

Evaluation of polyunsaturated to saturated fatty acid ratio in the fat of different chicken types

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Abstract

Samples were taken from the liver and the abdominal fat of the carcasses of male and female broilers (meat type), layers (egg type) and locals (dual purpose type). Lipids were extracted and subjected to volumetric analysis, iodine number determination, thin layer chromatography (TLC) with iodine staining and high performance liquid chromatography – mass spectrometry (HPLC-MS) to determine the polyunsaturated to saturated fatty acid (P/S) ratio. The result showed that female chickens, meat type chickens and abdominal fat samples had higher ($P < 0.05$) lipid quantities than male chickens, layer and dual purpose chickens and liver samples, respectively. The P/S ratio of lipids extracted from the abdominal fat and from the liver did not differ ($P < 0.05$) from each other.

Keywords: P/S ratio, fat, broiler, layer, dual purpose.

Introduction

Excessive intake of food having high saturated fatty acid (SFA) as a fraction of the total fatty acid composition predisposes humans to excess cholesterol accumulation by increasing low density lipoprotein cholesterol (National Cholesterol Education Program, 1991; Sacks and Byers, 2001). This has led to the reduction in the consumption of red meat due to its high content of SFA which predisposes to arteriosclerosis. On the contrary, consumption of chicken meat is increasing. Different parts of the chicken have been reported to have varied amounts of fats (USDA, 2016). Therefore, it is important to investigate the fatty acid composition of different chicken types. The objective of this study was to evaluate the effect of type, sex and carcass sampling sites on P/S ratio of chicken carcass fat.

Materials and methods

A total of thirty six chickens were used. Twelve meat chicken types were obtained from the Poultry Unit of the Obafemi Awolowo University Teaching and

Research farm, Ile-Ife, Osun State. Twelve layer type and twelve dual purpose chickens were obtained from Sabo and Oja Ife markets, Ile-Ife, respectively. The body weight ranges of the chickens were: 2.5 kg-2.7 kg (female meat type), 2.3 kg-2.5 kg (male meat type), 3.3 kg-3.5 kg (male layer type), 3.2 kg-3.4 kg (female layer type), 1.8 kg-2.0 kg (female dual purpose type), and 3.0 kg-3.2 kg (male dual purpose type). There were six experimental units: female meat type, male meat type, female layer type, male layer type, female dual purpose and male dual purpose.

The birds were slaughtered and dressed, liver and abdominal fat samples were taken from each experimental unit with three replicates per sample making a total of 108 samples. Samples were wrapped in black cellophane bags to prevent moisture and oil loss, or light penetration which could cause oxidation. The wrapped samples were sealed in a tightly covered bowl and kept in a freezer at -20°C .

Lipid was extracted and stored in the freezer (20°C) following the procedure of Folch *et al.* (1957) at Poultry Meat Research

P/S ratio of different chicken types.

Laboratory, Department of Animal Sciences, Obafemi Awolowo University, Ile-Ife, Nigeria. Volumetric analysis of extracted lipid using titration method was carried out following the procedure of Egan *et al.* (1991) to determine the saturated, monounsaturated and polyunsaturated fatty acids fraction.

At Lipid and Natural Products Laboratory, Department of Biochemistry, Obafemi Awolowo University, Ile-Ife, Nigeria, iodine number determination of lipid extracted from liver samples was done following Hanus' procedure and Thin Layer Chromatography analysis was carried out on the lipid extract. It involves the separation of lipid fractions on silica gel prepared glass plate. The separated spots on silica gel plates were identified by staining with vanillin-sulphuric acid solution to identify all spots and iodine crystals to identify unsaturated spots.

Fresh lipid extract was collected and dissolved in 5ml of chloroform poured into a 5-10 ml glass vial and sent to the Faculty of Health and Wellness Science, Biotechnology Laboratory, Cape Peninsula University of Technology, South Africa. Extracted lipid dissolved in chloroform (1 mL) was mixed with 6 mL of 2 M sodium methoxide (or sodium hydroxide) in anhydrous methanol and shaken vigorously. The solution was heated in a steam bath at 500°C for about 60 minutes. Afterwards, 10 mL of 2 M methanolic solution of hydrochloric acid was added and the sample reheated. Distilled water (5 mL) was added and the required esters were extracted into n-hexane. The hexane layer was washed with 4 ml dilute potassium bicarbonate (5%) to remove excess acid and dried over anhydrous sodium sulphate. The gradient elution was prepared using 0.1% formic acid in water as solvent A and acetonitrile as solvent B. The gradient was

set up as follows: 2% B to 100% B (30 min); held at 100% B (30 – 36 min); 2% B (37 – 45 min). Thermo Fischer Scientific C18 column (5 µm; 4.6 × 150 mm, Bellefonte, USA) was used through which the samples were eluted. The injection rate was 0.6 µl per min at oven temperature of 300°C. Samples eluted from the HPLC column were passed into a mass spectrometer for fragmentation and structure determination. A mass spectrometer with specifications ESI QTOF (Bruker Daltonik GmbH, Germany) was used. Mass spectrometer settings include negative mode with Electrospray voltage +3500 V at dry gas temperature of 300 °C, dry gas flow of 9 lmin⁻¹; nebulizer gas pressure 35 psi.

The experiment was designed as a randomized complete block design. Data collected were subjected to analysis of variance using General Linear Model (GLM) and t-test procedures in the SAS statistical package. Duncan's multiple range test was used to determine the significant differences among the means.

Results and discussion

From the result shown in Table 1, abdominal fat of meat type chicken gave the highest quantity of lipid extract while abdominal fat of dual purpose chicken had the lowest quantity of lipid extract. Lipid extracted from the liver of the three chicken types studied did not differ significantly from each other. Regardless of chicken type, lipid extracted from abdominal fat sample sites had significantly higher quantity of lipid than lipid extracted from liver sample sites (Table 1). From this result and the report of Pikul *et al.* (1985), abdominal fat has the highest fat composition and could be a good representation of the total quantity of lipid in the chicken body.

Table 1: Percent lipid content and polyunsaturated to saturated fatty acid ratio of lipid extracts of two sampling sites from three chicken types

Chicken type	Sampling Site	SFA \pm SEM (%)	PUFA \pm SEM (%)	P/S ratio \pm SEM	Lipid content \pm SEM (%)
Meat type	Abdominal fat	2.3 \pm 0.10	0.9 \pm 0.02 ^{ab}	0.39 \pm 0.05 ^a	34.67 \pm 2.09 ^a
	Liver	2.2 \pm 0.00	0.9 \pm 0.05 ^{ab}	0.41 \pm 0.05 ^a	8.17 \pm 1.66 ^c
Egg type	Abdominal fat	2.1 \pm 0.15	0.7 \pm 0.00 ^{ab}	0.33 \pm 0.45 ^a	30.50 \pm 3.89 ^{ab}
	Liver	2.0 \pm 0.25	0.6 \pm 0.05 ^b	0.30 \pm 0.01 ^a	5.33 \pm 0.61 ^c
Dual purpose type	Abdominal fat	2.2 \pm 0.35	0.9 \pm 0.25 ^{ab}	0.41 \pm 0.06 ^a	25.67 \pm 4.27 ^b
	Liver	2.4 \pm 0.35	1.1 \pm 0.00 ^a	0.46 \pm 0.04 ^a	6.17 \pm 0.79 ^c
Mean across sites	Abdominal fat	0.38 \pm 0.03			30.28 ^a
	Liver	0.38 \pm 0.03			6.56 ^b

^{a, b, ab} Means in the same column having the same superscript do not differ significantly at P<0.05.

Meat type = broiler chicken, egg type = layer chicken, dual purpose = local chicken

In this study, there was no significant difference in the polyunsaturated to saturated fatty acid (P/S) ratio between the liver and abdominal fat sample sites in the three chicken types. This implies that sample sites for lipid extraction and the interaction between sample site and chicken type did not have an effect on the

P/S ratio. This agrees with the result of Kralik *et al.* (2005) who observed no significant difference in P/S ratio of breast and thigh muscle sample sites. Zlender *et al.* (2000), however, reported that there was a significant difference in P/S ratio between breast muscle with skin and leg muscle with skin at p<0.05.

Table 2: Saturated fatty acid and polyunsaturated fatty acid composition of lipid extract from three chicken types

Chicken type	Sex	Iodine No. \pm SEM	SFA	PUFA
Meat type	Female	239 \pm 85.81 ^a	C12:0, C22:0	C18:2,C20:4,C20:5,C22:5,C22:6
	Male	41 \pm 5.13 ^b	C12:0, C22:0	C18:2,C20:4,C20:5,C22:6
	Mean	139.8 \pm 67.11 ^a	-	-
Egg type	Female	122 \pm 20.05 ^{ab}	C20:0, C22:0	C18:2,C18:3,C20:4,C22:5,C22:6
	Male	166 \pm 11.74 ^{ab}	C14:0, C20:0, C22:0	C18:2,C18:3,C20:4,C20:5,C22:5,C22:6, C28:6
	Mean	144.0 \pm 15.97 ^a	-	-
Dual purpose	Female	203	C10:0, C20:0, C22:0	C18:3,C20:4,C20:5,C22:5,C22:6
	Male	122 \pm 6.36 ^{ab}	C20:0, C22:0	C18:2,C18:3,C20:4,C20:5,C22:5,C22:6
	Mean	111.6 \pm 49.91 ^a	-	-

^{a, b, ab} Means in the same column having the same superscript do not differ significantly at P<0.05.

Meat type = broiler chicken, egg type = layer chicken, dual purpose = local chicken

P/S ratio of different chicken types.

Sex had significant effect on the degree of unsaturation of lipid using iodine number determination (Table 2). Female meat type chickens had more than 500% more unsaturation than males. In the egg type chickens, sex had no significant effect on the degree of unsaturation of lipid. Rondelli *et al.* (2004) had reported that sex had no effect on the saturation of fatty acids in mixed-sex broiler chickens.

The retardation factor (R_f) obtained for lipid extracts during TLC analysis was compared with standards with different degrees of unsaturation. Cod liver oil with iodine number of 157 and 23% PUFA had R_f of 0.92 while olive oil with iodine number of 86 and 11% PUFA had R_f of 0.84. Lipid samples from female meat type chickens had R_f of 0.94. Lipid samples from male egg type and male dual purpose chickens had R_f of 0.68 and 0.80, respectively. These results indicate that female chickens on this study contained more PUFA than male chickens.

Considering the polyunsaturated fatty acids (PUFA) in the chicken liver fat (Table 2), female meat type had similar PUFA with female dual purpose except for linoleic acid (C18:2) present in female meat type but absent in female dual purpose, and -linolenic acid (C18:3) present in female dual purpose but absent in female meat type. The PUFA in male egg type and male dual purpose were also similar but differ slightly with the presence of C28:6 in male egg type which was absent in male dual purpose type.

Conclusion

From this study, it can be concluded that: female chickens had higher lipid content than male chickens; that fat from the abdominal site had higher lipid quantity than fat from the liver; that meat type chickens had the highest lipid content as compared with egg type chickens which

had higher lipid content than dual purpose chickens; that there was no difference in the polyunsaturated to saturated fatty acid ratio of lipids extracted from abdominal fat and from the liver. Across the three chicken types, there was no difference in the degree of unsaturation of the lipids extracted from different chicken types. In egg type chickens only, there was a higher degree of unsaturation in lipids extracted from females than from males. Quantitatively, lipids extracted from male egg type chickens had the highest carbon number polyunsaturated fatty acid as C28:6, while lipids extracted from female egg type, male and female meat type and male and female dual purpose chickens had the highest carbon number polyunsaturated fatty acid as C22:6.

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