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Enhancing Nutritional Value and Production Efficiency of Feeds Through Biochemical Composition Optimization

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Abstract: The necessity to optimize feed crop cultivation in Kazakhstan's steppe zone is underscored by evolving climatic conditions and sustainable agriculture demands. This study, conducted from 2021 to 2023 in the Akmola region, evaluated the nutritional value and production efficiency of annual and perennial grass mixtures. A randomized complete block design was utilized for annual grasses, while a sequential scheme was applied for perennial grasses, each with three replications per plot. Statistical data processing was employed to analyze the outcomes. Results indicated that mixed-feed crops exhibited superior nutritional composition and energy value. Specific combinations of annual grasses, such as oats with peas or oats, peas, and vetch, alongside multicomponent mixtures incorporating legumes for perennial grasses, demonstrated optimal results. The ideal harvest timings for these grass mixtures were also established. It is recommended to cultivate combinations like Sudan grass with peas and vetch, or oats with peas and vetch for green feed, and for hay and pasture, combinations of red fescue, bluegrass, wheatgrass, and alfalfa, as well as red fescue, bluegrass, brome, sainfoin, and alfalfa. This research emphasizes the importance of diverse crop mixtures to enhance feed nutritional value, thereby contributing to sustainable agricultural practices, food security, and environmental resilience amid climate change.

Keywords: Annual feed grasses; Perennial feed grasses; Mixed sowing; Grass mixtures; Feed nutritional value; Chemical composition; Amino acid composition; Metabolizable energy

1. Introduction

According to the Organisation for Economic Co-operation and Development (OECD) and Food and Agriculture Organization (FAO) Agricultural Forecast for 2023-2032, global agricultural and food production will continue to grow over the next ten years. Since food remains the main use of crops, which currently account for 49% of global consumption, the importance of feed use has increased in recent decades. The growth of global livestock production has led to the need for significantly greater use of crops as feed, which currently account for 26% of global consumption. The main food products are milk and meat (OECD & FAO, 2023).

Global demand for meat and dairy products has increased dramatically in recent decades, and it will inevitably continue to grow due to a combination of global population growth, increased life expectancy, and improved economic well-being. Meat is an energy-intensive source of high-quality protein and is enriched with trace elements such as thiamine, niacin, vitamin B12, calcium, iron, zinc, potassium, and phosphorus (Kliem & Givens, 2011). Milk and dairy products are also important sources of protein and make a significant contribution to the intake of calcium, phosphorus, iron, vitamin A, and riboflavin (Salter, 2017). Today, the growth of economic well-being, population growth, and an increase in life expectancy contribute to an increase in demand for meat and milk. As the population grows and demographic changes take place, the resulting increase in demand for beef and milk requires an increase in animal productivity. The productivity of animals is influenced by numerous factors, such as genetic factors, environmental factors, and feeding. The types and quality of feed and feed additives determine the characteristics and quality of animal products (milk and meat). In terms of quality and quantity, the effect of high productivity can be achieved only when the diet fully meets the needs of the animal. To obtain good physicochemical parameters of milk, it is necessary to use high-quality feeds that meet the needs of animals for protein and energy and maintain a properly balanced mineral and vitamin content in the diet (Pecka-Kiełb et al., 2018).

The realization of the genetic potential of animals' dairy productivity largely depends on the feed base, which consists of annual and perennial feed crops such as oats, peas, vetch, Sudan grass, awnless brome, fescue, alfalfa, and sainfoin. An increase in the productivity of these annual and perennial feed crops and their resistance to abiotic factors can be achieved by cultivating them in grass mixtures (Shen et al., 2013; Zhang et al., 2010). Grass mixtures have an advantage over single-species crops since they make much better use of environmental factors and their components absorb sunlight more efficiently. They suffer less from weeds and persist longer, and the feeds made from them are balanced in nutrients (White et al., 2013). Grass mixtures increase phytoavailability and the acquisition of limited resources, and managing the interaction of the root and rhizosphere can increase the efficiency of resource use by crops. Signaling cascades of phytohormones regulating plant development are also activated. In mixed crops, unlike in single-crop sowings, the activity of a wide range of enzymes increases significantly. This complex molecular bond between species stimulates plant growth (Li et al., 2014; Monteiro et al., 2012; Zhou et al., 2011).

Feeds made from grass mixtures contain more proteins, carotenes, and energy and are optimal for animal consumption, as a variety of plant species has a positive effect on biomass and nutritional value. Studies have shown that legumes in combination with cereals, due to the influence of species diversity, increase the nutritional properties of feed (Tahir et al., 2022). Mixed sowing of oats with vetch demonstrates an increase in feed yield of 13.4-202.8%, which is statistically significant ($p<0.05$). In addition, the yield of crude protein (CP) increases by 52.5-150.1% compared with single-crop sowings ($p<0.05$) (Wang et al., 2022). Mixed sowings of alfalfa with cereals also increase the CP yield (2.5 t/ha), while the yield of fiber decreases from 4.6 to 2.8 t/ha (Tahir et al., 2022).

Current research has demonstrated the benefits of using mixed grass crops over monocultures (Nasiyev et al., 2023). Studies have shown that grass mixtures can lead to higher yields, better nutrient use efficiency, and improved resistance to environmental stresses (Kurmangozhinov et al., 2024). For instance, combining legumes with cereals was found to increase the nutritional properties of feed, including higher CP yields and better amino acid balance (Kradetskaya et al., 2024). However, there are gaps in the research concerning the optimal combinations of grass species for specific climatic and soil conditions, particularly in the steppe zones of Kazakhstan.

While some studies have explored the benefits of grass mixtures in general (Ainebekova et al., 2023), there is a lack of detailed research focusing on the specific combinations of annual and perennial grasses that would be most effective in the steppe regions of Kazakhstan. Thus, the purpose of the study is to evaluate the nutritional value of annual and perennial grass mixtures depending on the crop composition. This evaluation includes assessing the biochemical composition, amino acid composition, and metabolizable energy (ME) of different grass mixtures. The following research questions are stated:

- Do mixed-grass crops have a higher nutritional value compared to monocultures?
- Which specific combinations of annual and perennial grasses optimize the nutritional value of feed?
- What are the optimal cutting moments for different types of grass mixtures to maximize their nutritional benefits?

2. Methodology

The study was carried out in 2021-2023 in the steppe zone of the Akmola region (52°N, 69°E), which is located in the north-central part of the Republic of Kazakhstan. It borders the Kostanay region in the west, the North Kazakhstan region in the north, the Pavlodar region in the east, and the Karaganda region in the south. The terrain of the territory is diverse: most of it is occupied by steppes, hillocky areas, poorly broken valleys, river valleys in the lowland, and mountains covered with forests. The climate of the region is continental. The summers are short and warm, while the winters are long and frosty, with strong winds and blizzards. The minimum air temperature is more than -40°C, and the maximum reaches +44°C.

2.1 Study Variants

The main objects of the study are perennial and annual grass mixtures. By composition, grass mixture combinations consist of such crops as oats (*Avéna satíva*), peas (*Pisum sativum*), vetch (*Vicia sativa*), Sudan grass (*Sorghum sudanense*), alfalfa (*Medicágo satíva*), sainfoin (*Onobrychis viciifolia*), brome (*Bromopsis inermis*), red fescue (*Festuca arundinacea*), bluegrass, and Russian wildrye. Two field experiments were established (Table 1).

Table 1. Experiment design

Experiment	Grass Mixture	Variants
No. 1	Annual grasses	Oats
		Vetch
		Peas
		Oats and vetch
		Vetch and Sudan grass
		Oats and peas
		Peas and Sudan grass
		Sudan grass, peas, and vetch
		Oats, peas, and vetch
No. 2	Perennial grasses	Awnless brome
		Red fescue, bluegrass, Russian wildrye, and sainfoin
		Red fescue, bluegrass, Russian wildrye, alfalfa, and sainfoin
		Red fescue, bluegrass, awnless brome, and alfalfa
		Red fescue, bluegrass, awnless brome, alfalfa, and sainfoin
		Red fescue, bluegrass, wheatgrass, and alfalfa

2.2 Research Process

The first experiment with annual grasses was conducted according to a randomized complete block scheme. The repeatability of the experiment was three times, and the size of the plot was 1.8×25 = 45 m^2. The second experiment with perennial grasses was carried out according to a sequential scheme. The repeatability of the experiment was three times, and the size of the plot was 15×15 = 225 m^2.

Sowing of annual grasses (experiment No. 1) was carried out in the second decade of July, with summer sowing to a depth of 3-7 cm with a SZ-1.8 seeding machine (manufactured in Ukraine by BM Systems). Sowing of perennial grasses (experiment No. 2) was carried out in the first decade of May, with spring sowing to a depth of 3-5 cm with a CH-16 mounted seeder (manufactured in the Republic of Belarus by Agro-Resource). After sowing, rolling was performed with a 3KKSH-6 roller (manufactured in Russia by Agromash).

The technology of annual and perennial crop cultivation recommended for the hill-plain zone of northern Kazakhstan was used in this study. The cutting of grasses was carried out in the earing phase of cereals and at the beginning of the flowering of legumes.

The yield of the green mass was determined by the cutting method, followed by weighing. In each variant, the grass was cut from four accounting sites with a size of 2.5 m^2 each (1.0×2.5 m) on replications. Samples from all replications of each variant were mixed, and an average weight of 1.0 kg was taken.

Chemical analysis of plant samples was carried out in accredited laboratories of AgroComplexExpert LLP (Zhaksy) and A.I. Baraev Research and Production Centre for Grain Farming (NPTsZKh) LLP.

The ME content in the dry matter of the feed was calculated from the chemical composition of the feed using the following regression equation:

$$ME = 10.678 + 0.088 \times CP - 0.332 \times CFa - 0.075 \times CFi + 0.006 \times NFE \quad (1)$$

where, *ME* is the metabolizable energy, *CP* is the crude protein, *CFa* is the crude fat, *CFi* is the crude fiber, and *NFE* is the nitrogen-free extractives.

The mass fraction of protein was determined by calculation using a coefficient of 6.25, crude ash (CA), crude fiber (CFi), crude fat (CFa), based on the total nitrogen content in the analyzed sample, and CP (Baidalin et al., 2017; Baidalina et al., 2023).

The mass fraction of amino acids was determined by capillary electrophoresis using the Kapel 105M system. The technique establishes the procedure for determining the following amino acids: arginine, lysine, tyrosine, phenylalanine, histidine, leucine and isoleucine (in total), methionine, valine, proline, threonine, serine, alanine, and glycine. The methodology provides for the analysis of three weighed samples, which are hereinafter referred to as weighed samples No. 1, 2, and 3. The course of the sample analysis differs depending on the sample preparation procedure, the conditions of electrophoretic determination, and the list of amino acids to be determined. All amino acids were determined after modification with phenylisothiocyanate in the form of phenylisothiocarbamyl (PTC) derivatives.

In weighed sample No. 1, arginine, lysine, tyrosine, phenylalanine, histidine, the sum of leucine and isoleucine, methionine, valine, proline, threonine, serine, alanine, and glycine were determined at a temperature of 30℃. The order of enumeration corresponds to the order of the peak output on the electrophoregram. In weighed sample No. 2, aspartic and glutamic acids, as well as cystine (after preliminary oxidation to cysteine acid), were determined in the form of PTC derivatives. Electrophoretic separation was carried out in an accelerated mode at a temperature of 30℃. In weighed sample No. 3, tryptophan was determined in the form of a fluorescein isothiocyanate (FTC) derivative after alkaline hydrolysis. After neutralization and removal of excess barium hydroxide, the hydrolysate was diluted in an aliquot portion, and a PTC derivative of tryptophan was obtained, which was analyzed by capillary electrophoresis in a borate buffer solution at a temperature of 50°C under a pressure of 30 mbar.

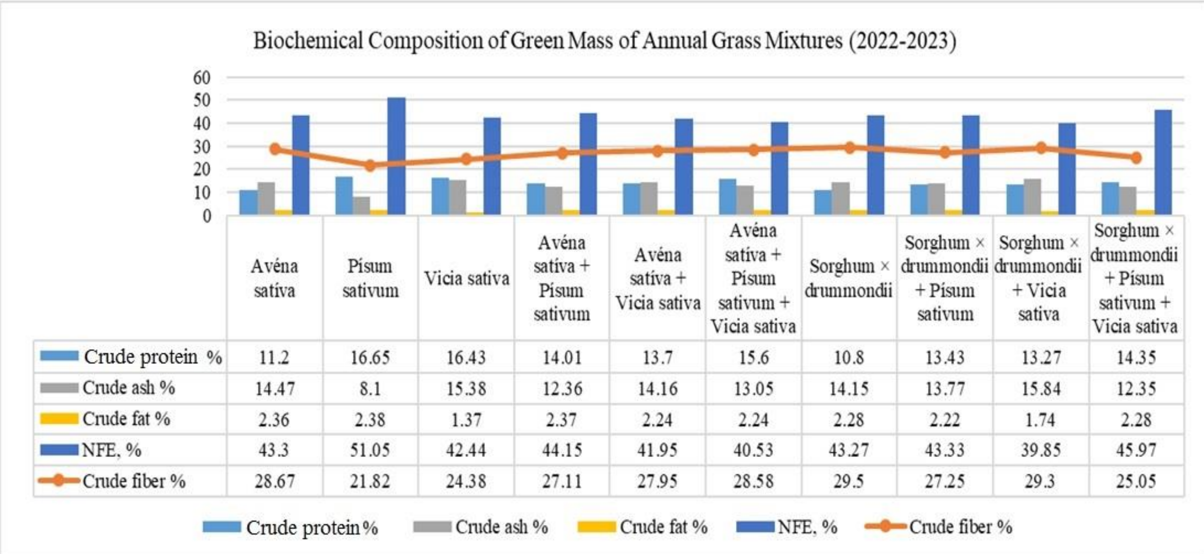
2.3 Statistical Analysis

The SNEDECOR software package was used for the statistical processing of experimental data. All measured parameters (e.g., CP content, fiber content, ME) were recorded for each plot and replication. The least significant difference (LSD) was determined between the variants at a probability level of 0.05 to compare the average values. The tables were created using Excel.

3. Results

The analysis of the biochemical composition of feed crops makes it possible to optimize the diet of animals, providing them with the necessary nutrients for health and productivity, helping to reduce feeding costs, and improving production results.

The biochemical composition of grass mixtures depends on the species composition of plants and their vegetation phase during harvesting. In the phase of the end of stem extension and the beginning of ear formation in cereal grasses, the highest level of yield of green mass is observed, while legume feed grasses in the budding phase and the beginning of flowering have the highest protein content. The annual grass mixtures were cut in the earing phase of cereals and at the beginning of the flowering of legumes. Data on the biochemical composition of grass mixtures are shown in Figure 1.



	Avéna sativa	Pisum sativum	Vicia sativa	Avéna sativa + Pisum sativum	Avéna sativa + Vicia sativa	Avéna sativa + Pisum sativum + Vicia sativa	Sorghum × drummondii	Sorghum × drummondii + Pisum sativum	Sorghum × drummondii + Vicia sativa	Sorghum × drummondii + Pisum sativum + Vicia sativa
Crude protein %	11.2	16.65	16.43	14.01	13.7	15.6	10.8	13.43	13.27	14.35
Crude ash %	14.47	8.1	15.38	12.36	14.16	13.05	14.15	13.77	15.84	12.35
Crude fat %	2.36	2.38	1.37	2.37	2.24	2.24	2.28	2.22	1.74	2.28
NFE, %	43.3	51.05	42.44	44.15	41.95	40.53	43.27	43.33	39.85	45.97
Crude fiber %	28.67	21.82	24.38	27.11	27.95	28.58	29.5	27.25	29.3	25.05

Figure 1. Biochemical composition of the green mass of annual grass mixtures (2022-2023)
Source: This figure was prepared by the authors.

The average values of the biochemical composition of annual crops in 2022-2023 demonstrate that the single-crop sowing of peas has the highest CP content (16.65%), surpassing oats and vetch, and the lowest protein content is observed in the single-crop sowing of Sudan grass (10.8%). In terms of protein content, the three-component grass mixtures of oats, peas, and vetch and Sudan grass, peas, and vetch, containing 15.6% and 14.35%, respectively, have the highest values. The lowest CFi content is observed in single-crop sowing of peas (21.82%), as well as in a grass mixture of Sudan grass, peas, and vetch (25.05%).

According to the CA content, the two-component grass mixture (Sudan grass and vetch) has the highest values (15.84%), and the single-crop sowing of vetch has a high ash content (15.38%), while peas have the lowest ash content (8.1%). In terms of CFa content, the highest value is observed in single-crop sowing of peas (2.38%), as well as in the oat and pea grass mixture (2.37%), and the lowest CFa content is observed in single-crop sowing of vetch (1.37%). According to the content of nitrogen-free extractives (NFE), peas (51.05%) and the three-component grass mixture (Sudan grass, peas, and vetch) (45.97%) demonstrate the highest content, while the two-component grass mixture (oats and vetch) has the lowest NFE content (41.95%).

Perennial grasses are rich in protein, vitamins, minerals, and fiber, which makes them a valuable source of nutrients for animals. Figure 2 shows the biochemical composition of perennial grass mixtures. The perennial grass mixtures were cut in the earing phase of cereals and at the beginning of the flowering of legumes in the second and third years of life.

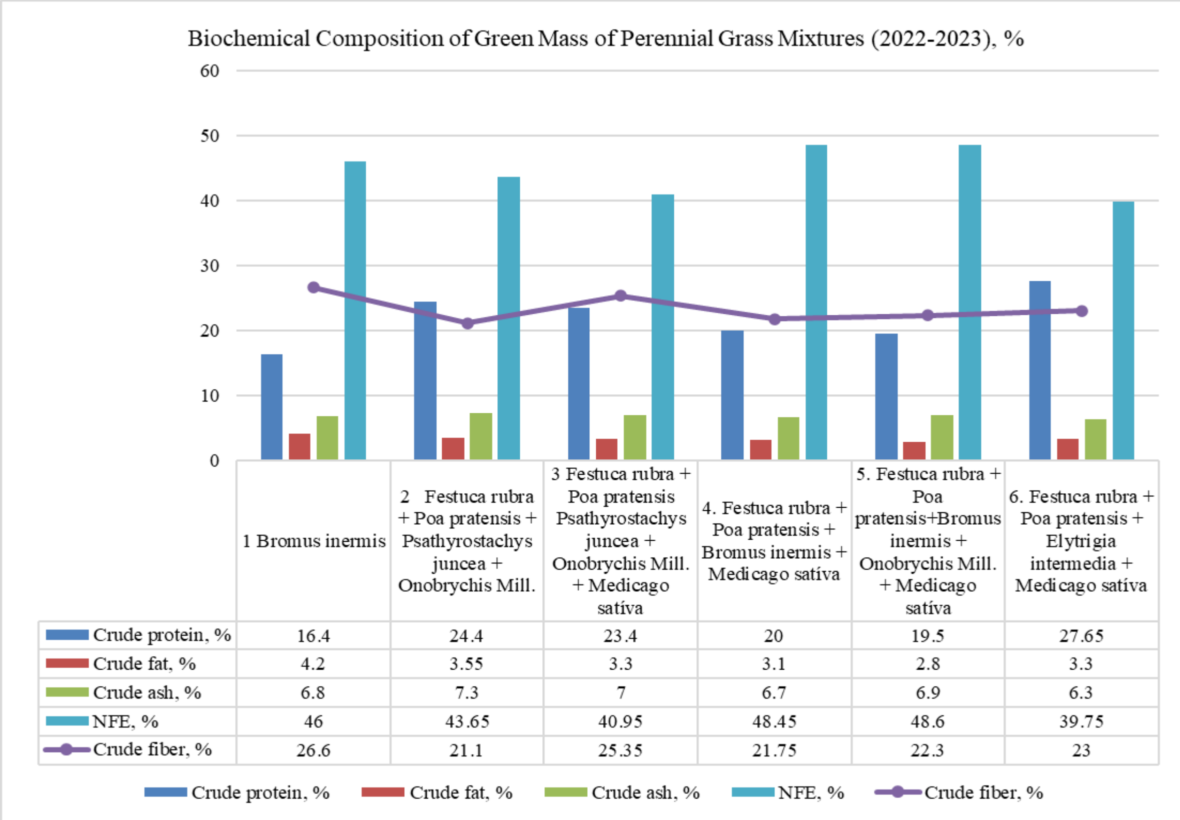


Figure 2. Biochemical composition of the green mass of perennial grass mixtures (2022-2023)
Source: This figure was prepared by the authors.

The data from the biochemical analysis of perennial grass mixtures in terms of protein content shows the highest content in four-component grass mixtures of red fescue, bluegrass, wheatgrass, and alfalfa (27.65%), while single-crop sowing of awnless brome shows the lowest protein content. The fiber content in the four-component grass mixture of red fescue, bluegrass, Russian wildrye, and sainfoin is the lowest (21.1%), while the largest proportion of fiber is observed in the single-crop sowing of awnless brome. By the CFa content, the highest value is observed in single-crop sowing of brome (4.2%) and the lowest content in a five-component grass mixture of red fescue, bluegrass, brome, sainfoin, and alfalfa (2.8%). By CA content, the four-component grass mixture (red fescue, bluegrass, Russian wildrye, and sainfoin) has the highest value (7.3%), and the lowest value is observed in the grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (6.3%). By NFE content, the highest value is demonstrated by a five-component grass mixture of red fescue, bluegrass, brome, sainfoin, and alfalfa (48.55%), while the lowest NFE index is observed in a four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (39.75%).

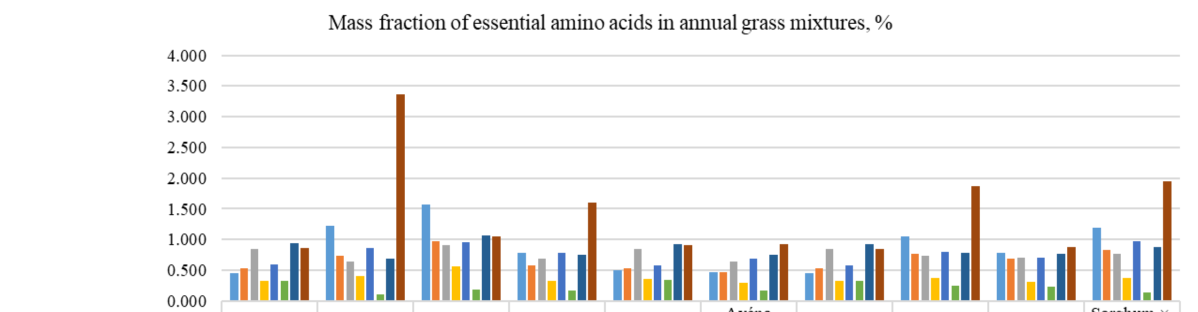
Table 2. Energy nutrition value of annual and perennial grass mixtures

Annual Grasses		Perennial Grasses	
Variants	ME (MJ)	Variants	ME (MJ)
Oats	9.59	Awnless brome	5.02
Peas	9.98	Red fescue, bluegrass, Russian wildrye, and sainfoin	7.75
Vetch	9.83	Red fescue, bluegrass, Russian wildrye, sainfoin, and alfalfa	6.035
Oats and peas	9.68	Red fescue, bluegrass, brome, and alfalfa	7.375
Oats and vetch	9.65	Red fescue, bluegrass, brome, sainfoin, and alfalfa	7.29
Oats, peas, and vetch	9.53		
Sudan grass	9.51		
Sudan grass and peas	9.67	Red fescue, bluegrass, wheatgrass, and alfalfa	7.98
Sudan grass and vetch	9.56		
Sudan grass, peas, and vetch	9.83		

The ME indicator is necessary to determine the nutritional value of feed and plays a key role in animal nutrition, determining the productivity and health of the animals, as well as the effectiveness of feed use in meeting their energy needs. The energy content depends on the composition of crops and the proportion of legumes. Data on the ME of annual and perennial grasses are presented in Table 2.

The highest ME indicator in annual crops is demonstrated by single-crop sowing of peas (9.98 MJ), and the oat-pea grass mixture has 9.68 MJ ME. The combination of Sudan grass with peas and vetch also increases the energy value of the feed, reaching 9.83 MJ. The lowest ME indicator is observed in the single-crop sowing of Sudan grass (9.51 MJ). The analysis of the ME of perennial grasses shows the highest value in a four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (7.98 MJ) and the lowest indicator in the single-crop sowing of awnless brome (5.02 MJ).

The protein nutritional value of feeds and diets, estimated by the amount of digested protein without taking into account its quality due to the content and ratio of essential amino acids, significantly reduces the effectiveness of feeding. The biological role of essential amino acids is determined by the fact that they are included in all the most important proteins of the animal body but are not synthesized and replaced by other amino acids in the body, since they must enter the body with food. A deficiency of one or more of them in the diet adversely affects animals. Eight amino acids (valine, isoleucine, leucine, lysine, methionine, threonine, tryptophan, and phenylalanine) are essential, and in the absence of at least one of them, protein synthesis, as well as protein substance synthesis, is impossible. The balance of amino acids plays a crucial role in many physiological processes in the animal body. In this study, 14 proteinogenic amino acids were investigated, of which nine were essential and five were non-essential amino acids. The composition and mass fraction of essential amino acids in annual grass mixtures are shown in Figure 3.



	Avéna satíva	Písum satívum	Vicia sativa	Avéna satíva + Písum satívum	Avéna satíva + Vicia sativa	Avéna satíva + Písum satívum + Vicia sativa	Sorghum × drummondii	Sorghum × drummondii + Písum satívum	Sorghum × drummondii + Vicia sativa	Sorghum × drummondii + Písum satívum + Vicia sativa
Arginine	0.452	1.220	1.572	0.789	0.498	0.465	0.445	1.050	0.779	1.194
Lysine	0.532	0.742	0.973	0.575	0.535	0.471	0.525	0.775	0.690	0.827
Phenylalanine	0.852	0.640	0.906	0.684	0.848	0.634	0.845	0.732	0.709	0.769
Histidine	0.331	0.398	0.564	0.322	0.361	0.301	0.324	0.368	0.311	0.380
Leucine and isoleucine	0.589	0.869	0.960	0.778	0.573	0.682	0.582	0.799	0.711	0.973
Methionine	0.331	0.108	0.190	0.177	0.340	0.173	0.324	0.252	0.235	0.145
Valine	0.937	0.686	1.072	0.754	0.918	0.748	0.930	0.780	0.769	0.871
Threonine	0.858	3.357	1.049	1.601	0.903	0.921	0.851	1.867	0.873	1.945

Arginine Lysine Phenylalanine Histidine Leucine and isoleucine Methionine Valine Threonine

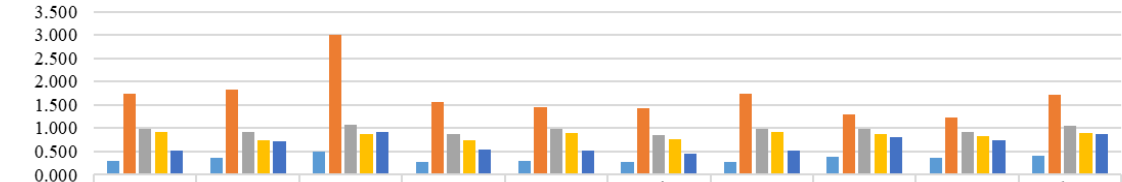
Figure 3. The mass fraction of essential amino acids in annual grass mixtures
Source: This figure was prepared by the authors.

According to the data on the amino acid composition of annual crops, the three-component grass mixture of Sudan grass, peas, and vetch has the highest arginine content with a level of 1.194%, while the minimum content of this amino acid is noted in the single-crop sowing of oats with a level of 0.452%. The three-component grass mixture (Sudan grass, peas, and vetch) has the highest lysine content (0.827%), while the minimum lysine content is observed in the grass mixture of oats and peas (0.471%). Phenylalanine is of the greatest importance in the composition of the three-component grass mixture (Sudan grass, peas, and vetch) with a level of 0.769%, while the lowest value of this amino acid is noted in the two-component grass mixture of oats and peas with a level of 0.634%. Histidine has the highest value in the three-component grass mixture of Sudan grass, peas, and vetch (0.380%), while its minimum content is noted in the grass mixture of oats and peas (0.301%).

The two-component grass mixture of Sudan grass and peas is characterized by the highest content of leucine and isoleucine with a level of 0.799%, and the minimum content of this amino acid is noted in the two-component grass mixture of oats and peas with a level of 0.778%. Methionine is observed in the largest amount in the single-crop sowing of oats (0.331%), while its minimum value is noted in the grass mixture of oats and peas (0.177%). The single-crop sowing of peas is characterized by the highest content of valine with a level of 3.357%, while the minimum value of this amino acid is noted in a two-component grass mixture of oats and peas with a level of 0.754%.

Threonine is observed in the largest amount in the three-component grass mixture of Sudan grass, peas, and vetch with a level of 1.945%, while its minimum value is noted in the two-component grass mixture of oats and peas with a level of 0.921%. The sum of essential amino acids is the most important in the three-component grass mixture of Sudan grass, peas, and vetch (7.10%). Such amino acids as tyrosine, proline, serine, alanine, and glycine were examined in this study. Data on the analysis of the composition of non-essential amino acids in annual grass mixtures and their mass fraction are shown in Figure 4.

Mass fraction of non-essential amino acids in annual grass mixtures, %



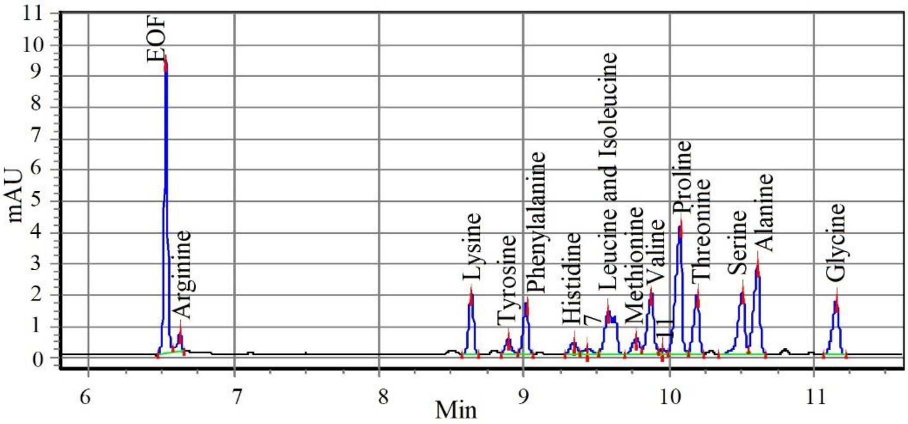
	Avéna satíva	Písum sativum	Vicia sativa	Avéna satíva + Písum sativum	Avéna satíva + Vicia sativa	Avéna satíva + Písum sativum + Vicia sativa	Sorghum × drummondii	Sorghum × drummondii + Písum sativum	Sorghum × drummondii + Vicia sativa	Sorghum × drummondii + Písum sativum + Vicia sativa
Tyrosine	0.290	0.364	0.493	0.278	0.295	0.268	0.283	0.395	0.366	0.401
Proline	1.739	1.833	2.999	1.571	1.447	1.431	1.732	1.289	1.239	1.711
Serine	0.990	0.921	1.073	0.864	0.991	0.848	0.983	0.991	0.920	1.056
Alanine	0.927	0.743	0.877	0.731	0.896	0.764	0.920	0.874	0.825	0.901
Glycine	0.525	0.723	0.919	0.552	0.520	0.463	0.518	0.806	0.735	0.866

Tyrosine Proline Serine Alanine Glycine

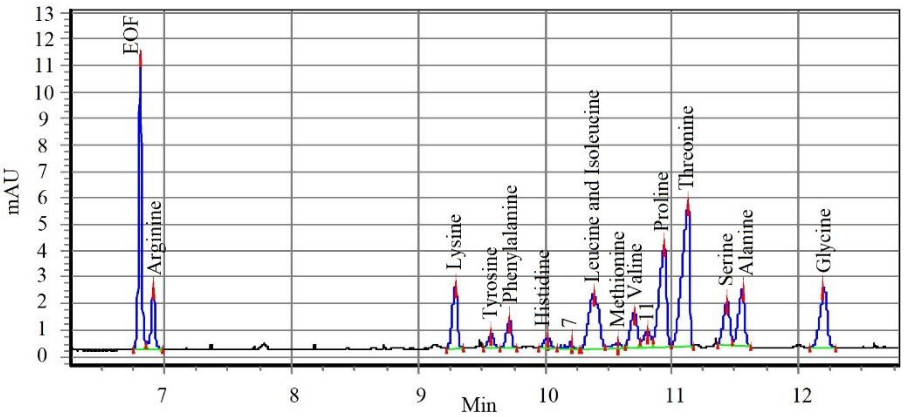
Figure 4. Mass fraction of non-essential amino acids in annual grass mixtures

The vetch single-crop sowing has the highest tyrosine content with a level of 0.493%, while the minimum content of this amino acid is noted in a three-component grass mixture of oats, peas, and vetch with a level of 0.268%. Proline has the highest value in the composition of single-crop sowing of vetch (2.999%), and the lowest proline content is observed in the three-component grass mixture of oats, peas, and vetch (1.431%). The content of serine is highest in the single-crop sowing of vetch (1.073%), and its minimum value is noted in the three-component grass mixture of oats, peas, and vetch (0.848%).

The single-crop sowing of vetch also has the highest content of alanine with a level of 0.877%, while the minimum value of alanine is noted in a three-component grass mixture of oats, peas, and vetch with a level of 0.764%. Glycine is observed in the largest amount in the three-component grass mixture of Sudan grass, peas, and vetch (0.866%), while its minimum value is noted in the three-component grass mixture of oats, peas, and vetch with a level of 0.463%. The amount of non-essential amino acids is the highest in the single-crop sowing of vetch (13.65%). The results of the electrophoregram determined by capillary electrophoresis using the Kapel 105M system are shown in Figure 5.

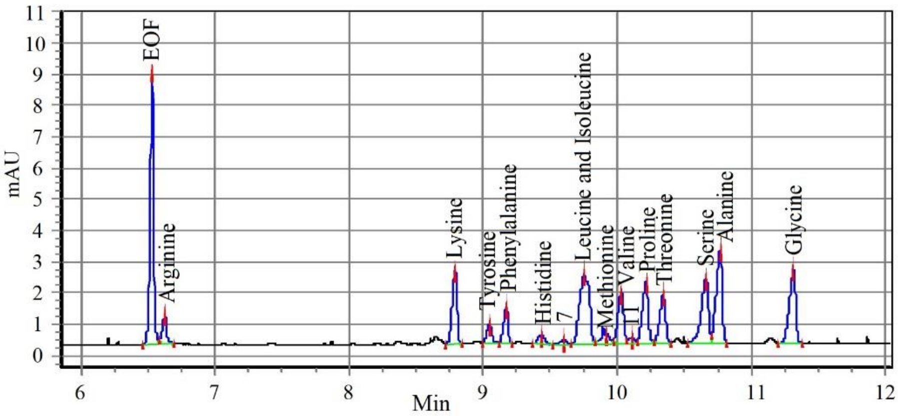


(a)



mAU
Min
EOF
Arginine
Lysine
Tyrosine
Phenylalanine
Histidine
7
Leucine and Isoleucine
Methionine
Valine
11
Proline
Threonine
Serine
Alanine
Glycine

(b)



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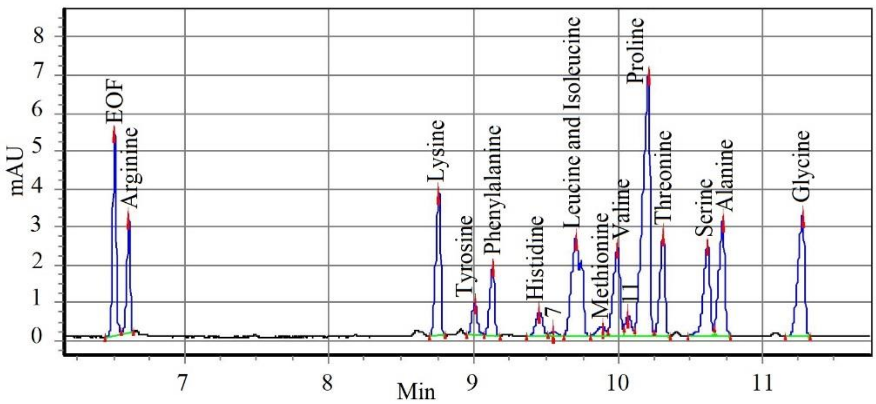

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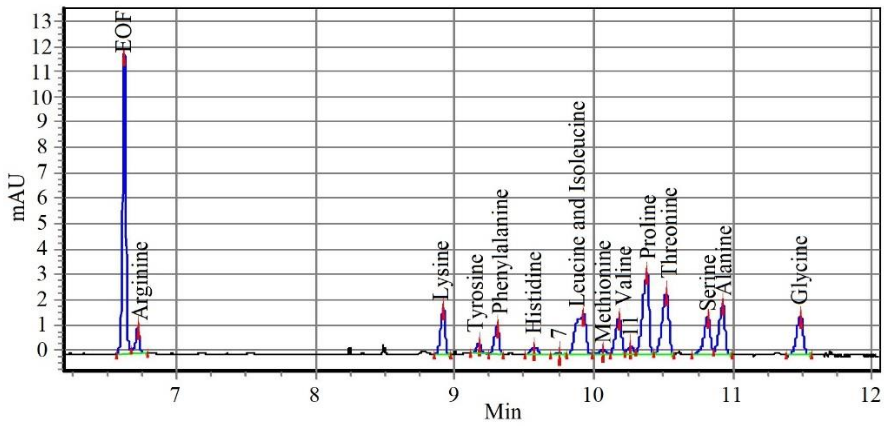

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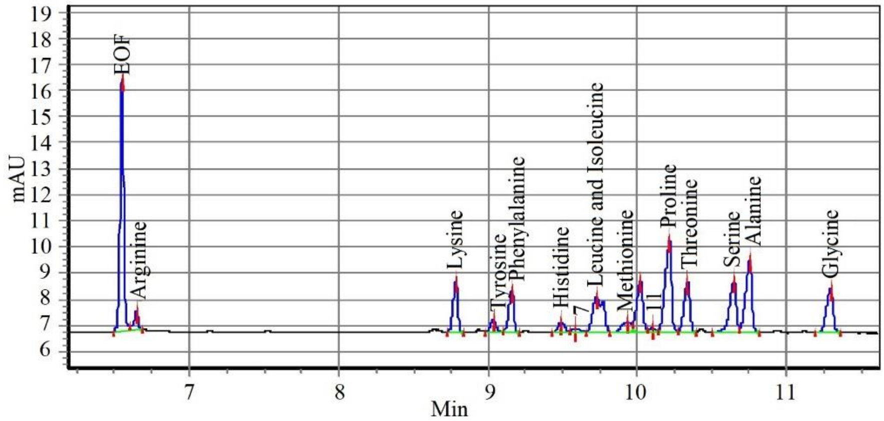

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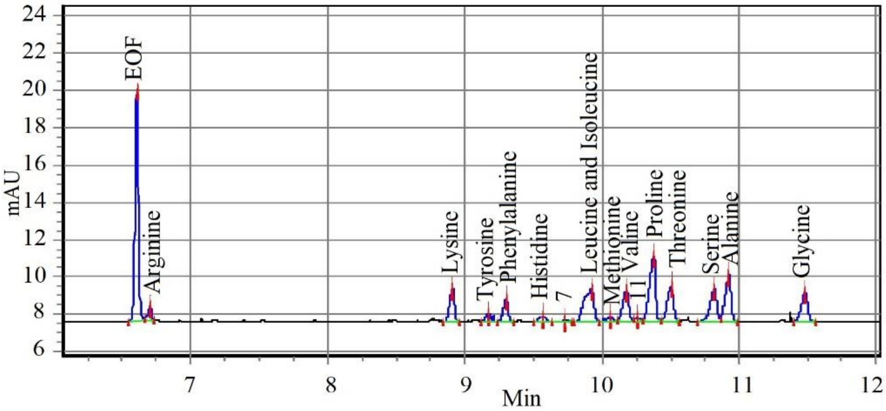

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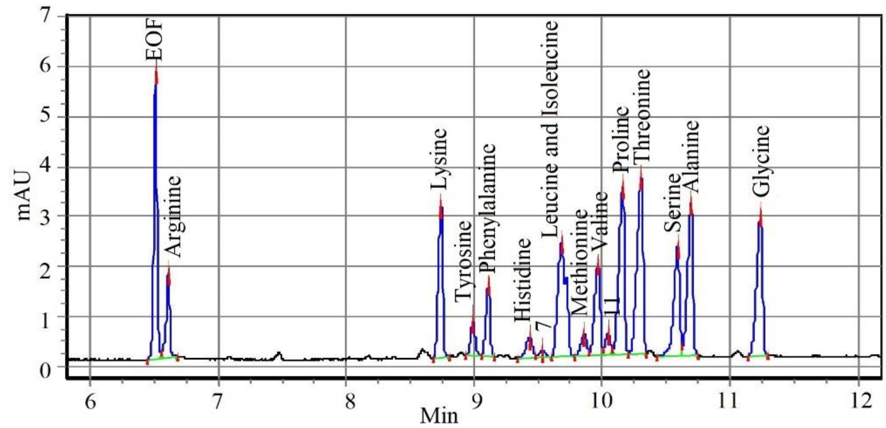

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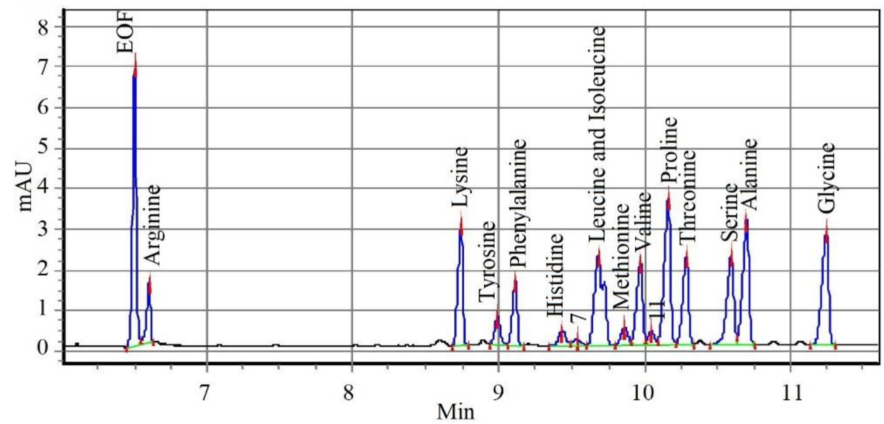

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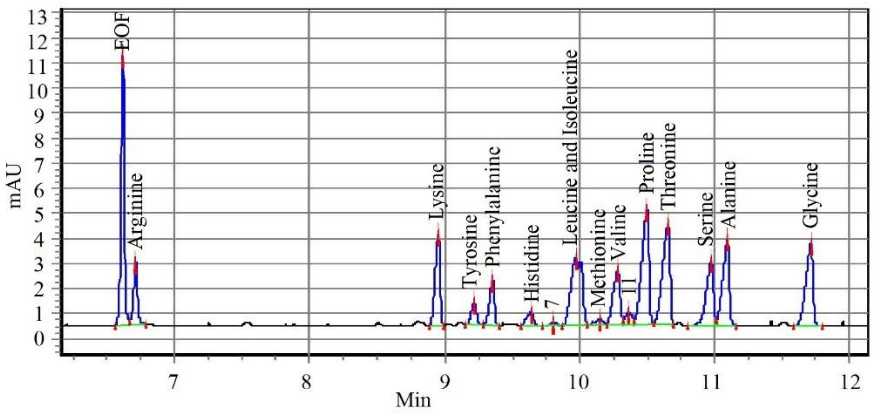

(j)

Figure 5. Electrophoregrams of annual grass mixtures. (a) Oats; (b) Peas; (c) Sudan grass; (d) Vetch; (e) Oats and peas; (f) Oats and vetch; (g) Oats, peas, and vetch; (h) Sudan grass and peas; (i) Sudan grass and vetch; (j) Sudan grass, peas, and vetch

Note: EOF is the electroosmotic flow.

Minerals do not have energy or carbohydrate nutritional value, but their importance in animal nutrition is extremely high since they are involved in all metabolic processes occurring in the body. The mineral elements in the diet in the required amount allow animals to make the most of the nutrients in the feed and ensure maximum productivity. The mineral content of perennial grass mixtures is shown in Figure 6.

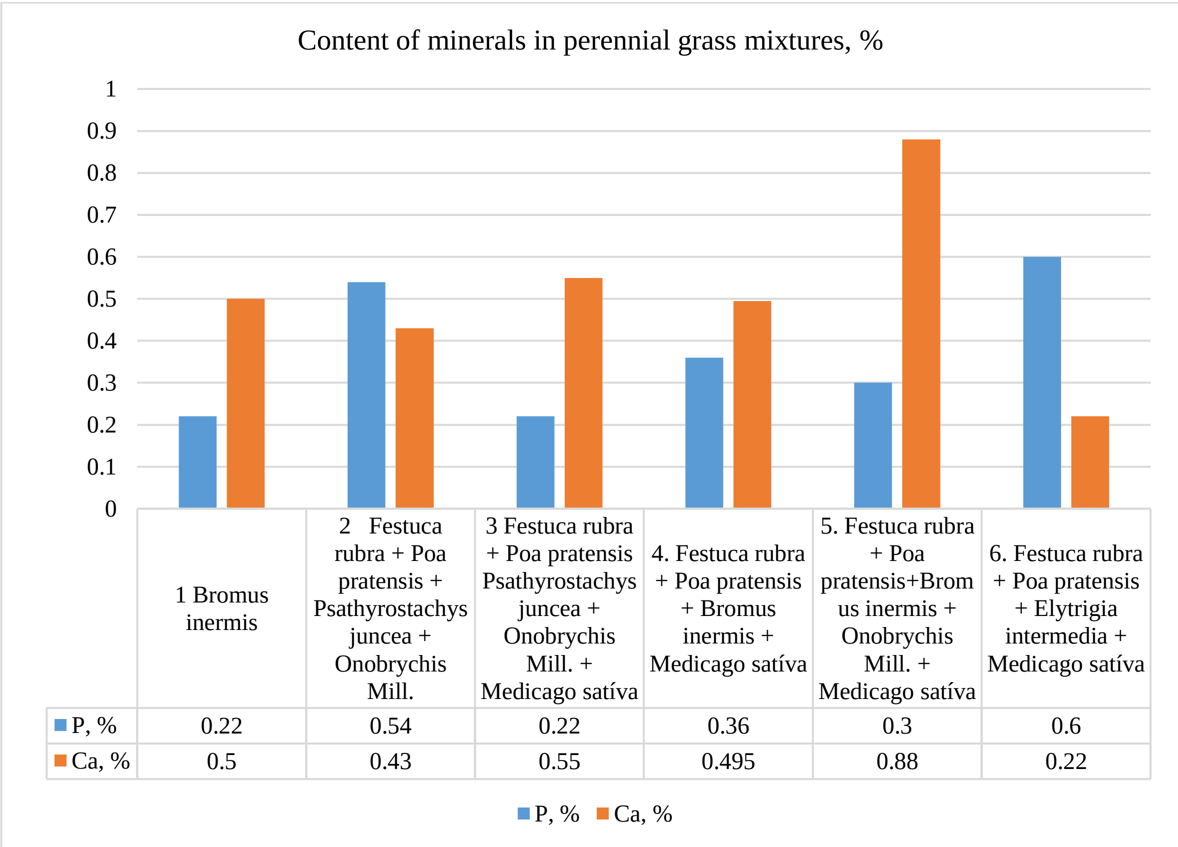

Figure 6. Mineral content in perennial grass mixtures

In terms of phosphorus content, the four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (0.6%) demonstrates the greatest value, while the minimum value is noted in the five-component grass mixture of red fescue, bluegrass, Russian wildrye, sainfoin, and alfalfa (0.22%). The calcium content is observed to have the highest value in the five-component grass mixture of red fescue, bluegrass, brome, sainfoin, and alfalfa

(0.88%), while the minimum value is noted in the four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (0.22%).

The sugar content with the highest value is demonstrated by the single-crop sowing of the awnless brome (3.03%), and the minimum value is noted in the four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (0.86%). In terms of starch content, the highest value is observed in a four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa (4.2%), while the minimum value is noted in the single-crop sowing of awnless brome (1.1%).

4. Discussion

Single-crop sowings of peas and vetch have the highest protein content. However, peas, vetch, and other legumes contain anti-nutritional substances like antigenic proteins, lectins, phytic acid, and others that are toxic and negatively affect the digestion of animals. The method of cultivating legumes together with cereals was used in this study, aiming to reduce the content of anti-nutritional substances, increase the digestibility of feeds, and increase the nutritional value of low-protein feeds. Combining legumes and cereals shows productivity advantages compared to single crops in studies by researchers from the Agricultural University of Inner Mongolia (China) (Qu et al., 2022) and the University of British Columbia (Canada). Higher yields were reported when combining peas with barley (Chapagain & Riseman, 2014). The biomass of joint sowing of vetch and oats increased in comparison with single sowing in the work of researchers from the College of Agricultural Science and Technology of Lanzhou University (China) (Li et al., 2020). The results of this study show that annual grass mixtures of oats, peas, and vetch and Sudan grass, peas, and vetch have a higher protein content compared to monocultures, which is consistent with the above studies. For instance, the grass mixture of oats, peas, and vetch has 39.2% more protein compared with the control variant of single-crop sowing of oats; the Sudan grass, peas, and vetch mixture has 28.1% more protein than the single-crop sowing of oats; and the same trend is observed with perennial grass mixtures, where the presence of legumes like alfalfa and sainfoin increases the CP content from 18.6 to 68.6% compared with single-crop sowing of awnless brome. The reason for the high protein content is the effect of the legume component on providing the herbage with nutrients through the root system and its biological ability to fix atmospheric nitrogen, activating the process of photosynthetic activity. Thus, the results of this study emphasize the importance of using a variety of cultural combinations to ensure the optimal biochemical composition of feed and increase its nutritional value for animals.

The results of this study show that the combination of species of feed grasses and legumes increases ME indicators compared to the same species grown in monoculture. The highest ME indicator is observed in the single-crop sowing of peas (9.98 MJ), which indicates the high energy value of this crop. The oat and pea grass mixture also has a high ME index (9.68 MJ), which indicates the potential of grass mixtures for enriching feed rations. The combination of Sudan grass with peas and vetch also shows a significant increase in the energy value of feed (9.83 MJ), which may be an interesting direction for further research in the field of mixed cultural systems. Comparison with annual and perennial grasses also shows interesting results. Even though perennial grasses usually have a lower ME content compared to annual cereals, a four-component grass mixture of red fescue, bluegrass, wheatgrass, and alfalfa demonstrates a significant ME value (7.98 MJ), which indicates its high potential for use as hay and pasture feed. The data of this study on the ME of crops and their combinations are consistent with the research conducted by Meza et al. (2022) at the Department of Soil Science and Crop Production at the University of Colorado (USA), which shows that combining cereal and legume feed grasses provides highly nutritious feeds with a high energy content.

Moreover, Wang et al. (2022) investigated the effects of seeding options on interspecific competition in oat and common vetch forage crops. They found that mixed sowing options led to better resource utilization and higher nutrient content compared to monocultures. This finding is consistent with the results of this study, which show that the combination of Sudan grass with peas and vetch has a high ME value, highlighting the advantages of diverse crop mixtures.

The analysis of the amino acid balance allows for choosing the optimal combinations of crops to provide animals with a full and balanced diet, which can increase their productivity. Combinations of crops in grass mixtures can enrich the amino acid complex due to the variety of protein components of legumes and cereals. Currently, the most biologically valuable diet in terms of amino acids (essential amino acids) for animals is considered to be a diet in which, on average, one part of tryptophan is combined with the following amount of other acids: lysine: 5.0, leucine: 4.5, valine: 4.0, phenylalanine: 4.0, methionine: 3.0, isoleucine: 2.5, threonine: 2.5, and histidine: 1.5 (Meza et al., 2022). Lysine is necessary for the absorption of calcium and its delivery to the bones. With a low content of lysine in feed, the growth of animals slows down. It supplies the body with substances for the production of the amino acid carnitine, which is involved in the transformation of fatty tissues. Methionine is involved in enzymatic methylation processes leading to the formation of choline, adrenaline, and other biologically important compounds. The lack of methionine in the diets of animals reduces their ability to absorb nutrients, which is the reason for a decrease in productivity. Threonine is necessary for the normal functioning of the immune system and the growth of the body. It promotes the formation of collagen and elastin, participates in the processes of metabolism and assimilation, and supports the work of the gastrointestinal tract. Histidine is used for the synthesis of hemoglobin and many other proteins. It is a source of the biologically active substance histamine, which affects many vital processes in the body. The amino acid isoleucine is important for building muscle tissue. Leucine ensures the growth of the body and is responsible for the normal functioning of the thyroid gland and kidneys. Phenylalanine acts as a building block of proteins, including insulin, papain, and melanin. It promotes the excretion of metabolic products and improves the secretory functions of the pancreas and liver. Valine is necessary to maintain a normal nitrogen metabolism in the body. Tryptophan is involved in the formation of nicotinic acid (vitamin PP) and serotine. With a lack of tryptophan in feed, functional and organic disorders occur in the body (D'Mello, 2003).

The green mass of the three-component grass mixture of Sudan grass, peas, and vetch is characterized by a high content of amino acids (12.04%) with a predominance of arginine, lysine, arginine, leucine and isoleucine, phenylalanine, and valine. The three-component grass mixture of oats, peas, and vetch has a lower amino acid content (8.17%) but has a higher methionine content compared to the grass mixture of Sudan grass, peas, and vetch. Among the two-component grass mixtures, the highest amino acid content is demonstrated by the Sudan grass and peas mixture (10.98%) in the level of threonine (1.86%) and proline (1.28%), and the grass mixture of oats and vetch contains 9.13% of essential and non-essential amino acids. As a result of studies of the amino acid composition, it was found that according to the content of non-essential and essential amino acids, annual grass mixtures could be ranked in descending order: Sudan grass, peas, and vetch (12.04%) > Sudan grass, and peas (10.98%) > oats, and peas (9.68%) > Sudan grass, and vetch (9.16%) > oats, and vetch (9.13%) > oats, peas, and vetch (8.17%). Of the studied components, the mixture of Sudan grass, peas, and vetch demonstrates the highest content of amino acids due to the high content of dry matter and the cutting period of grass mixtures during the budding of the legume component. Studies by researchers from the State Laboratory of the Crop Production and Agriculture System (China) have also confirmed that a higher content of essential amino acids is caused by the high leafiness of the legume component (Bo et al., 2022; Liu & Mahmood, 2015).

In this study, perennial grass mixtures have a high concentration of mineral substances, namely phosphorus and calcium, since the harvesting of grasses was carried out during the budding phase and the beginning of flowering of perennial legumes (alfalfa and sainfoin). The content of calcium and phosphorus in a multicomponent grass mixture exceeds the single-crop sowing of awnless brome by two times. The results are consistent with findings from similar studies, emphasizing the benefits of mixed cropping systems for enhancing forage quality.

Several researchers have reached a similar conclusion. The research by Bo et al. (2022) demonstrates that mixed cropping systems, particularly the combination of alfalfa with winter wheat and ryegrass, significantly enhanced forage quality, yielding higher fresh biomass and superior amino acid profiles compared to monocultures. Similarly, study by Pirhofer-Walzl et al. (2011) highlights that incorporating forage herbs into grassland mixtures improved the mineral composition of the herbage, with herbs showing higher concentrations of essential macro- and microminerals than grasses and legumes. Both studies underscore the importance of diverse species mixtures in enhancing the nutritional value of forage, whether through improved protein and amino acid content or increased mineral concentrations.

5. Conclusions

It was found in this study that mixed feed crops can increase the biochemical and mineral composition and improve the amino acid balance and energy content of the feed. It is recommended to cultivate annual feed crops in a combination of oats and peas, or oats, peas, and vetch. For haymaking and pasture use of perennial grasses, a multicomponent grass mixture is recommended with the inclusion of one or two legumes (alfalfa or alfalfa and sainfoin), such as red fescue, bluegrass, brome, sainfoin, and alfalfa or red fescue, bluegrass, brome, and alfalfa, which show the highest nutritional values. It is recommended to cut annual grass mixtures at the end of the stem extension phase and the beginning of ear formation of grasses, and perennial grass mixtures in the budding phase and the beginning of flowering of legumes. Thus, the results of the study emphasize the importance of using a variety of cultural combinations to ensure the optimal biochemical composition of feed and increase its nutritional value for animals.

To obtain highly nutritious feed in terms of biochemical, amino acid, and mineral composition with high ME in northern Kazakhstan, it is recommended to cultivate annual and perennial feed crops in grass mixtures.

Author Contributions

Conceptualization, S.B. and A.A.; methodology, S.B.; software, M.B.; validation, S.B., A.A., and M.B.; formal analysis, Z.B.; investigation, G.B.; resources, S.B.; data curation, S.B.; writing—original draft preparation, S.B.; writing—review and editing, A.A.; visualization, Z.B.; supervision, A.A.; project administration, A.A.; funding acquisition, M.B. All authors have read and agreed to the published version of the manuscript.

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Data Availability

The data used to support the findings of this study are available from the corresponding author upon request.

Conflicts of Interest

The authors declare no conflict of interest.

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