

# Comparison of breast muscle traits and meat quality characteristics in 2 commercial chicken hybrids

M. Petracci,\*<sup>1</sup> F. Sirri,\* M. Mazzoni,† and A. Meluzzi\*

\*Department of Agricultural and Food Sciences, and †Department of Veterinary Medical Sciences, Alma Mater Studiorum–University of Bologna, Ozzano dell’Emilia (BO), Italy

**ABSTRACT** A trial was conducted to compare muscle traits and meat quality characteristics of the pectoralis muscle in 2 chicken commercial hybrids having standard (SBY) and high breast yield (HBY), respectively. A total of 2,124 one-day-old male chicks, equally divided into 2 experimental groups represented by strains (SBY and HBY), were grown using homogenous conditions and fed the same standard diets until reaching live weight of 4.2 kg at 53 and 55 d for the SBY and HBY groups, respectively. Thirty-six birds per each genotype were randomly selected, and their pectoralis major muscles were used to assess meat quality properties (color attributes, pH, drip loss, cook loss, Allo-Kramer shear values after cooking, moisture, proteins, total lipids, and ashes) as well as histological traits (cross-sectional area, frequency of abnormal fibers, and intramuscular fat infiltration). As expected, HBY genotype had higher breast yield (31.0 vs. 30.0%;  $P \leq 0.05$ ). Histological evaluations showed that HBY pectoralis muscles had higher cross-sectional fiber area coupled with a

dramatically higher ( $P \leq 0.001$ ) incidence of abnormal fibers and more abundant infiltration of intramuscular fat. Moreover, histopathological anomalous features such as central nuclei, proliferation of endomysial and perimysial collagen, inflammatory infiltrate, and necrosis of the fibers were also observed. As for meat quality, SBY hybrid showed lower ultimate pH values (5.97 vs. 6.07;  $P \leq 0.01$ ), whereas overall color parameters were not affected by genotype. Breast meat from the HBY genotype also exhibited significantly lower ability to retain liquid during refrigerated storage (drip loss, 2.46 vs. 2.06%;  $P \leq 0.05$ ) and cooking (26.2 vs. 21.1%;  $P \leq 0.05$ ) as well as higher shear-force values (2.59 vs. 2.11 kg/g;  $P \leq 0.001$ ). Finally, with regard to chemical composition, significant differences ( $P \leq 0.05$ ) were detected in protein (22.8 vs. 23.5%) and lipid (1.65 vs. 1.82%) contents, which were significantly lower in the HBY hybrid, whereas moisture content tended ( $P = 0.07$ ) to be inferior in the SBY hybrid.

**Key words:** chicken hybrid, breast yield, histological muscle abnormality, meat quality

2013 Poultry Science 92:2438–2447  
<http://dx.doi.org/10.3382/ps.2013-03087>

## INTRODUCTION

In the last 50 yr, consumption of poultry meat has increased rapidly, and it is supposed that it will continue to grow in the future, particularly in the developing countries. It is well known that this huge demand for poultry meat has put pressure on breeders, nutritionists, and farmers to improve the growth rate of birds, feed efficiency, and breast-meat yield. Today, chickens and turkeys are marketed at about half the time and at about twice the BW compared with 50 yr ago (Barbut et al., 2008). Elevated heritabilities of BW and carcass composition are the main reasons for these successes (Le Bihan-Duval et al., 2003). These selection criteria

have created conditions favoring disease susceptibility and modifying skeletal-muscle cell structure, metabolism, and meat quality (Barbut et al., 2008). In general, the number of fibers is related to changes during muscle growth, and fast-growing birds have more muscle fibers than do slower-growing strains (Scheuermann et al., 2004). In birds, the large postnatal increase in muscle mass is achieved by the hypertrophy of the existing fibers because of the fusion of satellite cells with the fibers (Picard et al., 2010). This increase is also associated with the increase of the number of giant fibers, which typically have cross-sectional areas 3 to 5 times larger than normal ones, although these may also result from severe contraction (hypercontracted fibers; Dransfield and Sósnicki, 1999; Remignon et al., 2000). With the increase in growth rate and muscle size, there has been an increase in incidence of pectoral myopathies (e.g., focal myopathy, deep pectoral disease; Velleman et al., 2003; Lien et al., 2012).

©2013 Poultry Science Association Inc.

Received February 1, 2013.

Accepted May 19, 2013.

<sup>1</sup>Corresponding author: [m.petracci@umibo.it](mailto:m.petracci@umibo.it)

Moreover, an increase was observed in susceptibility to stress-induced myopathies, which can impair meat quality and favor the occurrence of abnormalities such as pale, soft, and exudative-like meat (Anthony, 1998; Duclos et al., 2007; Petracci et al., 2009; Strasburg and Chiang, 2009). At the present, a high increase of quality issues such as white striping (Kuttappan et al., 2012a; Petracci et al., 2013), poor cohesiveness, and hypercontraction in breast meat (Petracci and Cavani, 2012) has been observed at the poultry-industry level. The origin of these new quality defects is still not clear, but it can be hypothesized that there is a strict connection with still increasing growth rate and breast yield achieved in some broiler hybrids, together with increasing slaughter ages and weights employed by processors during the last years. Sósnicki and Wilson (1991) evidenced that the width of the muscle fiber exceeds that of the connective tissue, leading to a loss of muscle integrity or focal myopathy in turkeys selected for rapid growth. An increase was also observed in the number of giant fibers in pectoralis muscle from fast-growing birds (Dransfield and Sósnicki, 1999). More recently, Kuttappan et al. (2012b, 2013) observed that white-striping defect is usually associated with muscle degeneration and myopathic changes beneath the striation area and hypothesized a similarity with muscular dystrophies.

Overall, extreme fiber hypertrophy and increased occurrence of giant and abnormal fibers are strong indicators for the development of insufficient meat quality (Dransfield and Sósnicki, 1999; Mitchell, 1999; Rehfeldt et al., 2004). However, only a few studies focused on the consequences of such new emerging quality defects such as poor cohesiveness, hypercontraction, and white striping on technological properties and chemical composition of the meat. This study was aimed at comparing histological traits of pectoralis major muscle and quality characteristics of breast meat obtained from 2 commercial chicken hybrids having different breast yield.

## MATERIALS AND METHODS

### *Birds and Treatments*

For this experiment, 1,062 one-day-old male chicks for each broiler genotype were supplied by a commercial hatchery. The chicken hybrids were known to have different breast yields and are here referred to as high breast yield (**HB**) and standard breast yield (**SB**). The chicks were housed separately per hybrid at a stocking density of 10 chicks/m<sup>2</sup> (59 birds per pen in 18 pens per group of about 6 m<sup>2</sup> each). Pens were equipped with pan feeders to ensure at least 2 cm/bird of front space and an automatic drinking system (1 nipple/10 birds). Each pen was equipped with an individual bin as reservoir for the experimental feed, clearly labeled. On a daily basis, the experimental feed was manually

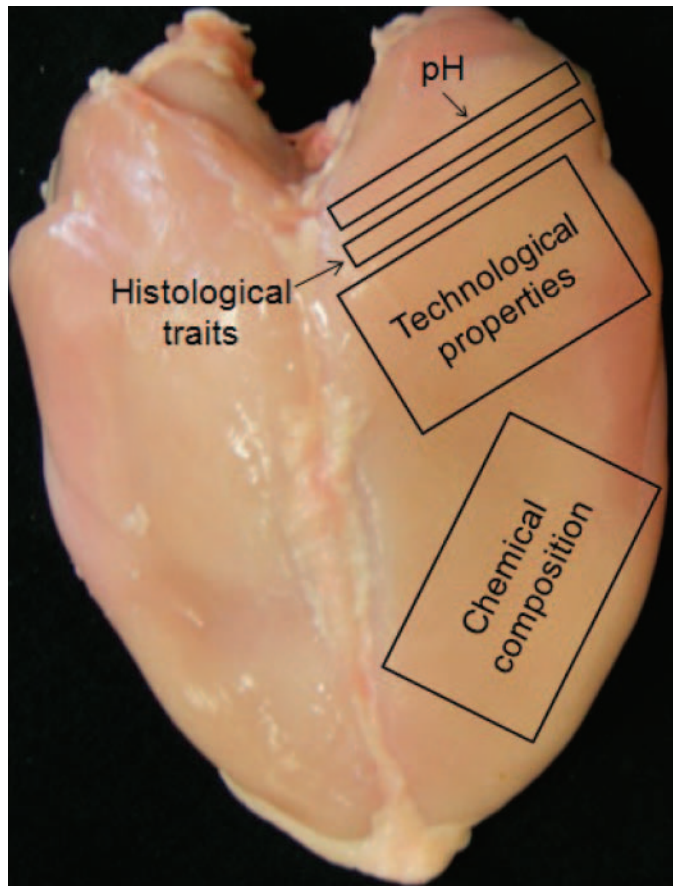
transferred from the bin to the feeder. Any changes in diet were made uniformly to all birds. Feed and water were provided for ad libitum consumption.

Birds were slaughtered at the same body live weight of 4.2 kg, reached at 53 and 55 d for the **SB** and **HB** hybrid, respectively. Two birds per each pen were randomly selected (36 birds/genotype), individually weighed, labeled, and subsequently subjected to a total feed withdrawal of 12 h, including the holding time of 2 h at the processing plant. The birds were processed under commercial conditions using electrical stunning (120 V, 200 Hz), and after chilling, carcasses were stored at 4°C for 20 h and used for subsequent meat-quality evaluation. Carcasses were obtained by removing head, neck, shanks, and abdominal fat from bled, plucked, and eviscerated birds. Carcass and breast yields were determined according to the method described by Working Group 5 of the World's Poultry Science Association (1984). After leg and wings removal, breast was dissected by making a cut from the caudal end of the cristu sterni toward the shoulder joint on both sides of each carcass and by pulling the breast apart from the back and breaking the connection at the shoulder joints. Color attributes, pH, drip loss, cook loss, Allo-Kramer shear values after cooking, moisture, proteins, total lipids, and ashes as well as histological traits (cross-sectional area and frequency of abnormal fibers and of intramuscular fat infiltration) were determined on each left breast fillet (pectoralis major muscle) according to Figure 1.

### *Histological Evaluation*

**Fiber Cross-Sectional Area.** Approximately a 2-cm<sup>3</sup> muscle sample of each pectoralis major breast muscle was removed at 20 h postmortem as shown in Figure 1. The samples were fixed in 10% buffered formalin for 24 h at room temperature. The specimens were oriented for the transverse fiber sectioning, dehydrated in a graded series of ethanol, and embedded in paraffin. From each sample, 10 serial transverse sections (6 μm thick) were obtained, mounted on poly-L-lysine coated slides, and stained with hematoxylin and eosin. For each bird, cross-sectional area of 100 fibers was estimated using KS 300 image-analysis software (Kontron Elektronik, Munich, Germany) by outlining the fibers profiles on the monitor screen using a computer mouse. Moreover, an index was calculated by subdividing values of fiber cross-sectional area into 4 ranges: <1,000 μm<sup>2</sup>, ≥1,000 to <2,000 μm<sup>2</sup>, ≥2,000 to <3,000 μm<sup>2</sup>, and ≥3,000 μm<sup>2</sup>. This subdivision has been chosen to better highlight the size distribution of the areas of the fibers within the respective experimental groups because, in some birds, fibers of small caliber were frequently associated with fibers of larger caliber and, consequently, a mean evaluation of all fibers for each experimental group would have been not entirely representative.





**Figure 1.** Positions for the determinations of pH, histological traits, technological properties (drip loss, cook loss, and Allo-Kramer shear force), and chemical composition (moisture, protein, lipid, and ash) in the pectoralis major muscle of the chicken hybrids investigated. Color version available in the online PDF.

**Abnormal-Fiber Index.** For each section of muscle, the presence of abnormal fibers (giant fibers, fibers with hyaline degeneration, and fibers with round profile) for

10 primary myofibers fascicles (**PMF**) was assessed. Results were expressed in the following way:

- when the number of abnormal fibers was 1 or 2 for each PMF, a score of F1 was assigned;
- when the number of abnormal fibers was 5 or 10 for each PMF, a score of F2 was assigned;
- when the number of abnormal fibers was more than 50% for each PMF, a score of F3 was assigned; and
- when the abnormal fibers represented almost all of the fibers for each PMF, a score of F4 was assigned.

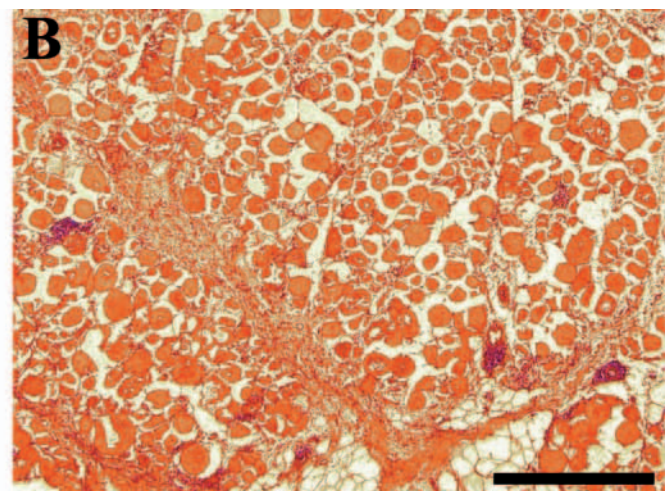
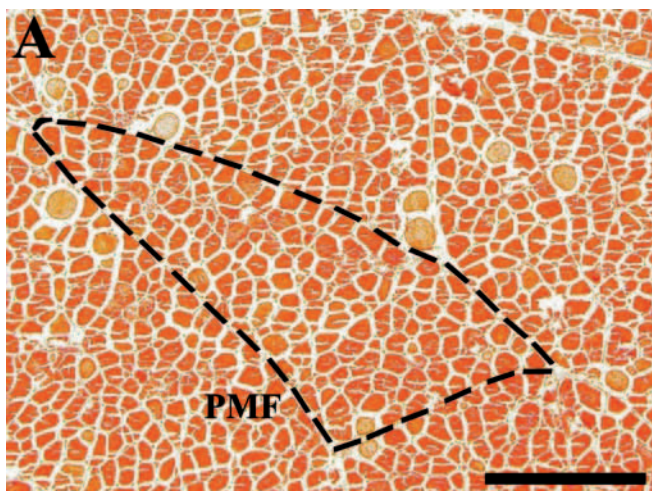
In Figure 2, low and abundant presence of abnormal fibers in PMF is given.

**Intramuscular-Fat-Infiltration Index.** For each section (about 1 cm<sup>2</sup>), the histological analysis was carried out evaluating the adipose tissue present in the perimysium connective tissue surrounding the PMF. The different histological features were classified as follows:

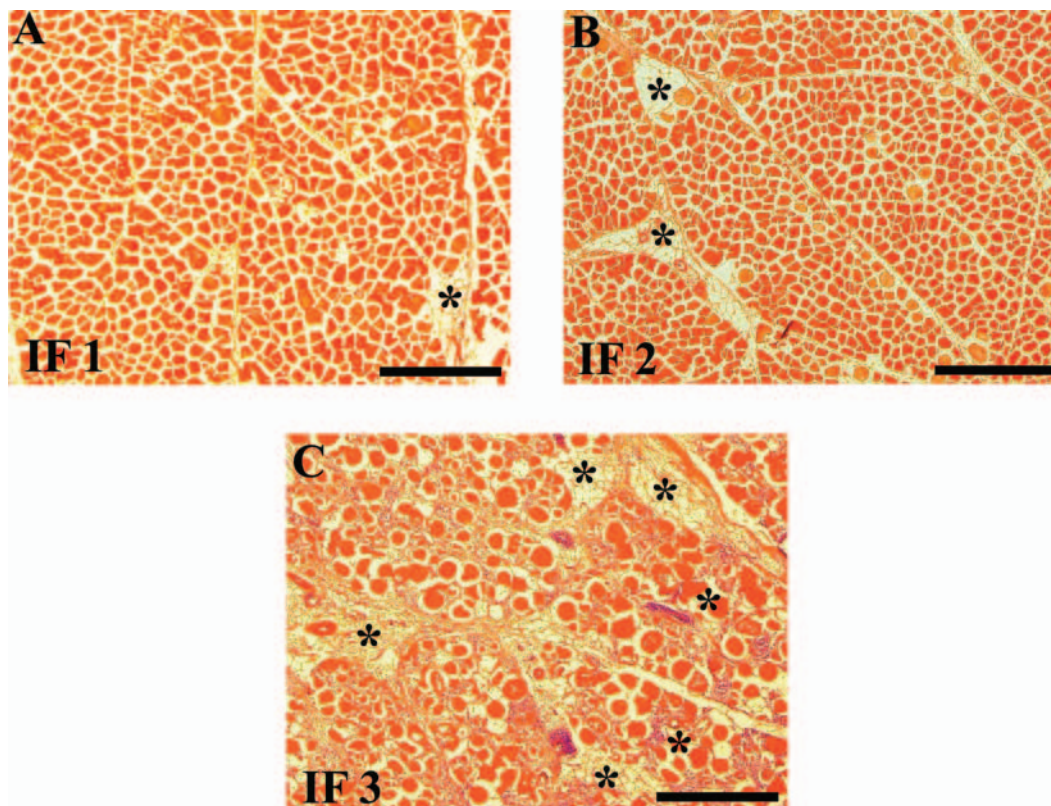
- when the intramuscular adipose tissue was represented by 1 or 2 areas of infiltration of the PMF, a score of IF1 was assigned (Figure 3A);
- when the intramuscular adipose tissue was represented by 3 to 4 areas of infiltration of the PMF, a score of IF2 was assigned (Figure 3B); and
- when the intramuscular adipose tissue was represented by more than 5 areas of infiltration of the PMF, a score of IF3 was assigned (Figure 3C).

### Meat-Quality Analyses

**Color Measurement.** The CIE (1978) system color profile of lightness ( $L^*$ ), redness ( $a^*$ ), and yellowness ( $b^*$ ) was measured by a reflectance colorimeter (CR-



**Figure 2.** Muscle fibers of the pectoralis major muscle stained with hematoxylin and eosin (H & E). Low presence of abnormal fibers in primary myofibers fascicle (PMF) is illustrated in (A). In (B), almost all of the fibers appear abnormal. (A) and (B) Bars: 800  $\mu$ m. Color version available in the online PDF.



**Figure 3.** Muscle fibers of the pectoralis major muscle stained with hematoxylin and eosin (H & E). Categorization of the amount of adipose tissue (asterisks) present in the perimysium connective tissue surrounding the primary myofibers fascicle (PMF). IF1 score was given when 1 area of infiltration of the PMF was detected (A); IF2 score was given when 3 to 4 areas were detected (B); IF3 score was given when more than 5 areas were detected (C). (A)–(C) Bars = 800  $\mu$ m. Color version available in the online PDF.

400, Minolta, Milano, Italy) using illuminant source C. The colorimeter was calibrated throughout the study using a standard white ceramic tile. Color was measured in triplicate on the bone-side surface of each fillet.

**pH Measurement.** The pH was determined using a modification of the iodoacetate method initially described by Jeacocke (1977). Approximately 2.5 g of meat samples was minced by hand and homogenized in 25 mL of a 5 mM iodoacetate solution with 150 mM potassium chloride for 30 s, and the pH of the homogenate was determined using a pH meter calibrated at pH 4.0 and 7.0.

**Drip-Loss Determination.** A sample weighing about 80 g (approximately 7  $\times$  4  $\times$  3 cm) obtained from the cranial portion of each fillet was individually weighed and suspended in a plastic box at 2 to 4°C. After 48 h, samples were blotted for the excess surface fluids, reweighed, and drip-loss determined as a percentage of weight lost by the sample during the refrigerated storage period (Petracci and Baéza, 2011).

**Cook-Loss Determination.** Meat samples after drip-loss determination were individually weighed, packaged in a plastic bag under vacuum, and cooked in a water bath (80°C) until their final internal temperature was 80°C following the recommendations given by Petracci and Baéza (2011). The samples were then allowed to equilibrate to room temperature and reweighed, and

cook loss was determined as a percentage of weight lost by the sample.

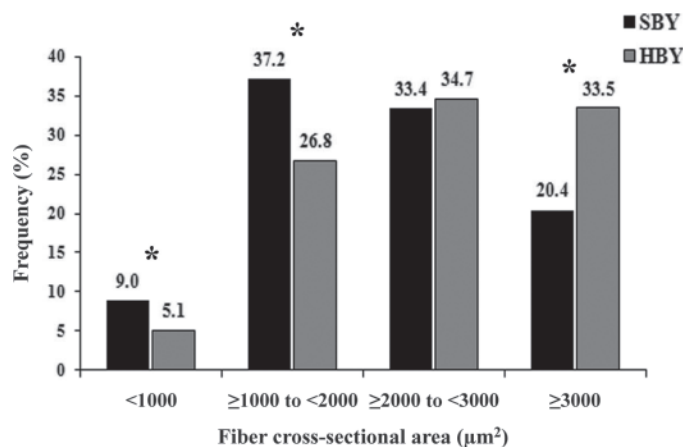
**Shear-Value Determination.** Shear values were determined using a TA.HDi Heavy Duty texture analyzer (Stable Micro Systems Ltd., Godalming, Surrey, UK) equipped with an Allo-Kramer shear cell using the procedure described by Sams et al. (1990). One meat sample (approximately 2  $\times$  4  $\times$  1 cm) from each sample used for cook-loss determination was cut parallel with the muscle-fiber direction, weighed, and sheared with the blades at a right angle to the fibers using a 250-kg load cell and cross head speed of 500 mm/min. Shear values are reported as kilograms shear per gram of sample.

**Chemical Analysis.** Moisture and ashes were determined in duplicate according to the Association of Official Analytical Chemists procedure (AOAC, 1990). Proteins were determined using the standard Kjeldahl copper catalyst method (AOAC, 1990). Total lipids were measured using a modification of the chloroform:methanol procedure described by Folch et al. (1957).

### Statistical Analysis

The influence of the hybrid (SBY vs. HBY) on slaughtering traits, cross-sectional fiber area, and meat



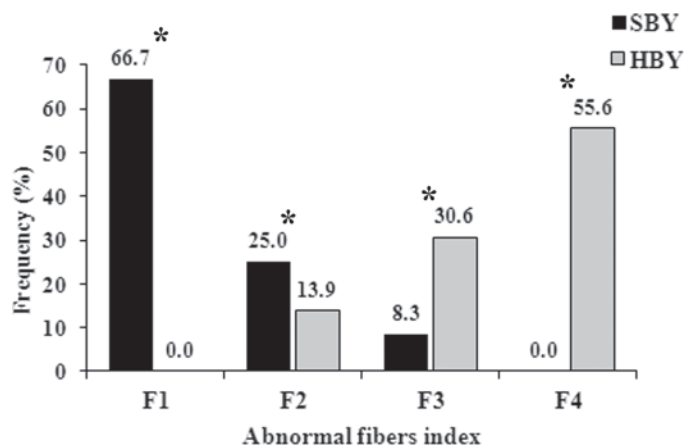


**Figure 4.** Size distribution of the fiber cross-sectional area of pectoralis major muscles from chickens belonging to standard-breast-yield (SBY,  $n = 36$ ) and high-breast-yield (HBV,  $n = 36$ ) hybrids. \*Means within each category differed at  $P < 0.05$ .

quality characteristics was evaluated by using one-way ANOVA (SAS, 1988). Moreover, fiber cross-sectional area, abnormal fibers, and intramuscular-fat-infiltration indexes were analyzed using chi-squared test. If the value of the frequency was expected to be less than 5, the Fisher's exact test was used (Fleiss, 2003). Individual birds were considered as the experimental unit for the entire analysis.

## RESULTS

The slaughtering performances and fiber cross-sectional area of pectoralis major muscle from birds belonging to the 2 hybrids are presented in Table 1. Body live and carcass weight did not differ, whereas hybrid HBV had higher carcass (74.1 vs. 72.9%;  $P \leq 0.01$ ) and, as expected, breast yield (31.0 vs. 30.0%;  $P \leq 0.05$ ). In accordance with this result, fiber cross-sectional area was greater in breast muscles from hybrid HBV (2,674 vs. 2,240  $\mu\text{m}^2$ ;  $P \leq 0.01$ ). Size distribution of the cross-sectional area of skeletal muscle fibers in the pectoralis major muscles is reported in Figure 4. The SBY hybrid had a higher number of muscle fibers included in the first 2 ranges (<1,000  $\mu\text{m}^2$  and  $\geq 1,000$



**Figure 5.** Prevalence of abnormal fibers in the pectoralis major muscles from chickens belonging to standard-breast-yield (SBY,  $n = 36$ ) and high-breast-yield (HBV,  $n = 36$ ) hybrids. \*Means within each category differed at  $P < 0.05$ . F1 = 1 or 2 abnormal fibers; F2 = 5 or 10 abnormal fibers; F3 = more than 50% of abnormal fibers; F4 = almost all abnormal fibers.

to <2,000  $\mu\text{m}^2$ ) with respect to group HBV. In the range  $\geq 2,000$  to <3,000  $\mu\text{m}^2$ , the number of the fibers is almost the same in the 2 hybrids, whereas the birds from group HBV have a larger number of fibers  $\geq 3,000$   $\mu\text{m}^2$ . Figure 5 and 6 show, respectively, the prevalence of abnormal fibers and intramuscular fatty infiltration areas in pectoralis major muscles obtained from birds belonging to the 2 hybrids. Histological observation of the PMF identified that the SBY hybrid has a limited number of abnormal fibers, scored F1 and F2, respectively. On the contrary, in HBV birds many abnormal fibers in the PMF scored F3 and F4, respectively. The intramuscular fatty infiltration is particularly abundant in the genotype HBV, where most of the histological samples were scored IF2 and IF3, whereas the samples from hybrid SBY were almost all scored IF1 and IF2.

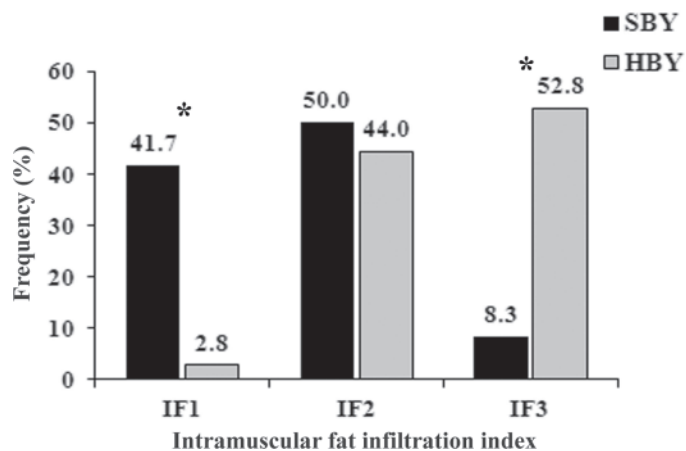
The histological observations, particularly in group HBV, have shown that most of the muscle fibers were characterized by amorphous-looking central area (central or eccentric; Figure 7A) weakly stained. This area may be referred to hyaline cytoplasm. In addition, these fibers exhibited central nuclei and a rounded profile (Figure 7A, B). In most sections, tiny fibers were

**Table 1.** Slaughtering traits and fiber cross-sectional area of pectoralis major muscle in chickens belonging to standard-breast-yield (SBY) and high-breast-yield (HBV) hybrids

Parameter	Hybrid		SEM	Probability
	SBY	HBV		
no.	36	36		
Body live weight (g)	4,236.1	4,226.3	76.0	NS
Carcass weight (g)	3,086.8	3,134.4	64.6	NS
Carcass yield (%)	72.9	74.1	0.27	**
Breast yield <sup>1</sup> (%)	30.0	31.0	0.51	*
Fiber cross-sectional area ( $\mu\text{m}^2$ )	2,239.6	2,674.3	146.8	**

<sup>1</sup>Based on carcass weight.

\*\* $P \leq 0.01$ ; \* $P \leq 0.05$ .



**Figure 6.** Prevalence of intramuscular fatty infiltration areas in the pectoralis major muscles from chickens belonging to standard-breast-yield (SBY,  $n = 36$ ) and high-breast-yield (HBY,  $n = 36$ ) hybrids. \*Means within each category differed at  $P < 0.05$ . IF1 = 1 area of fat infiltration; IF2 = 3 to 4 areas of fat infiltration; IF3 = more than 5 areas of fat infiltration.

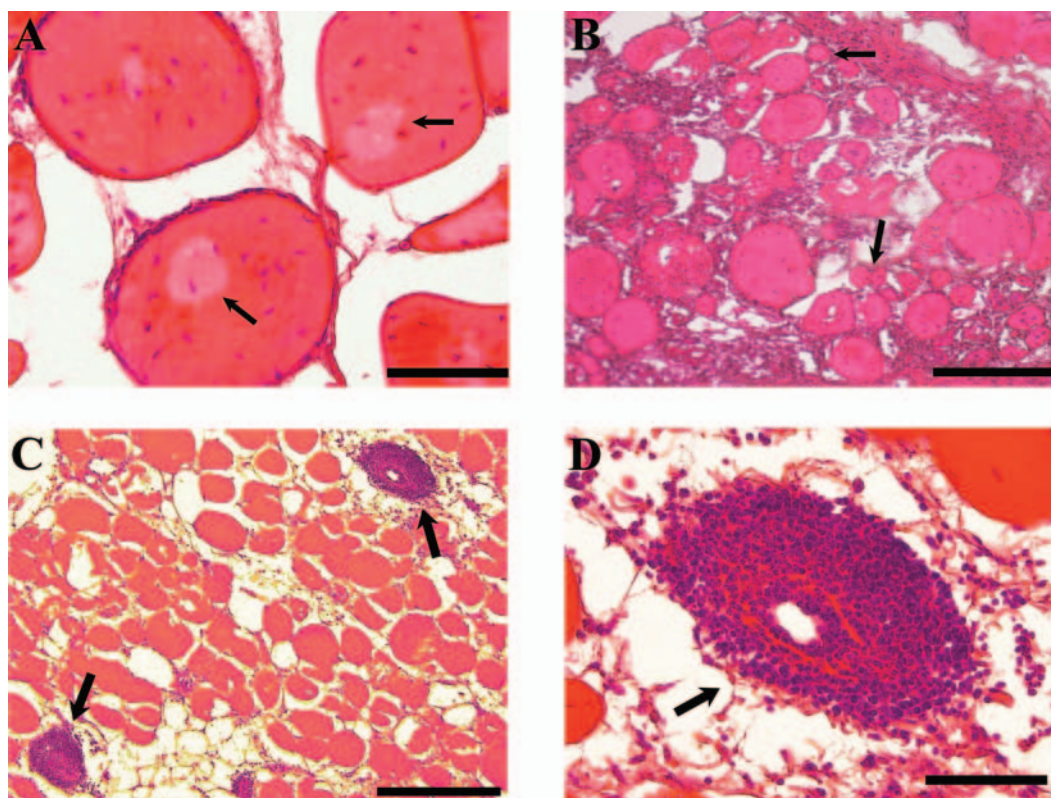
observed (Figure 7B). Proliferation or damage of the connective perimysial network (Figures 2B and 3C) and fatty tissue replacement (Figure 3C) with abundant infiltration of mononuclear cells were also seen. In some cases, necrotic fibers undergoing mononuclear

cells with consequent central or eccentric vacuolation were observed (Figure 7C, D).

As for meat quality characteristics (Table 2), the SBY hybrid showed lower ultimate pH values (5.97 vs. 6.07;  $P \leq 0.01$ ), whereas overall color parameters were not affected by genotype with the exception of yellowness ( $b^*$ ), which revealed significantly higher values in the hybrid HBY (17.5 vs. 16.4;  $P \leq 0.05$ ). Breast meat from genotype HBY also exhibited significantly lower ability to retain liquid during refrigerated storage (drip loss, 2.46 vs. 2.06%;  $P \leq 0.05$ ) and cooking (26.2 vs. 21.1%;  $P \leq 0.05$ ) as well as higher Allo-Kramer shear-force values (2.59 vs. 2.11 kg/g;  $P \leq 0.001$ ). Finally, with regard to chemical composition, significant differences ( $P \leq 0.05$ ) were detected in protein (22.8 vs. 23.5%) and lipid (1.65 vs. 1.82%) contents, which were significantly lower in the HBY hybrid, whereas moisture content tended ( $P = 0.07$ ) to be inferior in the hybrid SBY. Otherwise, ash content was not affected by genotype.

## DISCUSSION

The use of hybrids selected for different breast yields allowed us to obtain pectoralis major muscles having different fiber cross-sectional areas. Guernec et al.



**Figure 7.** Muscle fibers of the pectoralis major muscles from high-breast-yield hybrid stained with hematoxylin and eosin (H & E). (A) Most of the muscle fibers show hyaline cytoplasm characterized by an amorphous-looking central area (central or eccentric; arrows) weakly stained. In addition, these fibers exhibit central nuclei and a rounded profile. (B) Tiny fibers (arrows) that surround greater-caliber fibers and are immersed in a compromised connective tissue are shown. (C) A muscle fiber surrounded by inflammatory mononuclear cells (arrow) near to adipose tissue (probably adipose tissue replacement) is illustrated. (D) A necrotic fiber, magnification of (C), surrounded by mononuclear cells (arrows) exhibiting an eccentric vacuolation is shown. (A) and (D) Bars = 50  $\mu\text{m}$ . (B) and (C) Bars = 100  $\mu\text{m}$ . Color version available in the online PDF.

**Table 2.** Quality traits of breast meat from chickens belonging to standard-breast-yield (SBY) and high-breast-yield (HBY) hybrids

Parameter	Hybrid			Probability
	SBY	HBY	SEM	
no.	36	36		
pH	5.97	6.07	0.03	***
Color				
Lightness (L*)	53.84	54.04	0.51	NS
Redness (a*)	0.03	0.06	0.13	NS
Yellowness (b*)	16.36	17.53	0.48	*
Drip loss (%)	2.06	2.46	0.17	*
Cook loss (%)	21.05	26.23	1.16	*
Allo-Kramer shear force (kg/g)	2.11	2.59	0.15	***
Chemical composition				
Moisture (%)	75.07	75.52	0.26	0.0706
Protein (%)	23.48	22.82	0.37	*
Lipid (%)	1.82	1.65	0.10	*
Ash (%)	1.24	1.21	0.02	NS

\*\*\* $P \leq 0.001$ ; \* $P \leq 0.05$ .

(2003) evidenced that broiler selection focused on improved breast yield led to wider and thicker pectoralis major muscles essentially by increasing the diameter and length of fibers without significantly altering their number. Nevertheless, Scheuermann et al. (2004) suggested that increased muscle-fiber number may also participate to improve breast yield even though it was confirmed that fiber hypertrophy is an essential factor for an increase in muscle volume. Otherwise, in the present study, the increase in the area of muscle fibers was largely associated with a greater presence of abnormal muscle fibers. This feature was particularly evident in the genotype HBY. In addition, muscle fibers of very small size were observed in correspondence of abnormal fibers. Dransfield and Sónicki (1999) described how the increase of muscle-fiber size in broilers is associated with the increase in giant fibers having an area 3 to 5 times larger than normal fibers. In their review, Tumova and Teimouri (2009) supported the hypothesis that genetic selection to achieve a higher breast yield could be considered one of the main reasons for the presence of giant fibers in chicken muscle. Moreover, Miraglia et al. (2006) found a higher percentage of giant fibers in the pectoralis major muscle from Ross birds (fast growing, ADG >50 g/d) compared with Kabir ones (medium growing, ADG around 30–35 g/d). Interestingly, the same authors did not observe these differences in other muscles, such as the lateral iliotibial and the semimembranosus. On the contrary, Werner et al. (2008) observed a low prevalence of degenerated or giant fibers in turkeys from both selected and unselected genotypes. In an earlier study, Remignon et al. (2000) suggested that the occurrence of giant fibers is independent of the genetic background of the turkeys or the type of muscle, but it depends on the biochemical events occurring during rigor mortis. These structurally abnormal fibers are considered to arise from hypercontraction of the muscle fibers or parts of them that are not able to undergo normal relaxation after initial rigor mortis (Rehfeldt et al., 2004).

Besides giant fibers, additional fiber abnormalities have been found by analyzing the microstructure of the pectoralis major muscles, and these were particularly abundant in the HBY genotype. It is well known that improvements in the genetic selection for production traits such as growth rate, slaughter weight, and feed conversion efficiency have been associated, in some cases, with adverse effects in skeletal muscles (Mitchell, 1999).

In the present study, from the evaluations carried out under a microscope, most of the fibers showed an increase in the number of nuclei that were located in a central position (so-called internalization of the nuclei). In addition, these myonuclear abnormalities are again associated with fibers with hyaline sarcoplasm. The presence of nuclei in the central position of the muscle fibers is considered as an index of abnormality in mammals, and when the number of nuclei becomes conspicuous, it comes to a disorder due to a muscle myopathy (Dubowitz and Brooke, 1973); the same condition was also described by Pizzey and Barnard (1983) in dystrophic chickens.

Soike and Bergmann (1998) claimed that central nuclei and fibers with hyaline or necrotic features were symptomatic of structural damage of the muscle fiber; also Nakada et al. (1998) described the presence of hyaline cytoplasm such as a consequence of the muscle fiber degeneration. Finally, in a recent study, Polak et al. (2010) observed a considerable number of degenerated muscle fibers characterized by hyaline cytoplasm in broiler pectoralis major muscle.

In many animal species, in addition to the above anomalies of the muscle architecture, a distinct change of perimysial and endomysial connective tissue was evidenced. These alterations were frequently accompanied by inflammatory infiltrate (lymphocytes, micro- and macro-phages) and necrosis of some muscle fibers; in the latter, a sarcoplasmic vacuolation was sometimes identified. Histopathological anomalous features such as hypertrophy and modification of the muscle fiber



profile, central nuclei, proliferation of endomysial and perimysial collagen, inflammatory infiltrate, and necrosis of the fibers, entirely because of what we have above mentioned, have been reported by several authors in chickens (Pizzey and Barnard, 1983; Ashmore et al., 1988; Ishimoto et al., 1988; MacRae et al., 2006; Polak et al., 2009; Nishita et al., 2012) and in turkeys (Sósnicki et al., 1989, 1991). The same authors mentioned above described how these histopathological changes accompanied by ultrastructural and enzyme abnormalities were identified as characteristic of congenital muscular dystrophy.

The presence of muscle fibers of small caliber shown in correspondence of the abnormal fibers is a further histological appearance that supports the hypothesis of muscular dystrophy. Abnormal fibers surrounded by fibers of small caliber (also referred to as regenerative fibers) have been described in chickens and turkeys with various myopathies including dystrophy (Pizzey and Barnard, 1983; Ashmore et al., 1988; Sósnicki et al., 1989; Polak et al., 2009, 2010). In this context, the increase in infiltration of adipose tissue in the perimysial connective tissue is also included. Pizzey and Barnard (1983), in the pectoral muscle of dystrophic 60-d-old chickens, described degenerated muscle fibers that were subsequently removed and replaced with the adipose or collagen tissue: the same histological features have been described by Sósnicki et al. (1989) in turkey breast muscle.

Overall, extreme fiber hypertrophy and increased occurrence of giant and abnormal fibers are strong indicators for the development of insufficient meat quality (Mitchell, 1999; Rehfeldt et al., 2004). This assumption has been confirmed in the present study, because the higher degree of abnormalities observed in pectoralis major muscles from birds belonging to the HBY genotype is associated with an impairment of meat quality. Indeed, breast meat from the HBY hybrid exhibited a significant reduced ability to retain liquid during refrigerated storage and cooking as indicated by the higher drip-loss and cook-loss values. These results agree with some previous studies that stated that muscle alterations are related to selection for breast yield (Remignon et al., 1995; Berri et al., 2001; Guernec et al., 2003). However, in the present study poor water-holding capacity cannot be related to poultry pale, soft, and exudative-like abnormality, because it was not associated with low pH or pale meat color (Petracci et al., 2004; Barbut et al., 2008). Indeed, breast meat from the HBY hybrid had a higher pH if compared with the SBY group, whereas no differences were found in meat lightness. As a consequence, reduced ability to hold liquids found in the present study may be attributed to the set of microstructure observations previously described such as a diffused degeneration of muscle fibers with a replacement of myofibrillar and sarcoplasmic proteins by connective layers and infiltration of adipose tissue. It is well known that myofibrillar proteins (e.g.,

myosin, actin) are mainly responsible for the water-holding capacity of the meat, whereas connective-tissue proteins are water insoluble (Xiong, 2004). Hence, it is not surprising that breast meat from the HBY hybrid presented a reduced ability to retain its own water during storage and cooking. In a previous study (Petracci et al., 2013), it was found that HBY broiler hybrids exhibited a higher incidence of breast fillets showing white-stripping defect, which has been associated with a decreased water-holding and binding capacity. This higher amount of liquid loss during cooking can explain the increased toughness of breast fillets from HBY birds (Murphy and Marks, 2000). Moreover, it should be noted that breast meat from the HBY hybrid also exhibited a significantly lower protein content and tended to have a higher moisture content. This demonstrated that muscular microstructure degeneration is able to determine a macroscopic quantitative reduction of proteins with a correspondent increase of water. Also, Kuttappan et al. (2013) found a decrease in protein content in breast meat samples affected by myopathic lesions such as loss of cross striations, variability in fiber size, floccular or vacuolar degeneration and lysis of fibers, mild mineralization, and occasional regeneration. On the other hand, there is no association between the higher level of infiltration of adipose tissue in the perimysial connective tissue observed in samples from the HBY group and total lipids content, which is found to be higher in breast fillets from birds belonging to the SBY hybrid. This inconsistency can be due to the different localization of meat samples (Figure 1), because histological and chemical composition analyses were carried out in samples taken from cranial and caudal parts, respectively, of pectoralis major muscle. This hypothesis is supported by Kuttappan et al. (2013), who observed a higher lipid content corresponding to the cranial part of the pectoralis major muscle where the fillets showed more pronounced histological lesions. However, it should also be noted that in the present study, a specific staining to quantify (by means of morphometric measurements) the adipose tissue in the cross-sectional areas was not adopted, and these discrepancies may be due also to the lack of precision in the evaluation of the infiltration of adipose tissue.

In conclusion, this study showed that the ever-increasing genetic pressure to improve breast yield of broiler chickens has led to a very high occurrence of alterations in pectoralis major muscle such as tiny, split, abnormal, hyaline, and necrotic fibers referable to muscular dystrophy. As expected, this large extent of microstructural anomalies within breast fillets played a remarkable detrimental effect on meat-quality attributes by decreasing protein content and deeply reducing ability of the meat to hold liquids during processing and storage. Based on these results, it is strongly recommended to emphasize relevance of meat quality traits among the selection criteria of commercial chicken hybrids in the near future.



## ACKNOWLEDGMENTS

We thank Paolo Montagna and Filiberto Ceccaroni [both of the Gesco Consorzio Cooperativo, Cesena (FC), Italy] for the technical support. The authors also express their appreciation to Stefano Pignata and Roberto Donatini of the Department of Agricultural and Food Sciences of the University of Bologna for their technical assistance.

## REFERENCES

- Anthony, N. B. 1998. A review of genetic practices in poultry: Efforts to improve meat quality. *J. Muscle Foods* 9:25–33.
- AOAC. 1990. Meat and meat products. Pages 931–948 in *Official Methods of Analysis*. 15th ed. Vol. 2. Assoc. Off. Anal. Chem., Washington, DC.
- Ashmore, C. R., L. Hitchcock, and Y. B. Lee. 1988. Passive stretch of adult chicken muscle produces a myopathy remarkably similar to hereditary muscular dystrophy. *Exp. Neurol.* 100:341–353.
- Barbut, S., A. A. Sósnicki, S. M. Lonergan, T. Knapp, D. C. Ciobanu, L. J. Gatcliffe, E. Huff-Lonergan, and E. W. Wilson. 2008. Progress in reducing the pale, soft and exudative (PSE) problem in pork and poultry meat. *Meat Sci.* 79:46–63.
- Berri, C., N. Wacrenier, N. Millet, and E. Le Bihan-Duval. 2001. Effect of selection for improved body composition on muscle and meat characteristics of broilers from experimental and commercial lines. *Poult. Sci.* 80:833–838.
- CIE. 1978. Recommendations on uniform color spaces, color differences, equations. Psychometric color terms. CIE Publication 15, Suppl. 2. Comm. Int. de l'Eclairage, Colorimetry, Paris, France.
- Dransfield, E., and A. A. Sósnicki. 1999. Relationship between muscle growth and poultry meat quality. *Poult. Sci.* 78:743–746.
- Dubowitz, V., and M. H. Brooke. 1973. *Muscle Biopsy: A Modern Approach*. W. B. Saunders, London, UK.
- Duclos, M. J., C. Berri, and E. Le Bihan-Duval. 2007. Muscle growth and meat quality. *J. Appl. Poult. Res.* 16:107–112.
- Fleiss, J. 2003. *Statistical Methods for Rates and Proportions*. Wiley, New York, NY.
- Folch, J., M. Lees, and G. H. Sloane-Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.* 22:497–509.
- Guernec, A., C. Berri, B. Chevalier, N. Wacrenier-Cere, E. Le Bihan-Duval, and M. J. Duclos. 2003. Muscle development, insulin-like growth factor-I and myostatin mRNA levels in chickens selected for increased breast muscle yield. *Growth Horm. IGF Res.* 13:8–18.
- Ishimoto, S., I. Goto, and Y. Kuroiwa. 1988. Early morphological changes in the striated muscles in normal and dystrophic chickens. *J. Comp. Pathol.* 98:69–79.
- Jeacocke, R. E. 1977. Continuous measurement of the pH of beef muscle in intact beef carcasses. *J. Food Technol.* 12:375–386.
- Kuttappan, V. A., V. B. Brewer, J. K. Apple, P. W. Waldroup, and C. M. Owens. 2012a. Influence of growth rate on the occurrence of white striping in broiler breast fillets. *Poult. Sci.* 91:2677–2685.
- Kuttappan, V. A., S. D. Goodgame, C. D. Bradley, A. Mauromoustakos, B. M. Hargis, P. W. Waldroup, and C. M. Owens. 2012b. Effect of different levels of dietary vitamin E (DL- $\alpha$ -tocopherol acetate) on the occurrence of various degrees of white striping on broiler breast fillets. *Poult. Sci.* 91:3230–3235.
- Kuttappan, V. A., H. L. Shivaprasad, D. P. Shaw, B. A. Valentine, B. M. Hargis, F. D. Clark, S. R. McKee, and C. M. Owens. 2013. Pathological changes associate with white striping in broiler breast muscles. *Poult. Sci.* 92:331–338.
- Le Bihan-Duval, E., C. Berri, E. Baeza, V. Sante, T. Astruc, H. Réminon, G. Le Pottier, J. Bentley, C. Beaumont, and X. Fernandez. 2003. Genetic parameters of meat technological quality traits in a grand-parental commercial line of turkey. *Genet. Sel. Evol.* 35:623–635.
- Lien, R. J., S. F. Bilgili, J. B. Hess, and K. S. Joiner. 2012. Induction of deep pectoral myopathy in broiler chickens via encouraged wing flapping. *J. Appl. Poult. Res.* 21:556–562.
- MacRae, V. E., M. Mahon, S. Gilpin, D. A. Sandercock, and M. A. Mitchell. 2006. Skeletal muscle fibre growth and growth associated myopathy in the domestic chicken (*Gallus domesticus*). *Br. Poult. Sci.* 47:264–272.
- Miraglia, D., R. Mammoli, R. Branciarri, D. Ranucci, and B. T. Cenci Goga. 2006. Characterization of muscle fibre type and evaluation of the presence of giant fibers in two meat chicken hybrids. *Vet. Res. Commun.* 30:357–360.
- Mitchell, M. A. 1999. Muscle abnormalities—Pathophysiological mechanisms. Pages 65–98 in *Poult. Meat Sci.—Poult. Science Symp. Series Vol. 25*. R. I. Richardson and G. C. Mead, ed. CABI Publishing, Wallingford, UK.
- Murphy, R. Y., and B. P. Marks. 2000. Effect of meat temperature on proteins, texture, and cook loss for ground chicken breast patties. *Poult. Sci.* 79:99–104.
- Nakada, K., J. Mashima, Y. Yao, J. Miyazaki, and T. Hirabayashi. 1998. Developmental stage-dependent expression of troponin T isoforms in chicken embryonic breast grafted on chorio-allantoic membrane. *Zoolog. Sci.* 15:729–736.
- Nishita, T., D. Yorifuji, K. Orito, N. Ichihara, and K. Arishima. 2012. Muscle carbonic anhydrase III levels in normal and muscular dystrophias afflicted chickens. *Acta Vet. Scand.* 54:34.
- Petracci, M., and E. Baéza. 2011. Harmonization of methodologies for the assessment of poultry meat quality features. *World's Poult. Sci. J.* 68:137–153.
- Petracci, M., M. Bianchi, M. Betti, and C. Cavani. 2004. Color variation and characterization of broiler breast meat during processing in Italy. *Poult. Sci.* 83:2086–2092.
- Petracci, M., M. Bianchi, and C. Cavani. 2009. The European perspective on pale, soft, exudative conditions in poultry. *Poult. Sci.* 88:1518–1523.
- Petracci, M., and C. Cavani. 2012. Muscle growth and poultry meat quality issues. *Nutrients* 4:1–12.
- Petracci, M., S. Mudalal, A. Bonfiglio, and C. Cavani. 2013. Occurrence of white striping under commercial conditions and its impact on breast meat quality in broiler chickens. *Poult. Sci.* 92:1670–1675.
- Picard, B., C. Berri, L. Lefaucheur, C. Molette, T. Sayd, and C. Terlouw. 2010. Skeletal muscle proteomics in livestock production. *Brief Funct. Genomics* 9:259–278.
- Pizzey, J. A., and E. A. Barnard. 1983. Structural changes in muscles of the dystrophic chicken. II. Progression of the histopathology in the pectoralis muscle. *Neuropathol. Appl. Neurobiol.* 9:149–164.
- Polak, M., B. Przybylska-Gornowicz, and A. Faruga. 2009. Abnormal morphology of skeletal muscles in meat-type chickens—Ultrastructural observations. *Pol. J. Vet. Sci.* 12:473–479.
- Polak, M., B. Przybylska-Gornowicz, and A. Faruga. 2010. The effect of different rearing condition on muscle characteristic in broilers of two commercial lines. A light microscopic study. *Jpn. Poult. Sci.* 47:125–132.
- Rehfeldt, C., I. Fiedler, and N. C. Stickland. 2004. Number and size of muscle fibers in relation to meat production. Pages 1–30 in *Muscle Development of Livestock Animals: Physiology, Genetics, and Meat Quality*. M. F. W. te Pas, M. E. Haagsman, and H. P. Everts, ed. CAB Int., Wallingford, UK.
- Remignon, H., M. F. Gardahaut, G. Marche, and F. H. Ricard. 1995. Selection for rapid growth increases the number and the size of muscle fibres without changing their typing in chickens. *J. Muscle Res. Cell Motil.* 16:95–102.
- Remignon, H., J. Zanusso, G. Albert, and R. Babilé. 2000. Occurrence of giant myofibres according to muscle type, pre- or post-rigor state and genetic background in turkeys. *Meat Sci.* 56:337–343.
- Sams, A. R., D. M. Janky, and S. A. Woodward. 1990. Comparison of two shearing methods for objective tenderness evaluation and two sampling times for physical-characteristic analyses of early harvested broiler breast meat. *Poult. Sci.* 69:348–353.
- SAS. 1988. *SAS/STAT Guide for Personal Computers*. Version 6.03 ed. SAS Inst. Inc., Cary, NC.

- Scheuermann, G. N., S. F. Bilgili, S. Tuzun, and D. R. Mulvaney. 2004. Comparison of chicken genotypes: Myofiber number in pectoralis muscle and myostatin ontogeny. *Poult. Sci.* 83:1404–1412.
- Soike, D., and V. Bergmann. 1998. Comparison of skeletal muscle characteristics in chicken breed for meat or egg production. I. Histological and electron microscope examination. *J. Vet. Med.* 45:161–167.
- Sósnicki, A. A., R. G. Cassens, D. R. McIntyre, R. J. Vimini, and M. L. Greaser. 1989. Incidence of microscopically detectable degenerative characteristics in skeletal muscles of turkeys. *Br. Poult. Sci.* 30:69–80.
- Sósnicki, A. A., R. G. Cassens, R. J. Vimini, and M. L. Greaser. 1991. Histopathological and ultrastructural alternations of turkey skeletal muscle. *Poult. Sci.* 70:349–357.
- Sósnicki, A. A., and B. W. Wilson. 1991. Pathology of turkey skeletal muscle: Implications for the poultry industry. *Food Struct.* 10:317–326.
- Strasburg, G. M., and W. Chiang. 2009. Pale, soft, exudative turkey—The role of ryanodine receptor variation in meat quality. *Poult. Sci.* 88:1497–1505.
- Tumova, E., and A. Teimouri. 2009. Chicken muscle fibres characteristics and meat quality: A review. *Scientia Agriculturae Bohemica* 40:253–258.
- Velleman, S. G., J. W. Anderson, C. S. Coy, and K. E. Nestor. 2003. Effect of selection for growth rate on muscle damage during turkey breast muscle development. *Poult. Sci.* 82:1069–1074.
- Werner, C., J. Riegel, and M. Wicke. 2008. Slaughter performance of four different turkey strains, with special focus on the muscle fiber structure and the meat quality of the breast muscle. *Poult. Sci.* 87:1849–1859.
- World's Poultry Science Association, Working Group No. 5. 1984. Method of Dissection of Broiler Carcasses and Description of Parts. J. Fris Jensen ed. Papworth Everard, Cambridge, UK.
- Xiong, Y. L. 2004. Protein functionality. Pages 218–225 in *Encyclopedia of Meat Sciences*. W. K. Jensen, ed. Elsevier Ltd., Oxford, UK.