Effects of Carcass Weight and Muscle on Texture, Structure, Rheological Properties and Myofibre Characteristics of Roe Deer

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Abstract: Myofibre characteristic, texture, structure and rheological properties of selected muscles (BF, SM and L) of roe deer of different carcass weight were compared. Muscle texture and rheological properties was determined with the TPA and relaxation test, respectively, performed with a Instron 1140 device. Fibre type percentage and structural elements (muscle fibre cross sectional area and perimysium thickness) were measured in muscle samples using a computer image analysis programme. Of the muscles tested, the highest hardness, chewiness, sum of viscous and elastic moduli and the lower tenderness were found in BF which, at the same time, showed the highest fibre cross sectional area and the thickest perimysium. The most delicate histological structure and the lowest percentage of type I fibres as well as the lowest hardness, cohesiveness, chewiness and sum of viscous moduli were found in L. The young roe deer muscles showed lower percentage of I fibre type as well as lower values of textural parameters, while the sum of viscous and elastic moduli were higher than in old roe deer muscles. The muscle fibre cross sectional area and the perimysium thickness of young animal muscles were lower than those in the meat of older roe deer.

Key words: Venison, histology, muscle fibre types, mechanical properties of meat

INTRODUCTION

Meat quality can be defined as the total degree of satisfaction that meat gives the consumer[12] and can be assessed by measuring biophysical and histochemical properties. Żochowska et al.[39] and Żochowska et al.[40] observed correlation in wild boar meat between fibre diameter, connective tissue thickness, fibre type composition and meat texture parameters values. Some more studies suggest relationships between fiber type or size and texture or eating quality, especially tenderness in beef[6,25,31,34]. Although the results are variable and sometimes contradictory. No correlation between muscle fiber traits and tenderness were found by Whipple et al.,[37] however Klont et al.,[15] suggest relationships exist between muscle fiber type and meat quality, particularly in pork. Muscle fibre size and composition, is on one hand specific for different slaughter animal breed or lines and, on the other, it could be affected by growth rate[10,23,30]. However, no data on muscle fibre characteristic in roe deer of different carcass weight and structure, textural and rheological properties of their muscles could be found simultaneously in the available literature.

The objectives in this study were to compare fibre type, structure, texture and rheological properties in muscles of roe deer and to study the effect of the carcass weight on properties of three muscles: Biceps Femoris (BF), Semimembranosus (SM) and Longissimus (L) in roe deer.

MATERIALS AND METHODS

Source of animals: A total of eight carcasses from female of roe deer, of two different ages (four carcasses in each group), shot during winter in an enclosed area in the forest of the Western Pomeranian District were used. The carcass weights of the animals were 10±1 and 15±1 kg, while their ages were 0.5 and 2 years, respectively.

Carcass and sample preparation: Shortly after being shot (30-45 min) 1×1×0.5 cm samples were taken from the mid-part of muscles: Biceps Femoris (BF), Semimembranosus (SM) and Longissimus (L) frozen in liquid nitrogen and stored at -80°C for muscle fibre characteristics analysis. Carcasses were transferred to

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cold room of the Agricultural University of Szczecin. Half-carcasses of the experimental animals, kept at 4°C for 48 h from the moment of shooting were used to obtain the following muscles: BF, SM and L.

About 4 cm thick slices were cut perpendicularly to the fibres from each muscle. The muscles were placed in thermoresistant plastic bags and cooked in water at 85°C until the geometric centre reached 68°C. The cooked samples were cooled under tap water to about 12°C, wrapped in plastic prevent desiccation and stored at 4°C for 12 h.

Objective measurement of meat structure and texture myofibre classification and measurements: Myofibre characteristics were made on liquid-nitrogen-frozen samples of muscle. In order to classify the muscle fibres into type I, IIA and IIB groups, cross sections (10 μm) were cut at -26°C with a cryostat HM 505 EV. The sections were placed on glass slides, stained using the myosin ATPase method[11] with an alkaline preincubation solution (pH 10.4) and classified according to Brooke and Kaiser[4] into three groups: type I (slow oxidative), type IIA (fast oxidative-glycolitic) and type IIB (fast glycolytic).

Stained sections were examined with the image analysis system using a computer program (Multi Scan Base v.13). The following parameters were computed: percentage of different fibre types (%) (type I, type IIA and type IIB) per muscle fibre bundle and more than 10 bundles were examined for each muscle sample. A magnification of 100x was used.

Structure elements measurements: Histological assays were made on frozen samples cut from the BF, SM and L muscles of both groups of animals. The muscles were sectioned with a cryostat. The sections were placed on glass slides, contrast-stained with hematoxylin and eosin and sealed with Canada balsam[5].

The Multi Scan Base v.13 computer image analysis software was used to evaluate the mean fibre cross sectional area and perimysium thickness. The structural elements were measured in an area of fibre bundle and more than 200 muscle fibre and perimysium thickness/samples were analyzed.

Texture measurements: Texture measurements were made on the cooked meat at about 18°C. After removal of the plastic sheets, 20±2 mm thick slices were cut out from each sample to determine their texture on an Instron 1140 apparatus interfaced with a computer. The texture was evaluated using the double penetration test. The test involved driving a 9,6 mm diameter shaft twice, parallel to the muscle fibre direction into a sample down to 80% of its height (16 mm), using a crosshead speed of 50 mm min^{-1} and a load cell of 50 N. The force-deformation curve obtained served to calculate meat hardness, cohesiveness and chewiness[1]. The procedure was repeated 9-14 times on each sample.

Rheological characteristics: Rheological properties were determined with the relaxation test run on the Instron 1140. A sample was compressed to 10% of its original height (2 mm) and left for 90 s. Time-dependent changes in stresses were recorded by the computer every second during the first 15 s and at 15 s intervals thereafter.

The calculate the elastic and viscous moduli, the general Maxwell's body model was used, the model involving a parallel coupling of a Hookes body and two Maxwell's bodies. The following relaxation equation was applied:

$$\sigma = \varepsilon \cdot \left[E_0 + E_1 \cdot \exp\left(\frac{-E_1 \cdot t}{\mu_1} \right) + E_2 \cdot \exp\left(\frac{-E_2 \cdot t}{\mu_2} \right) \right]$$

where: σ, stress, ε, strain, E_0, E_1, E_2 elasticity moduli of Hook's body and of the first and second Maxwell's bodies, respectively, μ_1, μ_2, viscosity moduli of the first and second Maxwell's bodies, respectively, t, time.

Calculated values of the three elastic moduli are summarised in the figures as their sum; similarly, the values of the two viscous moduli are presented as their sum. The relaxation test was run 3 times on each muscle. During textural and rheological measurements, the muscle fibres were aligned in the direction of force.

Sensory texture evaluation: The sensory evaluation of the meat samples was assessed by a trained expert panel of 4 members with, in general, a minimum of four years experience in texture analysis of meat and meat products. The meat tenderness, game flavor, juiciness were assessed using a 5-points scale (Diagram 1).

Diagram 1: The 5-points sensory evaluation scale

Traits	1 point	2 points	3 points	4 points	5 points
Tenderness	The toughest	Tough	Average tough	Tender	The most tender
Perceptible of game flavor	Easily perceptible	Perceptible	Average perceptible	Weakly perceptible	Imperceptible
Juiciness	The most dry	Dry	Average dry	Juicy	The most juicy

Statistical analyses: Statistical analyses of the data involved the calculation of the mean values and standard deviations (SD) for each muscle and each group of roe deer. All the calculations were performed with Statistica® v.5.0 PL software.

RESULTS AND DISCUSSION

As shown in Table 1, the highest percentage of type I and IIB and lower percentage of types IIA fibres were found, regardless of the animal group, in the BF. Lower percentage of red and white fibres was characterized SM muscle, whereas L was the muscle with the high percentage type IIA fibre. Muscles from the roe deer of 15 kg carcass weight, compared to those from the younger animals showed a higher percentage of type I and type IIA fibres. Our findings are in agreement with those found for wild boar muscles by Żochowska et al.,[39], Żochowska et al.,[40], Ruusunen and Puolanne[30] and for cattle by Picard et al.,[28] and Brandstetter et al.,[3] who have shown that muscles of older animals compared to young ones contain a higher percentage of type I fibres. According to Swatland[35] muscle fibres undergo a continual alteration throughout life and that fibre type merely reflects the constitution of a fibre at any particular time.

When the roe deer muscles structure were compared Table 2, BF was characterized by the highest mean fibre cross sectional area and had the thickest perimysium. Lower values of this elements were found in SM muscle and L was characterized by the most delicate histological structure. Also Żochowska et al.,[39] showed that the BF of wild boar is

characterized by a less delicate structure than the other muscles tested. Whereas numerous authors comparing different domestic animal species have reported higher cross sectional areas or fibre diameters and thicker connective tissue in BF compared to SM[7,20,29].

A comparison of the values of the muscle structure elements in both groups of animals showed that the muscles from the young animals consisted of fibres of lower (by about 33-63%) cross sectional areas and had thinner by about 38-92%) perimysium than muscles from the roe deer of 15 kg carcass weight Table 2. The effect of age on meat structure has been the subject of research of Żochowska et al.,[39] and Żochowska et al.,[40], who also found an increase in average muscle fibre diameter and connective tissue thickness with increasing carcass weight of wild boar. Also Nishimura et al.,[26] found, in beef muscles an increase in the thickness of the collagen fibres in the perimysium with an increase in cattle weight. According to Fang et al.,[9] in pigs the perimysium thickens as a result of the increase in the number and thickness of perimysial sheets, which become thicker with growth.

Differences in muscle structure or different contents of red and white fibres in the roe deer muscles tested could be connected with the differences in the textural and rheological parameters observed in this study. Of all the muscles tested, the highest values of hardness, viscous and elastic moduli as well as the high chewiness were recorded in the BF muscle, the lowest values of this parameters including cohesiveness typically being in the L muscle Table 3 and 4. No

Table 1: Mean fibre type percentage of selected roe deer muscles

Muscle	Carcass weight (kg)											
	10						15					
	I (%)		IIA (%)		IIB (%)		I (%)		IIA (%)		IIB (%)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BF	20.78	1.29	30.02	1.05	49.20	2.31	25.33	1.88	29.33	1.71	45.34	2.36
SM	18.04	1.87	38.49	2.03	43.47	2.64	21.50	2.48	37.09	1.50	41.41	3.08
L	12.17	0.97	40.61	1.96	47.22	1.97	15.90	2.06	39.00	2.70	45.10	1.57

Table 2: Mean values of muscle structure elements of roe deer

Muscle	Carcass weight (kg)							
	10				15			
	Mean fibre cross sectional area (μm^2)		Perimysium thickness (μm)		Mean fibre cross sectional area (μm^2)		Perimysium thickness (μm)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BF	1487	87.9	13.79	3.1	2036	142.3	20.60	2.4
SM	1309	101.6	11.06	2.3	1862	106.5	16.94	1.7
L	1274	95.7	9.28	1.6	1605	142.6	15.60	2.6

Table 3: Mean values of muscle texture parameters of roe deer

Muscle	Carcass weight (kg)											
	10						15					
	Hardness (N)		Cohesiveness (-)		Chewiness (N×cm)		Hardness (N)		Cohesiveness (-)		Chewiness (N×cm)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BF	31.20	3.28	0.387	0.02	14.49	1.98	36.35	4.12	0.459	0.04	18.69	3.15
SM	27.61	1.67	0.412	0.03	13.42	2.42	33.60	2.74	0.387	0.03	14.30	2.04
L	24.11	2.02	0.341	0.03	9.46	2.16	28.67	3.06	0.354	0.02	10.66	2.15

Table 4: Rheological properties of roe deer muscles

Muscle	Carcass weight (kg)							
	10				15			
	Sum of viscous moduli (kPa)		Sum of elastic moduli (kPa×s)		Sum of viscous moduli (kPa)		Sum of elastic moduli (kPa×s)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BF	23252	785.9	359.1	20.6	20781	951.3	297.2	12.6
SM	21458	846.7	338.0	31.8	19037	1031.5	251.0	21.3
L	18443	1054.8	293.7	27.0	19344	641.3	267.7	14.0

Table 5: Sensory properties of roe deer muscles

Muscle	Carcass weight (kg)											
	10						15					
	Tenderness (pt.)		Game flavor (pt.)		Juiciness (pt.)		Tenderness (pt.)		Game flavor (pt.)		Juiciness (pt.)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BF	3.50	0.25	3.50	0.01	3.25	0.01	4.50	0.50	3.25	0.01	2.75	0.25
SM	2.75	0.01	3.00	0.25	4.25	0.01	3.75	0.25	1.75	0.25	3.75	0.50
L	2.75	0.25	3.75	0.01	3.00	0.25	3.50	0.01	2.50	0.01	2.00	0.25

significant differences in the textural parameters and rheological properties were found between SM and BF muscles. No information about comparison of muscle texture and rheological properties in roe deer of different weight was found in the present literature. However, numerous authors comparing muscles of wild boars[20,39,40] or different species of farm animals[8,21,32] have reported a similar order of hardness for muscles and showed BF to be tougher that either SM or L. Whereas higher values of viscosity moduli of BF compared with other ham muscles tested was reported also by Żochowska *et al.*,[40] for wild boars and Lachowicz *et al.*,[22] for pork meat.

A comparison among the textural parameters and rheological properties of the muscles from roe deer of different carcass weight showed that the old animal muscles were characterized by higher hardness and chewiness than the young ones, whereas cohesiveness was not dependent on carcass weight Table 3. Meat obtained from young roe deer compared to those from animal of 15 kg carcass weight showed higher values of viscous and elasticity moduli Table 4. Results obtained in this study are in agreement with results obtained for wild boars by Żochowska *et al.*[39], Żochowska *et al.*[40] and for farm animals by Bouton *et al.*[2], Shorthose and Harris[33] and Kołczak *et al.*[18], who observed that muscles from older animals were tougher than those obtained from the younger ones.

The sensory properties of the muscles studied Table 5 was evidenced also by data obtained by objective methods. Regardless of carcass weight, BF was the muscle with lowest tenderness and perceptible of game flavor. The most tender was L, the most juicy and with the highest perceptible of game flavor was SM. The lowest tenderness and juiciness and the highest perceptible of game flavor being typical of roe deer of 15 kg carcass weight muscles, compared to young animal muscles Table 5.

The differences in texture, rheological and sensory properties between roe deer of different carcass weight muscles, demonstrated in this study, according to numerous authors may have resulted from differences

in structural elements and a higher hardness being related to a thicker connective tissue[9,24,27,39], or/and a higher mean fibre cross sectional area[36,39]. The differences may have resulted also from different composition and properties of muscles proteins and lipids in particular muscle fibre types[38], or according to Dransfield[7] and Koohmaraie[19] higher content of calpastatin in red fibres being the most important factors. As shown by Karlsson *et al.,*[13] type I fibres contained neutral lipids, whereas only about 26% of type IIA and 1% of type IIB fibres contained neutral lipids and the same time had a higher content triglyceride which may be one factor of importance for meat quality, especially for meat tenderness. However according to Kłosowska[16], red fibres are characterized by thicker connective tissue, so the higher hardness of roe deer muscles with a high percentage of type I fibres found in this study could be connected with a thicker endomysium. Also Karlsson *et al.,*[13] and Kłosowska and Fiedler[17] reported that a higher percentage of white muscle fibres in muscles was inversely proportional to the shear forces.

To sum up, it can be concluded that BF is characterized by fibres of higher cross sectional areas, thicker perimysium; it is also harder, viscous and elastic, less chewy than the SM and also than the L, a muscle with the most delicate structure and at the same time with the lowest values of texture parameters, rheological properties, juiciness and the highest tenderness. Higher hardness and lower tenderness, juiciness, game flavor and values of rheological properties of muscles from older animals could also be connected with thicker perimysium, fibres of higher cross sectional area and probably with a higher content of red fibres, compared with muscles from animals of smaller carcass weight.

CONCLUSIONS

The highest values of hardness, chewiness, viscous and elastic moduli, the highest mean cross sectional area, the thickest perimysium were typical of BF, regardless of the animal group tested, the lowest values being recorded in L.

Compared with older roe deer muscles, those of young animals showed a lower mean fibre cross sectional area, a thinner perimysium and a higher percentage of type IIB fibres; they were also less hard, tender, juicy, viscous, elastic and easier to chew.

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