

Effect of vegetable oils on growth, lipid profile, and immunologic response in broiler chicken fed isoenergetic diet

Efecto de los aceites vegetales sobre el crecimiento, el perfil lipídico y la respuesta inmune en pollos de engorde alimentados con dietas isoenergéticas

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Received: 02/18/2011 First reviewing ending: 02/01/2012 First review received: 02/20/2012 Accepted: 02/25/2012

ABSTRACT

In the past, the main objective of the poultry meat industry was to improve body weight and feed efficiency of the birds. Modernly, there are other parameters that are essential to be taken into consideration in poultry industry such as low cholesterol deposition on the body. For this aim, the present study was conducted to evaluate the effects of dietary supplemental canola and olive oils alone or combined on lipid and cholesterol of serum and tissues in broiler chicks. A total of 120 male chicks were randomly distributed into four treatments (30 birds each) with three replications (10 birds each) for each treatment, a completely randomized design was used ($p \leq 0.05$). The blood and tissues samples were taken at the end of experiment (42 days) and values for triacylglycerol (TAG), total lipid, High-Density-Lipoprotein cholesterol (HDL-c), LDL-c, very low density lipoprotein cholesterol (VLDL-c) and free fatty acids levels were determined. In addition, feed intake, growth rate and feed conversion ratio, and immune response against Newcastle disease (ND) and sheep red blood cell (SRBC) were also determined. The results revealed that, dietary supplementation of either canola or olive oils to broiler chick increased immune response to ND, SRBC, final live body weight and daily gain compared to control without affecting TAG, LDL-c, VLDL-c and saturated fatty acids (SFA) when compared with the control. Interestingly, combined administration of both oils was more effective in such effect than if they supplemented alone.

Key words: broiler chicks, lipid profile, canola and olive oils

RESUMEN

En el pasado, el objetivo principal de la industria de la carne de ave fue mejorar el peso corporal y la eficiencia alimenticia de las aves. Modernamente, existen otros parámetros que son esenciales para tener en cuenta en la industria avícola tales como la deposición de bajo colesterol en el cuerpo. Para este fin, se realizó el presente estudio para evaluar los efectos de los aceites de canola y de oliva administrados solos o en una emulsión combinada sobre los lípidos y colesterol del suero y de los tejidos de pollos de engorde. Un total de 120 pollos machos se distribuyeron al azar en cuatro tratamientos (30 aves cada uno) con tres repeticiones (10 aves cada una) para cada tratamiento, el diseño estadístico fue completamente aleatorizado ($p \leq 0,05$). Las muestras de sangre y de los tejidos se tomaron al final del experimento (42 días) y se determinaron los valores de triacilglicerol (TAG), lípidos totales, colesterol de alta densidad de las lipoproteínas (HDL-c), colesterol de baja densidad de las lipoproteínas (LDL-c), colesterol de muy baja densidad de las lipoproteínas (VLDLc) y los patrones de ácidos grasos libres. Además, también se determinaron el consumo de alimentos, la tasa de crecimiento y la eficiencia de conversión alimenticia y también se determinó la respuesta inmune contra la enfermedad Newcastle (EN) y eritrocitos de oveja (SRBC). El presente estudio reveló que, suplementación dietética con aceite de canola o de oliva a los pollos de engorde aumentó la respuesta inmune a EN, SRBC, el peso vivo corporal y la ganancia de peso en la mayoría de los puntos de medición, mientras que ellos no afectaron los valores de TAG, LDL-c, VLDL-C y los ácidos grasos saturados en comparación con el control. Interesantemente, la administración combinada de ambos aceites fue más efectiva en ese efecto que si ellos se suplementaron de forma individual.

Palabras clave: pollo de engorde, perfil lipídico, aceite de canola y de oliva.

INTRODUCTION

It well known that, continuous increasing in population led to increasing demands of animal

proteins. Consequently, broiler industry is increasing dramatically all over the world. However, the adverse effect of animal protein is hypercholesterolemic causing heart and arteries diseases. It is possible to

control fatty acid profile in blood and meat of birds as a result of transferring certain components from the diet. It is generally accepted that dietary saturated fatty acids directly are related to plasma cholesterol levels. In the contrary, the consumption of Polyunsaturated Fatty Acids (PUFA) has been shown to have beneficial effects on human health (Kinsella *et al.*, 1990; Mensink and Katan, 1995). Oils rich in oleic acid (monounsaturated fatty acid; MUFA) were found also to be effective in lowering plasma cholesterol level. Thereby, the reasons for the increasing level of polyunsaturation in chicken meat are to reducing the intake of saturated fatty acids (SFA) because of its relationship with the development of cardiovascular diseases (Krauss *et al.*, 2001) and the use of animal fats has been reduced approximately in the World, in favor of vegetable oils that are more polyunsaturated with taking consideration of ecommendation of Conner *et al.*, (1986).

The use of vegetable oils in poultry diet increases long-chain n-3 FA level in meat (Chanmugam *et al.*, 1992; Pinchasov y Nir 1992). Vegetable sources, such as canola and olive oil may clearly increase the n-3 FA content in the form of linolenic acid (LNA), which is a precursor of the whole n-3 family to enhance the conversion to longer-chain n-3 FA from their precursors and to increase the nutritional quality of poultry meat.

Rapeseed oil (canola oil) is used as vegetable oil for cooking and in salads. It contains 55% of the oleic acid (MUFA), 25% linoleic acid and 10% alpha-linoleic acid (PUFA), and only 4% of SFA. Recommendations say that the diet should contain about 30% of calories as fat made up of less than 10% saturated fatty acids, and they consider canola oil having benefits on health in general (Dupont *et al.*, 1989). Canolol (4-vinyl-2, 6-dimethoxyphenol) is a highly potent free radicals scavenger, isolated from crude canola oil (rapeseed). After roasting the seed, the canolol content increases but its content is low in highly purified canola oil. Researcher found its potency was much greater than that of well-known antioxidants including alpha-tocopherol and vitamin C. Researchers found that canola oil has benefits on total cholesterol and low density lipoprotein cholesterol (LDL-C). Lipid-lowering diets containing either rapeseed oil or olive oil may have benefits on serum lipoprotein concentration in hyperlipidemic subjects.

Olive oil contains monounsaturated fatty acids, oleic acid with palmitic and lenoleic acids in smaller proportions (Pharmaceutical index 1979). Conner *et al.*, (1986) recommended that the dietary intact of polyunsaturated fat should not be increased in human beings due to its high caloric content and the association of polyunsaturated fat with the development of gall stones, breast cancer and cancers of the colon. Olive oil, however, has not been as consistent in this effect as other oleic-acid-rich oils (Sirtori *et al.*, 1986; Mata *et al.*, 1992). Researchers found that canola oil and olive oil reduced total serum cholesterol, low-density lipoprotein and the ratio between low-density and high-density lipoprotein cholesterol to the same extent in hyperlipidemic patients. However, there was a slightly greater decrease in low-density lipoprotein cholesterol with the diet containing rapeseed (canola) oil than with the olive oil diet. Some health organizations do not recommend use of canola oil in infant formula because of accumulation of triglyceride in heart due to erucic acid (22:1n-9) in the oil (Green *et al.*, 2000).

The aim of the present study was to determine the effect of a rapeseed oil (2%) and/or olive oil (2%) mixture on the profile of plasma lipid, lipoprotein and fatty acids profile in broiler chicken fed high fat diet.

MATERIALS AND METHODS

The current experiment was conducted using day old Ross broiler chicks which were obtained from a commercial hatchery (120, one day old male broiler chicks) and were placed in floor pens of 1.65 x 0.671 m with 10 birds per pen. Feed and water were provided *ad libitum*. The experiment arrangement consisted of 120 birds divided onto 4 treatments (30 birds each) with 3 replications (10 birds each) per each treatment:

Treatment 1: isoenergetic diet (control)

Treatment 2: isoenergetic diet and canola oil 2%

Treatment 3: isoenergetic diet and olive oil 2%

Treatment 4: isoenergetic diet and canola oil 1% and olive oil 1%

Experimental diets were prepared according to the NRC recommendation (NRC, 1994). These diets were formulated to meet nutrient requirements

according to NRC. Diets were containing the same level of methionine, lysine, vitamins and minerals. The treatment diets were isoenergetic and isonitrogenous treatment.

The chicks were weighed at the start of the experiment and during the experiment, live weight and total feed consumption per pen were recorded and feed conversion ratio was calculated at 7, 14, 21 and 42 days of the experiment. Mortality was also recorded for each treatment. Two birds from each replicate were sacrificed after slaughtering at day 42. Serum, liver, and muscle tissues were frozen at -20°C until the time of analysis of TAG, total lipid, HDL-c, LDL-c and free fatty acids content.

Liver, omentum and muscles were dissected, washed thrice in cold saline, blotted and preserved in freezer. One gram of each tissue was homogenized in 30 ml of extraction mixture of chloroform and methanol in ratio of 2: 1 in homogenizer for 10 minutes at 1500 r.p.m. and was filtered. The desiccated lipids were dissolved in 500ul chloroform for estimation of tissue total lipids, triglyceride and total cholesterol with commercially available kits.

The obtained sera were used for spectrophotometric determination of the activities of aspartate transaminase (AST) and alanine transaminase (ALT) as directed by Reitman and Frankle (1957). In addition, serum total protein, albumin and globulin values were determined spectrophotometrically as implied by the methods of Doumas *et al.*, (1981), Reinhold (1953) and Coles (1974), respectively. Serum urea and creatinine were determined according to the method described by Tabacco *et al.*, (1979) and Henry (1984), respectively. Furthermore, the obtained sera were used also for spectrophotometric analysis of serum triacylglycerol (TAG), total cholesterol and high density lipoprotein cholesterol (HDL-c) by using of enzymatic method of

spin react kits according to the methods of Sidney and Bernard (1973), Zak *et al.*, (1954) and Lopes-Virella *et al.*, (1977), respectively. Very low density lipoprotein cholesterol (VLDL-c) was calculated by division of TAG by 5 (mg/dl) while the low density lipoprotein cholesterol (LDL-c) was calculated (mg/dl) by subtracting the sum of HDL-c and VLDL-c from total cholesterol (Bauer 1982).

The obtained data on growth, feed utilization, biochemical and immunological parameters were subjected to one way ANOVA using the treatment as the main effect. All tests will perform using computer package of the statistical analysis system (SAS, 2000). The level of significance was ($p \leq 0.05$)

RESULTS AND DISCUSSION

Body weight, feed intake and feed conversion are shown in Tables 1, 2 and 3, respectively. Both oils increased significantly ($p \leq 0.05$) body weight compared to the control at the fifth week of age while no significant differences ($p > 0.05$) among other treatments at the same week. There were not any significant differences ($p > 0.05$) among all groups at any other weeks through the experimental period. In addition mixed oils group showed superiority over control group in food intake at fifth week of age. On spite of differences in body weight and feed intake there were not any differences in feed conversion rate among all treatments starting from the second week of age.

Colorimetric analysis of serum revealed that, TAG, total cholesterol and VLDL-c values were not significantly different among treatments ($P \leq 0.05$) (Table 4).

Spectrophotometric analysis of serum samples indicated that, during the whole experimental period broiler chicks fed diet supplemented with canola oil and olive oil either alone or in combination

Table 1. Body weight (g) of broiler chicks supplemented with canola and/or olive oils.

Treatments	Body weight (g)					
	Days of age					
	7	14	21	28	35	42
Control (ID)	104	287	531	901 ^{ab}	1335 ^b	1750
ID + CO 2%	104	287	570	904 ^{ab}	1380 ^{ab}	1820
ID + OO 2%	100	283	565	890 ^b	1393 ^{ab}	1720
ID + CO 1% + OO 1%	98	303	570	911 ^a	1432 ^a	1865

ID: Isoenergetic diet; CO: canola oil and OO: olive oil

Within columns, means with different letters differs significantly at $p \leq 0.05$

showed significant differences ($p \leq 0.05$) in total protein, albumin uric acid, and ALT (Table 5). Values of protein patterns showed significant differences ($p \leq 0.05$) between control and single oil supplement groups. Albumin concentration was the lowest for canola oil supplement compared to other treatments. Both mixed oils supplement and olive oil supplement harvested the lowest values for the uric acid.

The results concerning the effect of canola and olive oils on the activities of ALT indicated that, there were significant differences ($P \leq 0.05$) in the activities of ALT in broiler chicks fed ration containing canola and/or olive when compared with the control (Table 5). Both oils and canola oil supplement recorded the highest enzymatic activity compared to control, while olive oil supplement recorded the lowest enzymatic activity compared to control.

The results concerning immune response against Newcastle disease and SRBCs were remarkable. Over 5 of 6 weeks of the experimental period the mixed oil treatment recorded the highest significant ($P \leq 0.05$) antibody titer against ND compared to the control group, this trend is also recorded for the antibody titer against SRBC at 7 days post inoculation (Table 6). Other treatments differ from each other and from control variably.

The present finding demonstrated that the substitution of canola oils and olive oil either alone or in combination modulate immune response to ND and SRBC antigens, in addition both oils affect the body weight by increase the final live weight at the marketing time. The physiological activity indicators showed the significance of oil supplement on the physiological performance of the broilers.

Table 2. Feed intake (g) of broiler chicks supplemented with canola and/or olive oils.

Treatments	Feed intake (g)					
	Days of age					
	7	14	21	28	35	42
Control (ID)	113	240	434	661 ^{ab}	1016 ^b	1199
ID + CO 2%	112	240	465	663 ^{ab}	1051 ^{ab}	1247
ID + OO 2%	108	236	461	653 ^b	1061 ^{ab}	1185
ID + CO 1% + OO 1%	106	254	465	668 ^a	1090 ^a	1278

ID: Isoenergetic diet; CO: canola oil and OO: olive oil

Within columns, means with different letters differs significantly at $p \leq 0.05$

Table 3. Feed conversion ratio of broiler chicken supplemented with canola and/or olive oils.

Treatments	Feed conversion ratio					
	Days of age					
	7	14	21	28	35	42
Control (ID)	1.26 ^{ab}	1.32	1.78	1.79	2.36	2.89
ID + CO 2%	1.27 ^a	1.31	1.65	1.98	2.22	2.85
ID + OO 2%	1.25 ^b	1.30	1.64	2.00	2.11	3.74
ID + CO 1% + OO 1%	1.25 ^b	1.24	1.74	1.95	2.09	2.95

ID: Isoenergetic diet; CO: canola oil and OO: olive oil

Within columns, means with different letters differs significantly at $p \leq 0.05$

Table 4. Effect of canola and/or olive oils on serum profiles of TAG, cholesterol and VLDL-c of broiler chicken.

Parameters (mg dL ⁻¹)	Control (ID)	ID + CO 2%	ID + OO 2%	ID + CO 1% + OO 1%
TAG	47.94	54.04	58.16	40.71
Total cholesterol	74.46	94.74	71.99	81.94
VLDL-c	9.58	10.80	11.63	8.14

ID: Isoenergetic diet; CO: canola oil and OO: olive oil

TAG: Triacylglycerol and VLDL-c: Very low density lipoprotein cholesterol

Table 5. Effect of canola and/or olive oils on protein patterns and liver and kidney functions in broiler chicken.

Parameters	Control (ID)	ID + CO 2%	ID + OO 2%	ID + CO 1% + OO 1%
Total protein (g dL ⁻¹)	1.80 ^{ab}	1.34 ^c	1.48 ^{bc}	2.15 ^a
Albumin (g dL ⁻¹)	1.47 ^a	1.47 ^a	1.06 ^b	1.34 ^{ab}
ALT (μ L ⁻¹)	8.86 ^b	10.83 ^a	6.43 ^c	10.93 ^a
Uric acid (mg dL ⁻¹)	3.68 ^a	3.82 ^a	2.12 ^b	2.25 ^b
Creatinine (mg dL ⁻¹)	1.55	1.36	1.64	1.79

ID: Isoenergetic diet; CO: canola oil; OO: olive oil and ALT: Alanine transaminase

Within rows, means with different letters differs significantly at $p \leq 0.05$

Table 6. Effect of canola and/or olive oils on immune response to ND and SRBC of broiler chicken.

Parameters (log ₂)	Control (ID)	ID + CO 2%	ID + OO 2%	ID + CO 1% + OO 1%
ND 1	3.0 ^c	3.2 ^{bc}	4.0 ^{ab}	4.1 ^a
ND 2	5.0 ^b	5.2 ^b	5.7 ^b	6.6 ^a
ND 3	4.1 ^c	4.2 ^c	5.6	7.4 ^a
ND 4	6.2	6.2	7.0	7.1
ND 5	7.0 ^c	7.2 ^{bc}	8.1 ^{ab}	8.2 ^a
ND 6	6.5 ^b	6.3 ^b	7.2 ^{ab}	8.1 ^a
SRBC 3	2.0	2.3	2.1	2.0
SRBC 7	6.0 ^b	6.4 ^{ab}	7.0 ^a	7.1 ^a
SRBC 10	2.6	2.6	3.0	3.1

ID: Isoenergetic diet; CO: canola oil; OO: olive oil and ALT: Alanine transaminase

Within rows, means with different letters differs significantly at $p \leq 0.05$

ND 1, 2, 3, 4, 5 and 6 are antibodies titer against ND at 1, 2, 3, 4, 5 and 6 weeks of age.

SRBC 3, 7 and 10 are antibodies titer against SRBC 3, 7 and 10 days post injection.

ACKNOWLEDGMENTS

The authors are grateful for and acknowledges Deanship of Scientific Research for financial support and facilitate this research work. Thanks are also extended to Agriculture Research Station of King Faisal University for their support in conducting this research. The authors also acknowledge the technical assistance of all poultry research unit staff at the Agriculture Research Station of King Faisal University.

LITERATURE CITED

- Bauer, A., 1982. Enzymatic determination of cholesterol in egg yolk. *J. Assoc. Anal. Chem* 65 (9): 1222-1224.
- Chanmugam, P.; M. Boudreau, T. Boutte, R. S. Park, J. Hebert, L. Berrio and D. H. Hwang. 1992. Incorporation of different types of n-3 fatty acids into tissue lipids of poultry. *Poultry Science* 71 (3): 516-521.
- Coles, E. H. 1974. *Veterinary Clinical Pathology*. W.B. Saunders Company, Philadelphia and London
- Connor, S. L.; J. R. Gustafson, S. M. Artaud Wild, D. P. Flavell, C. J. Classick Kohn, L. F. Hatcher and W. E. Connor. 1986. The cholesterol/saturated-fat index: an indication of the hypercholesterolaemic and atherogenic potential of food. *Lancet* 1(8492): 1229-1232.
- Doumas, B.T., Bayson, D.D., Carter, R.J., Peters, T., Schaffer, R. 1981. Estimation of total serum protein. *Clin. Chem.* 27: 1642-1643.
- Dupont, J.; P. J. White, K. M. Johnston, H. A. Heggtveit, B. E. McDonald, S. M. Grundy and A. Bonanome. Food safety and health effects of canola oil. *Journal of the American College of Nutrition* 8 (5): 360-375
- Green, T. J. and S. M. Innis. 2000. Low erucic acid canola oil does not induce heart triglyceride

- accumulation in neonatal pigs fed formula. *Lipids* 35 (6): 607-612.
- Henry, R. J. 1946. *Clinical Chemistry*. Harper and Row publishers. New York. USA. P. 181.
- Katan, M. B., 1995. Commentary on the Supplement trans fatty acids and coronary heart disease risk. *American Journal of Clinical Nutrition* 62 (3): 518-519.
- Kinsella. I.E., B. Lokesh and R.A. Stone, 1990. Dietary n-3 polyunsaturated fatty acid and amelioration of cardiovascular disease: Possible mechanisms. *I. Food. Sci. Technol.*, 52: 1-28.
- Krauss, R. M.; R. H. Eckel, B. Howard, L. J. Appel, S. R. Daniels, R. J. Deckelbaum, J. W. Erdman, Jr, P. Kris-Etherton, I. J. Goldberg, T. A. Kotchen, A. H. Lichtenstein, W. E. Mitch, R. Mullis, K. Robinson, J. Wylie-Rosett, S. St. Jeor, J. Suttie, D. L. Tribble and T. L. Bazzarre. 2001. AHA Scientific Statement: AHA Dietary Guidelines Revision 2000: A Statement for Healthcare Professionals From the Nutrition Committee of the American Heart Association. *Journal of Nutrition* 131: 132-146.
- Lopes-Virella, M. F.; P. Stone, S. Ellis and J. A. Colwell. 1977. Cholesterol determination in high-density lipoproteins separated by three different methods. *Clinical Chemistry* 23 (5): 882-884.
- Mata, P., Alvarez-Sala, L. A., Rubio, M. J., Nuno, J. & De Oya, M. (1992). Effects of long-term monounsaturated vs polyunsaturated-enriched diets on lipoproteins in healthy men and women. *American Journal of Clinical Nutrition* 55, 846-850.
- Mensink, R. P. & Katan, M. B. (1989). Effect of a diet enriched with monounsaturated or polyunsaturated fatty acids on level of low-density lipoprotein cholesterol in healthy women and men. *New England Journal of Medicine* 321, 436-441.
- National Research Council (NRC). 1994. *Nutrient requirements of poultry*. 9th rev. ed. National Academy Press, Washington, D. C. USA.
- Pharmaceutical Codex. 1979. Olive oil. *In: The British Pharmaceutical codex* 11th ed. London. Pharmaceutical Press, p 615.
- Pinchasov, Y. and I. Nir. 1992. Effect of dietary polyunsaturated fatty acid concentration on performance, fat deposition, and carcass fatty acid composition in broiler chickens. *Poultry Science* 71 (9): 1504-1512.
- Reinhold, R. P. 1953. Determination of serum albumin. *Clin. Chem.* 21: 1370-1372.
- Reitman S.; and S. Frankel. 1957. Standard methods in *Clinical Chemistry*. *Am J Clin Chem* 28: 56-9.
- Statistical Analysis System (SAS). 2000. *User's Guide*. Statistics, SAS Institute Inc. Cary, North Carolina, USA.
- Sidney P. G. and Bernald R. 1973. Improved manual Spectrometric procedure for determination of serum triglyceride. *Clin Chem.* 19 (9): 1077-1078.
- Sirtori, C. R., Tremoli, E., Gatti, E., Montanari, G., Sirtori, M., Colli, S., Gianfranceschi, G., Maderna, P., Dentone, C. Z., Testolin, G. & Galli, C. (1986). Controlled evaluation of intake in the Mediterranean diet: comparative activities of olive oil and corn oil on plasma lipids and platelets in high-risk patients. *American Journal of Clinical Nutrition* 44, 635-642.
- Tabacco, A.; F. Meattini, E. Moda and P. Tarli. 1979. Simplified enzymatic/colorimetric serum urea nitrogen determination. *Clinical Chemistry* 25: 336-337.
- Zak, B.; Dickenman, R. C., White, E. G., Burnett, H., and Cherney, P. J. 1954. Rapid estimation of free and total cholesterol. *Am. J. Clin. Path.* 24: 1307-1315.