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Lipid Assessment, Cholesterol and Fatty Acid Profile of meat from broilers raised in four different rearing systems

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Abstract: Evaluated lipid and cholesterol concentration and fatty acid profile of raw breast, thigh and drumstick meat from broilers raised in different rearing systems. Were used 200 male broiler carcasses from four different rearing systems (n=50 from conventional intensive; n=50 from organic; n=50 from free-range; and n=50 from antibiotic-free) distributed in a completely randomized design with four rearing systems and 50 replications (carcasses). Breast meat from conventional broilers showed higher lipid (1.47) and cholesterol (34.13) concentration. Thigh and drumstick meat from free-range broilers had higher lipid (7.53/4.73) and cholesterol (45.55/53.65) concentration. Fat contained in breast, thigh and drumstick meat from free-range broilers showed higher levels of polyunsaturated fatty acids. Fat from breast and thigh meat from free-range broilers showed higher total concentration of ω 3 and ω 6 fatty acids. Fat from thigh meat from organic broilers showed higher levels of EPA (C20:5n3) and DHA (C22:6n3). Fat from drumstick meat from free-range broilers showed higher total concentration of ω 3 and ω 6 fatty acids. Meat from chickens raised in alternative rearing systems offers less risk to cardiovascular health because it presents lower concentrations of lipids and cholesterol, greater amounts of polyunsaturated fatty acids, which are beneficial for human health.

Key words: breast, drumstick, free-range, organic, thigh.

INTRODUCTION

Brazil currently ranks second in the ranking of production and the first place among the world exporters of chicken meat (ABPA 2016) and one of the reasons for the success of the Brazilian poultry industry is the consumer's search for healthier meat products than the traditional red meat. Chicken breast meat can be considered important in diets because it contains a higher proportion of polyunsaturated fatty acids, when compared to meat of other species (Berzaghi et al. 2005, Riovanto et al. 2012).

The strategies used in aviculture, related to the animal breeding, cause the meat produced satisfy desirable nutritional characteristics, such as low fat content and relatively high concentrations of polyunsaturated fatty acids (Nkukwana et al. 2014) which have a known beneficial action in reducing the risk of cardiovascular diseases, hypertension, diabetes, inflammatory and immunological disorders (Zhou et al. 2012). Thus, fat intake through food became a risk factor due to the possible consequences (Leosdottir et al. 2005). The American Heart Association (2004) recommends that daily fat intake does not exceed 30% of

total calories and that saturated fat intake is not more than 10%.

Lipids are essential fats for normal growth and development of all animals. The functions of these fats range from source of energy, transport and absorption of vitamins, protection of organs, physical and thermal insulation, hormone precursors in the organism and they are extremely important in texture, flavor, palatability, color and preservation of food. Meat is an important source of lipids in the human diet and its consumers are increasingly interested in fat composition, since nutritional guidelines recommend reducing total fat intake, especially saturated fat, and increasing polyunsaturated fat (Sierra et al. 2008, De Marchi et al. 2012).

Although the genetic (Zanetti et al. 2010, Riovanto et al. 2012) influences the functional and nutritional meat properties (Sirri et al. 2011), the feed (Rymer & Givens 2005, Riovanto et al. 2012) and the rearing conditions may also influence fatty acid composition and, consequently, the meat oxidative stability (Jung et al. 2010, Nkukwana et al. 2014). And the fatty acid profile of broiler fat can be directly influenced by the lipid source used in animal diet rations (Husak et al. 2008).

Moreover, the variation in fatty acid composition affects the technological, nutritional and sensory meat properties (Wood et al. 2003, De Marchi et al. 2012, Riovanto et al. 2012), however, high PUFA contents, although beneficial to health, is often associated with the occurrence of oxidation and rancidity (Wood et al. 2003, De Marchi et al. 2012), which negatively affects the shelf life (Flåtten et al. 2005, De Marchi et al. 2012, Zhou et al. 2012) and makes it important the monitoring the fatty acid profile so that the industries can assign each type of meat to direct consumption or to other processes (Riovanto et al. 2012).

Thus, the aim of this study was to evaluate the lipid and cholesterol concentration, as well as the fatty acid profile of raw breast, thigh and drumstick meat from broilers raised in different rearing systems.

MATERIALS AND METHODS

This research was developed at the Laboratory of Animal Products Technology of the Faculty of Agricultural and Veterinary Sciences at UNESP, Jaboticabal Campus, São Paulo, Brazil (21°08' S, 48°11' W, 583 m altitude).

Samples and experimental procedures

To analyze lipids, cholesterol and fatty acids, were used 200 male broiler carcasses from four different rearing systems (n=50 from conventional intensive rearing system; n=50 from organic rearing system; n=50 from free-range rearing system; and n=50 from antibiotic-free rearing system), which were acquired from commercial slaughterhouses (São Paulo, SP, Brazil) inspected by the Brazilian Federal Inspection Service.

Cobb broilers raised in conventional rearing system were confined in closed sheds at a maximum density of 40 kg/m², and slaughtered at 42 d of age. In organic system, Cobb broilers received feed containing certified organic ingredients; from 25 d of age, birds were confined in closed sheds with access to a grazing area at a maximum density of 30 kg/m², grazing area of 100 m² per m² shed and were slaughtered at 48 d of age. In free-range rearing system, ISA Label broilers were raised in closed sheds with a maximum density of 30 kg/m² and, from 28 d of age, birds had access to paddocks with density of, at least, 3 m² of area available for each bird housed. Broilers raised in free-range rearing system were slaughtered at 85 d of age.

In antibiotic-free rearing system, Cobb broilers were raised in conventional closed sheds, with no access to grazing area, at a maximum density of 30 kg/m² and were slaughtered at 45 d of age. Broilers were fed a diet containing certified organic ingredients, devoid of animal ingredients and antibiotics, and were slaughtered at 48 days of age. Broilers were fed the diet shown in Table I.

In the slaughterhouse, 50 broiler carcasses, from four rearing systems studied, were collected following the flowchart of commercial slaughtering, meeting the requirements of animal welfare and regulations of the federal inspection service of the Ministry of Agriculture, Livestock and Supply. After the completion of *rigor mortis* (around four hours after slaughter), carcasses were divided into their main cuts (breast, n=50 for each rearing system; thigh, n=50 for each rearing system; drumstick n=50 for each rearing system).

Methods

Fat was performed in triplicate and analyzed according to the procedure 991.36 recommended by AOAC (2005). The total cholesterol was determined according to the methodology described by Bohac et al. (1988), adapted by Bragagnolo & Rodriguez-Amaya (1992), in which 10 g of ground raw sample was submitted to the extraction of lipids with chloroform and methanol (2:1). Thereafter, a 10 mL aliquot of the chloroform extract was evaporated with gaseous nitrogen and submitted to saponification with KOH 12% alcoholic solution. The unsaponifiable fraction (cholesterol) was extracted with n-hexane, purified and submitted to color reaction with acetic acid and sulfuric acid, using ferrous sulfate as catalyst. Absorbance was read on a spectrophotometer at 490 nm.

Fatty acids were isolated from samples according to the method proposed by Bligh & Dyer (1959), which extracts the lipid phase from

the sample. The fatty acid esterification was carried out according to the method proposed by Maia & Rodriguez-Amaya (1993) using a gas chromatograph (Shimadzu 14B, Shimadzu Corporation, Kyoto, Japan) equipped with a flame ionization detector and a fused silica capillary column (Omegawax 250); H₂ was used as the carrier gas. The identification of peaks was made by comparison with the retention times of standards with known composition.

Statistical analysis

Data were analyzed using a completely randomized design with four rearing systems (conventional, organic, antibiotic free and free-range rearing systems) and 50 replications (carcasses). Results of the breast, thigh and drumstick meat from each carcass were analyzed. Data were analyzed by One-way ANOVA using SAS (SAS Institute Inc 2002, 2003). All data were tested by analysis of variance and compared by Tukey's test with a significance level of P < 0.05.

RESULTS

Lipid profile of breast meat

Breast meat from broilers raised in conventional rearing system showed higher lipid concentration than meat from broilers raised in antibiotic-free rearing system (Table II). Breast meat from broilers raised in organic and free-range rearing systems showed the same amount of lipid as meat from broilers raised in conventional and antibiotic-free rearing systems. Breast meat from broilers raised in conventional and antibiotic-free rearing systems had higher cholesterol levels than breast meat from broilers raised in organic and free-range rearing systems.

The fat contained in breast meat from broilers raised in conventional system had the highest (P<0.001) levels of saturated fatty acids (SFA) and monounsaturated fatty acids

Table I. Composition and calculated nutrient content of diets (as-fed basis).

Ingredient (%)	Conventional broilers			Antibiotic-free, Free-range and Organic broilers		
	Starter	Growing	Finisher	Starter	Growing	Finisher
Corn grain	57.28	63.90	65.00	59.92	67.50	71.05
Soybean meal	36.9	30.30	27.90	36.10	27.75	23.20
Soybean oil	1.80	2.50	3.90	0.00	1.00	2.00
Dicalcium phosphate	1.80	1.60	1.40	1.90	1.70	1.50
Calcitic limestone	1.30	0.80	0.90	1.10	1.20	1.40
Salt	0.30	0.30	0.30	0.40	0.30	0.30
Vitamin and mineral mix*	0.50	0.50	0.50	0.50	0.50	0.50
DL-methionine (98%)	0.12	0.10	0.10	0.08	0.05	0.05
Calculated nutrient composition						
Crude protein (%)	21.28	18.80	17.80	21.11	17.90	16.25
ME (MJ/kg)	12.27	12.83	13.28	11.94	12.58	13.01
Available phosphorus (%)	0.45	0.40	0.36	0.47	0.42	0.37
Calcium (%)	1.08	0.78	0.77	0.98	0.96	0.97
Total M+C (%)	0.78	0.70	0.67	0.74	0.63	0.58
Total methionine (%)	0.43	0.38	0.37	0.39	0.32	0.30
Total Lysine (%)	1.16	0.99	0.93	1.15	0.93	0.82

ME: Metabolizable energy; M+C: methionine + cysteine; Dry matter of the as-fed diets: 90%; *Product supply per kg of feed – A vitamin – 9.300 UI, D3 vitamin – 2.300 UI, E vitamin – 30 mg, K vitamin – 1.5 mg, B2 vitamin – 5.5 mg, B6 vitamin – 3.0 mg, B12 vitamin – 0.010 mg, biotin – 0.07 mg, pantothenic acid – 11 mg, folic acid – 0.77 mg, Fe – 55 mg, Cu – 11 mg, I – 1 mg, growth promoter – 20 mg, coccidiostatic – 50 mg, BHT – 100 mg, inert vehicle – 1.000 g. Dry matter of the as-fed diets: 90%. Not added to the diet provided to antibiotic-free, free-range and organic broilers.

(MUFA) and the lowest ($P < 0.001$) levels of polyunsaturated fatty acids (PUFA). Meat from broilers raised in free-range and antibiotic-free rearing systems showed higher ($P < 0.001$) levels of PUFA (36.38%) than meat from broilers raised in conventional (18.33%) and organic (29.88%) rearing systems. In the same way, breast meat from free-range and antibiotic-free broilers showed higher PUFA/SFA ratio than the breast meat from broilers raised in conventional and organic rearing systems.

There were no differences ($P > 0.05$) regarding to C10:0 and C20:0 fatty acids concentration in fat from breast meat from broilers raised in different rearing systems. Fat contained in breast meat from broilers raised in conventional rearing system showed higher values for almost all fatty acids, with exception for C24:1n9. Among PUFA, fat from conventional broilers had lower concentration than the others, mainly EPA (C20:5n3) and DHA (C22:6n3). Higher levels of PUFA were verified in fat from breast meat from broilers raised in alternative systems. There were no differences ($P > 0.05$) between analyzed groups regarding to C18:2c9,t11 concentration. Breast meat from organic broilers showed higher levels of C18:3n6, C20:2, C20:3n6, C20:4n6, C20:5n3, C22:4n6 and C22:6n3 fatty acids, highlighting organic system among all studied rearing systems, with respect to the concentration of polyunsaturated fatty acids.

The fat contained in breast meat from broilers raised in free-range and antibiotic-free rearing systems showed higher total concentration of $\omega 3$ (2.59%) and $\omega 6$ (33.48%) fatty acids than the others. Fat from breast meat from conventional broilers showed higher $\omega 6/\omega 3$ ratio (~36:1) than fat from breast meat from broilers raised in alternative rearing systems (~14:1, on average).

Lipid profile of thigh meat

Thigh meat from broilers raised in organic and free-range rearing systems showed higher ($P < 0.0001$) lipid concentration (7.05%, on average) than meat from broilers raised in conventional (4.82%) and antibiotic-free (3.30%) rearing systems (Table III). Thigh meat from broilers raised in free-range rearing system had higher ($P < 0.0001$) cholesterol levels (45.55 mg/100g) than thigh meat from broilers raised in conventional (35.33 mg/100g), organic (39.58 mg/100g) and antibiotic-free (33.59 mg/100g) rearing systems.

As the fat contained in breast meat, the fat contained in thigh meat from broilers raised in conventional system had the highest ($P < 0.001$) levels of saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) and the lowest ($P < 0.001$) levels of polyunsaturated fatty acids (PUFA). Fat contained in thigh meat from broilers raised in alternative rearing systems showed lower ($P = 0.0003$) levels of saturated fatty acids. Thigh meat from broilers raised in free-range rearing system showed higher ($P < 0.001$) levels of PUFA (34.48%) than thigh meat from broilers raised in conventional (21.25%), organic (26.70%) and antibiotic-free (26.51%) rearing systems. Thigh meat from free-range broilers showed higher PUFA/SFA ratio (1.18) than the thigh meat from broilers raised in conventional (0.58), organic (0.83) and antibiotic-free (0.78) rearing systems. There was no difference ($P > 0.05$) only for the C10:0 fatty acid concentration between studied groups. Fat contained in thigh meat from organic broilers showed higher ($P < 0.001$) levels of EPA and DHA. In this study no EPA levels were detected in thigh meat from conventional broilers.

The fat contained in thigh meat from broilers raised in free-range rearing system showed higher total concentration of $\omega 3$ (2.33%) fatty acids than the others. The fat contained in thigh

Table II. Lipids (%) and cholesterol (mg/100g) of breast meat and fatty acid composition (% of total fatty acids) of fat from breast meat from broilers raised in different rearing systems.

	Conventional (n=50)	Organic (n=50)	Free-range (n=50)	Antibiotic-free (n=50)	P-value
Lipids [‡]	1.47 ^A	1.09 ^{AB}	1.11 ^{AB}	0.93 ^B	0.0400
Cholesterol	34.13 ^A	23.51 ^C	30.78 ^B	36.78 ^A	<0.0001
Total SFA	38.47 ^A	33.59 ^B	29.20 ^C	29.20 ^C	<0.001
Total MUFA	43.21 ^A	36.63 ^B	33.77 ^B	33.77 ^B	<0.001
Total PUFA	18.33 ^C	29.88 ^B	36.38 ^A	36.38 ^A	<0.001
MUFA/SFA	1.12	1.09	1.16	1.16	
PUFA/SFA	0.48	0.89	1.25	1.25	
C10:0	0.01	0.01	0.01	0.01	0.4363
C12:0	0.04 ^A	0.02 ^B	0.02 ^B	0.02 ^B	<0.001
C14:0	0.75 ^A	0.53 ^B	0.48 ^C	0.48 ^C	<0.001
C15:0	0.13 ^A	0.10 ^B	0.08 ^B	0.08 ^B	<0.001
C16:0	27.77 ^A	23.41 ^B	21.99 ^C	21.99 ^C	<0.001
C17:0	0.20 ^A	0.15 ^B	0.14 ^B	0.14 ^B	<0.001
C18:0	9.49 ^A	9.30 ^A	6.40 ^B	6.40 ^B	<0.001
C20:0	0.08	0.08	0.08	0.08	0.8436
C14:1	0.16 ^A	0.09 ^B	0.10 ^B	0.10 ^B	0.0002
C16:1	5.18 ^A	3.00 ^C	3.38 ^B	3.38 ^B	<0.001
C17:1	0.27 ^A	0.15 ^B	0.16 ^B	0.16 ^B	0.0026
C18:1n9c	34.52 ^A	30.21 ^B	27.99 ^B	27.99 ^B	<0.001
C18:1n7	2.61 ^A	2.14 ^B	1.59 ^C	1.59 ^C	<0.001
C20:1n9	0.37 ^A	0.26 ^B	0.20 ^C	0.20 ^C	<0.001
C24:1n9	0.09 ^C	0.76 ^A	0.33 ^B	0.33 ^B	<0.001
C18:2n6c	15.19 ^C	20.60 ^B	30.09 ^A	30.09 ^A	<0.001
C18:3n6	0.05 ^C	0.21 ^A	0.18 ^B	0.18 ^B	<0.001
C18:3n3	0.40 ^C	0.96 ^B	2.23 ^A	2.23 ^A	<0.001
C18:2c9,t11	0.06	0.05	0.05	0.05	0.5048
C20:2	0.39 ^B	0.47 ^A	0.25 ^C	0.25 ^C	<0.001
C20:3n6	0.41 ^B	0.84 ^A	0.35 ^B	0.35 ^B	<0.001
C20:4n6	1.24 ^C	4.72 ^A	2.37 ^B	2.37 ^B	<0.001
C20:3n3	0.02 ^B	0.03 ^{AB}	0.04 ^A	0.04 ^A	0.0030
C20:5n3 (EPA)	0.01 ^C	0.20 ^A	0.04 ^B	0.04 ^B	<0.001
C22:4n6	0.50 ^B	1.39 ^A	0.49 ^B	0.49 ^B	<0.001
C22:6n3 (DHA)	0.05 ^C	0.41 ^A	0.28 ^B	0.28 ^B	<0.001
Total ω3	0.48	1.60	2.59	2.59	
Total ω6	17.39	27.76	33.48	33.48	
ω6/ω3	36.23	17.35	12.93	12.93	

^{A,B,C}Mean values, in the same row, followed by different letters are significantly different by Tukey's test ($P < 0.05$). [‡]: results expressed on a fresh matter basis; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid.

meat from broilers raised in free-range rearing systems showed higher total concentration of $\omega 6$ (31.90%) than conventional (19.95%), organic (25.02%) and antibiotic-free (24.80%) rearing systems. Fat from thigh meat from free-range broilers showed lower $\omega 6/\omega 3$ ratio (~14:1) than fat from thigh meat from broilers raised in other studied rearing systems (~17:1).

Lipid profile of drumstick meat

Drumstick meat from broilers raised in free-range rearing system showed higher ($P=0.0003$) lipid concentration (4.73%) than meat from broilers raised in other rearing system (3.45%, on average) (Table IV). As for thigh meat, drumstick meat from broilers raised in free-range rearing system had higher ($P<0.0001$) cholesterol levels (53.65 mg/100g) than drumstick meat from broilers raised in conventional (37.38 mg/100g), organic (46.89 mg/100g) and antibiotic-free (49.02 mg/100g) rearing systems.

Unlike what was observed for breast and thigh meat, the highest ($P<0.001$) level of saturated fatty acids (39.68%) was observed in fat from drumstick meat from antibiotic-free broilers, while fat from drumstick meat from free-range broilers showed the lowest level of SFA (27.69%) between evaluated groups. The fat contained in drumstick meat from free-range broilers showed higher PUFA concentration (37.94%) than fat contained in drumstick meat from conventional (14.33%), organic (22.78%) and antibiotic-free (26.08%) broilers.

Regarding to fatty acid composition of fat from drumstick meat from broilers raised in different rearing systems, it was detected significant difference for all fatty acids tested, wherein the highest levels of EPA were verified in drumstick meat from organic and free-range broilers and, the highest level of DHA was verified in drumstick meat from free-range broilers. The fat contained in drumstick meat

from broilers raised in free-range rearing system showed higher total concentration of $\omega 3$ (2.82%) and $\omega 6$ (34.75%) fatty acids than the others. Fat contained in drumstick meat from free-range and antibiotic-free broilers showed lower $\omega 6/\omega 3$ ratio (~12:1) than fat from drumstick meat from broilers raised in other studied rearing systems (~26:1, on average).

DISCUSSION

Lipid content

The consumer's preference is influenced by the variation of the quality of chicken meat available in the market and may be affected by factors such as bird feeding, strain, age, gender and slaughter processing (Rodrigues et al. 2008). Some studies (Castellini et al. 2006, Lonergan et al. 2003, Faria et al. 2009) have reported that broilers of fast-growing strains tend to have a higher concentration of fat in breast meat than chickens from slow-growing or crossbred strains; and Husak et al. (2008), observed a lower amount of fat in chicken meat from broilers raised in organic rearing system than in meat from conventional broilers, possibly due to the greater locomotion and other activities due to grazing access.

Animals from the same lineage may exhibit differentiated fat deposition (Faria et al. 2009), depending on the slaughter age, since increasing age results in greater fat deposition. In this study, it was observed that the breast meat from Cobb broilers raised in a conventional system and slaughtered at 42 days of age, had higher ($P = 0.0400$) lipid concentration (1.47) than breast meat from Cobb broilers raised in antibiotic-free (0.93) system and slaughtered at 45 days of age, suggesting that the breeding system, besides the slaughter age, influences the carcass fat deposition. The chicken meat shows variable fat concentration, depending on the evaluated

Table III. Lipids (%) and cholesterol (mg/100g) of thigh meat and fatty acid composition (% of total fatty acids) of fat from thigh meat from broilers raised in different rearing systems.

	Conventional (n=50)	Organic (n=50)	Free-range (n=50)	Antibiotic-free (n=50)	P-value
Lipids [‡]	4.82 ^B	6.57 ^A	7.53 ^A	3.30 ^C	<0.0001
Cholesterol	35.33 ^C	39.58 ^B	45.55 ^A	33.59 ^C	<0.0001
Total SFA	36.39 ^A	32.19 ^B	29.26 ^B	34.11 ^B	0.0003
Total MUFA	42.81 ^A	41.65 ^B	35.71 ^C	43.55 ^B	0.0003
Total PUFA	21.25 ^C	26.70 ^B	34.48 ^A	26.51 ^B	0.0004
MUFA/SFA	1.18	1.29	1.22	1.28	
PUFA/SFA	0.58	0.83	1.18	0.78	
C10:0	0.01	0.01	0.01	0.01	0.3272
C12:0	0.035 ^A	0.022 ^B	0.030 ^{AB}	0.025 ^{AB}	0.0381
C14:0	0.78 ^A	0.56 ^B	0.57 ^B	0.57 ^B	0.0003
C15:0	0.14 ^A	0.07 ^B	0.09 ^B	0.09 ^B	0.0003
C16:0	27.48 ^A	24.00 ^B	22.23 ^C	24.43 ^B	<0.001
C17:0	0.21 ^A	0.11 ^B	0.15 ^B	0.13 ^B	0.0013
C18:0	7.66 ^{AB}	7.33 ^{AB}	6.08 ^B	8.73 ^A	0.0051
C20:0	0.08 ^B	0.09 ^B	0.10 ^{AB}	0.13 ^A	0.0067
C14:1	0.18 ^A	0.12 ^B	0.11 ^B	0.12 ^B	0.0079
C16:1	6.11 ^A	4.33 ^{AB}	3.35 ^B	4.81 ^{AB}	0.0116
C17:1	0.16 ^A	0.08 ^B	0.08 ^B	0.07 ^C	<0.001
C18:1n9c	34.04 ^A	35.06 ^A	30.39 ^B	36.23 ^A	<0.001
C18:1n7	1.93 ^A	1.64 ^{AB}	1.50 ^B	1.75 ^{AB}	0.0332
C20:1n9	0.37 ^{AB}	0.27 ^{BC}	0.24 ^C	0.39 ^A	0.0022
C24:1n9	0.02 ^B	0.15 ^A	0.04 ^B	0.18 ^A	<0.001
C18:2n6c	19.73 ^C	22.93 ^B	31.02 ^A	22.76 ^B	<0.001
C18:3n6	0.04 ^C	0.22 ^A	0.17 ^B	0.22 ^A	<0.001
C18:3n3	1.17 ^C	1.35 ^B	2.25 ^A	1.42 ^B	<0.001
C18:2c9,t11	0.04 ^{AB}	0.04 ^A	0.04 ^{AB}	0.02 ^B	0.0132
C20:2	0.07 ^C	0.16 ^B	0.21 ^A	0.19 ^{AB}	<0.001
C20:3n6	0.04 ^C	0.28 ^A	0.15 ^B	0.23 ^A	<0.001
C20:4n6	0.11 ^C	1.24 ^B	0.43 ^B	1.27 ^A	<0.001
C20:3n3	0.01 ^B	0.01 ^B	0.02 ^A	0.01 ^B	<0.001
C20:5n3 (EPA)	---	0.04 ^A	0.03 ^{AB}	0.02 ^B	<0.001
C22:4n6	0.03 ^C	0.35 ^A	0.13 ^B	0.32 ^A	<0.001
C22:6n3 (DHA)	0.01 ^D	0.08 ^A	0.03 ^C	0.05 ^B	<0.001
Total ω3	1.19	1.48	2.33	1.50	
Total ω6	19.95	25.02	31.90	24.80	
ω6/ω3	16.76	16.90	13.69	16.53	

^{A,B,C,D}Mean values, in the same row, followed by different letters are significantly different by Tukey's test ($P < 0.05$). [‡]: results expressed on a fresh matter basis; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid.

cut (Givens et al. 2011, Dalziel et al. 2015) and, differently that was reported in the literature, in this study was found a greater amount of fat in thigh (6.57/7.53) and drumstick (3.51/4.73) meat from broilers raised with grazing access (organic and / or free-range) than in meat from confined broilers.

Cholesterol content

In contrast to that observed for breast meat, wherein was found higher ($P < 0.0001$) cholesterol concentration in breast meat from confined broilers, thigh and drumstick meat from free-range broilers showed higher ($P < 0.0001$) cholesterol concentration than thigh and drumstick meat from conventional, organic and antibiotic-free broilers.

Cholesterol is an important molecule of the membranes structure and is a precursor of steroid hormones, vitamin D and bile acids; it can be obtained directly from the diet or synthesized by biosynthesis *de novo*, allowing cholesterol production required to meet the needs of the large variety of biological processes which is involved (Ponte et al. 2008). The total cholesterol levels in meat plays an important biological role because they are closely related to cardiac diseases (Salma et al. 2007), such as obstruction of the coronary veins caused by the fatty material accumulation (cholesterol, calcium, blood cells, and the cells from the arterial wall), and may evolve over time, reducing the blood flow and also making the individual more susceptible to thrombus or clots (Chou & Friedman 2016). The compromising of normal heart functioning can affect consumer acceptance of some kinds of meat and, therefore, there has been increasing interest in recent years in modulating of the cholesterol concentration and fatty acid composition in chicken meat products (Sacks 2002, Salma et al. 2007).

Ponte et al. (2008) obtained results of cholesterol levels in breast meat from free-range chickens equal to 48 mg/100g, a lower concentration than that found in meat from conventional chickens (56 mg/100g). The results obtained in the present study are below than results published in the literature (47.11 mg/100g, Ponte et al. 2004, 66.79 mg/100g, Rosa et al. 2006, 45.96 mg/100g, Oliveira e Vieira et al. 2007, 93.6 mg/100g, Salma et al. 2007) for cholesterol levels in chicken breast meat; also are below the range of variation described by Bragagnolo & Rodriguez-Amaya (1992) (between 48 and 79 mg/100g), and below the average total cholesterol content considered by Ponte et al. (2008) (50 mg/100 g); the reasons for such low results are still unknown.

The cholesterol concentration in chicken meat may be influenced by the diet composition, by the bird's age and by the gender (Wang et al. 2005). Pastures consumed by broilers raised in some alternative rearing systems, such as organic and free-range, are a good source of tocopherols and tocotrienols, the latter being known to contribute to the reduction of cholesterol levels in blood plasma; however, the effects of pasture intake on cholesterol levels in free-range chicken meat is still unknown (Ponte et al. 2008).

Fatty acids profile

It is observed that, in general, the fat present in meat from broilers raised in alternative systems (mainly organic and free-range) had lower ($P < 0.05$) saturated (SFA) and monounsaturated (MUFA) fatty acid concentration, and higher concentration of polyunsaturated fatty acids (PUFA). Meat from alternative broilers presented higher PUFA/SFA ratio. Lower PUFA in relation to SFA levels may be risk factors for cardiovascular diseases, which are among the main causes of mortality in some countries (Hu et al. 2001,

Table IV. Lipids (%) and cholesterol (mg/100g) of drumstick meat and fatty acid composition (% of total fatty acids) of fat from drumstick meat from broilers raised in different rearing systems.

	Conventional (n=50)	Organic (n=50)	Free-range (n=50)	Antibiotic-free (n=50)	P-value
Lipids [‡]	3.30 ^B	3.51 ^B	4.73 ^A	3.55 ^B	0.0003
Cholesterol	37.38 ^C	46.89 ^B	53.65 ^A	49.02 ^B	<0.0001
Total SFA	37.53 ^B	34.79 ^B	27.69 ^C	39.68 ^A	<0.001
Total MUFA	50.85 ^A	42.81 ^A	33.34 ^B	49.52 ^A	<0.001
Total PUFA	14.33 ^C	22.78 ^B	37.94 ^A	26.08 ^B	<0.001
MUFA/SFA	1.35	1.23	1.20	1.25	
PUFA/SFA	0.38	0.53	1.37	0.66	
C10:0	0.01 ^B	0.04 ^A	0.01 ^B	0.01 ^B	<0.001
C12:0	0.04 ^B	0.05 ^A	0.02 ^C	0.03 ^{BC}	<0.001
C14:0	0.77 ^A	0.69 ^B	0.52 ^D	0.61 ^C	<0.001
C15:0	0.14 ^A	0.08 ^B	0.08 ^B	0.10 ^B	0.001
C16:0	27.98 ^B	25.04 ^C	20.54 ^D	29.17 ^A	<0.001
C17:0	0.19 ^A	0.12 ^B	0.14 ^B	0.13 ^B	0.001
C18:0	8.32 ^B	8.69 ^B	6.30 ^C	9.52 ^A	<0.001
C20:0	0.08 ^B	0.08 ^B	0.08 ^B	0.11 ^A	0.005
C14:1	0.21 ^A	0.15 ^B	0.11 ^B	0.14 ^B	0.001
C16:1	6.65 ^A	5.67 ^A	3.80 ^B	5.80 ^A	0.001
C17:1	0.23 ^A	0.09 ^B	0.11 ^B	0.12 ^B	<0.001
C18:1n9c	41.19 ^A	34.71 ^B	27.48 ^C	40.89 ^A	<0.001
C18:1n7	2.19 ^A	1.83 ^B	1.55 ^C	2.16 ^A	<0.001
C20:1n9	0.34 ^A	0.28 ^B	0.21 ^C	0.29 ^{AB}	0.0003
C24:1n9	0.04 ^B	0.08 ^B	0.08 ^B	0.12 ^A	0.0008
C18:2n6c	12.76 ^C	19.79 ^B	32.31 ^A	22.16 ^B	<0.001
C18:3n6	0.07 ^C	0.20 ^{AB}	0.16 ^{AB}	0.22 ^A	<0.001
C18:3n3	0.39 ^D	0.97 ^C	2.55 ^A	1.88 ^B	<0.001
C18:2c9,t11	0.05 ^A	0.05 ^A	0.03 ^B	0.04 ^{AB}	0.009
C20:2	0.15 ^C	0.18 ^{BC}	0.34 ^A	0.26 ^{AB}	0.0005
C20:3n6	0.12 ^B	0.33 ^A	0.30 ^A	0.28 ^A	0.0002
C20:4n6	0.59 ^C	0.89 ^B	1.50 ^A	0.89 ^B	<0.001
C20:3n3	0.01 ^B	0.01 ^B	0.03 ^A	0.01 ^B	0.015
C20:5n3 (EPA)	0.01 ^B	0.06 ^A	0.06 ^A	0.01 ^B	<0.001
C22:4n6	0.15 ^C	0.28 ^B	0.48 ^A	0.30 ^B	<0.001
C22:6n3 (DHA)	0.03 ^B	0.02 ^B	0.18 ^A	0.03 ^B	<0.001
Total ω3	0.44	1.06	2.82	1.93	
Total ω6	13.69	21.49	34.75	23.85	
ω6/ω3	31.11	20.27	12.32	12.36	

^{A,B,C,D}Mean values, in the same row, followed by different letters are significantly different by Tukey's test ($P < 0.05$). [‡]: results expressed on a fresh matter basis; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid.

Ganji et al. 2003, Ponte et al. 2008). Saturated fatty acids would be related to increased levels of total cholesterol, LDL and triglycerides, whereas monounsaturated fatty acids would be related to their reduction, and may promote the increase of blood levels of HDL (Hautrive et al. 2012) and benefit the body together with the polyunsaturated fats (Cuppari 2005, Mahan & Escott-Stump 2005, Hautrive et al. 2012). Data from the literature indicate that the PUFA/SFA ratio below 0.45, quite close to that found in this study for conventional chicken meat, may be considered unhealthy (Wood & Enser 1997, Hautrive et al. 2012). Based on this, it is suggested that meat from alternative chickens offers less risk to cardiovascular health because it presents a greater quantity of PUFA in relation to the amount of SFA.

Alternative rearing systems, in general, provided higher omega-3 and omega-6 concentrations to chicken meat. Conventional chicken meat showed higher n-6/n-3 ratio in breast and drumstick meat; such imbalance in the n-6/n-3 ratio may be responsible for the occurrence of cardiovascular, inflammatory and autoimmune diseases (Simopoulos 2004, Ponte et al. 2008). Data from the literature indicate that the proportion of n-6/n-3 fatty acids should be 5:1 to 10:1 (Hautrive et al. 2012), values quite different to those found in this study for chickens from all rearing systems studied. Some authors suggest that organically grown birds could use n-3 fatty acids as an essential nutrient for their own immune system's action against external agents rather than depositing it in the meat (Küçükyılmaz et al. 2012, Sales 2014) which could justify the results found in this research.

The chicken meat is considered one of the main sources of omega-3 fatty acids (Howe et al. 2006, Sioen et al. 2006, Ponte et al. 2008). In general, broiler meat raised in alternative rearing systems showed the highest levels of EPA

and DHA, which are important for the health of the retina, for phospholipids of the membranes of the brain and also help in reducing the risks of heart disease (Rymer & Givens 2005, Ponte et al. 2008). However, because the pasture is a poor source of EPA and DHA, it is still unknown whether broilers are able to utilize α -linoleic acid (18:3n-3) from the pasture as a precursor to the synthesis and deposition of these fatty acid in the meat (Ponte et al. 2008), and it is unknown if the higher levels of EPA and DHA present in meat from alternative chickens are influenced by the pasture.

CONCLUSION

Meat from chickens raised in alternative rearing systems offers less risk to cardiovascular health because it presents lower concentrations of lipids and cholesterol, and greater amounts of polyunsaturated fatty acids, which are beneficial for human health, once they have anti-inflammatory action, promotes heart health, blood circulation and improves cognitive processes.

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