. These results match the previously obtained marginal values. Prepartum values of NEFA indicating the risk of the occurrence of peripartal diseases range from 0.2 to 0.5 mml/l and regarding postpartum period the values range from 0.57 to 1 mmol/l (Oetzel, 2004; Ospina et al., 2010 Chapinal et al., 2011; Cincović and Belić, 2012) which is in accordance with the results we obtained. The same authors discovered that the concentration of BHB is important for the development of peripartal diseases if it exceeds 1.5 mmol/l, which is higher than the values obtained in this paper. Metabolic and hematologic findings from cows during the peripartal period depend on the intensity of lipid mobilization (González et al., 2011). During the peripartal period, due to the liver strain caused by ketones, various metabolic adaptations occur such as decreased glycemia, decreased albumin production in the liver, decreased cholesterol concentration, increased concentration of AST and bilirubin, and decreased urea concentration (Cincović et al., 2011; Đoković et al., 2009). This kind of situation is characteristic of the cows that are prone to fatty infiltration of the liver and ketosis. A significantly higher concentration of NEFA, BHB and AST was found along with decreased concentration of calcium and glucose which is in agreement with our results (Vanwinden et al., 2003). NEFA concentration in combination with leukocyte profile is important model to predict uterus and mammary gland health (Belić et al., 2012). One big research which included more than one thousand cows indicated that there is a significant variation in relation to the mean value of albumin, glucose, cholesterol and calcium in the cattle with a high incidence of peripartal diseases (Kida, 2002).

An increase in the number of leucocytes in cows suffering from metritis happens on account of the increase in the number of neutrophils. The increase in the number of neutrophils can be explained as the consequence of inflammation development, but this also occurs as a reaction to the increased concentration of cortisols during the labor. It is important to mention that the functionality of neutrophils declines during the peripartal metabolic stress which predisposes cows to metritis (Hammon et al., 2006). The reduction in the number of erythrocytes and the reduced concentration of hemoglobin which dominates in the cows prone to metritis can be explained as a consequence of bleeding from the inflamed injuries of the uterus. Haemogram of the cows with placenta retention which precedes metritis indicates leucocytosis and anemia (Ahmend et al., 2009), which was confirmed by our obtained results. The leukogram was obtained along with monocytosis by examining haemogram at the moment of diagnosing endoemetritis (Hanafi et al, 2008) which means that peripartal findings might be useful in estimating the occurrence of metritis. The increase in the number of leucocytes during the peripartal period in dairy cows suffering from mastitis is in accordance with the previously obtained results (Barnouin and Chassagne, 2000; Holtenius et al., 2004). The cows with lameness have significantly altered hematological parameters in comparison with the observation of healthy cows which is reflected in the decreased number of

erythrocytes and the reduced hemoglobin concentration while the total number of leukocytes and neutrophils was significantly higher (Meimandi Parizi and Khalafizadeh, 2006), which matches the results we obtained. A decreased number of leukocytes in diseased cows in the first week postpartum is an interesting finding which is considered to be a consequence of a significant effect of fatty acids, ketone and oxidative stress on immunological stress (Sordillo et al., 2009). Increased ration between neutrophils and lymphocytes occurs when the animals are under stress primarily due to the effect of the cortisols (Bertoni et al., 2003).

The body condition of the diseased cows significantly declines, exceeding 0.7 of measure unit in the first month whereas the decline of body condition of healthy cows is not so noticeable with the amount of 0.4 of measure unit. This change can be linked with higher catabolism which puts a strain on the diseased cows. The previous results indicated that cows losing from 0.5 to 1.0 of measure unit BCS (body condition score) showed that they carry a higher risk of the occurrence of metritis (Butler and Smith, 1989).

In the group of sick cows was found significantly higher proportion of cows that had values of metabolic and hematological profile outside the normal values and that had a significant loss of body condition in early lactation. Ospina et al. (2010a) were found that in herd in which more than 15% of animals sampled had prepartum NEFA concentration $\geq$0.30 mEq/L was 75%, BHBA $\geq$12 mg/dL was 40%, and postpartum NEFA $\geq$0.70 mEq/L was 65%. These results require further research on the impact of the metabolic profile in an independent sample of cows.

High values of NEFA and BHB can lead to significant changes in the metabolic profile and blood picture in healthy cows during early lactation, and many parameters can be outside of reference (Cincović et al., 2012). Therefore the monitoring of metabolic profile and blood test must be done in parallel with the clinical observation of cows in early lactation.

CONCLUSION

The cows suffering from peripartal associated diseases show significant changes in metabolic and hematologic profile in the first four weeks postpartum. During this period the body condition of the diseased cows shows more intensive decline. In group of diseased cows exists higher percent of cows with metabolic and hematological parameters which were out of reference for cattle. Cows with associated periparturient disease showed worst adaptation in early lactation which is reflected in metabolic profile, blood pictures and loss in body condition score.

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PERIPARTALNI METABOLIČKI PROFIL, KRVNA SLIKA I TELESNA KONDICIJA ZDRAVIH I KRAVA SA ZDRUŽENIM PERIPARTALNIM BOLESTIMA

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Izvod

Cilj ovog rada je bio da se ispitaju vrednosti metaboličkog profila, krvne slike i telesne kondicije u prve četiri postpartalne nedelje kod zdravih krava i krava sa različitim peripartalnim bolestima. U ogled je uključeno 39 krava: 15 zdravih i 24 obolelih krava. Ketoza je nađena kod 13 krava u kombinaciji sa mastitisom/metritisom (10 krava) i dislokacijom abomasuma (3 krave). Retencija palcente sa metritisom postojala je kod 8 krava,a kod 3 krave je nađena kombinacija dislokacije sirišta i jake šepavosti. Kod 13 od 24 obolelih krava dijagnostikovana je ketoza, koja je bila u kombinaciji sa mastitisom i metritisom (kod 10 krava) i sa dislokacijom abomasuma (kod 3 krave). Krave sa združenim peripartalnim bolestima imaju značajno višu koncentraciju NEFA, BHB, bilirubina, AST i broj neutrofila u prve četiri postpartalne nedelje. Koncentracija glukoze, albumina, uree, kalcijuma i holesterola je bila niža kod bolesnih krava. Hematološki profil obolelih krava odlikovao se nižim brojem eritrocita i limfocita uz sniženu koncentraciju hemoglobina. Ukupan broj leukocita kod bolesnih krava bio je niži u prvoj nedelji posle partusa, da bi potom značajno porastao iznad broja utvrđenog kod zdravih krava. Telesna kondicija obolelih krava pokazuje intenzivniji pad (0.7:0.4 jedinice) u prve četiri postpartalne nedelje. U grupi bolesnih krava nađena je značajno veća proporcija krava čije se vrednosti metaboličkog profila i krvne slike bile izvan referenrnih, a veći je bio i gubitak telesje kondicije. Krave sa združenim peripartalnim bolestima pokazuju lošiji adaptacioni kapacitet što je pokazano kroz vrednosti metaboličkog profila, krvne slike i telesne kondicije ispitivanih krava.

Ključne reči: mlečne krave, peripartalne bolesti, metabolički profil, krvna slika, telesna kondicija.

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