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# Effects of various protein levels in the ration of dairy heifers on their growth rate and biochemical status

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## ABSTRACT

When breeding dairy cattle, scientists and specialists focus special attention on their productive qualities (milk yield per lactation, fat and protein content, growth rate of young animals, etc.) and the influence of both genetic and paratypic factors on them. The impact of several dietary factors is especially relevant. Therefore, the purpose of this study was to determine the influence of dietary protein concentration, 20.87% (control), 23.49% (Mid), and 30.52%, on the growth of young dairy cattle bred in the northern region of Kazakhstan. The study involved 60 Holstein heifers, aged 90 days (± 5 days), with a body weight of 107.2 ± 0.48 kg. Their daily consumption of dry matter was 2.5 kg day$^{-1}$ which contained 10.10-10.60 MJ metabolizable energy, indigestible protein 14.4-17.8% and crude fiber 22.6-24.6%. The body weight of newborn calves in all groups did not differ and was 31.8-32.6 kg. At the age of 6 months, difference in body weight in the control, Mid, and High groups were 181.1 kg, 198.6 kg, and 200.8 kg respectively. The gain of calves in two experimental groups exceeded that of the control by 9.7% ($p < 0.001$) and 10.9% ($p < 0.001$), respectively. No differences in blood glucose were found. Highest AST values were observed in the control compared to Mid (5.14 U L$^{-1}$) and High (8.25 U L$^{-1}$). The Coefficient of Ritis value was within 2.74-3.11. Research indicates that a ratio in dry matter with an optimal proportion of indigestible protein 23.49% and degradable protein 70% was economically justified.

**Keywords:** Dairy herd, Growth and development intensity, Body weight, Average daily gain, Feed digestibility, Neutral-detergent fiber, Acid-detergent fiber.
**Article type:** Research Article.

## INTRODUCTION

Dairy production is one of the most labor-intensive areas of agriculture, where productivity is largely determined by high-quality feeding. To fully realize genetic potential, cattle must be provided with feed that is nutritionally balanced. An imbalance of the main nutrients leads to inefficient use feed, this, in turn, entails a significant increase in production costs. An integrated approach combining scientific developments, practical experience and economic aspects will effectively solve the problem of nutrient imbalance in feed and achieve a significant increase in the efficiency of animal husbandry (Lidauer *et al.* 2023; Dallago *et al.* 2025; Stephansen *et al.* 2025). With the development of modern technologies, requirements for animals are also growing. Correctly determining growth young animals at various stages plays a key role in ensuring highly productive livestock. This indicator

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significantly affects both life expectancy and efficiency of their using (Martin *et al.* 2021; Dallago *et al.* 2024). In order to achieve the goal of growing healthy, well-developed, resistant to adverse environmental factors and highly productive animals it is extremely important to take into account the peculiarities of their growth and development at different ages. Sufficient protein intake is vital for the normal development of muscle tissue, production essential enzymes and hormones, as well as for a strong immune system. If an animal does not receive enough protein, this leads to slower growth, weakened immunity and development various diseases. On the other hand, excessive consumption of nutrients in the early stages of development can accelerate puberty in females. Body weight at weaning, as well as maternal nutrition during pregnancy, can have a significant impact on the reproductive development of females, affecting formation of the neuroendocrine axis of fetus, and as a result, the subsequent onset of puberty. The health status of livestock, in particular biochemical indicators such as the concentration of amino acids in blood, and protein level in serum, are a direct reflection of the quality of protein nutrition. These indicators reflect the efficiency of absorption and use proteins supplied with feed and can indicate presence of a deficiency or excess of certain nutrients. Monitoring these biochemical indicators is an important tool for assessing the quality of protein nutrition livestock and early detection possible problems (Abilov 2023; Agustinho *et al.* 2024; Gaillard & Abarnou 2024; Huang *et al.* 2024). Blood parameters are important as they allow us to analyze the intensity of physiological processes occurring in body and to study health status. Changes in biochemical blood parameters help to assess current state of the animal, track the influence of various factors acting on body and determine relationship between blood parameters and productive qualities (Ping *et al.* 2024). So, the main goal of our research was to study the influence of different levels of protein component feed on the intensity of growth and development replacement young animal's dairy breeds in natural and climatic conditions of the northern region of Kazakhstan.

## MATERIALS AND METHODS

Preparatory stage of the experiment was carried out from birth to the age of 3 months. In this period calves received colostrum in the amount of 10% their body weight within 1 hour after birth using an individual drencher. The quality of colostrum (immunoglobulin level) from each cow was checked with an Atago PAL-Colostrum refractometer. The experimental calves were fed good quality colostrum with an average level of 22° Brix (lim 20-25°). Subsequent feeding colostrum and pasteurized milk was carried out from a nipple drinker for a month, then watering was carried out from buckets. On third day after birth, calves' blood serum immunoglobulin levels were determined using a refractometer. Experimental calves' immunoglobulin level was 6.1°Brix, with norm being at least 8.4°. Rations were standardized for main nutrients. The structure of ration by dry matter for all groups consisted: succulent feed 55%, roughage 9%, concentrate 36%. Calves of the control group received main ration among compound feed an indigestible protein content at the level of 20.87%. Their peers from first experimental group ("Mid" or Diet$^{23}$) – 23.49% and second experimental group ("High" or Diet$^{30}$) – 30.52%. To assess the chemical composition and nutritional value, samples of feed and feed mixtures (wheatgrass hay, corn silage, sweet clover haylage) were taken. Feed samples were dried at 60-65 ℃ and brought to an air-dry state contains. Hygroscopic moisture was determined by drying a sample feed in a drying cabinet at 100-105 ℃ until it was absolutely dried.

**Table 1.** Feeding ration for heifers.

| Feed name | Feed quantity (kg) | Dry matter (%) |
|---|---|---|
| Agropyron hay | 1 | 95.23 |
| Whole milk | 6.5 | 13 |
| Starter compound feed | 1.5 | 91.04 |
| Total feed | 9 | 100 |
| **The composition of ration** | **Ration** | **Standard** |
| Metabolizable energy, MJ | 25.29 | 40 |
| Dry matter (kg) | 3.16 | 3.8 |
| Indigestible protein (g) | 426.58 | 876 |
| Protein balance of the rumen (g) | -54.49 | 8.28 |
| Amino acids absorbed in intestine (g) | 255.73 | 325 |
| Crude fiber (g) | 368.37 | 398 |
| Neutral-detergent fiber (g) | 647.32 | 1000 |
| Acid-detergent fiber (g) | 488.17 | 715 |
| Starch (g) | 264.9 | 456 |
| Sugar (g) | 264.21 | 318 |
| Crude fat (g) | 92.28 | 221 |

| | | |
|---|---|---|
| Calcium (g) | 107.46 | 25 |
| Phosphorus (g) | 82.89 | 15 |
| Potassium (g) | 49.45 | 26 |
| Sulfur (g) | 2.16 | 8 |
| Sodium (g) | 70 | 5 |
| Zinc (mg) | 150.57 | 200 |
| Chlorine (g) | 80.28 | 9 |
| Magnesium (g) | 14.08 | 8 |
| Vitamin D (thousand IU) | 4.5 | 4.5 |
| Vitamin E (mg) | 78 | 135 |
| Vitamin A (IU) | – | 11.2 |
| **Characteristics of the ration:** | **Fact** | **Optimal** |
| The ratio of calcium to phosphorus | 1.3 | 1.67 |
| Concentrate content (% dry matter) | 43.18 | <50 |
| Indigestible protein content (% from dry matter) | 13.49 | 23.05 |
| Energy concentration in 1 kg dry matter (MJ) | 8 | 10.53 |
| Dietary Cation-Anion Difference | 124.04 | 152.79 |
| Crude fiber (% dry matter) | 11.65 | 10.47 |
| Neutral-detergent fiber (%) | 20.47 | 26.32 |
| Acid-detergent fiber (%) | 15.43 | 18.82 |
| The ratio of sugar + starch to indigestible protein | 1.24 | 0.88 |
| Starch + sugar (in % of the dry matter ration) | 16.73 | 20.37 |
| The ratio of potassium: sodium | 0.71 | 5.2 |
| The content of crude fat in dry matter (%) | 2.92 | 5.82 |
| The content of dry matter in ration (%) | 35.14 | 50 |

The content of indigestible protein was determined by Kjeldahl method using a semi-automatic distillation unit UDK 139 (Velp Scientifica) according to State standard 32044.1-2012 (ISO 5983-1:2005) "Feed, compound feed raw materials. Determination of the mass fraction nitrogen and calculation of the mass fraction indigestible protein" (https://files.stroyinf.ru/data2/1/4293775/4293775094.pdf).

– The dry matter content was determined by drying in a drying cabinet at 60 °C to constant weight, according to State standard 31640-2012 (ISO 3166:004-97) "Methods for determining dry matter content" https://files.stroyinf.ru/data2/1/4293787/4293787414.pdf.

– Crude fat was measured by defatted residue method in a Soxhlet apparatus according to State standard 13496.15-2016 (ISO 3166:004-97) "Methods for determining the mass fraction of crude fat" https://files.stroyinf.ru/data2/1/4293751/4293751219.pdf.

– Crude ash was measured by gravimetric according to State standard 32933-2014 (ISO 5984:2002) "Method for determination crude ash" https://online.zakon.kz/document/?doc_id=39695691.

– The amount of crude fiber was determined by Henneberg-Stoman method according to State standard 31675-2012 (ISO 3166:004-97) "Methods for determining the content of crude fiber using intermediate filtration" https://files.stroyinf.ru/data2/1/4293785/4293785612.htm. The content of fiber fractions was determined using FIWE-3 Fiber Analyzer with neutral-detergent: State standard ISO 16472-2014 https://meganorm.ru/data/573/57398.pdf and acid-detergent: State standard ISO 13906-2013 https://files.stroyinf.ru/data2/1/4293773/4293773728.pdf.

– Calcium was determined using complexometric method according to State standard 26570-95-2003 (ISO 6490-1-85) https://files.stroyinf.ru/data2/1/4294827/4294827866.pdf.

– Phosphorus was determined using photometric method according to State standard 26657-97-2002 https://files.stroyinf.ru/data2/1/4294827/4294827784.pdf.

– Macro- and microelements were determined using Quantum-2a atomic absorption spectrometer according to State standard 30692-2000 "Atomic absorption method for determining the content of copper, lead, zinc, and cadmium" https://files.stroyinf.ru/data2/1/4294813/4294813780.pdf.

Blood samples for biochemical analysis were collected in the morning 2 hours before feeding in vacuum tubes with a coagulation activator. Alkaline phosphatase activity, urea, magnesium, phosphorus, total protein, cholesterol, albumin, alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activity and serum glucose levels were determined using a StatFax-3300 biochemical analyzer with necessary reagent kits. Serum calcium was determined using complexometric method with fluorexone indicator, globulin content and protein index were calculated (Jung 2006). The de Ritis coefficient was determined by calculation as of AST to ALT ratio.

A reference manual with detailed standards was used to formulate balanced diets (Martins *et al.* 2025b). In addition, the determination digestibility of compound feeds was carried out using the method of studying "in vitro" on "artificial rumen" (Nikolić *et al.* 2024). During milk feeding period, which lasts 2 months, the calves were fed pasteurized milk in amount of 360 kg per head. Feeding with pre-starter compound feed and raw drinking water began at the age of 3 days according to palatability; by the 60th day, hay was added to ration. At the end of milk feeding period, at the age of 2.5 months, the replacement heifers were transferred to closed premises, where they were kept in groups of 20 head. A total of 60 Holstein heifers aged 90 days (±5 days) with a body weight of 107.2 ± 0.48 kg were selected. We allocated 3 sections only for experimental livestock 20 heads each, with a feeding front 0.36 $m^2$ (width 0.72 m × depth 0.5 m) per head. The width of feeding table was 43.2 m for the experimental groups, with a depth of 0.5 m. The individuality of feeding was ensured by the presence of headlocks. Feeding front per animal was 0.3 $m^2$. Feed was distributed to feed tables 2 times a day (morning and evening) by mobile Siloking brand feed mixers with automatic observance of the weight and ratio feeds according to a pre-calculated diet. After groups were formed, the growth of experimental livestock was monitored by monthly determination of the body weight each animal individually, followed by averaging indicators based on statistical methods (arithmetic mean, statistical error, coefficient variability) and determining the significance of indicators confirmed by Student's criterion, which eliminates the standard error of group studies.

The relative growth rate was calculated using the following formula:

$$RG = \frac{W_1 - W_0}{W_0} * 100\%$$

where, $W_0$ is body weight at initial and $W_1$ is body weight at final

Experiments were processed using Past v5.2.1 software.

When conducting experiments with animals, international laws and regulations were used: "European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes" (Strasbourg, 1987) and "OOSP Terrestrial Animal Health Code" (Paris 2017).

## RESULTS

The nutritional value of feed is ability to provide animals with necessary nutrients and energy to maintain life, produce products and reproduce. When determining the nutritional value of feed, a comprehensive approach was used, taking into account such parameters as energy value, content proteins, fats, carbohydrates, minerals (macro-, microelements) and vitamins. One of the main indicators of nutrition standardization is establishment of the optimal level of dry matter in ration, since the provision of animal's need for energy and nutrients depends on this. Using chemical analysis methods, the content of main nutritional components in feeds was assessed (Table 2).

**Table 2**. Main feeds chemical analysis.

| Indices | Agropyron hay | Corn silage | Melilot haylage |
|---|---|---|---|
| Dry matter (%) | 93.28 | 26.95 | 39.02 |
| Metabolic energy (Rumjanek *et al.*) | 9 | 10.25 | 8.98 |
| Indigestible protein (%) | 9.24 | 6.82 | 13.1 |
| Decomposable protein (%) | 60 | 85.40 | 78.6 |
| Non-decomposable protein (%) | 40 | 14.6 | 21.4 |
| Crude fiber (%) | 29.83 | 26.54 | 33.56 |
| Neutral-detergent fiber (%) | 67.34 | 54.03 | 48.95 |
| Acid-detergent fiber (%) | 39.22 | 29.72 | 38.83 |

According to data in Table 2, it was established that the studied feeds have a low level of indigestible protein and metabolizable energy. At the same time, the level of crude fiber was high, including its fractions: neutral-detergent and acid-detergent fiber. Table 3 presents the results of chemical analysis compound feed with different protein nutritional values. According to our data, it was found that the content of indigestible protein and crude fat in compound feed used in control and experimental groups complied with standard requirements including 20.87%, 23.49% and 30.52%; 5.54%, 3.81% and 4.9%, respectively. The ratio of protein decomposable and non-decomposable in rumen was within recommended limits (60:30). Table 4 presents the chemical analysis of feed mixtures used in experiment to ration. The daily dry matter intake in all of groups was 2.5 kg $head^{-1}$. In addition, 1 kg dry matter contained 10.1-10.6 MJ metabolizable energy, 14.4-17.8% indigestible protein and 22.6-24.6% crude fiber. In terms of metabolizable energy, the ration of experimental groups was inferior to control by 2% and 5%, respectively.

**Table 3.** Chemical analysis compound feeds by dry matter.

| Indices | "Control" | "Mid" | "High" |
|---|---|---|---|
| Indigestible protein (%) | 20.87 | 23.49 | 30.52 |
| Decomposable protein (%) | 66.7 | 61.6 | 66.7 |
| Non-decomposable protein (%) | 33.3 | 38.4 | 33.3 |
| Crude fat (%) | 5.54 | 3.81 | 4.9 |
| Metabolic energy (Rumjanek *et al.*) | 12.54 | 11.71 | 11.32 |
| Crude fiber (%) | 3.96 | 5.4 | 11.08 |
| Neutral-detergent fiber (%) | 10.42 | 8.78 | 18.29 |
| Acid-detergent fiber (%) | 8.43 | 7.14 | 14.21 |
| Dry matter digestibility (%) | 58.9 | 62 | 58.6 |

**Table 4.** Chemical analysis compound feeds by dry matter.

| Indices | "Control" | "Mid" | "High" | Norm |
|---|---|---|---|---|
| Metabolic energy (Rumjanek *et al.*) | 10.6 | 10.4 | 10.1 | 9.8 |
| Energy feed units | 1.06 | 1.04 | 1.01 | 1 |
| Costs per 1 kg gain (MJ) | 12.6 | 10.25 | 9.91 | – |
| Dry matter (kg) | 2.5 | 2.5 | 2.5 | 2.3-2.7 |
| Indigestible protein (%) | 14.4 | 15.3 | 17.8 | 14.3 |
| ±to control group (%) | – | +6.25 | +23.6 | – |
| Decomposable protein (%) | 71.9 | 68.8 | 71.42 | 66 |
| Non-decomposable protein (%) | 28.1 | 31.2 | 28.58 | 34 |
| Crude fiber (%) | 22.8 | 22.6 | 24.6 | 15-20 |
| Neutral-detergent fiber (%) | 35.2 | 34.6 | 38 | 28-40 |
| Acid-detergent fiber (%) | 25.8 | 25.4 | 27.9 | 16-24 |
| Rate (%) of dry matter digestibility | 42.2 | 48.7 | 38.3 | – |

The amount of indigestible protein in experimental groups was higher than in control by 6.25% and 23.6% respectively. The percentage of indigestible protein in the ration of experimental groups was higher than in control peers too. The ratio of decomposable to non-decomposable protein was 71.9:28.1, in the first experimental group was 68.8:31.2 and in the second was 71.42:28.58. The level of neutral-detergent fiber was close to upper limit norm (34.6-38%). An elevation in the content of indigestible protein of the first and second experimental groups by 6.25% and 23.6% led to an improvement in digestibility dry matter and, as a result, an increase in body weight gain. The growth intensity of animals was assessed based on absolute, average daily and relative increases in body weight during experimental period (Table 5).

**Table 5.** Experimental animals body weight.

| Groups | Body weight (kg) | | |
|---|---|---|---|
| | at birth | 3 months | 6 months |
| "Control" | 32.1 ± 0.6 | 106.9 ± 0.6 | 181.1 ± 3.11 |
| "Mid" | 31.8 ± 0.9 | 108.9 ± 0.9 | 198.6 ± 1.7* |
| "High" | 32.6 ± 0.8 | 108.9 ± 0.8 | 200.8 ± 2.5* |

Note: * the difference between of experimental and control groups is statistically significant at $p < 0.001$.

According to data in Table 5, it can be stated that the body weight of newborn calves in all groups did not differ significantly and was 31.8-32.6 kg. At the age of 6 months, absolute increased body weight in control, first and second experimental groups were 74.2 kg, 89.7 kg ($p < 0.001$) and 91.9 kg ($p < 0.001$), respectively. Fig. 1 shows diagrams of average daily and relative body weight gains over the study period. From the analysis of diagrams presented in Fig. 1 it was found that the average daily gain in body weight of calves in control group during experimental peers was 817 g, against 1015 g ($p < 0.001$) and 1020 g ($p < 0.001$) in the experimental groups (Mid and High) respectively. The coefficient of phenotypic variability ($C_v$) was stable and amounted to 8.1%. The greatest lability was characteristic of indices in heifers "Control" and "Mid", i.e. 18.7% and 11.2%, respectively.

Notably, there were no significant differences in relative body weight gains between the experimental groups received a diet with a protein content of 23.49% and 30.52%. Based on the results of determining the digestibility of dry matter in ration using "*in vitro*" method, it was found that the highest digestibility rate of 48.7% was in the first experimental group (Mid). To assess digestibility and identify problems with feed assimilation using washing on Komilfo sieves, calf manure was analyzed (Fig. 2). According to the diagram the most optimal ratio of undigested components feed mixture was observed in first experimental group, which is confirmed by the high degree of digestibility using "*in vitro*" method (48.7%).

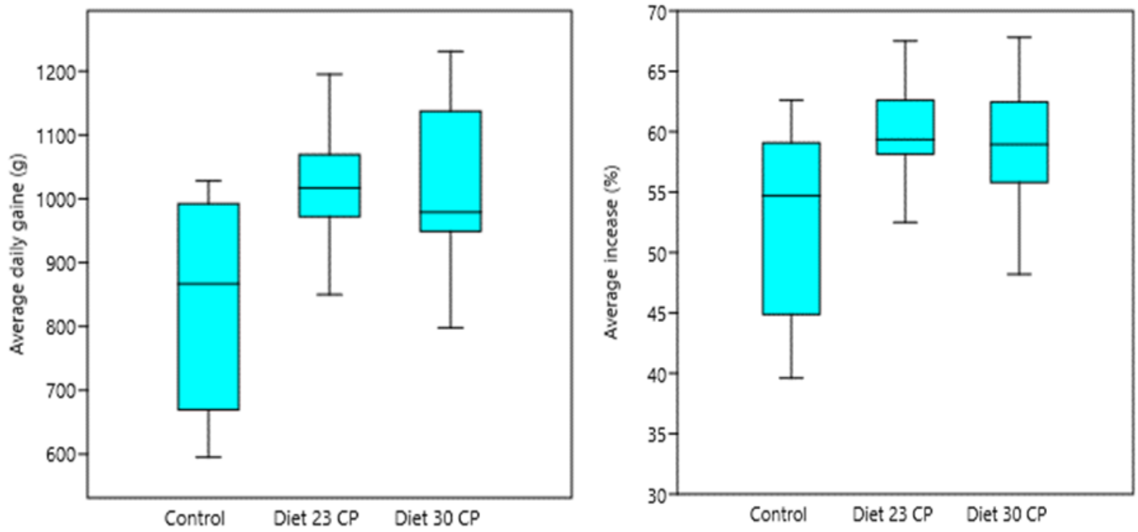


**Fig. 1.** Average daily (a) and relative (b) body weight.

The increased content of neutral-detergent and acid-detergent fiber in the ration of second experimental group affected the larger volume of undigested particles on upper sieve (20%), which is twice norm, while animals in first experimental group showed the optimal ratio of undigested fractions (11%, 18% and 71%). Blood is the main indicator of metabolism of any organism, therefore, to assess the adequacy of feeding, it is necessary to conduct a biochemical blood test. Biomarkers of protein metabolism in animals are the following parameters: total protein, albumins, globulins, urea, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and lactate.

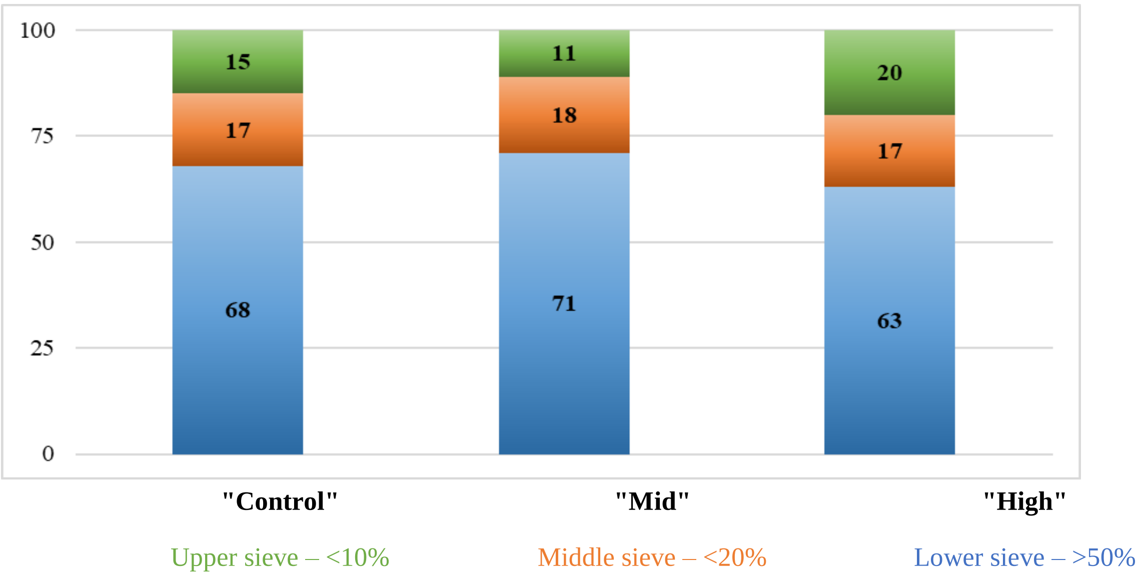


**Fig. 2.** Content of undigested components the ration (%).

Before experiment, all animals were clinically healthy and their blood biochemical parameters were normal: the content of total protein and its fractions (albumins and globulins) were 66.1 G $L^{-1}$, 25.6 G $L^{-1}$ and 38.6 G $L^{-1}$, in addition to urea (3.4 mMol $L^{-1}$); AST (83.6 U $L^{-1}$); ALT (31.6 U $L^{-1}$); and lactate (1.7 U $L^{-1}$), respectively.
Control measurement blood biochemical parameters were carried out at the end of the experimental period at the age of 6 months (Table 6). As established by the data of Table 6, the highest total protein content were observed in the second experimental group compared to "Control" by 8.7 G $L^{-1}$ ($p < 0.001$) and "Mid", difference: 4.9 G $L^{-}$

[1] ($p < 0.001$). It was found that the protein index values of replacement heifers were within 0.53-0.66. Statistically significant difference was revealed between the control and second experimental group (0.13) as well as between the first and second one (0.10). No reliable differences in the blood glucose level of calves were found. The highest AST activity rates were observed in control group compared to the first (5.01 U $L^{-1}$, $p < 0.01$) and second groups (8.24 U $L^{-1}$, $p < 0.05$).

**Table 6.** The biochemical parameters of blood experimental animals.

| Indices | "Control" | "Mid" | "High" |
|---|---|---|---|
| Total protein (G $L^{-1}$) | 61.23 ± 0.55 | 65.03 ± 0.21 | 69.93 ± 0.58 |
| Albumin (G $L^{-1}$) | 24.07 ± 0.53 | 24.98 ± 0.47 | 24.08 ± 0.75 |
| Globulin (G $L^{-1}$) | 37.16 ± 0.69 | 40.05 ± 0.49 | 45.84 ± 0.89 |
| Protein index | 0.66 ± 0.02 | 0.63 ± 0.02 | 0.53 ± 0.03 |
| Lactate (mMol $L^{-1}$) | 2.33 ± 0.26 | 2.25 ± 0.39 | 2.63 ± 0.31 |
| Glucose (mMol $L^{-1}$) | 4.13 ± 0.09 | 4.36 ± 0.07 | 4.25 ± 0.09 |
| Urea (mMol $L^{-1}$) | 4.32 ± 0.17 | 4.42 ± 0.21 | 4.41 ± 0.17 |
| Total bilirubin (mMol $L^{-1}$) | 2.46 ± 0.44 | 2.04 ± 0.23 | 1.78 ± 0.12 |
| Cholesterol (mMol $L^{-1}$) | 2.31 ± 0.11 | 2.54 ± 0.12 | 2.54 ± 0.09 |
| Alanine aminotransferase (U $L^{-1}$) | 26.95 ± 0.69 | 26.85 ± 0.75 | 27.45 ± 0.96 |
| Aspartate aminotransferase (U $L^{-1}$) | 82.65 ± 2.25 | 77.64 ± 3.15 | 74.41 ± 2.17 |
| de Ritis coefficient | 3.11 ± 0.13 | 2.91 ± 0.11 | 2.74 ± 0.09 |
| Calcium (mMol $L^{-1}$) | 2.47 ± 0.07 | 2.47 ± 0.02 | 2.51 ± 0.03 |
| Phosphorus (mMol $L^{-1}$) | 2.71 ± 0.03 | 2.91 ± 0.06 | 2.88 ± 0.05 |
| Magnesium (mMol $L^{-1}$) | 0.83 ± 0.02 | 0.81 ± 0.01 | 0.83 ± 0.01 |
| Alkaline phosphatase (U $L^{-1}$) | 387.14 ± 19.25 | 319.95 ± 17.73 | 328.75 ± 17.88 |

The values of de Ritis coefficient were within 2.74 and 3.11. The calcium and phosphorus concentrations, as well as Ca:P ratio in the blood of all groups were within physiological norm: 2.71-2.91 mMol $L^{-1}$, 2.47-2.51 mMol $L^{-1}$, and 1.1-1.5:1, respectively. To compare the efficiency of feed rations with different protein levels when growing young animals aged 3 to 6 months, it is necessary to take into account the cost of daily ration and cost per unit body weight gain (Table 7).

**Table 7.** Economic efficiency using compound feed with different protein levels.

| Indicators | "Control" | "Mid" | "High" |
|---|---|---|---|
| Feed costs per 1 kg weight gain (Zhao *et al.*) | 152.3 | 119.57 | 119.57 |
| Price of daily ration (Zhao *et al.*) | 751.5 | 804.94 | 126.5-845.02 |
| ± to the control group | – | 53.44 | 93.52 |
| Price 1 kg compound feed (Zhao *et al.*) | 287.24 | 313.96 | 334 |
| Cost of feed (tenge per 1 head) | 67635 | 72444.6 | 76051.8 |
| ± to the control group | – | 480.96 | 8416.8 |
| Cost of feed per 1 kg gain (Zhao *et al.*) | 920.5 | 792.92 | 828.32 |
| ±to the control group | – | 127.59 | 92.18 |
| Gross gain per head (kg) | 74.2 | 89.7 | 91.9 |
| Profitability level (%) | 8.5 | 19.4 | 18.1 |

Note: Conversion rate – 1 euro is equal to 627 Kazakhstan tenge.

The cost of daily ration depended on indigestible protein content in feed, therefore, lowest price was in control group (751.5 tenge), which is lower than cost the rations of experimental groups by 7.1% and 12.4%. As a result of the increased average daily body weight gain in experimental animals, feed costs per unit production decreased by 13.9% and 10%, thereby profitability level exceeded control by 10.9% and 9.6%, suitably. Based on the analysis of efficiency indices for using rations with different indigestible protein content, it was found that level of 23.49% in dry matter of feed and 15.3% in feed mixture with an optimal proportion of degradable protein up to 70% is economically justified. With this ratio, the absorption of nutrients is more efficient, which allows for a reduction in feed costs per 1 kg of body weight gain and an increase in the level of production profitability.

## DISCUSSION

In order to obtain high productivity from farm animals in accordance with their genetic potential, it is necessary to ensure that they consume more dry matter in rations with a variety quality of feeds and with a sufficient

concentration energy and nutrients. In this aspect, the issues of nutrition dietetics, improving the quality of feed, and the taste of rations, as well as the art of cooking and the use of various feed additives are brought to fore.
The digestibility of feed in rumen of cattle is one of the important indices reflecting quality compiled rations, as well as general condition of digestive system. Digestibility is main physiological process by which feed nutrients are transformed from complex chemical compounds into simpler ones that are available for assimilation by body, without which life would be impossible. The issue of digestibility of feed nutrients in rumen has not yet been sufficiently studied, which is due to the complexity of methods for studying this physiological, biochemical and microbiological process (Pazoki *et al.* 2017, Vieira *et al.* 2025, Xiao *et al.* 2025). Protein breakdown in forestomach feeds is considered one of the key indicators reflecting the quality of feed protein and affecting overall nitrogen metabolism animals. Protein cleavage is understood as the microbiological enzymatic destruction of protein and non-protein components of raw feed with the formation of end products – peptides, amino acids and ammonia (Choi *et al.* 2002; Beretta *et al.* 2020; Hong *et al.* 2025).
Proteins are complex chemical compounds, including oxygen, hydrogen, carbon, nitrogen, sulfur and sometimes phosphorus. The amount of indigestible protein in feed is determined by the nitrogen content multiplied by a factor of 6.25, based on assumption that the protein contains an average of 16% nitrogen. For dairy cattle, a good diet is one whose protein is well digested and optimally dissolved in the rumen, which ensures a relatively low concentration of ammonia and sufficient activity of rumen microorganisms (Martins *et al.* 2025a). It should be borne in mind that ruminants, as in monogastric animals, protein is broken down into amino acids in small intestine. In addition, although a number of amino acids in ruminants are formed during cicatricle digestion and they are less dependent on the usefulness of amino acid composition feed, nevertheless, the quality of protein entering small intestine has the same importance for them as for monogastric animals. To date, there has been a rationing system for the protein nutrition of ruminants, which is based on digestible and crude protein, according to which it is assumed that digestible protein is completely absorbed. However, as it has been established in some studies, this situation is valid only for monogastric animals (Nichols *et al.* 2024; Daddam *et al.* 2025). Ruminants have more complex processes converting raw and digestible feed protein, such as the formation of microbial protein in pre-intestines from feed nitrogenous substances and synthetic nitrogenous additives, nitrogen recycling in body and the use of amino acids.
According to modern concepts, when assessing the protein availability of ruminants, it is necessary to know the possibilities and quantitative parameters of microbial synthesis in pancreas, as well as the degree of assimilation along with using feed and microbial protein, amino acids contained in them under various physiological conditions and the level of productivity of animals.
In addition to the content of digestible or crude protein in feed, its solubility, cleavage and amino acid composition of the protein not cleaved in rumen become important indices in this system (Kalashnikov 2003). The content of cleavable fraction of feed protein (RP) should be known in order to normalize nitrogen available for microbial synthesis and the amount of protein not decomposed in rumen (NRP) as a source of amino acids of the feed itself, used in small intestine. Thus, the amino acid requirement of ruminant body is satisfied by microbial protein and protein that has not decomposed in rumen. The total expression of these two protein sources for ruminants is defined as a protein available for exchange. The quality of NRP in terms of amino acid composition should be high enough. This can be achieved by including high-protein feed additives, such as cake, meal, grain, granules and briquettes from legumes (alfalfa, and clover), protected from decomposition in rumen. In order to "protect" protein from decomposition in rumen, both chemical (treatment with formaldehyde, tannins, acetic, formic and other organic acids) and technological (drying, heating, granulation, briquetting, and extrusion) techniques are used. To meet the needs of a ruminant, it is important to ensure not only the total amount of crude protein in ration, but also the optimal ratio of its components in rumen. On average, optimal ratio is considered to be 60-70:30-40.

## CONCLUSIONS

As a result of the conducted research, it was established:
– The content of indigestible protein and crude fat in compound feed used in control and experimental groups meets the requirements of standard and amounts to 20.87%, 23.49% and 30.52%; as well as 5.54%, 3.81% and 4.9%, respectively. In addition, 1 kg dry matter contained 10.1-10.6 MJ metabolizable energy, 14.4-17.8% indigestible protein and 22.6-24.6% crude fiber.
– The amount of indigestible protein in experimental groups was higher than in control peers by 6.25% and 23.6% respectively. The ratio of decomposable to non-decomposable protein in the diets of replacement heifers in

control, the first experimental group and the second were 71.9:28.1, 68.8:31.2 and 71.42:28.58 respectively. An increased indigestible protein content in the ration of first and second experimental groups by 6.25% and 23.6% led to improved absorption dry matter and, as a result, increased body weight gain.
– The body weight of newborn calves in all groups was 31.8-32.6 kg. At the age of 6 months, difference in body weight in Control, Mid and High were 181.1 kg, 198.6 kg and 200.8 kg respectively. The average daily gain of calves in control, first and second groups during experimental period were 817 g, 1015 g ($p < 0.001$) and 1020 g ($p < 0.001$) respectively. In individuals of the first group, relative weight gain during study period was 59.7%, which is 0.5% more than in animals of the second group (59.2%).
– Based on the analysis efficiency indicators for using rations with different indigestible protein content, it was found that level of 23.49% in dry matter feed and 15.3% in feed mixture with an optimal proportion degradable protein up to 70% is economically justified.

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Data are unavailable due to privacy or ethical restrictions. West Kazakhstan Innovation and Technology University's Research Ethics Committee can be contacted to obtain data.

**Conflicts of Interest**
The authors declare no conflicts of interest.

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