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Effect of a multi-enzyme complex on morphometric and chemical parameters of carcasses of Ross 308 cross chickens

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Abstract. The dynamics of live weight growth, morphometric indices of carcasses, and chemical composition of muscles were studied on Ross 308 cross chickens up to 42 days of age raised on diets with increased fiber content and inclusion of multi-enzyme complex of protease and xylanase. Experimental studies were carried out in the feeding department of FSC ARRTPI, and vivarium of GSC "Zagorskoe EBF", and morphometric and chemical parameters were determined at the Department of Veterinary Medicine of the Peoples' Friendship University of Russia. When broilers reached the age of 28, 35, and 42 days, 3 chickens from each age group were slaughtered. Day-old chicks served as starting material. At the end of the growing period, the live weight of birds was determined, slaughtered, and morphometric indices of carcasses were studied; superficial pectoral muscles were subjected to histological studies, and shin muscles were subjected to chemical studies. Live weight of chickens from the control group reached 2432 ± 38.3 g by 42 days of age, experimental chickens — 2556 ± 28.5 g, at $p \le 0.05$ with a difference of 124 g, or more by 4.85%, carcass weight — 1778 and 1870 g respectively, with a difference of 4.20% ($p \le 0.05$). On the 42nd day, the thoracic part of the carcass is 35.19…36.39%, increasing with age — by 1.20%, and the relative mass of the femoral part decreased by 0.68%, the tibia — by 0.72%. Pectoral muscles increased in: protein content — by 22.10%, fat — by 1.89%, and moisture — by 6.15%. The experimental group of chickens showed an increase in the diameter of muscle fibers of superficial pectoral muscles by 42 days in 6.8 times. The structure of the muscle tissue of the superficial pectoral muscles was found to be disturbed at various stages.

Keywords: enzymes, broiler chickens, growth, weight, muscles, tissues, fibers, morphological composition, chemical composition

Authors' contributions: Nikitchenko D.V. — conception, analysis of the obtained data and research design; Rodionov V.D. — collection, processing of materials and text writing; Rystsova E.O. — material processing and

text writing; Snoz G.V. — data analysis; Bogolyubov I.D. — material collection and processing. All authors have read and agreed to the published version of the manuscript.

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Introduction

Growing broilers provide up to 36% of the country's population's needs for high-quality meat products and are accompanied by relatively lower economic costs, which puts the product in demand among representatives of different religions and socio-economic strata. The form of realization by anatomical parts is especially actual in conditions of the modern tendency to robotization of enterprises, as it simplifies automation of processes of storage, loading, and unloading. Therefore, the state allocates economic support to poultry enterprises [1].

The following crosses are widely used in meat poultry farming: Change 9, Cobb 500, Ross 308, Hubbard, and others. These crosses have a high genetic potential and high growth rate, increased proportion of muscle in carcasses, and reduced growing time [2–4]. But at the same time, some carcasses are found to have dystrophies and myopathies: White striping, Wooden breasts, and spaghetti meat [5, 6], reducing the quality of products [7, 8].

Poultry farms use prebiotics, phytobiotics, probiotics, and different feed additives to increase the meat productivity of poultry [9, 10].

Good nutrition is particularly important to unlock the genetic potential of poultry, as broiler feeding costs account for about 70% of the total costs [11, 12].

Vegetable feeds contain significant amounts of cellulose (fiber). The content of beta-glucans increases the viscosity of the feed. Therefore, the use of such crops as barley, oats, wheat, and bran reduces the efficiency of feeding birds. Enzyme supplements are used to break down fiber more efficiently. Thanks to enzymes, the assimilation of nutrition (substances of plant origin and trace elements) in the animal's body is not only much faster but also more efficient. Scientists report that: "without the use of enzyme-containing preparations, the body of livestock or poultry can digest only 15…30% of the feed or mixture. If enzyme preparations or enzyme-based bioactive additives are added to the feed, the digestion increases by an average of one-third" [12, 13].

The main components of compound feeds are peptides and amino acids formed as a result of protease-catalyzed hydrolysis and starchy polysaccharides formed as a result of hydrolysis of glycosidic bonds of non-starchy polysaccharides under the action of xylanase. Glucans, xylans, and araboxylans (pentoses) contribute to the breakdown of plant cell walls, leading to increased availability of starch, proteins, and fats. This

also improves the digestibility of non-starch polysaccharides, increases the metabolic energy of grain feeds, compensates for the lack of endogenous enzymes in young farm animals and poultry, reduces the viscosity of chyme, and decreases the moisture content of manure [14].

In addition, [15] indicates that Vitastimul preparation can be used to increase the viability and productivity of poultry under conditions of postvaccinal stress. The feasibility of its application is confirmed by high indicators of stock safety 97.9…98.1% and higher live weight gain of chickens at the level of 11.98 g compared to the control group.

The study aimed to investigate the dynamics of live weight growth, morphometric indices of chicken carcasses, as well as the chemical composition of muscles of Ross 308 cross broiler chickens, grown on diets with increased fiber content and addition of protease multi-enzyme complex.

Materials and methods

The funding allocated by the Government of the Russian Federation for the development of poultry farming in FSC ARRTPI was aimed at studying the feeding of broilers when enzymatic preparations are included in their diet. Experimental studies were conducted in the Feeding Department of ARRTPI and vivarium of GSC "Zagorskoe EBF", and morphometric and chemical parameters were measured in the Department of Veterinary Medicine of Agrarian and Technological Institute, RUDN University.

According to Lenkova et al. data, the producer of protease and xylanase enzymes is a strain of fungus *Penicillium verruculosum* PEP. These enzymes help to increase the digestibility of nutrients in feed and improve their assimilation. The preparations are available in the form of microgranulated powder [13].

As reported by Egorov et al. and Okolelova and Yengashev, enzymes used for dietary supplementation function effectively in all parts of the poultry intestine. Due to this, they are considered as an alternative to antibiotics, promoting the activity of digestive enzymes and providing the possibility of obtaining ecologically clean and safe meat. The use of enzyme supplements can significantly improve the quality of diets, reduce feed procurement costs, increase stock growth, and reduce maintenance and farming costs [11, 12].

The material for the study was broiler chickens of foreign Ross 308 cross, which were grown in the vivarium up to 42 days of age in batteries of P-15 type. The diets of meat broilers were balanced under the norms of ARRTPI. The experimental group additionally received a multi-enzyme complex.

When broilers reached 28, 35, and 42 days of age, 3 chickens from each age group were slaughtered. Day-old chicks served as starting material. At the end of the growing period, the live weight of birds was determined, slaughtered and morphometric parameters of carcasses were studied according to the method of anatomical cutting of carcasses[1].

[1] Lukashenko VS, Lysenko MA, Stollar TA, Kavtarashvili ASh et al. Methods of anatomical cutting of carcasses, organoleptic quality assessment of meat and eggs of poultry. Sergiev Posad, 2013. 35 c.

After slaughter and processing, the carcasses were weighed and stored in a refrigerator for 24 h at 0…4 °C. Then one-half of the carcasses were divided, separating the bones, muscles, fat, and other tissues, including skin with residual fat, as well as fragments of lungs and kidneys, and re-weighed. The other half was sent for analysis to the chemical and bacteriological laboratory of the Ostankinsky Meat Processing Plant, where the moisture content of the tibia and pectoral muscles was determined[2], including protein[3] and fat[4].

Microbiological studies of meat were conducted according to the established methods of microbiological control of food safety. The bacteriological analysis determined the total number of mesophilic aerobic and facultative-anaerobic microorganisms (QMA&OAMO)[5], Escherichia coli bacteria[6], *Staphylococcus aureus*[7], *Proteu* bacteria[8], sulfite-reducing clostridia[9], as well as *Salmonella spp*[10], and *Listeria monocytogenes*[11].

Superficial pectoral and tibia muscles served as material for histologic studies. Sampling and obtaining histological sections were performed following the methodological recommendations for the preparation of biological samples for histological studies[12]. Histological sections 5…8 μm thick were stained with hematoxylin and eosin. This method was used to detect dystrophies and myopathies in muscles.

The biological value of broiler meat was studied according to the recommendations[13].

JMP Trial software version 14.1.0 was used for statistical data processing. The reliability of differences was assessed using Student's t-criterion.

Results and discussion

During the experiment, it was possible to keep 100% of chickens. Feed consumption per 1 kg of live weight gain amounted to 11.55 kg. The live weight of slaughtered chickens by 42-day old age was 2432 ± 38.3 g in the control group, and 2556 ± 28.5 g in the experimental group, carcass weight was 1778 ± 20.7 and 1870 ± 22.6 g, respectively.

Morphological composition of carcasses. Dynamics of morphometric parameters (Fig. 1): in the period from day 1 to 28 the average daily live weight gain of chickens in the control group amounted to 44.13 g, while in the experimental group, it reached 48.85 g. In the following periods, from day 29 to 35, the gains amounted to 81.43

[2] GOST 9793—2016. Meat and meat products. Method for determining the mass fraction of moisture. Moscow, 2018.
[3] GOST 25011—2017. Meat and meat products. Method for determination of the mass fraction of total protein. Moscow, 2018.
[4] GOST 23042—2015. Meat and meat products. Method for determining the mass fraction of fat. Moscow, 2019.
[5] GOST 31747—2012. Food products. Method for determining the number of mesophilic aerobic and facultative-anaerobic microorganisms. Moscow, 2013.
[6] GOST R 54374—2011. Food products. Method of detection and counting of Escherichia coli bacteria (coliforms). Moscow, 2012.
[7] GOST 31746—2012. Food products. Method for detection and enumeration of Staphylococcus aureus. Moscow, 2013.
[8] GOST 28560—90. Food products. Method of detection and determination of bacteria of the genus Proteus. Moscow, 1991.
[9] GOST 7702.2.6—2015. Meat and meat products. Method for the detection of sulfite-reducing clostridia. Moscow, 2016.
[10] GOST 31468—2012. Food products. Method for the detection of Salmonella spp. Moscow, 2013.
[11] GOST 32031—2022. Food products. Method for the detection of Listeria monocytogenes. Moscow, 2023.
[12] GOST 31931—2012. Food products. Method of sampling and preparation of histological sections for microscopic examination. Moscow, 2013.
[13] Methodological recommendations for the use of express-method of biological evaluation of products and feeds. Approved by VASHNIL. Moscow, 1990.

and 81.71 g, respectively, and from day 36 to 42–83.43 and 82.00 g. During the whole rearing period, the average daily weight gain was 57.90 g in the control group and 60.86 g in the experimental group. The increase in live weight of 42-day-old chickens in comparison with day-old individuals amounted to 56.90 times in the control group and 59.85 times in the experimental group. The slaughter yield of chickens at the age of 42 days was 73.05% in the control group and 73.16% in the experimental group.

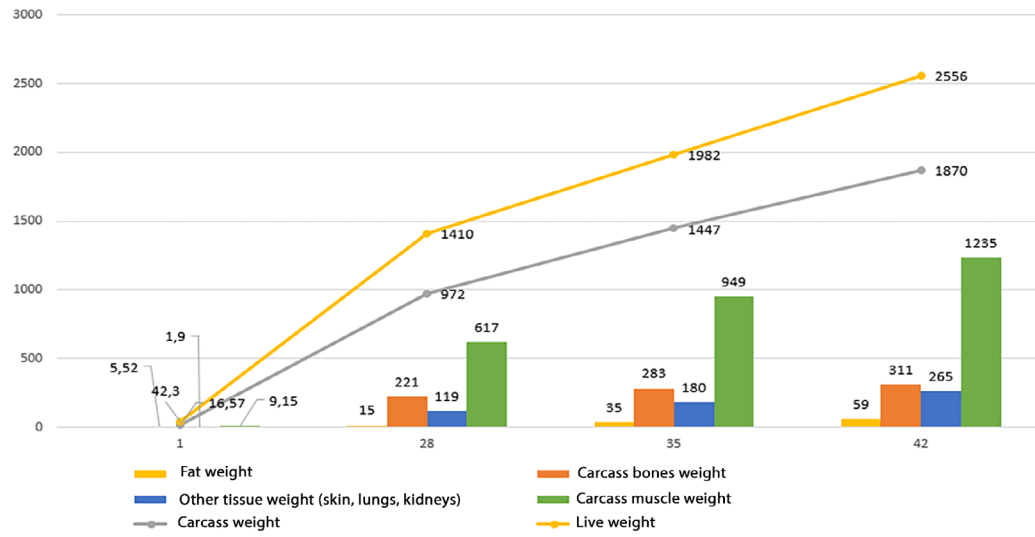

Fig. 1. Dynamics of live weight growth, carcass weight, and morphological composition
Source: compiled by V.D. Rodionov, D.V. Nikitchenko, E.O. Rystsova, G.V. Snoz, I.D. Bogolyubov.

Understanding the mechanisms that regulate muscle growth in poultry is of great importance in crossbreed evaluation. After all, an increase in the amount of muscle growth in the carcass shows an increase in carcass quality through the accumulation of complete protein and essential amino acids.

One of the main tasks of meat poultry breeding is the selection of poultry to increase growth rate and muscle mass while maintaining high-quality meat.

The formation of muscle fibers begins before chick hatching [16], and their subsequent growth occurs due to an increase in diameter associated with the formation of new myofibrils, as well as muscle fiber elongation due to the formation of new sarcomeres [17]. Thus, the increase in muscle mass is due to the number of muscle fibers and their hypertrophy. The growth dynamics of different morphological parts of carcasses in both groups demonstrate the preferential growth of muscle tissue compared to other tissue types.

Up to 28 days of age in the control group chickens, muscle weight reached 567 g, their relative weight amounted to 63.07%, and bone weight — 206 g or 22.91%. By the 42nd day, muscle mass increased to 1169 g, amounting to 65.73%, which is 10.51% more compared to the indicators on the 28th day. Bone mass by day 42 was 298 g (16.76%), which is 16.55% less compared to the values on day 28.

In the experimental group, by day 28, muscle mass amounted to 617 g or 63.48%, and bone mass — 221 g or 22.74%. By day 42, the muscle mass reached 1235 g (66.04%), which is 10.82% more compared to the values on day 28. Bone mass was 311 g (16.63%), which is 16.68% less compared to the values on day 28.

In comparison with day-old chicks, by the 42nd day, muscle mass in the control group increased 127.76 times and bone mass 53.99 times. In the experimental group, the increase in muscle mass amounted to 134.97 times, and bones — 56.34 times. The average daily gain of muscle mass for the whole period of growth amounted to 27.83 g in the control group and 29.40 g in the experimental group. For bones, the average daily gain amounted to 7.10 and 7.41 g, respectively.

In 1 group of chickens at 28 days of age, the ratio of muscles and bones was 2.75: 1, and at 42 days — 3.92: 1; in 2 groups — 2.79…3.97, respectively. Fat deposits in the carcass are an important qualitative indicator of the meat productivity of animals. Protein and fat synthesis in muscle tissue plays a key role in the evaluation of meat productivity. Fat can be deposited both in subcutaneous fatty tissue and in connective tissue between muscles and muscle fibers. Analyzing changes in the weight of edible and inedible parts of the carcass plays a key role in assessing productivity.

Biologically, poultry fat is superior to fats from other farm animals because it is rich in water-soluble essential fatty acids and contains vitamins A, D, and E.

At hatching, muscles contain exclusively structural fat. As the bird ages, reserve fat begins to be deposited, forming the so-called "fat depot", which plays an important role in the trophic and energy supply of the organism, providing it with both mechanical and chemical energy.

These studies show that at 28 days of age, the carcasses of chickens in the control group contain fat 15 g (1.67%), in the experimental group — 1.54%, at 42 days of age — 56 g (3.17%) and 59 g (3.16%), respectively. More than half of the fat is abdominal fat.

Other tissues (skin with fat remnants, kidneys, and lung remnants) in carcasses of group 1 of 28…42 days old ranged from 111…255 g (12,35…14,37%), group 2—119…265 g (12,24…14,17%).

Anatomical cutting of chicken carcasses was performed only in the experimental group because the data on the weight of anatomical parts in the control group were not reliable.

Fig. 2 shows that the highest yield in broiler carcasses of all age groups is in the breast. At the age of 28 days, its relative weight is 35.19%, and at 42 days it is 36.39%, which represents an increase of 1.20%. The carcass weight is 20.47% at 28 days of age and 20.73% at 42 days of age, which is an increase of 0.26%. At the same time, the relative mass of the other anatomical parts decreases: thigh, from 17.49 to 16.81% (a decrease of 0.68%); drumstick, from 14.51 to 13.79% (a decrease of 0.72%); wing, from 11.42 to 10.34% (a decrease of 1.08%).

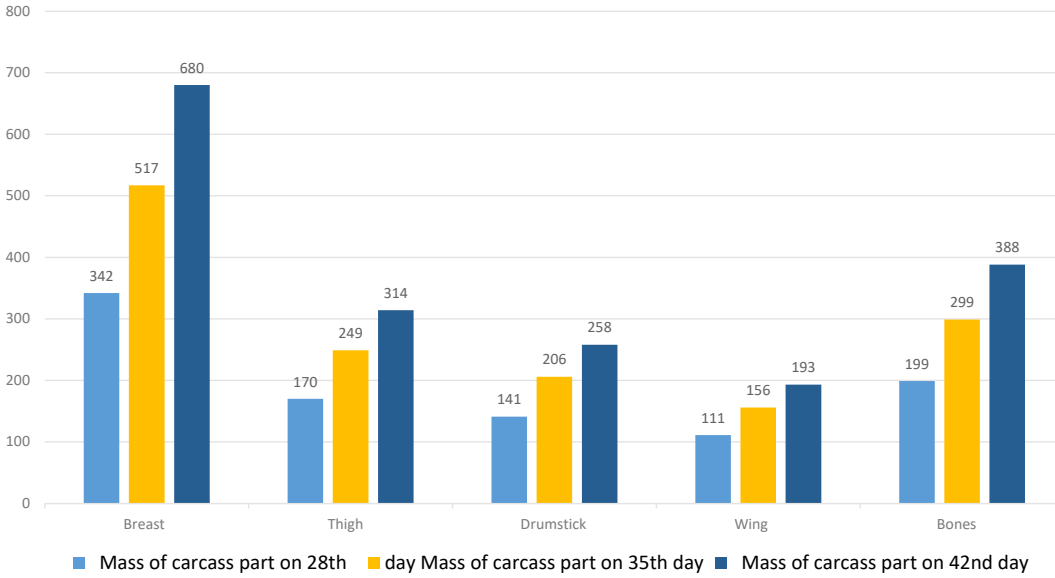

Fig. 2. Age dynamics of carcass parts weight changes obtained during anatomical cutting of broiler chickens of the experimental group (with the use of AGROPROT feed additive in the diet)

Source: compiled by V.D. Rodionov, D.V. Nikitchenko, E.O. Rystsova, G.V. Snoz, I.D. Bogolyubov.

Chemical composition of muscles. The studies showed that the chemical parameters of control and experimental groups of chickens of the same age had no statistically significant differences, so the data only for the experimental group are shown in Fig. 3.

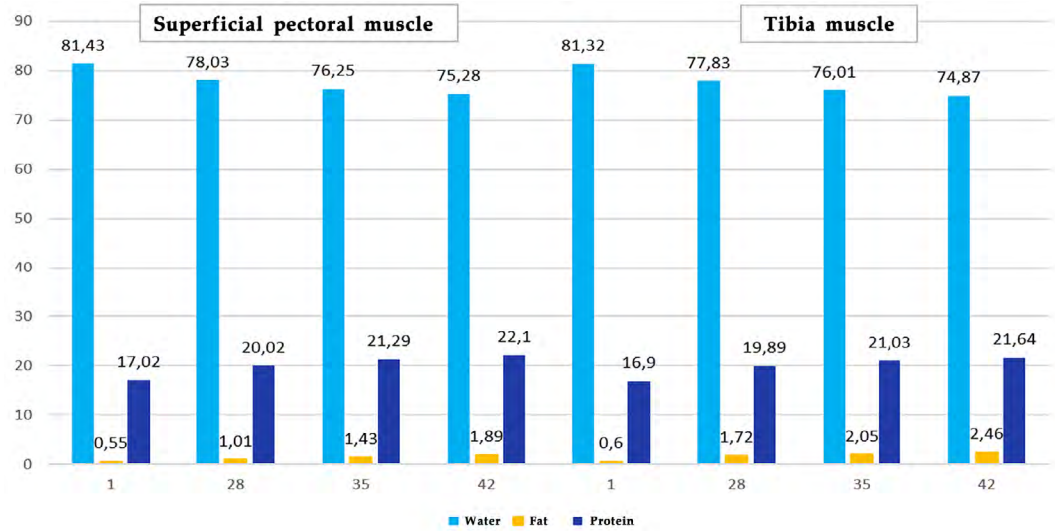

Fig. 3. Chemical composition of pectoral and tibia muscles

Source: compiled by V.D. Rodionov, D.V. Nikitchenko, E.O. Rystsova, G.V. Snoz, I.D. Bogolyubov.

In the superficial pectoral muscles of day-old chicks, the water content was 81.43%, and in the tibia muscles — 81.32%. By day 42, the water level had decreased to 75.30% in the pectoral muscles and 74.84% in the tibia muscles, a decrease of 8.06% over the entire rearing period.

Fat content increased from 0.55 to 1.89% in pectoral muscles and from 0.60 to 2.46% in tibia muscles. The protein content increased from 17.02 to 22.10% in pectoral muscles and from 16.90 to 21.64% in tibia muscles.

Histologic studies. Postembryonic muscle growth continues to increase immediately after chick hatching. If the diameter of muscle fibers of the superficial pectoral muscle in a day-old chick is 8.05 μm, then in 28-day-old chicks it is 28.68 μm, i.e. the daily increase in fiber diameter was 1.021 μm; further from 28 days to 35 days the daily increase increased — 1.76 μm, which in total for 7 days was 12.32 μm; by day 42 the daily increment decreased to 0.9 μm, which in total amounted to 6.3 μm, and in total for the whole period of rearing the diameter of muscle fibers increased to 55.62 μm, or in comparison with day-old chickens — in 6.91 times.

The muscles of poultry, unlike those of mammalian animals, are unevenly developed. They are maximally developed on the thoracic and pelvic limbs. According to our data, they account for about 65% of the live weight of all skeletal muscles.

Chickens of Change 9 cross at cellular technology of cultivation by 42-day-old age reach a little more live weight than Ross 308 cross 2573 g vs. 2556 g, carcass weight — 1897 g vs. 1870 g, but dystrophy of muscle tissue in Change 9 cross is less frequent [18, 19].

According to [16], at intensive technology of growing broilers of Ross 308 cross up to 42-day-old age in some individuals, the superficial pectoral muscle thickening of endomysium and accumulation of interfiber fluid with vacuolization of muscle fibers were revealed.

The analysis by Santos et al. and Barbut showed that the incidence of pectoral muscle myopathies (White striping, Wooden breast, spaghetti meat) increased from 10 to 70% in different companies, causing economic losses reaching hundreds of billions of dollars. The authors note that increased growth rate and increased breast meat yield in fast-growing lines of birds are associated with the development of these muscle pathologies. In myopathies, degeneration and regeneration of muscle fibers, fibrosis, fragmentation, necrosis, and infiltration of the affected areas are observed. It is also emphasized that the processing of meat affected by myopathies reduces the quality characteristics of the product, but this does not necessarily lead to a violation of its food safety [5, 6].

In our experience, 28-day-old chickens showed dystrophy of superficial pectoral muscles in the form of expansion of the diameter of endomysium and perimysium with accumulation of tissue fluid and formation of different volumes of vacuoles in muscle fibers with infiltration by immunocompetent cells (leukocytes, phagocytes). By 42 days, the process worsens, some muscle fibers become rounded, fibrosis develops, and their fragmentation and necrosis (Fig. 4). Some fibers are resorbed, replaced by adipose tissue, and the growth of loose collagenous connective tissue. There is increased infiltration by immunocompetent cells. These morphologic features are characteristic of myopathy — White striping.

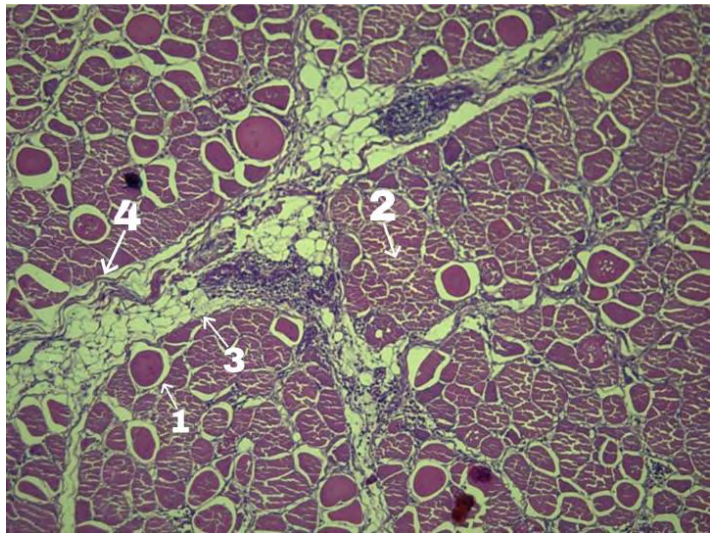

Fig. 4. Myopathy — White striping in superficial pectoral muscle, 42-day-old rooster: 1 — rounding of muscle fibers; 2 — fibrosis of muscle fibers; 3 — adipose tissue; 4 — loose connective tissue

Source: compiled by V.D. Rodionov, D.V. Nikitchenko, E.O. Rystsova, G.V. Snoz, I.D. Bogolyubov.

To prevent the occurrence of myopathies, Santos et al. and Barbut recommend additional genetic research aimed at breeding less susceptible birds, as well as improving feeding and technological factors of housing [5, 6].

Conclusion

It was found that by 42 days of age chickens of the control group reached a live weight of 2432 ± 38.3 g, experimental — 2556 ± 28.5 g, at ($p \leq 0.05$) with a difference of 124 g, or more by 4.85%, carcass weight — 1778 and 1870 g, by 4.20% ($p \leq 0.05$). In carcasses of chickens at 28…42 days of age, concerning other anatomical parts, the thoracic part is the largest — 35,19…36,39%, the increase of which with age amounted to 1,20%, while the relative weight of the thigh part decreased by 0,68%, tibia — by 0,72. During the period from one day to the end of the 42nd day of rearing, the protein content of pectoral muscles increased from 17.02 to 22.10%, the fat content increased from 0.55 to 1.89%, while there was a decrease in moisture content by 6.15%. By the end of the fattening period, the diameter of muscle fibers of superficial pectoral muscles of chickens of the experimental group reaches 60.01 μm, which is 6.8 times greater than the daily values; the average daily gain is 1.318 μm.

In some chickens from control and experimental groups under conditions of intensive industrial rearing technology characterized by stress and hypokinesia, disorders of structural organization of muscle tissue were revealed. These disorders include accumulation of tissue fluid, vacuolar dystrophy, fragmentation of muscle fibers, and Zenker's degeneration.

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Влияние мультиэнзимного комплекса на морфометрические и химические показатели тушек курочек кросса «Росс 308»

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Аннотация. На курочках кросса «Росс 308» до 42-суточного возраста, выращенных на рационах с повышенным содержанием клетчатки и включением в него мультиэнзимного комплекса протеазы и ксиланазы, изучали динамику роста живой массы, морфометрические показатели тушек и химический состав мышц. Экспериментальные исследования проводили в отделе кормления ФНЦ «ВНИТИП», виварии СГЦ «Загорское ЭПХ», а морфометрические и химические показатели определяли в департаменте ветеринарной медицины Российского университета дружбы народов. При достижения бройлерами возраста 28, 35 и 42 сут. осуществляли убой по 3 курочки из каждой возрастной группы. Исходным материалом служили суточные цыплята. По окончании срока выращивания определяли живую массу птицы, производили ее убой и изучали морфометрические показатели тушек, гистологическим исследованиям подвергали поверхностные грудные мышц, а химическим — мышцы голени. Живая масса к 42 сут. курочек из контрольной группы достигла 2432 ± 38,3 г, опытных — 2556 ± 28,5 г, при $p \leq 0,05$ с разницей 124 г, или больше на 4,85%, масса тушек — 1778 и 1870 г соответственно, с разницей на 4,20% ($p \leq 0,05$). На 42-е сутки грудная часть тушки составляет 35,19…36,39%, увеличение с возрастом — на 1,20%, а относитель-

ная масса бедренной части уменьшилась на 0,68%, голени — на 0,72%. В грудных мышцах увеличилось: содержание белка — на 22,10%, жира — на 1,89%, влаги — на 6,15%. Установлено у опытной группы цыплят увеличение диаметра мышечных волокон поверхностных грудных мышц к 42 сут. в 6,8 раз. Выявлены на различных стадиях нарушения структуры мышечной ткани поверхностных грудных мышц.

Ключевые слова: ферменты, цыплята-бройлеры, рост, масса, мышцы, ткани, волокна, морфологический состав, химический состав

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