ISSN 1682-8356
ansinet.org/ijps

INTERNATIONAL JOURNAL OF

POULTRY SCIENCE

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International Journal of Poultry Science 9 (7): 656-663, 2010
ISSN 1682-8356

Effect of n-3 and n-6 Fatty Acid Supplemented Diets on Semen Quality in Japanese Quail (*Coturnix coturnix japonica*)

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Abstract: The present work aimed to compare the effect of different dietary oil sources on semen characteristics of quail males. Japanese quail males (21 per diet) were fed one of four treatment diets: diet containing sunflower oil (T1), flax oil (T2), corn oil (T3), or fish oil (T4) as the oil source. Birds were 6 weeks old at the beginning of experiment. Following two weeks of adaptation period, semen was collected twice a week fortnightly from each male to evaluate semen traits included in this study. First semen collection was used to evaluate ejaculate volume, sperm concentration, live in total sperm, live normal sperm, sperm quality factor and abnormal sperm, while the second semen collection was used after pooled the semen of each replicate (7 male each) for determine semen glucose, protein, Aspartate Aminotrnsferase (AST), Alanine Aminotransferase (ALT) and Alkaline Phosphatase (ALP). Results of this study revealed that fish oil group (T4) surpasses other treatment groups as regards all semen characteristics involved in this study followed by the results of flax oil (T2), whereas the worst results for these traits were recorded when the diets of quail males supplemented with sunflower oil (T1) and corn oil (T3). In conclusion, fish oil and flax oil supplemented diets can be used as efficient tool for improving reproductive performance of Japanese quail males.

Key words: Japanese quail, oil sources, semen quality

INTRODUCTION

Bird semen contains high proportions of Polyunsaturated Fatty Acids (PUFA), making the semen susceptible to lipid peroxidation, which could lead to deterioration of sperm within sperm storage tubules (Surai *et al*., 1998). Docosahexaenoic acid (C22:6 n3) is the predominant PUFA found in mammalian sperm (Lin *et al*., 1993), but docosatetraenoic acid (22:4 n6) is the primary PUFA in chicken sperm (Cerolini *et al*., 1997a). Lipid composition is a major determinant of the membrane flexibility required for flagella movement of sperm and also of the fusing properties of the membrane associated with the acrosome reaction and fertilization (Cerolini *et al*., 1997b). Prior research has indicated that high content of n3 or n6 PUFA in the chicken sperm membrane influence sperm function and that modification to dietary fatty acids can affect spermatozoa traits (Cerolini *et al*., 2000). Kelso *et al*. (1997a) reported that long chain PUFA of chicken sperm are related to sperm quality and fertilizing ability of the male breeder. Dietary manipulation of polyunsaturated composition of sperm has been successfully used to improve fertility (Blesbois *et al*., 1997). Kelso *et al*. (1996) indicated that when cockerels received either 5% fish oil or corn oil in their diet, the former gave significantly higher fertility rates (96%) than the latter (89%). Another study found that inclusion of 3% menhaden oil in the diets of broiler breeder males, improved semen quality and increased fertility in 2nd week post-insemination and also improved hatchability of total eggs laid (Hudson and Wilson, 2003). Kelso *et al*. (1997b) reported that small increase in the proportion of n3 fatty acids in the sperm phospholipids induced by supplementation the diet with alpha-linolenic acid resulted in higher fertility at 39 wk of age. Improvements in fertility were attributed to a reduction in the n6: n3 ratio of fatty acids in the spermatozoa membrane, which may alter the physical properties of the membrane or its resistance to peroxidative damage. In turn, fertilizing capacity of the sperm is increased. Bongalhardo *et al*. (2009) suggested that if fish oil is provided to cockerels during rearing, then improvement in fertility may be more pronounced and persist for longer durations. It appears that 22: 6 n3 present in mammalian spermatozoa performs an essential function in promoting optimal fertility, as marked reduction in the amount of this fatty acid in spermatozoa are associated with impaired sperm number, motility and fertilizing ability (Gasanovas, 1999). However, it should be noted that the compositional data of avian reproductive performance are almost wholly confined to certain poultry species (the chicken and the turkey) and that little information is available for another species of birds. Therefore, the aim of the present study was to determine whether the nature of fatty acids (and especially PUFAs) in the diet can affect the semen quality of Japanese quail.

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Table 1: Ingredients and chemical composition of the diet fed to quails

Ingredients (%)	Sunflower oil (T1)	Flax oil (T2)	Corn oil (T3)	Fish oil (T4)
Yellow corn	12.00	12.00	8.50	10.00
Wheat	47.70	47.70	51.50	50.00
Soybean meal	20.00	20.00	19.70	19.70
Protein concentrate*	10.00	10.00	10.00	10.00
Lime stone	7.00	7.00	7.00	7.00
Oil	3.00	3.00	3.00	3.00
Sodium chloride	0.30	0.30	0.30	0.30
Calculated content**				
Crude protein (%)	21.05	21.05	21.10	21.05
Metabolisable energy (Kcal/kg)	2888.00	2879.00	2881.00	2885.00
Total calcium (%)	3.60	3.60	3.60	3.60
Available phosphorus (%)	0.30	0.30	0.30	0.30
Methionine (%)	0.35	0.35	0.34	0.34
Lysine (%)	1.00	1.00	0.99	0.99
Cystine (%)	0.27	0.27	0.27	0.27

*Golden protein concentrate provided per kg: : 2500 ME/kg; 40% crude protein; 9% crude fat; 4.5% crude fiber; 9% calcium; 2.3% available phosphorus; 2.3% lysine; 1.25 methionine; 1.8% methionine + cystine; 100000 IU vit A; 10 mg vit B1; 100 mg vit B12; 20 mg vit K_3; 50 mg copper; 700 mg manganese; 2 mg selenium; 200 mg vit E; 0.5 mg biotin; 5 mg folic acid; 200 mg niacin; 80 mg pantothenic acid; 10 mg iodine; 25000IU vit D3; 500 mg iron; 10 mg cobalt; 600 mg zinc; 10 mg vit B6.
**Calculated composition was according to NRC (1994)

MATERIALS AND METHODS

Birds and treatments: A total of 84, 6 weeks old Japanese quail males were used in this study. Following two weeks of adaptation period on experimental conditions and treatment diets the birds were weighed to provide an equal live weight in all groups at the beginning of the experiment. The males, separated into 4 groups with 3 replicates per group containing 7 males each. For 14 weeks (including adaptation period) the quail males were fed diets containing 3% oil from sunflower (T1), flax (T2), corn (T3), or fish (T4). The males were allowed free access to food and water and housed in stainless wire cages with 7 males for each cage. Ingredients and chemical composition of diets were shown in Table 1 and the fatty acid composition of the oils used in this study is presented in Table 2. A regime of 14 h constant lighting and continuous ventilation were provided and all males were reared under uniform management conditions throughout the experimental period.

Semen quality traits: Semen was collected twice a week fortnightly from each male by using the procedure reported by Al-Daraji (2007a). Extruded semen was collected from the everted copulatory organ with the use of a small glass collector fitted with rubber tubing and a mouthpiece into a small calibrated tube enabling measurement of ejaculate volume exact to 0.01 ml. For proper semen collection, two persons were necessary. First semen collection was used to evaluate ejaculate volume, sperm concentration, live in total sperm, live normal sperm, sperm quality factor and abnormal sperm by using the methods indicated by Al Daraji (2007b). However, the second semen collection was used after pooled the semen of each replicate for determine semen glucose, protein, Aspartate Aminotransferase (AST), Alanine Aminotransferase (ALT) and Alkaline Phosphatase (ALP) by using the procedures mentioned by Al-Daraji (2007b).

Statistical analysis: Statistical comparisons were based on 21 semen samples (each sample derived from an individual bird), for each experimental period which represented 14 days, from each dietary group as regards ejaculate volume, sperm concentration, live in total sperm, live normal sperm, sperm quality factor and abnormal sperm. Statistical analysis of semen glucose, protein, AST, ALT and ALP values were based on the replicate pooled semen samples, for each experimental period, from each dietary group, with each pooled sample obtained from 7 males in each replicate. The data was assessed by analysis of variance using the General Linear Model method (SAS, 2000). Test of significance for the difference between means of different oil treatments was done by Duncan's multiple range test (Duncan, 1955).

RESULTS

Results of this study revealed that dietary supplementation with fish oil (T4) recorded the best results ($p<0.05$) concerning ejaculate volume, sperm concentration, live in total sperm, live normal sperm and sperm quality factor, followed by the results of flax oil (T2), whereas sunflower oil (T1) and corn oil (T3) groups recorded the worst results with respect to these traits during all periods of experiment. However, there was no significant difference between T1 and T3 regarding these traits (Table 3-7). As shown from Table 8, T4 surpasses other treatments in relation to abnormal sperm when recorded the lowest value for this trait followed by T2 group, while T1 and T3 groups exhibited the highest abnormal sperm values during the whole period of experiment. Moreover, there was no significant difference between T1 and T3 groups respecting this

Table 2: Fatty acid composition (%) of oils included in the diets of quails

Numeric name	Common name	TI	T2	T3	T4
C12:0	Lauric acid	-	-	-	0.090
C14:0	Myristic acid	0.06	0.12	0.06	5.41
C15:0	None	0.02	0.08	0.03	0.47
C16:0	Palmitic acid	6.25	6.0	11.01	14.05
C17:0	Margaric acid	0.03	0.11	0.09	1.73
C18:0	Stearic acid	3.58	2.5	1.91	2.87
C20:0	Arachidic acid	0.238	0.5	0.36	0.15
C21:0	None	0.008	0.01	0.01	0.04
C22:0	Behenic acid	0.587	0.23	0.15	0.02
C23:0	None	0.028	0.02	0.02	0.06
C24:0	Lignoceric acid	0.203	0.08	0.16	0.15
C14:1	Myristoleic acid	-	-	-	0.03
C15:1	None	0.01	0.01	-	0.19
C16:1	Palmitoleic acid	0.09	0.4	0.13	8.25
C17:1	None	0.04	0.03	0.04	0.36
C18:1 n9	Oleic acid	23.0	19.0	24.0	21.94
C20:1 n9	Gadoleic acid	0.255	0.28	0.36	11.22
C22:1 n9	Erucic acid	0.007	0.01	0.01	7.65
C24:1 n9	Nervonic acid	0.005	0.02	0.12	2.30
C18:3 n3	Alpha linolenic acid	0.108	57.29	1.26	0.50
C20:3 n3	None	0.025	0.05	0.03	0.05
C20:5 n3	Eicosapentenoic acid (EPA)	0.118	0.63	0.09	10
C22:6 n3	Docosahexaenoic acid (DHA)	0.012	0.0	0.02	10.73
C18:2 n6	Linoleic acid	65	12.18	60	1.02
C18:3 n6	Gamma linolenic acid	0.016	0.02	0.06	0.13
C20:2 n6	11, 14-Eicosadienoic acid	0.155	0.08	0.06	0.19
C22:2 n6	13, 16-Docosadienoic acid	0.155	0.003	0.001	0.38
Total of saturated fatty acids		11.0	9.65	13.8	25.04
Total of mono unsaturated fatty acids		23.40	19.75	24.66	51.94
Total of polyunsaturated fatty acids		65.58	70.55	61.52	23.0
Total of omega-3 fatty acids		0.26	58.27	1.4	21.28
Total of omega-6 fatty acids		65.32	12.28	60.12	1.72
Total of omega-6/total omega-3 fatty acids ratio		251.23	0.21	42.94	0.08

Table 3: Effect of dietary supplementation with different oils on ejaculate volume (ml) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	0.018±0.008c	0.021±0.009b	0.017±0.007c	0.025±0.008a
2	0.017±0.009c	0.022±0.005b	0.018±0.009c	0.027±0.009a
3	0.019±0.006c	0.021±0.007b	0.019±0.007c	0.027±0.008a
4	0.018±0.009c	0.023±0.008b	0.019±0.007c	0.029±0.008a
5	0.020±0.008c	0.025±0.006b	0.021±0.008c	0.033±0.009a
6	0.022±0.007c	0.026±0.007b	0.023±0.005c	0.036±0.008a
Mean	0.019±0.004c	0.023±0.008b	0.019±0.008c	0.029±0.007a

Each period represented 14 days T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 4: Effect of dietary supplementation with different oils on sperm concentration (x 10^6/ml) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	432.1±75.3c	593.7±111.0b	440.3±65.2c	703.7±99.5a
2	445.3±66.9c	599.9±95.7b	449.6±50.1c	725.6±117.9a
3	485.1±88.2c	598.7±90.1b	480.1±73.9c	789.5±90.7a
4	499.7±55.9c	603.8±88.9b	483.9±80.8c	798.3±103.1a
5	502.3±77.9c	625.9±91.7b	509.1±81.9c	815.2±119.2a
6	500.1±83.6c	688.3±90.0b	519.2±81.9c	839.7±93.8a
Mean	477.4±109.7c	618.3±127.6b	480.3±119.7c	778.6±133.6a

Each period represented 14 days T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 5: Effect of dietary supplementation with different oils on live in total sperm (%) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	81.7±4.19c	84.1±3.77b	82.3±3.55c	89.1±4.39a
2	81.9±3.23c	84.2±5.11b	82.6±4.14c	90.0±6.07a
3	82.5±2.15c	84.7±3.25b	82.9±2.29c	90.6±5.33a
4	82.3±3.95c	86.9±4.44b	83.0±5.05c	91.3±7.09a
5	83.9±4.05c	87.5±4.20b	84.2±3.97c	93.8±8.11a
6	85.0±3.97c	87.9±2.29b	85.7±2.11c	95.9±8.85a
Mean	82.8±9.03c	85.9±8.80b	83.4±5.55c	91.7±4.18a

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 6: Effect of dietary supplementation with different oils on live normal sperm (%) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	71.2±2.99c	74.1±6.65b	71.1±2.77c	78.7±3.19a
2	71.3±3.55c	74.2±7.33b	71.9±3.25c	79.5±4.25a
3	71.9±4.00c	74.5±5.88b	72.1±2.61c	80.1±3.39a
4	72.1±4.15c	75.8±3.99b	72.8±4.24c	81.0±6.05a
5	72.8±3.33c	77.0±4.81b	73.0±3.95c	83.1±5.57a
6	74.9±5.03c	77.5±3.55b	74.9±4.41c	85.2±3.99a
Mean	72.3±2.22c	75.5±4.49b	72.6±2.29c	81.2±6.33a

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 7: Effect of dietary supplementation with different oils on sperm quality factor (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	5.53±0.99c	9.23±1.71b	5.32±0.83c	13.84±1.85a
2	5.39±0.97c	9.79±2.01b	5.81±1.02c	15.57±2.09a
3	6.62±1.03c	9.36±1.85b	6.57±1.11c	17.07±3.01a
4	6.48±1.09c	10.52±2.13b	6.69±1.78c	18.75±2.93a
5	7.31±1.17c	12.04±1.99b	7.80±1.33c	22.35±3.37a
6	8.24±1.33c	13.86±2.18b	8.94±2.06c	25.75±5.16a
Mean	6.59±1.00c	10.80±1.53b	6.85±1.09c	18.88±4.11a

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 8: Effect of dietary supplementation with different oils on abnormal sperm (%) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	26.3±1.77a	22.1±2.06b	25.9±1.02a	17.3±3.00c
2	26.0±1.08a	22.0±1.97b	25.5±2.11a	15.9±1.15c
3	25.6±2.11a	21.5±2.19b	24.9±1.77a	14.5±1.07c
4	24.7±3.07a	20.1±1.85b	24.0±3.15a	13.0±1.35c
5	23.1±2.85a	19.7±2.16b	22.5±1.95a	10.1±0.97c
6	20.2±2.55a	16.5±1.29b	19.5±2.06a	7.0±1.17c
Mean	24.3±3.30a	20.3±4.06b	23.7±4.18a	12.9±3.31c

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

trait. Treatment quail males with sunflower (T1) or corn oil (T3) resulted in significant ($p<0.05$) increase in semen glucose, followed by the results of flax oil (T2), whereas fish oil group (T4) recorded the lowest means for this characteristics throughout the total period of this study (Table 9). Supplementation quail males ration with sunflower oil (T1) resulted in significant ($p<0.05$) increase in semen protein and ALT activity, followed by the results of corn oil (T3) and then flax oil (T2), while T4 (fish oil) recorded the lowest values in respect of these two traits during all periods of experiment (Table 10 and 12). The data presented in Table 11 indicated that

Table 9: Effect of dietary supplementation with different oils on semen glucose (mg/100 ml) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	25.3±1.07a	20.1±1.85b	24.9±2.30a	17.3±1.80c
2	25.0±2.09a	19.7±2.33b	24.0±1.87a	16.5±1.39c
3	24.2±1.11a	18.3±1.66b	23.5±2.55a	15.0±1.11c
4	23.7±1.75a	16.7±1.08b	22.8±1.95a	13.2±1.78c
5	22.5±1.87a	15.5±1.93b	22.0±1.99a	11.5±1.55c
6	21.3±1.65a	13.2±1.67b	20.5±2.33a	9.8±1.30c
Mean	23.6±3.15a	17.2±1.18b	22.9±2.45a	13.8±1.01c

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 10: Effect of dietary supplementation with different oils on semen protein (g/100 ml) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	0.34±0.04a	0.28±0.05c	0.31±0.01b	0.24±0.01d
2	0.33±0.03a	0.28±0.03c	0.30±0.03b	0.22±0.03d
3	0.33±0.05a	0.26±0.02c	0.30±0.03b	0.21±0.04d
4	0.32±0.03a	0.25±0.03c	0.28±0.05b	0.19±0.02d
5	0.31±0.02a	0.25±0.04c	0.29±0.01b	0.17±0.03d
6	0.31±0.01a	0.23±0.05c	0.28±0.01b	0.15±0.02d
Mean	0.32±0.05a	0.25±0.03c	0.29±0.04b	0.19±0.01d

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c,d]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 11: Effect of dietary supplementation with different oils on semen AST (Unit/10^8 sperm/minute) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	100.3±6.48a	63.7±2.91b	99.8±7.88a	59.2±3.12c
2	98.1±7.55a	73.1±3.46b	99.0±6.45a	57.0±4.65c
3	95.7±5.33a	72.3±4.94b	96.2±5.55a	55.2±5.15c
4	91.6±4.18a	70.6±5.95b	92.0±6.77a	50.8±3.95c
5	89.2±6.35a	61.5±6.15	89.1±7.95a	45.1±2.96c
6	85.1±4.87a	55.2±3.96b	85.4±6.56a	40.0±4.14c
Mean	93.3±5.60a	67.7±4.13b	93.6±8.83a	42.8±3.29c

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c]Values within rows followed by different letters differ significantly ($p<0.05$)

Table 12: Effect of dietary supplementation with different oils on semen ALT (Unit/10^8 sperm/minute) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	1.55±0.31a	0.53±0.29c	1.13±0.45b	0.41±0.02d
2	1.53±0.24a	0.49±0.18c	1.10±0.62b	0.40±0.08d
3	1.48±0.43a	0.45±0.16c	1.03±0.39b	0.35±0.07d
4	1.40±0.32a	0.41±0.19c	0.99±0.25b	0.27±0.05d
5	1.37±0.26a	0.40±0.25c	0.98±0.15b	0.20±0.09d
6	1.33±0.29a	0.33±0.20c	0.91±0.19b	0.13±0.06d
Mean	1.44±0.26a	0.43±0.32c	1.02±0.36b	0.29±0.03d

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c,d]Values within rows followed by different letters differ significantly ($p<0.05$)

adding sunflower oil (T1) or corn oil (T3) to the ration of birds resulted in significant ($p<0.05$) increase in semen AST activity, followed by the means of flax oil (T2), while on the contrary T4 group denoted the lowest values with relation to this trait during the entire period of study (Table 11). Table 13 shows semen ALP activity of treatment groups. During the whole period of experiment the means of semen ALP activity of T4 group birds had increased significantly ($p<0.05$) in comparison with other treatment groups, followed by the results of T2 group and then T3 group. Furthermore, the lowest values of this trait recorded by T1 group. The means of this trait for

Table 13: Effect of dietary supplementation with different oils on semen ALP (King Armstrong unit) (Mean±SE) of quail males

	Treatments			
Periods	T1	T2	T3	T4
1	25.3±1.12d	34.5±3.77b	29.8±3.17c	36.3±2.19a
2	25.7±2.00d	35.1±1.16b	30.6±1.95c	36.9±3.15a
3	26.9±3.19d	35.3±2.22b	31.9±3.85c	37.3±3.16a
4	27.6±2.85d	35.9±3.15b	33.2±5.22c	37.3±2.25a
5	29.3±2.65d	36.0±2.19b	34.6±4.19c	38.0±1.19a
6	30.5±3.19d	36.9±1.27b	35.2±3.77c	39.2±3.23a
Mean	27.5±4.03d	35.6±3.26b	32.5±2.29c	37.5±4.19a

Each period represented 14 days. T1: Sunflower oil; T2: Flax oil; T3: Corn oil; T4: Fish oil.
[a,b,c,d]Values within rows followed by different letters differ significantly ($p<0.05$)

the total period of study were 27.5, 35.6, 32.5 and 37.5 King Armstrong units for treatments T1, T2, T3 and T4, respectively.

DISCUSSION

Results of this study clearly indicated that dietary supplementation with fish oil resulted in significant improvement as regards all semen quality traits included in this experiment, followed by the results of flax oil, whereas on the other hand the worst results of semen quality traits were recorded by sunflower and corn oil treatments. Blesbois *et al*. (1997) concluded that the transfer of essential fatty acids from the diet to the semen is effective and this transfer may have biological effects on semen quality and fertilizing ability of semen. However, those researchers found that there was a clear influence of dietary lipids on spermatozoa fatty acid profile: the proportion of n-3 fatty acids in spermatozoa from males fed fish oil compared to corn oil was higher (9.6% vs. 4.3%) and that of n-6 fatty acids was lower (22.4% vs. 33.3%). Furthermore, the total n-6/n-3 of the spermatozoa were 7.6 and 2.3 and of the seminal plasma was 154 and 2.8 for corn oil and fish oil, respectively. Hudson and Wilson (2003) reported that providing fish oil to broiler breeder males throughout their life may be a simple means to maintain fertilizing ability of spermatozoa of these birds. Kelso *et al*. (1997a,b) suggested that the small increase in the proportion of n-3 fatty acids in the sperm phospholipids induced by enriching the diet with alpha-linolenic acid is associated with significant improvement in semen quality of cockerel. Cerolini *et al*. (2000) reported that treatment the roosters with fish oil as a source of omega-3 fatty acids resulted in improvement in semen quality as compared with soybean or evening primrose oils as sources of omega-6 fatty acids. Bongalhardo *et al*. (2009) indicated that diets containing lipids from different sources would differentially modify the lipid contents of membranes from sperm heads and bodies, and they also found high correlations between these changes in lipid content and sperm concentration. Safarinejad *et al*. (2009) reported that fertile men were found to have higher blood and sperm levels of all three omega-3 fatty acids. Furthermore, infertile men had significantly higher blood ratios of omega-6 to omega-3 fatty acids. Cerolini *et al*. (2006) denoted that best sperm quality in n-3 rich sperm was found supplying 200 mg vitamin E/kg of feed to the broiler breeder males and in contrats in n-6 rich sperm supplying 300 mg vitamin E/kg. Zaniboni and Cerolini (2009) indicated the enrichment of turkey spermatozoa with n-3 long chain PUFA and vitamin E by dietary treatment prevent the negative effect of storage on sperm quality and sensitivity to induced *in vitro* peroxidation; however, it was efficient in prevent the increase of sperm death that occurred during liquid storage. The number of spermatozoa per ejaculate decreased by 50% between 26 weeks and 60 weeks of age in birds fed the maize oil diet. This age-related decrease in the number of spermatozoa was almost completely prevented by feeding the birds with the oils enriched in either 22: 6n-3 or 20: 4n-6. However, testis mass at 60 weeks of age was approximately 1.5 times greater in birds given of the tuna orbital and arasco oil diets compared with those given the maize oil diet (Surai *et al*., 2000). Estienne *et al*. (2008) reported that the number of sperm was increased and some of characteristics of sexual behaviour were altered, in boars fed a diet supplemented with omega-3 fatty acids. Waterhouse *et al*. (2006) suggested that male-to-male differences in sperm survival rate after freezing and thawing may be partly related to the amount of long- chain PUFA in the sperm plasma membranes. Al-Daraji (2001b) concluded that the highly significant negative correlation between numbers of spermatozoa and glucose concentration in seminal plasma suggests the utilization of glucose by spermatozoa. Al-Daraji (2002) indicated that spermatozoa utilized the glucose in their metabolism and postulated that glucose could be formed from glycogen and/or oligosaccharides by enzymatic break down of these products. These findings are in agreement with the results of this study, where the treatments that recorded the highest values of sperm concentration (T4 and T2) recorded at the same time the lowest values of semen glucose (Table 4 and 9). As shown from the results of this study T4 and T2 groups

exhibited the lowest numbers of abnormal sperm and the lowest concentration of semen protein (Table 8 and 10). Thurston (1976) has shown that the number of abnormal germinal cells and spermiophages present in turkey semen increases as the seminal protein concentration increased. Thus, the reduction in reproductive performance may be due to increased numbers of abnormal spermatozoa and spermatids in semen with high seminal plasma protein. Thurston *et al.* (1992) indicated that the level of seminal plasma protein can be used as a predictor of fertility and hatchability, since it was found highly significant negative correlation between seminal plasma protein concentration and fertility, hatchability of fertile eggs and hatchability of total eggs. However, they concluded that the individual determination of seminal plasma protein concentration can be used as a tool for determine roosters with low reproductive performance and the semen quality of roosters can be improved by selected them on the basis of lower seminal plasma protein concentration. Al-Daraji (2001a) reported that mean numbers of the spermatozoa exhibiting progressive motility and the mean germ cells concentration showed a highly significant negative correlation with the total seminal plasma protein content both in fresh and frozen-thawed semen samples.

Furthermore, it is widely accepted the number, viability, motility, survival and storage properties of spermatozoa are influenced by seminal plasma proteins (Al-Daraji *et al.*, 2001). The Lowest AST and ALT activities were obtained in semen of T4 and T2 groups. However, these treatments had best results of live in total sperm, live normal sperm and sperm quality factor and lowest number of abnormal sperm (Table 5, 6, 7 and 8). When sperm cell membrane damaged, AST and ALT enzymes are released into the extracellular medium (Al-Daraji *et al.*, 2002a). Al-Daraji *et al.* (2002b) reported significant correlation between seminal plasma AST and ALT activities following cellular disruption. Brown *et al.* (1971) examined several enzymes and selected AST and ALT release as the best indicator of cellular damage. Buckland (1971) suggested that the observed increase in AST and ALT activities of seminal plasma and semen during storage may be due to structural instability of the sperm. Al-Daraji (2001a) indicated that the ALT activity in seminal plasma was very weak as compared to the AST activity. Al-Daraji *et al.* (2000) found positive correlation between activities of AST and ALT in seminal plasma and percentages of dead and abnormal spermatozoa. Differences in semen ALP activity for different treatments included in this study closely resembled differences in spermatozoa liveability and concentration. A higher spermatozoa liveability and concentration were noticed in semen of T4 and T2 groups, which support high semen ALP activities, as compared with T1 and T3 groups, in which ALP activities were lowest (Table 4, 5, 6, 13). Al-Daraji *et al.* (2002b) found positive correlation between ALP activity and spermatozoa liveability and concentration. Al-Daraji *et al.* (2001) reported that both of alkaline and acid phosphatase are involved in the metabolism of spermatozoa *via* the hydrolysis of carbohydrates. Al-Daraji *et al.* (2002a) found highly significant positive correlation between the amount of ALP in the seminal plasma and the number of spermatozoa per ejaculate. An important consideration is the potential interaction of PUFA or their derived eicosanoids with the hypothalamo-pituitary-gonadal axis and the hormonal control of spermatogenesis (Etches, 1996).

Conclusion: The results of the present study show that adding fish and flax oils to the diets of quail males resulted in significant improvement with respect to semen quality traits included in this study in comparison with sunflower and corn oils. Therefore, fish and flax oils can be used as a beneficial tool for improving productive performance of quail males by inclusion these oils in their diets.

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