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CHANGES IN PORK MUSCLES STRUCTURE PARAMETERS IN THE COURSE OF CURING AND PASTEURISATION

Summary. The aim of the study was to perform a comparative evaluation of the histological structure of the following pork ham muscles: the semimembranosus muscle (*musculus semimembranosus*) and the quadriceps muscle of thigh (*musculus quadriceps femoris*) and to compare the extent of these changes following the injection with a curing brine under the pressure of 0.5 MPa, plasticization (massaging) in vacuum conditions and pasteurisation. Muscles for investigations were collected from carcasses 24 h after slaughter. The injected curing brine contained, among others, sodium chloride and polyphosphates. In order to perform histometric measurements at each phase of the experiment, histological preparations were made which were stained with the assistance of the HE method (hematoxylin and eosin). Measurements were conducted with the assistance of the computer image analysis (CIA) using for this purpose MultiScan software. Microscope pictures were characterised on the basis of the following muscle fibres parameters: cell cross-section area, cell circumference, as well as Feret's H and V diameters. In addition, percentage proportion of muscle fibres and their quantity in the analysed field of vision were calculated. The shape of the fibres cross-section was determined on the basis of the ratio of Feret's H and V diameters. The results of all measurements were tested for the significance level at $p = 0.05$. The performed comparative analysis of structure parameters showed that the cross-section area of cells of the quadriceps muscle of thigh was smaller than that of the semimembranosus muscle. Its increase in the former was caused, primarily, by the pasteurisation process, while in the second one – by the injection with brine. The massaging process resulted in more regular shapes of cells of the quadriceps muscle of thigh than of those of the semimembranosus muscle as evidenced by the value of the H/V quotient close to 1. Following pasteurisation, both the area and the circumference of the muscle fibres of the *m. quadriceps femoris* increased further, while the value of H/V in both muscles did not differ statistically significantly despite significant differences after massaging.

Key words: computer image analysis, muscle structure, curing, plasticization, pasteurisation

Introduction

Muscle fibres parameters provide one of the basic elements of meat histological structure associated with the quality of final products manufactured from whole muscles (AGUILERA 2005). Their size is connected with the anatomical position of a given muscle in the animal carcass. There are differences between individual animal muscles regarding their histological structure (WIKLUND et AL. 1998), texture (SHACKELFORD et AL. 1995) as well as in their susceptibility to the process of plasticization (MOTYCKA and BECHTEL 1983, SHACKELFORD et AL. 1989.). Morphological parameters of structural muscle parameters change under the influence of after slaughter maturation (MESTRE PRATES et AL. 2002, SOTELO et AL. 2004), as a result of technological processes to which meat is subjected in the course of processing as well as types of chemical compounds making up the curing mixture. The technological and technical curing conditions (LACHOWICZ et AL. 2003, SOBCZAK et AL. 2004.) and thermal treatment (PALKA 2004, TORNBERG 2005) exert a significant impact on the dynamics and direction of meat tissue structure changes. Brine which is applied intramuscularly remains in interfibrillar spaces in quantities which depend on the type of the raw material (GAJEWSKA-SZCZERBAL and KRZYWDZIŃSKA-BARTKOWIAK 2005, KRZYWDZIŃSKA-BARTKOWIAK and GAJEWSKA-SZCZERBAL 2007). The plasticization process – by loosening and destroying muscle structures – increases the sorption of the curing brine and diffusion of intrafibrillar proteins into intracellular spaces (LACHOWICZ et AL. 2003, XARAGAỳO et AL. 1998).

The aim of this study was a comparative assessment of the histological structure of swine ham muscles, namely: the semimembranosus (*musculus semimembranosus*) and thigh quadriceps (*musculus quadriceps femoris*) muscles and the range of its changes as a result of the introduction of the curing brine, plasticization and thermal treatment.

Material and methods

Muscles for analyses were collected 24 h after slaughter and divided into two parts, one of which was treated as the control, while the other – the experimental sample – was injected with curing brine which contained, among others, 5.0% sodium chloride and 0.5% polyphosphates converted into P_2O_5. The experimental samples injected with the brine were plasticized in 95% vacuum and then pasteurised in steel cans at the temperature of 72°C in the can geometric centre measuring the temperature with the assistance of a thermocouple sensor. Histological preparations were prepared at each of the four phases of the experiment staining them with the assistance of hematoxylin and eosin (HE). Histometric measurements were performed with the aid of the computer image analysis (CIA) using for this purpose the MultiScan software. The characterisation of the microscopic images was carried out on the basis of the following muscle fibres parameters: cell cross-section area, their circumference and Feret's H and V diameters. The fibres cross-section shape was determined on the basis of the ratio of the Feret's H and V diameters. In addition, the percentage proportion of muscle fibres and their quantity in the analysed field of vision were calculated. All the measurements were carried out three times from 10 fields of vision of the microscope eyepiece. The obtained results were tested for the significance level of $p = 0.05$.

Results and discussion

The results of parameter measurements of the porcine *musculus semimembranosus* and *musculus quadriceps femoris* structures are presented in Figures 1-6 as well as in Table 1. Using a visual assessment system, it was found that the scope of changes depended on the evaluated muscle and the stage of experiment.

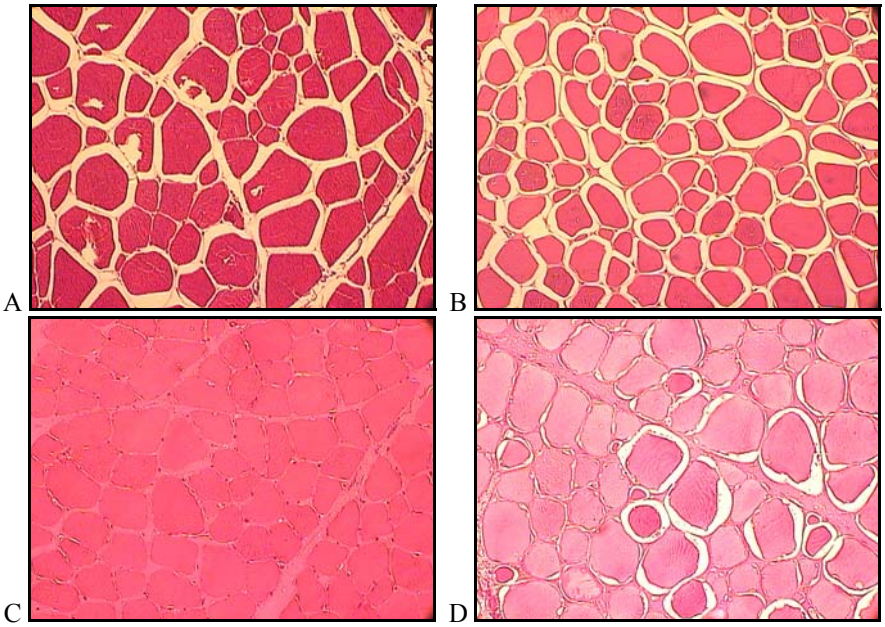

Fig. 1. Microstructure of the *musculus quadriceps femoris*: A – raw, B – injected with brine, C – plasticized, D – pasteurised
Rys. 1. Mikrostruktura *musculus quadriceps femoris*: A – surowy, B – nastrzyknięty solanką peklującą, C – plastyfikowany, D – pasteryzowany

Differences in the meat histological structure and its constitution depending on the animal species and/or type of muscle were reported earlier by other researchers, among others: LIU et AL. (1996), ORYL (2004), GAJOWIECKI et AL. (2001) and LACHOWICZ et AL. (1997). Analyzing the results obtained with the assistance of the computer image analysis it was found that the raw semimembranosus muscle was characterised by a greater area and circumference of muscle fibres and a fairly regular shape (H/V = 0.97) in relation to the thigh quadriceps muscle (Table 1). Similar experiments for raw muscles were conducted earlier and a similar correlation was obtained (GAJEWSKA-SZCZERBAL et AL. 2007).

The process of injection with the curing brine increased the cell capacity in the semimembranosus muscle in comparison with the raw muscle (Figs. 2, 3). This is confirmed by the measurement results of the cross section area and circumference of muscle fibres carried out in raw and injected muscles between which statistically significant differences were observed (Figs. 3, 4).

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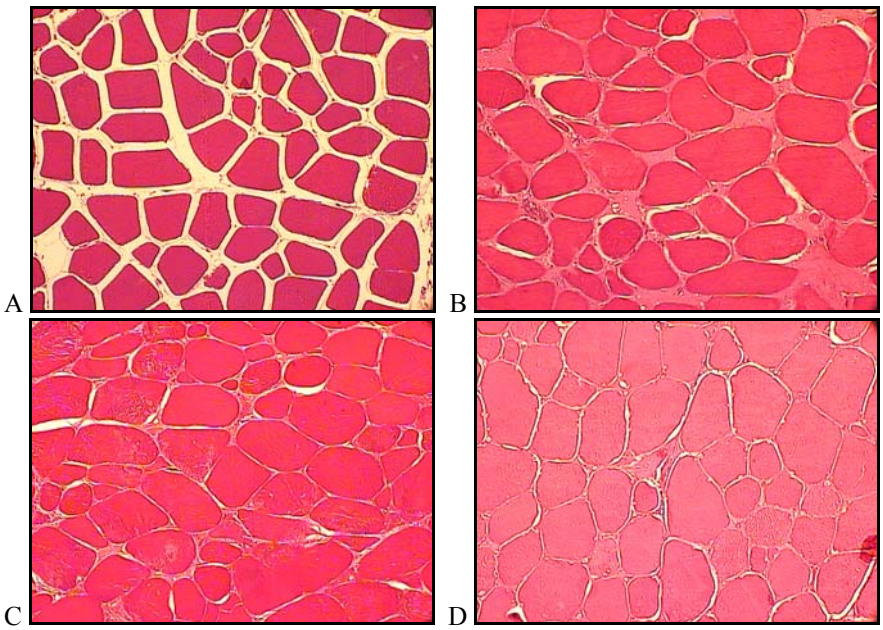

Fig. 2 Microstructure of the *musculus semimembranosus*: A – raw, B – injected with brine, C – plasticized, D – pasteurised
Rys. 2. Mikrostruktura *musculus semimembranosus*: A – surowy, B – nastrzyknięty solanką peklującą, C – plastyfikowany, D – pasteryzowany

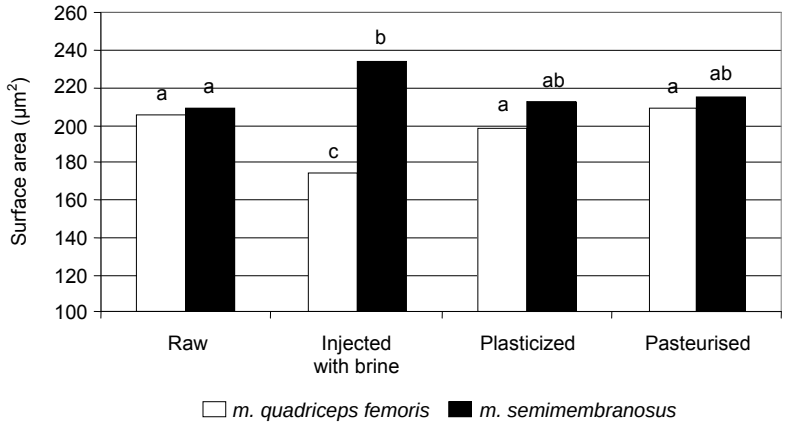

Fig. 3. Changes in the surface area (CSA) of muscle fibres caused by: the injection with brine, massaging and pasteurisation
Rys. 3. Zmiany powierzchni przekroju włókien mięśniowych spowodowane przez nastrzyk solanką peklującą, masowanie i pasteryzację

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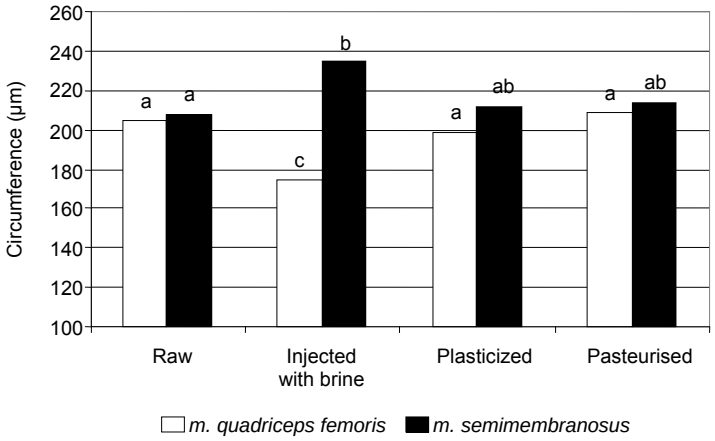

Fig. 4. Changes in the circumference of muscle fibres caused by: the injection with brine, massaging and pasteurisation
Rys. 4. Zmiany obwodu włókien mięśniowych spowodowane poprzez nastrzyk solanką peklującą, masowanie i pasteryzację

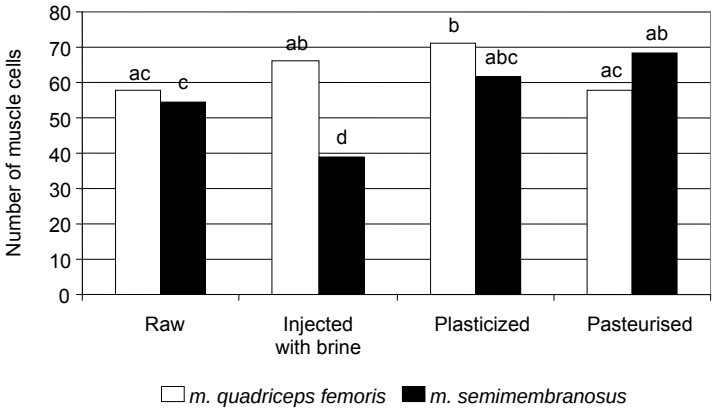

Fig. 5. Number of muscle cells in the field of view of the examined muscles: raw, injected with brine, massaged and pasteurised
Rys. 5. Liczba komórek mięśniowych w polu obrazu badanych mięśni: surowych, nastrzykniętych solanką peklującą, masowanych i pasteryzowanych

The increased area of muscle fibres in the semimembranosus muscle caused a simultaneous decline of their numbers in the microscopic field of vision (Fig. 5). The applied massaging and pasteurisation processes caused shrinkage of fibres and increased the amount of muscle cells counted in the field of vision and the area of the examined picture occupied by them. In contrast to the semimembranosus muscle, the injection of the thigh quadriceps muscle with the curing brine caused shrinkage and decrease of the cross-section area of its cells (Fig. 1). On the other hand, their numbers, circumference

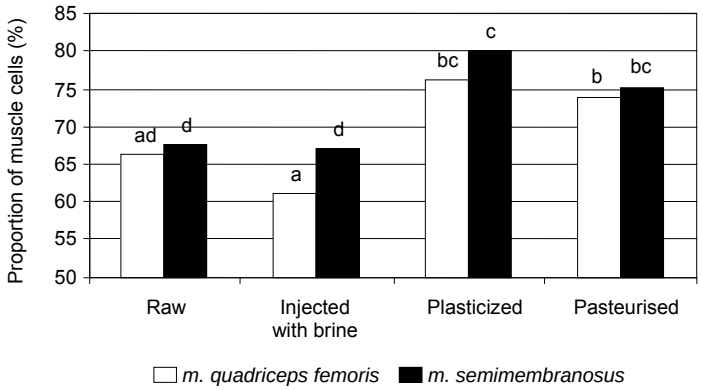

Fig. 6. Proportion of muscle cells in the field of view of the examined muscles: raw, injected with brine, massaged and pasteurised
Rys. 6. Zawartość komórek mięśniowych w polu obrazu badanych mięśni: surowych, nastrzykniętych solanką peklującą, masowanych i pasteryzowanych

Table 1. The effect of injection, massaging and pasteurisation on changes in structure parameters of *musculus quadriceps femoris* (I) and *musculus semimembranosus* (II)
Tabela 1. Wpływ nastrzyku, masowania i pasteryzacji na zmiany parametrów struktury *musculus quadriceps femoris* (I) i *musculus semimembranosus* (II)

Experiment's phase	The kind of muscle	Feret's diameter		
		H (μm)	V (μm)	H/V
Raw muscles	I	54.9^{ab} ±4.1	72.7^{a} ±6.9	0.75
	II	63.6^{abc} ±6.9	65.7^{abc} ±3.7	0.97
Muscles injected with brine	I	52.9^{a} ±3.4	59.5^{de} ±5.2	0.93
	II	76.2^{d} ±12.9	68.4^{ab} ±8.2	1.11
Massaged muscles	I	65.2^{bc} ±2.8	59.5^{de} ±5.2	1.09
	II	68.1^{cd} ±11.9	62.2^{bcde} ±6.4	0.91
Pasteurised muscles	I	65.7^{bcd} ±1.9	64.0^{bcd} ±2.4	1.02
	II	64.1^{bc} ±13.2	59.2^{cde} ±5.8	1.1

The same letters designate mean values which do not differ significantly at the level of $p \leq 0.05$.

and areas were found increased following the plasticization process. This fact appears to indicate that the penetration of the majority of brine into the cells of the semimembranosus muscle took place after the injection with the curing brine, whereas in the case of the thigh quadriceps muscle, only during the massaging process. However, the values of the above-mentioned histometric characteristics failed to differ statistically significantly in comparison with raw muscles (Figs. 3, 4, 5, 6).

Muscle fibres exposed to heat shrink as water derived both from intracellular spaces as well as from muscle fibres themselves are liberated. This phenomenon is caused by the denaturation and coagulation of sarcoplasmatic and myofibrillars proteins, as well as by reduced capability to hold water immobilized within myofibrils (SHACKELFORD et AL. 1989). In the presented experiments, the applied pasteurisation process consolidated the structure which developed in the course of the plasticization process.

Conclusions

1. A significant influence was determined of the curing brine constituents, as well as of the mechanical treatments and thermal processing on changes of the histometric parameters of the examined muscle structure.

2. When comparing the structure elements of both ham muscles, i.e.: the semimembranosus (*musculus semimembranosus*) and thigh quadriceps (*musculus quadriceps femoris*) muscles, it was found that the first of them was characterised by higher values of the examined characteristics in comparison with the second one.

3. Fibres of the semimembranosus muscle were more susceptible to the brine absorption during injection and plasticization processes than the cells of the thigh quadriceps muscle.

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ZMIANY PARAMETRÓW STRUKTURY MIĘSA WIEPRZOWEGO PODCZAS PEKLOWANIA I PASTERYZACJI

Streszczenie. Celem pracy była porównawcza ocena struktury histologicznej świńskich mięśni szynkowych: półbłoniastego (*musculus semimembranosus*) i czterogłowego uda (*musculus quadriceps femoris*) oraz zakresu jej zmian podczas nastrzyku solanką peklującą pod ciśnieniem 0,5 MPa, plastyfikacji w atmosferze próżni oraz pasteryzacji. Mięśnie pobierano z tusz po 24 h od uboju. W skład solanki nastrzykowej wchodziły m.in. chlorek sodu i wielofosforany. W celu wykonania pomiarów histometrycznych w każdej fazie doświadczenia sporządzano preparaty histologiczne, wybarwione metodą HE (hematoksyliną i eozyną). Pomiary wykonywano za pomocą komputerowej analizy obrazu (KAO), stosując program MultiScan. Obrazy mikroskopowe scharakteryzowano na podstawie następujących parametrów włókien mięśniowych: powierzchnia przekroju poprzecznego komórek, ich obwód oraz średnice Fereta H i V. Oprócz tego obliczono procentowy udział włókien mięśniowych oraz ich liczbę w analizowanym polu widzenia. Ze stosunku średnic Fereta H i V określono kształt przekroju włókien. Wyniki wszystkich pomiarów

testowano dla poziomu istotności p = 0,05. Analiza porównawcza parametrów struktury wykazała, że powierzchnia przekroju komórek mięśnia czterogłowego uda była mniejsza niż półbłoniastego. Na jej zwiększenie w pierwszym z mięśni wpłynął przede wszystkim proces pasteryzacji, natomiast w drugim – nastrzyk solanką. Proces masowania spowodował uzyskanie bardziej regularnych kształtów komórek mięśnia czterogłowego uda niż w mięśniu półbłoniastym, czego wyrazem była wartość ilorazu H/V zbliżona do 1. Po pasteryzacji nastąpiło dalsze zwiększenie powierzchni i obwodu włókien mięśniowych *m. quadriceps femoris*, a wartości H/V w obu mięśniach nie różniły się statystycznie mimo istotnych różnic po masowaniu.

Słowa kluczowe: komputerowa analiza obrazu, struktura mięśni, peklowanie, plastyfikacja, pasteryzacja

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