

## Article

# Development of a Lactic Bacteria Starter for Amaranth Silage and Investigation of Its Influence on Silage Quality

Ekaterina Anokhina <sup>1</sup>, Svetlana Obraztsova <sup>2</sup>, Anna Tolkacheva <sup>1,\*</sup>, Dmitrii Cherenkov <sup>2</sup>, Tatyana Sviridova <sup>2</sup> and Olga Korneeva <sup>2</sup>

<sup>1</sup> Laboratory “Metagenomics and Food Biotechnologies”, Voronezh State University of Engineering Technologies, Voronezh 394000, Russia; katya\_anoh@mail.ru

<sup>2</sup> Department of Biochemistry and Biotechnology, Voronezh State University of Engineering Technologies, Voronezh 394000, Russia; sveta5501pavlenkova@yandex.ru (S.O.); d.cherenkov@mail.ru (D.C.); sviridovatv@yandex.ru (T.S.); korneeva-olgas@yandex.ru (O.K.)

\* Correspondence: anna-biotech@yandex.ru

**Abstract:** The use of amaranth green mass as an alternative source of protein and fiber for canned feed for ruminants is very promising because of several reasons, including a high concentration of crude protein and dry matter digestibility, lower water requirement compared to corn, and a high yield. The purpose of this work is to develop a silage starter from lactic acid bacteria and study its effect on the quality of fermentation and the chemical composition of the silage. The selection of strains of lactic acid bacteria in the composition of the starter was carried out, taking into account their antibacterial activity against opportunistic microorganisms *Staphylococcus aureus* and *Escherichia coli*. Amaranth was treated with the developed silage starter (MPF) before ensiling. The study of the composition of the microflora and the acidity of the silage was carried out periodically during the amaranth ensiling process and after 45 days of storage. The use of MPF starter provided a rapid decrease in pH and, on the 10th day, reached a value of 4.2. Silage with the use of MPF starter was characterized by the largest amount of lactic acid—75.1%, the lowest content of acetic acid—24.9%, and the absence of butyric acid. The use of the MPF starter compared to other treatments resulted in the retention of crude protein up to 90%, which improved the quality of amaranth silage. Amaranth silage, in comparison with corn silage traditionally used in fodder production, was characterized by an increased content of all essential amino acids; in terms of lysine content, it exceeded corn silage 2.5 times. Based on the data obtained, it can be concluded that the developed silage starter can significantly improve the quality of amaranth silage.

**Keywords:** amaranth; silage; lactic acid bacteria; starter; essential amino acids



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## 1. Introduction

The forage base and feeding play the main and decisive roles in increasing the productivity of farm animals and ensuring the growth of the competitiveness of livestock products. Feed should be balanced in protein, amino acid, carbohydrates, and microelement composition and contain a sufficient number of vitamins and other physiologically active substances.

Currently, common crops for forage are the green mass of *Zea*, *Heliánthus*, *Heliánthus tuberosus*, *Sorghum*, and *Medicágo*. However, these fodder crops contain an insufficient amount of protein and are not fully able to provide animals with a balanced feed. Thus, the introduction of new, non-traditional agricultural crops with high nutritional value into the diet of animals is relevant [1]. One of these raw materials is amaranth. The use of amaranth green mass as an alternative source of protein and fiber for canned feed for ruminants is very promising [2–4]. There are several reasons why amaranth is a promising fodder crop, including a high concentration of crude protein and dry matter digestibility [5], lower water requirement compared to corn [5], and a high yield up to 85 t/ha [6,7]. Amaranth

significantly exceeds corn in protein content [6,8], balanced in the amount of essential amino acids such as lysine, methionine, and tryptophan [9,10]. In addition, this culture contains squalene, unique in its properties, vitamin C, riboflavin, folic acid, and carotene, as well as rutin and phenolic compounds [10,11]. Amaranth contains a large amount of minerals, such as calcium, potassium, magnesium, iron, and phosphorus. Smaller amounts include copper, zinc, sodium, chromium, manganese, nickel, lead, cadmium, and cobalt [10]. For the normal process of preserving plants, the content of soluble sugars should provide the formation of lactic acid in the amount necessary for acidification to a pH level of 4.2, at which putrefactive bacteria do not grow in the feed, which ensures the safety of the feed [12,13]. The content of soluble sugars in amaranth is more than 2 times less than in corn [7]; therefore, it is difficult to obtain high quality feed from the green mass of amaranth without the use of preservatives.

One of the ways to preserve nutrients when canning green fodder is to use lactic acid bacteria. The use of biological preservatives for obtaining feed from corn and alfalfa has been studied [14–18]. However, the effect of lactic acid bacteria on amaranth fermentation and the nutritional value of silage has not been studied enough [19]. There is data on the use of *Lactobacillus plantarum* as a microbial inoculum, as well as the co-use of *L. plantarum* with propionic acid [20], molasses [19] for ensiling amaranth, as well as molasses when ensiling amaranth with rice straw [21]. However, for the preservation of hard-to-silage, high-protein vegetable raw materials, which includes amaranth, it is recommended to use starters of lactic acid bacteria [22]. Undoubtedly, the search for associations of microorganisms in the preparation of plant food remains relevant, not only providing biological preservation, but also improving the availability of nutrients and enriching the feed with nutrients, especially amino acids and vitamins. The influence of the combined action of several types of lactic acid bacteria on amaranth fermentation and the nutritional value of silage remains understudied.

The aim of this work is the development of a consortium of lactic acid bacteria for the production of high-protein amaranth silage and the study of its chemical composition.

## 2. Materials and Methods

### 2.1. Lactic Acid Bacteria Screening

The objects of this research were the following strains of lactic acid bacteria from the All-Russian Collection of Industrial Microorganisms (VKPM, Russia): *Lactobacillus plantarum* B-10816 (source: *Betonica grandiflora*), *Enterococcus hirae* B-9069 (source: *Bosmutus*), *Lactobacillus fermentum* B-10888 (source: *Betonica grandiflora*), *Lactococcus lactis* subsp. *Lactis* B-6793 (source: raw milk), *Lactococcus lactis* subsp. *Cremoris* B-3873, *Lactococcus lactis* subsp. *lactis* var *diacetyllactis* B-5600, *Leuconostoc mesenteroides* subsp. *Dextranicum* B-3425 (source: sour cream), *Streptococcus thermophilus* B-4463 (source: self-fermented milk). Pure cultures were cultivated on MRS agar (Beckton Dickinson, Franklin Lakes, NJ, USA).

Antibacterial activity was determined in relation to opportunistic bacteria. Lactic acid bacteria were incubated in a MRS broth (Beckton Dickinson, Franklin Lakes, NJ, USA) for 16 h and then centrifuged in a CR3i centrifuge (ThermoFisherScientific, Waltham, MA, USA) at 5000 rpm for 5 min. Lactic acid in supernatants was neutralized with NaOH. Opportunistic bacteria were incubated until exponential growth, and the supernatants were added at a ratio of 1:5. After 6 h of incubation, optical density was measured at a wavelength of 600 nm on a Shimadzu UV-1240 mini spectrophotometer (Kyoto, Japan). Opportunistic bacteria incubated without supernatants were positive controls for active proliferation. The decrease in optical density of the opportunistic bacteria suspension under the influence of bacteriocins of lactic acid bacteria strains relative to the control was expressed as a percentage.

The study of the ability of strains of lactic acid bacteria to co-grow and reproduce was carried out using the method of perpendicular strokes [23].

## 2.2. Cultivation of Strains of Lactic Acid Bacteria

The strains of lactic acid bacteria selected for use in the composition of starters were cultured in 10 mL of liquid MRS medium for 24 h, and then 10 mL of seed material was sterilely transferred to 100 mL of a medium of the same composition. Cultures were incubated at 37 °C under static conditions for 24 h. Then, 10 mL of the broth was transferred to 100 mL of MRS medium; after incubation under the same conditions, generation III inoculum was obtained for seeding the fermenter. The inoculum of each type of lactic acid bacteria was added into a laboratory fermenter (volume 3 L) with MRS medium in an amount of 5% of the working volume of the fermenter. Cultivation was carried out at a temperature of  $37 \pm 1$  °C, a pH of  $6.5 \pm 0.2$ , and stirring at 200 rpm for 20 h. The population of viable cells in the resulting culture liquid was assessed using the plate culture method.

## 2.3. Silo Preparation

The green mass of amaranth of the giant variety (*A. hypochondriacus*), in the phase of the milky-wax ripeness of the seeds (harvested in October 2022), was crushed to a particle size of 1–3 cm, processed from a spray bottle with a liquid bacterial starter, mixed, and tightly packed into glass jars with a volume of 3 L, with 3 kg each. The starter with a lactic acid bacteria content of  $10^8$  CFU/mL was added at the rate of 100 mL per 1.0 kg of amaranth green mass. Samples of silage were prepared with the following different processing options: with the addition of lactic acid bacteria starter MPF (1); with the addition of MLF starter (2); with the addition of industrial starter “Baktosil”: *Lactococcus lactis*, *Lactobacillus plantarum*, *Streptococcus lactis diastaticus*, *Propionibacterium shermani* (OOO PO Sibbiopharm, Novosibirsk, Russia) (3); and without a starter (4). For a comparison of the chemical composition, silage was prepared from corn (*Zea mays*) and pre-crushed to a particle size of 2–4 cm without additional processing. Each experiment was carried out in triplicate. The silos were sealed and stored at room temperature for 45 days.

## 2.4. Study of Microflora and pH during Amaranth Ensiling

The study of the composition of the microflora and changes in acidity during the ensiling of amaranth was carried out on days 3, 10, 20, 30, and 45. The jars with silage were opened and, under sterile conditions, a sample of silage was taken into sterile dishes. To determine the microflora, 1 g of silage was mixed with 9 mL of sterile water. The resulting suspension was successively diluted  $10^3$ – $10^6$  times and sown on specific dense nutrient media; after incubation, the grown colonies were counted. To determine the amount of lactic acid bacteria, yeasts and ammonifying bacteria, the MRS, Sabouraud nutrient media, and peptone water with the addition of agar were used, respectively.

To determine the pH, a 30 g sample of silage was thoroughly mixed with 50 mL of distilled water. Afterwards, it was left for 30 min at 25 °C with occasional stirring. After decantation, the pH value was measured in the silage extract (pH-meter HANNA Edge HI 2002-02, Vöhringen, Germany).

## 2.5. Determination of Quality Indicators of Amaranth Silage

Part of the ensiled material was dried at a temperature of 45 °C for 48 h, crushed, and used to determine the chemical composition. Dry matter was measured by drying to constant weight (hygrometer Kett FD-610, Tokyo, Japan). Determination of the content of crude protein, crude fat, crude fiber, and carotene was carried out using the methods described in [24]. The mass fraction of crude protein was calculated using the amount of nitrogen that was determined using the Kjeldahl method, the amount of fat was determined using a Soxhlet extraction apparatus, the content of crude fiber was determined using the Genneberg and Stomann method, and the content of carotene was determined using the photometric method.

The determination of the mass fraction of organic acids was carried out in a wet silo using the Lepper method [25], based on the volatility of vapors of acetic and butyric acids mixed with water vapor. Lactic acid was determined using its oxidation to acetic acid

and subsequent distillation. The content of acids was determined using titration of exact volumes of distillates obtained as a result of their successive distillation from the water infusion of silage.

The determination of the content of amino acids in silage was carried out using ion-exchange chromatography with post-column derivatization with ninhydrin on a Shimadzu LC-20 Prominence liquid chromatograph (Kyoto, Japan) [26].

### 2.6. Statistical Analysis

The results were analyzed using one-way analysis of variance (ANOVA) in the program “Statistica 10”. Effects were considered significant at  $p < 0.05$ . Multiple comparisons between means were made using Duncan’s test.

## 3. Results and Discussion

### 3.1. Development of a Silage Starter for Amaranth Ensiling

Lactic acid bacteria play an important role in agriculture in the conservation of feed. They are part of the epiphytic microflora of plants and are used as inoculums, one of the most important effects of which is the suppression of foreign microflora due to the release of organic acids and a decrease in the pH of the medium [27], which makes it possible to use them as biopreservatives. The effect of *Lactobacillus plantarum* on the process of ensiling amaranth green mass has been studied the most [19–21].

However, the use of a consortium of lactic acid bacteria, in which the metabolites of some microorganisms are a substrate for other microorganisms, in our opinion, is the most promising line of research in the creation of silage starters. The combined action of different types of lactic acid bacteria would improve the microbiological composition of the silage, the formation of more lactic acid, reduce the loss of nutrients, and enrich it with vitamins and biologically active substances.

The main indicators when choosing lactic acid bacteria as inoculants for a long time were the growth rate and the ability to lower the pH of the medium [28,29]. Currently, there are works on the study of biofunctional lactic acid bacteria producing various metabolites, including bacteriocins, protein substances with antibacterial activity [30]. In this regard, in this work, the main criterion for choosing lactic acid bacteria strains when compiling a consortium for amaranth ensiling was their antibacterial activity, which manifests itself in the ability to produce bacteriocins and suppress the growth of opportunistic microorganisms [31]. An analysis of antibacterial activity (Table 1) showed that cell-free extracts of strains of *Lactobacillus plantarum* B-10816, *Lactobacillus fermentum* B-10888, *Lactococcus lactis* subsp. *lactis* B-6793, *Lactococcus lactis* subsp. *lactis* var *diacetylactis* B-5600, and *Leuconostoc mesenteroides* subsp. *dextranicum* B-3425 are able to suppress the growth of opportunistic microflora by more than 30%.

**Table 1.** Ability of lactic acid microorganisms to produce bacteriocins.

Lactic Acid Bacteria Strain	Decrease in Optic Density of Test-Culture Suspension, %	
	<i>Staphylococcus aureus</i>	<i>Escherichia coli</i>
<i>Lactobacillus plantarum</i> B-10816	49	37
<i>Enterococcus hirae</i> B-9069	27	14
<i>Lactobacillus fermentum</i> B-10888	35	50
<i>Lactococcus lactis</i> subsp. <i>lactis</i> B-6793	44	32
<i>Lactococcus lactis</i> subsp. <i>cremoris</i> B-3873	8	30
<i>Lactococcus lactis</i> subsp. <i>lactis</i> var <i>diacetylactis</i> B-5600	29	44
<i>Leuconostoc mesenteroides</i> subsp. <i>dextranicum</i> B-3425	34	45
<i>Streptococcus thermophiles</i> B-4463	29	0

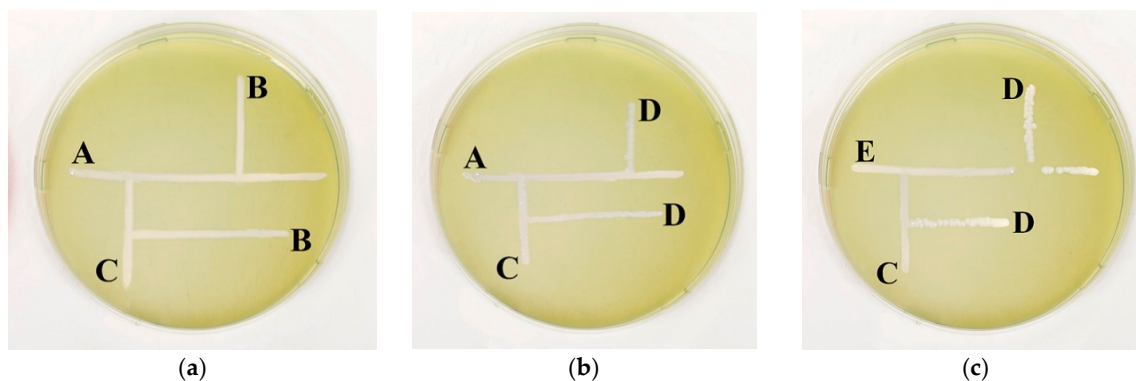
The studied strains of lactic acid bacteria have antibacterial activity against various opportunistic microorganisms, which characterizes the ability to synthesize bacteriocins as a strain-specific trait [31].

Subsequently, consortiums were formed from the most promising strains with antibacterial activity, distinctive in the optimal growth temperature and acid formation level, which ensured their consistent action during ensiling. Variants of starter cultures from selected bacterial strains are presented in Table 2.

**Table 2.** Microorganism consortia variants.

Consortium Variant No.	Starter Cultures Variants
MPF	<i>Leuc. mesenteroides</i> susp. <i>dextranicum</i> B-3425; <i>L. plantarum</i> B-10816; <i>L. fermentum</i> B-10888
MLF	<i>Leuc. mesenteroides</i> susp. <i>dextranicum</i> B-3425; <i>Lac. lactis</i> subsp. <i>lactis</i> B-6793; <i>L. fermentum</i> B-10888
LLF	<i>Lac. lactis</i> subsp. <i>lactis</i> var <i>diacetylactis</i> B-5600; <i>Lac. lactis</i> subsp. <i>lactis</i> B-6793; <i>L. fermentum</i> B-10888

The study of the ability of co-growth for lactic acid bacteria strains in starter cultures showed the absence of antagonism between lactic acid bacteria strains in microorganism consortiums MPF and MLF (Figure 1a,b). During the co-cultivation of bacteria included in consortium LLF, zones of antagonism were observed between microorganisms of the same *Lactococcus lactis* species (Figure 1c), which is probably due to the synthesis of bacteriocins that inhibit the growth of the bacteria of a homologous species. Thus, consortiums of microorganisms MPF and MLF were chosen for ensiling amaranth.

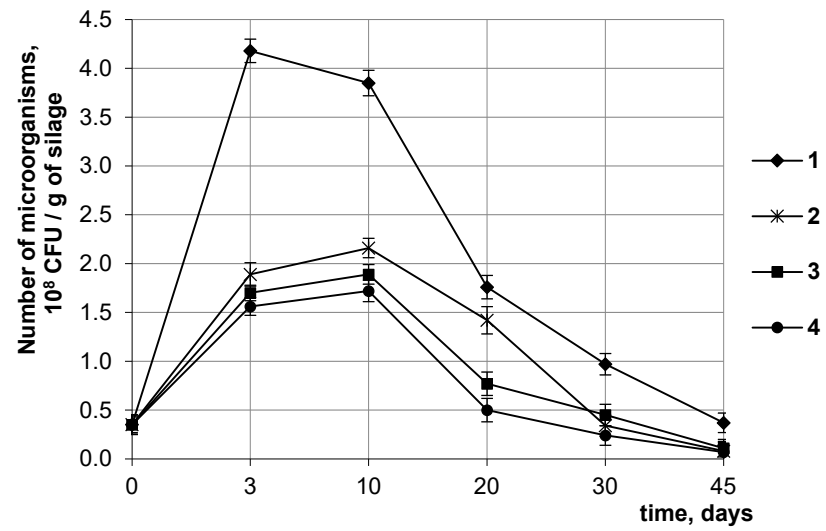


**Figure 1.** Mutual compatibility of lactic acid bacteria strains. (a) MPF: *Leuc. mesenteroides* subsp. *dextranicum* B-3425 (A); *L. plantarum* B-10816 (B); *L. fermentum* B-10888 (C). (b) MLF: *Leuc. mesenteroides* subsp. *dextranicum* B-3425 (A); *Lac. lactis* subsp. *Lactis* B-6793 (D); *L. fermentum* B-10888 (C). (c) LLF: *Lac. lactis* subsp. *Lactis* var *diacetylactis* B-5600 (E); *Lac. lactis* subsp. *Lactis* B-6793 (D); *L. fermentum* B-10888 (C).

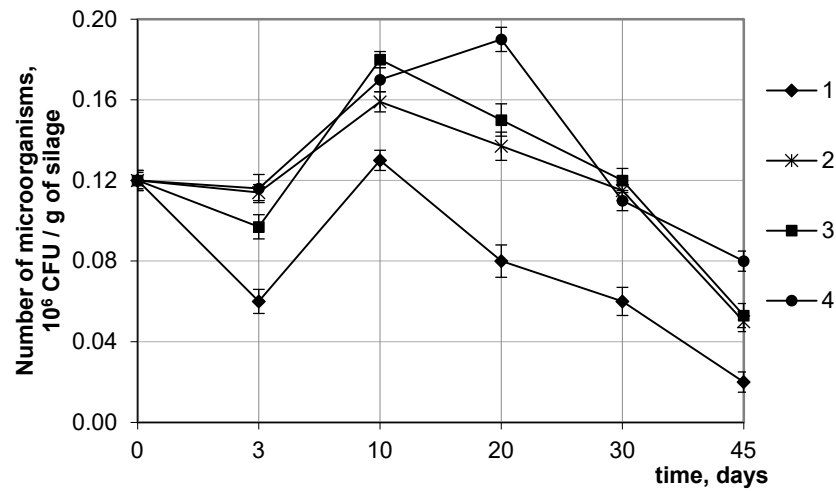
### 3.2. Study of the Microflora of Amaranth during Ensiling

Ensiling is a complex microbiological process. When ensiling, along with vegetable raw materials, a huge number of unwanted microorganisms enter the silage, which begin to grow intensively in vegetable juice. One of the main tasks of ensiling technology is to create optimal conditions for the vital activity of lactic acid bacteria and the inhibition of unwanted microflora. Identification of the features of the development of microflora and changes in its qualitative and quantitative composition in the process of ensiling largely determine the quality indicators of feed [32].

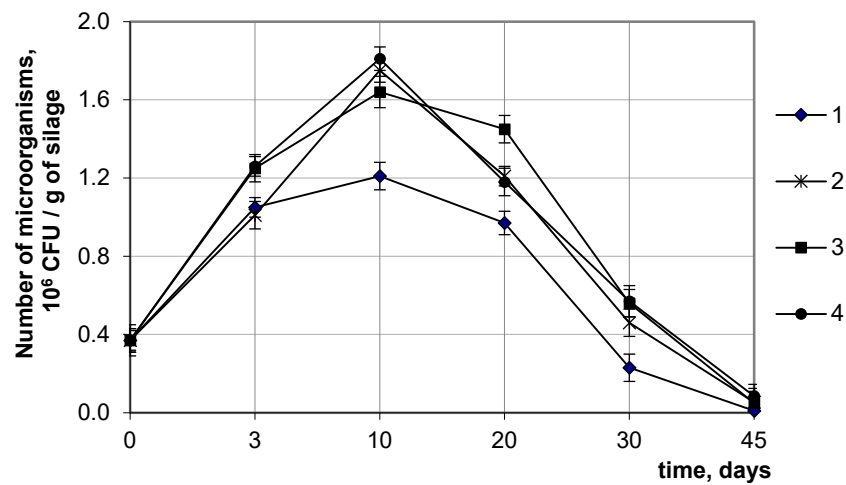
The study of the influence of the composition of experimental starter cultures of lactic acid bacteria on the silage microflora was carried out in comparison with the industrial starter culture “Baktosil”, widely used in the production of canned feed and without the addition of a starter culture. The dynamics of changes in the composition of microorganisms in the process of ensiling amaranth is shown in Figure 2.



(A)



(B)

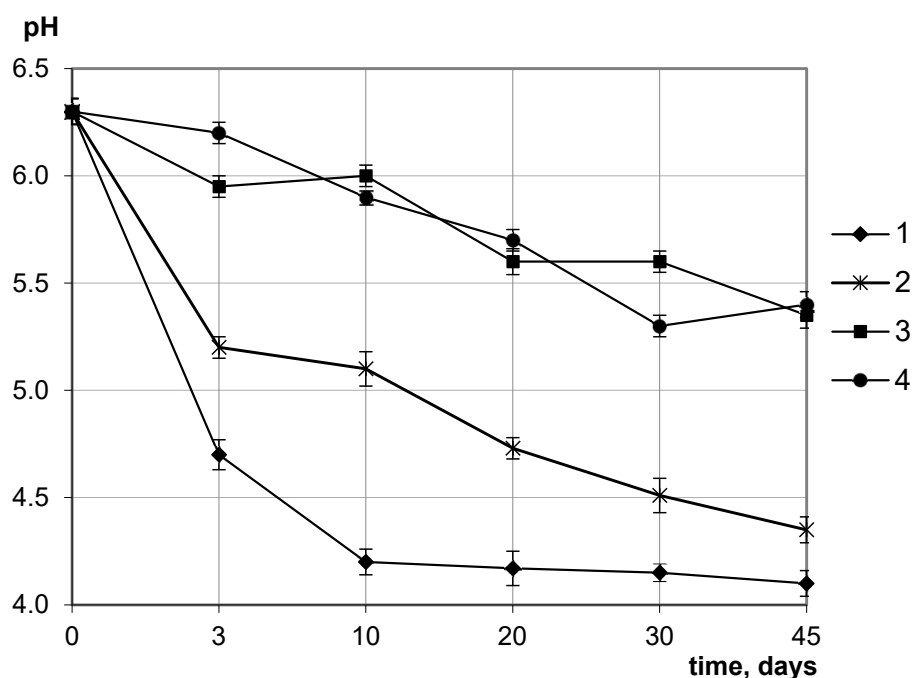


(C)

Figure 2. Dynamics of changes in the composition of the microflora in the process of amaranth ensiling. (A) lactic acid bacteria; (B) ammonifying bacteria; (C) yeast. 1—MPF starter, 2—MLF starter, 3—industrial starter culture “Baktosil”, 4—without starter culture.

The highest content of lactic acid bacteria (Figure 2A) throughout the entire ensiling process was in the silage sample using MPF (1) ( $p < 0.05$ ). The maximum amount of LAB was on the 3rd day ( $4.2 \times 10^8$  CFU/g), which is probably due to the symbiotic relationship of microorganisms in the composition of the MPF starter, in which the resulting metabolites of some bacteria prepare a favorable environment for the growth of others. The content of lactic acid bacteria in amaranth with MLF starter (2), in the period from 10 to 20 days of ensiling, differed from the silage obtained using the industrial starter (3) and without a starter (4) ( $p < 0.05$ ); however, by 45 days, significant differences between treatment options 2, 3, and 4 were not observed. Amaranth with MPF is characterized by the lowest level of ammonifying bacteria during 45 days of ensiling relative to other variants ( $p < 0.05$ ); a significant decrease in their number was noted in the first 3 days, probably due to the synthesis of bacteriocins with antibacterial activity (Figure 2B). The highest content of ammonifying bacteria in the silage was reached on the 10th day; their number significantly differed for different treatment options ( $p < 0.05$ ). However, in silage without a starter, an increase in the number of putrefactive bacteria was observed for 20 days. This may indicate insufficient acidification of the medium and the process of protein decomposition. In all silage samples in the first 10 days, there was a tendency for a sharp increase in the content of yeast microflora, which does not contradict the well-known fact of acid tolerance in most yeast cultures; by the end of silage, the amount of yeast decreased to a minimum (Figure 2C). The smallest amount of yeast was in the amaranth with MPF starter, which significantly differed from other treatment options starting from the 10th day of ensiling ( $p < 0.05$ ).

The change in acidity during ensiling amaranth is shown in Figure 3. After 45 days in untreated silage and silage with an industrial starter, the pH values did not differ significantly and were at the level of 5.4. At the same time, the pH in silage with MPF and MLF starter cultures differed significantly ( $p < 0.05$ ) and was at the level of 4.1 and 4.4, respectively. However, it should be noted that when using MPF, the pH reached a value of 4.2 on the 10th day, at which the growth of opportunistic bacteria was effectively suppressed, ensuring high-quality fermentation with minimal loss of nutrients [18]; on the 45th day, the pH was 4.11. In a past study [20], a pH value of 4.16 was only achieved by 60 days of ensiling with a single culture of *Lactobacillus plantarum*.



**Figure 3.** Dynamics of pH change during amaranth ensiling with different treatments. 1—MPF starter, 2—MLF starter, 3—industrial starter culture “Baktosil”, 4—without starter culture.

### 3.3. Influence of the Composition of Starter Cultures on the Quality of Amaranth Silage

The main indicator of silage quality is the ratio of organic acids in its composition. Along with lactic acid, lactic acid fermentation can produce acetic, propionic, and butyric acids, which reduce the quality of silage. In particular, the formation of butyric acid leads to a significant loss of dry matter [32]. The influence of the composition of the consortium of lactic acid bacteria on the accumulation of organic acids in amaranth silage is shown in Table 3.

**Table 3.** The content of organic acids in the silage.

Organic Acid	Mass Fraction of the Total Acid Content, %				SEM	p-Value
	Starter					
	MPF	MLF	Baktosil	Without Treatment		
Lactic acid	75.1 ± 0.8 <sup>a</sup>	65.4 ± 0.7 <sup>b</sup>	57.6 ± 0.8 <sup>c</sup>	29.1 ± 0.8 <sup>d</sup>	0.3	<0.0002
Acetic acid	24.9 ± 0.6 <sup>d</sup>	34.5 ± 0.6 <sup>c</sup>	42.2 ± 0.7 <sup>b</sup>	70.3 ± 0.6 <sup>a</sup>	0.2	<0.0002
Butyric acid	0	0.1 ± 0.08 <sup>c</sup>	0.2 ± 0.07 <sup>b</sup>	0.6 ± 0.07 <sup>a</sup>	0.03	<0.0476

Different lowercase letters indicate significant differences among different treatments ( $p < 0.05$ ); same letter indicates not significant ( $p > 0.05$ ).

The silage with the use of MPF starter was characterized by the largest amount of lactic acid—75.1%, the lowest content of acetic acid—24.9%, and the absence of butyric acid, which is comparable with the results of other authors [19,20]. In silage with the use of industrial starter “Baktosil” and without treatment, the content of lactic acid was 57.6% and 29.1%, respectively. In addition, butyric acid was found in these silage samples, which indicates the development of *Bacillus* spp., *Clostridium* spp., and *Enterobacter* spp. microorganisms in the silage microflora, which are involved in the decomposition of protein substances [33], and a rather high content of acetic acid, which reduces its nutritional value.

The content of nutrients in samples of silage with the introduction of starter cultures is presented in Table 4. For comparison, data are given for corn silage, traditionally used in cattle feeding.

**Table 4.** Main nutrient content (% of DM) in amaranth and corn silages.

Indicators	Raw Material	Amaranth Silage				Corn Silage	SEM	p-Value
		Starters						
		MPF	MLF	Baktosil	Without Treatment			
Crude protein, %	15.5 ± 0.2 <sup>a</sup>	13.9 ± 0.3 <sup>b</sup>	10.4 ± 0.3 <sup>c</sup>	8.1 ± 0.2 <sup>d</sup>	6.3 ± 0.3 <sup>e</sup>	6.3 ± 0.2 <sup>e</sup>	0.14	<0.0002
Crude fat, %	4.3 ± 0.3 <sup>a</sup>	4.0 ± 0.2 <sup>a</sup>	3.6 ± 0.3 <sup>b</sup>	3.3 ± 0.3 <sup>b</sup>	2.9 ± 0.4 <sup>b</sup>	0.9 ± 0.3 <sup>c</sup>	0.18	<0.0270
Crude fiber, %	21.3 ± 0.4 <sup>a</sup>	20.4 ± 0.2 <sup>a</sup>	21.1 ± 0.4 <sup>a</sup>	20.8 ± 0.4 <sup>a</sup>	20.7 ± 0.2 <sup>a</sup>	23.6 ± 0.4 <sup>b</sup>	0.17	<0.0002
Carotene, mg/kg	24.9 ± 0.3 <sup>a</sup>	24.1 ± 0.3 <sup>b</sup>	22.9 ± 0.2 <sup>c</sup>	21.8 ± 0.3 <sup>d</sup>	20.1 ± 0.2 <sup>e</sup>	19.3 ± 0.4 <sup>f</sup>	0.15	<0.0026

Different lowercase letters indicate significant differences among different treatments ( $p < 0.05$ ); same letter indicates not significant ( $p > 0.05$ ).

The raw green mass of amaranth in the phase of milky-wax ripeness was characterized by a higher content of DM (26.6%) and crude protein (15.5%), which favorably distinguished this amaranth variety from others [20]. The use of MPF starter compared to other treatments resulted in the retention of crude protein up to 90%. In terms of crude protein content, this sample of silage exceeded the silage obtained using the “Baktosil” starter culture and silage without treatment by 1.7 and 2 times, respectively. In addition, in this silage, the highest content of carotene was noted—24.1 mg/kg. In terms of the content of crude protein,



fat, and carotene, amaranth silage obtained using biological preservatives significantly exceeded corn silage ( $p < 0.05$ ).

Given the above, it is advisable to use the MPF starter for amaranth ensiling, which allows you to obtain high-quality, high-protein silage.

The efficiency of protein use as the most expensive and deficient feed nutrient is determined by the level and ratio of amino acids, primarily the essential ones. For dairy cows, the most important ones are lysine and methionine, which must be continuously supplied to the nutrition of animals with feed. In this regard, we conducted a comparative study of the content of amino acids in amaranth silage obtained using MPF starter culture and corn silage traditionally used in feeding cattle (Table 5).

**Table 5.** The content of essential amino acids in amaranth and corn silage.

No.	Amino Acids	Mass Rate in Silage, %	
		Amaranth Silage	Corn Silage
1	valine	0.73	0.37
2	methonine	0.11	0.09
3	isoleucine	0.61	0.26
4	leucine	0.97	0.70
5	phenylalanine	0.63	0.30
6	lysine	0.72	0.27
7	threonine	0.54	0.26
8	tryptophan	0.09	0.04
Sum of essential amino acids		4.40	2.32

According to Table 5, amaranth silage, in comparison with corn silage, is characterized by a high content of all essential amino acids and exceeds corn silage in lysine content by 2.5 times.

#### 4. Conclusions

In this work, the amaranth of the giant variety, which differs from many known varieties in its high content of dry matters and crude protein, was used to obtain high-protein silage. For ensiling amaranth, a starter from lactic acid bacteria (*Leuc. mesenteroides* spp. *dextranicum* B-3425, *L. plantarum* B-10816, and *L. fermentum* B-10888) has been developed. Used bacteria exhibit antibacterial activity against the opportunistic microorganisms *Staphylococcus aureus* and *Escherichia coli*, which characterizes their ability to synthesize bacteriocins that inhibit the growth of foreign microflora. The use of this starter provided a decrease in pH to 4.2 in the silage on the 10th day of ensiling, as well as a significant growth of lactic acid bacteria and the suppression of the growth of ammonifying bacteria and yeast. An analysis of the composition of organic acids in amaranth silage showed a high content of lactic acid with a relatively small amount of acetic acid and the absence of butyric acid, which is typical for high-quality silage. In addition, the preservation of crude protein in amaranth silage was 90%. Compared to corn silage traditionally used in feeding cows, it is characterized by a balanced amino acid composition, an increased content of all essential amino acids, and a lysine content exceeding corn silage by 2.5 times. Based on the data obtained, it can be concluded that the developed silage starter can significantly improve the quality of amaranth silage.

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