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ORIGINAL ARTICLE

Influence of amount and type of dietary fat on deposition, adipocyte count and iodine number of abdominal fat in broiler chickens

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Summary

This study described the relation between the type and amount of dietary fat on the deposition of abdominal fat by broiler chickens. It was hypothesized that at higher fat intakes, the well-known lowering effect of polyunsaturated fatty acids on the deposition of abdominal fat would be diminished. Experimental diets were formulated to contain three levels of added fat (3%, 6% and 9%). Each level had different proportions of the saturated fatty acids (SFA) and unsaturated fatty acids (UFA) by installing the ratios of 1:1, 1:2, 1:3, 1:4 and 1:5 with the use of tallow and soybean oil. Arbor Acres chicks, aged 7 days, were fed one of the 15 experimental diets until they were aged 42 days. Feed and water were provided *ad libitum*. There was no systematic effect of the dietary fat type and the amount on the weight gain and the feed intake. The lowest SFA:UFA ratio of 1:5 produced the lowest feed conversion rates, irrespective of the amount of the fat in the diet. The abdominal fat deposition was similar in the birds fed on diets containing either 3% or 6% added fat, but deposition was lower than in those fed 9% fat. A decrease in the SFA:UFA ratio of the diet was associated with a dose-dependent decrease in abdominal fat, irrespective of the amount of fat in the diet. This observation leads to the rejection of the hypothesis stated above. A decrease in the dietary SFA:UFA from 1:1 to 1:4 caused a decrease in the number of the fat cells per surface unit of breast meat. It is concluded that an increased intake of soybean oil at the expense of tallow reduced the abdominal fat deposition and the number of fat cells in the breast meat of broiler chickens.

Introduction

The fatty acid composition of the diet influences the composition and the amount of abdominal fat in broiler chickens. The relative percentage of dietary polyunsaturated fatty acids, such as linoleic and α-linolenic acid, is directly related to the percentage of these fatty acids in the adipose tissue of broilers (Bavelaar and Beynen, 2003). The addition to the diet of an oil rich in polyunsaturated fatty acids at the expense of a more saturated fat source reduces the amount of abdominal fat in broilers (Pinchasov and Nir, 1992; Zollitsch et al., 1997; Sanz et al., 1999, 2000a; Crespo and Esteve-Garcia, 2002a;

Newman et al., 2002; Villaverde et al., 2005). The dose-response relationship for the intake of polyunsaturated fatty acids and the amount of abdominal fat has not yet been described.

The metabolic basis for the diminishing effect of polyunsaturated fatty acids on the abdominal fat mass is little understood (Sanz et al., 2000b; Crespo and Esteve-Garcia, 2002b,c, 2003; Newman et al., 2002; Villaverde et al., 2006). One possible mechanism could be that the polyunsaturated vs. saturated fatty acids are preferentially oxidized (Beynen and Katan, 1985) and thereby yield ATP, so that the carbohydrates are shifted from the oxidative into the lipogenic pathway. The conversion of glucose into triglycerides is less efficient in terms of energy deposition than is the conversion of the fatty acids into triglycerides (Newsholme and Leech, 1984). Consequently, the feeding of polyunsaturated, instead of saturated, fatty acids may lead to less deposition of the abdominal fat. At high fat intakes, β-oxidation is enhanced through the increased activities of the enzymes involved (Newsholme and Leech, 1984) which could imply that preferences for the individual fatty acids become masked. It would follow that the different effect of polyunsaturated and saturated fatty acids on the deposition of the abdominal fat is less with the increasing fat intakes. In this study, the above reasoning was put to the test. Broiler chickens were fed diets with five different ratios of tallow:soybean oil at three dietary fat levels and the amount of abdominal fat was determined. In this way, we would be able to report the dose-response relationship between the intake of the polyunsaturated fatty acids and the amount of the abdominal fat, and it would be possible to indirectly evaluate the above considerations as to the metabolic effects of the dietary polyunsaturated fatty acids. In addition to the amount of the abdominal fat, the number of fat cells per unit of surface in the breast meat were measured. The iodine value of various tissues was analysed as an index of the incorporation of the polyunsaturated fatty acids. High tissue concentrations of the polyunsaturated fatty acids were associated with high iodine values (Gurr and Harwood, 1991).

Materials and methods

Animals and diets

A total of 390 (195 males and 195 females) 7-day-old Arbor Acres broiler chicks were used. They had been raised on a commercial diet. According to the

Table 1 Ingredient and nutrient composition of the experimental diets

Level of added fat, %	3					6					9				
SFA:UFA*	1:1	1:2	1:3	1:4	1:5	1:1	1:2	1:3	1:4	1:5	1:1	1:2	1:3	1:4	1:5
Ingredient (g/100 g)															
Tallow	2.87	1.45	0.72	0.28	–	5.52	2.85	1.48	0.65	0.10	8.15	4.26	2.25	1.03	0.22
Soybean oil	0.13	1.56	2.28	2.72	3.00	0.48	3.15	4.52	5.35	5.91	0.85	4.74	6.75	7.97	8.87
Tapioca starch	46.02	46.02	46.02	46.02	46.02	43.02	43.02	43.02	43.02	43.02	40.02	40.02	40.02	40.02	40.02
Constant components†	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98	50.98
Analysed composition (g/100 g)															
Dry matter	91.98	91.98	91.98	91.98	91.98	91.95	91.95	91.95	91.95	91.95	91.85	91.85	91.85	91.85	91.85
Crude protein	17.67	17.67	17.67	17.67	17.67	18.12	18.12	18.12	18.12	18.12	17.84	17.84	17.84	17.84	17.84
Crude fat	2.83	3.23	3.14	2.97	3.03	6.39	6.43	6.17	6.27	6.41	9.34	9.18	9.12	9.36	9.23
Crude fibre	3.14	3.14	3.14	3.14	3.14	3.22	3.22	3.22	3.22	3.22	3.13	3.13	3.13	3.13	3.13
Ash	4.14	4.14	4.14	4.14	4.14	4.37	4.37	4.37	4.37	4.37	4.22	4.22	4.22	4.22	4.22
Calculated composition (g/100 g)															
Protein:Energy ratio	799.66	799.66	799.66	799.66	799.66	811.26	811.26	811.26	811.26	811.26	855.94	855.94	855.94	855.94	855.94
Calcium	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Available phosphorus	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49	0.49
Sodium	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Nitrogen free extract	64.20	63.80	63.89	64.06	64.00	59.85	59.81	60.07	59.97	59.83	57.32	57.48	57.54	57.30	57.43

*Ratio of saturated to unsaturated fatty acids in the whole rations.

†The constant components consisted of the following (g): soybean meal, 41.05; rice bran, 4.0; di-calcium phosphate, 3.87; d,l-methionine, 0.3; l-lysine hydrochloride, 0.25; sodium chloride, 0.51; premix, 1.0. The premix supplied per kg of diet: vitamin A, 1,650 IU; vitamin D, 330 IU; vitamin E, 11 IU; vitamin K, 0.55 mg; thiamine, 198 mg; riboflavin, 3.96 mg; niacin, 3.30 mg; pyridoxine, 3.85 mg; vitamin B_{12}, 0.01 mg; calcium pantothenic acid, 10 mg; folacin, 0.61 mg; biotin, 0.17 mg; choline, 1,430 mg; manganese, 65.99 mg; iodine, 0.39 mg; potassium, 330 mg; zinc, 43.97 mg; copper, 8.80 mg; ferrous, 87.59 mg; selenium, 0.17 mg.

manufacturer, the diet contained 87% dry matter, 21% crude protein, 4% crude fat, 5% crude fibre and 4% ash. Within sex, they were randomly allocated to 15 groups consisting of 13 birds each. The birds were kept in individual cages. Until the chickens were aged 3 weeks, the temperature in each cage was kept at 30–34 °C with 200-W electric bulbs. Feed was provided *ad libitum* in the form of meal. Animals had free access to water. The experimental diets were formulated to contain three levels of added fat (3%, 6% and 9%) and five ratios of saturated to unsaturated fatty acids by mixing tallow with soybean oil. The experimental design had a $3 \times 5 \times 2$ factorial arrangement. However, extra fat was added to the diets at the expense of an identical weight of the tapioca starch component. Consequently, higher fat contents were associated with higher energy densities and, therefore, lower nutrient:energy ratios. The analysed (AOAC, 1975) gross energy contents of the diets containing 3%, 6% and 9% added fat were 14.1, 14.7 and 15.3 MJ/kg respectively. Table 1 shows the composition of the diets. The basal diet without added fats, but with the base content of tapioca starch, was analysed for dry matter, crude protein, crude fat, crude fibre and ash (AOAC, 1975). The macronutrient concentrations in the whole diets were then calculated. The nitrogen-free extract was calculated as the residual fraction. Table 1 shows the macronutrient composition of the diets. The ratios of saturated to unsaturated fatty acids (SFA:UFA) were calculated on the basis of feed table values (NRC, 1994) for the fatty acid compositions of the feed ingredients. Individual body weights and the feed intakes were determined.

Sample collection

When they were 42 days old, the broilers were stunned, slaughtered and bled at a local slaughter house. One bird per dietary group per gender was randomly taken for chemical analysis so that, in total, there were 30 animals. The abdominal adipose tissue from the proventriculus surrounding the gizzard down to the cloaca, breast, thigh and liver were removed and stored at −20 °C. A portion of the breast meat was stored in 10% buffered formalin solution.

Chemical and histological analyses

Feed samples were analysed for dry matter, crude protein, crude fat, crude fibre and ash (AOAC, 1975). The nitrogen-free extract was calculated as

Table 2 Growth performance of broilers fed the experimental diets

Level of added fat, %	3					Pooled	6					Pooled	9					Pooled
SFA:UFA	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE
Initial weight, g	156	159	158	159	156	4.07	156	156	155	156	155	4.04	153	153	153	153	155	4.01
Final weight, g	1429^{a}	1407^{a}	1512^{b}	1562^{bc}	1635^{c}	12.56	1458	1570	1435	1556	1472	12.54	1542^{a}	1519^{a}	1456^{b}	1561^{a}	1709^{c}	12.79
Average daily gain, g/b/d	36.3^{a}	35.7^{a}	38.7^{b}	40.1^{b}	42.3^{b}	2.01	37.2	40.4	36.6	40.0	37.6	2.01	39.7^{a}	39.0^{a}	37.2^{b}	40.2^{a}	44.4^{c}	2.05
Average daily feed intake, g/b/d	94.4	95.4	95.8	94.9	98.7	3.17	98.8^{a}	103.4^{a}	91.1^{b}	93.7^{b}	90.8^{b}	3.17	94.1	92.7	93.8	94.6	95.7	3.14
Feed conversion ratio	2.60^{a}	2.67^{a}	2.48^{b}	2.37^{bc}	2.34^{c}	0.51	2.67^{a}	2.55^{ac}	2.50^{abc}	2.34^{bc}	2.41^{c}	0.51	2.37^{a}	2.38^{a}	2.52^{b}	2.35^{a}	2.16^{c}	0.50
Energy intake:gain ratio, kJ/g of wt Gain	36.7^{a}	37.7^{a}	35.0^{b}	33.5^{bc}	33.1^{c}	7.21	39.3^{a}	37.5^{ac}	36.8^{abc}	34.4^{bc}	35.4^{c}	7.50	36.0^{a}	36.2^{a}	38.3^{b}	35.7^{a}	32.8^{c}	7.60

Data are means for 24–26 birds per dietary treatment and pooled SE's for each level.
SFA, saturated fatty acids; UFA, unsaturated fatty acids.
[abcd]Values within a row with different superscripts are significantly different; $p < 0.05$.

residual fraction. The iodine number of tissues was measured according to the Hanus method (AOAC, 1975). The number of fat cells per surface unit of the breast meat was counted by a histological method as described by Carson (2001).

Statistical analyses

As mentioned above, the nutrient:energy ratios of the diets with different amounts of fat were different. Therefore, only comparisons within the fat levels were made. ANOVA showed that there was no influence of sex on any of the variables. The data for males and females were then pooled. All data were subjected to Duncan's multiple range test within a fat level was used to identify statistically significant differences between group means (Steel and Torrie, 1980).

Results

A total of eight birds died during the course of the experiment. There was no association between mortality and the diet composition. Table 2 shows that the average daily gain (ADG) was not systematically influenced by the SFA:UFA ratio of the diet. This holds also for average daily feed intake (ADFI). At each level of the added fat, the feed conversion ratio (FCR) was significantly lower at an SFA:UFA ratio of 1:5 than at a ratio of 1:1. However, there was no clear relationship between the SFA:UFA ratio and the FCR.

The tissue iodine number increased in the order of the breast, the thigh, the abdominal fat and the liver (Table 3). For all tissues, an increase in the dietary SFA:UFA ratio produced an increase in the iodine number, but significant differences generally were seen only for the highest vs. the lowest SFA:UFA ratio.

The mean final body weights of the birds selected for the abdominal fat measurements (Table 4) differed from that of all birds per dietary group (Table 2). Within each fat level, the amount of abdominal fat expressed as percentage of the body weight fell with decreasing SFA:UFA ratio (Table 4). The same pattern was seen for the absolute weight of the abdominal fat. The percentage abdominal fat was lower for the diets containing 3% or 6% added fat than for the diets containing 9% fat. The diets with 3% or 6% fat induced similar values for the relative weight of the abdominal fat. The number of fat cells in the breast meat dropped when the SFA:UFA ratio decreased from 1:1 to 1:4, but there was no further drop when the ratio was decreased to 1:5 (Table 4).

Discussion

There was no clear pattern by which the dietary SFA:UFA ratio affected the growth performance, but, at each fat level, the highest intake of UFA was associated consistently with the lowest FCR. Also ADFI and ADG were not systematically low and high, respectively, when the broilers were fed on the diets with the lowest SFA:UFA ratio. It is thus difficult to see why the lowest SFA:UFA ratios were accompanied by the lowest FCRs. The low FCR for birds with the highest UFA intake could relate to a higher digestibility of UFA and/or to a higher protein:fat ratio in the carcass and/or to less heat production. Indeed, oils rich in UFA are digested by the broiler chickens more easily than are fats rich in SFA (Wiseman et al., 1991; Smits et al., 2000). In birds fed on a diet containing sunflower oil instead of tallow, there was no difference in carcass composition (Crespo and Esteve-Garcia, 2002a). Newman et al. (2002) provided evidence that the feeding of

Table 3 Iodine numbers for tissues from broilers fed the experimental diets

Level of fat, %	3					Pooled	6					Pooled	9					Pooled
SFA:UFA	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE
Broiler tissue																		
Breast	24^{a}	32^{b}	37^{b}	37^{b}	46^{c}	1.94	27^{a}	33^{ab}	36^{abc}	40^{bc}	44^{c}	1.96	27^{a}	32^{ab}	39^{b}	41^{bc}	47^{c}	2.00
Thigh	34^{a}	43^{a}	44^{a}	48^{a}	53^{b}	2.22	36^{a}	38^{a}	42^{ab}	49^{bc}	55^{c}	2.16	32^{a}	42^{b}	46^{bc}	53^{c}	51^{c}	2.19
Fat pad	47^{a}	52^{ab}	51^{a}	54^{ab}	60^{b}	2.36	48^{a}	51^{ab}	55^{abc}	57^{bc}	59^{c}	2.38	47^{a}	53^{b}	57^{c}	59^{c}	68^{d}	2.45
Liver	85^{a}	98^{b}	99^{b}	106^{b}	100^{b}	3.20	89^{a}	101^{b}	105^{b}	101^{b}	105^{b}	3.24	80^{a}	99^{b}	102^{b}	100^{b}	108^{b}	3.21

Iodine number is expressed as g iodine consumed by 100 g of tissue; it reflects the degree of unsaturation of fatty acids as the double bonds in fatty acids react with iodine.

Data are means for two birds per dietary treatment and pooled SE's for each level.

SFA, saturated fatty acids; UFA, unsaturated fatty acids.

^{abc}Values within a row with different superscripts are significantly different; $p < 0.05$.

Table 4 Abdominal fat deposition and fat cell counts for breast meat in broilers fed the experimental diets

Level of fat, %	3					Pooled	6					Pooled	9					Pooled
SFA:UFA*	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE	1:1	1:2	1:3	1:4	1:5	SE
Live weight, g	1330^{a}	1375^{a}	1575^{b}	1570^{c}	1550^{a}	12.49	1740^{a}	1610^{ac}	1408^{b}	1553^{c}	1475^{bc}	12.81	1465^{a}	1560^{b}	1655^{c}	1325^{d}	1635^{bc}	12.68
Abdominal fat, g	36.7	34.0	36.9	28.5	28.3	1.88	46.9	42.8	34.8	32.8	28.3	2.00	55.2	51.0	48.6	36.2	40.8	2.22
Abdominal fat, % live weight	2.76^{a}	2.47^{a}	2.44^{a}	1.82^{b}	1.79^{b}	0.49	2.70^{a}	2.69^{a}	2.44^{a}	2.11^{b}	1.92^{b}	0.50	3.77^{a}	3.26^{b}	2.95^{c}	2.75^{cd}	2.49^{d}	0.57
Fat cells/cm^2	69.0^{a}	51.5^{b}	43.5^{c}	41.0^{c}	38.5^{c}	2.29	91.5^{a}	81.5^{a}	51.5^{b}	39.5^{b}	43.0^{b}	2.62	89.5^{a}	82.0^{ab}	59.0^{bc}	44.0^{c}	42.5^{c}	2.64

Data are means for two birds per dietary treatment and pooled SE's for each level.
SFA, saturated fatty acids; UFA, unsaturated fatty acids.
abcValues within a row with different superscripts are significantly different; $p < 0.05$.
*SFA:UFA; ratio of saturated to unsaturated fatty acids in the whole ration, which calculated on the basis of feed table values (NRC, 1994) for the fatty acid compositions of the feed ingredients.

sunflower oil, instead of tallow, to broiler chickens influences nutrient partitioning and energy expenditure. However, this study does not support convincingly that a decrease in the SAF:UFA ratio improves the FCR, as there was no dose-response relationship. It should be noted that when formulating the diets, the higher digestibility of soybean oil vs. tallow (Smits et al., 2000) was not taken into account. Thus, a decrease in the dietary SFA:UFA ratio at the same dietary fat level would be associated with a higher digestibility of the fat component, and, thereby, with a decrease in protein:metabolizable energy ratio of the diet. The latter may tend to increase the FCR and would then counteract any positive effect of a decreasing SFA:UFA ratio on the FCR.

In keeping with earlier work (Pinchasov and Nir, 1992; Zollitsch et al., 1997; Sanz et al., 1999, 2000a; Crespo and Esteve-Garcia, 2002a; Newman et al., 2002), the abdominal fat deposition in the chickens was decreased by an increase in the UFA intake. When the diet contained either 3% or 6% added fat, the abdominal fat deposition was smaller than when the diet contained 9% fat. This observation may be explained by the relatively high energy density of the diet with 9% fat. However, it is clear that the effect of a decreasing SFA:UFA ratio on the abdominal fat deposition was similar for the three dietary fat concentrations. Thus, our hypothesis that the effect of UFA intake on the abdominal fat deposition would be smaller at higher fat intakes should be rejected.

Interesting results emerged from this study with regard to the number of fat cells per surface unit of the breast meat. The diets containing either 6% or 9% fat produced similar numbers of fat cells, but these numbers were higher than when the diet contained 3% fat. Irrespective of the amount of fat in the diet, a decrease in the dietary SFA:UFA ratio from 1:2 to 1:4 caused a dose-dependent decrease in the number of fat cells. There was no further decrease in the number of fat cells in the breast meat when the ratio dropped to 1:5. Possibly, polyunsaturated fatty acids regulate transcription factors, which, in turn, affect adipocyte development (Azain, 2004).

The present data may be important in order to enhance the quality of broilers' breast meat in terms of physical, colour and sensory characteristics (Liu et al., 2004). Poultry meat quality is a relative notion, being determined by its consistency and influence on consumer health. The consistency is related to the melting point of the fat component,

 Journal of Animal Physiology and Animal Nutrition.

which is associated with its fatty acid composition (Bavelaar and Beynen, 2003). An increase in the percentage of unsaturated fatty acids causes a decrease in the firmness and an increase in the oiliness of the poultry meat (Miller et al., 1990). Increasing the hardness of the tissue fat by extra saturated fatty acids can be advantageous in the marketing of broiler meat. However, an increase in the saturated fatty acids at the expense of the polyunsaturated fatty acids in the poultry meat may lead to an increase in the serum cholesterol of the consumer (Beynen, 1984). Thus, the ideal fatty acid composition of the poultry meat depends on whether the emphasis is on its consistency or on consumer health. On the basis of this study, a recommendation as to the SFA:UFA ratio of the diet cannot be made because neither the consistency nor the fatty acid composition of edible broiler meat was measured. It may be noted here that the fatty acid composition of broiler meat correlates well with that of the broiler adipose tissue (Bavelaar and Beynen, 2003).

In conclusion, this study clearly shows that an increased intake of soybean oil at the expense of tallow reduced the abdominal deposition in broiler chickens. A decrease in the dietary SFA:UFA ratio of the diet diminished the abdominal fat deposition in a dose-dependent manner, which was independent of the amount of fat in the diet. This study also demonstrates that the number of fat cells in the broilers' breast meat depends on both the amount and type of fat in the diet.

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