

## Section 1. Agricultural sciences

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### EFFICACY OF PROBIOTIC LABS IN FORAGE PROCESSING

**Abstract.** It has been shown that the use of a lactic acid bacterial consortium in silage production to increase lactic acid fermentation and suppress undesirable microbial processes (decomposition), resulting in reduced nutrient loss and higher quality forage, in addition to the enrichment of a specific lactic acid bacterial consortium leading to increasing the protein content and the amount of amino acids.

**Keywords:** probiotic, LAB, silage, protein, amino acids.

#### 1. Introduction

Currently, cost-effective livestock production is impossible to imagine without creating a sustainable feed base. It's no secret that feeding animals with normal high-quality food contributes to the maximum development of productive qualities in young animals, and the maintenance of this quality in adults. At the same time, the current economic situation of many farms does not allow to produce the right amount and good quality of fodder (silage, hay, grain). Loss of feed in the process of forage harvesting and subsequent storage is more than 30%, so the harvesting and storage of high-quality forage is one

of the most important problems in animal husbandry [6]. In developing countries, the use of microbes or their biological products is attracting more attention in various branches of agriculture. Many efficient technologies have been developed in science for the preparation of fodder and poultry feed using biological agents [3; 8].

Intestinal infections caused by opportunistic microbiota account for a large percentage of infant falls. As a result of the use of various antibiotics and chemical preparations, the microbial balance is disturbed, dysbacteriosis occurs. The activity of the digestive, hormonal and immune systems of animals and birds, the metabolism of mineral substances is

disrupted, as a result of which, mineral substances are lacking in the body and the products obtained from it [1; 2]. Today, alternative preparations are considered probiotics, which contain live cultures of symbiotic bacteria of the digestive system. They are used as biologically active substances, have growth-promoting, curative-preventive properties, are safe, and are free from the disadvantages specific to antibiotics and chemotherapeutic agents [4; 5]. The introduction of technology for the enrichment of fodder with probiotic microbes in agriculture will be beneficial not only financially but also from the point of view of increasing the quality, because the loss of fodder in the process of harvesting and further storage of fodder is more than 30%, and after the use of microbes – 5–10%, in addition the use of microbes reduces the loss of young animals, the degree of infection of animals with pathogenic microbes, shows an anti-inflammatory effect and also increases milk yield and meat quality.

The purpose of the work is: application of probiotics in fodder production technology and verification of their effectiveness.

## 2. Methodology

**The growing of lactic acid bacteria:** In the work we used *Ent. durans* P13, *L. acidophilus* 1991, *St. termophilus* 103, *Ent. faecium* KES, *St. lactis* 62 probiotic LAB bacteria. For bacterial growth we used MRS (ISO) and MRS (Himedia) broth, MRS agar (Himedia), lactoagar, nutritional agar (Nutrient, Himedia, India), whey and pasteurized, sterilized fat milk (3.6%) produced by MMY Company, as well as dry skim milk (fat content: 1.5). Milk sterilization was performed under a sterilizing device for 15 minutes under a pressure of 0.8 mm/Pa.

The strains were stored in the collection of the laboratory of Microbiology of Artsakh Scientific Center.

## The process of making silage in the laboratory

For the study we used mixed grass silage which we gathered from two different communities: Qrasni and Aygestan.

For the preparation of the silage, weather changes have been taken into account. Because the grass season is limited to late spring and summer, when the weather is favorable, the work has been done from late May to late July. The height of the grass should be 20–30 cm. After collecting the grass, the dried mass quickly was silaged within a day with two different preparations of lactic acid and yeast. We added 10% preparations of consortium lactic acid (50%) and yeast (50%) to the silosing mass.

In laboratory conditions, food (silage) after the enrichment with probiotic bacteria was stored in appropriate containers for 45–60 days, after which we first checked their physicochemical parameters.

**Bacteriological analyses of feedstuff:** Bacteriological analyses were carried out according to generally accepted methods [7].

The number of viable bacteria was determined by gradual dilution and characterized by the titration method. Microbiological parameters (fungi, mold, pathogens) were determined according to established methods and operating standards.

**Protein determination method:** Determination of protein in feedstuff is according to GOST PA13496.4–93

**Amino acid determination:** Determination of the amino acid content, thin-layer chromatography was used, followed by calculation of the content relative to the control data. For the quantitative determination of amino acids two methods of hydrolysed samples of grass were used, esparcet and green mass of corn. The samples after

silaging were hydrolyzed at 1 atm 120 °C in the following ratio: 4 grams of the test 8 ml of 4 N HCl or 8 ml of 6 N HCl was added to the sample under a hydrolysis mode of 130 °C for 20 min. Subsequently hydrolysis was selected hydrochloric acid embodiment hydrolysis at 130 °C for 20 minutes.

Based on the results on the amino acid content in all silage samples of three regions of Art-sakh, thin-layer chromatography was used to transfer the selected samples to determine the

amino acid content on an amino acid analyzer using High-performance liquid chromatography (HPLC) systems (Semi-preparative “Avex ODS” C18 column (8 by 250 mm, Waters and Shimadzu, Japan); Shimadzu LC-20 analytical C18 column (4.6 by 250 mm, Symmetry, USA, with a detector Diode array SPD-20 a, auto-sampler).

### 3. Results

Microflora contamination indicators of the prepared fodder sample are presented in table 1 .

Table 1 . – Microbiological indicators of the received feedstuff

	Sampling location	mixed grass silage		
		Control	Sample of silage	
			<i>Ent. durans</i> P13L. <i>acidophilus</i> 1991 <i>St. termophilus</i> <i>St. Lactis</i>	<i>Ent. faecium</i> KE5 <i>L. acidophilus</i> 1991 <i>St. termophilus</i> <i>St. lactis</i>
Contamination of feedstuff with fungi	Aygestan	–	–	–
	Krasni	–	–	–
Contamination of feedstuff with different pathogen bacteria	Aygestan	–	–	–
	Krasni	$4.5 \times 10^8$	–	–
Content of probiotic LABs (CFU/ ml)	Aygestan	–	$6.2 \times 10^7$	$1.4 \times 10^9$
	Krasni	–	$8 \times 10^7$	$9.8 \times 10^7$

Table 2 . – The content of amino acids and protein in mixed grass of Krasni and Aygestan communities

Aygestan	Amino acids mg / ml										The amount of amino acids mg/ml	pro-tein%
	lys	arg	ala	glut	val	isole	tre	met	fenilala	start		
Control	0.8	–	1.6	2.4	3.2	0.8	1.6	3.2	0.4	0.4	14.4	17.0
LAB	2.4	–	3.2	3.2	3.2	2.4	2.4	3.2	0.4	0.4	21.6	27.0
Krasni	Amino acids mg / ml										The amount of amino acids mg / ml	pro-tein%
	lys	arg	ala	glut	val	isole	tre	met	fenilala	start		
Control	1.6	–	3.2	2.4	3.2	1.6	1.6	3.2	0.4	0.4	16.0	15.0
LAB	2.2	0.4	0.4	3.2	3.2	3.2	3.2	3.2	0.4	–	20.4	16.0

As can be seen from the data in Table 2, different sources of mixed grass silage in the con-

tent of some essential amino acids in the control and experimental samples are differ. Thus, the

enrichment of a certain consortium of silage lactic acid bacteria leads to an increase in the protein content and the amount of amino acids. The content of lysine in the studied Aygestan samples increases three times on average, and in the Krasni sample – twice on average. Enrichment of silage with LAB increases protein content by an average of 27% and essential amino acids by 30% without the use of biological additives.

### **Conclusion**

Summarizing the studies, the following can be concluded:

Enrichment of fodder with antimicrobial probiotic bacteria is an innovative processing technology in the field of silage of forage crops. Due to the biological activity of probiotic bacteria, the growth of fungi, pathogenic microflora is suppressed, the quality indicators of fodder are increased and it is the promising for use in animal feeding.

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