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Full Length Research Paper

Correlation between serum lipoproteins and abdominal fat pad in broiler chickens

Bahman Navidshad*[1], Hamid Deldar[2] and Gholamreza Pourrahimi[1]

1 Department of Animal Science, University of Mohaghegh Ardabili, Ardabil, Iran.
2 Department of Animal Science, Sari Agricultural Science and Natural Resources University, Sari, Iran.

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In this experiment, four hundred day-old broiler chicks were assigned randomly to 16 floor pens with 25 chicks each. Three diets containing 0% fish oil + 7% soybean oil, 3.5% fish oil + 3.5% soybean oil and 7% fish oil + 0% soybean oil and a free oil control diet were formulated. The birds fed fish oil diet showed a lower body weight than control group (P < 0.05). Abdominal fat percent in birds fed soybean oil and fish oil diets, were lower than control group (P < 0.05). The high density lipoprotein (HDL) concentration in birds fed fish oil diet was higher than other treatments, but the serum low density lipoprotein (LDL) decreased in birds fed fish oil diet (P < 0.05). The live weight of birds was positively correlated with glucose and LDL and negatively correlated with HDL concentrations (P < 0.01). The abdominal fat pad percent was positively correlated with triglyceride, glucose, LDL and very low density lipoprotein (VLDL) and a negative correlation was observed with HDL concentrations (P < 0.01). Triglyceride, cholesterol and VLDL concentrations were positively correlated with each other and a negative correlation was observed between high-density lipoprotein and low-density lipoprotein (P < 0.01). The results of the present study showed that higher fish oil inclusion in broiler diets can result in some unfavorable reduction in live weight and a beneficial influence on abdominal fat reduction.

Key words: Broiler chickens, fish oil, high-density lipoprotein, low-density lipoprotein.

INTRODUCTION

There are several reports on the various effects of fat supplementation in poultry diets. Majority of previous researches have indicated a positive effect of single or mixed dietary fats on the feeding performance and the reducing effects of some polyunsaturated fatty acids on body fat accumulation in poultry (Tuncer et al., 1987; Kirkpinar et al., 1999; Abas et al., 2004). There are also some studies which have demonstrated the influence of the fatty acid profile of the dietary fat supplement on the body fat composition of birds (López-Ferrer et al., 1999; Mandal et al., 2000).

Currently, there is an increasing attention on the direct or indirect effects of fats on consumer health. There has been efforts to improve the wellbeing of human and animals by feeding formulations (Özdogan and Aksit, 2003; Alçiçek et al., 2003). It is reported that the source and fatty acid composition of dietary fat, especially the unsaturated to saturated fatty acids ratio, are important factors in poultry rations (Yalçin and Çiftçi, 1996). Fish oils contains long chain omega-3 fatty acids with well known physiological and metabolic effects and the possibility of including them into chicken rations has being and is being researched. There have been numerous researches on the role of omega-3 fatty acids in human and animal health (Tuncer et al., 1987; Bezard et al., 1994; Manilla et al., 1999; Pike, 1999; Mandal et

*Corresponding author. E-mail: bnavidshad@yahoo.com.

Abbreviations: HDL, High density lipoprotein; **VLDL,** very low density lipoprotein; **LDL,** low density lipoprotein; **TG,** triglycerides; **PUFA,** polyunsaturated fatty acid; **LPL,** plasma lipoprotein lipase.

Table 1. Ingredients and chemical composition of experimental diets.

Ingredients	Starter (1 – 10 days)	Grower (11 - 28 days of age)				Finisher (29 - 42 days of age)			
		Control	7% SO	7% FO	3.5% SO + 3.5% FO	Control	7% SO	7% FO	35% SO + 3.5% FO
Corn (%)	60.23	62.65	53.99	53.99	53.99	52.62	57.98	57.98	57.98
Soy meal (%)	30.81	30.76	32.27	32.27	32.27	27.76	30.27	30.27	30.27
Fish Meal (%)	5.37	3	3	3	3	3	1	1	1
Soy oil (%)	-	-	7	-	3.5	-	7	-	3.5
Fish oil	-	-	-	7	3.5	-	-	7	3.5
Oyster (%)	1.41	1.4	1.42	1.42	1.42	1.3	1.39	1.39	1.39
DCP (%)	0.51	0.57	0.66	0.66	0.66	0.69	0.84	0.84	0.84
Salt (%)	0.25	0.32	0.32	0.32	0.32	0.32	0.35	0.35	0.35
Vit-MinP (%)	1	1	1	1	1	1	1	1	1
DL-Met (%)	0.26	0.23	0.25	0.25	0.25	0.17	0.18	0.18	0.18
L-Lys (%)	0.15	0.07	0.09	0.09	0.09	0.22	-	-	-
Analysis									
ME (Kcal/Kg)	2860	2874	3205	3211	3208	3225	3235	3241	3238
CP (%)	22.5	21.03	21	21	21	19	19	19	19
Crude Fat (%)	2.86	2.1	9.52	9.52	9.52	1.34	9.55	9.55	9.5
Linoleic a (%)	1.46	2.63	5.37	1.39	3.38	2.74	5.45	1.46	3.46
Ca (%)	0.95	0.9	0.9	0.9	0.9	0.85	0.85	0.85	0.85
Ava P (%)	0.475	0.45	0.45	0.45	0.45	0.425	0.425	0.425	0.425
Na (%)	0.152	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16
Lys (%)	1.368	1.23	1.23	1.23	1.23	1.2	1.02	1.02	1.01
Met (%)	0.662	0.6	0.6	0.6	0.6	0.48	0.48	0.48	0.48
Met + Cys (%)	1.035	0.95	0.95	0.95	0.95	0.8	0.8	0.8	0.8

*FO = Fish oil; SO = soybean oil; CP = crude protein; DCP = dicalcium phosphate; Met = methionine; Lys = lysine, and Cys = cystine.

al., 2000; Abas et al., 2004) and transition of omega-3 fatty acids into animal products (Grashorn, 1995; López-Ferrer et al., 1999, 2001; Özpinar et al., 2002; Kahraman et al., 2004).

It seems that the energy and fatty acid characteristics of the fats from animal and vegetable origin affect the blood characteristics of broilers (Miller et al., 1962; Siegel et al., 1995; Oayzdog and Aksit, 2003). There are some reports on the sex differences of blood cholesterol in birds fed dietary fat supplements (Demir and Ozturkcan, 1991; Swierczewska and Niemiec, 1995).

Hassan et al. (2007) in a study on broilers showed that blood total cholesterol was positively correlated with high density lipoprotein (HDL) and low density lipoprotein (LDL). Similarly, triglyceride was positively correlated with HDL and very low density lipoprotein (VLDL). These authors reported that abdominal fat weight was positively correlated with some serum biochemical indices. They concluded that abdominal fat in broilers is affected by the levels of serum biochemical indices.

The aim of the present study was to test the hypothesis that fish oil can affect abdominal fat pad deposition, serum lipoproteins, cholesterol and glucose concentrations of broiler chickens and that there is a correlation between abdominal fat pad and serum parameters.

MATERIALS AND METHODS

In this experiment, four hundred day-old Ross 308 broiler chicks of both sexes were assigned randomly to 16 floor pens with 25 chicks in each pen. Birds were managed in accordance with the guidelines of the Ross 308 manual. Three Isoenergetic and isonitrogenous diets containing: 0% fish oil + 7% soybean oil, 3.5% fish oil + 3.5% soybean oil and 7% fish oil + 0% soybean oil and another free oil diet with an inevitable lower metabolisable energy as control (Table 1) were formulated according to the Ross 308 manual. These four diets formed the treatments of a completely randomized design. All chicks were fed a commercial starter diet from 0 to 10 days and were allowed free access to water and food. Chicks were fed the experimental diets from 11 to 24 days (grower phase) and 24 - 42 days (finisher phase).

Live weights of birds were recorded at 42 days of age. Birds were deprived of feed 12 h before being weighed. At the end of the experimental period, 2 male birds per replicate pen were randomly selected. Apparent characteristics like bird's comb and wattle shape and body size and weight were used to distinguish male from

Table 2. Effects of dietary fat type and level on live weight and abdominal fat pad percent of broiler chickens.

Dietary fats	Live weight (g)	Abdominal fat pad %
Control	1955.6c	2.2a
7% SO	2266.9a	2.0b
3.5% SO + 3.5% FO	2175.6a	2.1a
7% FO	1758.1d	1.9b
SEM	123.3	0.11

female birds, then samples were selected randomly from the male group. Bird's sex was reconfirmed by the post-slather sexual gland examination.

The selected birds were subjected to feed withdrawal overnight permitting gut clearance, then 5 ml blood samples were taken from the wing vein of fasting chickens. Serum was harvested with centrifugation at 3000 rpm for 10 min and then the serum was frozen for future analysis of serum lipid and lipoprotein concentrations and stored at -20℃.

The birds then were killed via neck cutting. Abdominal fat pad (including fat surrounding gizzard, bursa of fabricius, cloaca and adjacent muscles) was removed and weighed individually. The percentage of abdominal fat weight was expressed as a ratio of body weight. Results of abdominal fat and dressing percentage were analyzed using every chick as a replicate.

Total serum cholesterol (free cholesterol + cholesterol esters) and glucose were assayed according to the manufacturer's recommenddations using a commercial enzymatic kit supplied by Man Co., Ltd in Iran. Samples were incubated for 5 min at 37℃, their absorbance were measured at 500 nm with a spectrophotometer. The triglyceride absorbance was determined at 546 nm according to the guidelines (Man Co., Ltd, Iran).

High-density lipoprotein cholesterol was detected enzymatically after precipitation of LDL and VLDL by heparin and manganese; their absorbance was read at 540 nm. Likewise, very low density lipoprotein cholesterol was estimated as [Triglycerides/5] (Friedewald et al., 1972). Low-density lipoprotein cholesterol was estimated using the Friedewald et al. (1972) equation [Low-density lipoprotein cholesterol = Total cholesterol – High-density lipoprotein cholesterol – Triglycerides/5] (Friedewald et al., 1972).

Correlation coefficients were computed between cholesterol, triglycerides, lipoprotein concentrations, abdominal fat weight and percentage of abdominal fat weight. The general linear model (GLM) of the SAS software (SAS institute, 1997) was used for statistical analysis of data. Differences were considered significant at $P < 0.05$ and means were compared by Duncan test.

RESULTS

Final body weight in birds fed soybean oil and soybean oil + fish oil diets were higher than control group ($P < 0.05$), but the birds fed fish oil diet showed a lower body weight than control group ($P < 0.05$). The abdominal fat percent in birds fed soybean oil and fish oil diets were lower ($P < 0.05$) than control group (Table 2).

Table 3 demonstrates the effects of dietary fat types on serum lipids fractions. Dietary fat inclusion did not affect the triglycerides (TG) and VLDL concentrations of broilers serum. Total cholesterol decreased in the serum of birds fed oil containing diets as it was significantly lower in the group receiving soybean oil diet, than the group fed with the diet containing soybean oil + fish oil ($P < 0.05$). The HDL concentration was influenced by dietary oil such that birds fed fish oil diet showed higher level of HDL than other treatments ($P < 0.05$).

In contrast to the serum HDL level, the serum LDL decreased in birds fed fish oil diet. The live weight of birds was positively correlated with glucose and LDL and negatively correlated with HDL concentrations of serum ($P < 0.01$) (Table 4). The abdominal fat pad percent was positively correlated with triglyceride, glucose, LDL and VLDL and a negative correlation was observed with HDL concentrations ($P < 0.01$) (Table 4).

Table 5 shows the correlation coefficients between serum parameters. Triglyceride, cholesterol and VLDL concentrations were positively correlated with each other and a negative correlation was observed between high-density lipoprotein and low-density lipoprotein ($P < 0.01$).

Serum glucose concentration exhibited a positive correlation with TG, cholesterol and VLDL ($P < 0.01$).

DISCUSSION

One obvious difference that distinguishes the present study from previous reports is the high fish oil dosage (up to 7%) included in the experimental diets. The most common administered level of fish oil in poultry diets is lower than 5%, because of eventual unfavorable fishy taint in meat. The higher dietary fish oil decreased abdominal fat pad deposition. This is in agreement with the well known effects of omega-3 fatty acids on fat deposition reduction in chickens (Pinchasov and Nir, 1992). Safamehr et al. (2008) used diets containing up to 3% fish oil and did not observe any significant reduction in abdominal fat pad, but they reported higher body weight gain in birds fed fish oil containing diets. In the present study, dietary fat type (fish oil, soybean oil or their combination) did not affect chicken's serum TG and VLDL concentrations. This is in agreement with the findings of Alparslan and Özdogan (2006) and Safamehr et al. (2008).

A decrease in total cholesterol in serum of the birds fed 7% soybean oil or 7% fish oil was observed but there was no reduction for birds fed diet containing intermediate dosage of this oils (3.5% soybean oil + 3.5% fish oil) compared to the oil free control diet. This unexpected observation needs more examinations. Alparslan and Özdogan (2006) and Safamehr et al. (2008) using

Table 3. Effects of dietary fat type and level on serum glucose and lipid fractions of broiler chickens.

Dietary fats	TG (mg/dl)	TCH (mg/dl)	Glu (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
Control	53.8	143.6[a]	125.4[a b]	95.7b[c]	26.3[a]	10.8
7% SO	48.6	109.1[c]	108.1[b]	87.3[c]	24.9[a]	9.7
3.5% SO + 3.5% FO	53.8	134.0[ab]	142.3[a]	106.6[b]	16.2[b]	10.8
7% FO	50.3	114.4[bc]	96.6[b]	120.6[a]	11.0[c]	10.1
SEM	4.2	10.3	15.3	8.1	3.8	1.3

[a – d] Values in the same column in each comparison group, with no common superscript differ significantly ($P < 0.05$).
FO = Fish oil; SO = soybean oil; control = oil free diet; TG = triglycerides; TCH = total cholesterol; Glu = glucose; HDL = high-density lipoprotein; LDL = low-density lipoprotein; VLDL = very low-density lipoprotein.

Table 4. Correlation coefficient matrix of serum parameters, live weight and abdominal fat pad deposition in broiler chickens.

Serum parameters	TG (mg/dl)	TCH (mg/dl)	Glu (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
live weight	-0.107	-0.087	0.471**	-0.752**	0.522**	-0.107
AFP	0.776**	0.869**	0.836**	-0.519**	0.648**	0.777**

** Correlation is significant at the 0.01 level (2-tailed).
FO = Fish oil; SO = soybean oil; control = oil free diet; TG = triglycerides; TCH = total cholesterol; Glu = glucose; HDL = high-density lipoprotein; LDL = low-density lipoprotein; VLDL = very low-density lipoprotein; AFP = abdominal fat pad.

Table 5. Correlation coefficient matrix of serum parameters in broiler chickens.

Serum parameters	TG	TCH	Glu	HDL	LDL	VLDL
TG	1**	-	-	-	-	-
TCH	0.964**	1**	-	-	-	-
Glu	0.8164**	0.759**	1**	-	-	-
HDL	0.135	-0.0567	-0.217	1**	-	-
LDL	0.053	0.285	0.226	-0.942**	1**	-
VLDL	1**	0.964**	0.816**	0.135	0.053	1**

** Correlation is significant at the 0.01 level (2-tailed). TG = Triglycerides; TCH = total cholesterol; Glu = glucose; HDL = high-density lipoprotein; LDL = low-density lipoprotein; VLDL = very low-density lipoprotein.

diets containing up to 4 and 3% fish oil, respectively, did not found any difference in serum total cholesterol concentration.

Chin et al. (1994) and Manilla et al. (1999) reported that omega-3 fatty acids content of fish oil reduce the VLDL concentrations in blood and this is accompanied with a reduction in LDL concentration and hepatic triglyceride synthesis. We found a dose relationship changes in serum HDL according to dietary fish oil level that is in agreement with previous reports (Alparslan and Özdogan, 2006; Safamehr et al., 2008).It seems that this increase in HDL concentration occurred because of the fish oil, which is rich in omega 3 fatty acids (Pike, 1999; Manilla et al., 1999). An increase in HDL content of serum following feeding the chickens with polyunsaturated fatty acid (PUFA) rich diets is reported by Ozdogan and Aksit (2003). They used sunflower oil as PUFA rich oil. According to Bachorik et al. (1991), in a healthy body, the level of LDL is low and the level of HDL is high. Grundy (1991) also noted that increasing serum HDL has beneficial effects on decreasing unfavorable outcomes of high blood cholesterol.

The reduced serum LDL in birds fed fish oil containing diets is in agreement with Safamehr et al. (2008), but Alparslan and Özdogan (2006) did not observe any difference for this parameter. The reduced glucose concentration in the serum of birds fed the higher dietary fish oil is reported by Safamehr et al. (2008).

In the current study, positive relationships between abdominal fat pad deposition and serum triglyceride, glucose, LDL and VLDL concentrations was found. Hassan et al. (2007) in a study on two chicken breeds, reported same positive correlation only with serum total cholesterol and LDL and on the other hand, there were negative correlations between abdominal fat pad and serum triglyceride, HDL and VLDL. These authors reported a whole positive correlation between these parameters in another chicken breed.

Hermier and Dillon (1992) showed that the development of adipose tissue in birds depends directly on the VLDL-TG level. According to Wahl et al. (1984), the positive relationships between LDL and other lipoprotein lipids became inversed in the presence of high triglyceride levels. Changes of serum triglycerides and HDL may be mediated by alterations in the activities of the key lipoprotein metabolizing enzymes, such as increased activity of hepatic lipase (Després et al., 1989), decreased or unchanged activity of post-heparin plasma lipoprotein lipase (LPL) (Pollare et al., 1991) and high or normal activity of adipose tissue LPL (Ong and Kern, 1989).

The results of the present study showed that high dietary fish oil may result in some unusual changes in body weight, abdominal fat pad deposition or serum lipids fraction when compared with intermediate dosage of fish oil. It seems that higher fish oil inclusion in broiler diets can result in some unfavorable reduction in live weight and a beneficial influence on abdominal fat reduction.

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