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Original article

The effects of cooking on protein content and nutritional composition of fatty acid of broilers meat fed on green oak acorn (*Quercus ilex*)

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ABSTRACT

The aim of this work was to analyse the effects of cooking on proteins and nutritional composition of fatty acid of broilers fed on green oak acorn (GOA). The broilers were divided into two groups and they were given either experimental diet (50% corn and 50% of GOA) or control diet (C) (67% corn). At 56 days, twenty broilers from each group were slaughtered and their breast meat was prepared. Proteins and fatty acid (FA) composition of the muscle were determined. Dietary GOA affected fatty acid (FA) profile of breast muscle by significantly increasing polyunsaturated fatty acids (PUFA) and reducing saturated fatty acids (SFA) as compared with controls. After cooking, the lipids are in larger amounts in the muscle of control chickens compared to those fed on GOA. The results from this study showed that supplemented diet with GOA increased PUFA content of the meat.

1. Introduction

There is interest in the foods containing higher levels of polyunsaturated fatty acids (PUFA) because of their beneficial effects on human health, mainly in the prevention of cardiovascular disease (Cortinas et al., 2005). These fatty acids are bringing in food from plant fats and animal products containing a significant amount of PUFAs such as poultry meat (Mossab, 2001; Lessire, 2001). Previous reports suggest that in both birds and mammals,

PUFAs reduce lipid synthesis (Wilson et al., 1986, Sanz et al., 2000) and increase fatty acid oxidation (Sanz et al., 2000; Madsen et al., 1999), while saturated and monounsaturated fatty acids (MUFA) have no inhibitory effects on body fat deposition (Blake and Clarke, 1999). Diet composition can modulate the deposition of fatty acids because there is a direct relationship between the nature of dietary fat and fatty acids deposited in the meat (Kouba and Mourot, 2011). Among the diets used in food animals as a potentially rich source of n-3 polyunsaturated fatty acids (n-3 PUFAs) is acorn (Bouderoua et Selselet-Attou, 2003; Bouderoua et al., 2009). The acorn, or oak nut is usually contains a single seed (rarely two seeds) enclosed in a tough, leathery shell, and borne in a cup-shaped chapel. Acorns vary from 1–6 cm long and 0.8–4 cm broad. Acorns take between about 6 and 24 months (depending on the species) to mature. These products are used dried or boiled by the local neighbouring population's for fattening sheep and goats. The interests in using them are their high starch content. Some studies suggest that it is possible to use the acorn in pig feed without growth affecting performance and slaughter (Rey et al., 2005). Similarly, Meuret (1997) reports that the leaves of oaks are used and offered fresh to goats grazing on rangeland.

In North Africa, for instance, previous works have shown the possibility to use the green oak acorns (GOA) in the diet of broilers without altering growth performance and body composition (Bouderoua and Selselet-Attou, 2004; Bouderoua et al., 2009). These same authors reported that the meat of the sartorius muscle of broilers is characterized by a low-fat, low content in saturated fatty acids (SFA) and high in polyunsaturated fatty acids (PUFA) such as linoleic and linolenic acids recommended by nutritionists. However, the differences in fatty acid composition between raw and cooked meat of chicken were not approached. Poultry meat is mainly consumed after cooking, it is important to know the effect of cooking on the lipid fatty acids. Indeed, cooking seems to have an influence on the degree of unsaturated lipids that affects the sensory quality of chicken muscle (Sirbu et al., 1974; Veen and Strappers, 1975; Rabot, 1998; Kondjoyan and Peyron, 2006).

So, the objective of the present study was to determine the effects of a diet containing GOA on fatty acid composition of cooked chicken breast meat.

2. Materials and methods

2.1. Animals and experimental procedures

One-week male broiler chicks (*Gallus*) were reared conventionally for the first 10 days. During this pre-experimental period all birds received a standard starter diet (22% crude protein (CP) and 3100 kcal metabolizable energy/kg). Water was provided *ad libitum* throughout the experiment. In day 10 , 200 chickens were divided into two groups as control (C) and a test group (GOA) each having 100 birds. Birds were fed test diets until the end of the experiment (56 days). Corn-soybean (corn 65%, soybean meal 27%) based control diet was used in the present study. In control diet, GOA replaced a corn at 50%. The diets characteristics are presented in table 1. The green oak acorn *Quercus ilex* was harvested in the Algeria west forest in Chlef (latitude: 36° 13'N; longitude: 1° 20'E). Before formulating diets, the acorn is spread and dried at room temperature in a covered and well ventilated place

On day 56 of rearing, 20 chickens from each group were weighed and then slaughtered, scalded and then eviscerated. After chilling for 18h at 4°C, carcasses were fused longitudinally. Half of each carcass is then cut into two parts, one of them is used for tissue analysis as raw and the other after cooking. The muscles were stripped of skin and any visible trace of fat was removed. The cooking method chosen in our test was oven roasted, very widely used for the culinary preparation of whole chickens. It is roasted in a domestic oven.

The meat without ingredient is roasted during a proportional time in the weight (the 01 hours of cooking for 1kg of carcass in 180°C). The cut was performed as soon as cooked meat was cooled. The samples of Pectoralis major muscle (raw and cooked) were removed and stored at -20°C. The crude protein, polysaccharide fractions (starch and cellulose), ash and crude fat content of the diets were determined following the methodologies described in the AFNOR methods (1985). Phenolic compounds of the diets were determined using a spectrophotometric method Folin Ciocalteu (Scalbert et al., 1988).

2.2. Analysis of meat

Biochemical parameters selected for measurement of raw and cooked meat (*Pectoralis major*) are those necessary for understanding the changes undergone by the muscles during cooking. So we measured the dry matter, content water and mineral (AFNOR, 1985). The protein assay was developed using the method of Lowry et al. (1951). The lipids were extracted by cold chloroform, methanol (2:1) according to the method of Folch et al.

(1957). The fatty acid profile was determined by gas chromatograph after derivation to trifliorure boron (BF 3) method of Morrison and Smith (1964). The column is a capillary column 30m length and 0.25mm in diameter. The statitionary phase consists of biscianoprophyl and cyanopropylphenyl and the carrier gas is hydrogen. The temperatures of the injector and detector were respectively 220 and 280°C. Fatty acids are expressed as percentage of identifying fatty acids and total amount calculated using an internal standard (C17:0).

Table 1
Basal and experimental diet of broilers.

Item	Control	Green Oak Acorn
Ingredients (%)		
Corn	64,8	32,4
Green Oak Acorn (GOA)	-	32,4
Soya bean meal	27,0	270
Weat bran	5	5
Calcium	1,2	1,2
Phosphorus	1	1
Min-Vit premix1	1	1
Analysed composition (%)		
Starch	40,0	40,9
Cellulose	2,8	3,1
Crud Protein	21,0	19,1
Lipids	29	39
Ash	55	55
Total phenols (gallic acid equivalents milligram/gram)	ND	1
Calculated nutrient content		
ME (Kcal/Kg)	3616	3625
FA Analysis (% of identified FA)		
C14:0	0.1	0.2
C16:0	12.5	13.4
C16:1 (n7)	0.3	0.5
C18:0	3.1	2.7
C18:1(n9)	50.1	25.2
C18:2(n6)	31.0	55.1
C20:0	0.8	0.5
C18:3(n3)	1.6	2.4
C20:1 (n9)	0.6	0.0
SFA	16.2	16.9
MUFA	51.1	25.7
PUFA	32.7	57.7
n-6	31.0	55.1
n-3	1.7	2.4
n-6/n-3	18.7	23.0

Min-Vit premix1:vitamin E (d-alpha-tocophérol): 8.96UI; vitamin K3 (ménadione): 800µg; vitamin B1 (thiamine): 1mg; vitamin B2 (riboflavine): 3mg; Pantothenate of Ca: 6mg; vitamin B6 (pyridoxine): 1.5; vitamin B12 (cyanocobalamine): 6µg; folic acid: 200µg; nicotinic acid: 12mg; copper: 5mg; cobalt: 0.65mg; manganese: 65mg; zinc: 65mg; selenium: 250µg, iron: 50mg; iode: 800µg; magnesium: 100mg. ME=Metabolisable energy, ND=Not determined, FA=Fatty acids, SFA=Saturated fatty acids, MUFA=Monounsaturated fatty acids, PUFA=Polyunsaturated fatty acids, UI=Unity international, µg= microgramme, mg=milligramme.

2.3. Statistical analysis

Values are presented as means±SD. The data were processed using the Statview Software program, version 5 (SAS Institute, Cary, NC, USA). The homogeneity of variance between groups was tested by Bartlett's. The effects of treatments (GOA or C diets) and cooking on broilers breast meat characteristics are tested by ANOVA. The means were compared with Fisher's least significant difference test in the case of a significant effect and P<0.05 was considered statistically significant.

3. Results

3.1. Effect of cooking and diet on the nutritional composition of «Pectoralis major» muscle

3.1.1. Composition of meat

Whatever the nature of diet, cooking meat to an internal temperature of 180°C, significantly increased dry matter (DM) up to 39%, P<0.0001 (Table 2). In raw meat chickens, breast fillet water content of control group was slightly lower compared to GOA group 72.5% vs.72.9%, respectively. Similar results were observed for cooking meat of control and GOA chickens (62.5% *vs*. 61.7%), respectively (Table 2). No significant treatment effect was observed on ash contents. While a slight increase was recorded following cooking as compared to raw meat, i.e. (1.4% *vs*. 1.5%) for control and (1.4% *vs*. 1.5%) for GOA chickens. Whatever the treatment of meat (raw and cooked), the levels of protein were significantly lower in GOA group compared to control chickens by 15% (*P<0.0001*) (Table 2).

Table 2
Effect of cooking and the nature of diet on the chemical composition of (Pectoralis major) muscle (Expressed in g. 100g-1 of fresh tissue).

	Control group		GOA group		*P* values	
	Raw	Cooked	Raw	Cooked	Diet	cooking
Moisture	72.5±0.2b	61.7±1.5c	72.9±0.6a	62.5±1.5b	<0.01	<0.0001
Dry matter	27.5±0.2b	38.3±1.5a	27.1±0.6b	37.5±1.4a	NS	<0.0001
Ash	1.4±0.1	1.5±0.1	1.4±0.1	1.5±0.1	NS	NS
Protein	22.0±0.9a	21.2±0.3a	18.7±0.7b	18.0±0.8b	<0.0001	NS

For each group n=3. Data are expressed as mean and standard deviation
NS: Not significant
The values in the same line with different superscript letter are significantly different at 5%.

3.1.2. Fatty acid composition of breast fillet (Pectoralis major)

The lipid content and fatty acid compositions are presented in table 3. Whatever the diet, lipid proportion was significantly greater in cooked fillet as compared to raw fillet (1.2% vs. 2.1%, P<0.0001) (Table 3). In addition to that, the contents of PUFA were reduced with cooking by 10.1% in the control group and 6.54% in the GOA group (Table 3). While, SFA level was greater in chickens fed control diet compared to chickens fed GOA (14.4 vs. 13.1; P<0.0001%), respectively.

The amounts of palmitic and stearic acids in raw meat of control chickens were higher (P<0.0001) compared to chickens fed GOA with respective percentages 26.6% vs. 23.0% for C16:0 and 9.2% vs. 7.6% for C18:0. This effect was observed even after cooking (27.9% vs. 23.9% for C16:0 and 8.1% vs. 7.4% for C18:0). Moreover, the MUFA proportions were higher in meat of GOA chickens than in controls. This difference is about 7.27% and 5.1%, respectively, for raw and cooked meat (Table 3). Indeed, the proportion of oleic acid was higher significantly (P<0.0001) in the GOA group (44.5% vs. 40.0%) and remains constant after cooking (45.1% vs. 40.0%).

Despite the decrease in the amount of PUFA, a neat effect was observed in muscle quality of GOA group. Indeed, the proportion of PUFA reported by chicken muscle of GOA diet is higher than that observed by the chicken control, whatever the treatments (raw or cooked). The difference is significant (P<0.01) (8.3 vs. 11.8%) for the raw and cooked meat. The amount of essential fatty acids (C18:2, n-6; C18:3, n-3), are better represented in GOA group compared to the control group. Linoleic acid represents an amount of 75% of identified PUFA. In raw meat, linoleic acid level varying significantly from 17.3 % in GOA groups to 15.4% in control groups and 16.8% vs.

14.4% in cooked meat (P<0.0001). Linolenic acid is the most important polyunsatured fatty acids of the group of n-3 PUFAs which represents over 13% among the PUFA group. In our experimental conditions, its value is slightly higher, but not significant in the GOA group compared to the control group, especially in raw meat. However, a significant effect of n-6 PUFAs in the GOA diet was observed, with 8% difference for raw meat and 12% for cooking meat (P<0.05). In addition, the n-3 PUFAs content varied significantly after cooking, with a pronounced decrease in the GOA group compared to the control group (26% vs. 16%, P<0.05), respectively. The n-6/n-3 ratio was higher in the cooked meat of the GOA group than in the control group by 11% (P<0.01).

Finally, whatever the meat treatment (raw or cooked), a significant increase was shown in the PUFA/SFA ratio of chickens fed GOA diet compared to control chickens (0.7 vs. 0.5 in raw meat and 0.6 vs. 0.5 in cooked meat) (P<0.0001).

Table 3
Fatty acid Composition of Pectoralis major. Expressed in % of identified fatty acids).

	Control group		GOA group		*P* values	
	Raw	**Cooks**	**Raw**	**Cooks**	**Diet Effect**	**Cooking Effect**
Total Lipid ($g.100g^{-1}$ of muscle)	1.2±0.2a	2.2±0.7b	1.2±0.2a	2.0±0.2b	NS	<0 .0001
C14:0	0.4±0.0a	0.4±0.1a	0.4±0.1b	0.3±0.1b	<0. 005	NS
C16:0	26.6±0.6b	27.9±1.6a	23.0±0.8d	23.9±0.7c	<0.0001	< 0.005
C16:1	4.4±0.7a	5.2±1.2a	3.1±0.7b	3.4±0.5b	<0.0001	NS
C18:0	9.2±0.6a	8.1±0.9b	7.6±0.5c	7.4±0.7d	<.0.0001	< 0.01
C18:1(n-9)	40.0±1.6b	40.9±2.4b	44.5±2.3a	45.1±2.8a	<0.0001	NS
C18:2(n-6)	15.4±1.3b	14.4±1.8b	17.2±1.0a	16.8±1.5a	<0.0001	NS
C18:3(n-3)	0.4±0.1a	0.4±0.1a	0.4±0.1a	0.4±0.1a	NS	NS
C20:1(n-9)	0.3±0.2a	0.3±0.1a	0.4±0.2a	0.4±0.1a	NS	NS
C20:4(n-6)	2.8±0.9a	2.0±1.7b	2.8±0.7a	1.9±0.6b	NS	< 0.01
C22:5(n-3)	0.3±0.1a	0.2±0.2b	0.3±0.1a	0.2±0.1b	NS	< 0.01
C22 :6 (n-3)	0.2±0.1a	0.1±0.2b	0.2±0.1a	0.1±0.1b	NS	< 0.01
SFA	36.2±0.5a	36.4±1.0a	30.9±1.0b	31.7±0.6b	< 0 .0001	NS
UFA	63.9±0.5b	63.6±1.0b	69.0 ±1.0a	68.3±0.6a	< 0.0001	NS
MUFA	44.7±2.2b	46.4±3.3b	48.2±2.2a	48.9±2.5a	<0.001	NS
PUFA	19.1±2.1b	17.1±3.8b	20.8±2.0a	19.4±2.3a	<0.01	NS
n-6	18.2±2.1b	16.4±3.3b	19.8±1.8a	18.7±2.1a	<0.05	NS
n-3	0.9 ±0.2a	0.7±0.5b	1.0±0.2a	0.8±0.1b	NS	<0.05
n-6/n-3	21.4±5.5b	27.4±11.1a	20.0±5.1b	30.8±14.6a	NS	<0.01
PUFA/SFA	0.5±0.1b	0.5±0.1b	0.7±0.7a	0.6±0.1a	<0.0001	NS

For each group n=15. Data are expressed as mean and standard deviation. NS: Not significant. The values in the same line with different superscript letter are significantly different at 1% and 5%.

4. Discussion

The present study shows that cooking cause water loss and gain of fat irrespective of diet consumed. Our results show an increase in the DM content during cooking, especially in control chickens due to a high water loss compared to GOA chickens. Water loss during cooking can be explained by the physical protein denaturation of animal due to the combination of a pH of meat already low while the muscle temperature is still high (Astruc, 2007). The difference in water loss between the two groups is due to a high level of proteins in the meat of chickens fed a control diet compared to chickens fed GOA diet. Obtened results were in good agreement with previous results of Kim (1989) compared broiler chickens from quality label and standard chickens. The obtained results show that the quantity of minerals in the meat increases after the cooking because of a big loss of water.

An observation which confirms the observations of posati (1979) and combes (2004), but which does not suit those of rabot (1998), which showed that cooking pulls a loss in minerals.

Interestingly, muscles of chicken fed acorn (GOA) contained less protein compared to chicken corn regardless of the treatment of meat. This result is also agreement with that obtained by Rabot (1998). According to Blum (1984), this difference in content can be explained by the low proportion of protein contained in the oak acorn and the rate of which hardly exceeds 6%.

In this study, we showed a fat gain in the muscle after cooking for both groups due to the phenomenon of dehydration of the meat. These findings sustain those demonstrated that cooking increases lipid content of chicken's meat (Stadelman, 1978; Prusa and Louergan, 1987; Ackman et al, 1988; Rabot, 1998). During cooking, thermo-mechanical transformations were observed. Indeed, due to the osmotic pressure the water migrates to the surface of meat during cooking. These phenomena generate a crust that inhibits the migration of water, whiwch fundamentally alters the evolution of surface temperature and determines the final product quality (Kondjoyan and Peynon, 2006). After cooking, the lipid content observed in chickens fed GOA diet was lower (1.99 g/100g) than those of control chickens (2.21 g/100g). These levels are consistent with standard values (about 2 to 3 g/100 g) and these ones are indirect but convincing evidence of a reduced lipogenic synthesis in the liver of chickens fed acorn diet. A low lipogenic synthesis can be explained by the high level of PUFA on the acorn diet (Rabot, 1998; Lessire, 2001).

According to other studies, after cooking a gain of fat was observed in the pork chops and sausage, respectively (30% and 22%) and 100% of rabbit meat. The high content of lipid in rabbits after cooking can be explained by the nature of its meat that contains little fat and a lot of water (Mourot et al., 2006). Interestingly, fatty acid profile of *Pectoralis major* muscle reflects the balance between dietary fatty acids and endogenous hepatic synthesis, which was strongly influenced by dietary fatty acid composition (Mossab, 2001).

Besides, our results are in agreement with observations of Bouderoua et al. (2009) which showed that the introduction of GOA in the diet of broilers reduced the fat content of the thigh muscle and increase the proportion of linoleic and linolenic acids. In the present research, quantitative MUFA was greater in the muscle of GOA chickens (48.5% on average) compared to control chickens (45.5% on average). This result demonstrated that the GOA contains a high proportion of unsaturated fatty acid (UFA). Moreover, PUFA level was also elevated in chickens fed GOA compared to those fed the control diet with respective values about 20.1% and 18.1%. Therefore, the fatty acid compositions of the food affect strongly fatty acid composition of the muscle.

In addition, heat treatment, causes a partial degradation of PUFA during cooking. Thus, PUFA undergoes a partial destruction by oxidation in both types of chickens. This degradation was very low in control chickens but, high in GOA chickens. The proportion of PUFA altered during cooking increases with the number of double bonds in the fatty acid (Kim, 1989). During cooking, and in control chickens, (n-3) and (n-6) long chain PUFA undergo a slight deterioration compared to GOA chickens (28.8% vs. 32%) for C20:4 (n-6); (31% vs. 39%) for C22:5 (n-3) and (40% vs. 50%) for C22:6 (n-3).

The proportion of linoleic and linolenic acids remain constant after cooking and for the two types of chickens. It is well known that the sensitivity to oxidation of fatty acids increases with the number of double bonds in their carbon chain (Kim, 1989). Effects of cooking on the oxidation of PUFA were studied by Kim (1989), Posati (1979) and Igene et al. (1979). These authors demonstrated that in chicken label 20% to 50% of arachidonic acid (C20:4) (n-6) disappears during cooking, while this proportion exceeds 50% for docosapentaenoic acid C22:5 (n-6) and docosahexaenoic acid C22:6 (n-3). In our study, degradation of PUFA was very low in control chickens. It's known that, the proportion of polyunsaturated fatty acids deteriorated during the cooking of the pectoral muscles, increases with the number of double bonds of the fatty acid. Thus, the loss of linoleic acid varied from 0 to 20%, while they reach 30% to 50% for C20:4 (n-6), 50% for C22:5 (n-6) and 67% for C22:6 (n-3) (Gandemer, 1997).

Interestingly, the oxidation of PUFAs during cooking promotes the subsequent appearance of UFA with elevated levels. These results are consistent with those published by Posat (1979) and Kim, (1989) indicated that the muscles of label chickens had much greater oxidation during cooking (higher loss of PUFA) than the standard chickens. Differences in oxidation between the two types of chickens during cooking may be explained by differences in fatty acid composition. Indeed, the muscles of chickens fed GOA diet contain a greater level of PUFA than those of control chickens. The decrease in PUFAs (n-6) and (n-3) during cooking, can be explained by their oxidation that should be ensured by measuring oxidized products.

Finally, after cooking, we showed a high ratio of PUFA/SFA in GOA chickens compared to control chickens, which is certainly due to the richness of the GOA meat in PUFA. These results confirm the observations of Bouderoua et al. (2009).

In conclusion, whatever the diet fed to chickens, cooking roast type causes water loss and a gain of dry matter and fat. No differences in proteins and mineral levels of muscles were observed. The control and experimental chickens are differentiated by their fat contents. Indeed, muscle control *Pecroralis* chicken contains more total fat than chicken GOA (1.21% vs. 1.16%) respectively. Fatty acid compositions differ between the two types of chicken. The incorporation of the acorns in chicken diet has increased significantly the amount of UFA in particular linoleic acid and linolenic acid, and lower content of SFA. The chicken enriched UFA could be a major source of long chain PUFA immediately available to meet human needs and contribute to the prevention of cardiovascular disease risk.

A good diet and nutrition of the chicken of the GOA is linked to a high proportion of unsaturated fat, especially PUFA are very susceptible to oxidation during cooking. Net loss in PUFA of chicken's GOA after cooking was observed, which is not the case for control chickens. This marks a greater oxidation of PUFA in chickens than in chickens control under identical conditions of cooking. This result is explained by the presence of a larger proportion of PUFA in muscles of chickens GOA than in control chickens. Whatever the type of chicken, there is a decrease in long chain n-3 PUFAs and n-6 PUFAs, especially C22:5 (n-3), C22:6 (n-3) and C20:4 (n-6), whose proportions fall into the net much more acorns than corn nets, this is explained by the oxidation of PUFA during the heat treatment. But the proportions of linoleic acid and linolenic remain constant. The results mark a high content of PUFA/SFA ratio in chickens GOA than in control chickens after cooking, it confirms once more the richness of the chicken fillet acorns PUFA.

Finally, despite changes in the composition of meat from chickens fed GOA during cooking, dietary and nutritional value is still the best compared to control chickens. The tenderloin cooked chicken acorn contains low in SFA and high levels of UFA (especially linoleic acid and linolenic acid). All these properties make this a staple food of choice essential quality diet and nutrition currently highly sought after by consumers.

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