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The effect of adding N-carbomylglutamate to the diet on dairy productivity and product quality in cows

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Abstract. The study examined the effect of N-carbamylglutamate (NCG) additives on milk yield, nitrogen metabolism and milk quality in highly productive dairy cows. In the experimental group, the efficiency of nitrogen use increased by 2.1%, and milk production increased. Milk quality indicators remained unchanged. After 30 days of taking N-carbamylglutamate, the total milk yield in the experimental group was 5.6% ($p<0.05$) higher than the baseline level and 8.3% ($p<0.05$) higher than in the control group. In both groups, there was a natural decrease in productivity at the third stage of lactation, but in the experimental group, a higher level of productivity was maintained. The level of arginine in the blood increased significantly in the experimental group, which indicates an improvement in non-protein nitrogen absorption. Although there were no significant changes in the amino acid composition of milk, the experimental sample had a higher concentration of lactose (by 3.4%) and dry matter (by 1.68%), as well as casein (by 1.8%) compared to the control group. Analysis of the fatty acid content in the experimental sample revealed elevated levels of several types of fatty acids, including stearic, oleic, and long-chain monounsaturated and polyunsaturated acids. It was found that the use of N-carbamylglutamate does not have a negative effect on key indicators of the quality of dairy products.

1 Introduction

Ruminants have a unique ability to convert dietary nitrogen into essential amino acids, which are subsequently used to synthesize proteins such as muscle tissue and milk components. This process mainly occurs in the rumen, where microbiological fermentation decomposes the eaten food into simpler components [9, 10]. The resulting amino acids are subsequently absorbed and used by the animal's body for various physiological functions, including milk and meat production. Thus, the efficient conversion of nitrogen from fibrous feed and non-protein sources is crucial to maintain the productivity of ruminants. Nevertheless, up to 70% of the nitrogen contained in feed is excreted in feces and urine. Urea is the main product of nitrogen metabolism and is used to remove excess nitrogen from the body. Urea is formed

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from amino acids during the ornithinic cycle in the liver, which also helps to break down toxic ammonia. Finding effective and safe additives to improve nitrogen use by ruminants is a major challenge. The use of ornithinic cycle activators can increase the efficiency of ammonia utilization and the cleavage of other nitrogenous compounds [6,11]. Stimulation of the production of endogenous arginine, nitric oxide, and polyamines (putrescine, spermidine, spermine) in the ornithinic cycle can increase animal productivity [1,13]. Milk yield and milk composition are the most important economic indicators in the dairy industry and have been carefully studied. The fat and protein content in milk are the most important characteristics of raw milk that affect economic profitability. Amino acids play an important role in regulating protein and fat levels in milk. It has been proven that the addition of amino acids such as lysine and methionine to the diet increases milk yield, increases the efficiency of nitrogen use in lactating cows and increases the protein content in their milk [2]. Although ruminants are able to synthesize arginine, it is generally considered essential for dairy cows and plays an important role in various body functions. Acting as a precursor to polyamines and nitric oxide (NO), arginine can promote angiogenesis and lactation. The addition of arginine increases the synthesis of casein protein in the mammary gland of cattle. Nevertheless, due to the rapid decomposition in the rumen, free arginine is not an effective feed additive for dairy cows. In addition, unbalanced consumption of amino acids with the help of amino acid supplements can lead to an imbalance in the amino acid composition [17]. N-carbamylglutamate (NCG) is a stable analog of N-acetylglutamate (NAG) in the rumen and can be considered as a potential arginine enhancer for ruminants. The purpose of this production experiment is to study the effect of NCG on milk production in highly productive cows to develop a method for its use as an activator for endogenous arginine production. Milk yield and milk composition play a central role in the production of dairy products and have been the subject of numerous studies [8]. The fat and protein content plays an important role in determining the economic efficiency of raw milk [7]. Amino acids act as essential regulators of protein and fat levels in milk, and the addition of certain amino acids, such as lysine and methionine, to the cow's diet can increase milk yield, nitrogen efficiency, and protein content [14]. Despite the fact that cows have the ability to synthesize arginine, its role in milk production is significant. Arginine, as a precursor of polyamines and nitric oxide, plays an important role in stimulating angiogenesis and lactation [15]. Its use has been shown to increase casein synthesis in epithelial cells of mammary gland [7]. Nevertheless, due to its rapid decomposition in the rumen of cows, unprocessed arginine is ineffective as a feed additive for dairy animals. The introduction of amino acids into the diet of dairy cows requires careful consideration to avoid an imbalance in the amino acid composition [13].

2 Materials and Methods

In this study, the effect of N-carbamylglutamate (NCG) additives on productivity, nitrogen metabolism and milk quality of highly productive cows during lactation was studied. On the basis of the farm, experimental and control groups from the herd being used were formed to conduct an experiment using the method of pairs of analogues n = 20 in the second phase of lactation. The experimental groups received a standard diet according to the technological scheme of the farm. The cows of the experimental group were added the feed additive N-carbamylglutamate (a synthetic analogue of N–acetylglutamate (NAG) at a dosage of 20 g per head per day (continuously for 30 days). The feed was distributed evenly over two meals. The studied additive was added daily to the feed before the first meal and thoroughly mixed. This modification of the diet was carried out to assess the potential effect of NCG supplements on various indicators of animal dairy productivity. To evaluate the effectiveness of this supplement in terms of its effect on the level of amino acids in milk and blood, the results were compared at the initial stage and after 30 and 60 days of supplementation. Milk quality was assessed at the end of the 30-day supplementation phase. The amino acid composition of blood and milk samples was determined using capillary electrophoresis on the Kapel-105M device (Lumix, Russia), and the fatty acid composition of milk was determined using the CombiFOSS 7 analyzer (Foss, Denmark). To analyze the collected data, a statistical analysis was performed using the Statistica 8.0 software and Microsoft Office Excel.

3 Results and Discussion

The study showed that despite similar levels of productivity at the beginning of the production experiment, the group receiving NCG demonstrated an increase in productivity during lactation from 180 to 210 days and maintained an increased level throughout the follow-up period after the addition of a feed supplement. The efficiency of nitrogen use in the diet of the experimental group improved by 2.1% compared to the beginning of the study (based on changes in nitrogen consumption per unit of milk production in the experimental and control groups). Cows of the experimental group gave 0.9 liters more milk daily than cows of the control group, while milk quality indicators remained unchanged. In general, milk productivity in the experimental group increased by 5.6% ($p<0.05$) after 30 days of taking the ornithine activator compared to the baseline level and by 8.3% ($p<0.05$) compared to the control group. This increase in productivity can be explained by the use of an activator that stimulated endogenous nitrogen metabolism. The decrease in productivity in the control group may be associated with a natural decrease in productivity, which occurs at the third stage of lactation. Starting from the 210th day of lactation, the experimental group showed a steady tendency to decrease productivity compared to the beginning of the study. By day 210, milk productivity decreased by 6.7% ($p<0.05$) compared to the baseline level in the control group. When assessing milk productivity, NCG did not have a negative impact on the milk yield of highly productive cows.

Table 1. Milk quality indicators (M±m, n=10).

Indicators	Experimental group			Control group		
	Beginning of the experiment	30 days	60 days	Beginning of the experiment	30 days	60 days
Fat content %	3.6±0.3	3.6±0.8	3.6±0.4	3.6±0.6	3.6±0.4	3.6±0.4
Protein content, %	3.3±0.6	3.3±0.4	3.3±0.3	3.3±0.4	3.3±0.7	3.3±0.5
Number of somatic cells, thousand units/ml	142±28	126±25	125±30	158±26	162±32	182±36
Productivity, kg	40.7±0.6	42.9±0.71.2	40.9±0.51.2	40.1±0.9	39.6±0.4	37.4±0.72
Note: *P<0.05, according to the t-criterion when compared to the control						

These results indicate that the use of NCG led to an increase in productivity, which indicates the effectiveness of the use of endogenous nitrogen by experimental animals. During the study, significant differences were observed in the arginine content in the blood: on day 30, it increased by 5.7% ($p<0.05$), and at the end of the experiment - by 4.6% ($p<0.05$). These data indicate an increase in the efficiency of fixation of non-protein nitrogen due to the use of N-carbamylglutamate, as evidenced by an increase in arginine levels in the blood.

During the production process, there were no significant differences in the levels of individual amino acids in the milk of experimental cows compared with the experimental animals from the control group.

Table 2. The effect of N-carbamylglutamate on the amino acid profile.

Indicators	Experimental group			Control group		
	Beginning of the experiment	30 days	60 days	Beginning of the experiment	30 days	60 days
Amino acid composition of the blood (M±m, n=10).						
Arginine	1.72±0.05	1.83±0.03*	1.82±0.05*	1.71±0.04	1.73±0.05	1.72±0.05
NEAA	9.77±0.31	9.8±0.37	9.61±0.35	9.77±0.35	9.7±0.41	9.6±0.38
Total AA	19.52±0.58	19.61±0.66	19.35±0.61	19.48±0.57	19.54±0.95	19.35±0.59
Amino acid composition of milk (M±m, n=20).						
Arginine	0.08±0.005	0.09±0.004	0.08±0.005	0.08±0.004	0.08±0.005	0.08±0.005
NEAA	1.290±0.123	1.280±0.114	1.241±0.115	1.285±0.101	1.271±0.104	1.259±0.124
Total AA	2.416±0.221	2.389±0.248	2.365±0.261	2.410±0.239	2.378±0.266	2.370±248

Note: *P<0.05, according to the t-criterion when compared to the control
NEAA - nonreplaceable amino acids, Total AA - Total number of amino acids

To assess the effect of N-carbamylglutamate on product quality, the fatty acid composition of milk was analyzed. The quality assessment based on the fatty acid composition revealed several changes. The lactose level in the experimental group was 3.4% higher than in the control group, while the level of skimmed milk residue was 0.81% lower. The dry matter content was 1.68% higher, and the indicators of the experimental group in terms of casein exceeded those in the intact group by 1.8%. In addition, acetone and urea levels in the experimental group were 13% and 5.56% lower (P<0.05), respectively, compared to the control group. A significant decrease in urine urea levels correlated with a decrease in blood urea levels in the experimental group, which occurred simultaneously with an increase in milk production in this group. In addition, the fatty acid composition of milk showed an increase in the level of stearic acid by 12.46%, oleic acid - by 18.2%, long-chain fatty acids - by 22.7% ($p < 0.05$), medium-chain fatty acids - by 1.7%, monounsaturated fatty acids - by 18.8%. and the level of polyunsaturated fatty acids increased by 18.1% compared to the control group. In the experimental group, the level of saturated fatty acids was 5.8% higher, and the level of short-chain fatty acids was 2.6% higher. This general tendency to increase the content of fatty acids may be associated with increased metabolic activity. The results obtained indicate that the use of N-carbamylglutamate does not have a negative effect on key milk quality indicators. This can help to improve energy metabolism in animals by stimulating the production and activation of enzymes involved in carbohydrate, lipid, and protein metabolism, thereby optimizing the oxidation of nutrients and their conversion into energy. Improved nitrogen and energy metabolism leads to more efficient use of feed nutrients. By maintaining optimal levels of nitrogen and energy, NCG can help increase animal productivity.

Table 3. The content of fatty acids in milk on the 30th day of the study (M±m, n=10).

Indicators	Experimental group	Control group
Lactose	4.8956±0.17	4.7348±0.19
Casein	2.6104±0.18	2.5628±0.21
Acetone	0.0392±0.04	0.0444±0.05
Urea	43.54±0.70*	46.10±1.09
Milk fatty acids		
Myristic	0.28±0.05	0.28±0.06

Palmitic	0.73±0.16	0.73±0.18
Stearic	0.31±0.07	0.27±0.08
Oleic	1.10±0.16	0.93±0.21
Long-chain FA	1.35±0.07*	1.10±0.09
Medium-chain FA	1.14±0.23	1.12±0.27
Mono-unsaturated FA	1.01±0.16	0.85±0.19
Poly-unsaturated FA	0.13±0.02	0.11±0.02
Saturated FA	1.98±0.46	1.87±0.52
Short-chain FA	0.39±0.11	0.38±0.03
FA trans-isomers	0,11±0,11	0,08±0,03
Note: *P<0.05, according to the t-criterion when compared to the control		

4 Conclusions

In conclusion, the study examined the effect of N-carbamylglutamate (NCG) additives on productivity, nitrogen metabolism and milk quality of highly productive cows during lactation at an agricultural enterprise. The data obtained indicate that the addition of NCG had a positive effect on the productivity of lactating cows, as evidenced by an increase in milk yields in the experimental group compared with the control group, despite similar initial levels of productivity. In the experimental group, there was a steady improvement in indicators, which indicates the effectiveness of NCG supplements in stimulating milk production. Based on the collected data, it is worth noting the increase in the efficiency of nitrogen use observed in the experimental group with respect to milk production compared to intact animals. This indicates that NCG increases the efficiency of endogenous nitrogen use by cows, which leads to increased productivity without compromising milk quality. Analysis of the amino acid composition of the blood showed a significant increase in arginine levels in the experimental group, which indicates an improvement in the absorption of non-protein nitrogen due to NCG supplements. These data highlight the potential of NKG in optimizing nitrogen metabolism in lactating cows. As for the quality of milk, the use of N-carbamylglutamate (NCG) additives did not have a negative impact on the main quality indicators. The fatty acid composition of milk has indeed undergone some changes, including an increase in the content of certain fatty acids, which may indicate changes in metabolic activity caused by NCG introduction. The use of NCG in the experimental group receiving a standard diet did not have a negative effect on the overall quality of milk. Based on the data obtained, the addition of an ornithinic cycle activator is promising as one of the additional methods to stimulate the productivity of lactating cows at the second stage of lactation and optimize nitrogen metabolism in animals. This can lead to improved nutrient utilization and increased milk yields, as well as improved nitrogen utilization. Further research is needed to determine the long-term effect of NCG and the optimal dosage for feeding dairy cows.

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