Available online at http://www.ijabbr.com

International journal of Advanced Biological and Biomedical Research

Volume 2, Issue 3, 2014: 741-751

Effects of olive leaf on blood metabolites and humoral immunity response of broiler chickens

Siamak Parsaei[1#], Zahra Amini[2], Mohammad Houshmand[1]

[1#] Department of Animal science, Faculty of Agriculture, Yasouj University, Yasouj, Iran.

Email: parsaei@mail.yu.ac.ir

[1] Department of Animal science, Faculty of Agriculture, Yasouj University, Yasouj, Iran.

Email: hooshmand@yu.ac.ir

[2]Department of Animal science, Faculty of Agriculture, Yasouj University, Yasouj, Iran.

Email: Zahraamini448@yahoo.com

Abstract

This experiment was conducted to investigate the effects of olive leaf (OL) on immune response and some blood metabolites of broiler chickens. A total of 400 one-day-old broiler chicks (male and female) were randomly assigned to 5 experimental treatments with 4 replicates of 20 birds each. Birds of different treatments were given diets containing 0, 0.25, 0.5, 0.75 and 1% OL powder from 14 to 42 d of age. At 21 and 42 d of the experiment, blood samples were taken from two birds of each pen (8 birds/treatment) to evaluate different blood metabolites. Moreover, in order to determine immunity status, at the end of the experiment, antibodies titer against SRBC and weight of bursa of Fabricious were determined. The results indicated that significant difference was not observed for total Ig and IgM, while IgY was higher in birds fed with 0.75% OL compared to the control group. The heaviest bursa of Fabricious observed in birds fed with 0.25% OL. Feeding with OL resulted in significant reductions in blood levels of triglyceride, cholesterol, glucose, LDL, VLDL, HDL and liver enzymes. In conclusion, the findings show that dietary supplementation with OL had positive effects on blood lipids profile, liver enzymes and immunity of broiler chickens.

Keywords: Olive leaf, Broiler, Blood metabolites, Cholesterol, Lipoprotein, Immunity

Introduction

The olive tree (*Olea europea L.*) is widely cultivated in Mediterranean countries. This tree shows strong resistance against microorganism invasion and insect attacks (Kubo and Lunde, 1995). Methanolic extracts of olive leaves contain secoiridoids such as oleuropein, ligostroside, dimethyl oleuropein and

oleoside (EI and Karakaya, 2009), flavonoids including apigenin, kaempferol and luteolin, as well as phenolic compounds such as caffeic acid, tyrosol, and hydroxytyrosol (Chiou *et al*, 2007).

Olive leaf contains high level of a complex phenolic compound, is known as oleuropein (Ryan et al, 2002). Oleuropein has many positive and health-promoting effects which is mostly related to its antioxidant property (Al-Azzawie and Alhamdani, 2006). For example, it has been reported that oleuropein reduced infarct size, plasma lipid concentrations, and plasma markers of oxidative stress in cholesterol-fed rabbits (Al-Azzawie and Alhamdani, 2006; Andreadou *et al*, 2006). Also, some in vivo studies have shown that olive leaf extract reduced blood levels of cholesterol (Fki et al, 2005) and lipid (Jemai, 2008) in cholesterol-fed rats. Moreover, it has been reported that this extract reduced blood pressure in nitro-L-arginine methyl ester induced hypertensive rats (Khayyal *et al*, 2002) and normotensive rats (Lasserre *et al*, 1983). The potential of olive leaf extract in reduction of the serum levels of glucose, lipids, uric acid, creatinine and liver enzymes in streptozotocin-induced diabetic rats has also been reported (Eidi *et al*, 2009).

Olive leaf can improve immunity status (Gonzales, 1992). It also contains an antimicrobial compound (Markin *et al*, 2003, Pereira *et al*, 2007). In folk medicine, it is used in treatment of different diseases such as hypertonia, arteriosclerosis, rheumatism, gout, diabetes mellitus and fever (Gonzales, 1992, Al-Azzawie and Alhamdani, 2006). Hypoglycemic (Gonzales, 1992, Al-Azzawie and Alhamdani, 2006), hypotensive (Khayyal *et al*, 2002), anti-arrhythmic (Somova *et al*, 2003), anti-atherosclerotic (Wang *et al*, 2008) and vasodilator (Zarzuelo *et al*, 1991) properties of olive leaf have been shown in different animal studies. Its antimicrobial (Bisignano *et al*, 1999, Markin *et al*, 2003, Pereira *et al*, 2007), antiviral (Lee-Huang *et al*, 2003, Micol, 2005) and anti-inflammatory activities (Pieroni *et al*, 1996) were also reported.

OL is cheap and available in different seasons in some parts of the world, for instance, Iran. Also, as mentioned earlier, it is rich in beneficial compounds such as phenols. On the other hand, there is no report on the effects of OL on immunity and blood metabolites of broiler chickens. Therefore, the aim of this study was to investigate the effects of OL on serum levels of cholesterol, triglyceride, LDL, HDL, VLDL, glucose, alanin aminotransferase, aspartat aminotransferase and humoral immune response of broiler chickens.

MATERIALS AND METHODS

Birds and diets

A total of four hundred one-day-old male and female broiler chicks (Cobb 500) were obtained from a local commercial hatchery. Upon arrival, birds were weighed in group and distributed between 20 floor pens and reared under same conditions until 14 d of age. On day 14, pens were randomly assigned to one of the five experimental diets (treatments). A basal diet without OL was served as the control (Ctrl) and four diets were supplemented with 0.25, 0.5, 0.75 and 1% OL, respectively. Each treatment had four replicates of 20 chicks each. There was no significant difference among treatments for body weight at 14 d of age. Isocaloric and isonitrogenus diets were formulated according to NRC (1994) recommendations by using UFFDA software. The starter and finisher diets in mash form were given from 1-21 and 22-42 of age, respectively. The composition of the experimental diets are shown in Table 1. Birds had free access to feed and water throughout the experiment. Rice straw was used as litter. Fresh leaves were collected in October 2011 from olive trees of the south region of Iran and dried in shadow and then fine grinded and included in the diets from 14 to 42 d of age.

Table 1: Composition of experimental diets

	Starter					Grower& Finisher				
Ground olive leaf (g/kg)										
	0	0.25	0.5	0.75	1	0	0.25	0.5	0.75	1
Corn	550	549	547.5	546.2	54.5	625	623.7	622.5	621.2	620
Soyben meal	361	360	358.5	357.2	35.6	302	300.8	299.5	298.3	297
Soybean oil	45	45	45	45	45	30	30	30	30	30
Limestone	14	14	14	14	14	13	13	13	13	13
Dicalcium phosphate	18.4	18.4	18.4	18.4	18.4	15	15	15	15	15
Common salt	3.6	3.6	3.6	3.6	3.6	3	3	3	3	3
Vit. & Min. Premix*	5	5	5	5	5	10	10	10	10	10
DL-Met	3	3	3	3	3	2	2	2	2	2
Olive leaf	0	2.5	5	7.5	10	0	2.5	5	7.5	10
Calculated analysis										
ME (Kcal/kg)	3126	3118	3111	3104	3097	3148	3141	3133	3126	3119
Crude protein	223.8	223	222.4	221.7	22.11	187.6	187	186.3	185.7	184.9
Lysine	11.5	11.5	11.5	11.5	11.5	9.9	9.9	9.9	9.9	9.9
Methionine	5.9	5.9	5.9	5.9	5.9	5.4	5.4	5.4	5.4	5.4
Try	8.3	8.3	8.3	8.3	8.3	7.6	7.6	7.6	7.6	7.6
Arg	17.2	17.2	17.2	17.2	17.2	12.1	12.1	12.1	12.1	12.1
Ca	10.6	10.6	10.6	10.6	10.6	9.3	9.3	9.3	9.3	9.3
Ap	5.5	5.5	5.5	5.5	5.5	4.7	4.7	4.7	4.7	4.7
Na	1.7	1.7	1.7	1.7	1.7	1.5	1.5	1.5	1.5	1.5

*****Vitamin mix provided the following per kilogram of complete diet: vitamin A, 9,000 IU; vitamin D3, 2,000 IU; vitamin E, 18 IU; vitamin K3, 2 mg; thiamine, 2 mg, riboflavin, 6.6 mg, niacin 30mg, D-pantothenic acid, 10 mg, pyridoxine, 3 mg Folic acid, 15 mg cobalamin, 0.01 mg; biotin, 100 mg; antioxidant, 500 mg. Mineral mix provided the following per kilogram of complete diet: Colin, 400mg, Fe, 50 mg; Mn, 100 mg, Zn, 85 mg, Cu, 10mg; Se, 0.2 mg and I, 100 mg.

Measurements and sampling

On d 21 (end of the starter period) and 42 (end of the finisher period), blood samples were taken via jugular vein from two birds of each replicate (8 birds/treatment). Collected samples were centrifuged (2500×g) for 15 min and their serum were separated to measure levels of different metabolites. Serum concentrations of total cholesterol (TC), total triglycerides (TG), low-density lipoprotein (LDL-C) and high density lipoprotein (HDL-C) were determined by commercial kits (Kyokuto Pharmaceutical, Japan) using the colorimetric methods. The standard enzymatic methods using commercial kits were used to determine serum glucose and liver enzymes (alanin aminotransferase and aspartat aminotransferase). Antibody response against sheep red blood cells (SRBC) was measured to evaluate bird immunity status. For this purpose, two birds from each replicate were injected intravenously (brachial vein) with SRBC (3 % suspension in PBS, 1 ml per bird) at 28 and 35 d of age. On d 42, blood samples were taken from injected birds and serum was taken. Serum antibodies levels were determined using heamagglutination technique (Cheema *et al*, 2003). Moreover, at the same time, bursa of Fabricius was carefully removed and weighted. Relative bursa weight was calculated as bursa weight / live body weight × 100.

Statistical Analysis:

Data of the experiment were analyzed by analysis of variance using General Linear Models (GLM) procedures of SAS. Duncan's Multiple Range Test was used to compare the means. The level of significance was reported at $P<0.05$.

RESULTS AND DISCUSSION

Blood lipid profile at 21 d of age

Effects of experimental treatments on blood lipid profile at 21 d of age are shown in Table 2. Blood cholesterol and HDL were significantly lower in 0.75% group compared to the other treatments. Also, birds fed diets supplemented with 0.75 and 1% OL, had lower levels of triglyceride and VLDL compared to those fed the control diet. Dietary treatments had no significant effects on LDL.

Table 2: Effects of experimental treatments on blood cholesterol, triglyceride, LDL, HDL and VLDL (mg/dl) at 21d of age

Treatment	cholesterol	triglyceride	LDL	VLDL	HDL
Ctrl	114.9^{a}	116.0^{a}	29.4	23.2^{a}	87.6^{ab}
0.25	112.7^{a}	99.0^{ab}	27.8	19.9^{ab}	86.6^{ab}
0.5	117.5^{a}	111.5^{a}	25.0	22.3^{a}	89.9^{a}
0.75	89.6^{b}	77.5^{c}	25.2	15.7^{b}	66.4^{c}
1	108.6^{a}	83.1^{bc}	27.7	16.6^{c}	78.2^{b}
SEM	4.46	7.06	1.50	1.62	3.41

Means within a column with different superscript letters are significantly different at P<0.05.
Ctrl: a basal diet without OL, 0.25: diet containing 0.25% OL, 0.5: diet containing 0.5% OL, 0.75: diet containing 0.75% OL, 1: diet containing 1% OL.

Blood lipid profile at 42 d of age

Effects of experimental treatments on blood lipid profile at 42 d of age are shown in Table 3. The results indicated that dietary inclusion of 0.5 and 0.75% OL resulted in a significant decrease in blood cholesterol compared to the control group. Moreover, supplementation with OL, regardless of OL level, significantly decreased blood HDL and LDL. Birds fed diets containing 0.5% OL, had less VLDL relative to the control group. Briefly, The current findings suggest that dietary addition of OL could reduce the blood levels of cholesterol, triglyceride and LDL.

Table 3: Effects of experimental treatments on blood cholesterol, triglyceride, LDL, HDL and VLDL (mg/dl) at 42 d of age

Treatment	TC	TG	LDL	VLDL	HDL
Ctrl	133.7^{a}	115.5^{a}	43.3^{a}	23.1^{a}	73.9^{a}
0.25	132.7^{a}	110.6^{ab}	31.3^{c}	22.1^{a}	66.5^{b}
0.5	104.5^{b}	90.5^{c}	30.2^{c}	16.9^{b}	61.8^{c}
0.75	106.9^{b}	95.2^{bc}	33.9^{c}	19.0^{ab}	60.6^{c}
1	152.2^{a}	97.5^{abc}	39.1^{b}	19.0^{ab}	57.7^{c}
SEM	8.32	6.08	1.38	1.44	3.42

Means within a column with different superscript letters are significantly different at P<0.05.
Ctrl: a basal diet without OL, 0.25: diet containing 0.25% OL, 0.5: diet containing 0.5% OL, 0.75: diet containing 0.75% OL, 1: diet containing 1% OL.

Previously, Gorinstein *et al* (2002) reported that olive oil polyphenols reduced plasma LDL-C levels. Hypocholesterolemic property of olive oil may be related to the inhibitory effect on cholesterol. Olive oil can reduce the intestinal absorption of cholesterol, or decrease its synthesis by liver (Krzeminski, 2003). Also, olive oil stimulates the biliary secretion of cholesterol and its

excretion in the faeces (Prasad and Kalra, 1993). Due to these effects, it has hypocholesterolemic property. In vitro studies have shown that flavonoids are able to decrease the availability of lipid substrates for hepatic synthesis of VLDL (sung *et al*, 2004). Isoflavones inhibit the catalytic domain of 3-hydroxy-3-methyl glutaryl (HMG) CoA reductase for cholesterol synthesis (sung *et al*, 2004). Srinivasan *et al* (2007) reported that lipid peroxidation decreased in the olive leaf extract-treated animals. Oleuropein, oleuropein aglycone and hydroxytyrosol have antioxidant properties, because they are able to decompose free radicals by quenching reactive oxygen species and by trapping radicals before reaching their cellular targets (Srinivasan *et al*, 2007).

Blood glucose

Effects of experimental treatments on blood glucose at 21 and 42 d of age are presented in Table 4. On day 21, birds fed diets containing 0.75 and 1% OL, had less blood glucose compared to the other treatments. Indeed, at the end of the experiment (42 d of age), birds fed diets supplemented with 0.25 and 0.75% OL had less glucose level than those fed the control diet.

Table 4: Effects of experimental treatments on blood glucose (mg/dl) at 21 and 42 d of age

Treatment	21d	42 d
Ctrl	262.5^{a}	235.6^{a}
0.25	253.8^{a}	206.3^{b}
0.5	265.8^{a}	227.1^{a}
0.75	173.3^{b}	216^{b}
1	191.2^{b}	221.1^{ab}
SEM	6.8	4.2

Means within a column with different superscript letters are significantly different at $P<0.05$.
Ctrl: a basal diet without OL, 0.25: diet containing 0.25% OL, 0.5: diet containing 0.5% OL, 0.75: diet containing 0.75% OL, 1: diet containing 1% OL.

Hypoglycemic property of olive leaf in diabetic animals has previously been reported by Gonzalea *et al* (1992). They concluded that this effect could be related to a compound, known as oleuropein. Two mechanisms were suggested by these researchers for anti diabetic property of olive leaf: a) potentiation of glucose induced insulin release and b) increased peripheral uptake of glucose. The results of Al-Azzawie and Alhamdani, (2006) indicated that oleuropein had beneficial effects on alloxan-diabetic rabbits. They reported that during 16 weeks of treatment of diabetic animals with oleuropein (20 mg/kg body weight), hypoglycemia and oxidative stress induced by diabetes were significantly reduced.

Liver enzymes

Effects of experimental treatments on liver enzymes at 21 and 42 d of age are presented in Table 5. At 21 d of age, 0.75 and 1 % groups had the lowest levels of SGOT and SGPT, respectively, which indicate significant decreases compared to the Ctrl group. At the end of the experiment (42d of age), all supplemented groups (except for 1%) had less levels of liver enzymes compared to the Ctrl group.

To the best of our knowledge, there is no previous information in the literature on the effects of olive leaf (or other parts of olive tree) on animals liver enzymes. As discussed earlier, high levels of oleuropein are present in olive products. Many of pharmacological properties of olive products are related to their antioxidant property (Visioli et al., 2002). Recently, there is increasing interest to use natural antioxidants such as polyphenols, due to their known abilities to

scavenge free radicals (Pyo et al., 2004). Some beneficial effects of these compounds are as follows: Inhibition of oxidative stress, prevention of amino transferase enzymes departure and treatment of liver cells and liver toxicity (Tiot et al., 2001). Thus it is expected that OL has beneficial effects on liver.

Table 5: Effects of experimental treatments on the levels of liver AST and ALT at 21 and 42 d of age

	SGOT(AST)		SGPT(ALT)	
Treatment	21d	42d	21d	42d
Ctrl	192.8^{b}	140.1^{a}	8.1^{a}	33.9^{a}
0.25	258.4^{a}	138.5^{b}	7.8^{a}	20.1^{b}
0.5	193.6^{b}	132.9^{c}	6.5^{ab}	11.4^{c}
0.75	146^{c}	136.5^{b}	6.6^{ab}	16.8^{bc}
1	177.6^{bc}	141^{a}	5^{b}	30.3^{a}
SEM	12.7	22	0.62	5.3

Means within a column with different superscript letters are significantly different at P<0.05.
Ctrl: a basal diet without OL, 0.25: diet containing 0.25% OL, 0.5: diet containing 0.5% OL, 0.75: diet containing 0.75% OL, 1: diet containing 1% OL.

Antibody response to SRBC and bursa of Fabricious Weight

Effects of experimental treatments on antibody response to SRBC and bursa of Fabricious weight are shown in Table 6. Total Ig and IgM were significantly not influenced by treatments, while significant differences were observed among treatments for IgY. Supplementation with 0.75% OL resulted in a significant increase in IgY compared to the control group. Birds fed diet containing 0.25% OL had heavier bursa of Fabricious than the other groups. Herbs may influence immune system via four mechanisms: activation of phagocytotic property, stimulation of fibroblasts, increscent of respiratory activity and leukocyte movement (Bauer *et al*, 1989). Olive leaf contains flavonoids. These compounds are able to increase IgY and improve immunity status (Christake et al, 2004). Also, anti-inflammatory, anti-allergic and anti-viral properties of flavonoids have been reported (Cushnie, 2005). Some plant extracts can increase antibody production and therefore immunity. They are able to influence gastrointestinal mucosal lymphocyte and also stimulate local immunity (Shams-Ghafarokhi *et al*, 2003). In vitro studies have shown that olive leaf compounds have strong inhibitory effects against viruses (parainfluenza, herpes, pseudorabies, and some forms of polio) (Renis, 1969). Olive leaf prevent viral infection by different mechanisms such as inhibition of assembly at the cell membrane, interfering with critical amino acids production and stopping viral shedding (Renis, 1969; Micol *et al*, 2005). Inhibitory effects of olive leaf on many gram negative and positive bacteria, yeast and parasites have been reported *(*Elliott *et al*, 1969; Markin *et al*, 2003).

Table 6: Effects of experimental treatments on the relative weight of bursa of Fabrecious (g/ 100 g body weight) and antibody response against SRBC (Log 2)

Treatment	IgTotal	IgY	IgM	bursa of Fabrecious
Ctrl	6.75	4.25^{b}	2.50	0.04^{b}
0.25	7.50	4.25^{b}	3.25	0.06^{a}
0.5	7.25	5.25^{ab}	2.00	0.04^{bc}
0.75	8.00	5.5^{a}	2.50	0.04^{bc}
1	8.25	4.75^{ab}	3.50	0.03^{c}
SEM	0.72	0.36	0.75	0.003

Means within a column with different superscript letters are significantly different at P<0.05.
Ctrl: a basal diet without OL, 0.25: diet containing 0.25% OL, 0.5: diet containing 0.5% OL, 0.75: diet containing 0.75% OL, 1: diet containing 1% O.

In conclusion, the findings of the current study showed that dietary inclusion of olive leaf had positive effects on blood lipid profiles, liver enzymes and immunity of broiler chickens, but more research and investigations are needed in this field to use it as a beneficial organic feed additive in broiler diets.

Acknowledgements

Thanks to financial support provided by Yasouj University.

References

Al-Azzawie HF, Alhamdani MS (2006). Hypoglycemic and antioxidant effect of oleuropein in alloxan-diabetic rabbits. Life Science. 78:1371–7.

Al-Qarawi A.A, Al-damegh M.A, El-Mougy S.A (2002). Effect of freeze dried extract of *Olea europaea* on pituitary-thyroid axis in rats. Phytother Research. 16: 286-287.

Andreadou I, Iliodromitis EK, Mikros E, Constantinou M, Agalias A, Magiatis P, Skaltsounis AL, Kamber E, Tsantili-Kakoulidou A, et al (2006). The olive constituent oleuropein exhibits anti-ischemic, antioxidative, and hypolipidemic effects in anesthetized rabbits. Journal of Nutrition. 136:2213–9.

Andrikopoulos N.K, Kaliora A.C, Assimopoulou A.N, Papageorgiou V.P (2002). Inhibitory activity of minor polyphenolic and nonpolyphenolic constituents of olive oil against in vitro low-density lipoprotein oxidation. Journal of Medicinal Food. 5, 1 –7.

Barton M.d (2000). Antibiotic use in animal feed and its impact on human health. Nutrition Research Reviews. 13:279-299.

Bauer R, Reminger P (1989). TLC and HPLC analysis of alkamides in Echinacea drugs. Journal of Medicinal Plant. 55: 367-371.

Bisignano G, Tomaino A, LoCascio R, Crisafi G, Uccella N, Saija A (1999). On the in vitro antimicrobial activity of oleuropein and hydroxytyrosol. Pharm. Pharmacol. 51, 971–974.

Cheema M.A, Qureshi M.A, Havenstein G.A (2003). A comparison of the immune response of a 2001 commercial broiler with a 1957 random bred broiler strain when fed representative 1957 and 2001 broiler diets. Poultry science association. 9, 1591/1529.

Chimi H, Cillard J, Cillard P, Rahmani M (1991). Peroxyl and hydroxyl radical scavenging activity of some natural phenolic antioxidants. Journal of the American Oil Chemists' Society. 68, 307–312.

Chiou A, Salta FN, Kalogeropoulos N, Mylona A, Ntalla I, Andrikopoulos NK (2007). Retention and distribution of polyphenols after pan-frying of French fries in oils enriched with olive leaf extract. Food Science. 72:S574-84.

Christake E. P, Paneri I, Giannenas M, Papazahariadou N, Botsoglou A, Spais A.B (2004). Effect of mixture of herbal extract on broiler chicken infected with eimeria tenella. Animal Research. 53: 137-144

Corona G, Tzounis X, Assunta Dessi M, Deiana M, Debnam ES, Visioli F, Spencer JP (2006). The fate of olive oil polyphenols in the gastrointestinal tract: implications of gastric and colonic microflora-dependent biotransformation. Free Radic Research. 40:647–58.

Cushnie T.P.T, Lamb A.J (2005). Antimicrobial activity of flavonoids: review. Int. J. Antimicrob Agents. 26: 343-356.

dela Puerta R, Ruiz Gutierrez V, Hoult J.R (1999). Inhibition of leukocyte 5-lipoxygenase by phenolics from virgin olive oil. Biochemical Pharmacology. 57, 445– 449

Edgecombe S.C, Stretch G.L, Hayball P.J (2000). Oleuropein, an antioxidant polyphenol from olive oil, is poorly absorbed from isolated perfused rat intestine. Journal of Nutrition. 130, 2996–3002.

EI SN, Karakaya S (2009). Olive tree (Olea europaea) leaves: potential beneficial effects on human health. Nutrition Review. 67:632–8.

Eidi A, Eidi M, Darzi R (2009). Antidiabetic effect of Olea europaea L. in normal and diabetic rats. Phytother Research. 23:347–50.

Elliott GA, Buthala DA, DeYoung EN (1969). Preliminary 18. safety studies with calcium elenolate, an antiviral agent. Antimicrob Agents Chemother (Bethesda). 9:173-176.

Fki I, Bouaziz M, Sahnoun Z, Sayadi S (2005). Hypocholesterolemic effects of phenolic-rich extracts of Chemlali olive cultivar in rats fed a cholesterolrich diet. Bioorganic & Medicinal Chemistry. 13:5362–70.

Gonzalez M, Zarzuelo A, Gamez M. J, Utrilla M. P, Jimenez J, Osuna I (1992). Hypoglycemic activity of olive leaf. . Journal of Medicinal Plant and Natural Product Research. *58*, 513-515.

Gordon M.H, Paiva-Martins F, Almeida M (2001). Antioxidant activity of hydroxytyrosol acetate compared with that of other olive oil polyphenols. Journal of Agricultural and Food Chemistry. 49, 2480–2485.

Gorinstein S, Leontowicz H, Lojek A, Leontowicz M, Ciz M, Krzeminski R, Gralak M, Czerwinski J, Jastrzebski Z, Trakhtenberg S, Grigelmo-Miguel N, Soliva-Fortuny R (2002). Olive oils improve lipid metabolism and increase antioxidant potential in rats fed diets containing cholesterol, Journal of Agricultural and Food Chemistry. 50 6102–6108.

Inouye M, Mio T, Sumino K (1999). Link between glycation and lipooxidation in red blood cells in diabetes. Clinica Chimica Acta. 285, 35– 44.

Jemai H, Bouaziz M, Fki I, El Feki A, Sayadi S (2008). Hypolipidimic and antioxidant activities of oleuropein and its hydrolysis derivative-rich extracts from Chemlali olive leaves. Chemico-Biological Interactions. 176:88–98.

Jennings P.E, McLaren M, Scot N.A, Saniabadi A.R, Belch J.J.F (1991). The relationship of oxidative stress to thrombotic tendency in type I diabetic patients with retinopathy. Diabetic Medicine. 8, 860– 865.

Khayyal MT, el-Ghazaly MA, Abdallah DM, Nassar NN, Okpanyi SN, Kreuter MH (2002). Blood pressure lowering effect of an olive leaf extract (Olea europaea) in L-NAME induced hypertension in rats. Arzneimittelforschung. 52:797–802.

Komaki E, Yamaguchi S, Maru I, et al (2003). Identification 27. of anti-alpha-amylase components from olive leaf extracts. Food Science and Technology Research. 9:35-39.

Krzeminski R, Gorinstein S, Leontowicz H, Leontowicz M, Gralak M, Czerwinski J, Lojek A, Ciz M, Martin-Belloso O, Gligelmo-Miguel N, Trakhtenberg S (2003). Effect of different olive oils on bile excretion in rats fed cholesterol-containing and cholesterol-free diets, J. Agric. Food Chemistry. 51 5774–5779.

Kubo A, Lunde C.S, Kubo I, (1995). Antimicrobial activity of the olive oil flavor compounds. Journal of Agriculture and Food Chemistry. 43, 1629–1633.

Lasserre B, Kaiser R, Huu Chanh P, Ifansyah N, Gleye J, Moulis C (1983). Effects on rats of aqueous extracts of plants used in folk medicine as antihypertensive agents. Naturwissenschaften. 70:95–6.

Lee-Huang S, Zhang L, Huang PL, et al (2003). Anti-HIV 16 activity of olive leaf extract (OLE) and modulation of host cell gene expression by HIV-1 infection and OLE treatment. Biochem Biophys Reserch Commun. 307:1029-1037.

Madar Z, Maayan N, Sarit O, Eliraz A (2004). Antioxidants modulate thenitric oxide system and SOD activity and expression in rat epithelial lungcells. Asia Pacific Journal of Clinical Nutrition. 13, S101.

Mak D.H, Ip S.P, Poon M.K, Ko K.M (1996). Alterations in tissue glutathione antioxidant system in streptozotocin-induced diabetic rats. Molecular and Cellular Biochemistry. 162, 153–158

Manna C, Migliardi V, Golino P, Scognamiglio A, Galletti P, Chiariello M, Zappia V (2004). Oleuropein prevents oxidative myocardial injury induced by ischemia and reperfusion. J Nutrition Biochemistry. 15:461–6.

Markin D, Duek L, Berdicevsky I (2003). 19. In vitro antimicrobial activity of olive leaves. Mycoses. 46:132-136.

Martinez-Gonzalez MA, Bes-Rastrollo M, Serra-Majem L, Lairon D, Estruch R (2009). Trichopoulou A. Mediterranean food pattern and the primary prevention of chronic disease: recent developments. Nutrition Review. 67 Suppl 1:S1116.

Masella R, Vari R, D'Archivio M, Di Benedetto R, Matarrese P, Malorni W, Scazzocchio B, Giovannini C (2004). Extra virgin olive oil biophenols inhibit cell-mediated oxidation of LDL by increasing the mRNA transcription of glutathione-related enzymes. Journal of Nutrition. 134, 785– 791.

Micol V, Caturla N, Pérez-Fons L, et al (2005). The olive 17. leaf extract exhibits antiviral activity against viral haemorrhagic septicaemia rhabdovirus (VHSV). Antiviral Reserch. 66:129-136.

Pereira A.P, Ferreira I.C.F.R, Marcelino F, Valentao P, Andrade P.B, Seabra R, Estevinho L, Bento A, Pereira J.A (2007). Phenolic Compound and Antimicrobial Activity of Olive (*Olea europaea L.Cv.Cobrancosa*) leaves. Journal of Molecules. 12, 1153-1162.

Petkov V, Manolov P (1978). Pharmacological studies on substances of plant origin with coronary dilatating and antiarrhythmatic action. Comparative Medicine East and West. 6, 123– 130.

Pieroni A, Heimler D, Pieters L, et al (1996). 24. In vitro anti-complementary activity of flavonoids from olive (Olea europaea L.) leaves. Pharmazie. 51:765-768.

Prasad K, Kalra J (1993). Oxygen free radicals and hypercholesterolemic atherosclerosis, American Heart Journal. 125 958–971.
Pyo Y.H, Lee T.C, Logendra L, Rosen R.T (2004). Antioxidant activity and phenolic compounds of Swiss chard extracts. Food chemistry 85: 19-26.

Renis HE (1969). 15. *In vitro* antiviral activity of calcium elenolate. Antimicrob Agents Chemother (Bethesda). 9;167-172.

Ryan D, Antolovich M, Prenzler P, Robards K, Lavee S (2002). Biotransformations of phenolic compounds in Olea europea L. Scientia Horticulturae. 92, 147–176.

Shams-Ghafarokhi M, Razafsha M, Allumeh A, Razzaghi Abyaneh M (2003). Inhibitory Effect of Aqueous Onion and Garlic Extracts on Growth and Keratinase Activity in Trichophyon Mentagro Phytes. J of Iran Biomed. 7: 113-118.

Soler-Rivas C, Espin JC, Wichers HJ (2000). Oleuropein and related compounds. Journal of Science Food Agricultur. 80:1013–23.

Somova L.I, Shode F.O, Ramnanan P, Nadar A (2003). Antihypertensive ,antiatherosclerotic and antioxidant activity of triterpenoids isolated from olea europaea, subspecies Africana leaves. Journal of Ethnopharmacology, 84:299-305.

Srinivasan P, Sabitha K.E, Shyamaladevi C.S (2007). Attenuation of 4-nitroquinoline 1-oxide induced in vitro lipid peroxidation by green tea polyphenols. Life Science, 80:1080–1086.

Sung JH, Choi SJ, Lee SW, Park KH, Moon TW (2004). Isoflavones found in Korean soybean paste as 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors. Biosci Biotechnol Biochem, 68:1051-1058.

Tiot R, Volsteedt Y, Apostolides Z (2001). Comparison of the antioxidant content of fruits, vegetables and teas measured as vitamin C equivalent. Toxicology, 166: 63-69.

Visioli F, Bellomo G, Montedoro G.F, Galli C (1995). Low-density lipoprotein oxidation is inhibited in vitro by olive oil constituents. Atherosclerosis, 117, 25– 32.

Visioli F, Galli C (2001). Antiatherogenic components of olive oil. Current Atherosclerosis Reports 3, 64– 67.

Visioli F, Poli A, Galli C (2002). Antioxidant and other biological activities of phenols from olives and olive oil. Medicinal Research Reviews, 22, 65– 75.

Walker C, Reamy BV (2009). Diets for cardiovascular disease prevention: what is the evidence?. American Family Physician, 79:571–8.

Wang L, Geng C, Jiang L, et al (2008). The anti-31. atherosclerotic effect of olive leaf extract is related to suppressed inflammatory response in rabbits with experimental atherosclerosis. European Journal of Nutrition, 47:235-243.

Wolffe S.P, Jiang Z.Y, Hunt J.V (1991). Protein glycation and oxidativestress in diabetes mellitus and ageing. Free Radical Biology & Medicine, 10, 339– 352.

Young C.E, Karas R.H, Kuvin J.T (2004). High-density lipoprotein cholesterol and coronary heart disease. Cardiology Reviews, 12 (2) 107–119.

Zarzuelo A, Duarte J, Jimenez J, Gonzales M (1991). Utrilla Vasodilator effect of olive leaf. Planta Medica, *57*, 417-424.

Available online at http://www.ijabbr.com

International journal of Advanced Biological and Biomedical Research

Volume 2, Issue 3, 2014: 741-751

Corresponding Author E-mail: parsaei@mail.yu.ac.ir