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Effect of feeding marine fishmeal on fatty acids profile, plasma lipid profile and production traits of layers

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Abstract

This study was conducted to find the effect of feeding marine fish (1 and 2%) on egg yolk fatty acids, blood cholesterol and egg quality in laying hens. Forty five laying hens (Hisex), 20 weeks old, were divided into three groups (15 birds per treatment). The diets were formulated to meet the requirement of egg production. Two formulae of diets were prepared by inclusion of marine fishmeal (1 and 2%). Results revealed that feeding fish meal at a concentration of 1 and 2% in a basal diet of laying hens for two months resulted in significant elevation of poly unsaturated fatty acids and omega-3 FA, contents of egg yolk compared to the control group with no effect on production traits except the egg number and egg shell thickness. Fish meal had significantly reduced plasma total lipids, triglycerides, cholesterol and low density lipoproteins, while high density lipoprotein was increased. The results showed that inclusion of 1 and 2% marine fish improved egg quality, egg yolk fatty acids and blood cholesterol profile in laying hens.

Keywords: Marine fish; egg quality; blood cholesterol; yolk fatty acids; layers

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Introduction

Dietary intake of omega-3 fatty acids decreases the risk of heart disease, provides an inhibitory effect on the growth of prostate, colon and breast cancer, and delays the loss of immunological functions. The n-3 fatty acids have hypocholesterolemic properties are important in foetal brain and retinal development (Lewis et al, 2000; Carrillo-Dominguez et al., 2005).

Docosahexaenoic acid (DHA) can strongly induce apoptosis in human MCF-7 breast cancer cells both *in vivo* and *in vitro*. The induction of apoptosis in these cells is selectively mediated via caspase 8 activation (Kang et al., 2010). DHA reduces the ability of J774A.1 cells to control *tuberculosis* in response to activation by modulation of IFNϒ receptor signalling and function, suggesting that n-3 PUFA-enriched diets may have a detrimental effect on host immunity to tuberculosis (Bonilla et al., 2010).

Several epidemiological studies suggest co-variation between seafood consumption and rates of mood disorders. Biological marker studies indicate deficits in omega-3 fatty acids in people with depressive disorders, while several treatment studies indicate therapeutic benefits from omega-3 fatty acids supplementation. A similar contribution of omega-3 fatty acids to coronary artery diseases may explain the described links between coronary artery diseases and depression.

Dietary manipulation of the n-3 fatty acid content of laying hen rations results in the production of eggs containing substandard amount of n-3 fatty acids, whereas, attempts to modify whole-egg cholesterol content to meet demands of health conscious consumers have been largely unsuccessful (Mary and Van elswyk, 1997).

Food typically considered being major contributor of PUFA (n-3) in the diets including, fish and other

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types of seafood. Other common dietary sources of n-3 PUFA include chicken, eggs, canola oil and soybean oil. Less common food products that are high in n-3 PUFA are wheat grass, fresh liquid lecithin, flaxseeds, flaxseeds oil and hemp seed oil. Other foods that provide smaller quantities of n-3 PUFA include green vegetables, whole milk and ground beef (Lewis et al., 2000). Fish omega-3 FAs (namely EPA and DHA) posses a lot of properties that can explain their positive impact on cardiovascular events have seen both in epidemiological and interventional studies. EPA and DHA differ in their ability to promote various effects of omega-3 FAs supplementation. Obviously, the two occur always together in natural sources–fish meals and fish oil. However, as highly purified EPA and DHA became available, evidence documenting individual effects of EPA and DHA has been accumulated (Vrablik and Zlatohlavek, 2009).

In general, fish oils are rich sources of omega-3FAs and poor sources of omega-6, and the contents of linoleic acid (LA) are also low. The FA profile of the different oils varies with the time, the processing method and the predominant fish species from which they were extracted (Alparsan and Ozdogan, 2005).

The aim of this paper was to find the effect of feeding marine fishmeal on fatty acids profile, plasma lipid profile and production traits of layers.

Materials and Methods

The experiment was conducted in the Central Veterinary Research Laboratories Centre (VRI), at Soba, from January to March, 2010. The duration of the experiment was two months. Birds were kept in wire cages. Each cage was of 1.5 meter in height, and 2.5 meter wide. The capacity of each cage was 15 birds. The cages were placed in an open poultry house.

Forty five laying hens (Hisex), 20 weeks old, were obtained from Animal Production Research Centre (Kuku) and were divided into three groups (15 birds per treatment). The diets were formulated to meet the requirement of egg production according to the directions of the National Research Council (1994). Two formulae diets were prepared by inclusion of marine fishmeal (1 and 2%).

Twenty kilograms of marine fish were brought from Port Sudan. Versa and their heads were removed, minced and dried at an open yard. Fish was subjected to proximate analysis, to determine its content of protein, fat, fibre, N.F.E and energy (AOAC, 1995). Table 1 shows the experimental and control diet composition, and Table 2 shows the nutritional composition of experimental and control diet.

Fifteen eggs from each group were collected randomly throughout the eighth week for egg yolk lipids profile determination. Eggs were broken, the yolk was separated and each two yolk were pooled together and placed into a glass container and stored at -20°C until analysis.

Three ml of blood were collected from seven birds of each group in EDTA coated vials. The samples were centrifuged at 3000 rpm, and plasma was transferred into plane vials. Plasma samples were stored at -20°C until analysis.

Lipids were extracted in chloroform-methanol (2:1 v/v) following method of Floch et al. (1957). Methyl esters of the lipid extract were prepared according to Wang et al. (2000). The analysis was performed using 2010 Shimadzu, Japan gas chromatograph, fitted with flame ionization detector (FID). Separation of fatty acids was achieved using DB-WAX column, serial number (US 6551263 H), of 0.25 um film thickness, 30 meter length and 0.25 mm inner diameter.

Fatty acids methyl esters were identified by comparison of retention times with standards, and expressed as percentage of methyl esters. The plasma lipid profile was determined using commercial kits by Unicam 8625 Spectrophotometer.

Fifteen eggs of each group were collected randomly on the last two days of the experiment, for determination of egg external and internal quality. The shell thickness was measured using a modified Starrett Model 1010 M thickness gauge, according to the method described by Anderson and Heckey (1972).

For yolk height measurement, a needle provided with a movable loop was dipped in the centre of the

Table 1: Experimental and control diet composition

Diet Composition%	Control Group (A)	Marine fish 2% (B)	Marine fish 1% (C)
Corn	62	67.4	67.8
Wheat hull	19	11.4	12
Ground cake	3.5	3.4	3.4
Concentrate	5	5	5
Calcium carbonate	9	10	10
Salt (NaCl)	0.125	0.125	0.125
Methionine	0.15	0.34	0.36
Lysine	0	0.15	0.15
Mycofix	0.1	0.1	0.1
Premix	0.1	0.1	0.1
Marine fishmeal	0	2	1

Table 2: Nutritional composition (%) of experimental and control diet calculated

Nutritional Parameter (%)	Control (A)	Marine fish 2% (B)	Marine fish 1% (C)
Protein	18	18.2	18
Either extract	5	3.1	3
Fiber	4	3.3	3.3
Lysine	0.86	0.87	0.84
Calcium	4.1	4	4
Phosphorous	0.31	0.64	0.62
Methionine + cystine	0.73	0.72	0.72
Energy (cal/kg)	2800	2750	2753

Table 3: Effect of feeding fishmeal on egg yolk fatty acids content (%)

Groups	Palmitic Acid	Stearic Acid	Oleic Acid	Linoleic Acid	Linolenic Acid	Arachidonic Acid	EPA	DPA	DHA
Control A	21. 77±4.49^{b}	1.02±1.42^{c}	35.11±8.35^{b}	0.00±1.25^{b}	0.00±0.82^{a}	0.00±0.13^{b}	0.00±0.17^{b}	0.00±0.12^{b}	0.00±0.03^{b}
Group B	30.7±4.95^{ab}	8.37±1.35^{a}	47.9±8.26^{ab}	9.16±1.27^{a}	0.43±0.43^{a}	0.17±0.45^{a}	1.94±0.57^{a}	0.67±0.42^{a}	0.51±0.13^{a}
Group C	33.28±4.93^{a}	5.63±1.31^{b}	52.13±8.35^{a}	8.91±1.23^{a}	0.00±0.63^{a}	0.04± 0.65^{ab}	0.00±0.05^{b}	0.00±0.04^{b}	0.00±0.33^{b}

Means in the same columns followed by the same letters are not significantly different at (P<0.05); Group A: control, Group B: 2% Fish meal, Group C: 1% Fish meal; Docosahexaenoic acid (DHA), Docosapenta-enoic acid (DPA) and Eicosapentaenoic acid (EPA)

Table 4: Effect of feeding 2 and 1% marine fish meal on plasma lipid profile (mg/dl)

Group	Total lipids	Cholesterol	Triglycerides	LDL-cholesterol	HDL-cholesterol
Control A	592.7±8.33^{a}	124.28±1.68^{a}	334.9±2.91^{a}	49.14±1.28^{a}	9.131.23^{a}
Group B	539.9±8.37^{c}	96.28±1.64^{c}	219.13±2.93^{c}	32.70±1.22^{c}	18.94±1.2^{a}
Group C	551.2±8.34^{b}	99.76±1.66^{bc}	227.61±2.91^{bc}	36.26±1.28^{b}	17.54±1.25^{bc}

Means in the same columns followed by the same letters are not significantly different at (P<0.05); Group A: control, Group B: 2% Fish meal, Group C: 1% Fish meal

Table 5: Effect of feeding 2 and 1% marine fish meal on egg external and internal quality

Group	Egg weight (g)	Egg size (cm^{3})	Shell thickness (mm)	Yolk weight (g)	Yolk height (mm)
Control (A)	52.98± 1.33^{b}	47.00± 2.53^{a}	0.091±6.38^{c}	13.59± 0.43^{a}	1.63± 0.05^{a}
Group (B)	56.46 ± 1.35^{a}	48.5± 2.57^{a}	0.105± 6.35^{b}	14.60± 0.45^{a}	1.62± 0.05^{a}
Group (C)	54.56±1.31^{ab}	50.00±2.54^{a}	0.109± 6.33^{a}	14.73±0.47^{a}	1.60±0.05^{a}

Means in the same columns followed by the same letters are not significantly different at (P<0.05); Group A: control, Group B: 2% Fish meal, Group C: 1% Fish meal

Table 6: The accumulative effect of feeding different diet types on egg yolk content of fatty acids (%)

Group	∑Poly unsaturated %	∑Omega 3%
Control(A)	0.00 ± 1.53^{c}	0.00 ± 0.63^{b}
Group (B)	12.91 ± 1.41^{a}	3.56 ± 0.51^{a}
Group (C)	8.91 ± 1.61^{b}	0.00 ± 0.71^{b}

Means in the same column followed by the same letters are not significantly different at (P<0.05); Group A: control, Group B: 2% Fish meal, Group C: 1% Fish meal

Table 7: The effect of feeding different diet types on egg production (%)

Group	First month	Second month
Control(A)	51.78 ± 16.42^{a}	84.49 ± 2.01^{b}
Group (B)	50.95 ± 16.52^{a}	94.52 ± 2.42^{a}
Group (C)	46.45 ± 16.96^{a}	93.57 ± 2.62^{a}

Means in the same column followed by the same letters are not significantly different at (P<0.05); Group A: control, Group B: 2% Fish meal, Group C: 1% Fish meal

yolk, the lower end of the loop was adjusted and the dipped portion of the needle was measured in centimetres on a scale. Egg and albumin weight were determined using digital scale (Shimadzu, electronic balance, type: BL-620S). Figure 2 shows the effect of feeding 1 and 2% marine fishmeal on egg production.

For yolk height measurement, a needle provided with a movable loop was dipped in the centre of the yolk, the lower end of the loop was adjusted and the dipped portion of the needle was measured in centimetres on a scale. Egg and albumin weight were determined using digital scale (Shimadzu, electronic balance, type: BL-620S).

Statistical Analysis

Statistical analysis was done using Analytical Software, 2008. Statistix 9 User's Manual, Analytical Software, Tallahassee, FL. P values less than 0.05 was considered to be statistically significant.

Results

As shown in Table 3, except linolenic acid, egg yolk fatty acids were significantly high in treated groups compared to control. Palmitic acid, oleic acid, linoleic acid was significantly higher in group fed 1% marine fish meal compared to the control. While stearic acid, DHA, EPA and DPA were significantly higher in 2% group compared to control and 1%.

Similarly, total lipids, cholesterol, triglycerides, LDH-cholesterol were significantly lower in birds fed 2% marine fish, while HDL cholesterol was significantly higher in the same group (Table 4).

Egg weight was significantly high in group fed 2% marine fish meal while shell thickness was significantly high in group fed 1% marine fish. Other parameters did not differ between control and treated groups (Table 5).

Percentage of poly unsaturated and omega 3 were significantly high in group fed 2% marine fish (Table 6). No significant change was observed in egg production percentage in first month, however, it was significantly higher in treated groups compared to the control (Table 7).

Discussion

Menhadin oil, fishmeal and marine algae are frequently used to enrich egg yolk with EPA, DPA and DHA (Gonzalez-Esquerra and Leeson, 2000). In this study, the layers of group B (fed 2% marine fishmeal), deposited high levels of DHA, DPA and EPA in their egg yolk. The significant levels of DHA, DPA and EPA, deposited in the yolk were also reported by Gonzalez-Esquerra and Leeson (2000).

Fishmeal showed a significant reduction in plasma total lipids, cholesterol, triglycerides and LDL-cholesterol concentrations compared to the control group. While the HDL-cholesterol level was significantly increased and these findings agreed with Al-Sultan (2005) who reported that feeding 1.5% and 3% of fish oil to laying hens resulted in low concentration of total lipids, cholesterol, triglycerides and LDL-cholesterol in plasma, while the concentration of HDL-cholesterol was significantly elevated.

Harris and Connor (1984) reported a reduction in total cholesterol, triglycerides and LDL-cholesterol levels in normal subjects fed diets containing fish oil as a source of omega-3 fatty acids. These authors justified the decreased level of LDL-cholesterol in human subjects to reduction in LDL-cholesterol synthesis, an increased fractional rate of catabobolism of LDL, or combination of both. Simopoulos (1991) reported that the effect of dietary omega-3 fatty acids on factors and mechanisms involved in the development of inflammation, atherosclerosis and immune diseases may be due to reduction in LDL-cholesterol, triglycerides and an increase in levels of HDL-cholesterol.

Egg production was significantly increased in treated groups compared to the control. This result agrees with what was reported by Al-Sultan (2005) who showed increased egg production percentage in response to feeding 1.5 and 3% fishmeal. The accumulative effect of feeding fishmeal on external and internal quality resulted in significantly high yolk weight compared to the control. This result disagreed with what was reported by Mary and Van (1997) who reported decrease in yolk weight in response to feeding fish oil or fishmeal. This contradictory result may be due to the different percentage of fishmeal used, or may be because of different birds strains.

Egg size and yolk thickness did not vary significantly between the treated and the control and this goes with what Al-Sultan (2005) reported that feeding fish oil had no effect on production parameters, except the number of egg produced. Egg weight was not significantly influenced by feeding fishmeal, except for group B, which was fed 2% marine fishmeal. Also a significantly high shell thickness agreed with what reported by Farrel (2002) who found that feeding 3% fishmeal increased eggshell thickness significantly. The difference in results with what was reported by Al-Sultan (2005) may be due to the differences of experimental environment, or diets composition, strains, or the percentages of additives.

The present study showed a significant increase in egg production in response to consuming 1% marine fish compared to the control group. Similar results were also found by Scheideler and Forning (1996). The same authors reported decreased eggshell thickness in birds fed flaxseed for eight month, these results agree with the findings of the present study.

In conclusion, the used percentage of marine fish improved the egg yolk fatty acids, cholesterol profile and egg quality.

Abbreviations

DHA= Docosahexaenoic acid, n-3= Omega 3, PUFA= Polyunsaturated fatty acid, EPA= Eicosapentaenoic acid or icosapentaenoic acid, FAs= Fatty acids, N.F.E= Nitrogen Free Extract

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