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Full Length Article

Effect of Supplementing Olive, Blackseed and Flaxseed Oils on Growth Performance and Serum Biochemistry of Broiler and Economics of its Production

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Abstract

This study was conducted to investigate the effect of olive oil, black seed oil and flaxseed oil on growth performance and serum biochemistry of broiler chicks. Three hundred broiler chicks were grouped into 30 experimental units having 10 chicks each. Three levels (0.5, 1.0 and 1.5%) of each of the oils were offered to the chicks such that each treatment was replicated thrice. The chicks fed on basal diet served as a control. The data on weekly body weight gain, feed intake, weekly feed conversion ratio, Newcastle Disease (ND) and IBD titers, serum triglycerides, serum cholesterol, low density lipoprotein and high density lipoprotein were recorded. The overall weight gain (1720.6 g) was significantly ($P<0.05$) highest in the treatment having 0.5% flax seed oil. Highest value of titer (213.33) against ND was observed in the treatment having black seed oil 0.5%. Non-significant results were observed regarding serum triglycerides, serum cholesterol, low and high density lipoprotein. Overall results depicted that 0.5% flax seed oil improves growth performance in broiler chicks. © 2017 Friends Science Publishers

Keywords: Plant oil; Growth promoter; Immune booster; FCR

Introduction

Antibiotic growth promoters (GPs) are being used in poultry feed to enhance their performance for the last few decades. No doubt these antibiotics inhibit the growth of pathogenic microorganisms, prevent intestinal problems and improve performance of chicken (Dibner and Richards, 2005; Adil *et al.*, 2011), but antibiotic residues found in the meat and eggs are great risk for human health (Jang *et al.*, 2007). Due to the continuous use of antibiotics as GPs, pathogenic bacterial flora has become resistant to these antibiotics and it has become dominant in the environment. It is expected that in next few years birds will show no response to antibiotics due to continuous use of antibiotics as GPs. These drug resistant pathogens will also affect the human health and diseases with drug resistant pathogens will emerge as a serious issue. Due to these facts, strong protest against the use of antibiotics as GP and immune booster has been generated (Gustafson and Bowen, 1997; Waldroup *et al.*, 2003) and European Union has put a ban on use of antibiotics in poultry feed since 2006. That is why scientists are working on different alternates of these antibiotic GPs.

Fats and oils are good alternate of these antibiotic GPs. Vegetable oils are added in poultry feeds as energy source. They not only increase feed intake but also have growth promoting effects. Many fats and oils have been reported in literature as GP (Korver and Klasing, 1997; El-Sheikh *et al.*, 1998; Growell, 1999; Akhtar *et al.*, 2003; Ramadan, 2007; Windisch *et al.*, 2008). Plant oils have different biological activities like antibacterial, antioxidant and antifungal (Steiner, 2009) and supplementation of poultry feed with these oils improve the performance by changing the gut microflora, profile of digestive enzymes and stimulation of immune system (Haijto *et al.*, 1989; Helander *et al*., 1998; Jang *et al.*, 2004). A large number of studies have been conducted on the antibacterial effect of plant oils *in vitro* (Jamroz *et al.*, 2003; Mitsch *et al.*, 2004). Improvement in growth performance of broilers may also be due to improvement in gut equilibrium, by lowering the fermentation, bacterial colony counts and stimulating the digestive secretions. It has been observed that vegetable oils increase the mucous production in intestine and reduces the bacterial adhesion in broilers (Jamroz *et al.*, 2006; Windisch *et al.*, 2008). Improved digestive

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enzyme activities (trypsin and amylase) and absorption of nutrients in intestine promote growth in broilers (Lee *et al.,* 2003; Jang *et al.,* 2007).

Black seed, flax seed and olive oils have also been studied for anti-oxidative and antimicrobial effects (Gokce *et al.,* 2000; Ramadan, 2007). Black seed oil also helps in the reduction of the cholesterol synthesis in poultry by inhibiting the activity of hepatic 3-hydroxy-3-methylglutaryl coenzyme A. Reduction of total cholesterol, triglycerides and LDL-cholesterol and increase in HDL cholesterol content in the egg yolk has been reported by use of black seed oil. Use of black seed not only improve the overall performance, feed efficiency, growth rate and digestibility but also reduce the deposition of abdominal fat in broilers (Ashayerizadeh *et al.,* 2009). Olive oil helps to increase monounsaturated fatty acids and decrease low density lipoprotein cholesterol level without reducing high density lipoprotein cholesterol in serum (Bolukbasi and Erhan, 2005). Olive oil has 55.0-83.0% oleic acid, birds fed with the diet having olive showed higher live weight, dressing percentage and weights of visceral organs (Zarei *et al.,* 2011).

Dietary inclusion of oils has been also reported to have immuno-regulatory effects in broilers (Fritsche *et al.,* 1992). Extract from *Achillea talagonica* have found to stimulate the immune system (Rezaeipoor *et al.,* 1999). Oils rich in PUFA affect the inflammatory response and results in improved immune system functions (Korver and Klasing, 1997). Fish oil fed to male broilers resulted in improved cellular immunity but did not affect the humoral immunity (Korver and Klasing, 1997; Maroufyan *et al.,* 2012) reported that the titer of both ND and IBD was improved by using combinations of tuna oil and sunflower oil in broiler diets.

But major hindrance in use of these plant oils as commercial feed additives is lack of knowledge on economics of its use, exact mechanism of action and dose level to be included in feed. Present study was, therefore, conducted to evaluate the economics of broiler production when plant oils are used as feed additives.

Materials and Methods

To explore the effect of olive, black seed and flax seed oils on the growth promotion, Serum biochemistry and economics of broiler production this project was executed at the Poultry Research Center, Department of Poultry Science, University of Agriculture, Faisalabad, Pakistan. Experimental room was thoroughly cleaned, white washed, disinfected and fumigated before the arrival of chicks. Each experimental unit was kept in a separate pen having similar floor space and environmental conditions like temperature, relative humidity, ventilation and light etc. and drinking water was offered *ad libitum* to all experimental units. Three hundred day old broiler chicks (Hubbard strain) were purchased from hatchery and were divided into 30 experimental units having ten chicks each. All birds were vaccinated according to recommended vaccination schedule (Table 1). These units were randomly allocated to 10 treatment groups (Table 2).

Treatments were started from day 1 to 35th day of the experimental period. Commercial feed was used with or without supplementation of olive oil, black seed oil and flax seed oil. Body weights of the all individual birds were recorded on first day and every week thereafter. Growth rate and feed consumption of each group were recorded on weekly basis. Feed conversion ratio (FCR) was calculated for each experimental unit on weekly basis.

$$\text{Feed Conversion Ratio} = \frac{\text{Feed intake (g)}}{\text{Weight gain (g)}}$$

Blood samples were collected from brachial vein of birds in 5 mL sterilized disposable syringes and placed at room temperature in upright tilted position for blood clotting. The syringes were placed in the same position in refrigerator for shrinkage of clot to increase harvesting of the serum. The serum were taken in 1.5 mL labeled Eppendorf tubes and were frozen (-20°C) till analysis. Antibody titer against Newcastle Disease (ND) and Infectious Bursal Disease (IBD) were estimated from serum samples collected on day 32nd and 35th day, using methods described by MAFF (1984) and Cullen and Wyet (1975), respectively. Serum triglycerides, serum cholesterol, low density lipoprotein and high density lipoprotein were estimated from samples collected at the end of the experiment on MICROLAB 300 and TECHNO-786 using reagents from HUMAN and MERCK (Schettler and Nussel, 1975), respectively.

For the interpretation of results the data were subjected to analysis of variance technique under completely randomized design. Treatment means were compared by Least Significant Difference test (Montgomery and Runger, 2010).

Results

Effect on Growth Performance

Overall weight gain of the birds differed significantly ($P<0.05$) among treatments. Maximum weight gain was observed in the treatment having flax seed oil 0.5% (1720.6.g) followed by the treatment having olive oil 1.0% (1701.5 g) and olive oil 0.5%(1691.4 g), while minimum weight gain was recorded in –ve control that was 1489 g (Table 3). As far as comparison of weight gain is concerned all the treatment groups showed significantly better growth rate than the control but between the treatments difference was non-significant. Similarly, maximum overall feed intake was observed with flax seed oil 0.5% that was 3089.7 g. Effect of inclusion of dietary oils in broiler showed statistically significant ($P<0.05$) effect on FCR.

Table 1: Vaccination Schedule of experimental birds

Age (day)	Vaccine	Method of administration
7	Newcastle Disease (Lasota)	Eye drop
12	Infectious Bursal Disease (Bur-706)	Eye drop
22	Newcastle Disease (Lasota)	Drinking water
25	Infectious Bursal Disease (Bur-706)	Drinking water

Table 2: Dose of oils administered to different experimental groups

Treatment Group	Feeding Plan
A1	Commercial Ration + 0.5% olive oil
A2	Commercial Ration + 1% olive oil
A3	Commercial Ration + 1.5% olive oil
B1	Commercial Ration + 0.5% black seed oil
B2	Commercial Ration + 1% black seed oil
B3	Commercial Ration + 1.5% black seed oil
C1	Commercial Ration + 0.5% flaxseed oil
C2	Commercial Ration + 1.0% flaxseed oil
C3	Commercial Ration + 1.5% flaxseed oil
D (-ve Control)	Commercial Ration + No supplementation

The best FCR was observed in the treatment having olive oil 1.0% that was 1.72 followed by the treatments having flax seed oil 0.5% and black seed oil 0.5% that was 1.75 and 1.77, respectively (Table 3).

Serum Biochemistry

Detailed results of serum biochemistry have been presented in Table 4. Maximum level of cholesterol was observed in the treatment having flax seed oil 0.5% that was 199.5 mg/dl followed by control group that was 192.5 mg/dl. Minimum value of cholesterol was found in the treatment having flax seed oil 1.5% that was 152.33 mg/dl. Similarly, maximum value of triglycerides was observed in treatment having black seed oil 1.0% that was 232.67 mg/dl and minimum value of triglycerides was found in the treatment having black seed oil 1.5% that was 124.17 mg/dl. Evaluation of Serum High Density Lipoproteins (HDL) the highest value in group fed on black seed oil 1.5% that was 46.83 mg/dl followed by control and treatment having olive oil 0.5% that is 46.33 mg/dl and 45.67 mg/dl. Birds fed on 1.5% showed 86.33 mg/dl of low density lipoproteins (LDL) followed by treatments having black seed oil 0.5% and flax seed oil 1.0% that is 86.5 mg/dl and 90.17 mg/dl, while maximum value of LDL was found in treatment having flax seed oil 0.5% that is 114.5 mg/dl. Statistical analysis of the experimental data showed non-significant ($P<0.05$) effect on cholesterol, triglycerides, LDL and HDL due to supplementation of different oils in the feed of broiler birds.

Antibody Titer after Vaccination

Maximum value of ND titer was observed in the treatment having black seed oil 0.5% that was 213.33. Minimum value of ND titer was observed in control that was 53.33. Statistical analysis of the experimental data showed significant ($P<0.05$) effect on ND titer due to varying levels of different oils in the feed of broiler birds. Comparison of the mean values with least significant difference test showed that groups having flaxseed oil 0.5% showed significantly maximum ND titer than all the treatments including control. Similarly, feeding of different oils also affected the Infectious Bursal disease (IBD) titer. Maximum titer was observed in olive oil 0.5% and flax seed oil 1.0% that was 106.67. followed by treatments having olive oil 1.0% & 1.5% and flax seed oil 1.5% that was 85.33. Detailed results have been presented in Table 5.

Economics of Oil Based GPs

Mortality of only one bird was observed in group treated with black seed oil 1.5% on the 11th day of the trial. No disease symptoms were observed in the dead bird. Maximum profit was calculated in the group feed on flaxseed oil 0.5% that was 50.292 rupees followed by olive oil 0.5% and flaxseed oil 1.0% that was 41.934 and 41.335 rupees, respectively. Whereas minimum profit was recorded in the treatment with black seed oil 1.5%that was 1.187 rupees (Table 6).

Discussion

All the groups fed with the oils (olive, black seed and flaxseed) differed non significantly from each other but showed significantly higher weight gain than the control. Similar findings were reported by Maroufyan *et al.* (2012) who found that omega-3 and omega-6 fatty acids in broiler diets from various oil sources with different levels gained significantly more body weight. These oils could also be used as energy sources for growth (Gardiner, 1973). These results have also been supported by the findings of Du and Ahn (2002) who reported that use of oils in the commercial diet significantly improved the weight gain. In contrast Ebeid *et al.* (2011) and Bou *et al.* (2005) observed that the use of oils had no effect on the body weight gain of the poultry birds. Gonzalez-Esquerra and Leeson (2000) also stated that feeding of vegetable oils had no effect on body weight of broilers. The differences in the results of present study with those observed by the scientists mentioned above may be due to the use of different oils. As different plants contain different group of chemicals. This difference in chemicals results in difference in biological activity of each plant.

Though the feed intake by birds do not differ due to addition of plant oils but a statistically significant difference in weight gain was observed. This increase in weight gain of birds due to addition of plant oil in feed have been also reported by many researchers previously (Olomu and Baracos, 1991; Bou *et al*., 2005; Febel *et al*., 2008; Ebeid *et al*., 2011). In present study, improve in weight gain might be due to the improved digestibility as a result of presence of oils in diets of broilers (Balevi and Coskun, 2000).

Table 3: Five week cumulative weight gain, feed intake and feed conversion ratio of broilers fed various levels of different oils

Treatment	Weight Gain (g)	Feed Intake (g)	Feed Conversion Ratio
Olive Oil (0.5%)	1691.4±40.90[a]	3063.3±4.28[a]	1.77±0.04[ab]
Olive Oil (1.0%)	1701.5±11.25[a]	3004.7±22.71[a]	1.72±0.01[a]
Olive Oil (1.5%)	1662.2±66.74[a]	3074.6±24.85[a]	1.81±0.07[ab]
Black Seed Oil (0.5%)	1678.7±14.27[a]	3055.7±48.94[a]	1.77±0.01[a]
Black Seed Oil (1.0%)	1674.1±20.74[a]	3051.1±34.88[a]	1.78±0.02[ab]
Black Seed Oil (1.5%)	1618.2±6.37[a]	3036.5±14.25[a]	1.83±0.00[bc]
Flax Seed Oil (0.5%)	1720.6±56.77[a]	3089.7±41.33[a]	1.75±0.04[a]
Flax Seed Oil (1.0%)	1676.6±55.45[a]	3052.2±54.99[a]	1.77±0.03[a]
Flax Seed Oil (1.5%)	1666.1±6.25[a]	3050.2±28.96[a]	1.78±0.02[ab]
Control	1489±24.89[b]	3031±4.93[a]	1.98±0.03[c]

[a-c] Values in the same column with different superscripts are significantly (P<0.05) different

Table 4: Serum Lipid Profile of Broilers fed Different Levels of Various Oils (mg/dL ±Std. Error)

Treatment	Cholesterol	Triglycerides	High Density Lipoproteins	Low Density Lipoproteins
Olive Oil (0.5%)	188.5±8.79	184.17±9.21	45.67±2.92	105.83±6.06
Olive Oil (1.0%)	171±12.27	189.67±16.47	41.83±0.93	90±8.39
Olive Oil (1.5%)	179.83±46.96	146.17±15.47	42.5±7.52	107.83±36.47
Black Seed Oil (0.5%)	169.83±29.94	187±19.79	45.67±6.13	86.5±24.67
Black Seed Oil (1.0%)	180.5±17.2	232.67±96.26	45.5±4.5	90±25.97
Black Seed Oil (1.5%)	178.33±14.62	124.17±21.68	46.83±2.62	106.33±12.81
Flax Seed Oil (0.5%)	199.5±52.42	194.17±41.21	45.67±6.77	114.5±37.92
Flax Seed Oil (1.0%)	164.67±11.42	171.17±51.56	40.83±4.28	90.17±9.43
Flax Seed Oil (1.5%)	152.33±16.17	136.5±16.77	38.67±3.24	86.33±15.09
Control	192.5±19.29	178.67±25.84	46.33±3.71	110.17±13.29

Table 5: Newcastle Disease and Infectious Bursal Disease titer of broilers fed on various levels of different oils (Mean±Std. Error)

Treatment	Newcastle Disease Titer	Infectious Bursal Disease Titer
Olive Oil (0.5%)	170.67±42.67[ab]	106.67±21.33[a]
Olive Oil (1.0%)	170.67±42.67[ab]	85.33±21.33[a]
Olive Oil (1.5%)	170.67±42.67[ab]	85.33±21.33[a]
Black Seed Oil (0.5%)	213.33±42.67[a]	42.67±10.67[a]
Black Seed Oil (1.0%)	170.67±42.67[ab]	53.33±10.67[a]
Black Seed Oil (1.5%)	128±0.00[abc]	42.67±10.67[a]
Flax Seed Oil (0.5%)	106.67±21.33[bc]	64±0.00[a]
Flax Seed Oil (1.0%)	64±0.00[c]	106.67±21.33[a]
Flax Seed Oil (1.5%)	170.67±42.67[ab]	85.33±21.33[a]
Control	53.33±10.67[c]	53.33±10.67[a]

[a-c] Values in the same column with different superscripts are significantly (P<0.05) different

Table 6: Economics of broilers fed various levels of different oils when cost of chick was Pakistani Rs. = 35/- and cost of management per bird was Rs. = 5.97

Economic Factors	Olive (0.5%)	Olive (1.0%)	Olive (1.5%)	Black Seed (0.5%)	Black Seed (1.0%)	Black Seed (1.5%)	Flax Seed (0.5%)	Flax Seed (1.0%)	Flax Seed (1.5%)	Control
Feed Consumed (Kg/Bird)	3.06	3	3.07	3.05	3.05	3.03	3.8	3.05	3.05	3.03
Feeding Cost/Bird (feed/kg = Rs. 40.6)	124.236	121.8	124.642	123.83	123.83	123.018	125.048	123.83	123.83	123.018
Oil Used Per Bird (ml)	15.3	30	46.05	15.25	30.5	45.45	15.4	30.5	45.75	0
Cost Of Oil / Treatment (Rs.)	9.18	18	27.63	13.725	27.45	40.905	3.85	7.625	11.4375	0
Body Weight/Bird (Kg)	1.69	1.7	1.66	1.67	1.67	1.61	1.72	1.67	1.66	1.48
Income/Bird (Live Weight/Kg = Rs. 128)	216.32	217.6	212.48	213.76	213.76	206.08	220.16	213.76	212.48	189.44
Expenditure Per Bird (Rs.)	174.386	180.77	193.242	178.525	192.25	204.893	169.868	172.425	176.2375	167.988
Profit Per Bird (Rs.)	41.934	36.83	19.238	35.235	21.51	1.187	50.292	41.335	36.2425	25.452

(All prices are in Pakistani Rs.)

In contrast to our findings Maroufyan *et al*. (2012) observed that omega-3 and omega-6 fatty acids in the broiler diets from various oil sources showed significantly improved feed intake. Gonzalez-Esquerra and Leeson (2000) also concluded in their study that the feed intake may be improved by the inclusion of oils in the diet of broilers.

Similarly, in this trial the overall FCRs of the groups supplemented with the vegetable oils were significantly better than the control group. These results are in line with the findings of the Maroufyan *et al.* (2012) and Gardiner (1973). They concluded that the use of vegetable oils such as flaxseed oil, olive oil, black seed oil or fatty acids like omega-3 and omega-6 improves the Feed Conversion Ratio and feed efficiency of the broiler birds. The improved FCR and performance might be due to the presence of PUFA (Polyunsaturated fatty acids) in the vegetable oils. These PUFA decrease the rate of passage of digesta in the digestive system and as a result adsorption of nutrients increased resulting in improved digestibility, which ultimately improved the body weight and performance of birds (Balevi and Coskun, 2000). It is also reported that the feeding linoleic acid and lenoleinic acid did not affect the feed consumption in broilers but helpful for the growth as these fatty acids produce dynamic heat and as a result FCR improves (Schreiner *et al.,* 2005; Mridula *et al.,* 2011).

In present study, supplementation of olive oil, black seed oil and flaxseed oil in feed of broilers did not showed significant effect on lipid profile (serum cholesterol, triglycerides, high density lipoprotein and low density lipoprotein). These findings are similar to the findings of An *et al*. (1997) and Crespo and Garcia (2003). They used different dietary oil and fat sources and found non-significant effects on cholesterol, triglycerides, high density lipoprotein and low density lipoprotein. It is evident that n6 fatty acids have the ability to lower the cholesterol level in the serum where as n3 fatty acids decreases cholesterol and triglycerides (An *et al*., 1997).

Statistical analysis of the experimental data showed significant ($P<0.05$) effect on ND titer after feeding the birds with different levels of plant oils. These results are in line with the findings of Fritsche *et al.* (1991) who reported that the use of different vegetable oils such as corn oil, canola oil and flax seed oil significantly improves the antibody titer. Maroufyan *et al*. (2012) also reported that the combination of tuna oil and sunflower oil significantly improved the titer of ND and IBD. This improvement in titer might be due to presence of PUFA (Polyunsaturated fatty acids) in plant oils. As PUFA affects inflammatory response which ultimately results in improvement in immune response (Koryer and Klasing, 1997). Poly-unsaturated fatty acids may improve the membranes of the immune cells and these fatty acids have the anti-microbial properties. That's why there might be improvement in ND titer (Pathponysiriporn and Scheideler, 2005).

Conclusion

It can be concluded from the present study that the broilers fed with feed having flax seed oil 0.5% showed the cumulative best FCR, no doubt this FCR was non-significantly different ($P>0.05$) with other treatments except black seed oil 1.5% and control. But it was also observed that flax seed oil 0.5% showed better ND titer than control. Other oils showed best ND titer but flax seed oil 0.5% was economical also, so it can be recommended that flax seed oil 0.5% level is the best for production of economical broiler having best FCR and disease resistance.

References

Adil, S., M.T. Banday, G.A. Bhat and M.S. Mir, 2011. Effect of dietary supplementation of organic acids on performance, intestinal histomorphology and serum biochemistry of broiler chicken. *Vet. Med. Int.*, doi: 10.4061/2010/479485

Akhtar, M.S., Z. Nasir and A.R. Abid, 2003. Effect of feeding powered *Nigella sativa* L. Seeds on poultry egg production and their suability for human consumption. *Veterinarski Archiv.*, 73: 181-190

An, B.K., C. Banno, Z.X. Xia, K. Tanaka and S. Ohtani, 1997. Effect of dietary fat sources on lipid metabolism in growing chicks (*Gallus domesticus*). *Comp. Biochem. Physiol. B Biochem. Mol. Biol.*, 116: 119-125

Ashayerizadeh, O., B. Dastar, M.S. Shargh, A. Ashayerizadeh, E. Rahmatrnejad and S.M. Hossani, 2009. Use of garlic (*Allium sativum*), Black cumin seeds Nigella sativa L. and wild mint (*Mentha longifolia*) in broiler chickens diets. *J. Anim. Vet. Adv.*, 8: 1860-1863

Balevi, T. and B. Coskun, 2000. Effects of some oil used in broiler rations on performance and fatty acid compositions in abdominal fat. *Revue. Med. Vet.*, 151: 937-944

Bolukbasi, S.C. and M.K. Erhan, 2005. Effect of dietary conjugated linoleic acid on lying hen performance, egg yolk fatty acids composition and egg quality during refrigerated storage. *J. Amin. Feed Sci.*, 14: 685-693

Bou, R., F. Guardiola, A.C. Barroeta and R. Codony, 2005. Effect of dietary fat sources and zinc and selenium supplements on the composition and consumer acceptability of chicken meat. *Poult. Sci.*, 84: 1129-1140

Crespo, N. and E. Garcia, 2003. Polyunsaturated fatty acids reduce insulin and very low density lipoprotein levels in broiler chickens. *Poult. Sci.*, 82: 1134-1139

Cullen, G.A. and P.J. Wyet, 1975. Quantitation of antibodies to infectious bursal disease. *Vet. Rec.*, 93: 315

Dibner, J.J. and J.D. Richards, 2005. Antibiotic growth promoters in agriculture: history and mode of action. *Poult. Sci.*, 84: 634-643

Du, M. and D.U. Ahn, 2002. Effect of dietary conjugated linoleic acid on the growth rate of live birds and on the abdominal fat content and quality of broiler meat. *Poult. Sci.*, 81: 428-433

Ebeid, T., A. Fayoud, S. Abou-el-soud, Y. Eid and M. Al-habbak, 2011. The effect of omega-3 enriched meat production on lipid peroxidation, antioxidative status, immune response and tibia bone characteristics in Japanese quail. *Czech J. Anim. Sci*., 56: 314-324

El-Sheikh, A.M.A., A.E. Amin and A.A. Khadiga, 1998. The effect of feeding different levels of Niggela sativa seeds on layers performance and egg quality characteristics. *Sudan J. Vet. Sci. Anim. Hus.*, 37: 121-128

Febel, H., M. Mezes, T. Palfy, A. Herman, J. Gundel, A. Lugasi, K. Balogh, I. Kocsis and A.Blazovics, 2008. Effect of dietary fatty acid pattern on growth, body fat composition and antioxidant parameters in broilers. *J. Anim. Nutr*., 92: 369-376

Fritsche, K.I., N.A. Cassity and S.C. Huang, 1992. Dietary (n-3) Fatty acid and vitamin E interaction in rats: Effects on vitamin E status, immune cell prostaglandin E production and primary antibody response. *J. Nutr.*, 122: 1009-1018

Fritsche, K.L., C.A. Nancy and S.C. Huang, 1991. Effect of dietary fat source an antibody production and lymphocyte profile ration in chickens. *Poult. Sci.*, 70: 611-617

Gardiner, E.E., 1973. Comparison of linseed oil with animal tallow as a source of energy in broiler diets. *Can. J. Anim. Sci.*, 53: 557-560

Gokce, R., I. Akhus, M. Yontem, A.Y.A. Gurel, O. Caglayan, S. Bodur and S. Ergun, 2000. Effects of dietary oils on lipoproteins, lipid peroxidation and thromboxane A_2 production in chicks. *Turk. J. Vet. Anim. Sci.*, 24: 473-478

Gonzalez-Esquerra, R. and S. Leeson, 2000. Effects of menhaden oil and flaxseed in broiler diets on sensory quality and lipid composition of poultry meat. *Brit. Poult. Sci.,* 41: 481-488

Growell, P.L., 1999. Prevention and therapy of cancer by dietary monoterpenes. *J. Nutr.*, 129: 7755-7785

Gustafson, R.H. and R.E. Bowen, 1997. Antibiotic use in animal agriculture. *J. Appl. Microbiol.*, 83: 531-541

Haijto, T., K. Hostanska and H.J. Gabius, 1989. Modulatory potency of the B-galactoside-specific lectin from mistletoe extract (Iscador) on the host defense system *in vivo* in rabbits and patent. *J. Cancer Res.*, 49: 4803–4808

Helander, I.M., H.L. Alakomi, K. Latva-Kala, T. Mattila-Sandholm, I. Pol, E.J. Smid, L.G.M. Gorris and A. Von Wright, 1998. Characterization of the action of selected essential oil components on gram-negative bacteria. *J. Agric. Food Chem.*, 46: 3590–3595

Jamroz, D., I. Orda, C. Kamel, A. Wiliczkiewicz, T. Wertelecki and I. Skorupinska, 2003. The influence of phytogenic extracts on performance, nutrient digestibility, carcass characteristics, and gut microbial status in broiler chickens. *J. Anim. Feed Sci. Technol.*, 12: 583–596

Jamroz, D., T. Wertelecki, M. Houszka and C. Kamel, 2006. Influence of diet type on the inclusion of plant origin active substances on morphological and histochemical characteristics of the stomach and jejunum walls in chicken. *J. Anim. Physiol. Anim. Nutr. (Berl.)*, 90: 255–268

Jang, I.S., Y.H. Ko, H.Y. Yang, J.S. Ha, J.Y. Kim, S.Y. Kang, D.H. Yoo, D.S. Nam, D.H. Kim and C.Y. Lee, 2004. Influence of essential oil components on growth performance and the functional activity of the pancreas and small intestine in broiler chicken. *Asian-Aust. J. Anim. Sci.*, 17: 394–400

Jang, I.S., Y.H. Ko, S.Y. Kang and C.Y. Lee, 2007. Effect of a commercial essential oil on growth performance, digestive enzyme activity and intestinal microflora population in broiler chickens. *Anim. Feed Sci. Tehcnol.*, 134: 304-315

Korver, D.R. and K.C. Klasing, 1997. Dietary fish oil alters specific and inflammatory immune responses in chicken. *J. Nutr.*, 127: 2039-2046

Lee, K.W., H. Everts, H.J. Kappert, M. Frehner, R. Losa and A.C. Beynen, 2003. Effects of dietary essential oil components on growth performance, digestive enzymes and lipid metabolism in female broiler chickens. *Brit. Poult. Sci.*, 44: 450–457

MAFF, 1984. *Manual of Veterinary Investigation*, Vol. 2, 3rd edition. Reference Book, Her Majesty Stationary Office, London

Maroufyan, E., A. Kasim, M. Ebrahimi, T.C. Loh, M.H. Bejo, H. Zerihun and F. Hosseni, 2012. Omega-3 polyunsaturated fatty acids enrichment alters performance and immune response in infectious bursal disease challenged broilers. *Lipids Health Dis.*, 25: 11: 15

Mitsch, P., K. Zitterl-Eglseer, B. Kohler, C. Gabler, R. Losa and I. Zimpernik, 2004. The effect of two different blends of essential oil components on the proliferation of *Clostridium perfringens* in the intestines of broiler chickens. *Poult. Sci.*, 83: 669–675

Montgomery, D.C. and G.C. Runger, 2010. *Applied Statistics and Probability for Engineers*, 5th edition. Jhon Wily and Sons, Inc, USA

Mridula, D., S.D. Kaur, S. Nagra, P. Barnwal, S. Gurumayum and K.K. Singh, 2011. Effect of flaxseed meal on growth performance, carcass traits and meat quality in broilers. *Asian J. Anim. Sci.*, 12: 1729-1735

Olomu, J.M. and V.E. Baracos, 1991. Influence of dietary flaxseed oil on the performance, muscle protein deposition, and fatty acid composition of broiler chicks. *Poult. Sci.*, 70: 1403-1411

Pathponysiriporn, U. and S.E. Scheideler, 2005. Effects of dietary ratio of linoleic to linoleinic acid on performance, antibody production and in-vitro lymphocyte proliferation in two strains of leg horn pullet chicks. *Poult. Sci.*, 84: 846-857

Ramadan, M.F., 2007. Nutritional value, functional properties and nutracetuical application of black cumin (*Nigella sativa* L.). *Int. J. Food Sci. Technol.*, 42: 1208-1218

Rezaeipoor, R., S. Saeidnia and M. Kamalinejad, 1999. Immunosuppressive activity of *Achillea talagonica* on humoral immune responses in experimental animals. *J. Ethnopharmacol.*, 65: 273–276

Schettler, G. and E. Nussel, 1975. Massnahmen zur prevention der artherosklerose. *Arb. Med. Soz. Med. Prav. Med.*, 10: 25

Schreiner, M., H.W. Hulan, E. Razzazi-Fazeli, J.B. Bohm and R.G. Moreira, 2005. Effect of different sources of dietary omega-3 fatty acids on general performance and fatty acid profiles of thigh, breast, liver and portal blood of broilers. *J. Sci. Food. Agric.*, 85: 219-226

Steiner, T., 2009. *Phytogenics in Animal Nutrition—natural Concepts to Optimize Gut Health and Performance*. Nottingham University Press, Nottingham, UK

Waldroup, P.W., E.O. Oviedo-Rondon and C.A. Fritts, 2003. Comparison of Bio-Mos and antibiotic feeding programs in broiler diets containing copper sulfate. *Int. J. Poult. Sci.*, 2: 28-31

Windisch, W., K. Schedle, C. Plitzner and A. Kroismayr, 2008. Use of phytogenic products as feed additives for swine and poultry. *J. Anim. Sci.*, 86: 140-148

Zarei, M., M. Ehsan and M. Torki, 2011. Productive performance of laying hens fed wheat-based diets included olive pulp with or without a commercial enzyme product. *Afr. J. Biotechnol.*, 10: 4303-4312

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