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ENZYMATIC ACTIVITY OF THE RUMEN MICROBIOTA AND INTENSITY OF GROWTH OF YOUNG EWES UNDER THE ALIMENTARY ACTION OF PREBIOTIC SUPPLEMENT ISGD

The article presents the results of experimental research on changes in the enzymatic activity of the rumen microbiota and the intensity of growth of young ewes when using as a part of the compound feed of a new prebiotic made on the basis of baking yeast *Saccharomyces cerevisiae* "Inactivated Dry Glutathione Yeast" (ISGD), manufactured by the "Enzym" company (Lviv city).

The research was carried out in the conditions of the sheep farm of the State Enterprise "Experimental Farm "Hrusiatychi" (Lviv region, Zhydachiv district, Hrusiatychi village) and the department of small animal husbandry of the Institute of Agriculture of the Carpathian region of the National Academy of Agrarian Sciences of Ukraine. For this purpose, 4 groups of 11-month-old young ewes with a live weight of 38.0–38.2 kg, 5 animals each, were formed. The duration of the experiment – 2 months (February – March).

The main ration of the control group young ewes consisted of 1.1 kg of meadow cereal-herb hay and 0.5 kg of standard compound feed. Animals of the first, second, and third experimental groups were additionally fed ISGD prebiotic as part of compound feed in amounts, respectively: 1.0; 1.4 and 1.8 % of its mass.

At the end of the experimental period, after the morning feeding, from 3 animals of each investigited groups, with the help of oro-esophageal probe, ruminal fluid was collected, in which the enzymatic activity of the microbiota was determined. In order to determine the intensity of growth of the young ewes, their monthly weighing was carried out. The live weight of the animals was determined at the beginning and at the end of the experimental period.

It has been established that the use of the prebiotic in the feed of animals in doses of 1.0; 1.4 and 1.8 % of the weight of concentrate feed increases the enzymatic activity of amylolytic, proteolytic and cellulolytic microorganisms of the rumen by 13.7–120.5; 1.9–19.0 and 5.2–10.5 %, compared to animals that did not receive such a supplement as part of the diet.

The use of ISGD prebiotic in the indicated doses as part of the combined fodder for young ewes during the experimental period increases the average daily increase in the live weight of animals by 1.3–1.6 times. The most pronounced stimulatory effect on the digestive activity of the symbiotic microbiota of the rumen and the intensity of animal growth is revealed by 1.4 % of the prebiotic ISGD from the mass of compound feed.

Keywords: prebiotic, young sheeps, feeding, ruminal metabolism, growth intensity of the animals.

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Ферментативна активність мікробіоти рубця та інтенсивність росту ярок за аліментарної дії пребіотичної добавки ISGD

Наведено результати експериментальних досліджень щодо змін ферментативної активності мікробіоти рубця та інтенсивності росту ярок при застосуванні у складі комбікорму нового пребіотика, виготовленого на основі хлібопекарських дріжджів *Saccharomyces cerevisiae* «Інактивовані сухі глютатіонові дріжджі» (ISGD), виробництва фірми «Ензим» (м. Львів).

Дослідження проведено в умовах вівцеферми Державного підприємства «Дослідного господарства “Грусятичі” (Львівська область, Жидачівський район, село Грусятичі) та відділу дрібного тваринництва Інституту сільського господарства Карпатського регіону Національної академії аграрних наук України. З цією метою було сформовано 4 групи ярок-аналогів 11-місячного віку живою масою 38,0–38,2 кг, по 5 тварин у кожній. Тривалість досліду 2 місяці (лютий – березень).

Основний раціон ярок контрольної групи складався із 1,1 кг лучного злаково-різнотравного сіна і 0,5 кг стандартного комбікорму. Яркам першої, другої і третьої дослідних груп у складі комбікорму додатково згодовували пребіотик ISGD у кількостях відповідно: 1,0; 1,4 і 1,8 % від його маси.

Після завершення експериментального періоду після ранкової годівлі від 3 ярок кожної із піддослідних груп за допомогою рото-стравохідного зонда відбирали рубцеву рідину, в якій визначали ферментативну активність мікробіоти. Для встановлення інтенсивності росту ярок проводили їхнє щомісячне зважування, а також визначали живу масу тварин на початку і після завершення експериментального періоду.

Встановлено, що застосування у годівлі ярок вказаного пребіотика у дозах 1,0; 1,4 і 1,8 % від маси комбікорму підвищує ферментативну активність амілолітичних, протеолітичних і целюлозолітичних мікрорганізмів рубця відповідно на 13,7–120,5; 1,9–19,0 і 5,2–10,5 % порівняно до тварин, які не отримували такої добавки у складі раціону.

Застосування пребіотика ISGD у наведених дозах у складі комбікорму ярок за експериментальний період підвищує середньодобові прирости живої маси тварин в 1,3–1,6 рази. Найбільш виражений стимулюючий вплив на ферментативну активність симбіотичної мікробіти рубця та інтенсивність росту тварин виявляє 1,4 % пребіотика ISGD від маси комбікорму.

Ключові слова: пребіотик, молодняк овець, годівля, рубцевий метаболізм, інтенсивність росту тварин.

Introduction. Prebiotics are non-digestible components of various types of microorganisms and some plants, which, entering the digestive tract of animals, selectively stimulate the vital activity of the microbiota present in it [1, 20, 28, 33]. An important property of prebiotics is that they are resistant to stomach acidity, absorption and hydrolysis by enzymes of the gastrointestinal tract of animals [8, 14, 18]. Currently, a wide range of prebiotic preparations are used in the feeding practice of animals of various species, which include: mannan oligosaccharides, fructooligosaccharides, galactooligosaccharides, lactulose, lacthiol, beta-glucans, inulin and others [10, 16, 17, 23, 27]. The most common prebiotics, which have been used in recent years as bioadditives to ruminant diets, are preparations made on the basis of microscopic fungi, including baking yeast *Saccharomyces cerevisiae* [6, 22, 31].

In ruminants, the alimentary use of yeast prebiotics is characterized by the specificity of metabolic action compared to monogastric animals [5, 7, 8, 19]. The mechanism of biological action of yeast prebiotics due to the use of their additives in ruminant diets is only partially elucidated, but it is known to be complex and multifaceted. Experimental studies have established that these prebiotics, when used in cattle and sheep feed rations, have a positive effect on ruminal digestion processes, especially cellulolysis and fungal protein synthesis [1, 25]. Prebiotics made on the basis of yeast fungi activate the rate of enzymatic processes in the symbiotic microflora of the rumen, while inhibiting the production of carbon dioxide, ammonia and methane [8, 33]. Prebiotic preparations obtained from *Saccharomyces cerevisiae* yeast strains are particularly effective in ruminants for optimizing the acidity of the rumen environment. They stimulate the metabolic activity of ciliated populations, the enzymatic activity and vital activity of the rumen microflora, which produces organic acids, oligosaccharides, vitamins of group B, amino acids [8, 15, 20].

The use of yeast prebiotic supplements in the diets of cows stimulates fiber fermentation by rumen microorganisms and increases their milk productivity [3, 19, 21, 32]. In fattening cattle, optimization of ruminal digestion processes due to the use of yeast prebiotic additives in diets increases the efficiency of assimilation of feed nutrients, as a result of which the average daily gains and live weight of animals increase [17, 19, 30, 33].

The market of yeast feed prebiotics and their assortment for the needs of animal husbandry is constantly growing. In recent years, in our country, the “Enzym” company (Lviv city) has established the production

of a number of prebiotic preparations based on *Saccharomyces cerevisiae* yeast. However, their metabolic and productive effect when used as food in ruminants, remains unclear. Based on the above, the aim of our work was to study the effect of the use of supplement of the new domestic prebiotic "Inactivated Dry Glutathione Yeast" (ISGD) in feed rations of young sheeps, on the enzymatic activity of the rumen microbiota and the intensity of animal growth.

Research materials and methods. The research was conducted in the conditions of the sheep farm of the State Enterprise "Experimental Farm "Hrusiatychi" (Lviv region, Zhydachiv district, Hrusiatychi village) and the department of small animal husbandry of the Institute of Agriculture of the Carpathian region of the National Academy of Agrarian Sciences of Ukraine.

In order to perform this work, using the method of analogues by live weight and age (the live weight of the animals at the beginning of the experiment was 38.0–38.2 kg, the age was 11 months), four groups of Askanian meat-wool breed young ewes, of 5 animals each were formed. The scheme of the experiment is shown in Table 1. The main diet of the young ewes of control group consisted of 1.1 kg of meadow cereal-herb hay and 0.5 kg of standard compound feed, which provided the animal's body with nutrients, vitamins, macro- and microelements. The ewes of the first, second and third experimental groups were additionally fed prebiotic "Inactivated dry glutathione yeast" (ISGD) produced by the company "Enzym" (Lviv city) in the following quantities: 1.0; 1.4 and 1.8 % of its mass.

1. The scheme of the experiment

Groups of animals	The number of animals in the group	The composition of the daily ration
Control	5	Basic ration (BR) (1.1 kg of meadow hay + 0.5 kg of compound feed)
Experimental 1	5	BR + 1.0 % ISGD from the mass of compound feed
Experimental 2	5	BR + 1.4 % ISGD from the mass of compound feed
Experimental 3	5	BR + 1.8 % ISGD from the mass of compound feed

The composition of the compound feed for experimental ewes is given in Table 2. The duration of the experiment was 60 days (February-March).

2. Compound feed recipe for experimental ewes

Name of feed	Content, %
Corn	10
Oats	15
Barley	30
Wheat	15
Wheat bran	15
Sunflower meal	12
Monocalcium phosphate	1
Salt	1
Premix (Composition is given in Table 3)	1
1 kg of compound feed contains:	
Dry matter, g	846
Exchangeable energy, MJ	10.7
Crude protein, g	146
Crude fat, g	32
Crude fiber, g	84
Calcium, g	3
Phosphorus, g	6

Feed consumption was recorded every 10 days by weighing the amount of given feed and unconsumed residues to the nearest 0.1 kg. The average daily consumption per 1 head of the experimental ewes during the experiment period was: dry matter – 1.3 kg, exchangeable energy – 13.2 MJ, crude protein – 176 g, crude fat – 39 g, crude fiber – 390 g, calcium – 9 g, phosphorus – 5 g.

At the end of the 60-day experimental period, after morning feeding from 3 ewes of each of the 4 groups, with the help of an oro-esophageal probe, the rumenal fluid was collected, in which the enzymatic activity of the microbiota was determined. In order to determine the intensity of growth of the ewes, their monthly weighing was carried out and the live weight of the animals was determined at the beginning and at the end of the experimental period.

Determination of the amylolytic, proteolytic and cellulolytic activity of the rumen microbiota was carried out colorimetrically, according to the methods described in the handbook by Vlizlo V. V. et al. [2].

3. The composition of the premix used in the combined feed of experimental ewes

Components	Units of measurement	Quantity
Vitamins		
D_3	IU/kg	200
E	mg/kg	2000
K_3	mg/kg	20
B_1	mg/kg	230
B_2	mg/kg	880
B_6	mg/kg	300
B_{12}	mg/kg	5
C	mg/kg	3000
Calcium pantothenate (B_3)	mg/kg	2000
Folic acid (B_c)	mg/kg	30
Choline chloride (B_4)	mg/kg	0
Niacin (B_5)	mg/kg	1950
Biotin (H_2)	mg/kg	3
Microelements		
Manganese	mg/kg	8000
Zinc	mg/kg	6000
Cuprum	mg/kg	1300
Iodine	mg/kg	50
Cobalt	mg/kg	175
Selenium	mg/kg	5
Filler	g	Up to 1000

The obtained digital data were processed by standard methods of variational statistics using the Excel 2003 program package.

Research results. A number of scientific studies have proven that in ruminant animal species, yeast prebiotics activate the enzymatic activity of rumen microflora, suppress the growth and development of pathogenic bacteria in the digestive tract, have an immunomodulatory effect in the body and increase the productive qualities [11, 12–15, 24, 29, 34, 35].

In recent years, the "Enzym" company (Lviv city) in Ukraine has established the production of a number of prebiotic bioadditives for animal rations based on *Saccharomyces cerevisiae* yeast. One of them is the prebiotic “Inactivated Dry Glutathione Yeast” (ISGD). However, the metabolic and productive effect of the alimentary use of this prebiotic in

ruminant feeding rations has not been clarified at this time. Based on this, the goal of our work was to study the effect of different doses of the specified prebiotic in the feeding diet of young sheeps on the metabolic activity of the rumen microbiota and the intensity of growth.

The results regarding changes in the enzymatic activity of rumen microbiota in experimental animals are shown in Table 4.

Determination of the amylolytic, proteolytic and cellulolytic activity of the rumen microbiota (Table 4) shows that their parameters, compared to the control, are the highest in the rumen of the third group, which received the prebiotic ISGD in quantity of 1.8 % by the weight of the compound feed, slightly lower in the second and lowest in the animals of the first experimental group, which were fed, respectively, 1.0 and 1.4 % of the prebiotic additive by weight of compound feed.

The use of supplements in the indicated amounts of prebiotic ISGD in the compound feed for young sheeps compared to animals that did not receive them, increases the amylolytic activity of the rumen microbiota by 13.7–120.5 %; proteolytic – by 1.9–19.0 %; cellulolytic activity – by 5.2–10.5 % (P<0.01–0.001).

4. Enzymatic activity of rumen microbiota in test ewes (M±m, n=3)

Enzymatic activity	Groups of animals			
	control	exp. 1	exp. 2	exp. 3
Amylolytic (amyl units)	0.73±0.05	0.83±0.09	1.34±0.07***	1.61±0.06***
Proteolytic (microequiv. of act. tyrosine in 100 ml/min)	2.11±0.07	2.15±0.08	2.43±0.06**	2.51±0.09**
Cellulolytic (% of activity)	17.2±0.43	18.1±0.93	18.6±0.75	19.0±1.01

Note: In this and the following table, asterisks indicate the probability of the obtained data in relation to the animals of the control group. Accordingly: *P<0.05; **P<0.01; ***P<0.001.

From the data presented in Table 4, it can be concluded that the ISGD prebiotic, introduced into the composition of the combined fodder, due to the normalization of the acidity of the rumen environment, optimization of the quantitative and qualitative composition of the symbiotic microbiota of the rumen, stimulates its vital activity, which is evidenced by the increase in amylolytic, proteolytic and cellulolytic activity.

The scientific works of a number of researchers indicate the activation of ruminal digestion processes, an increase in the number of ruminal bacteria, amoebae, infusoria, microscopic fungi, and an increase in their metabolic activity due to the use of prebiotic yeast biosupplements in ruminant diets [4, 21, 26, 27].

It was also established that the introduction of ISGD prebiotic in the tested doses into the compound feed for young sheeps stimulates their growth and development (Table 5). In particular, it is shown that live weight of experimental groups of animals, compared to the control, increased by 1.3–1.6 times during the two-month experimental period (P<0.05).

5. Indicators of body weight and intensity of growth of experimental ewes (M±m, n=5)

Indicators	Groups of animals			
	control	exp. 1	exp. 2	exp. 3
Body weight, kg				
Beginning of the experiment	38.0±1.06	38.0±1.00	38.2±1.39	38.2±1.47
End of the experiment	42.0±0.35	43.2±0.55	44.4±0.67*	43.4±0.35*
Increase				
Gross, kg	4.00	5.20	6.20	5.20
Daily average, g	66.7	86.7	103.3	86.7
Relative, %	5.00	6.40	7.51	6.37

The significant increase in live weight gains of the ewes of the second research group is obviously explained by the activating effect of the prebiotic ISGD in a dose of 1.4 % of the mass of compound feed, on the qualitative and quantitative composition of the ruminal microbiota, its increase in the production of microbial protein, which is broken down in the small intestine into amino acids, provides an increase in protein synthesis of animal organs and tissues. These data indicate that despite the rather high enzymatic activity of the rumen microbiota in the third experimental group of animals, the highest increases in live weight were found in the ewes of the second experimental group.

Regarding the productive effect of the use of the prebiotic supplement ISGD in the compound feed of experimental lambs, the data obtained by us are consistent with the results of other researchers conducted on calves and lambs [9, 12–15, 18, 25, 34, 35], in which it was established that prebiotic preparations manufactured using modern technologies on the basis of yeast fungi not only activate the metabolic activity of the symbiotic

microbiota in the rumen, but also significantly stimulate the growth and development of animals.

The results obtained by us also indicate that the most pronounced stimulating effect on the enzymatic activity of the rumen microbiota and the intensity of the growth of ewes is revealed by the introduction of 1.4 % of the prebiotic ISGD by its weight into the composition of animal feed.

As for the economic efficiency of using ISGD prebiotics, it is most appropriate to use it in compound feed for young sheeps at a dose of 1.4 % of the weight of the compound feed, thereby increasing the assimilation of nutrients in the body, and due to the increase in live weight gain, get an additional 18 UAH of profit per animal.

Conclusions. It was established that the use of domestically produced prebiotic ISGD as part of compound feed for feeding young sheeps, in doses of 1.0; 1.4 and 1.8 % of the mass of combined feed, stimulates the amylolytic, proteolytic and cellulolytic enzymatic activity of the rumen symbiotic microbiota respectively by 13.7–120.5; 1.9–19.0 and 5.2–10.5 % and increases live weight gain by 1.3–1.6 times, compared to animals that did not receive the specified additives in the diet.

Based on the obtained results regarding the productive effect, it is recommended to add ISGD prebiotic at a dose of 1.4 % of the compound feed weight for feeding young sheeps.

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