# **Fatty acid composition of meat and estimated indices of lipid metabolism in different poultry genotypes reared under organic system**

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 **ABSTRACT** According to EC regulation 889/08, different European countries should draw up a list of slowgrowing strains adapted to an organic system, and in the meantime, provide this information to operators and the European Union commission. Thus, the aim of the present work was to evaluate the effect of poultry genotype on fatty acid composition and lipid indices of poultry meat. Six poultry genotypes (100 birds each), each with a different growth rate (slow-growing: Leghorn, Ancona, Cornish  $\times$  Leghorn; medium-growing: Kabir, Naked neck; fast-growing: Ross), were reared under an organic system. Breast meat fatness, fatty acid composition, and indices were largely related to genotype, as slow-growing strains had higher elongase, thioesterase, and  $\Delta^5/\Delta^6$  desaturase indices accompanied by a lower  $\Delta^9$ . Differences in the fatty acid profiles were observed by varying contents of total saturated fatty acids, with a higher value seen in Leghorn chickens and a lower value seen in commercial lines. On the contrary, Leghorn and Ancona chickens exhibited higher amounts of stearic acid and total polyunsaturated fatty acids compared with commercial genotypes, both in the total content and in the different fractions (total n-3 and total n-6). Despite the increased consumption of fresh forage, the lower linolenic acid in meat of the slow-growing strain could be explained by the higher conversion of this fatty acid to its long-chain derivatives.

**Key words:** organic rearing system, chicken genotype, fatty acid

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## **INTRODUCTION**

 Consumer interest in organic and natural poultry products is expanding, and consequently, the organic market has grown by 20% annually for the past decade (Fanatico et al., 2005, 2007; Hughner et al., 2007). In Europe, organic poultry production is regulated by different national and international rules regarding the choice of genotype. EC regulation 1804/99 and the Network for Animal Health and Welfare in Organic Agriculture's final recommendation (Hovi et al., 2003) suggest to use local, slow-growing breeds for their higher rusticity and capacity to use outdoor areas and pasture.

 In a recent review, Sossidou et al. (2010) reported various meanings of the term pastured poultry and defined the different pasture-based poultry production systems. The authors raised attention to production topics such as poultry genetics, outdoor access, and pasture management as critical points necessary to obtain successful results. Indeed, many findings revealed that fast-growing chickens are not adapted to extensive rearing conditions, as they exhibit muscular-skeletal problems and very low motor activity and foraging behavior (Castellini et al., 2002a,b,c; Fanatico et al., 2005; Branciari et al., 2009; Sirri et al., 2010). On the other hand, slow-growing chickens possess a good aptitude for pasture, which enhances the dietary intake of bioactive substances (vitamins, antioxidants, and fatty acids) contained in the forage (Mugnai et al., 2009). Hughes and Dun (1983) estimated that a free-range laying hen can consume up to 30 to 40 g of DM/d (forage, insects, earthworms), which could reduce the feed cost that represents approximately 70% of the total variable costs (Walker and Gordon, 2003).

 Unfortunately, slow-growing birds displayed very poor productive performance, and the use of such strains in commercial production would be unprofitable. One possible option could be to create crossed lines capable of increasing live weight and feed efficiency, and at the same time, maintaining suitable grazing behavior. Pasture exerts relevant effects both on product quality and on animal health (Lopez-Bote et al., 1998; Sossidou et al., 2010); indeed, it improves the oxidative stability of meat (Castellini et al., 2006b) and plays an important role in the immune system of birds (Mugnai et al., 2011). Hence, one of the aims of an organic production system (Council Regulation, 1999) is to enhance the

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rearing and diffusion of local genotypes, which in the last 50 yr have been replaced by hybrid birds.

From a qualitative point of view, organic chickens are expected to eat variable amounts of forages that could modify fatty acid profiles in their meat, but it is not well known by how much the chicken is able to elongate and desaturate the linolenic acid (**ALA**, C18:3n-3) of the pasture into eicosapentaenoic acid (**EPA**, C20:5n-3) and docosahexaenoic acid (**DHA**, C22:6n-3; Rymer and Givens, 2005). Thus, the aim of this study was to evaluate the fatty acid composition and fatty acid indices of meat from different chicken genotypes reared under organic conditions.

### **MATERIALS AND METHODS**

#### *Birds and Diets*

This trial was conducted at the experimental section of University of Perugia (Italy) from March to May 2010. All animals were reared according to EU regulation 834/07 and Italian directives (Gazzetta Ufficiale, 1992) on animal welfare for experimental and other scientific purposes.

Six hundred male birds were compared and categorized with regard to their growth rhythm: slow  $\langle$  <20  $g/d$ ), medium (20 > 35 g/d) or fast (>35 g/d) (RCE, 2007; RCE, 2008). The genotypes were the following: Ancona  $(A)$ , Leghorn  $(L)$ , crossbreed Cornish  $\times$  Leghorn (**CL**), Kabir (**K**), Naked neck (**NN**), and Ross (**R**).

The L and A genotypes are originated from a conservation flock at the Department of Applied Biology in the 1960s. The CL chicks were produced by crossing L hens with Cornish fowl (from local farmer), whereas K (strain KR4), NN (strain NN1), and Ross 308 were furnished by a commercial poultry farm (Avicola Berlanda, Italy).

Chickens were kept separate after hatching until 20 d of age in an environmentally controlled poultry house with temperatures ranging from 20 to 32°C and with RH ranging from 65 to 75%. Incandescent light (30 lx) placed at bird level was used for heating and illumination. Chicks were vaccinated against Marek and Newcastle diseases.

At 21 d of age, the chicks were transferred to strawbedded indoor pens  $(0.10 \text{ m}^2/\text{bird})$  each equipped with feeders and drinkers and with free access to forage paddock  $(4 \text{ m}^2/\text{bird})$ . Each genotype was represented in 4 replicates containing 25 chicks each. Birds were confined to indoor pens during night.

The pasture lands were not treated with pesticides or herbicides during the 3 yr before organic production. The pasture area also contained mature trees, bushes, and hedges.

Birds were raised until the minimum slaughter weight (81 d); NN, K, and R reached 2.50 kg, 2.38 kg, and 4.5 kg, respectively, whereas the crossbreed (1.8 kg) and the pure breeds (1.4 kg and 1.3 kg for A and L, respectively) were much lighter.

**Table 1.** Ingredient composition and calculated analysis of diets (%, unless otherwise indicated)

Item	Starter	Finisher	
Ingredient			
Maize	52.0	46.0	
Full-fat soybean	30.5	12.5	
Wheat		20.0	
Soybean meal <sup>1</sup>	9.00	14.0	
Alfalfa meal	2.80	2.80	
Gluten feed	3.00	2.00	
Vitamin-mineral premix <sup>2</sup>	1.00	1.00	
Dicalcium phosphate	1.00	1.00	
Sodium bicarbonate	0.50	0.50	
NaCl	0.20	0.20	
Chemical composition			
DM	90.9	90.8	
CP	22.3	18.0	
Ether extract	7.95	4.98	
Crude fiber	4.67	4.01	
Ash	5.76	5.59	
Neutral detergent fiber	10.7	10.1	
Acid detergent fiber	5.58	5.06	
Cellulose	4.22	3.56	
Acid detergent liquid	1.03	1.11	
Hemicellulose	5.16	5.05	
ME (MJ/kg of DM)	12.5	12.9	

<sup>1</sup>From conventional crops.

2Amounts per kilogram: vitamin A, 11.000 IU; vitamin D3, 2.000 IU; vitamin  $B_1$ , 2.5 mg; vitamin  $B_2$ , 4 mg; vitamin  $B_6$ , 1.25 mg; vitamin  $B_{12}$ , 0.01 mg;  $\alpha$ -tocopheryl acetate, 30 mg; biotin, 0.06 mg; vitamin K, 2.5 mg; niacin, 15 mg; folic acid, 0.30 mg; pantothenic acid, 10 mg; choline chloride, 600 mg; Mn, 60 mg; Fe, 50 mg; Zn, 15 mg; I, 0.5 mg; Co, 0.5 mg.

Chickens were fed ad libitum the same starter (1–21 d) and grower-finisher (22 d to slaughter) diets (Table 1) containing 100% organic ingredients certified by a national agency. Access to feed and water was freely available, and all diets were formulated to contain adequate nutrient levels as defined by the NRC (1994).

The chemical composition in each pasture pen  $(n =$ 24) was estimated by cutting a  $1-m^2$  fenced area using garden scissors (at 5 cm above soil) at the beginning and at the end of the trial.

A sample of 20 birds per strain, each weighing between  $\pm 10\%$  of the population mean, were slaughtered in the department processing plant 12 h after feed withdrawal. Chickens were not transported and were electrically stunned (110 V; 350 Hz) before killing. After killing, the carcasses were placed in hot water (56.5°C for 1 min) and then plucked, eviscerated (nonedible viscera: intestines, proventriculus, gall bladder, spleen, esophagus, and full crop), and stored for 24 h at 4°C.

From the carcass, the pectoralis major muscle was excised for analysis. Samples were immediately analyzed in duplicate to determine lipid amounts. Total lipids were extracted in duplicate from 5 g of each homogenized sample and calculated gravimetrically (Folch et al., 1957). Fatty acids were quantified as methyl esters (**FAME**) with a Mega 2 Carlo Erba gas chromatograph (model HRGC, Milano, Italy), using a D-B wax capillary column  $(0.25 \text{ mm } \varnothing, 30 \text{ m long}).$ 

The FAME peaks were identified by comparing the retention time with the commercially available FAME standards. The fatty acid compositions were calculated using the peak areas and expressed on percentage basis. The average amount of each fatty acid was used to calculate the sum of the total saturated (**SFA**), total monounsaturated (**MUFA**), and total polyunsaturated (**PUFA**) fatty acids.

Several indices were used to estimate desaturase and elongase activity of muscle tissue. Estimated desaturase activities are often used and, among many other authors, Vessby et al. (2002) reported that the calculated activities of  $\Delta^9$ -,  $\Delta^5$ -, and  $\Delta^6$ -desaturase can be used as surrogates of the measure of the true desaturase activity.

In particular, the activities of  $\Delta^9$ -desaturase and elongase were estimated by relating the percentage of product to the percentage of precursor (Okada et al., 2005). Specifically, the  $\Delta^9$ -desaturase index for the C18:1, which is the main MUFA of poultry meat, was calculated as 100 times the ratio of oleic acid (C18:1) to the sum of C18:1 and stearic acid (C18:0). The total  $\Delta^9$ -desaturase index (both 16 and 18) was calculated as 100 times the ratio of the sum of C16:1 and C18:1 to the sum of C16:1, C16:0, C18:1, and C18:0.

The elongase index was calculated as the ratio of C18:0 to C16:0, whereas the thioesterase index was calculated as the ratio of C16:0 to myristic acid (C14:0; Zang et al., 2007).

To evaluate the activity of both  $\Delta^5$ -desaturase and  $\Delta^{6}$ -desaturase, the enzymes catalyzing the formation of long-chain n-6 and n-3 PUFA starting from the precursors C18:2n-6 and C18:3n-3, the following equation was used (Sirri et al., 2010):

 $\Delta^5$ -desaturase +  $\Delta^6$ -desaturase =  $[C20:2n-6 + C20:4n-6 + EPA + C22:5n-3$  $+$  DHA/C18:2n-6 (LA)  $+$  ALA  $+$  C20:2n-6  $+$  C20:4n-6 + EPA + C22:5n-3 + DHA]  $\times$  100.

#### *Statistical Analyses*

The data were analyzed with a linear model (Stata-Corp., 2005) to evaluate the effect of genotype. The significance of differences  $(P < 0.05)$  was evaluated by multiple *t*-tests.

## **RESULTS AND DISCUSSION**

The fatty acid compositions of both the diets and the forage are shown in Table 2.

According to other authors (Ponte et al., 2008), linoleic acid (**LA**, C18:2n-6) is the major fatty acid in feed, whereas ALA is predominant in forage. In particular, Gurr (1984) reported that ALA is esterified in structural lipids, including galactolipids from chloroplasts.

The main floristic species found in pasture in the different pens were represented by *Lolium perenne*, *Lotus corniculatus*, *Sorgum halepense*, and *Trifolium pretense*, which represented 72% of the pasture floristic composition (data not shown). Concerning the differences in the fatty acid concentrations between plant species, legumes have a higher amount of ALA than grass, as reported by Dewhurst et al. (2001), Van Ranst et al. (2009), and Wyss and Collomb (2010). The pasture presented LA/ALA ratios of 0.19, whereas the feed had an LA/ALA ratio of 8.13 and 9.26 for the starter and finisher diets, respectively.

The content of antioxidants, according to our previous studies (Castellini et al., 2006a; Mugnai et al., 2009), confirmed the notable contribution of forage antioxidants that are crucial for the organic rearing system given that the use of synthetic vitamins in organic feed was banned (Council Regulation, 1999). In a cereal-based diet supplemented with 20 mg/kg of α-tocopheryl acetate for free-range chickens, Ponte et al. (2008) found  $\alpha$ -tocopherol levels of 36.9 mg/kg.

Alpha-tocopheryl acetate is the most-used antioxidant in feed industry because it is very resistant to light, oxygen, and to technological processes of production. In the intestine esterase removes the acetate group of the molecule and releases  $\alpha$ -tocopherol, which passes into the lymph stream and is transported by chylomicrons in the muscles and other tissues where tocopherol is stored (Liu et al., 1995).

The lipid content and the fatty acid profile of the breast meat are shown in Table 3. The genotype affected the lipid content of the meat: A and L chickens showed lower values, whereas the commercial strains (NN, K, and R) showed higher values. The crossbreed showed an intermediate lipid level.

The genotype effect reported herein also implicates a different behavior of birds in terms of the amount of feed/forage intake and motor activity. Commercial breeds, mainly fast-growing, showed some dynamism problems, especially in the last phase of the cycle as observed in our previous studies (Castellini et al., 2006b; Branciari et al., 2009). These genotypes, which were selected on the basis of precocity and ability to reach their market live weight at an early age, are not compatible with longer periods of raising. The combination of age, low kinetic activity, and the high feed intake resulted in a higher fat accumulation in muscles.

These results confirm that fatness is largely related to genotype (Berri et al., 2007). Sirri et al. (2010), comparing the lipid composition of different chicken strains (the fast-growing Cobb 700 strain, the medium-growing NN strain, and the slow-growing Brown Classic Lohman strain) reared under organic conditions, observed an increase in the lipid content of the fast-growing strain.

Breeds were also a source of variation for the main fatty acids. Differences in the fatty acid profiles were observed in the content of total SFA, where the higher value was observed in L and the lower value in commercial lines. Additionally, L and A chickens exhibited higher amounts of stearic acid (C18: 0).

The total MUFA concentrations, which in chickens are related either to the endogenous synthesis or to the

	Diet				
Item	Starter	Finisher	Mean pasture $(\text{initial} + \text{final})^1$		
Fatty acid <sup>2</sup>					
C16:0	625.2	601.8	299.0		
C18:0	396.1	334.6	40.4		
Others	45.1	47.5	16.2		
<b>SFA</b>	1,066.4	983.9	355.6		
$C16:1n-7$	16.9	17.7	6.80		
$C18:1n-9$	1,001.3	1,113.6	181.8		
Others	55.4	62.7	17.0		
<b>MUFA</b>	1,073.6	1,194.0	205.6		
$C18:2n-6$	1,298.8	1,345.1	256.4		
Others	18.2	14.8	3.21		
$n-6$	1,317.0	1,359.8	259.6		
$C18:3n-3$	159.6	145.2	1,342.9		
Others	28.1	22.1	5.05		
$n-3$	187.7	167.3	1,347.9		
<b>PUFA</b>	1,504.7	1,527.1	1,607.4		
$n - 6/n - 3$	7.00	8.13	0.19		
Carotenoids	18.3	19.1	102.1		
$\alpha$ -tocopherol	12.5	15.2	175.2		

**Table 2.** Fatty acid composition (mg/100 g feed or grass), total carotenoids, and α-tocopherol (mg/ kg of DM) of diets and grass

 $1<sup>n</sup> = 48.$ 

 ${}^{2}$ SFA = saturated fatty acid; MUFA = monounsaturated fatty acid; PUFA = polyunsaturated fatty acid.

gut absorption from the diet, showed the highest levels in commercial genotypes; these MUFA concentrations were mainly represented by oleic and by palmitoleic acid. The low MUFA levels observed in pure breeds can be attributed to the higher intake of pasture (average value MUFA 227 mg/100 g) with respect to feed (average value MUFA  $1,134 \text{ mg}/100 \text{ g}$  and to the different intramuscular fat content of birds (Sirri et al., 2010).

Meat of slow-growing genotypes compared with commercial genotypes was characterized by a high concentration of total PUFA, both in the total content and in the different fractions (n-3 and n-6). Despite the increased consumption of fresh forage, lower levels of ALA in the meat of slow-growing strains could be explained by the higher conversion of this fatty acid in the long-chain derivatives.

**Table 3.** Lipid content  $(g/100 g)$  of meat) and fatty acid composition  $(g/100 g)$  of fatty acids) of breast of different genotypes (n =  $20 \text{ per group}$ <sup>1</sup>

Item <sup>2</sup>	L	А	CL	K	NN	$\mathbf R$	Pooled $\rm SE$
Lipid	$0.32^{\mathrm{a}}$	0.22 <sup>a</sup>	$0.50^{\rm ab}$	0.85 <sup>b</sup>	0.73 <sup>b</sup>	1.30 <sup>c</sup>	0.16
C14:0	0.48 <sup>a</sup>	0.66 <sup>a</sup>	1.68 <sup>b</sup>	1.56 <sup>b</sup>	$0.47^{a}$	1.04 <sup>b</sup>	0.52
C16:0	28.8 <sup>a</sup>	$28.3^{\mathrm{a}}$	32.5 <sup>b</sup>	30.3 <sup>ab</sup>	$29.6^{ab}$	$31.5^{\rm ab}$	1.50
C18:0	13.3 <sup>c</sup>	$13.4^c$	8.76 <sup>a</sup>	10.1 <sup>b</sup>	9.81 <sup>a</sup>	11.0 <sup>b</sup>	2.3
Others	4.53c	3.56 <sup>b</sup>	2.00 <sup>a</sup>	2.06 <sup>a</sup>	3.72a	2.07 <sup>a</sup>	1.69
Total SFA	47.1 <sup>b</sup>	45.9 <sup>b</sup>	44.9 <sup>b</sup>	44.0 <sup>b</sup>	43.6 <sup>a</sup>	43.7 <sup>b</sup>	2.9
$C14:1n-6$	0.09 <sup>b</sup>	0.10 <sup>b</sup>	0.18 <sup>c</sup>	0.04 <sup>a</sup>	0.12 <sup>b</sup>	0.01 <sup>a</sup>	0.14
$C16:1n-7$	0.60 <sup>a</sup>	0.58 <sup>a</sup>	3.26 <sup>c</sup>	3.10 <sup>c</sup>	3.30 <sup>b</sup>	$1.41^{ab}$	1.24
$C18:1n-9$	16.9 <sup>a</sup>	$18.5^{\mathrm{a}}$	21.6 <sup>ab</sup>	23.0 <sup>b</sup>	24.3 <sup>b</sup>	24.1 <sup>b</sup>	2.71
Others	0.24	0.26	0.24	0.22	0.22	0.17	0.18
Total MUFA	17.8 <sup>a</sup>	$19.4^{\mathrm{a}}$	25.3 <sup>b</sup>	26.4 <sup>b</sup>	27.94 <sup>b</sup>	$25.7^{\rm b}$	1.93
$C18:2n-6$	20.6 <sup>b</sup>	$18.4^{\mathrm{a}}$	$22.2^{b}$	20.9 <sup>b</sup>	20.8 <sup>b</sup>	20.7 <sup>b</sup>	1.97
$C20:2n-6$	0.75 <sup>c</sup>	0.68 <sup>c</sup>	0.44 <sup>b</sup>	$0.34^{ab}$	$0.42^{ab}$	0.28 <sup>a</sup>	0.21
$C20:3n-6$	$0.15^{ab}$	0.17 <sup>ab</sup>	0.24 <sup>b</sup>	0.24 <sup>b</sup>	0.26 <sup>b</sup>	$0.16^{ab}$	0.09
$C20:4n-6$	$9.80^{ab}$	10.6 <sup>b</sup>	$5.33$ <sup>a</sup>	6.34 <sup>a</sup>	4.20 <sup>a</sup>	8.14 <sup>ab</sup>	2.01
Total n-6	31.3 <sup>b</sup>	29.8 <sup>b</sup>	$28.2^{ab}$	$27.8^{ab}$	25.8 <sup>a</sup>	26.3 <sup>a</sup>	1.43
$C18:3n-3$	$0.64^{\rm a}$	1.03 <sup>ab</sup>	$0.87^{a}$	0.93 <sup>a</sup>	1.46 <sup>b</sup>	1.03 <sup>ab</sup>	0.43
$C20:3n-3$	0.02	0.03	0.02	0.02	0.01	0.03	0.03
$C20:5n-3$	0.17 <sup>b</sup>	0.17 <sup>b</sup>	0.08 <sup>a</sup>	$0.12^{ab}$	$0.10^{ab}$	0.09 <sup>a</sup>	0.05
$C21:5n-3$	0.65 <sup>b</sup>	0.87 <sup>b</sup>	0.06 <sup>a</sup>	0.05 <sup>a</sup>	0.03 <sup>a</sup>	0.03 <sup>a</sup>	0.18
$C22:5n-3$	1.34 <sup>c</sup>	1.37 <sup>c</sup>	$0.45^{\mathrm{a}}$	$0.54$ <sup>a</sup>	0.94 <sup>b</sup>	1.02 <sup>b</sup>	0.40
$C22:6n-3$	0.94 <sup>b</sup>	1.32 <sup>b</sup>	0.09 <sup>a</sup>	0.14 <sup>a</sup>	$0.14^{a}$	0.20 <sup>a</sup>	0.39
Total n-3	3.76 <sup>c</sup>	4.79c	1.57 <sup>a</sup>	1.80 <sup>a</sup>	2.68 <sup>b</sup>	2.40 <sup>b</sup>	1.11
$n - 6/n - 3$	8.32 <sup>b</sup>	6.23 <sup>a</sup>	17.9 <sup>c</sup>	15.5 <sup>b</sup>	9.62 <sup>a</sup>	10.4 <sup>ab</sup>	2.21
Total PUFA	35.1 <sup>b</sup>	34.7 <sup>b</sup>	29.8 <sup>a</sup>	29.6 <sup>a</sup>	$28.5^{\mathrm{a}}$	$28.7^{\mathrm{a}}$	4.66

 $a$ <sup>-c</sup>Values with different superscripts within a column differ at  $P < 0.05$ .

<sup>1</sup>L: Leghorn; A: Ancona; CL: crossbreed Cornish  $\times$  Leghorn; K: Kabir; NN: Naked neck; R: Ross.

 ${}^{2}$ SFA = saturated fatty acid; MUFA = monounsaturated fatty acid; PUFA = polyunsaturated fatty acid.

**Table 4.** Main estimated indices of fatty acid metabolism (on the basis of fatty acid composition expressed as mg/100 g of meat) in breast muscle of different genotypes  $(n = 20 \text{ per group})^1$ 

Item			CL		NΝ		Pooled <b>SEM</b>
Elongase	0.46 <sup>b</sup>	$0.47^{b}$	0.37 <sup>ab</sup>	$0.27^{\rm a}$	$0.33^{a}$	$0.33^c$	0.11
Thioesterase	59.8 <sup>d</sup>	$42.7^{\circ}$	$28.3^{b}$	$19.4^{\rm a}$	$20.3^{\rm a}$	30.9 <sup>b</sup>	5.33
$\Delta^9$ -desaturase (18)	55.9 <sup>a</sup>	$58.0^{\rm a}$	$62.1^{\rm a}$	71.1 <sup>b</sup>	$68.5^{ab}$	$68.7^{\circ}$	7.47
$\Delta^9$ -desaturase (16+18)	$29.3^{\rm a}$	$31.4^{\rm a}$	$32.4^{\rm a}$	37.6 <sup>b</sup>	34.9 <sup>b</sup>	$37.5^{\rm b}$	4.30
$\Delta^{5}/\Delta^{6}$ -desaturase	$52.5^{\rm b}$	$52.4^{b}$	$23.6^{\rm a}$	$28.0^{\rm a}$	$26.4^{\rm a}$	$23.5^{\rm a}$	4.75

a<sup>-d</sup>Values with different superscripts within a column differ at  $P < 0.05$ .

<sup>1</sup>L: Leghorn; A: Ancona; CL: crossbreed Cornish x Leghorn; K: Kabir; NN: Naked neck; R: Ross.

As already affirmed, the differences in the fatty acid profiles of breast meat may be attributed to genetic and epigenetic effects or to causal interactions between genes and their products, which brings the phenotype into play, which could affect lipid metabolism and fatty acid deposition.

Indeed, some estimated indices of fatty acid metabolism (Table 4) showed that the pure breeds diverged from the other breeds. In particular, slow-growing genotypes had higher elongase, thioesterase, and  $\Delta^5/\Delta^6$ desaturase indices accompanied by a lower  $\Delta^9$  index.

In fatty acid synthesis, thioesterase is responsible for terminating the reaction and releasing the newly synthesized fatty acid. The ratio of C16:0 to C14:0 was used to reflect the selective division of thioesterase on C14-acyl-acyl carrier protein or C16-acyl-acyl carrier protein; the higher thioesterase index observed in our trial is related to less cleavage of C14-acyl-acyl carrier protein.

The  $\Delta^9$ -desaturase specific for the major MUFA of chicken lipids and the total  $\Delta^9$ -desaturase that catalyzes the conversion of C16:0 and C18:0 to C16:1 and C18:1 (Jeffcoat, 1979) were always lower in L and A birds, followed by CL and medium-fast growing strains. Some studies on beef suggested that different concentrations of C16:1 between breeds could be attributed to increased  $\Delta^9$ -desaturase activity (Sturdivant et al., 1992; Laborde et al., 2001). According to Kouba et al. (2003), in pigs, the  $\Delta^9$ -desaturase activity decreased as a result of a higher intake of ALA. It could be assumed that our slow-growing strains eat more forage ALA, partly contributing to such  $\Delta^9$ -desaturase reduction.

Poureslami et al. (2010) analyzed the effect of diet and age on saturated and monounsaturated fatty acid metabolism in broilers; they observed that both factors affected deposition, elongation,  $\Delta^9$  desaturation activity, and fatty acid oxidation.

More evident changes, however, are observed in  $\Delta^5$ plus  $\Delta^6$ -desaturase. This index represents a valid tool to estimate the ability of birds to synthesize long-chain fatty acids from precursors. Pure breeds and their crossbred counterparts exhibited lower concentrations of LA and ALA and higher long-chain family derivatives (Table 4). Together, with the highest  $\Delta^5$ - and  $\Delta^6$ desaturase index, this finding demonstrates a higher efficiency in long-chain fatty acid synthesis. It is known that a competition between the n-6 and n-3 fatty acids exists in which n-3 fatty acids are used as the preferred substrate in the desaturation and elongation pathway (Lands, 1992). The rate-limiting step in the enzymatic pathways of PUFA biosynthesis is thought to be  $\Delta^{6}$ desaturase (Yamazaki et al., 1992). The commonly accepted pathway for the synthesis of DHA consists of the elongation of EPA to 22:5n-3 followed by a  $\Delta^4$ desaturation (Cook, 1991).

Cherian and Sim (2001), investigating the effects of dietary ALA of laying hens on the fatty acid composition of liver microsomes and activity of  $\Delta^6$ -desaturase in hatched chicks, observed an increase in long-chain 20- and 22-carbon fatty acids in the liver, which may be attributed to the use of ALA as the preferred substrate over LA.

Our results are in accord with those of Sirri et al. (2010), who observed lower concentrations of LA and ALA and higher proportions of their long-chain family derivatives (C20:2n-6, C20:4n-6, EPA, C22:5n-3, and DHA) along with the highest  $\Delta^{5}/\Delta^{6}$ -desaturase index in slow-growing chickens which appeared more efficient than medium- and fast-growing birds in long-chain fatty acid synthesis. In the same study, PUFA gradually decreased in slow- to medium- and fast-growing birds: slow-growing chicken breast contained approximately 2- and 3-fold higher amounts of long-chain n-3 fatty acid and arachidonic acid, respectively, than that in medium- and fast-growing strains. This trend could be explained by the  $\Delta^5$ - plus  $\Delta^6$ -desaturase index, which was 54.0, 34.4, and 23.6 for slow-, medium-, and fastgrowing birds, respectively. The drumstick of slow-, medium-, and fast-growing birds showed the same tendency (36.3 vs. 21.4 vs. 15.2, respectively; data not shown).

The different lipid metabolisms and the higher efficiency of EPA and DHA deposition in slow-growing animals could be explained by considering the following facts:

- A specific gene determinism (FADS gene) has been reported to be involved in elaborating long chain n-3 and n-6 (Khang et al. 2007);
- The intake of pasture, that is, more forage, increases the intake of ALA, antioxidants, and phytoestrogens; in turn, it is reported that LNA modulates estrogen receptors (Holst-Schumacher et al., 2010) and the conversion to the longer chain n-3 PUFA. Phytoestrogens are mainly contained

in leguminous (Whitten and Patisaul, 2001), which were highly represented in the pasture.

- Fast-growing strains, selected for meat traits, had a different hormonal profile (Mao et al., 1998). In our case, the slow-growing strains used are eggtype lines, and laying hens seem to have a higher efficiency in EPA and DHA deposition respecting meat-type chickens, being that elongation is partly affected by the estrogen level (Alessandri et al., 2012).
- Slow-growing birds showed higher kinetic activities (walking, running, foraging, exploring, crouching at pasture) that produced different metabolism and fiber characteristics of muscle. Indeed, in our previous study (Branciari et al., 2009), we found that an organic system affected muscle fiber characteristics in L chickens with the presence of  $\alpha R$ fibers and an increased cross section area in pectoralis major; on the contrary, fiber characteristics, muscle enzyme functions, and lipid metabolism were not modified in fast-growing birds.

In conclusion, the results of this study indicate that the differences in the fatty acid content of chicken breast is affected by the breed; therefore, we demonstrated that in organic farming, chicken genotypes play an important role in the fatty acid composition of meat. This finding assumes great importance because health concerns over fat intake are one of the main factors contributing to the decline of meat intake.

The observed differences among poultry genotypes may be sufficient to be exploited through selection to reach a suitable compromise between rusticity and economic sustainability and design a healthier chicken meat for nutrition-conscious consumers, also matching the biodiversity goal of organic production. Further studies are needed to confirm our hypothesis through the direct measure of different enzymatic activity and gene expression of the above-mentioned complex in liver mitochondria.

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