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HISTOLOGICAL CHARACTERISTICS AND FUNCTIONAL PROPERTIES OF RED AND WHITE PARTS OF M. *SEMITENDINOSUS* OF SLAUGHTER PIGS

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Keywords: *meat quality, meat defects, PSE, hypercontraction, m. semitendinosus, type of muscle tissues, histology*

Abstract

A unique muscle of pigs (Sus scrofa domesticus) is m. semitendinosus, which contains the "red" (dark) part located mainly in the depth of the leg cut and the "white" (light) part located in the close proximity to the subcutaneous fat layer. Differences in the characteristics of its "red" and "white" parts can exert a significant effect on quality and economic indicators of meat products. The aim of this research was to study histological features of the microstructure and technological properties of muscle tissue from different parts of m. semitendinosus, obtained from slaughter pigs of Russian production. M. semitendinosus was excised from chilled porcine carcasses (N=20) 24 hours after slaughter in the process of deboning. Histological examination showed that the dark part of the muscle was characterized by a higher package density of fibers, higher number of capillaries and higher sarcomere length. On the contrary, the light part was characterized by a higher diameter of muscle fibers. Analysis of muscle fiber types showed that the proportion of type I, intermediate and type IIb fibers was higher by 9.3, 5.2 and 4.1%, respectively, in the dark part. Significant differences between the dark and light parts of m. semitendinosus were revealed in terms of the number and size of giant fibers: the light part was characterized by a larger number (by more than 5 times) of giant fibers with the fibers of a larger size (almost by 11%). The samples of minced meat from the dark and light parts showed significant ($p<0.05$) differences in the mean values of lightness, redness and yellowness (L^*, a^* and b^*) by 6.00, 4.68 and 3.01 units, respectively, in raw samples, and by 6.53, 2.99 and 1.81, respectively, after curing with the nitrite mixture and cooking ($p<0.05$). The dark part of m. semitendinosus had higher pH values ($p<0.05$) both for raw and cooked samples. The consistency of the samples from the light part was less elastic, looser and more crumbly than that in the samples produced from the dark part of m. semitendinosus, which was confirmed by the structural-mechanical investigations. Therefore, this study showed significant differences between the dark and light parts of m. semitendinosus by microstructural and functional-technological characteristics. Significant variability by muscle fiber diameter, which was observed in the light part of this muscle, apparently should be taken into account in breeding work and quality assessment of pork from slaughter animals.

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Introduction

Two quality defects of meat are widely known in the meat industry: pale, soft and exudative (PSE) and dark, firm and dry (DFD) meat. DFD meat has the dark red or red color and pH values higher than 6.0. On the contrary, PSE meat has the pale pink color and pH values lower than 5.6. It is traditionally believed that the PSE defect is typical of "white" muscles with the high content of white glycolytic fibers (IIb type). At the same time, "white" muscles are more prone to the development in the process of autolysis of the so-called "giant" fibers emerging mainly as a result of hypercontraction of IIb type muscle fibers [1]. "Red" muscles, which have a significantly lower content of such fibers, are distinguished, as a rule, by a high pH value after rigor mortis and high uniformity of muscle fibers by diameter. Muscles that are different in color usually have different metabolism: "red" muscles are characterized by aerobic metabolism and "white" muscles by anaerobic metabolism. Prominent examples of "white" and "red" muscles in pigs are *m. longissimus dorsi* and *m. masseter*, respectively [2].

Skeletal muscles of mammals, birds and fish demonstrate a great variety of shapes, sizes, anatomic location and physiological functions. Certain muscles of slaughter animals can simultaneously have different types of metabolism, various physiological functions and can show different ultimate result of meat quality formation during ageing after slaughter [3]. Such a unique muscle in pigs (*Sus scrofa domesticus*) is *m. semitendinosus*, which contains the "red"

(dark) part located mainly in the depth of the leg cut (ham) and the "white" (light) part located in the close proximity to the subcutaneous fat layer. However, both light and dark parts constitute a single muscle having the same origin and location [3,4].

Characteristics of muscle tissue are of fundamental importance for meat and meat product quality. Producers, distributors and consumers impose various and specific requirements for quality that depend on the use of products [3]. The leg cut of pork carcass is a valuable raw material for production of expensive delicacy products such as cooked, cooked-smoked, raw smoked and raw air dried hams and other meat products from whole muscles (group of muscles), for which color uniformity is an important characteristic of production quality [5]. The presence of PSE meat (PSE zones) in ham also negatively affects sensory characteristics such as consistency, juiciness, aroma and taste [3]. For processors, the best technological qualities of pork are always associated with low losses in production of the final product, including upon cold storage, cutting, cooking and other operations. Due to the low water holding capacity of PSE pork, its processing leads to a significant decrease in the final product yield and an increase in the risks of manufacturing products with low quality or defects [3,6].

The structure and composition of skeletal muscles and, consequently, the technological properties of pork are influenced by many factors *in vivo* and *post mortem*, such as species and genotypes of animals, nutritional and environmental factors, conditions of slaughter and primary processing of carcasses. As there are a large number of such factors, their interaction with meat quality is not always clear due to their mutual influence [3]. Studying this interrelation is expedient when undesirable reduction of certain characteristics of raw materials and their effect on meat technological properties were established.

In the Russian Federation, fast growing hybrid animals from three-breed crossing (LWxDxL) are mainly used for growing pigs for slaughter [7]. Earlier, we studied manifestation of myopathic changes, which are typical of PSE meat and emerge under an effect of lifetime and slaughter factors, in the microstructure of *m. longissimus dorsi* taken from slaughtered hybrid animals [8,9]. This muscle is the most frequently chosen object of investigations and its characteristics are studied quite well. Several foreign studies aimed at investigation of *m. semitendinosus* from regional phenotypes of animals showed significant differences in characteristics of "red" and "white" parts and their impact on quality and economic indicators of meat products. The number of studies dedicated to pork quality carried out on the samples of *m. semitendinosus* has been increasing over the last years [10–14]. There are only few studies aimed to investigate the microstructure of "red" and "white" parts of *m. semitendinosus*, as well as a possible effect of special features of growing, transportation and slaughter of pigs in Russia on quality of muscles from the leg cut. Therefore, the aim of this study was to investigate the histological characteristics and technological properties of muscle tissue of the dark and light parts of *m. semitendinosus* obtained from slaughter pigs of Russian production.

Objects and methods

Objects of research

Objects of research were *m. semitendinosus* excised from chilled porcine carcasses 24 hours after slaughter in the process of deboning of leg cuts.

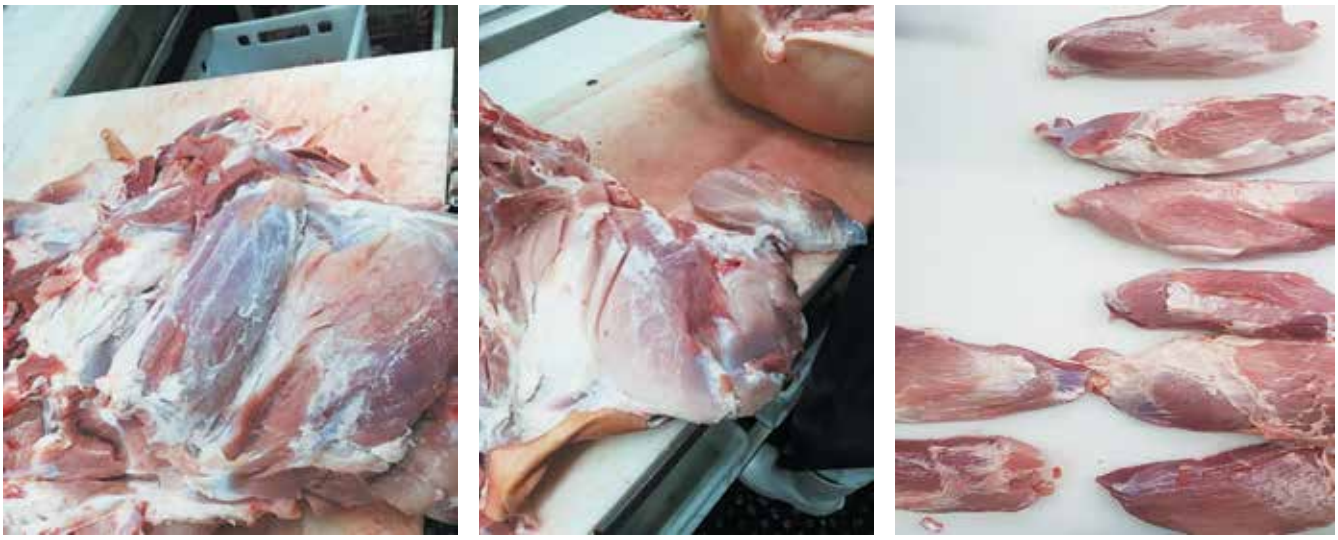
Sampling of muscle tissue was carried out in an industrial enterprise that slaughtered hybrid pigs (LWxDxL) in the quantity of 800 heads/day. Immediately after slaughter, 60 hot porcine carcasses with a weight of 85 ± 3 kg were taken for investigations. Twenty carcasses were randomly selected from 60 chilled porcine carcasses 24 hours after slaughter before cutting and deboning. Their leg cuts were sent to deboning to excise *m. semitendinosus*. To this end, a leg cut was opened by cutting intermuscular connective tissue interlayers with a knife providing access to *m. semitendinosus* (Figure 1a). Then, *m. semitendinosus* was excised without compromising muscle integrity (Figure 1b). After the muscle was excised, it was additionally cleaned from adjacent tissues (Figure 1c). Two muscles were excised from each carcass from the left and right leg parts, respectively.

When sampling, each muscle was cut in half for visibility of the boundary between the dark and light parts (Figure 2a). To this end, a knife was moved at first through "red" meat and then through "white" meat. After that, pieces with a size of about (2.5–3.0) × (2.5–3.0) × (2.5–3.5) cm were cut out from the dark and light parts of each sample (Figure 2b) for histological investigations. The rest of flesh was divided into "red" and "white" meat and minced by forcing meat through a grinder plate with 3 mm holes. The obtained minced meat (Figure 2c) was used for investigations of the functional-technological characteristics.

Histological investigations

To study the microstructure, muscle tissue samples taken from the dark and light parts of each muscle were fixed in the 10% neutral buffered formalin solution (BioVitrum, Russia) for 72 hours at room temperature (21 ± 1 °C). Two pieces (1.5 × 1.5 × 0.5 cm) with the longitudinal and cross orientation of muscle fibers were taken from each sample for the following study. The pieces were washed with cold running water for four hours and, after that, they were embedded in gelatin solutions (AppliChem GMBH, Germany) in an ascending concentration (12.5%, 25%) at a temperature of 37 ± 1 °C for 8 hours in each solution using a thermostat TS-1/20 SPU (Smolensk SKTB-SPU, Russia).

Preparation of serial sections with a thickness of 16 μm was carried out on a cryostat «MIKROM–HM525» (Thermo Scientific, USA). Three sections were made from each piece. The prepared sections were mounted on Menzel-



a) b) c)

Figure 1. Sampling during deboning of the leg cut:
a) opening the cut to excise *m. semitendinosus* (the oval marks the location of the muscle);
b) the leg cut after excision of *m. semitendinosus*; c) samples of *m. semitendinosus* after cleaning from adjacent tissues

a) b) c)

Figure 2. Sampling: a) appearance of cut *m. semitendinosus* (a boundary between the dark and light parts is well seen);
b) pieces cut out from the dark and light parts of *m. semitendinosus* for histological investigations;
c) appearance of minced meat obtained from meat of the dark and light parts of *m. semitendinosus*

Glaser slides (Thermo Scientific, USA) and stained with Ehrlich's hematoxylin and 1% aqueous-alcoholic solution of eosin (BioVitrum, Russia) by the conventional method [15]. Investigation of the histological preparations and their photographing were carried out using an Axio Imager A1 light microscope (Carl Zeiss, Germany) with the connected AxioCam MRc-5 camera (Carl Zeiss, Germany).

Morphometric investigations were carried out using the image analysis system AxioVision 4.7.1.0 (Carl Zeiss, Germany). The general scheme of the investigations corresponded to the methodology published earlier [9]. The diameter of muscle fibers, sarcomere length, and cross-sectional area of giant fibers were measured in the interactive mode. No less than 100 objects were calculated for each section. A fiber diameter was measured with an accuracy of ± 1.0 μm. A sarcomere length was determined with an accuracy of ± 0.1 μm. In addition, the number of giant fibers/1 cm^2 of the section and packing density of muscle fibers / 1 mm^2 were also calculated.

When studying cross sections of histological preparations, investigations were performed to determine a shape of muscle fibers, their packing density, condition of nuclei, thickness and condition of connective tissue layers, as well as to reveal giant fibers. In longitudinal sections, investigations were aimed to determining a condition and shape of muscle fibers, condition of sarcolemma, presence of striation (cross-striation and longitudinal striation), presence of destructive changes (ruptures, cracks, fragmentation) as well as to revealing knots of hyper-contraction.

To determine muscle fiber types, sections with cross orientation of muscle fibers were additionally stained with Sudan B according to MR001–00496254/00419779–2021[1] and PAS reaction (in Shabadash's modification) was performed

[1] MR001–00496254/00419779–2021. Performance of histological investigations on determination of myopathy. Approved by the director of L. K. Ernst Federal Science Center for Animal Husbandry N. A. Zinovieva and the director of V. M. Gorbatov Federal Research Center for Food Systems of RAS O. A. Kuznetsova, 2021, Moscow, 11 p.

according to [15]. Determination of muscle tissue types, their diameter and packing density was carried out.

Investigation of the functional-technological characteristics

Minced samples of *m. semitendinosus* obtained from its dark and light parts were investigated by the following methods:

— pH measurement by the potentiometric method using a pH-meter Testo 205 (Testo, Germany) with a measurement error of ±0.02; measurement was carried out in raw and cooked minced meat (minced meat was cooked as described below);

— determination of color characteristics CIELab — lightness (L*), redness (a*) and yellowness (b*) — with a spectrocolorimeter Konika Minolta CM-2300d (Konika Minolta, Japan). Before investigation, a spectrophotometer was calibrated using standard white and black plates;

— cooking losses in minced meat: before thermal treatment, 2% of nitrite salt with the a mass faction of sodium nitrite of 0.4% was added and mixed, cured samples with a weight of 100 g were placed into polymer packages, polymer packages were tightly tied, then packages with minced meat were placed into a water bath with an initial water temperature of 78 ± 1 °C. Thermal treatment was carried out at 76 ± 1 °C until reaching a temperature of 75 ± 1 °C in the geometrical center of a sample. After that, a package was opened and a moisture was removed; a piece of minced meat was placed on filter paper for chilling and draining. After chilling samples to room temperature, the samples were weighed. Weight losses were calculated in% to the initial weight of samples by a difference between weights before and after thermal treatment;

— structural-mechanical characteristics were determined using a texture analyzer "Structurometr ST-2" (Quality laboratory, Russia). For each test, a test portion with a size of no less than 2.5 × 2.5 × 3.9 cm was cut out from the core of the cooked sample; the test portion was placed in the testing field of a texture analyzer and subjected to compression between the lower unmovable platform and Bloom indenter fixed on the upper movable platform. Load force was registered upon introducing the indenter into the test portion to a depth of 20 mm at its speed of movement (introduction) of 1.0 mm/s, after touch force of 7 g, using the software of the texture analyzer. The maximum value of load force on the indenter expressed in grams was taken as a result of the test.

All investigations of functional-technological characteristics were carried out in triplicate.

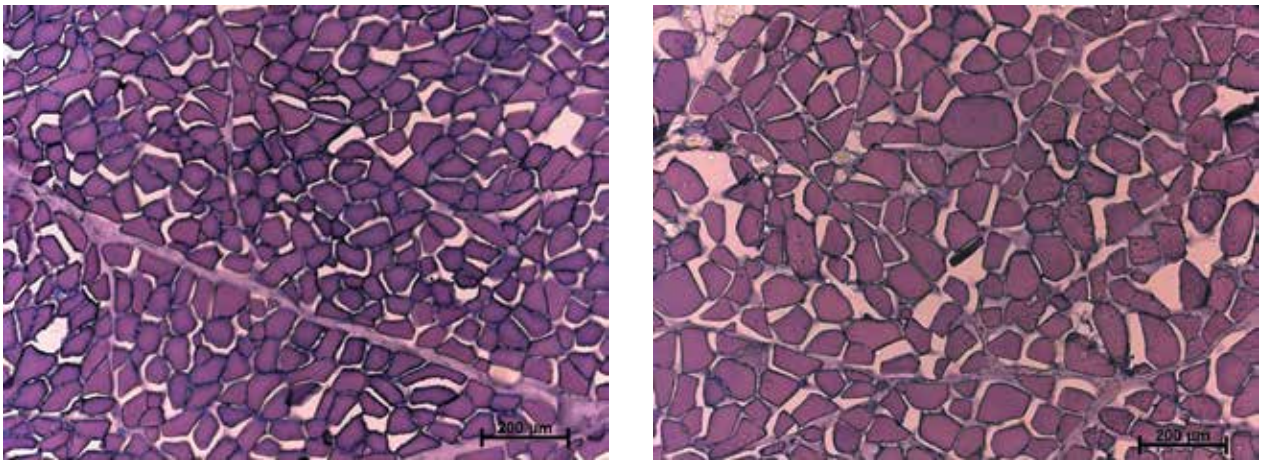
Statistical analysis

Statistical analysis of the experimental data was carried out using the software R (version 4.3.0). Quantitative data are presented as the arithmetic mean (Mean), standard deviation (SD), standard error of the mean (SE), minimum and maximum values (Min/Max), interquartile range (P 25/75), confidence interval (CI) и median. The Kolmogorov–Smirnov test was applied to assess the normality of distribution of parameters of quantitative variables. The coefficient of variation (CV) was used as the main method for assessing parameters of distribution. Correlation between indicators of muscle fibers and data of standard investigations was evaluated by the Pearson parametric method. Differences were considered significant and the presence of a relationship between parameters was recognized at a probability level of not higher than 0.05.

Results and discussion

Results of the histological investigations

All studied samples both from the dark part and from the light part of *m. semitendinosus* (Figure 3) exhibited the uniform condition of muscle tissue. On the cross section, muscle fibers had the polygonal or weakly round shape. The interlayers of the endomysium were well defined; the boundaries between individual muscle fibers were revealed without major difficulties. Giant fibers with a round-oval shape and large diameter were observed.



a) b)

Figure 3. Typical microstructure of the samples of the dark (a) and light (b) parts of *m. semitendinosus* (cross section, staining with hematoxylin and eosin; magnification: 10 x, scale bar = 200 µm)

In the longitudinal section, most muscle fibers showed clearly defined cross-striation and straightened shape. Individual wavy fibers with longitudinal striation were revealed, which pointed to the presence of contraction zones.

The nuclei in muscle fibers were well stained, had the oval shape and were located directly under the sarcolemma.

The connective tissue interlayers of the perimysium were wavy, tightly adjacent to the bundles of muscle fibers. The nuclei in the connective tissue interlayers were clearly seen in the histological preparations. Individual adipocytes or their small groups having the typical histological structure were found between the bundles of muscle fibers in the areas of the perimysium.

The functional condition of muscle tissue in each group of samples was quite uniform. Sporadic cross microcracks and ruptures of sarcolemma were noticed. Destruction of myofibrils, multiple ruptures and fragmentation of "normal" muscle fibers were not found. Destruction of sarcomeres and appearance of cracks and ruptures of fibers were observed in the knots of hyper-contraction (giant fibers on the longitudinal section).

Statistical processing of the morphometry results for the samples taken from the dark and light parts of *m. semitendinosus* is presented in Tables 1–3.

The dark part of the muscle was characterized by a higher packing density of fibers on the cross section, higher number of capillaries and sarcomere length. It is necessary to note that the sarcomere length is interrelated in a complicated way with biochemical reactions of proteolysis and metabolism of glycogen. The sarcomere length also influences the palatability of prepared meat and water holding capacity of meat products [16].

On the contrary, the light part was characterized by a higher diameter of muscle fibers with higher variability of this indicator, although high variability of this indicator (CV higher than 20%) was also observed in the dark part of the muscle. All differences by the main morphometric indicators were significant ($p < 0.05$).

Figure 4 presents a histogram and "heat map" for the distribution of muscle fibers of the dark and light parts of *m. semitendinosus* created for each of the studied samples (N = 20). The character of distribution of muscle fibers by diameter in the dark and light parts of *m. semitendinosus* differed in all samples. Comparison of the histogram data (Figures 4a, 4b) showed that the scatter by fiber diameter was shifted to the left and a range of variability was less pronounced in the dark part compared to the samples from the light part of the muscle. Such a narrow range of variability of the attribute can be associated with a lower effect of the internal and external factors on the muscle tissue condition before and after animal slaughter.

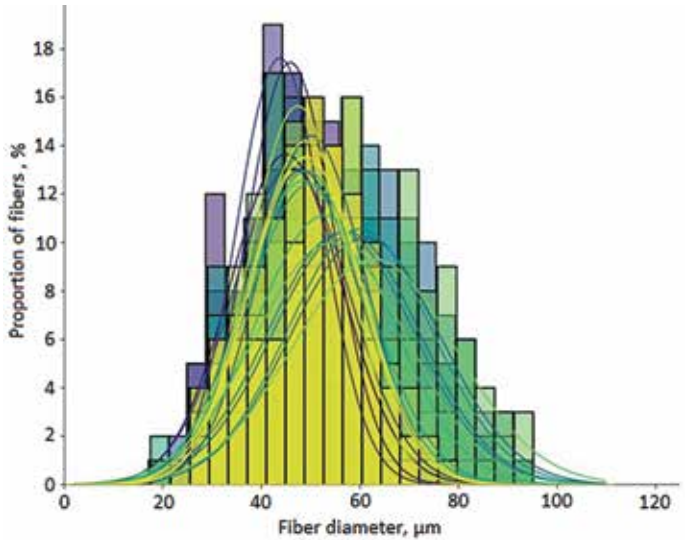
For pork, fibers with a lower diameter are especially desirable as they exert a beneficial effect on its quality and are considered an indicator of its tender structure [17]. An increase in the muscle fiber diameter reduces tenderness and water holding properties of meat [18]. Therefore, the dark part of the muscle represented by muscle fibers of lower diameter is more desirable in terms of technological characteristics.

The composition of muscle fibers is the main criterion for classifying muscles as "red" and "white". The study of the composition of muscle fibers allows predicting biochemical changes in muscle tissue and, consequently, meat quality as the rate of post mortem metabolism depends on a ratio between quantities of muscle fibers of different types (oxidative and glycolytic) [11].

Investigation of muscle fiber types (Table 2) in the *m. semitendinosus* samples showed that the proportion of type I, intermediate and type IIb fibers was higher by 9.3, 5.2 and 4.1%, respectively, in the dark part.

Table 1. Results of the statistical processing of the main morphometric characteristics of muscle tissue in the dark and light parts of *m. semitendinosus*

Statistical indicator	Value of the statistical indicator for			
	fiber diameter, μm	fiber density fibers/mm^2	sarcomere length, μm	Number of capillaries, capillaries/mm^2
Dark part				
Mean (Mean ± SE)	51.17 ± 0.31	273 ± 6	2.81 ± 0.03	34.0 ± 0.3
Min/Max	17.00/94.50	162/359	2.15/3.58	30/39
Interquartile range (P25/75)	41.50/60.11	239/310	2.58/3.06	33/36
Median	50.56	295	2.78	35
Confidence interval (CI)	0.60	12.80	0.07	0.59
Coefficient of variation (CV), %	26.79	18.49	12.20	6.80
Light part				
Mean (Mean ± SE)	59.54 ± 0.36	190 ± 4	2.12 ± 0.02	29.7 ± 0.4
Min/Max	18.43/99.37	128/253	1.74/2.88	24/38
Interquartile range (P25/75)	48.30/70.90	168/215	1.96/2.30	28/31
Median	60.20	188	2.01	30
Confidence interval (CI)	0.71	7.56	0.05	0.77
Coefficient of variation (CV), %	27.33	15.72	11.10	10.21



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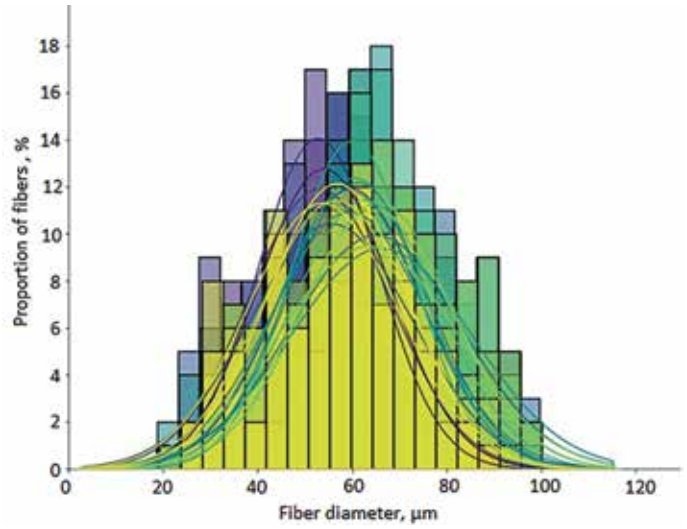

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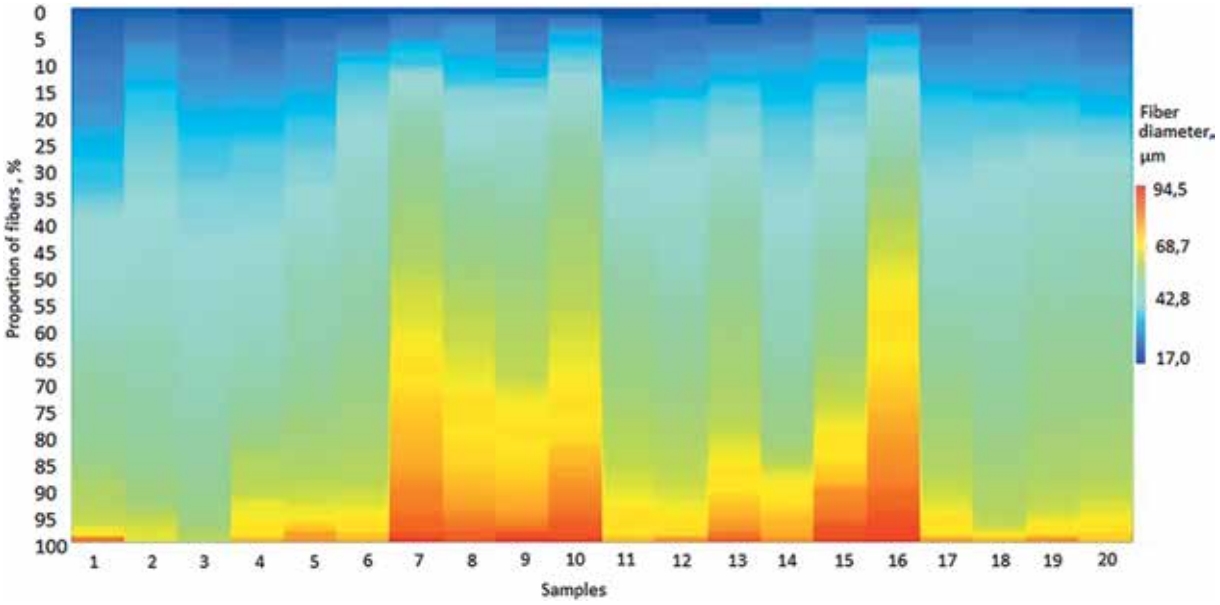

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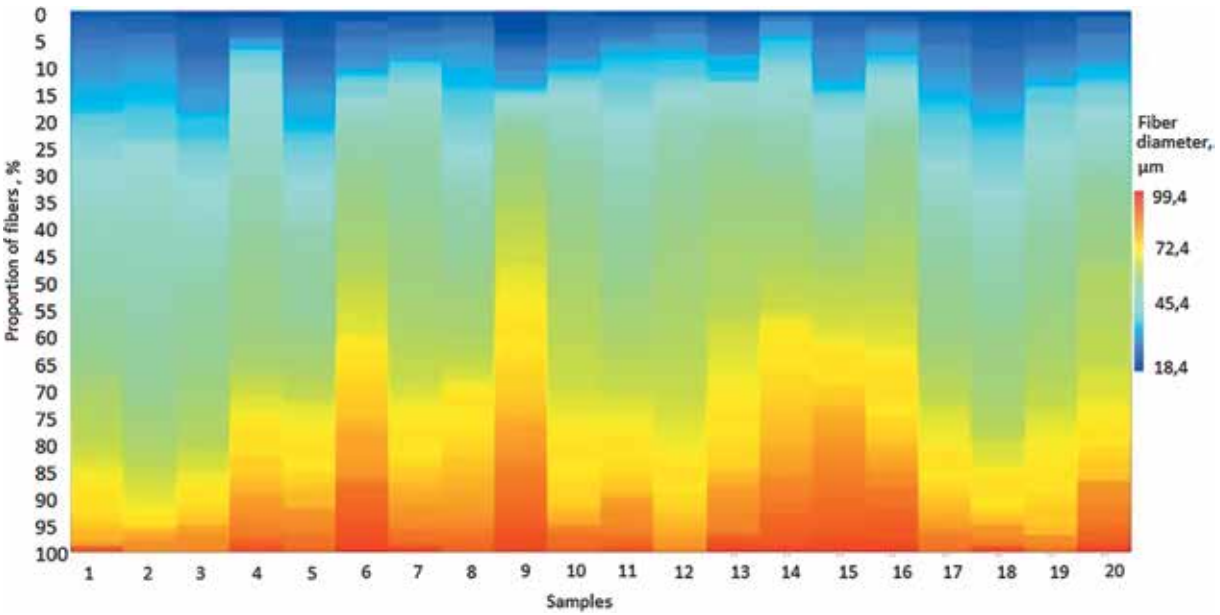

d)

Figure 4. Distribution (a, b — histogram, c, d — "heat map") of muscle fibers by diameter in the samples of *m. semitendinosus*: a, c — dark part and b, d — light part

Table 2. Differences in the dark and light parts of *m. semitendinosus* by muscle fiber types

Statistical indicator	Value of the indicator for			
	Dark part		Light part	
	Proportion, %	Diameter, μm	Proportion, %	Diameter, μm
Type I (oxidative) muscle fibers				
Mean (Mean ± SE)	19.4 ± 1.0	32.69 ± 0.24	10.1 ± 0.6	30.79 ± 0.27
Min/Max	11.0/25.0	17.01/43.26	5.0/16.0	18.44/38.77
Interquartile range (P25/75)	14.8/23.0	29.61/36.41	8.0/11.25	28.30/33.60
Median	20.5	33.35	10.0	30.87
Confidence interval (CI)	1.98	0.48	1.20	0.53
Coefficient of variation (CV), %	23.31	14.74	27.05	12.38
Type IIa (intermediate) muscle fibers				
Mean (Mean ± SE)	15.8 ± 1.1	42.35 ± 0.14	11.0 ± 1.0	41.32 ± 0.20
Min/Max	8.0/25.0	36.12/48.92	5.0/17.0	34.06/48.08
Interquartile range (P25/75)	0.0/0.0	45.92/43.98	6.8/15.3	39.09/43.72
Median	15.00	42.44	10.0	41.70
Confidence interval (CI)	2.19	0.27	2.01	0.40
Coefficient of variation (CV), %	31.77	5.74	41.85	7.32
Type IIb (glycolytic) muscle fibers				
Mean (Mean ± SE)	64.9 ± 2.0	58.82 ± 0.28	79.0 ± 1.5	65.75 ± 0.30
Min/Max	52.0/81.0	42.46/94.50	67.0/89.0	44.23/99.37
Interquartile range (P25/75)	57.0/74.0	51.04/64.96	73.5/84.3	56.42/73.98
Median	64.5	56.89	80.5	64.34
Confidence interval (CI)	3.91	0.55	2.87	0.59
Coefficient of variation (CV), %	13.74	17.17	8.29	18.13

Type I and intermediate type fibers did not have significant differences in terms of diameter in the samples from the dark and light parts. At the same time, light muscle tissue demonstrated not only the relatively high content of IIb type fibers but also their larger size in terms of diameter (by about 12%).

Significant variability in the proportion of type I muscle fibers was observed both in the dark part and in the light part of the muscle (CV was 23.31 and 27.05%, respectively). Even higher variability (CV more than 30%) was revealed for intermediate (IIa) type fibers. With that, CV of this indicator in the light part of *m. semitendinosus* was more than 10% higher than the corresponding value in the dark part, which suggests heterogeneity of the muscle tissue samples from the light part of the muscle by the number of type IIa muscle fibers. On the contrary, moderate variability with regard to the proportion of IIb muscle types was observed both in the samples from the dark part and light part of the muscle. However, the light part was characterized by a higher value of the CV coefficient than the dark part (18.13 and 13.74%, respectively).

These differences are also clearly seen on the histograms of distribution of muscle fibers of different types by diameter that were built using the results of data analysis for each sample (Figure 5). Fiber diameters varied in all samples depending on their type; with that, the maximum individual variability was observed in the dark and light parts of the muscle for the diameter of type IIb fibers.

Difference in the ratio of muscle fiber types can be the main reason for different meat quality, including color and its stability, pH, water holding capacity. It is known that even the high content of glycogen in dark skeletal muscles of the oxidative type does not lead to the sharp post mortem drop in pH due to the low content (or absence) of

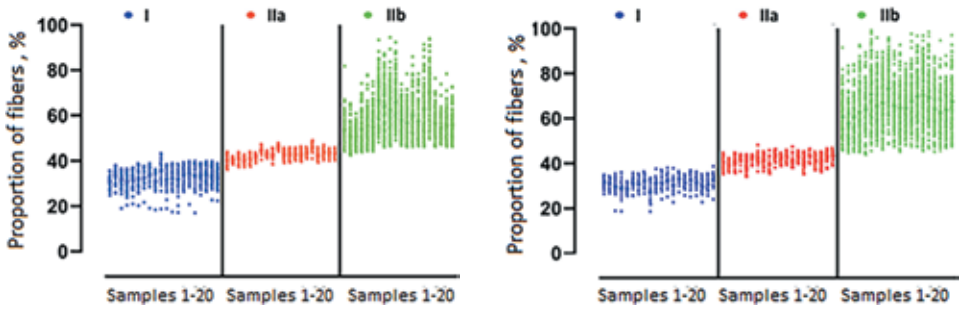

Figure 5. Distribution of muscle fibers by diameter in the samples of *m. semitendinosus*: a — dark part, b — light part

fibers of the glycolytic type [2]. With that, it is type I fibers that are considered to be linked with the high quality of pork [11,19].

The development of giant fibers is regarded as one of significant reasons for a decrease in the technological quality of pork. These structural abnormalities are linked with the fast growth of muscles and stress loads. Hypercontraction (giant fibers) is usually accompanied by a decrease in the capillary density and myoplasmic calcium loading. Typically, anaerobic fast-twitch muscles with low lactate metabolism but accelerated onset of rigor mortis are most prone to the development of giant fibers [20].

In our study, significant differences were established between the dark and light parts of *m. semitendinosus* by the quantity and size of giant fibers (Table 3).

As can be seen from Table 3, the light part was characterized by a larger quantity (more than 5 times) of giant fibers with the fibers of a larger size (almost by 11%). The high variability of the quantity of giant fibers (84.14%) in the dark part of the muscle apparently suggests that the presence of a significant quantity of giant fibers is not typical of the dark part: giant fibers were revealed only in three samples (5–6 fibers/cm^2 each). In the light part, the quantity of giant fibers was up to 20 fibers/cm^2. With that, the giant fibers in the light part of the muscle were characterized by an insignificant variability of sizes (CV < 20%). Appearance of giant fibers in the porcine muscle tissue due to the enhanced and long post mortem glycolysis is closely linked with the PSE defect [21,22] and suggests that the light part of *m. semitendinosus* is more prone to show poor functional-technological characteristics.

Results of the study of the functional-technological characteristics

An effect of histological indicators on pork quality is underestimated. Various studies continue to reveal interrelation between the microstructure of muscle tissue and functional-technological indicators, such as pH, cooking losses, sensory tenderness, color and others [23].

Meat color is one of the most important indicators of meat quality that influences a desire to consume it [24]. The color of the final product remains to be a criterion of freshness and quality for consumers, as well as a stimulus to make a buying decision [5]. The minced meat samples made from the dark part and light part of *m. semitendinosus* had significant differences in color both in the raw (Figure 2c) and in the cooked (Figure 6) state.

Results of the instrumental determination of color (Table 4) confirmed color differences between the samples in the raw and cooked state. In the raw samples, significant ($p<0.05$) differences were recorded in the mean values of lightness, redness and yellowness by 6.00, 4.68 and 3.01 units, respectively. Color differences (by 6.53, 2.99 and 1.8, respectively) also retained in the cooked samples after chilling for mean values of L*, a* и b* ($p<0.05$).

Table 3. Quantity of giant fibers and their area in the dark and light parts of *m. semitendinosus*

Statistical indicator	Value of the indicator for			
	Dark part		Light part	
	Quantity, fibers/cm^2	Area, μm^2	Quantity, fibers/cm^2	Area, μm^2
Mean (Mean ± SE)	2.1 ± 0.4	13,415.74 ± 398.18	11.9 ± 1.0	15,062.39 ± 190.53
Min/Max	0.0/6.0	7,621.51/20,102.66	5.0/20.0	9,233.93/23,737.56
Interquartile range (P25/75)	1.0/2.8	11,306.31/14,906.29	9.0/14.0	12,952.09/16,807.18
Median	2.0	13,051.34	12.5	14,947.44
Confidence interval (CI)	0.80	644.10	0.19	369.99
Coefficient of variation (CV), %	84.14	20.35	36.59	18.50

a) b)

Figure 6. Cooked minced meat samples from the dark and light parts of *m. Semitendinosus*:
a) appearance in packages immediately after thermal treatment; b) appearance on the cut surface after chilling
(left – samples from the dark part; right – samples from the light part of the muscle)

Table 4. Color characteristics of the *m. semitendinosus* samples in the raw and cooked state

Indicator	Value of the indicator for					
	Dark part			Light part		
	L	a*	b*	L	a*	b*
Raw minced meat						
Mean (Mean ± SE)	57.76 ± 0.80	7.92 ± 0.25	20.34 ± 0.14	63.76 ± 0.40	3.24 ± 0.17	17.33 ± 0.32
Min/Max	49.90/60.96	6.65/10.06	19.22/21.25	61.00/67.97	2.08/4.49	14.49/19.37
Interquartile range (P25/75)	56.22/60.23	7.17/8.31	19.99/20.88	62.38/64.90	2.58/3.83	16.14/18.66
Median	59.15	7.69	20.38	63.29	3.35	17.44
Confidence interval (CI)	1.56	0.48	0.28	0.79	0.32	0.64
Coefficient of variation (CV), %	6.02	13.49	3.01	2.74	22.28	8.15
Cooked minced meat						
Mean (Mean ± SE)	69.61 ± 0.39	8.08 ± 0.07	10.05 ± 0.10	76.44 ± 0.51	5.09 ± 0.17	11.86 ± 0.10
Min/Max	67.28/70.99	7.84/8.63	9.50/10.47	74.49/79.57	4.07/5.69	11.52/12.84
Interquartile range (P25/75)	69.29/70.52	7.89/8.15	9.78/10.30	75.39/76.89	4.53/5.61	11.66/11.94
Median	70.02	8.04	10.20	76.02	5.32	11.74
Confidence interval (CI)	0.76	0.14	0.20	1.01	0.34	0.2
Coefficient of variation (CV), %	1.93	3.11	3.51	2.33	11.65	2.96

Color differences between the minced meat samples point to the different content of muscle pigments in the dark and light parts [24] of *m. semitendinosus* and different amounts of nitrosomyoglobin formed upon curing with nitrite salt.

The highest variability of color characteristics of the samples was observed in raw minced meat for the redness values. On the contrary, the cooked samples were characterized by insignificant variability of color (CV from 1.93 to 3.51 excluding redness (11.65) for the samples from the light part of *m. semitendinosus*).

When processing meat, it is extremely important to reveal raw materials with lower technological quality. Measuring pH values is the main and the most often used criterion of quality [25,26].

Measurement of pH and statistical processing of the results (Table 5) show that the dark part of *m. semitendinosus* was characterized by higher pH values; with that, differences in pH between the dark and light parts were significant ($p < 0.05$) both for the raw and cooked samples.

It is believed that a pH range at 24 hours after slaughter of 5.7 ~ 6.1 is the most suitable for consumers and processors [26]. As can be seen from Table 5, the minimum values of pH of raw meat both in the dark and light samples were higher than 5.7, which indicated the absence of the PSE defect. For the samples from the light part of *m. semitendinosus*, the mean and maximum pH values were lower than 6.1, which is characteristic for pork with the normal course of autolysis. On the contrary, the samples from the dark part of *m. semitendinosus* showed the mean and maximum values of pH higher than 6.1, which is typical of DFD meat.

A slight increase (by 0.48 and 0.43, respectively) in the mean value of pH was observed in the cooked samples from the light and dark parts of *m. semitendinosus* as a result of thermal treatment. In general, however, the cooked samples from the dark part had a higher mean value of pH (by 0.30 units). The highest variability of pH values was characteristic of the samples from the dark part of the muscle both before and after thermal treatment. A pH value of a product also determines the shelf life of foods [27]. The understanding of the variability of this indicator in a product and reduction of its values due to the choice of raw materials can be used in combination with other hurdles to increase shelf life of pork products.

Cooking test is of great importance in pork quality assessment [28]. The dark and light samples did not show significant differences in cooking losses (Table 6). However, the tendency ($p = 0.052$) was observed toward higher losses in the samples of muscle tissue from the light part of *m. semitendinosus*: the mean values differed by 1.83% and medians by 1.43%. At the same time, high variability

Table 5. Results of pH changes in the samples of *m. semitendinosus* in the raw and cooked condition

Indicator	Value of the indicator for			
	Dark part		Light part	
	Raw minced meat	Cooked minced meat	Raw minced meat	Cooked minced meat
Mean (Mean ± SE)	6.15 ± 0.05	6.58 ± 0.03	5.80 ± 0.02	6.28 ± 0.01
Min/Max	5.74/6.63	6.23/6.98	5.69/5.93	6.15/6.38
Interquartile range (P25/75)	6.01/6.22	6.52/6.65	5.72/5.89	6.23/6.33
Median	6.16	6.58	5.79	6.29
Confidence interval (CI)	0.09	0.07	0.03	0.02
Coefficient of variation (CV), %	4.06	2.88	1.50	1.02

was observed for the value of cooking losses in all samples: 28.45% and 26.04%, respectively.

Table 6. Cooking losses (%) in the dark and light parts of *m. semitendinosus*

Indicator	Value of the indicator for	
	Dark part	Light part
Mean (Mean ± SE)	6.67 ± 0.60	8.50 ± 0.70
Min/Max	3.54/9.13	6.71/13.83
Interquartile range (P25/75)	5.76/7.94	6.83/8.76
Median	6.76	8.19
Confidence interval (CI)	1.17	1.37
Coefficient of variation (CV), %	28.45	26.04

Higher moisture losses upon cooking did not influence tightening of consistency of the samples made from the light part of *m. semitendinosus*. Tactile evaluation demonstrated that their consistency was less elastic, looser and more crumbly than that in the samples made from the dark part of *m. semitendinosus*.

The results of the structural-mechanical investigations of the samples of cooked minced meat confirmed this observation, and statistical data analysis showed that the differences in the structural-mechanical characteristics between the samples from the dark and light parts of *m. semitendinosus* were significant ($p < 0.05$) (Table 7).

Table 7. Results of determination of load force (g) in the cooked samples from the dark and light parts of *m. semitendinosus*

Indicator	Value of the indicator for	
	Dark part	Light part
Mean (Mean ± SE)	1,369.74 ± 112.54	1,984.78 ± 97.53
Min/Max	869.67/2,259.98	1,515.03/2,599.88
Interquartile range (P25/75)	1,030.73/1,712.12	1,666.64/2,333.49
Median	1,202.95	1,878.77
Confidence interval (CI)	220.57	191.15
Coefficient of variation (CV), %	31.82	19.03

It should be noted that the samples taken in an enterprise in our study were characterized by pH values higher than 5.7, which points to the absence of the PSE defect. Nevertheless, the light part demonstrated a reduction in the functional-technological characteristics of meat. Heat stress in pigs can lead to an increase in differences in the quality of the dark and light parts due to the differences between the dark and light parts of *m. semitendinosus* in the rate of the metabolic response to external factors [29].

At the same time, it is well known that some diets have a positive effect on reduction of the proportion of glycolytic muscle fibers in *m. semitendinosus*. Such diets include, for example, addition of microalgae [30], low content of amylose/amylopectin [31].

For the time being, however, it is acknowledged that the strongest effect on the microstructure of muscle tissue is exerted by a breed rather than a raising system. Less intensive growth of pigs shows lower quantity of glycolytic muscle fibers also influencing quality of *m. semitendinosus* [10].

Conclusion

Pork is the most consumed meat after poultry meat both in Russia and in the world. Consumers and processors are interested in increasing pork quality. This interest should stimulate animal husbandry to improve characteristics of industrially raised slaughter animals. Apparently, some special features of pork muscles, in particular, *m. semitendinosus*, must be taken into account in breeding work to produce meat raw materials with required technological characteristics for production of delicacy whole-muscle (whole-piece) products. Our study of *m. semitendinosus* showed significant differences in the microstructure of the dark and light parts of this muscle. The dark part was characterized by a higher density of muscle fibers, higher length of sarcomeres and higher number of capillaries. On the contrary, the light part was characterized by a larger diameter of muscle fibers with predominance of type IIa and type IIb fibers, as well as by a higher quantity of giant fibers, which presence is linked with the PSE defect of meat quality and myopathy. A significant range of variability of microstructural indicators in the light part, in particular, by fiber diameter, can be associated with less conservative heredity and a possibility of the further breeding work. Differences in the microstructure are the main reason for different quality of meat from the dark and light parts of *m. semitendinosus*. The dark part was characterized by higher pH values, better color development with nitrite and better consistency after cooking. With that, there was a tendency toward lower cooking losses. It should be noted that even at pH in a range of 5.7 ~ 6.1, which is optimal for pork, the special features of the microstructure of the light part of *m. semitendinosus* from domestic hybrid pigs can cause problems with quality of final products with regard to the development of color and consistency. Therefore, it is expedient to include *m. semitendinosus* along with *m. longissimus dorsi* into the plan of investigations. The results of such investigations can be useful for animal breeders and meat processors for the further improvement of quality of Russian pork and pork products.

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