Annals of Warsaw University of Life Sciences – SGGW
Animal Science No 52, 2013: 113–120
(Ann. Warsaw Univ. of Life Sci. – SGGW, Anim. Sci. 52, 2013)

Histological profile of breast and leg muscles of Silkies chickens and of slow-growing Hubbard JA 957 broilers

MONIKA ŁUKASIEWICZ[1], NATALIA MROCZEK-SOSNOWSKA[1], AGNIESZKA WNUK[1], MACIEJ KAMASZEWSKI[2], DOBROCHNA ADAMEK[2], LEON TARASEWICZ[3], PETER ŽUFFA[4], JAN NIEMIEC[1]
[1]Department of Poultry Breeding, Warsaw University of Life Sciences – SGGW
[2]Department of Ichthyobiology and Fisheries, Warsaw University of Life Sciences – SGGW
[3]Faculty Media Art and Stage Design
[4]Czech and Slovak Breeders Association (race pigeons, chickens original, rabbits, exotic birds)

Abstract: *Histological profile of breast and leg muscles of Silkies chickens and of slow-growing Hubbard JA 957 broilers.* The experiment was conducted with 60 slow-growing Hubbard JA 957 broilers and with 60 Silkies chickens reared until the 63rd day of life, in three replications, 20 birds each, in a closed building on litter. A three--stage feeding program was applied in the rearing period. Feed mixtures used in the experiment did not differ in contents of nutrients. In the first stage of rearing (till 21 days of life) the chickens were fed a standard starter feed mixture containing: 20.98% of total protein, 2,845 kcal/kg of metabolizable energy, 1.17% of lysine and 0.98% of met. + cys. In the period from 22 to 56 days of life they were receiving a grower type mixture containing: 20.0% of total protein, 2,900 kcal/kg of energy, 1.04% of lysine and 0.88% of met. + + cys., whereas contents of these components in the finisher type mixture administered till the end of the rearing period were as follows: 18% of protein, 2,965 kcal/kg of EM, 0.90% of lysine and 0.78% of met. + cys. On day 63 of rearing, 12 cockerels and 12 hens were selected random from each group. The birds were slaughtered in a poultry slaughter house, and specimens of their major breast muscle (*m. pectoralis superficialis*) and thigh muscle (*biceps femoris*) were sampled for analyses. The breast and leg muscles of the birds were characterized by diversified diameters of muscle fibers depending on bird genotype. The greatest diameters were found in breast and leg muscles of the slow-growing Hubbard JA 957 broilers, which was confirmed statistically ($P \leq 0.01$). It was also demonstrated that the mean diameter of muscle fibers of Silkies chickens was more than two times smaller compared to the Hubbard JA 957 chickens.

Key words: Silkies and Hubbard chicken, meat, histology, morphometry of muscle fibers

INTRODUCTION

Contemporarily only highly-selected hybrids of slaughter chickens and turkeys are used in the production of poultry meat in Poland. White meat of chickens, compared to red meat, is deemed as basic in a healthy diet of man owing to, among other things, relatively low contents of fat and cholesterol. In addition, consumers are convinced by a relatively low price and usually convenient portions of this meat as well as by a lack of religious restrictions in its consumption (Jaturasitha 2004).

Quality traits of poultry meat are determined by many factors, including species, breed or genetic line of birds, rearing conditions, pre-slaughter handling and meat processing procedures. The key one include the origin of birds and

their age at slaughter. These two factors influence mainly the organoleptic traits of meat, but also affect its technological properties. A significant factor determining meat quality is the rearing system. In the intensive production of slaughter chickens, constituting the main source of poultry meat in Poland, use is mainly made of hybrids with a high genetically--determined production potential. Consumers of poultry meat are increasingly more interested in the welfare of animals as well as in the quality and safety of final products. Many of them pay attention to housing conditions, rearing system and color of carcass. Today also Polish consumers are considering the purchase of meat originating from chickens reared in less intensive systems and additionally having access to free ranges, as they believe that this meat is characterized by higher quality. Furthermore, they search for meat with good flavor and health--promoting values. Meat of slow-growing chickens is characterized by a higher content of protein and a low fat content desired by consumers (Khantaprab et al. 1997, Połtowicz et al. 2003). The elongation of rearing period affects the concentration of chemical compounds in breast and leg muscles, their more attractive aroma, taste and, thus, sensory properties (Fujimura et al. 1994, Qinghua 1994). A faster rate of these birds body weight gain, at unequal development of their entire bodies, often leads to metabolic disorders, exerts an adverse effect on their health status and – most of all – on meat quality, which is especially significant to consumers.

Meat and animal products are essential components of a man diet. According to PN-65/A-82000, meat constitutes "skeletal muscles and adherent adipose, connective and bone tissues, derived from carcasses, half carcasses or quarter carcasses of particular types of animals for slaughter". One of the most valuable, not only from the dietetic point of view, is poultry meat being a source of balanced protein, amino acids (e.g. leucine, isoleucine, valine or arginine), fatty acids and minerals (Wangang et al. 2010). High digestibility of poultry meat, its assimilability, availability and low price make it very popular amongst consumers (Nowak and Trziszka 2010). The histological structure of muscles is subject to some changes depending on their functions (Klont et al. 1998). Factors that affect changes in the character of muscle fibers include: gender (Ozawa et al. 2000), age (Candek-Potokar et al. 1998), breed (Ryu et al. 2008) and physical activity (Karlsson et al. 1999). The quality of meat is determined by the content of three tissues: crosswise striated muscle tissue, connective tissue and fatty tissue. Mutual proportions and type of particular muscle fibers may lead to changes in the sensory assessment of meat (Damez and Clerjon 2008).

Meat of Silkies chickens has been for years appreciated in the Asian cuisine owing to its health-promoting values. Meat of these chickens is characterized by dark (blue) color of skin and tissue (Siriwan et al. 2004). Nowadays, they are also becoming popular in America where their meat is deemed to be "super food" and "exclusive meat". They owe this status to, among other things, high contents of antioxidants, proteins, vitamins (mainly B-group vitamins and fat-soluble vitamins) as well as macro- and microelements (calcium, iron, phosphorus).

The Silkies chickens came to Europe from China or from eastern India, whereas old literate says also about Singapore. Amateur breeders keep these hens mainly due to their strong hatching instinct and great care over chickens. The Silkies hens are trustful and are reared for their atypical fluffy plumage (silky). They are characterized by a dark mulberry color of comb and wattles turquoise color of earlobes, dark eyes and exceptional crop of hair. Worthy of special attention is dark (blue) color of their skin as well as five toes in each foot. Today, this ornamental hen in several colors is also reared in Europe.

The goal of this study was to determine differences in the histological and morphological structure of fibers of breast and leg muscles of Silkies chickens and slow-growing Hubbard JA 957 chickens.

METHODS AND MATERIALS

The experiment was conducted with 60 slow-growing Hubbard JA 957 broilers and with 60 Silkies chickens reared until the 63rd day of life, in three replications 20 birds each, in a closed building on litter. A three-stage feeding program was applied in the rearing period. Feed mixtures used in the experiment did not differ in contents of nutrients. In the first stage of rearing (till 21 days of life) the chickens were fed a standard starter feed mixture containing: 20.98% of total protein, 2,845 kcal/kg of metabolizable energy, 1.17% of lysine and 0.98% of met. + cys. In the period since 22 to 56 days of life they were receiving a grower type mixture containing: 20.0% of total protein, 2,900 kcal/kg of metabolizable energy (EM), 1.04% of lysine and 0.88% of met. + cys., whereas contents of these components in the finisher type mixture administered till the end of the rearing period were as follows: 18% of protein, 2,965 kcal/kg of EM, 0.90% of lysine and 0.78% of met. + cys.

One-day chicks, after weighing and tagging, were allocated to two groups (each of 60 birds), in three replications (20 birds each). On day 63 of rearing, from each group 12 cockerels and 12 hens were selected random from each group. The birds were slaughtered in a poultry slaughter house, and specimens of their major breast muscle (*m. pectoralis superficialis*) and thigh muscle (*biceps femoris*) were samples for analyses. The samples (0.5×0.5×1.0 cm) were collected within 15 minutes since slaughter after appropriate chicks bleeding, and subjected to 24-hour fixing. Next, the samples were rinsed in ethyl alcohol to remove the fixing agent and dehydrated using a series of ethyl alcohols with increasing concentration. The dehydrated preparation were saturated with paraffin. The saturation process was conducted in an incubator, at a melting point of paraffin. Its duration was adjusted to the sampled specimens of muscles and lasted a few hours. In result, paraffin blocks were formed. Paraffin preparations were sectioned using a Leica RM 2265 rotary microtome (Leica Microsystems, Nussloch, Germany) by cutting the muscles crosswise into 6 μm thick sections that were next stained with a standard H+E method (Ostaszewska et al. 2008). In each preparation, diameters of 300 muscle fibers were measured with a Nikon Ellipse E200 light microscope equipped

in a Nikon DS-Fi2 camera and COOL view 2.7 software.

Results were developed statistically using the variance analysis, computed with the method of the least squares using statistical software SPSS 19.0 PL for Windows (SPSS Inc., Chicago, IL, USA). Differences were found significant at $p \leq 0.05$ and $p \leq 0.01$.

RESULTS AND DISCUSSION

Results of microstructural analysis of the major breast muscle (*m. pectoralis superficialis*) and thigh muscle (*biceps femoris*) of chickens were presented in Table 1. Both the breast (Figures 1 and 2) and leg muscles of the experimental birds were characterized by diversified diameters of muscle fibers depending on genotype. Cross sections showed a distinct shelf structure in most of the fibers. The largest diameter was determined in the breast muscle of slow-growing Hubbard JA 957 birds, which was confirmed statistically ($P \leq 0.01$) – Table 1. The diameter of breast muscle fibers in the group of Hubbard JA 957 birds reached 75.61 μm, whereas in the group of Silkies chickens it was more than two times smaller (33.23 μm). More advantageous to consumers is fine-fiber meat as fine fibers improve its juiciness. In the group of Hubbard JA 957 chickens, diameters of breast muscle fibers were relatively equal, 75–90% of the fibers in particular bundles had diameters in the range of 70–80 μm, and 10–30% of the fibers had diameters in the range of 25–40 μm. In the group of Silkies chickens, diameters of breast muscle fibers were more equalized and reached 25–35 μm in ca. 80–90% of the fibers and less than 25 μm in 10–15% of the fibers.

TABLE 1. Fiber diameter of breast and leg muscles of chickens (♀ + ♂)

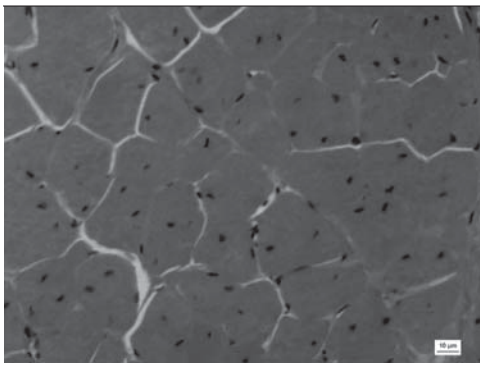
Group	Fiber diameter (μm)	
	breast muscles	leg muscles
Silkies	33.23^{B}	28.73^{B}
Hubbard JA 957	75.61^{A}	62.51^{A}
SEM	1.311	1.262

A, B – statistically significant differences at $P \leq 0.01$.

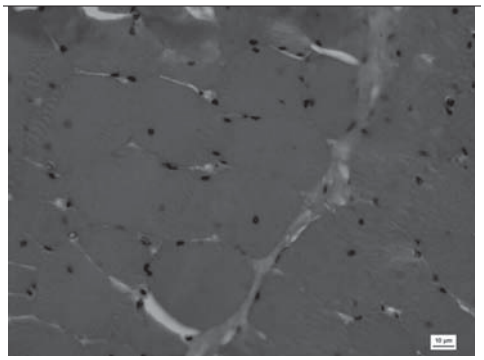
The same tendency was observed in case of leg muscles – larger diameters were demonstrated in Hubbard JA 957 birds. Diameters of leg muscle fibers in the group of Hubbard JA 957 chickens were relatively equalized, i.e. 70–90% of fibers in particular bundles had diameters between 60 and 70 μm, and 10–30% of fibers had diameters in the range of 25––50 μm. In the group of Silkies chickens, the diameters of leg muscle fibers, likewise these of breast muscles, were more equalized compared to the Hubbard JA 957 chickens because 80–90% of fibers in particular bundles had diameters in the range of 20–35 μm, and in 10% of the fibers diameters were lesser than 20 μm.

According to Dransfield and Sośnicki (1999), the fast-growing chickens are characterized by a larger diameter of muscle fibers compared to the chickens with a slow growth rate. As reported by other authors, increased diameter and length of muscle fibers may be due to intensive selection (Brocka et al. 1998, Guernec et al. 2003) and changes appearing in the size and shape of muscle fibers (Bogucka and Kapelański 2004).

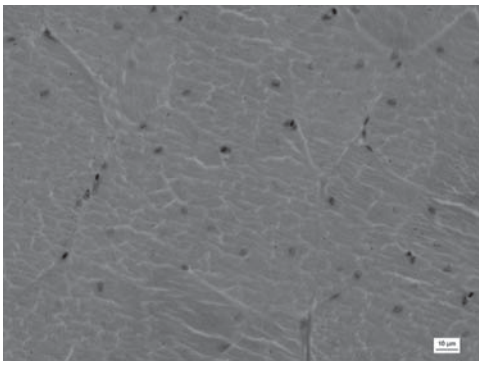
Skeletal muscles are a heterogenous tissue that is characterized by multiple types of muscle fibers that affect



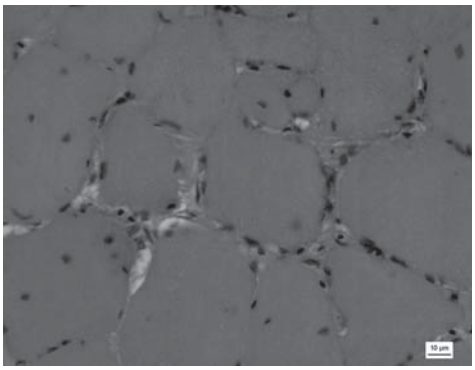
breast muscle – Silkies chickens



leg muscle – Silkies chickens



breast muscle – Hubbard JA957 chickens



leg muscle – Hubbard JA957 chickens

FIGURE 1. Cross-section of breast and leg muscles of Silkies and Hubbard JA 957 chickens

diversified characteristics of particular muscles (Bottinelli and Reggiani 2000). There are many factors that contribute to changes in the character of muscle fibers, these including: gender (Ozawa et al. 2000), age (Candek-Potokar et al. 1998), breed (Ryu et al. 2008), and physical activity (Jurie et al. 1999). Investigations conducted by Castellini et al. (2002a, 2002b) and Branciari et al. (2009) demonstrate that the access to free runs may also influence the character of muscle fibers.

The number and type of muscle fibers are determined already in the fetal life (Petersen et al. 1998, Karlsson et al. 1999). Dankowiakowska et al. (2012) showed that an increased temperature of hatching to 38.5 or 39.0°C resulted in an increased number of fibers with a larger diameter in the breast muscle of broiler chickens. The experiment conducted by Sobolewska et al. (2011) indicates that in the period of 35-day rearing of broiler chickens, a more intensive increase of muscle fiber diameters was observed between days 8 and 21. The breast muscle fibers of chickens are fully developed already around the 8th week of birds life.

The impact of muscle fibers character on meat quality has been investigated for years. The character of muscle fibers is

determined genetically and is typical of particular hybrids and breeds. Histological and biochemical properties of muscles, including the type, number, proportions and diameter of muscle fibers as well as their metabolic character affect the pH value and water absorbability of meat, whereas these traits influence meat quality (Bereta and Eckert 2010). From a consumer's point of view, proportions and diameters of particular types of fibers may lead to changes in the sensory assessment. A higher number of fibers with small and medium diameters improves meat quality (Choi and Kim 2009). Additional increase in the thickness of white muscle fibers has a positive impact on tenderness but a negative impact on juiciness of meat. In case of red fibers, this dependency is opposite (Cameron et al. 1998, Migdał et al. 2005). In hens, almost entire breast muscle is constituted by white fibers.

CONCLUSION

Results of histomorphometric analyses demonstrate that breast and leg muscles of slow-growing Hubbard JA 957 chickens were characterized by a significantly greater diameter of muscle fibers compared to muscles of Silkies chickens. These differences were most of all due to genetic factors. Though slowly growing, the Hubbard JA 957 chickens are selected for body weight gains. The Silkies chickens are a native breed of Chinese hens that have for years been valued for the health-promoting properties of their meat (higher content of carnosine). Chickens of this breed could be successfully used for small-scale production. The end product may be especially appreciated by a taste-loving consumer, a connoisseur or a dietitian owing to its originality and to the fact of being a valuable source of animal protein.

REFERENCES

BERETA A., ECKERT R., 2010: Profil histologiczny mięśni ma ścisły związek z jakością mięsa wieprzowego. Wiad. Zoot. 43, 4, 65–70.

BOGUCKA J., KAPELAŃSKI W., 2004: Histopathological changes in *longissimus lumborum* muscle of Polish Landrace and crossbred Stamboek and Torhyb pigs. Anim. Sci. Pap. Rep. 22, Supp. 3, 67–72.

BOTTINELLI R., REGGIANI C., 2000: Human skeletal muscle fibres: molecular and functional diversity. Prog. Biophys. Mol. Biol. 73, 195–262.

BRANCIARI R., MUGNAI C., MAMMOLI R., MIRAGLIA D., RANUCCI D., DAL BOSCO A., CASTELLINI C., 2009: Effect of genotype and rearing system on chicken behavior and muscle fiber characteristics. J. Anim. Sci. 87, 4109–4117.

BROCKA L., HULSEDGGE B., MERKUS G., 1998: Histochemical characteristic in relation to meat quality properties in the *longissimus lumborum* of fast and lean growing lines of large white pigs. Meat Sci. 50 (4), 441–420.

CAMERON N.D., OKSBJERG N., HENCKEL P., NUTE G., BROWN S., WOODN J.D., 1998: Relationships between muscule fibre traits with meat eating in pigs, BSAS Annual Meeting, 123.

CANDEK-POTOKAR M., ZLENDER B., LEFAUCHEUR L., BONNEAU M., 1998: Effects of age and/or weight at slaughter on *longissimus dorsi* muscle: biochemical traits and sensory quality in pigs. Meat Sci. 48, 287–300.

CASTELLINI C., MUGNAI C., DAL BOSCO A., 2002a. Effect of conventional versus organic method of production on the broiler carcass and meat quality. Meat Sci. 60, 219–225.

CASTELLINI C., MUGNAI C., DAL BOSCO A., 2002b: Meat quality of three chicken genotypes reared according to the organic system. Ital. J. Food Sci. 14, 403–414.

CHOI Y.M., KIM B.C., 2008: Muscle fiber characteristics, myofibrillar protein isoforms, and meat quality. Livest Sci. 122, 105–118.

DAMEZ J.-L., CLERJON S., 2008: Meat quality assessment using biophysical methods related to meat structure. Meat Sci. 80, 132–149.

DANKOWIAKOWSKA A., SOBOLEWSKA A., BOGUCKA J., WALASIK K., ELMINOWSKA-WENDA G. KOZŁOWSKA I., BEDNARCZYK M., 2012: Zróżnicowanie grubości włókien mięśniowych kurcząt 1-dniowych stymulowanych temperaturą podczas inkubacji. Proc. XXIV Międzynarodowe Sympozjum Drobiarskie PO WPSA, 22–24.

DRANSFIELD E., SOSNICKI A.A., 1999: Relationships between muscle growth and poultry quality. Poult. Sci. 78, 743–746.

FUIJMURA S., MURAMOTO T., KATSUKAWA M., HATANO T., ISHIBASHI T., 1994: Chemical analysis and sensory evaluation of free amino acids and 5-inosinic acid in meat of Hinari-dori, Japanese native chicken. Comparison with broilers and layer pullets. Animal Sci. and Technology 65, 610–618.

GUERNEC A., BERRI C., CHEVALIER B., WACRENIER-CÉRÉ N., LE BIHAN-DUVAL E., DUCLOS M.J., 2003: Muscle development, insulin-like growth factor-I and myostatin mRNA levels in chickens selected for increased breast muscle yield. Growth Hormone and IGF Res. 13, 8–18.

JATURASITHA S., KHIAOSAARD R., PONGPAEW A., LEAWTHARAKUL A., SAITONG S., APICHATSARANGKUL T., LEAUNGWUNTA V., 2004: Carcass and indirect meat quality of native and Kai Baan chickens with different sex and slaughter weight. Proc. 42nd Annu. Conf. Kasetsart Univ., Bangkok, 116–126.

JURIE C., PICARD B., GEAY Y., 1999: Changes in the metabolic and contractile characteristics of muscle in male cattle between 10 and 16 months of age. Histochem. J. 31, 117–122.

KARLSSON A.H., KLONT R.E., FERNANDEZ X., 1999: Skeletal muscule fibres as factor pork quality. Livestock Production Science. 60, 255–269.

KHANTAPRAB S., NIKKI T., NOBUKUNI K., 1997: Effect of re- stricted feed intake on the growth of muscle and fat deposition in broiler chickens. Jpn. Poult. Sci. 34, 363–372.

KLONT R.E., BROCKS L., EIKELENBOOM G., 1998: Muscle fibre type and meat quality. Meat Sci. 49, 219–229.

MIGDAŁ W., WOJTYSIAK D., PAŚCIAK P., 2005: Profil histochemiczny mięśni tuczników w zależności od rodzaju mięśnia, płci, rasy, masy ciała i żywienia. Żywność 3 (44), Suppl., 157–168.

NOWAK M., TRZISZKA T., 2010: Zachowania konsumentów na rynku mięsa drobiowego. Żywność. Nauka. Technologia. Jakość 1 (68), 114–120.

OSTASZEWSKA T., DĄBROWSKI K., WEGNER A., KRAWIEC M., 2008: The effects of feeding on muscle growth dynamics and the proliferation of miogenic progenitor cells during Pike Perch development (*Sander lucioperca*). J. World Aquaculture Society 39 (2), 184–195.

OZAWA S., MITSUHASHI T., MITSUMOTO M., MATSUMOTO S., ITOH N., ITAGAKI K., KOHNO Y., DOHGO T., 2000: The characteristics of muscle fiber types of *longissimus thoracis* muscle and their influences on the quantity and quality of meat from Japanese Black steers. Meat Sci. 54, 65–70.

PETERSEN J.S., HENCKEL P., OKSBJERG N., SØRENSEN M.T., 1998: Adaptations in muscle fibre characteristics induced by physical activity in pigs. J. Anim. Sci. 66, 733–740.

PN-65/A-82000. Mięso i podroby zwierząt rzeźnych. Wspólne wymagania i badania.

POŁTOWICZ K., WĘŻYK S., CYWA-BENKO K., 2003: Wykorzystanie rodzimych ras kur w produkcji mięsa bezpiecznego dla zdrowia konsumenta. Praca zbiorowa. Zakrzewo, 21–32.

QINGHUA Z., 1994: Palace chicken superior Chinese breed. Misset World Poultry 10, 47.

RYU Y.C., CHOI Y.M., LEE S.H., SHIN H.G., CHOE J.H., KIM J.M., HONG K.C., KIM B.C., 2008: Comparing the histochemical characteristics and meat quality traits of different pig breeds. Meat Sci. 80, 363–369.

SIRIWAN P., RATTANAWARAHA A., PIMSARN S., SONLOY W., NAGKITSET S., 2004: A preliminary study in order to develop and improve black-boned chickens. J. Anim. Husbandry 68, 44–53 (In Thai).

SOBOLEWSKA A., ELMINOWSKA-WENDA G., WALASIK K., BOGUCKA J., SŁAWIŃSKA A., SZCZERBA A., ŻMUDA-TRZEBIATOWSKA M., 2011: Increase in thickness of the pectoral muscle fibers in meat-type chicken beetween 1st and 35th day of fattening. Proc. XXIII International Poultry Symposium PB WPSA, Poznań.

WANGANG Z., XIAO S., SAMARAWEERA H., LEE E. J., AHN D., 2010: Improving functional value of meat products. Meat Sci. 86, 15–31.

Streszczenie: *Profil histologiczny mięśni piersiowych i nóg kurcząt Jedwabistych oraz wolno rosnących Hubbard JA 957*. Badanie przeprowadzono na 60 brojlerach wolno rosnących Hubbard JA 957 oraz 60 kurczętach Jedwabistych odchowywanych do 63. dnia życia, w trzech powtórzeniach po 20 szt., w zamkniętym budynku na ściółce. W okresie odchowu zastosowano trzyfazowy program żywienia. Mieszanki użyte w doświadczeniu nie różniły się pod względem zawartości składników pokarmowych. Kurczęta w pierwszym okresie odchowu do 21. dnia życia żywiono standardową mieszanką typu starter: o zawartości 20,98% białka ogólnego, 2845 kcal na kg energii metabolicznej, 1,17% lizyny i 0,98% met. + cys. W okresie od 22. do 56. dnia stosowano mieszankę typu grower, zawierającą 20,0% białka ogólnego, 2900 kcal na kg energii metabolicznej (EM), 1,04% lizyny i 0,88% met. + cys. Zawartość wcześniej wymienionych składników w mieszance finiszer, stosowanej do końca odchowu, wynosiła odpowiednio: 18% białka, 2965 kcal na kg EM, 0,90% lizyny i 0,78% met. + cys. W 63. dniu odchowu wybrano z każdej grupy po 12 kogutów i 12 kur o masie ciała zbliżonej do średniej w grupie. Kurczęta ubito w ubojni drobiu i pobrano wycinki mięśnia piersiowego powierzchniowego (*m. pectoralis superficialis*) oraz dwugłowego uda (*biceps femoris*). Mięśnie piersiowe i nóg ptaków doświadczalnych charakteryzowała zróżnicowana średnica włókien mięśniowych w zależności od genotypu. Największą średnicą charakteryzowały się mięśnie piersiowe i nóg ptaków wolno rosnących Hubbard JA 957, co zostało potwierdzone statystycznie ($P \leq 0,01$). Stwierdzono, że średnica włókien mięśniowych kurcząt Jedwabistych była o ponad połowę mniejsza w porównaniu do kurcząt Hubbard JA 957.

MS. received in November 2013

Authors' address:
Monika Łukasiewicz
Wydział Nauk o Zwierzętach SGGW
Katedra Szczegółowej Hodowli Zwierząt
Zakład Hodowli Drobiu
ul. Ciszewskiego 8, 02-786 Warszawa
Poland
e-mail: monika_lukasiewicz@sggw.pl