EFFECTS OF METABOLASE® ON SEVERAL BLOOD INDICES, AND PRODUCTIVITY IN FRESH DAIRY COWS

Ramūnas Antanaitis, Mindaugas Televičius, Robertas Stoškus

Department of Non-Infectious Diseases of the Veterinary Academy of the Lithuanian University of Health Sciences Tilžės 18, LT-47181 Kaunas, phone +370 336 34 53; e-mail: antanaitis@lva.lt

Abstract. The aim of this work was to assess the possible effects of the medicinal product "Metabolase®" on cows health by several biochemical blood indices and productivity in fresh dairy cows.

Depending on the cow's age, breed, and productivity 30 fresh dairy cows (7 (±1)) days on the average after calving) were selected for the research. During the research, the cows were divided into two groups: experimental group (n=15) and control group (n=15). The cows in the test group were injected 500 ml of solution METABOLASE® into vena jugularis on the 1st and 7th day after calving. During the first stage, blood samples were taken before injecting the solutions. During the second stage, blood samples were taken a week later, before injecting the solution the second time. During the third stage, blood samples were taken a week after the second injection of the solution. During the fourth stage, blood samples were taken 30 days after the last injection. Milk samples for determining milk composition were taken at the same intervals.

The effects of the medicinal product Metabolase® were as folows: reduced beta-hydroxybutyrate (BHB) concentration and milk fat to protein ratio; increased amounts of glucose in blood; increased milk production and thus reduced risk of ketosis; increased albumin concentrations, and reduced aspartate aminotransferase (AST) concentration, which signals a better function of liver.

Keywords: cow, postpartum diseases, blood indices, productivity

Introduction. It is well known that severe negative energy balance in the periparturient period increases the risk for postpartum diseases such as retained placenta, milk fever, metritis, mastitis, clinical ketosis, and displaced abomasum (LeBlanc, 2010).

The productivity of high-yielding dairy cows has increased so that milk yield has doubled over the past 40 years. This has increased cow susceptibility to metabolic diseases, mainly related to ketosis, fatty liver, and hypocalcemia (Oltenacu and Broom, 2010). Ketosis and fatty liver are closely linked and are responsible for severe economic losses in dairy farms due to drop in milk yield and increase in culling rates. Clinical ketosis in dairy cows usually occurs between the second and seventh week of lactation (Duffield et al., 1997).

The resolution of post-partum uterine infection and inflammation has been identified as one of the most important events for successful establishment of pregnancy in dairy cattle (Walsh et al., 2011).

Several epidemiological studies have demonstrated that these diseases cause major financial losses as a result of the costs related to diagnosis and treatment of diseases, reduced milk yield, and decreased reproductive performance in dairy herds. Approximately 75% of diseases in dairy cattle occur during the first month postpartum (LeBlanc, 2010), and 50% of dairy cattle suffer from metabolic and infectious diseases during the transition period (LeBlanc, 2010). The severity and duration of NEB are reflected by increase in circulating NEFA and BHBA and the degree of decrease in glucose concentrations (Drackley, 1999). The decreased DMI prepartum causes NEB and increases NEFA and BHB concentrations (Suriyasathaporn et al., 1999; LeBlanc, 2010; Gumen et al., 2011).

The indices for blood enzymes and minerals are used for diagnosing reproductive diseases, metabolic diseases, and ketosis in cattle, and are important for analyzing the reasons for reduced milk yield and calculating feeding errors (Yameogo et al., 2008). Milk production could be an additional diagnostic index for cow's postpartum condition. It has been noticed that in the case of ketosis, milk yield reduces by approximately 3 kg per day during lactation period (Fourichon et al., 1999)

The aim of this work was to assess the possible effects of the medicinal product "Metabolase®"* on cows health by several biochemical blood indices, productivity and composition of milk in fresh dairy cows.

* Metabolase® constituents: l-Carnitine hydrochloride (equivalent to l-Carnitine - 500.0 mg) - 613.3 mg; Thioctic acid - 20.0 mg; Pyridoxine hydrochloride - 15.0 mg; Cyanocobalamin - 3.0 mg; D,L-acetylmethionine - 2.0 g; l-Arginine - 240.0 mg; l-Ornithine hydrochloride (equivalent to l-Ornithine - 120.0 mg) - 153.2 mg; l-Citrulline - 120.0 mg; l-Lysine hydrochloride (equivalent to l-Lysine - 50.0 mg) - 62.5 mg; Glycine - 150.0 mg; Aspartic acid - 150.0 mg; Glutamic acid - 150.0 mg; Fructose - 5.0 g; Sorbitol - 8.0 g; Excipients up to 100.0 ml

Material and methods

The research was conducted in one of the Lithuanian dairy cattle farms during the period from 01.02.2014 to 01.04.2014. The average milk yield amounted to 7000 kg of milk per year. Cows were kept in cold loose housing barns. Each group of cows (depending on lactation day and productivity) was fed individual balanced diet.

Depending on the cow's age, breed, and productivity, 30 fresh dairy cows (7 (±1)) days on the average after calving) were selected for the research. The research was conducted following the general principles of clinical research. Only clinically healthy cows were included into

the research. During the research, the cows were divided into two groups: experimental group (n=15) and control group (n=15). The cows in the experimental group were injected 500 ml of solution METABOLASE® into vena jugularis on the 1st and 7th day after calving. At the same time, and at the same frequency and amounts, the cows from the control group were injected 0.9 % sodium chloride solution into vena jungularis. Solutions to be injected were heated up to the cow's body temperature, and then injected slowly. The withdrawal period for milk and meat of the used solutions was 0 days.

Blood samples for measuring biochemical indices were taken from the coccygeal vessels in four stages. During the first stage, blood samples were taken before injecting the solutions. During the second stage, blood samples were taken a week later, before injecting the solution the second time. During the third stage, blood samples were taken a week after injecting the solutions the second time. During the fourth stage, blood samples were taken 30 days after the last injections. Blood samples were taken at 10:00 a.m. before feeding.

Blood samples were collected into vacuum test tubes (BD Vacutiner, England). The samples were delivered for examination to the Laboratory of Clinical Tests of the Large Animal Clinic of the Veterinary Academy of the Lithuanian University of Health Sciences, and centrifuged for 5 minutes at the speed of 3000 rpm. The obtained blood serum was examined using the analyzer Hitachi 705 (Hitachi, Japan), reagents DiaSys (Diagnostic Systems GmbH, Germany) determining the concentrations of beta-hydroxybutyrate (BHB), albumins (ALB), aspartate aminotransferase (AST), and glucose (GLU) in blood serum.

During all the stages of research, the average daily milk yields (kg/day) of cows were recorded with the help of herd management software Dairy Plan C21. Milk samples for determining milk composition were taken at the same intervals. The collected milk samples were examined for fat content (R%) and milk protein content. The research was conducted following the provisions of the Law of the Republic of Lithuania No. 11-2271 on Protection, Keeping and Use of Animals, dated 03/10/2012 (*Valstybės žinios* (Official Gazette) No. 122-6126 dated 20/10/2012). and of the by-laws, Education and training purposes of animals used in storage, maintenance and conditions of use No. B1-866, dated 31/10/2012 *(Valstybės žinios* (Official Gazette) No. 130-6595 dated 10/11/2012).

The test data were processed using the SPSS statistical package (SPSS for Windows 15.0, SPSS Inc.,Chicago, IL, USA, 2006). The data were considered reliable from the statistical point of view when $P<0.05$.

Results and discussion

BHB concentrations before injecting the solution in blood serums of cows from both groups differed insignificantly: the average BHB concentration was 0.9 (±0.1) mmol/l. in the experimental group, and 0.8 (±0.05) mmol./l in control group. A week after injecting the medicinal product under investigation, the average BHB concentration reduced to 0.5 (±0.05) mmol/l. in the test group of cows, and increased to 1.1 (±0.1) mmol/l. ($P<0.05$) in the control group. A week later, the average BHB concentration reached 0.5 (±0.05) mmol/l in the experimental group, and 0.9 (±0.05) mmol/l ($P<0.05$) in control group. A month later after the last injection of the medicinal product, under consideration no statistical differences were observed in the investigated parameters. The average BHB concentration was 0.7 (±0.07) mmol/l in the test group, and 0.8 (±0.07) mmol/l in the control group (Fig. 1).

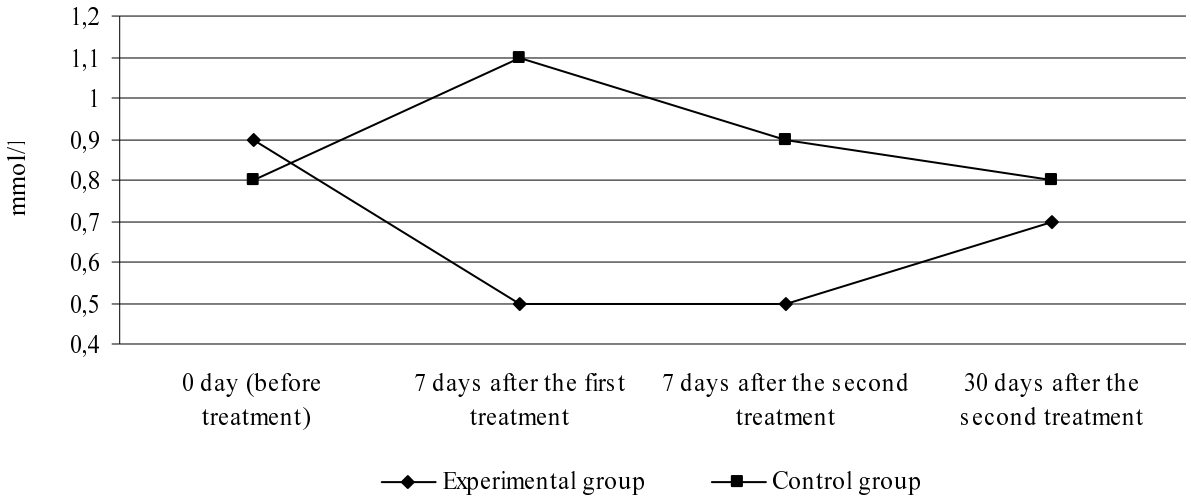

Fig. 1. **BHB concentration in blood serum of cows from test and control groups**

The severity and duration of NEB is reflected by the increase in circulating NEFA and BHB and the degree of decrease in glucose concentrations (Drackley, 1999). The decreased dry matter intake (DMI) prepartum causes negative energy balance (NEB) and increases non-esterified fatty acids (NEFA) and BHBA concentrations

(LeBlanc, 2010; Gumen et al., 2011). The case definition of subclinical ketosis is characterized by serum BHBA levels >1.0 to 1.4 mmol/L in the absence of clinical signs of ketosis (Iwersen et al., 2009; Rollin et al., 2010). Ketosis may be diagnosed based on specific changes in blood serum: increase in non-esterified fatty acids, hydroxybutyric acid (BHB), or acetone concentration (Holtenius et al., 2004; Sakha et al. 2007). The highest efficiency level of the medicinal product on subclinical ketosis was observed a week after the first injection, and after the second injection.

Albumin concentration was statistically reliably ($P<0.05$) higher in blood serum of the experimental group cows one week after the first injection. During this period, the average albumin concentration was 36 (±2) g/l in the experimental group, and 32.5 (±1.85) g/l in the control group. A statistically reliable difference ($P<0.05$) remained throughout the entire period of the research (Fig. 2). Gonzalez et al (2011) have stated that in ketotic cows, albumin levels may decrease related to inadequate synthesis of albumin which results from the fatty acids cumulating in hepatocele hindering the liver from functioning. Cumulation occurs as a result of hepatic changes.

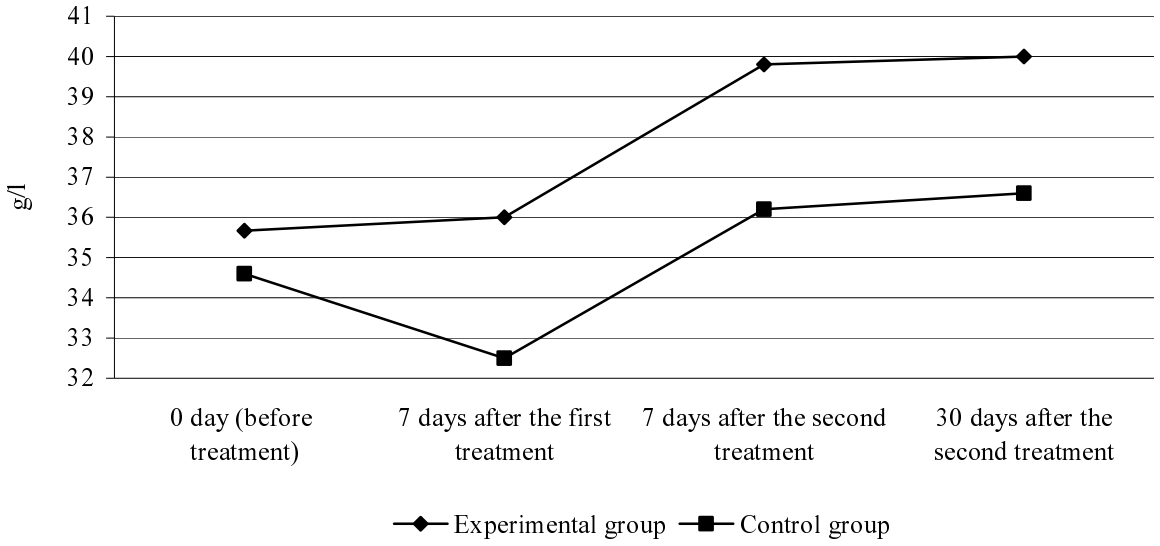

Fig. 2. **Albumin concentrations in blood serum of cows from test and control groups**

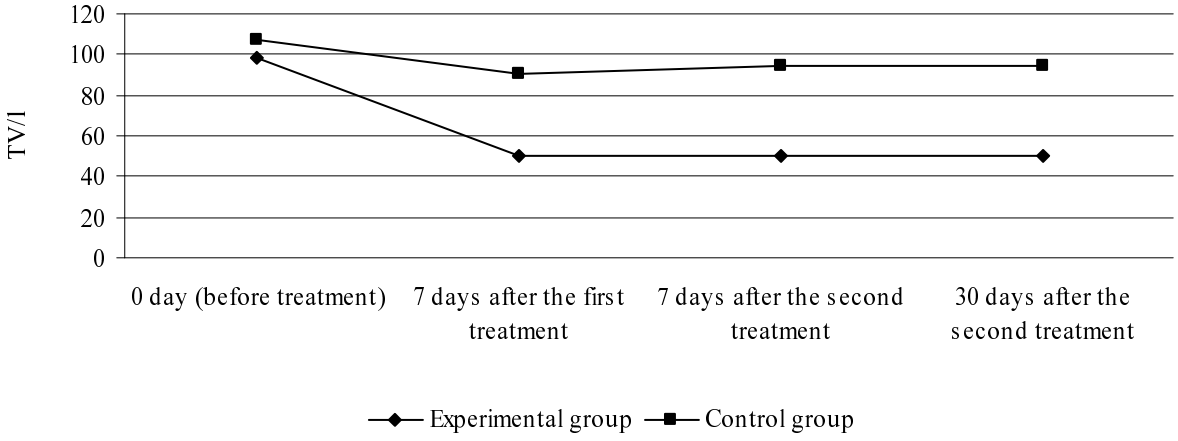

Fig. 3. **AST concentration in blood serum of cows from test and control groups**

Aspartate aminotransferase concentrations before injecting the solution in blood serums of cows from both groups differed insignificantly, however a week after the first injection, the difference between experimental and control groups was statistically reliable ($P<0.05$) (Fig. 3). When fat infiltrates the liver, a lesion appears in the hepatic tissues and the levels of enzymes that indicate liver injury (AST, gamma–glutamyl transferase (GGT), and glutamate dehydrogenase (GLDH)) are generally augmented (Bobe et al., 2004). AST values are more

sensitive than GGT as indicators of hepatic lipidosis in early lactation cows (Gonzalez et al., 2011). According to the data presented by Žymantienė and other researchers (2010), AST activity in healthy cows may vary depending on lactation period. Žilaitis et al (2008) claimed that AST activity in ketotic cows can reach 109–134 TV/L. Based on the data presented by Sutkevičius and Černauskas (2003), the amounts of blood enzymes alanine aminotransferase (ALT), AST, alkaline phosphatase (ALP) and the alkali reserve determine the functional capacity of liver and hepatocellular damage in case of ketosis or other metabolic diseases.

Glucose concentrations at the beginning of the research were similar in blood serums of both experimental and control groups (Fig. 4). A statistically reliable ($P<0.05$) difference in glucose concentrations was observed a week after the second injection. Serious decreases in serum glucose levels during the early stages of lactation are reported in dairy cows with SCK (Sakha et al 2006, Gonzalez et al 2011).

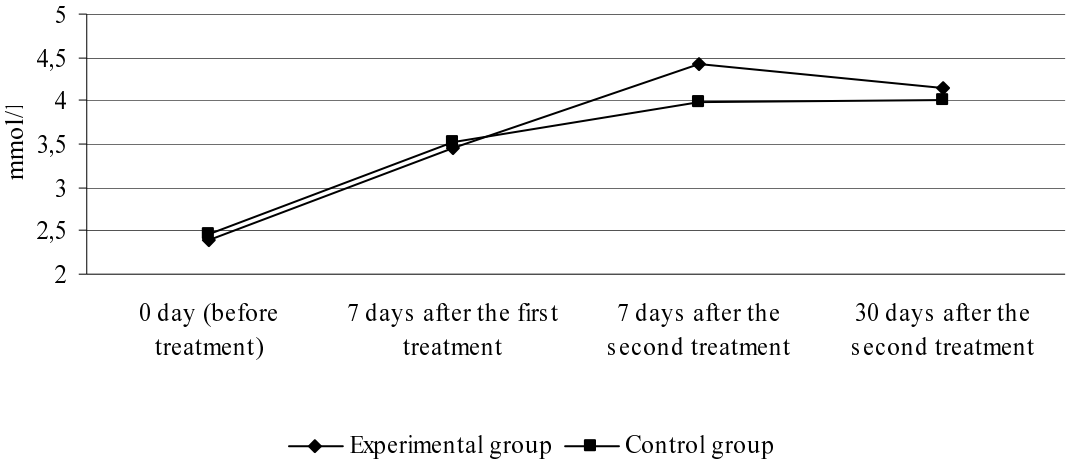

Fig. 4. **Glucose concentration in blood serum of cows from the test and control groups**

A week after the first treatment, a statistically reliable difference ($P<0.05$) was observed in milk yields from experimental and control groups. During this stage of research, the average milk yield was 22.1 (±1,2) kg/day in the experimental group, and 19 (±1.1) kg/day in the control group. A month after the second treatment, the average milk yield was 26.36 (±1.4) kg/d in the experimental group, and 23.5 (±1.2) kg/d in the control group (Fig. 5). Edwards and Tozer (2004) have determined that milk yield reduces statistically reliably 5 days before clinical symptoms of displaced abomasum. Klimienė et al (2005) claims that when cows suffer from mastitis, not only milk quality deteriorates but also its amount is reduced.

Milk production declines 2–4 weeks before diagnosis of ketosis, therefore this indicator may be a valid argument in assessing the health of cows (Rajala-Schultz et al., 1999).

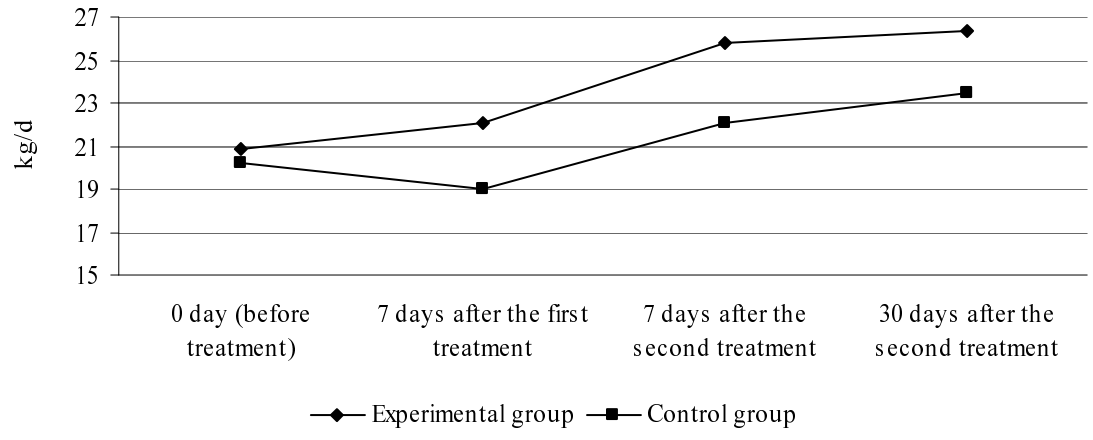

Fig. 5. **Average milk yields from test and control groups in the course of the research**

A week after the first treatment, a statistically reliable difference ($P<0.05$) in milk fat to protein ratio was observed between the experimental and control groups (Fig. 6). During this stage of the research, the average milk fat to protein ratio was 1.41 (±0.08) in the experimental group, and 1.47 (±0.05) in the control group. The difference remained throughout the entire period of the research (Fig. 6). Milk fat to protein ratio is very important for diagnosing certain early-lactation health disorders. According to Heuer et al (1999), cows with milk fat to protein ratio > 1.5 demonstrate higher risk of ketosis, displaced abomasums, and reproductive diseases. Cows with milk fat to protein ratio > 1.5 produce more milk, however their service period is shorter (Heuer et al., 1999). Cows with milk fat to protein ratio <1 have a higher probability of developing lactic acidosis, laminitis. Variation of this milk indicator depends on diet, and the amounts of proteins, carbohydrates and fiber content in the diet.

It is argued that ketotic cows produce milk with higher fat and protein content (Duffield et al., 1997). If cows mobilize energy reserves more intensely, the risk of inflammation in udder increases. The milk produced by such cows demonstrate a lower concentration of lactose, proteins and urea (Rezamand et al., 2007). The data on milk yields, protein and fat content may be used for calculating cow's energy balance (Friggens et al., 2007). Milk composition is affected by HB concentration, and ketosis prophylaxis can affect milk quality (Žilaitis et al., 2008). Increased fat content during early lactation period and reduced concentration of lactose may be considered as symptoms for ketosis risk. One of the ways to improve milk quality is prevention of ketosis (Žilaitis et al., 2008). BHB concentration in blood serum is statistically reliably related to milk fat content (Žilaitis et al., 2008).

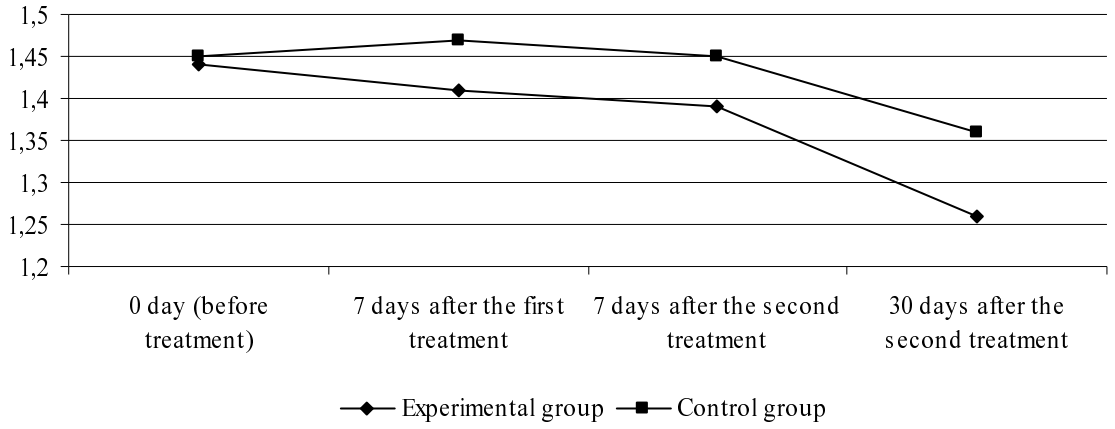

Fig. 6. **Comparison of cow milk fat to protein ratios in test and control groups**

Conclusion

The effects of the medicinal product Metabolase® were as follows:

reduced BHB concentration and milk fat to protein ratio; increased amounts of glucose in blood; increased milk production and thus reduced risk of ketosis; the increased albumin concentrations and reduced AST concentration signal a better function of liver.

Recommendation: in order to reduce the risk for ketosis in cows, it is recommended to inject 500 ml of the medicinal product Metabolase® twice every 7 days.

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