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Effect of Feeding Olive Leaves Extract (Oleuropein) on the Performance, Nutrient Utilization, Small Intestine and Carcass Characteristics of Broiler Chickens

[1]Tarek M. Shafey, [2]Ibrahim M. Al-Ruqaei and [1]Saud I. Almufarij
[1]Department of Animal Production, College of Food and Agriculture Sciences,
King Saud University, Riyadh, Saudi Arabia
[2]The National Center for Agriculture Technology,
King Abdulaziz City for Science and Technology, P.O. Box 6086,
11442 Riyadh, Saudi Arabia

Abstract: The effects of dietary levels of olive leaves extract (0, 1.8, 3.6 and 6.25 g Oleuropein (OLE)/kg) on the performance (weight gain, feed intake and feed conversion ratio) from 1-35 days of age, nutrient utilization (apparent nitrogen retention and corrected nitrogen apparent metabolizable energy between 18 and 21 days of age, Small Intestine (SI) measurements (weight, length and weight/length (thickness)) at 21 and 36 days of age and carcass characteristics at 36 day of age of broiler chickens were investigated. Dietary supplementation of OLE did not influence the performance at 35 day of age, nutrient utilization between 18 and 21 days of age, measurements of SI at 21 and 36 days of age, weights of eviscerated carcass and its components of thigh, drums, wings, breast and back as proportions of live weight. Chickens at 36 day of age had significantly ($p<0.01$) higher body and carcass weights and SI measurements than those of chickens at 21 day of age. The addition of 6.25 g OLE/kg diet significantly ($p<0.05$) reduced carcass abdominal fat when compared with those of the control diet. It was concluded that OLE supplementation with up to 6.25 g kg^{-1} of broiler diets did not influence performance, nutrient utilization, SI measurements and weights of eviscerated carcass components but reduced abdominal fat of chickens.

Key words: Broiler, oleuropein, performance, nutrients utilization, small intestine, carcass characteristics

INTRODUCTION

There is a growing interest in the use natural products in animal feed as feed additives (Bozkurt *et al*., 2009). Herbs, spices and various plant extracts or essential oils were used in poultry diets to control diseases and improve performance, carcass characteristics and economic return (Vogt, 1991; Huang *et al*., 1992; Dickens *et al*., 2000; Al-Harthi, 2002; Abd El-Latif *et al*., 2002; Emadi and Kermanshahi, 2006; Singh *et al*., 2007). Recently, Eevuri and Putturu (2013) reviewed the use of some herbal preparations in broiler feeds. These feed additives include *Acacia nilotica* (Schragle and Muller, 1990); *Aloe* sp. (Boudreau and Beland, 2006) Chinese medicinal herbs (Huang *et al*., 1992); cinnamon (Zita *et al*., 2009), combretum mole, pepper, khasanda kwata, imbululusi concoctions, sisal, omusirangokho, tithonia concoctions and neem (Okitoi *et al*., 2007; Bonsu *et al*., 2012) extracted oils from thyme, mace and caraway or coriander, garlic and onion (Vogt, 1991), garlic (Choi *et al*., 2010), ginger (Ali *et al*., 2008), oregano (Bampidis *et al*., 2005), tulsi (Gupta and Charan, 2007), thyme, black cumin, dianthus or fennel (Abd El-Latif *et al*., 2002), turmeric (Mehala and Moorthy, 2008; Al-Kassie *et al*., 2011). Combinations of two or more of these additives have been used in various trials to maximize the benefits from using them (Mehala and Moorthy, 2008).

The olive tree (*Olea europaea*), native to Mediterranean region, produces a variety of natural phytochemicals compounds with medicinal properties (Privitera, 1996). Among the phytochemicals that have interest is Oleuropein (OLE) a substance found in the olive leaf extract (Panizzi *et al*., 1960). OLE is successful against a broad spectrum of microbial agents including viruses, bacteria and even parasites and can considered one of the most useful and safe natural anti-microbial herbal extracts (Walker, 1997; Aziz *et al*., 1998; Bisignano *et al*., 1999).

There is a lack of information regarding the effects of OLE on the performance of broiler chickens. Therefore, the objectives of this study were to investigate the effects

Corresponding Author: Tarek M. Shafey, Department of Animal Production, College of Food and Agriculture Sciences, King Saud University, P.O. Box 2460, 11451 Riyadh, Saudi Arabia

of dietary supplementation of OLE on the performance, nutrients utilization, intestinal measurements and carcass characteristics of broiler chickens.

MATERIALS AND METHODS

Olive Leaves (OL) were collected from olive trees (*Olea europaea*) and were sun dried by spreading on drying trays and exposing directly to solar radiation and natural air circulation. Dried OL were then ground to pass a 1.4 mm steel screen using a grinder (Moline M-06, Italy) and OLE was extracted according to Markin *et al*. (2003). Briefly, OLE was extracted in water at a 20% concentration (w/v, 200 g OL powdered materials per liter of water) for 24 h. The extract was filtered and evaporated to dryness under room temperature. The residue was dissolved in methanol and filtered through a 0.40 μm. The extract was injected into the HPLC and concentration of OLE was measured in triplicate. The OLE content of OL was 12.5 mg g^{-1} OL.

A total of 220, day old Ross male broiler chickens were individually weighed and randomly sorted into 44 replicates to minimize differences in initial body weight among replicates, with 5 birds each. Birds were housed in electrically heated battery cages. Lighting was incandescent and continuous throughout the experimental period. Eleven replicates were randomly assigned to either one of four starter diets to 21 days, followed by finisher diets to 35 days of age. The four experimental diets contain OLE levels of 0, 1.8, 3.6 and 6.25 mg kg^{-1}. Acid washed sand (10 g kg^{-1}) was added to the diet as a source of Acid Insoluble Ash (AIA). Sand with particle size of 40 mesh (~595 μm) was soaked in 4 N HCl for a day and washed thoroughly to remove all acid. Sand was then oven dried at 100°C, cooled and stored for inclusion in the diet. The experimental diets were formulated to be isocaloric and isonitrogenous (Table 1). Feed and water were available *ad libtium*. Excreta samples were collected every 12 h from 18-21 days of age from three randomly selected replicates of birds from each dietary treatment. Trays lined with plastic sheets were fitted under the cages for excreta collection. After removing feathers, feed residues and other contamination sources, excreta from each experimental unit were collected early in the morning and in the evening. Feed and excreta samples were oven dried at 80°C and finely ground prior to analysis. Nitrogen (N) in both feed and excreta was determined by Kjeldahl procedure (AOAC, 1990), gross energy by using an adiabatic bomb calorimeter and AIA by Dourado *et al*. (2010). The calculations of nutrient utilization as described by Scott *et al*. (1982) were as follows:

Table 1: Composition of the basal diets (g kg^{-1})

Ingredients	Starter phase (1-21 days of age)	Finisher phase (21-35 days of age)
Soybean meal	375.50	302.7
Corn	258.50	329.9
Wheat	200.00	200.0
Corn oil	63.30	64.0
Wheat bran	50.00	50.0
Dicalcium phosphate	16.10	16.6
Limestone	14.10	14.3
Sand	10.00	10.0
Premix[1]	5.00	5.0
Salt	4.30	4.3
DL-methionine	1.50	2.3
L-lysine	0.50	0.4
Choline	1.20	1.2
Analysis		
ME (kcal g^{-1})[2]	2.95	3.0
CP (N%×6.25)[3]	23.00	20.0
Lysine (g kg^{-1})	12.00	11.0
Met + Cys (g kg^{-1})[4]	9.00	9.0
Ca (g kg^{-1})	10.00	9.0
AP (g kg^{-1})[5]	5.00	4.5

[1]The composition of vitamins and minerals in the premix (per ton of diet): vitamin A, 6000,000 IU; vitamin D, 1500,000 IU; vitamin E, 20,000IU; vitamin K 1,000 mg; vitamin B1, 1 mg; vitamin B2, 3000 mg; vitamin B6, 2000 mg; vitamin B12, 10 mg; niacin, 20,000 mg; folic acid, 500 mg; pantothenic acid 5,000 g; biotin, 50 mg; antioxidant, 60.000 mg; cobalt, 100 ppm; copper, 5,000 ppm; iodine, 500 ppm; iron, 20.000 ppm; manganese, 40.000 ppm; selenium, 100 ppm; zinc, 30.000; [2]Metabolizable energy; [3]CP = Crude protein; [4]Met + Cys = Methionine + Cysteine; [5]AP = Available Phosphorus was calculated on the basis of 30% availability of phosphorus in plant products

$$\text{Apparent N Retention (ANR)} = 100-(100\times F^{*}\times F^{**})$$

Where:

F* = Percentage of AIA in diet/Percentage AIA in excreta

F** = N in excreta (mg g^{-1})/N in diet (mg g^{-1})

N corrected apparent metabolizable energy:

$$\text{Energy } (AME_n) = \text{Energy/g diet-Excreta energy/g diet+ } 8.22\times\text{mg N retaind/g diet}$$

Where:

Excreta energy/g diet = Energy per g excreta×F*

N retention (mg g^{-1} diet) = N in diet (mg g^{-1})−N in excreta (mg g^{-1})×F*

At 21 and 36 days of age, four and seven birds per diet were randomly selected, respectively and processed at King Saud University to determine Small Intestine (SI) measurements, processing yields and carcass quality. Intestinal measurements were determined at 21 and 36 days of age and carcass characteristics were determined at the end of the experiment at 36 day of age. Birds were weighed, killed by cervical dislocation after 9 h of feed and water deprivation, bled, scalded, defeathered in a rotary picker and eviscera and abdominal fat were removed. Body components, edible offal (liver

plus heart plus gizzard) and SI were harvested for measurements. The SI segments of duodenum (from the pylorus to distal point of entry of bile duct), jejunum (from entry of the bile ducts to Meckel's diverticulum) and the ileum (from Meckel's diverticulum to the ileocecal junction) were removed and then gently flushed with saline solution to remove any intestinal contents remained. Weight and length of each segment were recorded. Weight of intestinal segment was expressed on the basis of absolute value (g) or as a percentage of live weight (%). Data from carcass weight, abdominal fat and cut parts (back, breast, wings, thigh and drumstick) were recorded.

Measurements were made of body weight gain, feed intake and Feed Conversion Ratio (FCR) during the starter, finisher and whole experimental periods (1-21, 22-35 and 1-35 days of age, respectively), SI segments measurements (weight, length and thickness (weight/length of duodenum, jejunum and ileum)) at 21 and 36 days of age and carcass composition at 36 day of age. Data collected were subjected to analysis of variance using GLM procedures (SAS, 2005). Where significant variance ratios were detected, differences between treatment means were tested using the Least Significant Difference (LSD) procedures.

RESULTS AND DISCUSSION

The effects of dietary levels of OLE on the performance, nutrient utilization, SI measurements and carcass characteristics of broiler chickens are shown in Tables 2-5, respectively. Dietary supplementation of OLE did not influence the performance (body weight gain, feed intake and FCR) of chickens during the starter, finisher and the whole experimental periods (1-21, 22-35 and 1-35 days of age, respectively, Table 2), nutrient utilization (ANR and AMEn, Table 3) between 18 and 21 days of age and weights of eviscerated carcass and edible offal (liver plus heart plus gizzard) as proportions of live body weight, eviscerated carcass components of thigh, drums, wings, breast and back at 36 day of age (Table 4) and the measurements of SI and SI segments (weight, length and weight/length) at 21 and 36 days of age (Table 5).

The addition of 6.25 g OLE/kg diet significantly ($p<0.05$) reduced carcass proportions of abdominal fat when compared with that of the control diet and neck when compared with those fed 1.8 and 3.6 g OLE/kg diets (6.25<1.8 = 3.6 g OLE/kg diet). Chicken at 36 day of age had significantly ($p<0.01$) higher body and carcass weights and SI and SI segments measurements than those of chickens at 21 day of age. There were significant interactions ($p<0.05$) between dietary level of OLE and age of chickens on live and carcass weights. There was no significant difference in live and carcass weights among dietary levels of OLE at 21 day of age. Whilst the addition of 6.25 g OLE/kg diet significantly ($p<0.05$) reduced live and carcass weights of chickens at 36 day of age when compared with that of the control or 3.6 g OLE/kg diet (0 = 3.6>6.25 g OLE/kg diet).

Results from this study indicated that the addition of OLE to the diet of broiler chickens did not influence the performance (body weight gain, feed intake, FCR), nutrient utilization (ANR and AMEn), SI measurements and weights of eviscerated carcass and its components of thigh, drums, wings, breast and back. To our knowledge, no information is available concerning the effects of dietary supplementation of OLE on the performance and carcass characteristics of chickens. However, OLE has been shown to boast a wide range of pharmacological activities, including antioxidant (Visioli *et al*., 2002), anti-inflammatory (Visioli *et al*., 1998), anti-atherogenic (Carluccio *et al*., 2003), anti-cancer (Owen *et al*., 2000), antimicrobial (Aziz *et al*., 1998; Bisignano *et al*., 1999; Fredrickson, 2000; Tripoli *et al*., 2005. The non-beneficial effects obtained from the supplementation of OLE to broiler diets are in agreement with results from other trials conducted with broiler chickens fed non-antibiotic, feed additives with medicinal properties. Hernandez *et al*. (2004) found that the addition of two plant extracts (essential oil extract from oregano, cinnamon and pepper and labiatae extract from sage, thyme and rosemary) to the diet had no positive effect on the performance, proventriculus, gizzard, liver, pancreas or large or small intestine weight at 42 day of age and fecal digestibility of crude protein during the starter phase of

Table 2: Body Weight Gain (BWG), feed intake and Feed Conversion Ratio (FCR, feed (g)/gain (g)) of broiler chickens fed diets containing different levels of Olive Leaves Extract (oleuropein, OLE) during the starter and finishing phases (1-21 and 22-35 days of age, respectively)

	Age (days)								
Dietary level of OLE	1-21			22-3			1-35		
(g kg^{-1})	BWG (g)	Feed intake (g)	FCR (Feed/gain)	BWG (g)	Feed intake (g)	FCR (Feed/gain)	BWG (g)	Feed intake (g)	FCR (Feed/gain)
0	846.9	1117.3	1.33	1266.7	2037.0	1.61	2113.7	3154.3	1.49
1.8	816.4	1096.2	1.36	1212.8	1989.9	1.65	2029.2	3086.2	1.53
3.6	856.7	1134.9	1.33	1300.1	2119.6	1.63	2156.7	3254.5	1.51
6.25	832.9	1105.5	1.33	1202.0	1982.1	1.65	2034.8	3087.1	1.52
SEM[1]	24.6	30.1	0.04	31.8	46.7	0.02	59.9	44.7	0.02

[1]Standard Error of Mean

broiler chickens. Horton *et al.* (1991), Dey and Samanta (1993), Mehala and Moorthy (2008) and Rahmatnejad *et al.* (2009) reported that dietary supplementation of garlic and turmeric powders had no significant effect on weight gain and FCR. Similarly, Madrid *et al.* (2003) reported that dietary supplementation of plant extract (blend of oregano, cinnamon and pepper essential oil) did not influence the performance of broiler chickens. Hernandez *et al.* (2006) reported that performance of chickens was not influenced by dietary supplementation of organic acids. Also, Landy *et al.* (2011) reported that carcass traits and internal organ weights of broilers fed diets supplemented with neem or antibiotic were not affected by dietary treatments. In contrast, Shi *et al.* (1999) and Kumar *et al.* (2005) reported beneficial effects for dietary supplementation of garlic powder and turmeric powder on the performance of broiler chickens. Denli *et al.* (2004) reported positive effects of dietary supplementation of essential oil on body weight of broiler chickens. Also, Narimani-Rad *et al.* (2011) found that dietary supplementation of medicinal plants mixture (Oregano, Ziziphora and Peppermint) improved body weight gain and carcass quality of broiler chickens.

Results of this study and others conducted with non-antibiotic, feed additives with medicinal properties may suggest that other factors influence responses of broiler chickens to dietary supplementation of plant extracts with medicinal benefits. These factors include environment, management, nutrition, additive type and level of supplementation (Yang *et al.*, 2009). Coates *et al.* (1963) showed that dietary antibiotics had no effect on the growth performance of chickens raised in a germ free environment as compared to those raised in a conventional environment. This suggested that rearing environment of chickens is a major determining factor on the performance of chickens fed antibiotic and non-antibiotic feed additives. This suggestion is

Table 3: Apparent Nitrogen Retention (ANR) and Nitrogen corrected Metabolizable Energy (AMEn, kcal kg^{-1}) between 18 and 21 days of age of broiler chickens fed diets containing different levels of Olive Leaves Extract (oleuropein, OLE)

Dietary level of OLE (g kg^{-1})	ANR (%)	AMEn (kcal kg^{-1})
0	66.00	3470.7
1.8	66.10	3498.2
3.6	67.12	3418.7
6.25	67.00	3447.2
SEM[1]	24.60	50.7

[1]Standard Error of Mean

Table 4: Carcass composition of broiler chickens at 36 days of age fed diets containing different levels of Olive Leaves Extract (oleuropein, OLE)

	Body composition								
	Live body weight (g kg^{-1})				Eviscerated carcass (g kg^{-1})				
Dietary level of OLE (g kg^{-1})	Abdominal fat	Edible offal[1]	Neck	Evis cerated carcass	Thigh	Drums	Wings	Breast	Back
0	8.75[a]	54.6	46.75[ab]	749.4	156.0	142.5	50.5	354.0	222.8
1.8	8.26[ab]	56.0	48.18[a]	743.2	164.4	142.4	50.5	353.2	214.0
3.6	8.14[ab]	54.9	49.08[a]	747.0	154.4	141.2	48.2	368.8	210.7
6.25	7.15[b]	54.1	43.30[b]	738.6	167.1	133.1	48.6	363.5	219.3
SEM[2]	0.54	1.7	1.57	4.6	5.5	3.3	0.9	6.8	4.7

[1]Edible offal = Liver weight+heart weight+gizzard weight; [2]Standard Error of Mean; [a, b]Means within column followed by different superscripts are significantly different ($p<0.05$)

Table 5: Small intestine measurements at 21 and 36 days of age of broiler chickens fed diets containing different levels of Olive Leaves Extract (oleuropein, OLE)

	OL (g kg^{-1} diet)			Age (days)				Probability		
Characteristics	0	1.8	3.6	6.25	21	36	SEM[1]	OLE	Age	OLE×Age
Live weight (g)	1721.10	1657.40	1693.10	1659.0	837.60	2165.70**	16.50	NS	**	*
Carcass weight (g)	1240.60	1176.30	1213.50	1182.9	486.20	1613.10**	16.80	NS	**	*
Duodenum weight (W, g)	9.69	10.57	8.33	8.8800	7.6400	10.36**	0.51	NS	**	NS
Duodenum weight (live weight (%))	0.64	0.72	0.58	0.6000	0.9100	0.48**	0.03	NS	**	NS
Duodenum length (L, cm)	27.00	26.86	26.00	26.770	23.750	28.32**	0.78	NS	**	NS
Duodenum thickness (W/L, g cm^{-1})	0.36	0.39	0.32	0.3300	0.3200	0.37**	0.02	NS	**	NS
Jejunum weight (W, g)	17.01	19.04	16.79	18.430	12.370	20.93**	1.15	NS	**	NS
Jejunum weight (live weight (%))	1.18	1.24	1.08	1.1800	1.4800	0.97**	0.06	NS	**	NS
Jejunum length (L, cm)	68.27	65.54	64.09	70.180	63.000	69.03**	1.74	NS	**	NS
Jejunum thickness (W/L, g cm^{-1})	24.40	0.29	0.26	0.2600	0.2000	0.30**	0.02	NS	**	NS
Ileum weight (W, g)	16.14	15.09	13.74	15.010	8.8700	18.50**	0.85	NS	**	NS
Ileum weight (live weight (%))	0.97	0.96	0.84	0.9400	1.0600	0.85**	0.05	NS	**	NS
Ileum length (L, cm)	67.27	64.00	62.27	65.450	53.690	71.07**	1.74	NS	**	NS
Ileum thickness (W/L, g cm^{-1})	0.23	0.23	0.21	0.2200	0.1600	0.26**	0.01	NS	**	NS
Small intestine weight (W, g)	42.84	44.70	38.86	42.320	28.880	49.78**	1.92	NS	**	NS
Small intestine weight (live weight (%))	2.71	2.92	2.51	2.7200	3.4500	2.30**	0.11	NS	**	NS
Small intestine length (L, cm)	162.54	156.41	151.36	162.41	140.94	168.43**	3.32	NS	**	NS
Small intestine thickness (W/L, g cm^{-1})	0.26	0.28	0.25	0.2600	0.2000	0.30**	0.01	NS	**	NS

[1]Standard Error of Mean; *Significant difference ($p<0.05$); **Significant difference ($p<0.01$); NS = Non Significant

supported by the finding of Angel *et al.* (2005) who reported that under favorable rearing condition without any disease or stress, dietary supplementation with probiotics had no beneficial effects on broiler growth performance. Also, Timmerman *et al.* (2006) concluded that the effect of non-antibiotics feed additives is negatively related to the performance of broiler chickens. It seems that the effect will be lower on high performing birds than in low performing broilers. Results from the current study showed that dietary supplementation of OLE did not affect the performance of broiler chickens. Similarly, Hernandez *et al.* (2006) concluded that the lack of significant effects for dietary supplementation of non-antibiotic feed additives such as organic acids on the performance of chickens could be related to good rearing conditions of their study. It appears that the non-beneficial effects of OLE dietary supplementation on broiler chickens are more likely related to the good housing conditions. Results from this study may suggest that the effect of dietary supplementation of OLE is insignificant under good husbandry conditions.

CONCLUSION

It is concluded that OLE supplementation with up to 6.25 g OLE/kg diet produces no significant effect on performance, nutrient utilization, intestinal measurements and weights of eviscerated carcass as a proportion of live weight and eviscerated carcass components of thigh, drums, wings, breast and back but reduced abdominal fat of broiler chickens.

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REFERENCES

AOAC, 1990. Official Method of Analysis. 15th Edn., Association of Official Analytical Chemists, Washington, DC., USA., pp: 66-88.

Abd El-Latif, S.A., F.A. Ahmed and A.M. El-Kaiaty, 2002. Effect of feeding dietary thyme, black cumin, dianthus and fennel on productive and some metabolic responses of growing Japanese quail. Egypt. Poult. Sci., 22: 109-125.

Al-Harthi, M.A., 2002. Performance and carcass characteristics of broiler chicks as affected by different dietary types and levels of herbs and spices as non classical growth promoters. Egypt. Poult. Sci., 22: 325-343.

Al-Kassie, G.A.M., A.M. Mohseen and R.A. Abd-Al-Jaleel, 2011. Modification of productive performance and physiological aspects of broilers on the addition of a mixture of cumin and turmeric to the diet. Res. Opin. Anim. Vet. Sci., 1: 31-34.

Ali, B.H., G. Blunden, M.O. Tanira and A. Nemmar, 2008. Some phytochemical, pharmacological and toxicological properties of ginger (Zingiber officinale Roscoe): A review of recent research. Food Chem. Toxicol., 46: 409-420.

Angel, R., R.A. Dalloul and J. Doerr, 2005. Performance of broiler chickens fed diets supplemented with a direct-fed microbial. Poult. Sci., 84: 1222-1231.

Aziz, N.H., S.E. Farag, L.A. Mousa and M.A. Abo-Zaid, 1998. Comparative antibacterial and antifungal effects of some phenolic compounds. Microbios, 93: 43-54.

Bampidis, V.A., V. Christodoulou, P. Florou-Paneri, E. Christaki, P.S. Chatzopoulou, T. Tsiligianni and A.B. Spais, 2005. Effect of dietary dried oregano leaves on growth performance, carcase characteristics and serum cholesterol of female early maturing Turkeys. Br. Poult. Sci., 46: 595-601.

Bisignano, G., A. Tomaino, R.L. Cascio, G. Crisafi, N. Uccella and A. Saija, 1999. On the *in-vitro* antimicrobial activity of oleuropein and hydroxytyrosol. J. Pharm. Phrmacol., 51: 971-974.

Bonsu, F.R.K., J.K. Kagya-Agyemang, W.K.J. Kwenin and H.K. Zanu, 2012. Medicinal response of broiler chickens to diets containing neem (Azadirachta indica) leaf meal, haematology and meat sensory analysis. World Applied Sci. J., 19: 800-805.

Boudreau, M.D. and F.A. Beland, 2006. An evaluation of the biological and toxicological properties of Aloe barbadensis (miller), Aloe vera. J. Environ. Sci. Health Part C: Environ. Carcinog. Ecotoxicol. Rev., 24: 103-154.

Bozkurt, M., A. Alcicek, M. Cabuk, K. Kucukyilmaz and A.U. Catli, 2009. Effect of an herbal essential oil mixture on growth, laying traits and egg hatching characteristics of broiler breeders. Poult. Sci., 88: 2368-2374.

Carluccio, M.A., L. Siculella, M.A. Ancora, M. Massaro and E. Scoditti *et al.*, 2003. Olive oil and red wine antioxidant polyphenols inhibit endothelial activation: Antiatherogenic properties of Mediterranean diet phytochemicals. Arterioscler. Thromb. Vasc. Biol., 23: 622-629.

Choi, I.H., W.Y. Park and Y.J. Kim, 2010. Effects of dietary garlic powder and α-tocopherol supplementation on performance, serum cholesterol levels and meat quality of chicken. Poult. Sci., 89: 1724-1731.

Coates, M.E., R. Fuller, G.F. Harrison, M. Lev and S.F. Suffolk, 1963. A Comparison of the growth of chicks in the Gustafsson germ-free apparatus and in a conventional environment, with and without dietary supplements of penicillin. Br. J. Nutr., 17: 141-150.

Denli, M., F. Okan and A.N. Uluocak, 2004. Effect of dietary supplementation of herb essential oils on the growth performance, carcass and intestinal characteristics of quail (Coturnix coturnix japonica). S. Afr. J. Anim. Sci., 34: 174-179.

Dey, A. and A.R. Samanta, 1993. Effect of feeding garlic (Allium sativum L.) as a growth promoter in broilers. Indian J. Anim. Health, 32: 17-19.

Dickens, J.A., M.E. Berrang and N.A. Cox, 2000. Efficacy of an herbal extract on the microbiological quality of broiler carcasses during a simulated chill. Poult. Sci., 79: 1200-1203.

Dourado, L.R.B., J.C. Siqueira, N.K. Sakomura, S.R.F. Pinheiro, S.M. Marcato, J.B.K. Fernandes and J.H.V. Silva, 2010. Poultry feed metabolizable energy determination using total or partial excreta collection methods. Rev. Bras. Cienc. Avic., 12: 129-132.

Eevuri, T.R. and R. Putturu, 2013. Use of certain herbal preparations in broiler feeds-A review. Vet. World, 6: 172-179.

Emadi, M. and H. Kermanshahi, 2006. Effect of turmeric rhizome powder on performance and carcass characteristics of broiler chickens. Int. J. Poult. Sci., 5: 1069-1072.

Fredrickson, W.R., 2000. Method and composition for antiviral therapy with olive leaves. US Patent No. 6117844.

Gupta, G. and S. Charan, 2007. Exploring the potentials of Ocimum sanctum (Shyama tulsi) as a feed supplement for its growth promoter activity in broiler chickens. Indian J. Poult. Sci., 42: 140-143.

Hernandez, F., J. Madrid, V. Garcia, J. Orengo and M.D. Megias, 2004. Influence of two plant extracts on broilers performance, digestibility and digestive organ size. Poult. Sci., 83: 169-174.

Hernandez, F., V. Garcia, J. Madrid, J. Orengo, P. Catala and M.D. Megias, 2006. Effect of formic acid on performance, digestibility, intestinal histomorphology and plasma metabolite levels of broiler chickens. Br. Poult. Sci., 47: 50-56.

Horton, G.M.J., M.J. Fennell and B.M. Prasad, 1991. Effect of dietary garlic (Allium sativum) on performance, carcass composition and blood chemistry changes in broiler chickens. Can. J. Anim. Sci., 71: 939-942.

Huang, Y.F., H.L. Ma, D.F. Wu, J.I. Zhou, K.S. Zhou and Z.Y. Qi, 1992. Effect of Chinese medicinal herbs additives on the growth of broilers. J. Fujian Agric. College, 21: 93-96.

Kumar, M., R.S. Choudhary and J.K. Vaishnav, 2005. Effect of prebiotic, probiotics and turmeric in diet on the performance of broiler chicks during summer. Indian J. Poult. Sci., 40: 137-141.

Landy, N., G.H. Ghalamkari and M. Toghyani, 2011. Performance, carcass characteristics and immunity in broiler chicks fed dietary Neem (Melia azadirachta) as alternative for an antibiotic growth promoter. Livestock Sci., 142: 305-309.

Madrid, J., F. Hernandez, V. Garcia, J. Orengo, D. Magias and V. Savilla, 2003. 14th European symposium on poultry nutrition. August 10-14, Norway.

Markin, D., L. Duek and I. Berdicevsky, 2003. *In vitro* antimicrobial activity of olive leaves. Mycoses, 46: 132-136.

Mehala, C. and M. Moorthy, 2008. Production performance of broilers fed with Aloe vera and Curcuma longa (Turmeric). Int. J. Poult. Sci., 7: 852-856.

Narimani-Rad, M., A. Nobakht, H.A. Shahryar, J. Kamani and A. Lotfi, 2011. Influence of dietary supplemented medicinal plants mixture (ziziphora, oregano and peppermint) on performance and carcass characterization of broiler chickens. J. Med. Plants Res., 5: 5626-5629.

Okitoi, L.O., H.O. Ondwasy, D.N. Siamba and D. Nkurumah, 2007. Traditional herbal preparations for indigenous poultry health management in Western Kenya. Livestock Res. Rural Dev. Vol. 19.

Owen, R.W., A. Giacosa, W.E. Hull, R. Haubner, G. Wurtele, B. Spiegelhalder and H. Bartsch, 2000. Olive-oil consumption and health: The possible role of antioxidants. Lancet Oncol., 1: 107-112.

Panizzi, L., M.L. Scarpati and G. Orient, 1960. The constitution of oleuropein, a bitter glucoside of the olive with hypotensive action. Gazzetta Chimica Italiana, 90: 1449-1485.

Privitera, J.D., 1996. Olive Leaf Extract: A New/Old Healing Bonanza for Mankind. NutriScreen Inc., Covina, CA, USA.

Rahmatnejad, E., H. Roshanfekr, O. Ashayerizadeh, M. Mamooee and A. Ashayerizadeh, 2009. Evaluation the effect of several non-antibiotic additives on growth performance of broiler chickens. J. Anim. Vet. Adv., 8: 1757-1760.

SAS, 2005. SAS User's Guide. 9th Edn., SAS Institute Inc., Cary, NC., USA.

Schragle, R. and W. Muller, 1990. The influence of selected Tannin-containing plants species on the tenacity of pathogenic bacteria *in vtro* rumen system. 1-Communication: The influence of selected plant species on the tenacity of CI. Derferingens. Zentraib Veterinarmed B, 37: 181-186.

Scott, M.L., M.C. Nesheim and R.J. Young, 1982. Nutrition of the Chicken. 3rd Edn., ML Scott and Associates, Ithaca, New York.

Shi, X.H., S.Z. Li, Z.P. Liu, X.H. Shi, S.Z. Li and Z.P. Liu, 1999. A trial on the use of garlic as a feed additive for meat chickens. Poult. Husbandry Dis. Control, 10: 19-20.

Singh, N., J.P. Singh and V. Singh, 2007. Effect of dietary supplementation of herbal formulation on dressing percentage and mortality in broiler chicks. Indian J. Field Vet., 2: 22-24.

Timmerman, H.M., A. Veldman, E. van den Elsen, F.M. Rombouts and A.C. Beynen, 2006. Mortality and growth performance of broilers given drinking water supplemented with chicken-specific probiotics. Poult. Sci., 85: 1383-1388.

Tripoli, E., M. Giammanco, G. Tabacchi, D. Majo, D. Majo, S. Giammanco and M. La Guardia, 2005. The phenolic compounds of olive oil: Structure, biological activity and beneficial effects on human health. Nutr. Res. Rev., 18: 98-112.

Visioli, F., A. Poli and C. Gall, 2002. Antioxidant and other biological activities of phenols from olives and olive oil. Med. Res. Rev., 22: 65-75.

Visioli, F., S. Bellosta and C. Galli, 1998. Oleuropein, the bitter principle of olives, enhances nitric oxide production by mouse macrophages. Life Sci., 62: 541-546.

Vogt, H., 1991. Essential oils in broiler diets. Landbauforshung Volkenrode, 41: 94-97.

Walker, M., 1997. Natures Antibiotic: Olive Leaf Extract. Kensington Books, New York, USA., pp: 120-128.

Yang, Y., P.A. Iji and M. Choct, 2009. Dietary modulation of gut microflora in broiler chickens: A review of the role of six kinds of alternatives to in-feed antibiotics. Worlds Poult. Sci. J., 65: 97-114.

Zita, F., P. Elena, M. Zuzana, T. Jana, C. Klaudia, L. Leng and S. Faix, 2009. Effects of borneol on blood chemistry changes in chickens. Acta Veterinaria, 59: 177-184.