Effect of various fats on digestibility of nutrients in diet of calves

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Abstract. Article presents results of study on effect of various vegetable fats on digestibility of the main nutrients in young cattle. We obtained new data on effects of fats on absorption of amino acids, analyzed blood of experimental animals, depending on source of lipids. We found that use of native form of fat contributes to less intensive digestion of raw fat, in contrast to prepared. In this case, fatty acid composition plays significant role. Transformation of amino acids showed slight difference between the samples. As result of our research, thanks to unique surgical operation on transplantation of the pancreatic duct into isolated segment of intestine, new knowledge was obtained about exocrine function of pancreas, absorption of nutrients, and biochemical parameters of blood in cattle when exposed to fats in animals. Thus, obtained the data reflect body's response to various types of fats, administered in doses that can be digested by body, without disrupting metabolism of other substances.

1. Introduction

Intake and absorption of chemically complex feed compounds in gastrointestinal tract is the most important part in animal nutrition [1]. When entering the gastrointestinal tract proteins, carbohydrates and fat, which is the most energy-intensive compound, these substances begin to transform into components under influence of the microbiota and gastrointestinal juice, containing various enzymes [2].

Sufficient amount of data has been accumulated by the science of use of fatty supplements in feeding cattle, input standards have been established, and methods for preparing fats before feeding have been developed [3,4].

Effect of various fatty acids on microflora of rumen was also established [5]; process of biohydrogenation of rumen bacteria with respect to fatty acids was studied [6]. Fat, falling into body, breaks down into fatty acids and they are in contact with the microflora, which may be accompanied by violation of scar.

In our work, we studied digestibility of nutrients, depending on input of fat in diet of animals. We paid attention to fatty acid composition, effect of fat on absorption and assimilation of amino acids, on biochemical composition of cattle blood, as well as the adaptation of the pancreas to fatty diet through changes in activity of pancreatic enzymes.
2. Materials and methods
In experiment, calves of the Kazakh white-headed breed with average weight of 115-120 kg, at the age of 7 months, were used. Animal service and experimental studies were performed in accordance with the instructions and recommendations Russian Regulations, 1987 (Order No. 755 on 12.08.1977 the USSR Ministry of Health) and «The Guide for Care and Use of Laboratory Animals (National Academy Press Washington, D.C. 1996)».

In carrying out research, measures were taken to minimize suffering of animals and reduce number of samples used.

Calves were kept in special cage with free access to water and feed. Basic diet (BD) was balanced for basic nutrients, according to detailed norms of VNIIMS. The diet included: mixed grass hay (6.5 kg), mixture of concentrates (2.3 kg), dicalcium phosphate 35 g, salt 35 g, additionally injected fat (sunflower oil, palm fat) in the amount of 3% of the dry matter of diet.

To study the pancreatic secretion of animals, original operation was performed to impose duodenal anastomosis [7].

Studies conducted on empty stomach. Before start of experiment, animal was placed in special machine for purpose of fixation during experiment. Special container for collecting juice was attached to the fistula. Juice collection was performed for 8 hours with interval of 30 minutes. After taking first sample, animals were fed (amount of feed is 30% of the daily value) and continued to collect juice. Determination of the quantity and enzymatic activity of the juice was carried out «in cito».

Studies were carried out in the laboratory "Agroecology of technogenic nanomaterials" and the Testing Center (Federal Scientific-Research Center "Biological Systems and Agrotechnologies of the RAS", accreditation certificate RA. RU.21PF59 from 02.12.15).

Blood was collected from jugular vein into vacuum tubes with addition of anticoagulant, for biochemical parameters - into vacuum tubes with clotting activator (thrombin). Morphological blood analysis was performed on an automatic hematology analyzer URIT-2900 Vet Plus («URIT Medical Electronic Group Co., Ltd», China), biochemical analysis of blood serum - on an automatic analyzer CS-T240 («DIRUI Industrial Co., Ltd», China) using commercial kits for veterinary (CJSC "DIAKON-DS", Russia).

Amylase activity was measured according to Smith-Roy in modification to determine the high enzyme activity [8], proteases - by hydrolysis of casein purified by Hammersten with calorimetric control (wavelength 450 nm), lipase - using automated biochemical analyzer CS-T240 («Dirui Industrial Co., Ltd», China) using commercial biochemical kits for veterinary DiaVetTest (Russia).

Trypsin was determined by method of V.G. Vertiprahova et al. [9].

Level of NO metabolites (i.e., the total concentration of nitrates and nitrites, NOx) was determined by colorimetric method for development of color in diazotization reaction of sulfonamide nitrite, which is part of the Griss reagent. The color intensity was determined on a Labsystems Multiskan MCC/340 instrument, measuring optical density of samples in standard 96-well plate (for ELISA analysis) at wavelength of 540 nm (principle of action is vertical photometry) [10].

Statistical analysis was performed using ANOVA methods (Statistica 10.0 software package, StatSoft Inc., USA) and Microsoft Excel. The significance of differences in the compared indicators was determined by Student's t-test. The level of significant difference was set at P <0.05.

3. Discussion of the results
Changes in activity of pancreatic enzymes under influence of various diets is example of adaptation that is inherent in various types of mammals. Therefore, we established dependence of amount of pancreatic juice secreted and level of enzyme activity on structure of diet and, as consequence, change in absorption of nutrients of diet. In our studies, we obtained differentiated results on nutrient digestibility. Thus, digestibility of dry matter for sunflower oil averaged 94.1%, for palm oil 93.6%, the difference was 0.5%.

Difference in digestibility of organic matter was 1.4%, more digested in group with palm oil. Digestibility of raw protein was greater in group with prepared palm fat (on average, 86.1%), which is 4.7% more than in group with use of sunflower oil.
In group with use of sunflower oil, digestibility of crude fat averaged 44.8%, which is 15.0% lower than in group with use of palm fat; digestibility of crude fat in this group averaged 59.8%.

In our experience, it was not established that digestibility of crude fiber decreased in all groups, which averaged in groups 85.4% and 84.5% for group with sunflower oil and palm fat, respectively. Difference in groups was 0.8%. Nitrogen-free extractives are more digested in group with use of palm fat by 1.7% than in group with use of sunflower oil.

To understand the mechanisms of adaptation of activity of enzymes of excretory system of pancreas to various sources of fat, dynamics of pancreatic juice secretion and activity of enzymes over 8-hour period were studied. As result of introduction of feed, including sunflower oil, in diet, increase in amylolytic activity was observed against the background of decrease in protease activity, while lipolytic activity increased in gastric phase and gradually decreased in intestinal phase by 15.3%. It should be noted that amount of pancreatic juice secreted also decreased in intestinal phase of regulation relative to gastric phase by 25%. The same pattern was noted in relation to activity of enzyme amylase.

Amylolytic activity increased in gastric phase and decreased in intestinal by 28%, proteolytic activity gradually decreased in second phase of regulation by 10.6%, in intestinal by 59.3%.

When replacing sunflower oil in diet with palm oil, activity of pancreatic juice enzymes had following pattern: there was marked increase in lipolytic and amylolytic activity almost 2 times during intestinal phase of regulation against background of reduced protease activity. This is probably due to pronounced pancreatic secretion response to higher dietary fat content.

It should be noted that proteolytic activity in presence of palm fat, as well as sunflower oil, gradually decreased to intestinal phase of regulation by more than 2 times, and in gastric phase by 43.3%. At the same time, level of pancreatic secretion was higher in non-reflex phase of regulation, and gradually decreased to third phase of regulation by 9.3% (to gastric phase by 4%).

Comparing the dynamics of secretion of pancreatic juice, we observed differences characterizing increase in the release of juice in the gastric phase of regulation of pancreatic secretion in the presence of sunflower and palm oil in the reflex-phase regulation and gradual decrease towards the intestinal phase.

Considering the increase in level of lipolytic activity of enzymes on background of fatty diet in connection with increase in activity of amylase, we can conclude about the adaptation of pancreas to fat composition of diets [8].

The metabolism of protein, calcium and phosphorus in body did not change, these indicators were stable.

Important in terms of changes in environment and diets is to support normal physiological state of body, which is ensured by active supply of oxygen cells and depends on intensity of the exchange of gases between the cells and the environment. Blood is the most important, as being internal environment of body, it has constant composition, and changes in qualitative or quantitative composition mainly indicate metabolic disorder and process of growth, development and productivity associated with it.

Consequently, analysis of morphological and biochemical blood parameters is important in normalizing feeding of animals and at different sources of protein in body of calves. Introduction to diet of animal feed containing sunflower oil and palm fat, morphological and biochemical blood parameters were within physiological norms [11]. However, it is necessary to note tendency to decrease or increase in some of studied parameters, which may indicate violation of metabolic processes in body.

Thus, in group that received palm fat, when evaluating hemostasis system, decrease in almost all indicators was recorded, except for alkaline phosphatase and α-amylase relative to indicators of group that received sunflower oil [12].

Inclusion of sunflower oil in diet contributed to increase in hemoglobin level of 30.6% relative to group that received palm oil.

When studying level of NO-metabolites, we noted significant difference in exchange of NO-metabolites between samples in diets containing sunflower or palm oil.

To date, convincing evidence has been obtained that nitric oxide (NO) mediates number of physiological processes, including regulation of vascular tone, plasma and platelet hemostasis, the formation of immune response and the neurotransmission, inhibition of smooth muscle cell proliferation. At the same time, decrease in concentration of NO is considered one of the main causes
of endothelial dysfunction, which occurs in such pathological conditions as hypercholesterolemia, type 2 diabetes, arterial hypertension (AH), heart failure [12].

Insufficient production of NO is associated with development of disorders in the cardiovascular and other systems of the body; At the same time, its excessive production, due to which antimicrobial effect is provided during inflammation, can turn from adaptation link into link of pathogenesis and become no less dangerous damaging factor for body than NO deficiency [12]. In our studies, we recorded increase in level of nitric oxide metabolites in all animals, but level of NO metabolites in animals whose diet included palm oil was 3 times lower than in animals treated with sunflower oil. We consider increase in level of NO metabolites to be consequence of impaired metabolic processes in body during high-fat diet.

Analysis of scientific literature shows that pancreatic enzymes, along with admission to pancreatic juice, also enter blood. Existing hypothesis of Laporte and Tremolyer reveals mechanism of regulation of pancreatic enzyme production: entry of trypsin into blood reduces release of enzymes with pancreatic juice, and administration of trypsin inhibitor, on the contrary, is accompanied by increase in separation of enzymes. Determination of trypsin activity showed that inclusion of palm oil in diet contributed to increase in trypsin activity by 9.1%.

4. Conclusion

One of the ways to increase productivity of farm animals is their assimilation of nutrients of feed used. Assimilation depends on many factors, including structure, composition and volume of diet, as well as on physiological state of animal and proper functioning of digestive system, mainly pancreas.

As result of our studies, we found that when cattle of sunflower and palm fat were included in diet, digestibility of nutrients, in particular, of raw protein and crude fat, was higher in group that received palm fat. When replacing sunflower oil in diet with palm oil, there is slight difference of 0.5% digestibility of dry matter and crude fiber in direction of sunflower oil.

The amount of pancreatic juice produced correlated with enzyme activity.

In our work, we noted that the saturation of diets with fats increases the activity of enzymes amylase and lipase and decreases proteolytic activity.

When sunflower oil was included in diet, the greatest lipolytic and amyloytic activities were observed in the gastric phase of pancreatic secretion regulation, with a gradual decrease in protease to the second phase of regulation. The inclusion of palm fat in the diet was accompanied by a pronounced increase in lipolytic and amyloytic activities, with an increase in activity up to the intestinal phase. The level of protease activity decreased gradually towards the intestinal phase of regulation.

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