



Dietary inclusion of flaxseed oil, fish oil and their combinations on production performance and egg quality traits in White Leghorn hens[#]

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Abstract

A total of 120, 28-week-old White Leghorn hens were randomly selected to study the effect of dietary inclusion of flaxseed oil (FSO), fish oil (FO) and their combinations on production performance and egg quality traits in White Leghorn hens and the birds were divided into six treatments that had four replicates of five birds each. The experimental groups were fed with control diet and diet containing 5.0 per cent FSO, 5.0 per cent FO, 2.5 per cent FSO + 2.5 per cent FO, 3.5 per cent FSO + 1.5 per cent FO and 1.5 per cent FSO + 3.5 per cent FO. The experiment was conducted from 29 to 40 weeks of age comprising three, 28-day periods. The obtained data on statistical analysis, did not reveal any significant difference in body weight, egg production, feed intake, feed conversion ratio, egg weight and egg quality traits of White Leghorn hens among all the treatment groups at all the periods of age.

Keywords: Flaxseed oil, fish oil, production performance, egg quality

The changes in food habits and life style of people have led to an increase in the incidence of heart failure, hypertension and diabetes. As a result, nowadays consumers are more aware of

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what they eat. The egg is accepted world-wide and is considered as a complete food that contains all the essential nutrients such as protein, fat, vitamins and minerals (Miranda *et al.*, 2015).

Designer eggs have high demand because of the consumer's willingness to purchase them owing to specific nutritional qualities over and above that of regular eggs (Megha *et al.*, 2021). Enrichment of egg by incorporating omega-3 fatty acid sources into the layer diet is getting popular in order to get the benefits of omega-3 fatty acid. Major omega-3 fatty acid sources are flaxseed oil and fish oil. The flaxseed oil contains a greater amount of short-chain alpha-linolenic acid, which is the precursor of long-chain polyunsaturated fatty acids (PUFA) like eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Fish oil contains high levels of EPA and DHA (Omar *et al.*, 2014). However, studies reported that dietary inclusion of these omega-3 fatty acid sources reduced the body weight (Soliman and El-Afifi, 2020), feed intake (Ebeid *et al.*, 2008) and egg weight (Dong *et al.*, 2018) of birds and also increased egg production (Promila *et al.*, 2017a) and yolk colour score (Saleh, 2013). The enrichment of omega-3 fatty acid can be done in egg by feeding the birds with ingredients rich in such fatty acids without affecting the production and egg quality parameters (Valavan *et al.*, 2013).

Hence, the present study was aimed at evaluating the influence of dietary inclusion of flaxseed oil, fish oil and their combinations on production performance and egg qualities of

White Leghorn hens to serve as a platform for producing omega-3 fatty acid enriched eggs in future.

Materials and methods

The experiment was carried out at All India Coordinated Research Project (AICRP) on Poultry for eggs, Mannuthy, Thrissur, Kerala, India.

Experimental birds, layout and diets

A total of one hundred and twenty, 28-weeks old White Leghorn hens housed in individual cages were reared from 29 to 40 weeks of age. They were randomly distributed in a complete randomised design with six treatments that had four replicates of five birds each. The experimental groups were fed with control diet formulated using maize and soya bean meal as the major ingredients as per BIS (2007) specifications (T1), diet containing 5.0 per cent FSO (T2), 5.0 per cent FO (T3), 2.5 per cent FSO + 2.5 per cent FO (T4), 3.5 per cent FSO + 1.5 per cent FO (T5) and 1.5 per cent FSO + 3.5 per cent FO (T6). The birds were fed with *ad-libitum* feed and water and provided with a photoperiod of 16 h. The ingredient composition of experimental diets is presented in Table 1 and the chemical composition of experimental diets (Table 2) were analysed as per AOAC (2016).

Additional supplements (g/100 kg feed) vitamin premix and DL- Methionine – 50g, Toxin binder and liver tonic- 100g, Choline chloride- 200g, Trace mineral mixture and sodabicarb- 150g,

Table 1. Ingredient composition of experimental diets, per cent

Ingredient	T1	T2	T3	T4	T5	T6
Maize	55.50	37.55	35.80	36.20	37.50	35.80
Wheat Bran	2.60	11.00	11.75	11.35	10.75	11.8
De-oiled rice bran	5.00	10.00	12.30	11.80	11.30	11.85
Soya bean meal	26.10	25.80	24.50	24.90	24.80	24.90
Calcite powder	4.00	4.00	4.00	4.00	4.00	4.00
Dicalcium phosphate	1.40	1.40	1.40	1.40	1.40	1.40
Shel Grit	5.00	5.00	5.00	5.00	5.00	5.00
Salt	0.40	0.25	0.25	0.25	0.25	0.25
Fish oil	0.00	0.00	5.00	2.50	1.50	3.50
Flaxseed oil	0.00	5.00	0.00	2.50	3.50	1.50
Total	100	100	100	100	100	100

Table 2. Chemical composition of experimental diets, per cent

Composition (%)	T1	T2	T3	T4	T5	T6
Dry matter	89.36	89.98	89.88	89.68	89.71	89.80
Total ash	14.63	17.07	16.82	17.65	18.01	18
Crude protein	19.66	19.26	19.80	19.32	19.73	19.55
Crude fibre	2.29	2.68	2.98	2.54	2.89	2.89
Ether extract	2.85	6.76	6.70	6.04	6.21	7.64
Acid insoluble ash	0.13	0.87	0.54	0.82	0.62	0.75
NFE	60.57	54.23	53.70	54.45	53.16	51.92
Calcium	3.04	3.45	3.38	3.27	3.82	3.72
Phosphorus	0.63	0.52	0.80	0.58	0.68	0.71
ME (kcal/kg)*	2603.40	2603.00	2603.00	2602.00	2608.00	2604.00

*Calculated Value

The individual body weight and egg weight at 28, 32, 36 and 40 weeks of age, egg production from 29 to 40 weeks of age and feed intake at 28 days interval and egg quality traits at 32, 36 and 40 weeks of age were recorded. From the above data, replicate-wise feed intake per bird per day, hen housed and hen day egg production percentage and replicate-wise feed conversion ratio (kilogram of feed consumed per dozen egg produced) for each 28-day periods and cumulative period were calculated. The eggs collected from individual hens in each treatment group for the last three consecutive days of each 28-day period were used to measure external and internal qualities like shape index, shell thickness, yolk colour, yolk index, yolk weight, albumen index, albumen weight and Haugh unit score. The length and breadth of egg, diameter yolk and breadth and length of thick albumen were measured using a Vernier caliper. The height of albumen and yolk was measured using Ame's tripod stand micrometer. Yolk colour was measured using Roche yolk colour fan. The shell thickness was measured using screw gauge. The shape, albumen and yolk indices and Haugh unit score were calculated as per Arsha (2019).

The formula applied for calculating shape, albumen and yolk indices and Haugh unit score are listed below.

$$\text{Shape index} = \frac{\text{Breadth of egg (mm)}}{\text{Length of egg (mm)}} \times 100$$

$$\text{Yolk index} = \frac{\text{Height of yolk (mm)}}{\text{Diameter of yolk (mm)}} \times 100$$

$$\text{Albumen index} = \frac{\text{Height of thick albumen (mm)}}{\text{Average of breadth and length of thick albumen (mm)}} \times 100$$

$$\text{Haugh unit} = 100 \times \log (H - 1.7W^{0.37} + 7.6)$$

Where, H stands for height of thick albumen (mm) and W stands for weight of egg (g).

Egg was broken and yolk was carefully separated and weighed to the nearest gram. Albumen was scrapped by a scraper in to a petridish and weighed.

Statistical analysis

The data of present study were statistically analysed with one-way ANOVA using SPSS version 24.0.

Results and discussion

There was no significant difference in body weight of White Leghorn layers fed with diet containing flaxseed oil, fish oil and their combinations up to 5.0 per cent level at 28, 32, 36 and 40 weeks of age (Table 3). The result was in accordance with the findings of Ceylan *et al.* (2011) and Valavan *et al.* (2013). However, Saleh (2013) found significantly ($p < 0.05$) higher body weight in hens fed with a diet containing fish oil at 3.5 per cent level compared to control.

The results depicted in Table 4 showed that there was no significant difference in hen-housed egg production percentage of birds during the entire experiment period (29-40

Table 3. Mean (\pm SE) body weight of White Leghorn layers in different dietary treatments at four-week intervals, g

Age in weeks	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
28	1396.50 \pm 24.78	1406.00 \pm 36.91	1401.80 \pm 44.07	1384.85 \pm 32.47	1419.85 \pm 36.63	1388.50 \pm 28.65	0.984
32	1431.75 \pm 26.70	1429.50 \pm 33.15	1435.25 \pm 41.01	1416.84 \pm 22.88	1440.50 \pm 28.60	1407.75 \pm 34.35	0.980
36	1438.00 \pm 26.39	1426.32 \pm 35.08	1426.75 \pm 43.84	1424.21 \pm 15.20	1460.56 \pm 34.79	1428.42 \pm 30.34	0.971
40	1422.50 \pm 28.04	1379.73 \pm 44.10	1486.67 \pm 45.70	1462.63 \pm 28.00	1383.82 \pm 91.88	1411.58 \pm 36.23	0.58

week period) and that in Table 5 revealed that the difference in mean per cent hen day egg production (HDEP) was not significant among the White Leghorn layers fed with diet containing flaxseed oil, fish oil and their combinations at all periods except during 33-36 weeks when the mean per cent HDEP was significantly ($p < 0.05$) higher in hens fed with control diet and diet containing 3.5 per cent and above flaxseed oil added group. The HDEP of hens fed with a diet containing 5.0 per cent fish oil and 2.5 per cent each flaxseed oil and fish oil was intermediate. The results is in agreement with the results

obtained by Omar *et al.* (2014), Ebeid (2011) by feeding different levels of flaxseed oil, fish oil and their combinations and Arsha (2019) and Saleh (2013) by feeding different levels of fish oil. On the other hand, Promila *et al.* (2017) observed significantly ($p < 0.05$) higher hen day egg production in 2.5 per cent flaxseed oil added group compared to control group.

The feed intake of birds was significantly ($p < 0.05$) lower in groups fed with both flaxseed oil and fish oil during the first two 28-day period that might be due to the sudden

Table 4. Mean (\pm SE) hen-housed egg production of White Leghorn layers in different dietary treatments at four-week intervals, per cent

Period (in age)	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
29-32 wk	96.96 \pm 1.11	95.54 \pm 1.42	89.29 \pm 4.05	92.86 \pm 3.78	96.25 \pm 1.08	95.18 \pm 1.58	0.273
33-36 wk	96.07 \pm 1.29	96.25 \pm 1.20	88.21 \pm 4.71	91.17 \pm 4.56	96.07 \pm 1.66	85.15 \pm 4.24	0.075
37-40 wk	81.43 \pm 4.79	80.45 \pm 4.87	71.43 \pm 6.64	82.89 \pm 5.82	75.40 \pm 6.86	79.89 \pm 5.71	0.730
Overall mean (29-40 wk)	91.49 \pm 1.77	89.40 \pm 2.44	82.98 \pm 4.53	86.07 \pm 5.20	86.73 \pm 3.13	83.99 \pm 4.14	0.592

Table 5. Mean (\pm SE) hen-day egg production of White Leghorn layers in different dietary treatments at four-week intervals, per cent

Period (in age)	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
29-32 wk	96.96 \pm 1.11	95.54 \pm 1.42	89.29 \pm 4.05	93.69 \pm 3.49	96.25 \pm 1.08	95.18 \pm 1.58	0.268
33-36 wk	96.07 ^a \pm 1.29	96.40 ^a \pm 1.09	88.21 ^{ab} \pm 4.71	91.17 ^{ab} \pm 4.56	97.68 ^a \pm 0.75	85.15 ^b \pm 4.24	0.038 [*]
37-40 wk	83.24 \pm 3.89	80.45 \pm 4.87	71.57 \pm 6.60	82.89 \pm 5.82	81.49 \pm 5.42	84.33 \pm 3.80	0.529
Overall mean (29-40 wk)	92.54 \pm 1.26	90.97 \pm 1.87	83.07 \pm 4.52	88.92 \pm 3.93	91.47 \pm 2.18	85.65 \pm 3.12	0.200

*Means bearing different superscripts within a row differ significantly ($p < 0.05$)

change in feed which got adjusted during the later period and there was no significant difference in feed intake of birds during the overall period (Table 6). The result was in accordance with the results of Lawlor *et al.* (2010), Ebeid (2011) and Omar *et al.* (2014) by feeding different levels of flaxseed oil and fish oil. Contrary to the present finding, significantly ($p<0.05$) lower feed consumption was observed by feeding fish oil in laying hens (Saleh, 2013; Arsha, 2019).

There was no significant difference in feed conversion ratio (FCR) of birds during all the three periods and also in entire experiment period from 29-40 weeks (Table 6). The present

finding was in accordance with that of Ceylan *et al.* (2011), Valavan *et al.* (2013) and Omar *et al.* (2014) by feeding flaxseed oil, fish oil and their combinations and Arsha (2019) by feeding fish oil in layers. Contrary to the present finding, Soliman and EL-Alfifi (2020) reported significantly ($p<0.05$) better FCR in hens fed with 3.0 per cent flaxseed oil compared to control.

There was no significant difference in egg weight of hens at 28, 32, 36 and 40 weeks of age as presented in Table 7. The present finding is in line with the result obtained by Ebeid (2011) and Omar *et al.* (2014) by feeding laying

Table 6. Mean (\pm SE) daily feed consumption of White Leghorn layers in different dietary treatments at four-week intervals, g

Period (in age)	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
	Feed consumption (g/bird/day)						
29-32 wk	99.01 ^a ±0.41	94.84 ^b ±0.11	94.62 ^b ±0.43	95.48 ^b ±1.48	94.09 ^{bc} ±0.94	91.67 ^c ±1.38	0.001 [*]
33-36 wk	102.93 ^a ±0.79	97.26 ^b ±1.60	97.21 ^b ±1.90	101.51 ^{ab} ±1.44	99.42 ^{ab} ±1.41	97.49 ^b ±1.07	0.04 [*]
37-40 wk	101.84 ±1.27	94.63 ±6.28	90.36 ±3.91	97.98 ±7.18	96.65 ±4.28	95.26 ±5.22	0.724
Overall mean (29-40 wk)	101.26 ±0.76	95.58 ±2.19	94.06 ±1.95	98.32 ±2.49	96.72 ±1.66	94.8 ±1.60	0.122
	Feed conversion ratio						
29-32 wk	1.18 ±0.02	1.15 ±0.01	1.23 ±0.04	1.18 ±0.04	1.13 ±0.01	1.11 ±0.01	0.05
33-36 wk	1.24 ±0.03	1.17 ±0.02	1.28 ±0.06	1.3 ±0.06	1.18 ±0.003	1.38 ±0.11	0.165
37-40 wk	1.46 ±1.17	1.47 ±0.09	1.6 ±0.20	1.42 ±0.05	1.56 ±0.24	1.42 ±0.03	0.947
Overall mean (29-40 wk)	1.28 ±0.03	1.23 ±0.05	1.34 ±0.07	1.29 ±0.04	1.24 ±0.04	1.29 ±0.04	0.643

*Means bearing different superscripts within a row differ significantly ($p<0.05$)

Table 7. Mean (\pm SE) egg weight of White Leghorn layers in different dietary treatments at four-week intervals, g

Age in weeks	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
28	46.29 ± 0.68	46.20 ± 0.40	46.61 ± 0.37	48.53 ± 0.54	47.02 ± 0.52	47.36 ± 0.39	0.631
32	47.30 ± 0.80	47.97 ± 0.67	48.04 ± 0.71	49.89 ± 0.82	47.75 ± 0.71	47.60 ± 0.82	0.264
36	47.76 ± 0.70	47.88 ± 0.65	48.87 ± 0.74	49.56 ± 0.68	47.87 ± 0.52	47.82 ± 0.76	0.319
40	48.41 ± 0.68	49.29 ± 0.69	49.43 ± 0.84	49.59 ± 0.63	50.09 ± 0.65	48.47 ± 0.95	0.571
Overall mean (28-40)	47.73 ± 0.62	48.18 ± 0.61	48.75 ± 0.70	47.32 ± 2.55	48.16 ± 0.53	47.63 ± 0.69	0.969

hens with the diet containing fish oil, flaxseed oil and their combinations. Similar result was observed by Celebi and Utlu (2006) and Duan *et al.* (2022) by feeding flaxseed oil and Arsha (2019) by feeding fish oil. On contrary to the present study, significantly ($p<0.05$) higher egg weight was obtained by Mariod *et al.* (2015) and Brown *et al.* (2018) by feeding fish oil and significantly ($p<0.05$) lower egg weight was obtained by Dong *et al.* (2018) by feeding fish oil.

There was no significant difference in egg quality traits *viz.*, shape index, shell

thickness, yolk colour score, yolk index, yolk weight, albumen index, albumen weight and Haugh unit score of eggs from birds among all treatments at all ages as presented in Table 8. The result of present study was supported by the findings of Omar *et al.* (2014), Valavan *et al.* (2013) and Ebeid (2011) by feeding different levels of flaxseed oil and fish oil. On the other hand, Saleh (2013) reported significantly ($p<0.05$) higher yolk colour score in fish oil added groups compared to control and Lawlor *et al.* (2010) found significant ($p<0.05$) reduction in yolk weight of hens fed with different levels of fish oil in laying hens.

Table 8. Mean (\pm SE) egg quality traits of White Leghorn layers in different dietary treatments at four-week intervals

Age in weeks	Treatment groups						p-value
	T1	T2	T3	T4	T5	T6	
	Shape index						
32	75.07±0.007	74.65±0.006	73.79±0.007	73.22±0.004	73±0.007	74.73±0.005	0.084
36	75.65±0.018	75.49±0.006	73.62±0.008	74.73±0.007	72.54±0.008	74.24±0.004	0.238
40	74.41±0.007	75.25±0.006	75.16±0.10	74.18±0.006	74.17±0.007	74.95±0.007	0.826
	Shell thickness (mm)						
32	0.347±0.003	0.347±0.006	0.344±0.005	0.336±0.007	0.345±0.006	0.351±0.01	0.728
36	0.363±0.003	0.362±0.003	0.355±0.004	0.357±0.004	0.353±0.002	0.359±0.003	0.145
40	0.331±0.006	0.323±0.007	0.317±0.006	0.316±0.006	0.331±0.008	0.316±0.009	0.436
	Yolk colour score						
32	5.58±0.08	5.3±0.16	5.49±0.12	5.37±0.09	5.47±0.12	5.22±0.16	0.376
36	4.76±0.07	4.48±0.10	4.47±0.10	4.67±0.08	4.74±0.07	4.64±0.11	0.088
40	4.88±0.19	4.58±0.15	4.44±0.20	4.63±0.19	4.14±0.16	4.24±0.21	0.078
	Yolk index						
32	0.437±0.005	0.418±0.006	0.429±0.004	0.425±0.004	0.433±0.005	0.427±0.006	0.114
36	0.432±0.004	0.416±0.007	0.424±0.007	0.416±0.007	0.413±0.006	0.422±0.005	0.213
40	0.418±0.004	0.412±0.008	0.396±0.005	0.398±0.01	0.403±0.008	0.397±0.007	0.156
	Yolk weight (g)						
32	13.22±0.19	13.4±0.23	13.29±0.29	13.96±0.28	13.46±0.19	13.61±0.18	0.257
36	13.71±0.22	14.13±0.26	13.98±0.40	14.22±0.16	13.83±0.27	13.42±0.26	0.336
40	13.87±0.22	14.38±0.27	14.43±0.37	14.21±0.17	14.34±0.28	13.88±0.25	0.455
	Albumen index						
32	0.096±0.004	0.09±0.003	0.089±0.002	0.089±0.002	0.087±0.004	0.089±0.004	0.373
36	0.101±0.006	0.09±0.005	0.091±0.005	0.089±0.004	0.089±0.005	0.104±0.006	0.175
40	0.088±0.007	0.076±0.005	0.082±0.004	0.08±0.005	0.081±0.003	0.088±0.006	0.552
	Albumen weight (g)						
32	27.01±0.47	26.9±0.46	26.74±0.45	27.86±0.50	27.26±0.33	26.73±0.44	0.503
36	27.43±0.50	27.03±0.44	27.6±0.77	27.82±0.57	26.7±0.35	26.99±0.37	0.671
40	26.52±0.54	27.56±0.66	27.43±0.74	27.46±0.50	28.3±0.79	27.84±0.67	0.512
	Haugh unit score						
32	87.73±1.13	86.47±1.24	85.86±0.77	86.04±0.74	84.84±0.81	83.06±2.54	0.203
36	87.38±1.21	84.86±1.84	85.59±1.68	84.12±1.39	84.7±1.71	88.91±2.06	0.306
40	83.07±2.78	79.59±2.24	82.83±1.48	81.22±1.76	83.58±1.18	84.26±2.11	0.655

Conclusion

It is concluded that the dietary inclusion of flaxseed oil, fish oil and their combinations up to 5.0 per cent level did not cause any adverse effect on body weight, egg production, feed intake, feed conversion ratio and egg quality traits like shape index, shell thickness, yolk colour score, yolk index, yolk weight, albumen index and albumen weight in White Leghorn hens. Future investigations on the enrichment of eggs with omega 3 fatty acids after dietary inclusion of this combination need to be taken up.

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Conflicts of Interest

The authors declare that they have no conflict of interest.

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