

Breast meat quality in males and females of fast-, medium- and slow-growing chickens fed diets of 2 protein levels

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ABSTRACT The aim of this study was to evaluate the effects of genotype, sex, dietary protein level, and their interactions on select carcass characteristics and meat quality of fast- (Ross 308), medium- (Hubbard JA757) and slow-growing (ISA Dual) chickens (n = 2,520). The diet of the low-protein group of chickens had 6% lower CP than the commercial diet fed to the control group. When the chickens reached an average live weight of 2 kg, 10 males and 10 females of each genotype and the diet were selected for slaughter and breast meat–quality analysis. The dressing out and breast percentages were lower in the JA757 (−2.0 and −5.9%, respectively) and ISA Dual chickens (−9.9 and −14.3%, respectively) than those in the Ross 308 chickens. The ISA Dual chickens had higher abdominal fat percentage, higher DM and protein contents and

lower ether extract content and shear force value in breast meat than the other genotypes. Significant interaction effects of genotype, sex, and diet were found on the color of breast skin. Among the various combinations of genotype, sex, and diet group, Ross 308 females fed the low-protein diet had the highest redness and yellowness of breast skin, highest pH₄₅ value, and largest fibers, whereas ISA Dual females had the lowest color parameters and pH₄₅ value, and ISA Dual males had the smallest muscle fibers. The low-protein diet was associated with decreased abdominal fat percentage and changes in meat quality parameters, including increased darkness, meat color intensity, drip loss, and muscle fiber area, in all genotypes. The results indicated greater differences in meat quality owing to genotype than to sex or dietary protein level.

Key words: chicken, genotype, sex, diet, meat quality

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INTRODUCTION

Currently, chicken meat production is mainly conducted with fast-growing genotypes of chickens housed indoors under climate-controlled conditions, including controlled photoperiod, light intensity, temperature, and balanced diets that ensure high growth intensity and a low feed conversion ratio (Sirri et al., 2011). Owing to advances in genomics and improved nutrition, fast-growing chickens reach the market weight of 2 kg as early as 5 to 6 wk of age (Devatkal et al., 2019). However, there are negative consequences of selection for intensive growth. Fast-growing chickens can be susceptible to sudden death syndrome, ascites, and leg and cardiovascular

disorders. Another potential adverse effect of selection on growth rate is lower meat quality and increased risk of muscle abnormalities such as pale, soft, exudative meat, giant muscle fibers, wooden breast, and white striping of breast meat (Soglia et al., 2016). In recent year, the demand for chickens from alternative housing systems lacking well-controlled conditions (Fanatico et al., 2007, 2008) has been growing, for which medium- and slow-growing chickens are especially suitable. Medium- and slow-growing chickens show more foraging behavior and could be more suitable for alternative production systems because of their higher vitality, disease resistance, and adaptability to outdoor conditions than fast-growing chickens (Sirri et al., 2011), which result in lower mortality and lower incidence of limb defects. Medium-growing chickens are characterized by moderate daily weight gain (20–35 g/d; Dal Bosco et al., 2012). Slow-growing chickens may be defined as chickens with low growth rates, a daily weight gain of up to 20 g (Dal Bosco et al., 2012) and a live weight of 2.2–2.5 kg in 56–81 d. Slow-growing birds are usually male layer

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hybrids used for meat production and dual-purpose chickens, the hens of which have satisfactory laying performance, and the males exhibit good growth and carcass quality (Mueller et al., 2018). In dual-purpose chickens, it is possible to raise both sexes together during the first week, as sex sorting is not necessary. A mixed-sexes rearing period requires a broiler diet for males to achieve adequate growth performance. In the final fattening period, males are separated from females based on feather and comb condition, and the females are maintained for an entire laying period.

Regarding carcass parameters, medium- and slow-growing chickens have lower dressing out percentages (DOP) and breast muscle percentages but higher thigh percentages and lower abdominal fat percentages (Dal Bosco et al., 2014) than fast-growing genotypes. Selection on growth induces changes in the number of myofibers and postnatal hypertrophy; it can also lead to changes in muscle metabolism and thus probably affects meat quality. Among meat quality characteristics, tenderness, color, odor, and taste are crucial factors affecting consumer choice.

Balancing the nutritional requirements and growth rate of chickens can be important for achieving optimal productive capacity of individual chicken genotypes. A conventional diet was developed for fast-growing chickens as per their nutritional requirements (Fanatico et al., 2007), but little information about the nutrition of medium- and slow-growing chicken genotypes is available. Morris and Njuru (1990) showed that slow-growing chickens need less CP in the diet than fast-growing chickens. Kreuzer et al. (2020) proposed that dual-purpose chickens might perform similarly well on a lower-quality diet as their nutrient requirements are expected to be lower than those of fast-growing chickens.

Genotype and nutrition interactions can be very important in determining meat quality characteristics. Therefore, the aim of our study was to evaluate the effects of genotype, sex, and dietary CP level and their interactions on meat quality traits and muscle fiber characteristics in fast-, medium- and slow-growing genotypes of chickens.

MATERIALS AND METHODS

The effects of dietary protein level in males and females of fast- (Ross 308), medium- (JA757), and slow-growing (ISA Dual) chickens were evaluated. The experiment was approved by the Ethics Committee of the Central Commission for Animal Welfare of the Ministry of Agriculture of the Czech Republic and carried out in accordance with Directive 2010/63/EU for animal experiments.

Animals and Experimental Design

In the experiment, fast- (Ross 308), medium- (Hubbard JA757), and slow-growing (ISA Dual) chickens were used. A total of 2,520 1-day-old chickens (male:female ratio, 1:1) were wing banded and assigned to 12 indoor floor pens (12

groups as per genotype, sex, and diet combination, 210 chickens per pen, 11.35 birds per 1 m²). Two diets differing in protein level were used. The control groups received commercial types of feed mixture for growing chickens, and the experimental groups were fed a diet with 6% lower CP than the control diet (Table 1). The feed mixtures, that is, starter, grower and finisher, were provided as per the scheme in Table 2. Throughout the experiment, water was provided *ad libitum* for all groups. The environmental conditions were in accordance with the requirements for growing chickens and were the same for all groups. The lighting regime was 23 h of light from 1 to 7 d of age, 18 h of light from 8 to 67 d of age, and 23 h of light from 68 to 70 d of age.

When the chickens reached an average live weight of 2 kg, 10 males and 10 females from each genotype and the diet were selected for slaughter (yielding a total of 120 chickens). Ross 308 chickens reached the slaughter weight of 2 kg at 35 d of age, JA757 chickens at 42 d of age, and ISA Dual at 70 d of age. Chickens were slaughtered at the experimental slaughterhouse of the International Poultry Testing Station, Ústřašice. Birds were electrically stunned, bled, and defeathered after hot bath. Birds were eviscerated manually, and the head and distal portions of the legs were removed. The carcass weight without giblets was determined for calculation of hot DOP. Among carcass characteristics, breast percentage was selected for evaluation because it is the main valuable part, and measurements of meat quality were performed in breast meat. Breast percentage was calculated as breast meat weight divided by hot carcass weight without giblets. Abdominal fat percentage was selected for analysis owing to its relationship with chemical meat composition and was calculated by dividing the weight of the abdominal fat by the hot carcass weight.

Samples of right *pectoralis major* (PM) were collected for histochemical analysis 45 min postmortem. The remaining breast meat was chilled at 4°C for 24 h for analyses of chemical and physical meat characteristics.

Meat Chemical Composition

The left breast was collected for determination of chemical composition. Chemical analyses (of DM, CP, ether extract, and ash) were performed as per Association of Official Analytical Chemists methods (AOAC, 1995). DM was determined by drying the samples at 105°C to a constant weight. CP was detected by the Kjeldahl method (using a factor of 6.25), and ether extract was obtained by the Soxhlet method (AOAC, 1995). Ash content was performed after sample combustion at 550°C in a muffle furnace. For cholesterol content analysis, 2 g of PM was saponified with potassium hydroxide in ethanolic solution, and cholesterol was extracted with n-hexane. A validated gas chromatographic method was used for determination of cholesterol content (Perkin Elmer, model 5000). The content of total cholesterol in PM was calculated based on the external standard technique from a standard curve of

Table 1. Composition of experimental diets.

	Starter		Grower		Finisher	
	C	LP	C	LP	C	LP
Ingredient (%)						
Wheat	45.16	49.15	57.63	57.57	63.99	62.59
Corn	15.00	17.00	8.00	15.00	5.00	15.00
Soybean meal	31.05	28.75	26.85	22.95	22.35	18.60
Fish meal	1.00	-	-	-	-	-
Monocalcium phosphate	0.88	1.01	0.63	0.61	0.57	0.44
Calcium carbonate	1.44	1.52	1.12	1.24	1.08	0.84
Salt	0.28	0.27	0.25	0.29	0.28	0.28
Soybean oil	3.41	1.20	1.00	1.00	-	1.20
Animal fat	-	-	2.93	-	5.58	-
Sodium sulfate	0.11	0.12	0.12	0.08	0.08	0.08
Amino acid premixes	0.80	0.66	0.77	0.82	0.69	0.62
Vitamin and mineral supplement	0.88	0.33	0.70	0.44	0.37	0.35
Calculated composition						
CP (g/kg)	21.59	20.39	19.72	18.59	18.05	17.03
ME (MJ/kg)	12.55	11.86	12.90	12.12	13.50	12.43
Ether extract (g/kg)	5.44	3.26	5.76	3.04	7.32	3.26
Digestible lysine (g/kg)	1.29	1.18	1.16	1.12	1.03	0.97
Digestible methionine (g/kg)	0.60	0.53	0.52	0.49	0.46	0.41
Calcium (g/kg)	0.94	0.84	0.77	0.64	0.70	0.46
Nonphytate phosphorus (g/kg)	0.45	0.62	0.39	0.51	0.35	0.46

Abbreviations: C, control group fed a conventional diet for growing chickens; LP, chickens fed a low-protein diet.

peak area vs. concentration. Results were expressed as mg cholesterol/1000 g of meat.

Physical Meat Quality

For determination of physical parameters of meat quality, the right breast was used. pH values were detected 24 h postmortem using a Jenway 3510 pH meter (Jenway, Essex, England), with a glass injection probe introduced 1 cm deep into the PM. Meat color was measured 24 h postmortem on the transversal section of the PM using a Minolta SpectraMagic NX analyzer (Konica Minolta Sensing, Inc., Osaka, Japan) with the CIE 1976 Lab system. Instrumental meat color was expressed as L* (lightness), a* (redness), and b* (yellowness). The color of the skin surface in the upper third of the PM was also measured. Drip loss was determined by calculating the difference between the weight of the right breast at the time of slaughter and after storage for 24 h at 4°C.

Meat tenderness was detected in PM muscle by the Warner-Bratzler method. After dissection, the samples of meat were frozen at -20°C. Before analysis, the samples were defrosted at 4°C for 24 h and then packaged in zip-tied plastic bags and heated in a water bath at 75°C

for 1 h. The cooled meat samples were cut into 2 × 1 cm² cuboids, with the cuts running perpendicular to the muscle fibers. Meat tenderness was measured using an Instron Model 3342 instrument (Instron, Norwood) with a Warner-Bratzler shear blade containing a triangular hole to detect maximum shear force (F_{max}; N); the load cell was 20 N with a crosshead speed of 100 mm/min and a sampling rate of 20 points/s. In addition to meat tenderness, the freezing and cooking losses were calculated. The freezing loss was measured from the weight of PM before freezing (at -20°C) and after thawing (at 4°C for 24 h). The cooking loss was calculated from the differences between the weight of the raw and cooked (for 1 h at 75°C) PM samples.

Histochemical Analysis

Samples from the right part of the PM were collected immediately after slaughter and frozen in 2-methylbutane, cooled with liquid nitrogen (-156°C), and stored at -80°C until histochemical analysis. Cross sections (12-μm thickness) were then obtained from each sample with a Leica CM 1850 cryostat (Leica Microsystems Nussloch GmbH, Nussloch, Germany) at -20°C. Staining for basic histochemical characteristics was performed using hematoxylin and eosin.

The muscle fiber characteristic number of muscle fibers per 1 mm², fiber cross-sectional area (CSA), and diameter were determined with NIS Elements AR 3.1 software (Nikon, Tokyo, Japan).

Statistical Analysis

The data were processed with SAS software 9.4 (SAS Institute Inc., 2013). The data of carcass characteristics, meat quality, and histologic parameters of muscle fibers

Table 2. Feeding scheme.

Genotype and diet	Feeding scheme (day of age)		
	Starter	Grower	Finisher
Ross 308 C	1-14	15-28	29-35
Ross 308 LP	1-14	15-28	29-35
JA 757 C	1-14	15-35	36-56
JA757 LP	1-21	22-35	36-56
ISA Dual C	1-21	22-42	43-70
ISA Dual LP	1-21	22-42	43-70

Abbreviations: C, control group fed a conventional diet for growing chickens; LP, chickens fed a low-protein diet.

were analyzed by three-way ANOVA, including the interaction of genotype, sex, and diet (general linear model procedure). Genotype, sex, and diet were considered as fixed effects. Differences between means with $P < 0.05$ were considered statistically significant and tested by the Duncan test. $P < 0.05$ was considered significant for all analyses, and statistically significant differences were indicated by different superscript letters. For all statistical analyses, the individual bird was the experimental unit.

RESULTS

Carcass Characteristics

There was no 3-way interaction effect of genotype, sex, and diet on the selected carcass characteristics (Table 3), but an effect of genotype was observed on all of the selected carcass characteristics. The medium-growing JA757 chickens and slow-growing ISA Dual chickens had lower (-2.0 and -9.9% , respectively) DOP ($P < 0.001$) than the fast-growing Ross 308 chickens. The effect of genotype ($P < 0.001$) on breast percentage was similar to that on DOP. The highest ($P < 0.001$) abdominal fat percentage was found in the slow-growing ISA Dual chickens, followed by the medium-growing JA757 chickens; the lowest percentage was found in the Ross 308 chickens.

Females had higher breast meat ($P = 0.034$) and abdominal fat percentages ($P < 0.001$) than males. The greatest differences in abdominal fat percentage between males and females were observed in the medium-growing chickens of genotype JA757 and the slow-growing ISA Dual chickens, with a significant interaction effect of genotype and sex on abdominal fat percentage observed ($P < 0.001$).

There was a significant main effect of diet ($P < 0.001$), with the low protein level associated with a lower abdominal fat percentage (-0.3%)

Meat Chemical Composition

There was no significant 3-way interaction effect of genotype, sex, and diet on the chemical composition of breast meat (Table 4). Among the fixed effects, genotype had stronger effects on all parameters of meat chemical composition except ash and cholesterol contents. The highest DM and CP contents ($P < 0.001$ for both) and lowest ether extract content ($P < 0.001$) in breast were detected in slow-growing ISA Dual chickens, following by the medium-growing JA757 chickens; the fast-growing chickens of genotype Ross 308 had the lowest DM and CP contents and the highest ether extract content. Ash content was affected only by sex ($P = 0.048$), with higher values in females than in males.

The diet had a minor effect on the chemical composition of PM. A significant effect of diet was observed for only cholesterol content ($P = 0.042$), with the chickens receiving the low-protein diet having higher cholesterol content than those receiving the control diet.

Physical Meat Quality

Significant interaction effects of genotype, sex, and diet on the a^* (redness) and b^* (yellowness) parameters of breast skin were observed (Table 5). Females of Ross 308 fed the diet with lower protein level had the highest a^* ($P < 0.001$) and b^* ($P = 0.007$) of breast skin, whereas ISA Dual females fed the control diet had the lowest values of these color parameters. Significant effects of genotype on the L^* (lightness) and a^* (redness) of breast skin ($P < 0.001$ for both parameters) were observed. Medium-growing JA757 chickens had lighter skin color than the other genotypes, and Ross 308 had greater redness of breast skin than the other genotypes. The color of PM measured on transversal section was lighter ($P < 0.001$) with lower redness ($P < 0.001$) and higher yellowness ($P < 0.001$) in ISA Dual chickens than in the other genotypes.

Table 3. Effect of genotype, sex, and diet on selected carcass characteristics.

Genotype	Sex	Diet	DOP (%)	Breast meat (%)	Abdominal fat (%)
Ross	Male	C	76.08	28.35	0.89
		LP	75.84	27.17	0.96
	Female	C	76.18	27.30	1.04
		LP	77.14	29.13	0.98
JA757	Male	C	74.97	21.25	2.35
		LP	75.21	21.45	1.91
	Female	C	74.34	22.08	3.45
		LP	74.58	23.41	2.77
ISA Dual	Male	C	67.93	13.60	2.37
		LP	68.98	13.16	2.29
	Female	C	69.27	13.91	3.67
		LP	68.80	13.96	3.04
SEM			0.33	0.57	0.10
<i>P</i> -value					
Genotype			<0.001	<0.001	<0.001
Sex			0.411	0.034	<0.001
Diet			0.258	0.429	0.003
G*S*D			0.107	0.355	0.668

Abbreviations: C, control group fed a conventional diet for growing chickens; D, diet; DOP, dressing out percentage; G, genotype; LP, chickens fed a low-protein diet; S, sex.

Table 4. Effect of genotype, sex, and diet on chemical composition of breast meat.

Genotype	Sex	Diet	DM (%)	CP (%)	Ether extract (%)	Ash (%)	Cholesterol (mg/kg)
Ross 308	Male	C	24.95	21.43	1.18	1.15	428.31
		LP	25.29	21.92	0.99	1.18	399.92
JA757	Female	C	24.49	21.19	0.83	1.17	299.21
		LP	24.92	21.60	0.99	1.18	448.23
	Male	C	24.71	22.15	0.50	1.12	343.77
		LP	24.89	22.44	0.40	1.11	321.55
ISA Dual	Female	C	25.43	22.78	0.47	1.21	315.88
		LP	25.37	22.83	0.43	1.17	330.10
	Male	C	26.42	23.83	0.22	1.11	270.47
		LP	26.87	24.07	0.39	1.17	380.71
SEM	Female	C	26.46	23.86	0.31	1.15	298.39
		LP	26.60	23.94	0.31	1.17	363.18
			0.10	0.11	0.03	0.01	9.02
<i>P</i> -value							
Genotype			<0.001	<0.001	<0.001	0.552	0.065
Sex			0.904	0.747	0.255	0.048	0.522
Diet			0.219	0.158	0.975	0.638	0.042
G*S*D			0.925	0.983	0.141	0.994	0.190

Abbreviations: C, control group fed a conventional diet for growing chickens; D, diet; G, genotype; LP, chickens fed a low-protein diet; S, sex.

Females had significantly darker ($P = 0.003$) breast skin and lighter ($P < 0.001$) meat than males.

The dietary protein level influenced the yellowness of skin ($P < 0.001$) and both meat color intensity parameters ($P < 0.001$), with higher values observed in chickens fed the low-protein diet, which had darker meat ($P < 0.001$).

The pH value measured 45 min postmortem was affected by the 3-way interaction of genotype, sex, and diet ($P = 0.001$; Table 6), with the highest value observed in females of Ross 308 fed the low-protein diet and the lowest in ISA Dual males fed the control diet. Genotype affected almost all of the meat quality characteristics. The slow-growing ISA Dual chickens had lower pH45 and pH24 values ($P < 0.001$) than the JA757 or Ross 308 chickens. The drip loss was not significantly affected by genotype but the freezing and cooking losses were. The medium-growing chickens JA757

had the highest freezing and cooking losses; the losses of ISA Dual chickens did not differ from those of Ross 308 chickens. Genotype also influenced meat tenderness as measured by maximum shear force. ISA Dual chickens had the highest shear force ($P < 0.001$), followed by JA757 chickens; the tenderest meat was observed in Ross 308.

Sex had a significant effect only on meat tenderness, with males having more tender meat than females ($P < 0.001$).

The low-protein diet increased ($P = 0.006$) drip loss, but the diet had no effect on pH, freezing or cooking loss, or shear force value, all of which were affected by genotype ($P < 0.001$).

Histochemical Analysis

The muscle fiber characteristics of the PM are provided in Table 7. The number and area of muscle fibers

Table 5. Effect of genotype, sex, and diet on breast skin color and PM color.

Genotype	Sex	Diet	Skin color			Meat color		
			L*	a*	b*	L*	a*	b*
Ross	Male	C	67.71	3.89 ^B	15.11 ^{B,C,D,E}	50.02	-0.67	7.82
		LP	64.27	2.08 ^{C,D,E}	10.99 ^F	47.82	0.60	12.49
	Female	C	64.92	3.28 ^{B,C}	13.61 ^{C,D,E,F}	52.35	-0.69	8.30
		LP	66.99	5.97 ^A	19.85 ^A	49.95	0.64	13.08
JA757	Male	C	64.53	0.83 ^{D,E,F}	13.08 ^{D,E,F}	49.14	-1.02	6.61
		LP	62.64	2.33 ^{B,C,D}	18.90 ^{A,B}	47.38	0.10	11.01
	Female	C	60.41	1.11 ^{D,E,F}	11.53 ^{E,F}	52.99	-1.27	6.88
		LP	58.25	0.44 ^{E,F}	15.02 ^{B,C,D,E}	47.34	0.74	12.21
ISA Dual	Male	C	68.56	0.46 ^{E,F}	13.23 ^{C,D,E,F}	59.21	-2.36	7.84
		LP	71.02	1.09 ^{D,E,F}	17.53 ^{A,B,C}	53.81	-0.99	13.46
	Female	C	65.66	0.28 ^F	11.31 ^{E,F}	58.38	-1.60	9.16
		LP	62.45	0.33 ^F	16.64 ^{A,B,C,D}	55.31	-0.42	15.74
SEM			0.61	0.22	0.47	0.49	0.11	0.31
<i>P</i> -value								
Genotype			<0.001	<0.001	0.963	<0.001	<0.001	<0.001
Sex			0.003	0.721	0.860	0.039	0.076	0.006
Diet			0.351	0.246	<0.001	<0.001	<0.001	<0.001
G*S*D			0.121	<0.001	0.007	0.204	0.360	0.864

^{A-F}Values in the same subgroup of variables with different superscripts differ ($P \leq 0.01$).

Abbreviations: C, control group fed a conventional diet for growing chickens; D, diet; G, genotype; LP, chickens fed a low-protein diet; S, sex.

Table 6. Effect of genotype, sex, and diet on pH value, water-holding capacity and meat tenderness measured in PM.

Genotype	Sex	Diet	pH 45	pH 24	Drip loss (%)	Freezing loss (%)	Cooking loss (%)	F max (N)
Ross	Male	C	6.40 ^{B,C,D}	5.91	0.56	6.84	23.90	8.91
		LP	6.22 ^{C,D,E}	5.91	0.85	8.05	25.89	8.05
	Female	C	6.53 ^{A,B}	5.80	0.85	6.59	23.47	7.78
		LP	6.74 ^A	5.86	0.79	6.17	24.12	9.51
JA757	Male	C	6.52 ^B	5.67	0.73	10.11	28.52	18.32
		LP	6.48 ^{B,C}	5.72	1.05	9.73	28.13	15.96
	Female	C	6.48 ^{B,C}	5.60	0.74	11.00	27.11	11.87
		LP	6.56 ^{A,B}	5.75	0.82	11.20	27.98	12.98
ISA Dual	Male	C	6.17 ^E	5.66	0.79	6.36	23.94	16.68
		LP	6.52 ^B	5.61	1.01	5.77	24.27	17.15
	Female	C	6.28 ^{C,D,E}	5.64	0.71	7.78	24.30	14.69
		LP	6.22 ^{D,E}	5.64	1.07	7.75	24.01	16.62
SEM			0.03	0.02	0.04	0.31	0.26	0.24
<i>P</i> -value								
Genotype			<0.001	<0.001	0.327	<0.001	<0.001	<0.001
Sex			0.052	0.190	0.987	0.277	0.145	<0.001
Diet			0.189	0.195	0.006	0.995	0.210	0.367
G*S*D			0.001	0.848	0.344	0.647	0.420	0.543

^{A-E}Values in the same subgroup of variables with different superscripts differ ($P \leq 0.01$).

Abbreviations: C, control group fed a conventional diet for growing chickens; D, diet; F max, maximum shear force; G, genotype; LP, chickens fed a low-protein diet; PM, pectoralis major; S, sex.

are known to be negatively correlated with each other; the present results were consistent with this observation. Slow-growing ISA Dual chickens had a lower number of muscle fibers ($P < 0.001$) than medium-growing JA757 chickens and fast-growing Ross 308 chickens. The cross-sectional area ($P < 0.001$) and diameter ($P < 0.001$) were significantly affected by the interaction of genotype, sex, and diet. The largest cross-sectional area and diameter of fibers were found in females of the Ross 308 genotype fed the low-protein diet, whereas the smallest CSA was observed in males of the slow-growing ISA Dual chickens fed the normal-composition diet. As the growth rate increased, the CSA and diameter increased, demonstrating the main effect of genotype ($P < 0.001$). Slow-growing chickens

had lower fiber CSA and diameter ($P < 0.001$ for both) in PM than both JA757 and Ross 308 chickens. In addition, the low-protein diet was associated with an increased CSA of muscle fibers ($P = 0.047$).

DISCUSSION

Carcass Characteristics

Commercial broilers usually reach marked weight at around 35 d of age. In our study, the fast-growing Ross 308 chickens reached 2 kg of live weight at 35 d of age. The JA757 chickens reached this weight at 42 d of age and the ISA Dual chickens reached it at 70 d of age. Comparisons of animals at similar slaughter weight

Table 7. Effect of genotype, sex and diet on histological characteristics of PM.

Genotype	Sex	Diet	Number of muscle fibers per 1 mm ²	Cross-sectional area (μm ²)	Diameter (μm)
Ross	Male	C	260.0	3,296 ^A	224.6 ^A
		LP	238.7	3,021 ^B	208.6 ^{B,C,D}
	Female	C	223.3	3,299 ^A	222.3 ^{A,B}
		LP	228.7	3,417 ^A	227.3 ^A
JA757	Male	C	249.3	2,960 ^B	212.0 ^{B,C}
		LP	254.7	2,986 ^B	209.5 ^{C,D}
	Female	C	276.0	2,753 ^C	201.1 ^D
		LP	259.3	2,975 ^B	211.3 ^C
ISA Dual	Male	C	450.4	1,440 ^E	146.4 ^G
		LP	424.7	1,750 ^D	163.9 ^E
	Female	C	429.1	1,598 ^D	152.3 ^{F,G}
		LP	458.0	1,654 ^D	157.0 ^{E,F}
SEM			12.23	20.90	1.00
<i>P</i> -value					
Genotype			<0.001	<0.001	<0.001
Sex			0.969	0.290	0.586
Diet			0.771	0.047	0.101
G*S*D			0.513	0.001	<0.001

^{A-G}Values in the same subgroup of variables with different superscripts differ ($P \leq 0.01$).

Abbreviations: C, control group fed a conventional diet for growing chickens; D, diet; G, genotype; LP, chickens fed a low-protein diet; PM, pectoralis major; S, sex.

may be useful for practical commercial purposes (Hernández et al., 2004). Despite similar slaughter weight, the DOP of chickens differed among the genotypes, being 76.3, 74.8, and 68.7% for the fast-, medium-, and slow-growing genotypes, respectively. These results agree with those of the study by Devatkal et al. (2019). Similarly, as expected, breast percentage decreased in the order fast growth > medium growth > slow growth. This is in accordance with genetic selection for growth and carcass quality in commercial hybrids. Abdominal fat has little economic value and is recognized as the main source of waste in poultry production. Fat tissue undergoes very late maturation and has an allometry coefficient greater than 1 (Tůmová and Chodová, 2018), which probably explains why the fast-growing chickens, having a lower degree of maturity, had a lower abdominal fat percentage.

Shafey et al. (2013) reported positive correlations between live weight and the weights of carcass parts. Accordingly, the similar live weight of females and males in this study resulted in no effect of sex on DOP and breast percentage. Lipid synthesis is under estrogenic control, and lipid deposition in peripheral tissues is increased in females (Baéza et al., 2012). The different fatness between females and males could also arise because females start to store fat earlier (from 6 wk) than males (from 8 wk of age) (Almasi et al., 2012).

The protein level of the diet had no significant effect on DOP or breast percentage, consistent with Kamran et al. (2008). Moreover, the low-protein diet reduced the deposition of abdominal fat. This effect conflicts with those reported by Kamran et al. (2008), who showed increased accumulation of abdominal fat in chickens receiving a low-protein diet, which ate significantly greater amounts than the control birds. However, in our study, the lower abdominal fat percentage in the birds receiving the low-protein diet might have been owing to a similar ME:CP ratio between the dietary treatments in contrast with an increased ME:CP ratio in the study of Kamran et al. (2008).

Meat Chemical Composition

In our study, genotype appeared to be the main factor influencing the chemical composition of breast meat, being more important than sex and diet. Slow-growing birds exhibited a higher protein content in PM than fast-growing ones, which may have been related to the age at slaughter. In the present study, the slow-growing birds were 35 d older than the fast-growing ones. As an animal ages, the protein content increases, and the moisture content decreases (Metzger et al., 2011). Intramuscular fat is a precursor of meat flavor substances, and ether extract can enhance the juiciness and tenderness of meat. Although the amount of abdominal fat was lowest in the fast-growing chickens, the ether extract content of breasts was higher in these chickens than in the slow-growing chickens, consistent with the findings of Fanatico et al. (2007) and Mueller et al. (2018). It can be explained by the finding that in animals

with high growth rates, fat is rapidly incorporated into cells, replacing water (Metzger et al., 2011). These authors found that within groups of the same BW, the fat content of meat was lower in older animals, which agrees with the finding in chickens in the present study.

Sex did not influence any component of the proximate meat composition except ash content.

The effect of the low-protein diet on the chemical composition of the meat was minor. In contrast, Wang et al. (2013) reported that a low-protein diet led to a lower protein content and higher lipid content of breast meat, whereas Fanatico et al. (2007) reported that a conventional diet led to a higher fat content than a low-protein diet because the conventional diet was higher in energy. The discrepancies between these previous studies and our results could be related to study differences in the experimental diets and conditions.

Physical Meat Characteristics

Among the physical characteristics of meat, color is most important to consumers and is affected by numerous factors, including the presence and concentrations of heme pigments, genetics, and diet (Batkowska et al., 2015). Meat color is most often measured in cross section of muscle; however, as the chicken is also offered to consumers as whole carcass, we believe that skin color is also important to consumers. In the present study, darker and redder skin color was observed in the medium-growing chickens fed the low-protein diet than in the other bird combinations. The darker color of PM in the medium-growing chickens is in accordance with the results of Sirri et al. (2011). However, the differences in meat color characteristics among the genotypes could have been a consequence of differences in slaughter age/maturity. In chickens, older birds have darker and redder breast meat than younger ones (Baéza et al., 2012). In our experiment, the fast-growing chickens, which were slaughtered at younger age than the slow-growing ones, had higher lightness and yellowness values; however, the ISA Dual chickens in our experiment had lighter meat than the JA757 chickens despite being of greater age. The L^* value indicates the degree of paleness. Breast meat with L^* values higher than 54 is considered light and tends to be pale, soft, exudative meat (Woelfel et al., 2002). In our study, the ISA Dual genotype had such light meat; however, the pH value was in the range for normal meat, so the light color may be a characteristic of this genotype.

The reason for the difference in lightness and yellowness of meat from males and females could be the greater amount of subcutaneous fat in females.

The low-protein diet significantly increased the lightness and color intensity of the meat. The larger area of muscle fibers found in the chickens fed the low-protein diet might explain the greater reflection of light rays and thus the greater lightness of meat from these birds.

In the present study, genotype affected most of the meat quality characteristics. The pH value declines postmortem, and this process is very important in the conversion

of muscle to meat as it affects meat characteristics such as color, water-holding capacity, and tenderness (Chodová et al., 2019). Low pH is associated with poor meat quality. In the present study, the PM of medium-growing chickens had a higher pH45 than that of fast- or slow-growing birds; the latter had the lowest pH45. Debut et al. (2005) reported that shackling stress before slaughter led to rapid muscle acidification. It is possible that the slow-growing chickens experienced more stress such that postmortem glycolysis was accelerated, contributing to the low pH45 in these birds.

The pH measured at 24 h postmortem should be in the range 5.6–6. Fanatico et al. (2007) observed lower pH24 values for slow-growing chickens than for fast-growing ones at similar slaughter weight, in accordance with our results. The differences among genotypes were significant, but no pale, soft, exudative meat was observed. Selection on BW and carcass traits could have caused a reduction in the glycogen reserves of breast muscles, thereby contributing to the observed differences in pH24 among the genotypes (Quentin et al., 2003).

Water-holding capacity affects the functionality, processing ability, and sensory characteristics of meat. It can be measured from drip, freezing, or cooking loss. A negative correlation between drip loss and pH24 of chicken breasts has been reported (Le Bihan-Duval et al., 1999). Consistent with this observation, higher drip losses and lower pH24 values were observed in the slow- and medium-growing chickens than in the fast-growing ones in our experiment. Fanatico et al. (2007) and Sirri et al. (2011) showed that the higher drip loss in slow-growing chickens was probably owing to smaller size and larger surface area of muscle in those birds than in fast-growing birds. However, in the present study, the effect of genotype on drip loss was not significant. On the other hand, after subsequent adjustments of the meat, such as freezing or boiling, the effect of genotype on these characteristics was significant, with the highest freezing and cooking losses observed in the medium-growing chickens. In rabbits, a higher water-holding capacity was observed in older animals (Hernández et al., 2004) because of the lower maturity of the meat. This reason might also explain the greater water-holding capacity of meat from ISA Dual chickens, which were slaughtered at 70 d of age.

Meat tenderness is determined by muscle structure, collagen content, and postmortem biochemical changes. The hardness of the meat from slow-growing birds could have been due to the higher age at slaughter of this genotype; with increasing age, the intramuscular connective tissue content increases, which decreases meat tenderness. Moreover, the observed differences in meat tenderness among genotypes can be explained by the combined effects of intramuscular fat and protein turnover and differences in collagen structure. Fast-growing chickens had more intramuscular fat in PM than the other genotypes, and greater intramuscular fat is usually associated with higher tenderness of meat.

The females and males had similar pH values and water-holding capacity, which indicates a lack of structural differences in muscle between the sexes, as also

evidenced by the muscle fiber characteristics. The more tender meat in females is probably related to smaller size of muscle fibers in females.

It seems that a diet with only a 6% reduction in protein has no negative effect on the measured physical properties of meat quality, in agreement with other experiments (Fanatico et al., 2007; Wang et al., 2013).

Histochemical Analysis

Muscle mass is determined by the number of muscle fibers that are formed before hatching and by the cross-sectional area after hatch. Muscle fiber characteristics are affected by numerous factors, including genotype, sex, rearing system, nutrition, age, and muscle location. Chicken breast contains only type IIB muscle fibers. In the present study, significant interaction effects of genotype, sex, and diet on fiber CSA and diameter in PM were observed. As similar interaction effects have not been studied previously, comparisons of our results with the literature are not possible. A main effect of genotype was observed, with the fast-growing genotype, selected for enhanced growth, having the highest breast percentage among the genotypes owing to the higher muscle accretion and increased fiber CSA in this genotype than in the other genotypes in agreement with Devatkal et al. (2019). Larger myofibers have reduced glycolytic potential than smaller ones; thus, muscle with these fiber characteristics has higher pH measured 24 h postmortem, as detected in our experiment. Increased CSA and diameter of PM fibers contribute to increased meat tenderness, which could explain the lower shear force of muscle from fast-growing birds in our experiment.

In the present study, no significant differences in the muscle fiber characteristics in PM were detected between male and female chickens, consistent with Chiang et al. (1995) who reported that sex had no influence on either the proportions of muscle fiber type or fiber CSA.

An adequate nutrient supply plays an important role in the postnatal period; however, the dietary effects on muscle fiber characteristics remain unclear. The present experiment revealed no effect of diet on the number of muscle fibers. On the other hand, the low-protein diet was associated with a larger CSA of fibers in PM, indicating increased muscle fiber hypertrophy, consistent with our finding of a higher breast percentage in chickens fed the low-protein diet.

CONCLUSION

In conclusion, the studied main effects had stronger influences on the interaction effects on the investigated carcass characteristics and meat quality traits. The interaction of genotype, sex, and diet had negligible effect on the meat quality parameters, and all genotypes responded similarly to dietary protein level at the slaughter weight of 2 kg. The results showed that selection on growth rate in fast-growing chickens could be

associated with lower CP and higher ether extract contents and increased pH in PM than those observed in medium- and slow-growing chickens. The meat from slow-growing chickens can be considered more suitable for specialized markets owing to its higher values of color intensity parameters and lower tenderness.

It seems that the low-protein diet had a particularly negative effect on meat quality, as it significantly increased cholesterol content in breast meat, drip loss, and size of muscle fibers.

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DISCLOSURES

The authors declare no conflicts of interest.

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