

## ASPECTS CONCERNING THE HISTOLOGICAL STRUCTURE OF THE BICEPS BRACHIALIS MUSCLES IN CHICKEN BROILERS

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The researches goal was to assess the histological features of the Biceps brachialis muscles from chicken broilers reared within the intensive system. The biological material comprised 120 broilers, belonging to "COBB-500" (30♀ and 30♂) and "ROSS-308" (30♀ and 30♂), slaughtered at 42 days old. The muscular samples have been processed through formalin 10% fixation and paraffin impregnation then by serial sectioning and by HEA coloration. A digital trinocular Motic DMWB1-223 microscope has been used to study the smears and to run the micrometric measurements, via the Motic Image Plus ML software. The large and small diameters of the myocytes have been assessed then the average thickness and the cross-surface area have been calculated. The ANOVA single factor algorithm was used for statistical analysis. The results indicated higher values for the myocytes average thickness in ROSS-308 hybrid (32.19μ in males, 30.80μ in females, distinguished significance), respectively thinner texture in the muscles of COBB-500 chickens (27.90μ at cockerels and 26.51μ in pullets, distinguished significance). These data led to various values for the cross-section area, comprised between the limits of 547.95-601.59μ<sup>2</sup> (Cobb-500), respective of 739.96-801.80μ<sup>2</sup> (ROSS-308). The results revealed thinner texture of these muscles, compared with the results achieved during previous researches, which referred to the histology of white muscles or of red muscles from the rear limbs, in chicken broilers.

**Key words:** chicken broiler, *Biceps brachialis*, histology, structure, myocytes

### INTRODUCTION

Scientific literature, mainly the publications dealing with human nutrition and customer safety, emphasizes on aspects which pass over the quantitative side of meat production in poultry. While most of the data spread by the companies producing high value broilers refers to some technological and economical features of their products (microclimate, nutrition requirements, weight gains, FCR, slaughtering efficiency), this paper brings some partial results from a study onto the textural quality of the poultry meat produced by two of the most used hybrids in Romanian aviculture.

Previous researches [5] stated that the myocytes having mainly glycolitic metabolism, usually found in white muscles (pectoralis) are thicker than those with oxidative preponderant metabolism (red muscles – limbs) which have higher density. As compared to chicken broilers, in waterfowl domestic species (duck and goose), all skeletal muscles are mainly made of red fibers, which are thicker in wings [7, 8].

Although previous researches recommend pectoral muscles as high qualitative, mainly concerning those physical, chemical and nutritional features (pH value, high protein content, low energy level) [1, 6, 9, 10], this researches tried to find what are the histological – textural features of the wing muscles (*Biceps brachialis*), theoretically knowing that tenderness qualities should be better in red muscles (especially in wings and thighs ones) [2, 3, 4].

### MATERIALS AND METHODS

A group of 120 broilers, meaning 60 "Cobb 500" chickens (30♀ and 30♂) and 60 "ROSS-308" individuals (30♀ and 30♂), issued from two halls accommodating each 9500 chickens, have been used as biological material to sample muscular tissue from the *Biceps brachialis* muscles. The fowl have been fed with a classical corn-soymeal diet (3012 KCal ME and 24% CP-starter; 3175 KCal ME and 22.5% CP-grower; 3226KCal ME and 20% CP - finisher). Muscular samples

have been processed through formalin 10% fixation, paraffin impregnation at +56°C, microtome sectioning, mounting on blades, acid fuchsin and Evans blue coloration, resulting histological smears. These have been studied at a photonic microscope (Motic DMB1-30), endorsed with objective micrometer, ocular micrometer, micrometric grid and calibrated for three OBXOC associations: 10X10; 20X10 and 40X10. The ocular micrometer served to run the assessments, while an 8MP digital camera has been used to take microphotography shots. The measurements have been back-up through the usage of the built-in microscope software: Image Plus 2.0. The studies comprised cytometric and histometric measurements of myocytes, whose results have been introduced in several mathematical relations, in order to achieve some histological indexes: fibers mean thickness and cross section areas. The formulas are listed below:

(1) *mean thickness*: 
$$\overline{D}(\mu) = (D + d) / 2,$$

meaning: D=large diameter, d=small diameter;

(2) *ratio between large and small diameter* (shape index): 
$$\text{Ratio} = D/d$$

(3) *cross section area*: 
$$S(\mu^2) = D \times d \times \pi / 4,$$

meaning:  $\pi = 3.1416$ .

100 readings and/or computations have been carried on for each analyzed feature. The achieved values have been statistically processed running the ANOVA single factor algorithm, obtaining the main statistical estimators and the significance degree between for the differences between mean.

## RESULTS AND DISCUSSION

The acquired data concerning main histometric features of the *Biceps brachialis* muscles (fibers diameters, ratio between large and small diameters, cross-section areas) are presented in table 1. Microscopic imagery from both hybrids and genders is presented in fig. 1 (magnifications of 100 folds).

Thus, in “COBB-500” males, the myocytes thickness varied between 20.94 and 34.77 $\mu$ , with an average value of 27.90 $\pm$ 0.38 $\mu$ , while the variation coefficient reached 13.87%. Although the uniformity was poor, the fact is quite normal for this kind of assessments, knowing that, within the microscopic field, there are plenty of cells with multiple sizes and shapes. The ratio between large and small diameter was close to 1.5 (1.44/1), indicating an ellipsoidal shape of the contractile cells on cross-section. The area of the myocytes was found within the 331.11 – 914.82 $\mu^2$  interval, while the calculated average reached 601.59 $\pm$ 17.33 $\mu^2$  (variation increased = 28.80%, as absolute values geometrically progressed). In females muscles, histometric measurements revealed variation limits of 14.43-32.20 $\mu$  for the average diameter of the muscular fibers, while the calculated mean was situated at the 26.51 $\pm$ 0.34 $\mu$  ( $v=13.26\%$ ). The cellular shape index (ratio between small and large diameters) had lower average value than in males, meaning 1.34/1, indicating thus less ovoid shape. The cross-section surface of the myocytes reached an average value of 547.95 $\pm$ 13.34 $\mu^2$ . The data indicate the occurrence of the muscular hypertrophy in males, compared to the female broilers, fact which is translated through higher carcasses and meat yield at slaughtering. Statistically, the differences between genders were found as high significant ( $\hat{F} > F\alpha$  0.001 at 1;198 FD – myocytes large diameter), distinguished significant  $\hat{F} > F\alpha$  0.01 at 1;198 FD – myocytes average diameter and shape index) and just significant  $\hat{F} > F\alpha$  0.05 at 1;198 FD – myocytes cross-section area) (table 1).

Other researches revealed different values for the histometric features of the myocytes within the *Biceps brachialis* muscles, issued from different fowl categories. Thus, in domestic chicken (*Gallus bankiva*, var. *domesticus*), common breed, the average thickness reached 26.47 $\pm$ 0.45 $\mu$  [7, 11], while for the wings muscles of the domestic goose (*Anser anser*), the fibers were thicker (31.1 $\pm$ 0.45 $\mu$ ) [7].

Table 1  
 Thickness and cross-section area of the myocytes within the *Biceps brachialis* muscles

The hybrid	Gender	Statistical estimators	Large diameter (D) ( $\mu$ )	Small diameter (d) ( $\mu$ )	Average diameter ( $D \bar{X}$ ) ( $\mu$ )	DM/Dm ratio	Cross-section area ( $\mu^2$ )
"COBB 500"	♂	$\bar{X}$	32.73 <sup>d</sup>	23.07	27.90 <sup>c</sup>	1.44 <sup>c</sup>	601.59 <sup>b</sup>
		$\pm s_{\bar{x}}$	0.45	0.40	0.38	0.02	17.33
		V%	13.89	17.36	13.72	13.87	28.80
		Min.	24.78	16.43	20.94	1.15	331.11
		Max.	42.19	28.11	34.77	2.00	914.82
	♀	$\bar{X}$	30.17 <sup>a</sup>	22.86	26.51 <sup>a</sup>	1.34 <sup>a</sup>	547.95 <sup>a</sup>
		$\pm s_{\bar{x}}$	0.39	0.37	0.34	0.02	13.34
		V%	13.04	16.01	12.72	13.26	24.35
Min.		15.54	12.43	14.43	0.78	162.65	
"ROSS 308"	♂	$\bar{X}$	37.39 <sup>d</sup>	26.98	32.19 <sup>c</sup>	1.40 <sup>c</sup>	801.80 <sup>b</sup>
		$\pm s_{\bar{x}}$	0.46	0.42	0.40	0.02	20.76
		V%	12.24	15.57	12.27	12.38	25.89
		Min.	29.35	19.42	24.87	1.16	469.82
	♀	$\bar{X}$	34.83 <sup>a</sup>	26.77	30.80 <sup>a</sup>	1.31 <sup>a</sup>	739.96 <sup>a</sup>
		$\pm s_{\bar{x}}$	0.41	0.39	0.36	0.01	16.49
		V%	11.84	14.45	11.67	11.21	22.28
		Min.	17.94	14.98	16.46	0.82	211.04
		Max.	42.05	39.09	36.12	1.80	997.28

ANOVA test – for each character and hybrid, compared between genders:

<sup>ab</sup> significant differences ( $\hat{F} > F\alpha$  0.05 at 1;198 GL);

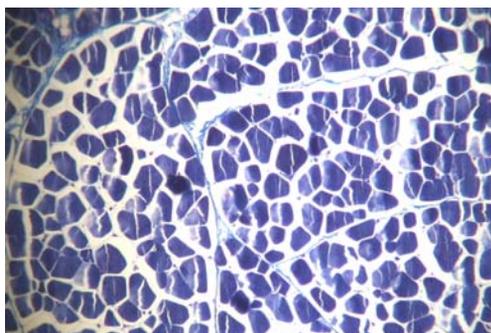
<sup>ac</sup> distinguished significant differences ( $\hat{F} > F\alpha$  0.01 at 1;198 FD);

<sup>ad</sup> high significant differences ( $\hat{F} > F\alpha$  0.001 at 1;198 FD)

Studying the smears issued from "ROSS-308" broilers, it was found that for males, the average diameter oscillated between the 24.87 $\mu$  and 40.10 $\mu$  limits, while the mean value was calculated at 32.19 $\pm$ 0.40 $\mu$  (table 1). The cells shape on cross section was close to those calculated at the "Cobb-500" hybrids, meaning 1.40/1. The cross surface area was calculated at an average value of 801.80 $\pm$ 20.76 $\mu^2$ . In females, the average thickness of the myocytes oscillated between 16.46 $\mu$  and 36.12 $\mu$ , with a mean calculated value was of 30.80 $\pm$ 0.36 $\mu$ . The shape index also indicated more rounded shape on section, while the area of the cross sections was found within the 211.04 $\mu^2$  and 997.28 $\mu^2$ , with an average value of 739.96 $\pm$ 16.49 $\mu^2$ . The variability oscillated between 11.67% (av. diameter in females) and 25.89% (cross-section area in males). Statistical computation revealed the same kind of

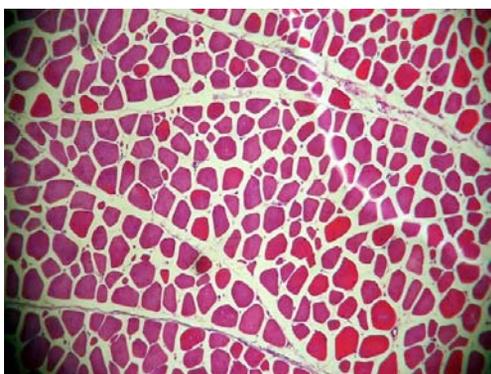
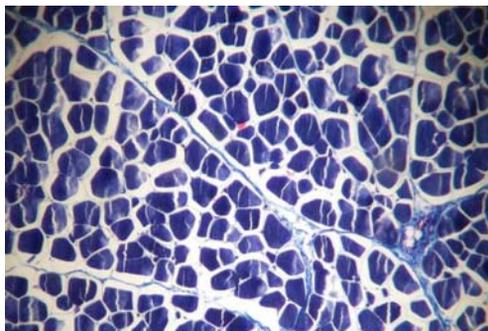
differences, as they were found in the other studied hybrid, meaning significant degree ( $\hat{F} > F\alpha$  0.05 at 1;198 FD – myocytes cross-section area), distinguished significant differences ( $\hat{F} > F\alpha$  0.01 at 1;198 FD – fibers average diameters and shape index) and high significance, as well ( $\hat{F} > F\alpha$  0.001 at 1;198 FD – myocytes large diameter). The hypertrophied myocytes in „ROSS-308” broilers indicated higher values of the histometric values, even than those in some waterfowl species [7].

The differences between the histometric features of the myocytes measured at the samples issued from the two studied hybrids have been also analyzed as percentage differentiations (table 2). It was found that the muscular fibers were hypertrophied in „ROSS-308” chickens, compared to the "Cobb-500" ones, no matter their gender.



“COBB – 500” broilers  
Sample from males  
(100 X, OC 10 X OB 10)

“COBB – 500” broilers  
Sample from females  
(100 X, OC 10 X OB 10)



“ROSS – 308” broilers  
Sample from males  
(100 X, OC 10 X OB 10)

“ROSS – 308” broilers  
Sample from females  
(100 X, OC 10 X OB 10)



**Fig. 1** –Myocytes and 1<sup>st</sup> order muscular fascicles within the *Biceps brachialis* muscle, sampled from both studied hybrids

Table 2

Comparisons between the myocytes histometric features of the *Biceps brachialis* muscles, sampled from both studied hybrids

Gender	The hybrid	Large diameter (D) (μ)	Small diameter (d) (μ)	Average diameter (D $\bar{X}$ ) (μ)	Cross-section area (μ <sup>2</sup> )
♂	"COBB-500"	32.73	23.07	27.90	601.59
	"ROSS-308"	37.39	26.98	32.19	801.80
	± "Ross" vs. "Cobb" (%)	+14.24%	+16.95%	+16.53%	+33.28%
♀	"COBB-500"	30.17	22.86	26.51	547.95
	"ROSS-308"	34.83	26.77	30.80	739.96
	± "Ross" vs. "Cobb" (%)	+15.44%	+17.10%	+16.18%	+35.04%

Thus, the average diameter was found 16.18%-16.53% higher, while the cross-surface area was 33.28%-35.04% increased in "ROSS-308" broilers. The analysis of variance has not been carried on between the absolute values achieved by both hybrids, due to the slight peculiar conditions of microclimate in the accommodation halls, therefore to certain degree of different environments, which could affect the ANOVA test relevance. Moreover, the aspects related to muscular fibers development must be further studied alongside with the existing ratio between connective and pure muscular tissue within muscles, in order to better estimate histological features of the skeletal muscles.

Compared to the thickness values of the contractile cells in other muscles, even white or red [7, 11], the mean diameter of the myocytes in *Biceps brachialis* muscles was found thinner.

## CONCLUSIONS

In both studied hybrids, the myocytes dimensions (average thickness and cross-surface area) were higher in males than in females, statistical significance occurring for the differences between calculated means.

It was found that "ROSS-308" chickens had thicker muscular fibers than the "COBB-500" ones, no matter the studied gender.

*Biceps brachialis* muscles proved to have the thinnest myocytes, compared to other studies which assessed the histometric features of breast, thighs and shanks muscles contractile cells.

However, the data concerning myocytes thickness and overall muscle tenderness must be considered in relation with the nutritional value assessments, to be able to achieve an appropriate picture of meat quality.

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