UDK: 338.439.4:637.4 664:28-442-027.568 | Ekonomski izazovi/ Economic Challenges, Year 11, no. 22, pg. 81-91
doi: 10.5937/EkoIzazov2222081S - Date of receiving: 16.05.2022.
„Original science paper“ - Date of acceptance: 25.09.2022.

HALAL PRODUCTION OF OMEGA 3 ENRICHED EGGS ON THE FARM PEĆIGRAD, B&H

Amela Semić
Faculty of Agriculture and Food Sciences,
Sarajevo, Bosnia and Herzegovina
amela.semic@asholding.ba

Arman Hodžić
Third year student of University of Sarajevo – Veterinary
Sarajevo, Bosnia and Herzegovina

Aida Hodžić
Department of Veterinary Medicine Basic Sciences, University of Sarajevo – Veterinary Faculty, Sarajevo, Bosnia and Herzegovina

Abstract
The aim of this research is to develop a functional food and halal Omega 3 eggs by dietary manipulation of laying hens as well as to investigate the influence of nutrient composition on egg yolk content of PUFAs.
The research was conducted on the farm Pećigrad, Velika Kladuša, which has 21,162 laying hens, with an egg production rate of 70%. Laying hens were fed with halal and NON GMO feed mixtures from three suppliers.
Omega 3 eggs contained 318 mg of Omega 3 PUFAs, which is 6 times more than the standard eggs.The n-6:n-3 PUFAs ratio was 18.4:1.9=9.9 in egg yolks of laying hens fed with standard feed mixture, while in egg yolks produced by laying hens fed with feed mixture enriched with flaxseed this ratio was 17.8:5.3=3.4. Consumption of eggs with the latter n-6:n-3 ratio can prevent cardiovascular diseases and provides the intake of RDA of Omega 3 FAs.
Traceability and established HrCCP in the chain of suppliers, in processes of primary and industrial production, application of good practices, sustainable development resulted in halal products with additional functional properties.

Keywords: omega 3 eggs, functional food, NON GMO product, halal product.
JEL codes: L66

INTRODUCTION

The energy and biological value of a food depends on the amount and mutual relationship of individual nutrients contained in it. Functional food satisfactorily demonstrates a beneficial influence on one or more target functions of the organism, above and beyond the corresponding nutritional effects, in a way that contributes to either good health or disease risk reduction. In that product, bioactive, functional components will be concentrated, or particularly energetically and biologically favorable products will be produced. The applied technology enables the production of food with fewer calories, less fat, with a changed ratio of fatty acids, less salt, increased nutritional density, which suits not only each group of consumers, but also their subgroups. This product can be plant and animal foods with a changed percentage of fats, fibres, proteins, vitamins and minerals. There are five approaches to making food functional: elimination of an ingredient that has a harmful effect; increasing the level of a natural ingredient of some food to reach a concentration that can have the desired effect; the addition of a substance that is not a common ingredient of a food, but whose beneficial effects have been proven; replacement of a macronutrient, whose intake is usually excessive and causes harmful consequences, with an ingredient that has a positive effect on health; and increasing the bioavailability of ingredients that have been proven to have positive effects on health. A number of epidemiological and controlled experiments have reported an inverse relationship between consumption of n-3 polyunsaturated fatty acids (PUFA) and cardiovascular disease risk (Bang et al. 1980; Kromhout et al. 1985; Kinsella et al. 1990; Pauletto et al. 1996; Leaf and Kang 1998). The Western-world diet is deficient in n-3 fatty acids and high in n-6 fatty acids mainly because of increase of consumption of vegetable oils such as sunflower, corn and safflower oils, which are rich in n-6 polyunsaturated fatty acids (PUFA) (Simopoulos and Robinson 1998).

Chicken eggs are an inexpensive source of high-value nutrients present on the table every day, the only nutritional deficiency of which can be a relatively high cholesterol content and a high proportion of saturated fat compared to unsaturated fatty acids. The composition of fatty acids in eggs can be changed by modifying the composition of fatty acids in the hen's diet. Such changes have been demonstrated in many studies (Caston and Leeson 1990; Cherian and Sim 1991; Caston et al. 1994; Cherian et al. 1995; Nash et al. 1995; Ayerza and Coates 1999). Currently on the market, n-3 enriched eggs are produced by including flax seeds, chia seeds, fish oil or flour and seaweed in the hen's diet. Both mentioned seeds are rich in α-linolenic acid, while marine sources contain long-chain n-3 acids, docosahexaenoic (DHA) and eicosapentaenoic acid (EPA).

Of the components used in the diet of hens to produce eggs rich in n-3 PUFA, only flax (Linum usitatissimum L.) and chia (Salvia hispanica L.) are produced by traditional agricultural methods. These two species have the highest concentration of n-3 α-linolenic acid (Oomah and Kenasehuk 1995; Coates and Ayerza 1999, 2000).

There have been many attempts to use flax as a source of n-3 fatty acids in poultry diets due to the low price and its availability. Despite the fact that flaxseed does not contain DHA, eggs contain this acid due to the process of synthesis of DHA from the precursor linolenic acid under the influence of two enzymes: desaturase and elongase (Cherian and Sim, 1991). The increase in PUFA content in the yolk is accompanied by a significant decrease in saturated fatty acids, resulting in a healthy fat profile and more nutritious eggs. It is assumed that this effect is realized by a diet with a plant supplement that could help in more efficient transformation of a-linolenic acid into DHA. In addition, Bean and Leeson (2003) reported that birds consuming flaxseed simultaneously had higher ($P<0.0001$) amounts of linolenic acid, DHA, and total n-3 PUFA in their eggs. According to Cherian and Sim (1991), PUFA (n-6 and n-3 series) in the diet are more effective in reducing monounsaturated than saturated acids in eggs, through the inhibition of desaturase enzyme activity to produce mainly oleic acid. In this study, it was shown that a rich source of PUFA such as flaxseed results in significantly lower concentrations of monounsaturated acids (MUFA), mainly oleic.

A number of researchers have reported a fishy odor or taste in eggs produced by hens fed seafood (Jiang et al. 1992; Van Elswyk et al. 1992; Caston et al. 1994; Nash et al. 1995; Van Elswyk et al. 1995; Scheideler et al. 1997) and this is the consequence of oxidation of polyunsaturated fatty acids (Jiang et al. 1992; Van Elswyk et al. 1995; Cherian et al. 1995). Other researchers, however, suggest that fish odor arises from lipid and non-lipid substances present in the food (Leskanich and Noble 1997). In any case, this negative side effect is avoided by adding flax seeds to the feed of laying hens.

In conditions of quantitative and qualitative lack of food, we are witnessing the increasingly frequent use of GMO raw materials and food. A special problem related to GMO-based food is the assessment of its risk to human health. There are several questions that must be considered during the assessment of the safety of using new plant varieties obtained by genetic modifications: the content of natural toxic substances, the potential of transferring allergens from one source to another, the concentration and bioavailability of important nutrients, the possibility of gene transfer from GMOs to microorganisms of the digestive tract, identity, composition and biological value of modified carbohydrates or fats. This technique can improve agronomic and quality properties such as nutritional value, composition, taste, smell, reserve substances, resistance to some diseases, herbicides, tolerance to stressful conditions. In agriculture, genetic engineering has become very important, and is most often applied to wheat, rice, corn, tomatoes, etc. Some authors advocate the thesis that genetic modification, genetic engineering, is a radical and dangerous change in the genetic structure of food that can result in unexpected phenomena, e.g. due to the appearance of toxins, allergens and damage to the consumer's immune system. The allergic reaction is most often related to proteins, and less often to smaller molecules, e.g. haptens. Genetic modification creates unrecognizable proteins in the human body, so it can be determined that GMO products have the characteristics of an allergen. A person can react to these unrecognizable proteins with an allergy and even anaphylactic shock. The dangers of releasing GMOs into the environment can be greater than radioactive radiation and the release of chemicals because GMOs can reproduce, multiply, spread, mutate and transfer their genetic material to related organisms. Therefore, it is extremely important to use NON-GMO raw materials in the production chain of potential functional food.

Halal quality implies a set of applied procedures in the production process as well as the totality of product characteristics that are in accordance with Islamic regulations and determine both the product and the service acceptable for use or consumption by all consumers who want to know what they are eating. Today's trends have shown that the market has recognized Halal food as a kind of guarantee of premium quality. In the standardization of halal, a proactive preventive approach is applied to prevent the possibility of haram entering the entire production process.

The aim of this research was to examine the influence of adding flax seeds to feed for laying hens on the fatty-acid composition of eggs, as well as the ratio of n-6:n-3 polyunsaturated fatty acids, and in this way potentially develop a new functional and halal product - "omega 3 enriched halal eggs".

MATERIAL AND METHODS

The research was conducted on the farm Pećigrad, Velika Kladuša, which has a total of 21,162 laying hens, with a laying capacity of 70%. Hens were kept in cages, separately for the production of regular table eggs and omega 3 enriched eggs. For the production of omega 3 eggs, the flock numbered 7,000 hens, and the rest were fed with a standard feed mixture. The feeding regimen for all hens was the same, 120 g/laying hen/day. In the event of increased breakage and shards due to the thin egg shell, calcium is included as a dietary supplement. The number of chicken deaths was 2-3 per day, which is in the expected percentage of mortality.
The egg producer proved that he does not use haram raw materials (all mixtures were of halal and NON GMO status), as well as that the production process is carried out in accordance with halal requirements. A halal product requires, in addition to precisely defined raw materials, also appropriate processing, preparation, packaging, labeling, storage and transportation.
For the production of table eggs, a standard feed mixture was used, the declared basic chemical composition of which is shown in Table 1.

Table 1. Chemical composition of fodder for the production of table eggs

Chemical component	Value (%)
Proteins	16,9
Fats	6,6
Calcium	2,9
Phosphorus	0,4

Table 2 shows the nutritional components contained in the feed mixture for the production of omega 3 eggs. Carophyl Yellow 10% (0.001%) and Xamacol (0.040%) were added to the mixture in order to prevent less intensively colored yolks in hens fed with a mixture enriched with flax seeds, because lutein and zeaxanthin give the natural yellow color.

Table 2. Content of nutrient components in the feed mixture for the production of omega 3 enriched eggs

Nutrient component	Content (%)
Maize - 6,8 % SB 2018	55,364
Soybean meal - 45 %	27,742
Soybean oil - refined	0,921
Flaxseed - ekstruded	5,5
Sea salt	0,226
Coarse calcite	5,5
Limestone	2,984
Baking soda	0,15
Mono-potassium phosphate	0,833
px-NSK - 0,5%	0,5
Alimet - 88%	0,181

Carophyl Yellow 10%	0,001
Yellow color - Xamacol	0,04
Ronozyme Hiphos - 90 g	0,009
Choline chloride - 75% liquid	0,05

Analyzes of the fatty acid composition of omega 3 eggs were performed using the gas chromatography method at the Emona Development Center for Nutrition l.l.c. from Ljubljana, Slovenia.

RESULTS AND DISCUSSION

Table 3 shows the content of nutrients in the feed mixture enriched with extruded flaxseed, which was used for the production of omega 3 eggs.

Table 3. Content of nutrients in the feed mixture for the production of omega 3 eggs

Ingredient/substance	Unit of measure	Value
Crude protein	g/kg	175
Crude fat	g/kg	53,34
Crude fiber	g/kg	32,25
Raw ash	g/kg	123,30
Metabolic energy	MJ/kg	11,50
Metabolic energy	kcal/kg	2.747, 26
Phosphorus - total	g/kg	5,3
Phosphorus - usable	g/kg	4,53
Calcium	g/kg	38,5
Magnesium	g/kg	2,14
Potassium	g/kg	7,89
Sodium	g/kg	1,70
Chlorine	g/kg	1,9
Iron	mg/kg	108,42
Manganese	mg/kg	115,94
Zinc	mg/kg	108,46
Copper	mg/kg	20,47
Iodine	mg/kg	1,01
Selenium	mg/kg	0,36
Iron - inorganic	mg/kg	50
Copper - inorganic	mg/kg	12,5
Zinc - inorganic	mg/kg	80
Manganese - inorganic	mg/kg	101,68
Cobalt - inorganic	mg/kg	0,32
Selenium - inorganic	mg/kg	0,25
Lysine	g/kg	8,94
Methionine	g/kg	4,1
Methionine+cystine	g/kg	6,72

Threonin	g/kg	6,28
Tryptophan	g/kg	1,98
Real preb lysine	g/kg	7,65
Real preb methionine	g/kg	3,85
Real preb cystine	g/kg	2,13
Real preb threonine	g/kg	5,31
Real preb tryptophan	g/kg	1,65
A	IE/kg	11.286,75
D-3	IE/kg	3.200
E	mg/kg	34,43
K	mg/kg	6,11
Biotin H-2	mg/kg	0,27
Thiamine B-1	mg/kg	1,86
Riboflavin B-2	mg/kg	7,1
Pyridoxine B-6	mg/kg	5,27
B-12	mg/kg	0,03
Niacin	mg/kg	42,89
Ca pantothenate	mg/kg	13,33
Choline	mg/kg	375
Folate	mg/kg	0,63
A - added	IE/kg	10.000
D-3 - added	IE/kg	3.200
E - added	mg/kg	25
K - added	mg/kg	6,11
Biotin H-2 - added	mg/kg	0,10
Thiamine B-1 - added	mg/kg	1
Riboflavin B-2 - added	mg/kg	6,24
B6 pyridoxine -added	mg/kg	2,5
B 12 - added	mg/kg	0,03
Niacin - added	mg/kg	0,53
Ca pantothenate - added	mg/kg	10
Choline -added	mg/kg	375
Folate - added	mg/kg	0,5
Linoleic (18:2, n-6)	g/kg	17,05
α-linoleic (18:3, n-3)	g/kg	10,05
Color	g/kg	0,03

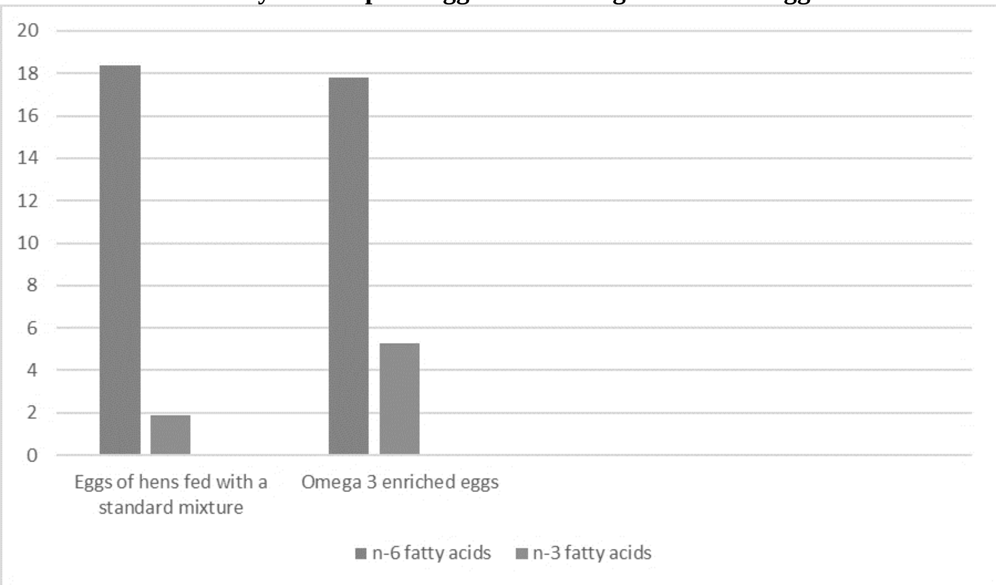
The feed mixture qualitatively fully satisfied the metabolic needs of laying hens, and the addition of extruded flaxseed resulted in a ratio of polyunsaturated fatty acids - linoleic from the n-6 series and α-linolenic from the n-3 series of 17.05:10.05=1.70. Table 4 shows the fatty-acid composition of ordinary table eggs and omega 3 eggs. Compared to ordinary table eggs, omega 3 eggs contained more unsaturated fatty acids, especially those with more double bonds and 2.78 times more of those from the n-3 series.

Table 4. Fatty acid composition of ordinary table eggs and omega 3 eggs

1. **Fatty acid (formula)**	**Fatty acid (name)**	**Consumable eggs (%)**	**Omega 3 eggs (%)**
C 14 : 0	Myristinc	0,4	0,3
C 14:1	Myristoleic	0,1	0,1
C 15:0	Pentadecane	0,1	0,1
C 16:0	Palmitinc	24,1	21,0
C 16:1 c+t	Palmitoleic	3,6	2,6
C 17:0	Margarine	0,1	0,2
C 18:0	Stearic	7,1	7,7
C 18:1 c+t	Oleic	41,9	42,6
C 18:2 c+t, n-6	Linoleic	14,7	15,0
C 18:3 c, n-6	y-linolenic	0,2	0,1
C 19:0	Nonadecane	-	<0,1
C 18:3 c, n-3	α-linolenic	0,6	2,9
C 20:0	Arachidin	<0,1	<0,1
C 20:1 c+t	Eicosaenic	0,2	0,2
C 20:2 n-6	Eicosadiene	0,1	0,1
C 20:3 n-6	Homo-g-linolenic	0,2	0,2
C 20:4 n-6	Arachidin	2,5	2,1
C 20:3 n-3	Eicosatrienic	-	0,1
C 20:5 n-3	Eicosapentaenoic (EPA)	<0,1	0,1
C 22:4 n-6	Docosatetraene	0,2	0,1
C 22:5 n-6	Docosapentaenoic	0,5	0,2
C 22:5 n-3	Docosapentanoic	0,2	0,2
C 22:6 n-3	Docosahexaenoic (DHA)	1,1	2,0
Unidentiefied peaks		2,1	2,2
Total area		100,0	100,0
Σ n−3		**1,9**	**5,3**
Σ n−6		**18,4**	**17,8**
n-6 / n-3		**9,9**	**3,4**
Monounsaturated (MUFA)		45,8	45,4
Polyunsaturated (PUFA)		20,3	23
Saturated (SFA)		31,7	29,2
Unsaturated (UFA)		66,1	68,4
PUFA / SFA		2,1	2,3

One of the very important parameters in evaluating the fatty acid composition of eggs is the ratio between n-6 and n-3 polyunsaturated fatty acids. In the case of eggs produced by laying hens fed with a standard feed mixture, this ratio was 18.4 : 1.9 = 9.9, i.e. about 10, while in the case of eggs produced by laying hens fed with linseed enriched feed mixture, the ratio was between n-6 and n-3 polyunsaturated fatty acids was 17.8 : 5.3 = 3.4, or about 3 (Graph 1). This ratio in the omega 3 enriched eggs produced in this study is even lower than the 4:1 ratio reported by Kouba and Mourot (2011).

Figure 1. Comparison of the content of n-6 and n-3 polyunsaturated fatty acids in ordinary consumption eggs and in omega 3 enriched eggs.



Numerous studies show various health benefits of consuming eggs enriched with n-3 polyunsaturated fatty acids, especially in people with cardiovascular diseases, and they have the status of a functional food. Some of the benefits of consuming eggs with a lower ratio between n-6 and n-3 polyunsaturated fatty acids include reducing risk of many chronic diseases (Simopoulos, 2002; Harris et al., 2006), beneficial effects in patients with rheumatoid arthritis (James and Cleland, 1997), reduction of pro-inflammatory cytokines (Cotogni et al., 2011), reduction atherosclerotic changes and general anti-inflammatory effect (Hagi et al., 2010). In addition, Simopoulos (2011) indicated that a lower ratio of omega-6 : omega-3 fatty acids is important for homeostasis and normal development of organisms. The development of visual and cognitive functions in fetuses and young children is associated with an adequate intake of n-3 PUFA, especially DHA and EPA. The content of these acids in omega 3 eggs was 2 and 0.1%, respectively. At the same time, the content of DHA in omega 3 eggs was almost twice as high as in ordinary table eggs (Tab. 4).
The Omega 3 egg, produced in our experimental setup, contains 318 mg of n-3 PUFA, which is about six times more than a regular table egg. The consumption of such two omega 3 eggs can significantly contribute to the recommended daily intake of n-3 PUFA, which is 900 mg/d/2000 Cal (Hibbeln and Davis, 2009).
Researchers report that chickens fed by flaxseed show reduced growth compared to those fed a conventional diet (Caston et al. 1994; Scheideler and Froning. 1996; Novak and Scheideler, 2001). Birds fed by flaxseed have also been reported to exhibit diarrhoea resulting from accelerated intestinal peristalsis (Scheideler and Froning. 1996; Gonzalez-Esquerra and Leeson 2000). A high rate of peristalsis could reduce

the bird's ability to digest and absorb most nutrients. It is also important to note the presence of anti-nutritive cyanogenic glycosides and antagonists of vitamin B6 (pyridoxine) present in flaxseed (Kung and Kummerow 1950; Homer and Schaible 1980; Bhatty 1993; Bond et al. 1997). Because of all the above, the percentage of linseed used in the diet of our chickens is strictly calculated to avoid potential toxic and anti-nutritional effects. It has been proven that the use of 5% flaxseed in the total diet increases the percentage of omega-3 fatty acids in eggs but at the same time decreases the percentage of omega-6 fatty acids (Ferrier et al. 1995; Bean and Leeson 2003; Ansari et al. 2006; and Yalcin and Unal 2010), which is also confirmed by our results (Tab.4).

In the end, it is important to point out that the certification by the B&H Halal Quality Certification Agency, the verification of the List of Halal Certified Products, confirmed that the following products are on the list and bear Halal status: fresh eggs of S, M and L class, fresh eggs of commercial called M Bazar 30/1 and 10/1, XL class, and omega 3 enriched eggs.

CONCLUSION

The addition of extruded linseed in the amount of 5.5% to the feed mixture of laying hens increased the PUFA content in the eggs, especially those of the n-3 series, which resulted in an even more favorable n-6 : n-3 PUFA ratio from current official recommendations. By consuming two omega 3 enriched eggs produced in this way on a daily basis, the recommended daily amount of omega 3 fatty acids, which are necessary for the optimal functioning of the body, is consumed. Furthermore, this concept achieves uniqueness on the B&H market, which is more of an argument for a good consumer reaction, regardless of the slightly more expensive product. Through traceability and established HrCCP in the chain from suppliers, processes in primary and industrial production, application of good practices, sustainable development, a halal product with additional functional properties - "omega 3 halal eggs" was obtained.

BIBLIOGRAPHY

1. Ansari, R., Azarbayejani, A., Ansari, S., Asgari, S. & Gheisari, A. (2006). Production or egg enriched with omega-3 fatty acids in laying hens. A. R. Y. A. J., 1, 242-246.
2. Ayerza, R. (h) & Coates, W. (1999). An omega-3 fatty acidenriched chia diet: its influence on egg fatty acid compo-sition, cholesterol and oil content. Can. J. Anim. Sci., 79. 53–58.
3. Ayerza, R. (h) & Coates, W. (2000). Dietary levels of chia influence on yolk cholesterol, lipid content and fatty acidcomposition for two strains of hens. Poult. Sci., 79, 724–739.
4. Bang, H. O., Dyerberg, J., & Sinclair, H. M. (1980). The composition of the Eskimo food in Northwest Greenland. Am. J. Clin. Nutr., 33, 2657–2661.
5. Bean, L. D. & Leeson, S. (2003). Long-term effects of feeding flaxseed on performance and egg fatty acid composition of brown and white hens. Poult. Sci., 82, 388–394.

6. Bhatty, R. S. (1993). Further compositional analyses of flax: mucilage, trypsin inhibitors and hydrocyanic acid. J. Am. Oil Chem. Soc., 70, 899–904.
7. Bond, J. M., Julian, R. J., & Squires, E. J. (1997). Effect of dietary flaxseed on broiler growth, erythrocyte deformability and fatty acid composition of erythrocyte membranes. Can. J. Anim. Sci., 77, 279–286.
8. Caston, L. J., & Leeson, S. (1990). Dietary flax and egg composition. Poult. Sci., 69, 1617–1620.
9. Caston, L. J., Squires, E. J. & Leeson, S. (1994). Hen performance, egg quality and the sensory evaluation of eggs from SCWL hens fed dietary flax. Can. J. Anim. Sci., 74, 347–353.
10. Cherian, G., & Sim, J. S. (1991). Effect of feeding full fat flax and canola seeds to laying hens on the fatty acid composition of eggs, embryos, and newly hatched chicks. Poult. Sci., 70, 917–922.
11. Cherian, G., Li., S. X., & Sim, J. S. (1995). Dietary alpha-linolenic acid and laying hen strain: Fatty acids of liver, adipose tissue, white meat, dark meat, and egg yolk. J. Agric. Food. Chem., 43, 2553–2559.
12. Cotogni, P., Muzio, G., Trombetta, A., Ranieri, V. M., Canuto, R. A. (2011). Impact of the omega-3 to omega-6 polyunsaturated fatty acid ratio on cytokine release in human alveolar cells. J. Parenter. Enteral. Nutr., 35(1), 114-21.
13. Ferrier, L. K., Caston, L. J., Leeson, S., Squires, J., Weaver, B. J., & Holub, B. J. (1995). Alpha-linolenic acid and docosahexaenoic acid enriched eggs from hens fed flaxseed: influence on blood lipids and platelet phospholipid fatty acids in humans. Am. J. Clin. Nutr., 62, 81–86.
14. Gonzalez-Esquerra, R., & Leeson, S. (2000). Studies on the metabolizable energy content of full-fat flaxseed in mash, pellet, and crumbled diets assayed with birds of different ages. Poult. Sci., 79, 1603–1607.
15. Hagi, T., Kobayashi, M., & Nomura, M. (2010). Molecular-based analysis of changes in indigenous milk microflora during the grazing period. Biosci. Biotechnol. Biochem., 74, 484–487.
16. Harris, K., Miller, M. F., Loneragan, G. B., & Brashears, M. M. (2006) Validation of the use of organic acids and acidified sodium chlorite to reduce Escherichia coli O157 and salmonella Typhimurium in beef trim and ground beef in a simulated processing environment. J. Food Prot., 69(8), 1802-1807.
17. Hibbeln, J.R., & Davis, J.M. (2009). Considerations regarding neuropsychiatric nutritional requirements for intakes of omega-3 highly unsaturated fatty acids. Prostaglandins Leukot. Essent. Fatty Acids, 81, 179–186.
18. Homer, P. & Schaible, P. J. (1980). Poultry: Feeds and nutrition. Westport, CT, AVI Publishing.
19. James, M. J. & Cleland, J. G. (1997). Dietary n-3 fatty acids and therapy for rheumatoid arthritis. Semin. Arthritis Rheum., 27, 85–97.
20. Jiang Z., Ahn, D. U., Ladner, L., & Sim J. S. (1992). Influence of feeding full fat flax and sunflower seed on internal and sensory qualities of eggs. Poult. Sci., 71, 378-382.
21. Kinsella, J. E., Lokesh, B., & Strong, R. A. (1990). Dietary n-3 polyunsaturated fatty acids and amelioration of cardiovascular disease: Possible mechanisms. Am. J. Clin. Nutr., 52, 1–28.
22. Kouba, M. & Mourot, J. (2011). A review of nutritional effects on fat composition of animal products with special emphasis on n-3 polyunsaturated fatty acids. Biochimie, 93, 13-17.
23. Kromhout, D., Bosschierter, E. B., & Coulnader, C. D. (1985). The inverse relation between fish consumption and 20-year mortality from coronary heart disease. N. Eng. J. Med., 312, 1205-1209.

24. Kung, T. K. & Kummerow, F. A. (1950). The deposition of linolenic acid in chickens fed linseed oil. Poult. Sci., 29, 846–851.
25. Leaf, A., and Kang, J. X. (1998). Omega 3 fatty acids and cardiovascular disease. World Rev. Nutr. Diet., 83, 24-37.
26. Leskanich, C. O., & Noble, R. C. (1997). Manipulation of the n-3 polyunsaturated fatty acid composition of avian eggs and meat. World's Poult. Sci. J., 53(2), 155-183.
27. Nash, D. M., Hamilton, R. M. G. & Hulan, H. W. (1995). The effect of dietary herring meal on the omega-3 fatty acid content of plasma and egg yolk lipids of laying hens. Can. J. Anim. Sci., 75, 247–253.
28. Novak, C. & Scheideler, S. E. (2001). Long-term effects of feeding flaxseed-based diets. 1. Egg production parameters, components, and eggshell quality in two strains of laying hens. Poult. Sci., 80, 1480–1489.
29. Oomah, B. D. & Kenaschuk, E. O. (1995). Cultivars and agronomic aspects. Pages 43–45 in Flaxseed in human nutrition, edited by S. C. Cunnane and L. U. Thompson. American Oil Chemists' Society Press, Champaign, IL.
30. Pauletto, P., Puato, M., Angeli, M. T., Pessina, A. C., Munhambo, A., Bittolo-Bon, G., & Galli, C. (1996). Blood pressure, serum lipids, and fatty acids in populations on a lake-fish diet or on a vegetarian diet in Tanzania. Lipids, 31, s309–s312.
31. Scheideler S. E., Froning G., & Cuppett S. (1997). Studies of consumer acceptance of high omega-3 fatty acid-enriched eggs. J. Appl. Poult. Res., 6, 137-146.
32. Scheideler, S. E., & Froning, G. W. (1996). The combined influence of dietary flaxseed variety, level, form, and storage conditions on egg production and composition among vitamin E supplemented hens. Poult. Sci., 75, 1221–1226.
33. Simopoulos, A. P. (2002) The importance of the ratio of omega-6/omega-3 essential fatty acids. Biomed. Pharmacother., 56, 365-379.
34. Simopoulos, A. P. (2011). Evolutionary aspects of diet: the omega-6/omega-3 ratio and the brain. Mol. Neurobiol., 44(2), 203-215.
35. Simopoulos, A. P., & Robinson, J. (1998). The omega plan: The medically proven diet that gives you the essential nutrients you need. HarperCollins, New York.
36. Van Elswyk, M. E., Dawson, P. L., & Sams, A. R. (1995). Dietary menhaden oil influences sensory characteristics and head-space volatiles of shell eggs J. Food Sci., 60(1), 85-89.
37. Van Elswyk, M. E., Prochaska, J. F., Carey, J. B., & Harris, P. S. (1992). Physiological parameters in response to dietary menhaden oil in molted hens. Poult. Sci., 71(Suppl. 1), 144.
38. Yalçin, H., & Unal, M. K. (2010). The enrichment of hen eggs with omega-3 fatty acids. J. Med. Food, 13(3), 610–614.