

INVESTIGATIONS ON THE HISTOLOGICAL STRUCTURE OF THE SUPERFICIAL PECTORAL MUSCLE IN COBB-500 COMMERCIAL MEAT-TYPE HYBRID HEN

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Received June 22, 2009

ABSTRACT - There are few data concerning the somatic muscles and carcass quality of commercial hybrid hens. Meat quality is determined by a variety of factors, including fibre thickness and muscle fascicles. For the elucidation of some of the above-mentioned elements, we have collected histological samples of the superficial pectoral muscle from 10 individuals, males and females, belonging to the COBB-500 commercial meat-type poultry hybrid. Samples were processed by using the "paraffin section" technique, 30 histological blades being obtained. They were studied with the MC₃ - type photon microscope, while the thickness of fibres and of muscle fascicles was measured by using cito- and histometric techniques. The obtained results were myocyte thickness that ranged from 31.6 to 34.08 μ , according to gender. The thickness of the primary muscle fascicles ranged from 320.25 to 354 μ and of the secondary muscle fascicles,

from 568.25 to 628.5 μ . No notable differences were found between the two genders, some being statistically significant, while others were not.

Key words: myocyte, fascicles, muscle, pectoral, hen

REZUMAT - Cercetări privind structura histologică a mușchiului pectoral superficial la hibridul comercial de găină pentru carne – Cobb-500. La hibridii comerciali de găină pentru carne se cunosc puține date privind calitatea musculaturii somatice și, implicit, a carcaselor. Calitatea cărnii este determinată de o multitudine de factori, printre care și grosimea fibrelor și a fasciculelor musculare ce o compun. Pentru elucidarea, fie și parțială, a unora dintre elementele mai sus menționate s-au recoltat probe histologice din mușchiul pectoral superficial de la 10 indivizi, masculi și femele, aparținând hibridului comercial de

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găină pentru carne COBB-500. Probele au fost procesate după tehnica "secționării la parafină", obținându-se 30 de lame histologice. Acestea au fost studiate la un microscop fonic binocular de tip MC₃, măsurându-se prin tehnici cito- și histometrice, grosimea fibrelor și fasciculelor musculare. S-au obținut următoarele rezultate: grosimea miocitelor a variat între 31,6 și 34,08μ, în funcție de sex; grosimea fasciculelor musculare a fost de 320,25-354μ, pentru cele de ordinul I și de 568,25-628,5μ, pentru cele de ordinul II. Între cele două sexe s-au găsit diferențe notabile, unele fiind semnificative statistic, iar altele nu.

Cuvinte cheie: miocite, fascicule, mușchi, pectoral, găină

INTRODUCTION

New food sources and raw matters are reclaimed by the quick and uncontrolled increase of human population and of its higher life standards, as well. Meat is found on the top of food matters and nutrition resources, providing high nutritional and biological value, being also easy to be produced.

Numerous animal species, breeds and hybrids are raised and used by humans for obtaining meat as raw matter and food, under proper conditions. Poultry and, especially, poultry hybrids could provide high quality meat at large amounts, at relatively low prices. It is already known that the poultry meat has dietetic features and can be easily processed at a large variety of specific products. Nevertheless, there is little knowledge about the chicken hybrids bred for meat in Romania (COBB-500

or ROSS-308), mainly concerning those aspects related to the quality of somatic muscles from carcasses. Sporadic studies were carried out on the thickness of muscle fibres from some somatic muscles in poultry (chickens, ducks and geese) (Radu-Rusu et al., 2006; Radu-Rusu et al., 2007; Teușan, 2000). However, there are few scientific data concerning the above-mentioned hybrids. Our study is trying to bring some contributions to the elucidation of the studied theme.

MATERIALS AND METHODS

The used biological material included 10 individuals (five males and five females), belonging to the COBB-500 hybrid, slaughtered at 42 days old (***, 2008). Poultry had an average bodyweight of 2296 grams for males and 2168 grams for females. The obtained carcasses had the weight values between 1677 and 1795 grams, as influenced by gender. Histological samples were taken from each carcass, of about 1 cm³, from right and left superficial pectoral muscles (PS) (*Pectoralis superficialis*) (Sandu, 1995).

Anatomic and histological tools were used for sampling and processing the muscle tissue (knives, scalpels, clips, scissors, cotton pads, Berzelius glasses, graded cylinder, cap jars, batteries for fixation, clarification, dehydration, paraffin inclusion, oven, Leuckart frames, microtome, colouring pools, photonic binocular microscope, ocular and objective micrometer and digital photo camera). The used reagents are formalin 10%, ethylic alcohol, amylic alcohol, Mayer albumin, toluene, benzene, xilene, Canada balm and several histological

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colorants. The histological samples have been processed according to the paraffin sectioning technique, proceeding through the fixation, sizing, dehydration, clarification, inclusion, sectioning, spreading on glass smears, drying, coloration, mounting. A number of 30 histological smears have been obtained, using the "HE" bichromic coloration (hematoxylin-eosine). Smears were analysed at a MC₃ type-photonic binocular microscope, previously adjusted and calibrated for several ocular and objective associations: OC10 x OB6; OC10 x OB10; OC10 x OB20, several micrometric values being also calculated: 15.000, 9.011 and 4.441. The best sections from the analysed muscles were studied and micro-captured from the microscopic field. Small and great diameters of myocytes and of the first and second order muscle fascicles have been also analysed, through cito- and histometry (Radu-Rusu *et al.*, 2007; Teușan, 2000). Fibres were counted at each analysed muscle fascicle. Calculations were done in order to assess the average diameter of fibres and fascicles, the ratio between small and great diameters and the cross-sectional area of the histological elements. The following mathematical relations were used:

$$(1) D\bar{x} = \frac{LD + sd}{2} D, \text{ where } D\bar{x} = \text{mean}$$

diameter (μ); LD = large diameter (μ); sd = small diameter (μ)

$$(2) LD/sd \text{ ratio} = \frac{LD}{sd}$$

$$(3) C.s.a. = \frac{LD \times sd}{4} \times \pi, \text{ where } C.s.a. =$$

cross-sectional area (μ^2); π = coefficient

All the obtained data from measurements and calculations (750) have been statistically processed, the usual statistical estimators being initially

calculated (mean and mean standard error, variance, standard deviation and variability coefficient) (Sandu, 1995). The "ANOVA SINGLE FACTOR" algorithm was applied for testing the statistical significance of the difference between the means of studied traits (Sandu, 1995).

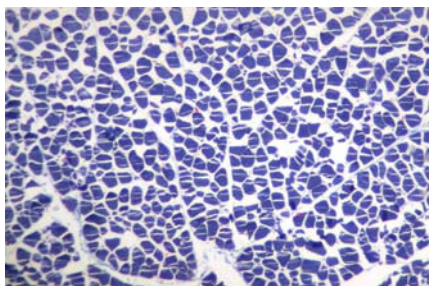
RESULTS AND DISCUSSION

The breast musculature is very developed in poultry, reaching 22-25% of the whole carcass weight, being influenced by species, breed, hybrid and gender. The pectoral musculature is represented by both superficial pectoral muscles (*Pectoralis superficialis dexter* and *Pectoralis superficialis sinister*) and by profound pectoral muscles (*Pectoralis profundus dexter* and *Pectoralis profundus sinister*) (Coțofan *et al.*, 2000; Gheție *et al.*, 1976; Nickel *et al.*, 1977).

These muscles are involved in the wing movement of all poultry species and breeds, having different dimensional, gravimetric, histological and physiological features. In meat-type hybrids, the pectoral muscle was especially studied as selection and genetic improvement character, reaching impressive sizes (Sandu, 1995). Superficial pectoral muscles have a parallelogram shape with dimensions of 12-15 cm in length and 7-8 cm in width and weights of 300-320 grams (in the COBB-500 broiler chickens) (Radu-Rusu *et al.*, 2008). As concerns the histological features, our investigations revealed a very interesting structural picture of the

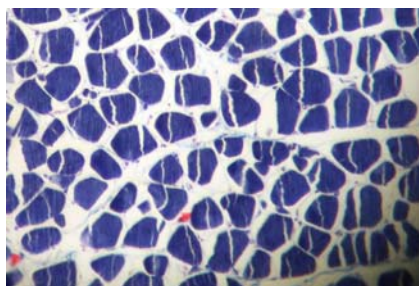
superficial pectoral muscles. Thus, the muscular fibres of these muscles are grouped in the second and the third order muscular fascicles. The myocytes from these muscles have cylindrical shape with ovoid ends and

lengths of centimetres. The thickness of these cells has been estimated by the measurement of both large and small diameters, by the calculation of the average diameter.

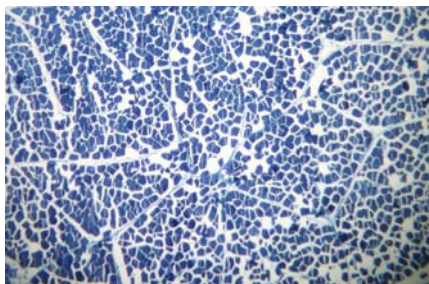


Histological sample from males
(40 X, OC 10 X OB 4)

Histological sample from males
(100 X, OC 10 X OB 10)



Histological sample from females
(40 X, OC 10 X OB 4)



Histological sample from females
(100 X, OC 10 X OB 10)

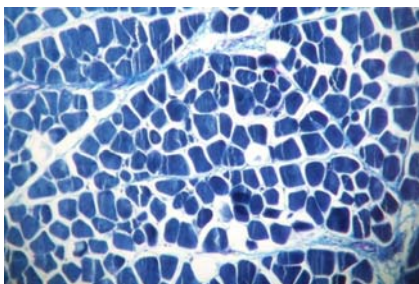


Fig. 1 – Myocytes and the first order muscular fascicles from the *Pectoralis superficialis* muscle of the COBB-500 hybrid

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In samples issued from male chickens, the myocytes of the analysed muscle (PS) had a large diameter of $39.58 \pm 0.63 \mu$ ($v=16.01\%$), a small diameter of $28.58 \pm 0.51 \mu$ ($v=17.80\%$) and an average diameter of $34.08 \pm 0.51 \mu$ ($v=15.07\%$) (Table 1) (Figure 1). The cross-sectional area had an average value of $904.02 \pm 26.54 \mu^2$ ($v=29.40\%$) (Table 1).

In female chickens, the muscular fibres of the pectoral superficial

muscle (PS) had a large diameter of $35.85 \pm 0.62 \mu$ ($v=17.32\%$), a small diameter of $27.35 \pm 0.57 \mu$ ($v=20.76\%$) and an average diameter of $31.60 \pm 0.53 \mu$ ($v=16.83\%$). The cross-sectional area of myocytes in the muscles (PS) of females had an average of $786.54 \pm 25.97 \mu^2$ ($v=33.02\%$), while their shape is slightly ellipsoid (Figure 1). (GD/Ds ratio = 1.342-1.405/1) (Table 1).

Table 1- Statistical indices concerning the myocyte thickness of the *Pectoralis superficialis* muscle from the COBB-500 hybrid chickens

Specification	Chicken gender	n	Statistical estimators			Variation limits	
			$\bar{x} \pm s \cdot \bar{x}$	s	V%	Minimum	Maximum
Myocyte large diameter (LD)	Male	100	39.58 ± 0.63	6.337	16.01	24.42	53.29
	Female	100	35.85 ± 0.62	6.208	17.32	22.20	46.63
Myocyte small diameter (Ds)	Male	100	28.58 ± 0.51	5.087	17.80	17.76	39.97
	Female	100	27.35 ± 0.57	5.679	20.76	13.32	39.97
Myocyte mean diameter (\bar{Dx})	Male	100	34.08 ± 0.51	5.136	15.07	22.20	46.63
	Female	100	31.60 ± 0.53	5.318	16.83	18.87	43.30
Myocyte LD/Ds ratio (x/1)	Male	100	1.405 \pm 0.022/1	0.218	15.50	1.00/1	2.00/1
	Female	100	1.342 \pm 0.03/1	0.256	19.14	1.00/1	2.25/1
Cross-sectional area of the muscular fibres	Male	100	904.02 ± 26.57	265.737	29.40	371.73	1672.90
	Female	100	786.54 ± 25.97	259.697	33.02	255.47	1463.83

As concerns the structure of the first order muscular fascicles from the pectoral superficial muscle, data presented in Table 2 have shown that their average thickness reached $320.25 \pm 14.63 \mu$ in males and $354 \pm 11.86 \mu$ in females, containing an average number of 48.9-50.43 myocytes, as gender influenced this character (Table 2). In the same muscle (PS), the second order muscular fascicles were obviously

greater, having a great diameter of $423 \pm 33.08 \mu$ in males and of $451.5 \pm 41.2 \mu$ in females (Table 3).

The average diameter of the second order muscular fascicles reached $569.25 \pm 37.58 \mu$ in males and $628.5 \pm 27.92 \mu$ in females. The ratio between the measured diameters indicates a strongly ellipsoidal shape (GD/Ds ratio = 1.82/1 – 1.956/1) of these second order muscular fascicles. Their average cross-sectional area was

found between 234111.55 and 284457.47 μ^2 , as influenced by chickens' gender (Table 3). From the analysis of data shown in Tables 1, 2 and 3, notable differences were found between genders, which are quantified in the following paragraphs. Thus, for the muscular fibres (myocytes) of the *Pectoralis superficialis* muscle, the values of the female gender were lower by 9.42% than of males for the large diameter, by 4.30% for the small diameter, by 7.28% for the average diameter and by 13% for the cross-sectional area of these myocytes (Table 4). This fact could be considered normal, knowing that the thickness of muscle cells is influenced, beside other factors, by

gender, in males being larger than in females (Teuşan, 2000).

The statistical analysis of the differences between genders has revealed a highly significant degree for great and average diameters and for the cross-sectional area, as well ($\hat{F} > F\alpha$ (1;198 FD). As concerns the small diameter, the differences had no statistical significance ($\hat{F} < F\alpha$ (1;198 FD). (Table 5). In the first and the second order muscular fascicles, females had higher values than males. Thus, the first order muscular fascicles were by 10.50-10.61% thicker in females than in males, containing a greater number of myocytes, by 3.13% (Table 4).

Table 2 - Statistical indices concerning the thickness and structure of the first order muscular fascicles from the *Pectoralis superficialis* muscle in the COBB-500 hybrid chickens

Specification	Chicken gender	n	Statistical estimators			Variation limits	
			$\bar{x} \pm s_x$	s	V%	Minimum	Maximum
MF ₁ * large diameter (LD) (μ)	Male	30	409.5 \pm 29.73	162.854	39.77	225	1020
	Female	30	452.5 \pm 24.06	131.777	29.12	240	900
MF ₁ small diameter (Ds) (μ)	Male	30	231.0 \pm 7.8	42.718	18.49	135	315
	Female	30	255.5 \pm 8.91	48.802	19.10	165	345
MF ₁ average diameter (\bar{D}_x) (μ)	Male	30	320.25 \pm 14.63	80.106	25.01	217.5	600.0
	Female	30	354.0 \pm 11.86	64.979	18.36	225	547.5
GD/Ds ratio of the FM ₁ (x/1)	Male	30	1.87 \pm 0.18/1	0.989	52.92	1.07/1	5.67/1
	Female	30	1.87 \pm 0.15/1	0.831	44.44	1.09/1	4.62/1
Number of m.f.** in the MF ₁ (n)	Male	30	48.9 \pm 3.17	17.377	35.54	23	82
	Female	30	50.43 \pm 2.28	12.511	24.81	22	78
Cross-sectional area of the MF ₁ (μ^2)	Male	30	73296.89 \pm 4906.19	26872.33	36.66	37110.06	144199.10
	Female	30	89717.99 \pm 4661.48	25831.99	28.46	39584.07	137837.38

*MF₁ = first order muscular fascicle; **m.f. = muscular fibres

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Table 3 - Statistical indices concerning the thickness and structure of the second order muscular fascicles of the *Pectoralis superficialis* muscle in the COBB-500 chicken hybrids

Specification	Chicken gender	n	Statistical estimators			Variation limits	
			$\bar{x} \pm s_x$	S	V%	Minimum	Maximum
MF _{II} * large diameter (LD) (μ)	Male	10	715.5±65.18	236.120	33.00	450	1125
	Female	10	805.5±41.90	132.486	16.45	570	1005
MF _{II} small diameter (Ds) (μ)	Male	10	423.0±33.08	104.594	24.73	240	600
	Female	10	451.5±41.20	130.278	28.85	240	720
MF _{II} average diameter (\bar{D}_x) (μ)	Male	10	569.25±37.58	118.872	20.88	420	750
	Female	10	628.5±27.92	88.303	14.05	517.5	787.5
GD/Ds ratio of the MF _{II} (x/1)	Male	10	1.82±0.25/1	0.788	43.28	1.00/1	3.00/1
	Female	10	1.956±0.26/1	0.810	41.42	1.06/1	3.94/1
Number of m.f.** in the MF _{II} (n)	Male	10	146.7±16.22	51.290	34.96	76	209
	Female	10	151.3±13.28	42.008	27.76	98	233
Number of MF _I in FMF _{II} (n)	Male	10	3.0±0.33	1.054	35.14	2	5
	Female	10	3.0±0.26	0.816	27.22	2	4
Cross-sectional area of the MF _{II} (μ^2)	Male	10	234111.55 ±2100.95	6643.780	2.84	113097.33	339998.86
	Female	10	284457.47 ±2888.88	9135.453	3.21	173180.29	483491.11

*MF_{II} = the second order muscular fascicle; **m.f. = muscular fibres

This situation is caused by the fact that muscular fibres are thinner in females, their density is higher and the pure muscular tissue proportion is higher to the prejudice of the connective tissues (lax and adipose). It is already known that there is an inverse proportion between the thickness and the density of muscular fibres from the skeletal muscles (Teuşan, 2000; Teuşan, 2000). At the second order muscular fascicles,

values have evolved similarly to those of the first order fascicles, meaning that the values of females were by 6.74%-21.51% higher than the values of males (Table 4). Statistically, almost all the differences found between males and females, at the first and the second order muscular fascicles, were not significant ($\hat{r} < F\alpha=0.05$ (1;58FD), ($\hat{r} < F\alpha=0.05$ (1;18FD)).

Table 4 - Thickness and structure of myocytes and of the first and the second order muscular fascicles from the *Pectoralis superficialis* muscle in the COBB-500 chicken hybrids, depending on gender

Specification	MU	Chicken gender				± % F vs. M	
		Males (M)		Females (F)			
		Absolute values	Relative values	Absolute values	Relative values		
At the myocyte level (m.f.)	Myocyte large diameter (LD)	μ	39.58	100	35.85	90.58	-9.42
	Myocyte small diameter (Ds)	μ	28.58	100	27.35	95.70	-4.30
	Myocyte mean diameter (\bar{Dx})	μ	34.08	100	31.60	92.72	-7.28
	Myocyte LD/Ds ratio	x/1	1.405/1	100	1.340/1	95.37	-4.63
	Cross-sectional area of the m.f.	μ ²	904.02	100	786.53	87.00	-13.00
	At the MF _I level*	MF _I * great diameter (LD)	μ	409.5	100	452.5	110.50
MF _I small diameter(Ds)		μ	231.0	100	255.5	110.61	+10.61
MF _I average diameter (\bar{Dx})		μ	320.25	100	354.0	110.54	+10.54
MF _I LD/Ds ratio (x/1)		x/1	1.87/1	100	1.87/1	100.0	--
Number of m.f.** in MF _I		n	48.90	100	50.43	103.13	+3.13
Cross-sectional area of the MF _I		μ ²	73296.89	100	89717.99	122.40	+22.40
At the MF _{II} level **	MF _{II} *** large diameter (LD)	μ	715.5	100	805.5	112.58	+12.58
	MF _{II} small diameter(Ds)	μ	423.0	100	451.5	106.74	+6.74
	MF _{II} average diameter (\bar{Dx})	μ	569.25	100	628.5	110.41	+10.41
	MF _{II} LD/Ds ratio of (x/1)	x/1	1.820/1	100	1.956/1	107.47	+7.47
	Number of m.f.** in the MF _{II} (n)	n	146.7	100	151.3	103.14	+3.14
	Number of MF _I in the MF _{II} (n)	n	3	100	3	100.0	--
	Cross-sectional area of the MF _{II}	μ ²	234111.55	100	284457.47	121.51	+21.51

*MF_I = the first order muscular fascicle;**m.f. = muscular fibres; ***MF_{II} = the second order muscular fascicle

CONCLUSIONS

The muscular fibres of the *Pectoralis superficialis* muscle of the COBB-500 hybrid are by 4.3-9.42% thinner in females than in males.

The muscular fascicles of the first and the second order from the *Pectoralis superficialis* muscle are thicker by 10.50-22.40% in females than in males, at the same studied hybrid.

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