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Artichoke (*Cynara scolymus* L.) Leaves and Heads Extracts as Hypoglycemic and Hypocholesterolemic in Rats

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Abstract Artichoke (*Cynara scolymus* L., *Asteraceae* family), an edible vegetable from the Mediterranean area, is a good source of phenolic compounds. Two varieties of artichoke were used in this study Green Globe (G) and Violet (V). Five major active phenolic compounds were identified into the aqueous methanolic extracts of artichoke leaves and heads. These compounds were identified as Chlorogenic acid, Cynarin, 1, 5-di-o-Caffeoylquinic, luteolin and apigenin. On the other hand, the artichoke aqueous leaves extract (ALE) and aqueous heads extracts (AHE) for the two varieties were used as hypoglycemic and hypocholesterolemic experiments by using albino rats. ALE was used in the concentration of (1.5 g/kg/day) for the two varieties. AHE was used in two different concentrations (1.5 and 3 g/kg/day). Rats were administrated orally by these different concentrations. Results show the effect of ALE and AHE extracts on the glucose level of diabetic rats. The superior effect was with G4 (Group No. 4) rats administrated 1.5 g LEG/kg/day (Leaves Extract of Green Globe). On the other hand results of the influence of artichoke leaves and heads as hypocholesterolemic action was in a positive way on the level of total cholesterol and reduced LDL and triglycerides levels and increased the level of glutathione peroxides, meanwhile it reduced the level of malondialdehyde (MDA) in rats serum. G3 (Group No. 3) [HFD (high fat diet) +1%cholesterol+1.5g LEG/kg/day] recorded the best results as hypocholesterolemic effect which could be attributed to their phenolic content. Our results indicated that, artichoke especially leaves extract of Green Globe (LEG) has good action as hypoglycemic and hypocholesterolemic.

Keywords: *artichoke, leave, head, hyperglycemic, hypercholesterolemia, rats*

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1. Introduction

Artichokes (*Cynara scolymus* L., *Asteraceae*) are an ancient crop and medicinal plant, the therapeutic potential of which was known to the ancient Egyptians, Greeks and Romans (Lattanzio et al., 2009). It contains caffeoylquinic acid derivatives (cynarin and chlorogenic acid) and flavonoids (luteolin and apigenin) [32,48]. Artichoke is of considerable economic importance for Spain and other countries. In Italy annual production of artichoke is estimated at 486600 tons FAO [10]. It is followed by Spain (198900 tons) and Egypt (180000 tons) as reported by FAO [10]. Reports from folk medicine as well as preliminary data from clinical studies have suggested that artichoke may have cholerectic, hypocholesterolemic and hypolipidemic properties [11,30,49].

Globe artichoke is considered a healthy food, due to its nutritional and phytochemical composition. It contains proteins, minerals, a low amount of lipids, dietary fiber and a high proportion of phenolics [13]. The phenolics include cynarin (1,3-di-O- caffeoylquinic acid), luteolin, cynaroside (luteolin-7-o-glucoside), scolmoside (luteolin-7-o-rutinoside); phenolic acids such as caffeic, coumaric, hydroxycinnamic, ferulic, caffeoylquinic acid derivatives; mono- and dicaffeoylquinic acids, including chlorogenic acid; acid alcohols; flavonoid glucosides, among others [13,41]. The extract obtained from the leaves has been shown to be hepatoprotective [34]. Artichoke leaf extract (ALE) is also reported to have a cholesterol reducing effect in hypercholesterolemic subjects (Joy and Haber, 2007). In addition, ALE has been found to decrease the production of reactive oxygen species, the oxidation of low density lipoproteins (LDL), and lipid peroxidation [26].

Although artichoke leaves are used as a herbal medicine and have been recognized since ancient times for their beneficial and therapeutic effects [15,31,32], the main use of this vegetable is the consumption of the edible immature flower heads (capitula), commonly referred to as "artichoke heads", which are eaten as fresh, canned or frozen vegetable [31]. More recently, the consumer demand for artichokes has increased because of their

reputation as a health food due to their nutritional and phytochemical composition [18,31].

The objective of this work was to find the chemical composition of two varieties of Artichoke " Green Globe and Violet" and extraction of phenolic compounds from the artichoke leaves and heads and also the fractionation and identification of these phenolic compounds as well as study the availability of artichoke phenolic compounds as hypoglycemic and hypocholesterolemic action.

2. Materials and Methods

2.1. Materials

Two varieties of fresh Artichoke (*Cynara scolymus* L.) "Green Globe and Violet" were obtained from the Horticultural Research Institute, Agriculture Research Centre, Giza, Egypt, in April 2013 at their full maturity. The samples were authenticated by Assistant Prof. Mostafa Kamal, Vegetable Crop Department, Horticultural Research Institute, Agriculture Research Centre, Giza, Egypt. Standards of phenolic acids: Chlorogenic acid; Cynarin; 1,5-di-o-Caffeoylquinic acid; Apignin 7-rutionside; Luteolin 7-rutionside; Narirutin; Caffeic acid; Salicylic acid; Gallicacid; Coumarin; Ferulic acid; and 2,2diphenyl-1-picrylhydrazyl (DPPH) (>95%) were purchased from Sigma Aldrich Company.

The fresh samples of artichoke were dried in an oven at 50°C for two days, ground, sealed in polypropylene bags, and stored at 4°C until analysis. One hundred and two male Albino rats weighing 180–200 g were purchased from the Research Institute of Ophthalmology, Giza, Egypt. Diagnostic kits were purchased from Bio-Diagnostics Company, Giza, Egypt. Alloxan monohydrate was purchased from Kemet Medical Company, Cairo, Egypt.

2.2. Determination of Proximate Composition of Dried Samples

The Analyses of moisture, ash, total carbohydrates, crude protein, crude fiber and fat were determined for the dried leaves and heads of the two varieties of artichoke (Green Globe and Violet) as described by the AOAC [3].

2.3. Extraction of Polyphenols

Amounts of 4 g of dried artichoke leaves or heads were homogenized with a homogenized IKA RW20 Germany and extracted by stirring with 100 ml aqueous methanol (60%, v/v) for 1 h at ambient temperature according to the method described by Schutz et al. [42]. After filtration through a filter paper, the extracts were evaporated to dryness in vacuo at 30°C. The residue was dissolved in methanol to yield a concentration (w/v) of 10 mg/mL.

2.4. Determination of Total Phenolic Content

The total phenolic content was determined according to the Folin–Ciocalteu method, described by Zheng and Wang [52], with some modifications, as suggested by Wang et al. [48]. Plant extracts were dissolved in methanol to yield a concentration (w/v) of 10 mg/mL. 50 µL aliquots were mixed with 1.25 mL of Folin–Ciocalteu reagent (diluted 1:10 fold) and 1 mL of 7.5% sodium carbonate solution. After 30min, absorbance was measured by Janway model 6705 spectrophotometer (England) at a λ=765 nm, at room temperature. The results were expressed gram as Chlorogenic acid equivalents per 100 g dried plant (g Chlorogenic acid /100 g).

2.5. Fractionation and Identification of Phenolic Compounds

The polyphenolic compounds of artichoke extracts were fractionated and identified for phenolic compounds by HPLC, according to the method described by Pinto et al [39]. Identification of individual phenolic compounds was performed on Hewlett- Packard HPLC (Model 1100), using a hypersil C18 reversed- phase column (25 × 4.6 mm) with 5µm particle size. Injection was done by means of a Rheodyne injection value (Model 7125) with 50µl fixed loop. The mobile phase was composed of solvent A (4.5% formic acid) and solvent B (80% of acetonitrile and 20% of solvent A). The program began with isocratic elution with 95% A (0-1min); then a linear gradient was used until 16 min, lowering A to 20%; from 17 min to 24 min, and A decreased to 0%. The flow rate was 1 ml min^{-1}, and the runs were integrated at 280 and 320, 360 nm for hydroxycinnamic acid and flavonoid derivatives, respectively. Scanning was performed from 200 to 600 nm. Phenolic compounds were identified by comparing retention times and UV-VIS spectra with those of pure standards and the range of calibration curves. The repeatability of the quantitative analysis was ± 4%. The analysis were replicated (n=3), and the contents given as means values, plus or minus the standard deviation. The results were expressed as grams of each compound per total phenolic compounds.

2.6. Determination of Antioxidant Activity

The diphenyl picrylhydrazyl (DPPH) radical scavenging activity was performed as described by Silva et al [44]. 250µg/ml methanolic solution of extract was prepared. The reaction mixture was 0.3 mL of extract with 2.7ml of a stock solution of DPPH (40µg/ml). The decrease in absorbance was determined at 516 nm until it reached a plateau (after 30 min), in the dark. The DPPH scavenging activity was calculated as $(A_0\text{-}A_1)/ A_0 \times 100$, where A_0 is the absorbance of the control (methanolic solution of DPPH) after 30 min incubation and A_1 is the absorbance of sample after 30 min incubation.

2.7. Animal Studies

One hundred and two male Albino rats weighing 180–200 g were obtained from the Research Institute of Ophthalmology, Giza, Egypt. The animals were housed individually in well aerated cages with screen bottom and fed on basal diet as described in A. O.A.C (2000) for 12 days as an adaptation period. Salt mixture and vitamin mixture were prepared as described in A.O.A.C [1,2] respectively. Temperature and humidity were maintained at 25°C and 60% respectively, food and water were given ad libitum.

2.7.1. Preparation of Artichoke Leaves and Head Aqueous Extracts

The artichoke leaves and head extracts were prepared by adding 1000 ml of distilled water to 100 g of leaves and head powder and kept at 60°C for 60 min. The aqueous extracts were filtered and the filtrate was evaporated to dryness at 50°C. The dry fraction was dissolved in distilled water just prior to use in the animal studies.

2.7.2. Extracts Administration to Animals

The rats were divided into two main groups as follows:

(I) Diabetic group
(II) hypercholesterolemic group

The group of diabetic experiment was carried out to 4 weeks. This main group contained 54 rats, divided into 9 sub-groups of rats, each having 6 rats as recommended by Fantini et al. (2011). All the diabetic rats were administrated orally (by stomach tube) with ALE or AHE. The designs of the groups of experimental rats are shown in Table 1.

Table 1. Composition of basal and different diabetic diets (g/100g diet)

Ingredient	(G1) Control basal diet	(G2) diabetic control	(G3) diabetic treated with insulin	(G4) diabetic +1.5g LEG	(G5) diabetic +1.5g HEG	(G6) diabetic +3g HEG	(G7) diabetic +1.5g LEV	(G8) diabetic +1.5g HEV	(G9) diabetic +3g HEV
Starch	65	65	65	65	65	65	65	65	65
Casein*	15	15	15	15	15	15	15	15	15
Corn oil	10	10	10	10	10	10	10	10	10
Cellulose	5	5	5	5	5	5	5	5	5
Vit- Mix	1	1	1	1	1	1	1	1	1
Salt-Mix	4	4	4	4	4	4	4	4	4
Artichoke Leaves extract (ALE)	-	-	-	1.5	-	-	1.5	-	-
Artichoke head extract (AHE)	-	-	-	-	1.5	3	-	1.5	3

* Casein contained 90% protein.

Diabetes was induced in overnight fasted animals by a single intraperitoneal injection of alloxan monohydrate, dissolved in 5% w/v normal saline at a dose of 150 mg/kg BW. The dose of alloxan was injected periodically for 3 days. Five days later, blood samples were collected from the eye plexuser by a fin capillary glass tube. The samples were centrifuged for 10 min at 3000 rpm and the serum was collected, blood glucose level was measured. The rats with blood glucose level ≥300 mg/dL were considered to be diabetic as recommended by Bagri et al. [4]. The groups were as follows: (G1) Control basal diet, (G2) diabetic control, (G3) diabetic injected with 20 units insulin, (G4) diabetic +1.5g LEG "Leaves extract Green Globe", (G5) diabetic +1.5g HEG "Heads extract Green Globe" , (G6) diabetic +3g HEG "Heads extract Green Globe" , (G7) diabetic +1.5g LEV "Leaves extract Violet" ,(G8) diabetic +1.5g HEV, "Heads extract Violet " (G9) diabetic +3g HEV "Heads extract Violet ".

The second main group of rats "hypercholesterolemia group" was carried out to 6 weeks. This second main group contained 48 rats, divided into 8 sub-groups, each having 6 rats as recommended by Kusku-Kiraz et al. [30]. All the hypercholesterolemic rats were administrated orally using stomach tube. The groups of experimental rats are shown in Table 2. The groups of rats were treated as follows: (G1) Control basal diet, (G2) high fat diet (HFD)+1%cholesterol, (G3) HFD+1%cholesterol+1.5g LEG, (G4) HFD+1%cholesterol+1.5g HEG, (G5) HFD + 1%cholesterol + 3g HEG, (G6) HFD+1%cholesterol+1.5g LEV, (G7) HFD + 1%cholesterol + 1.5g HEV, (G8) HFD + 1%cholesterol+3g HEV.

Table 2. Composition of basal and HF diets (g/100g diet)

Ingredient	(G1) Control basal diet	(G2) HFD +1%cholesterol	(G3) HFD +1%cholesterol +1.5g LEG	(G4) HFD +1%cholesterol +1.5g HEG	(G5) HFD +1%cholesterol +3g HEG	(G6) HFD +1%cholesterol +1.5g LEV	(G7) HFD +1%cholesterol +1.5g HEV	(G8) HFD +1%cholesterol +3g HEV
Starch	65	50.62	50.62	50.62	50.62	50.62	50.62	50.62
Casein*	15	15	15	15	15	15	15	15
Corn oil	10	8	8	8	8	8	8	8
Cellulose	5	5	5	5	5	5	5	5
Vit- Mix	1	1	1	1	1	1	1	1
Salt-Mix	4	4	4	4	4	4	4	4
Beef Tallow	-	15	15	15	15	15	15	15
Cholesterol	-	1	1	1	1	1	1	1
Choline bitartarate	-	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Cholic acid	-	0.18	0.18	0.18	0.18	0.18	0.18	0.18
Artichoke Leaves extract (ALE)	-	-	1.5	-	-	1.5	-	-
Artichoke head extract (AHE)	-	-	-	1.5	3	-	1.5	3

* Casein contained 90% protein.

At the end of experimental period rats were anaesthetized using diethyl ether and sacrificed. The blood was collected in tubes and centrifuged at 3000 rpm to obtain serum. Serum glucose, total cholesterol, HDL-

cholesterol, LDL - cholesterol, triglycerides (TG), and malondialdehyde (MDA) in organs and serum were determined according to the method of Trinders, [47], Richmond, [40], Burstein et al. [7], Wieland and Seidel, [50], Fassati and Prencipe, [12]; Ohkawa et al. [36] respectively. Glutathione peroxidase (GSH- PX) level in organs (liver and heart) and blood was determined according to the method of Paglia and Valentine, [37]. Results were calculated by using extinction coefficient (6.22×10^3 $M^{-1}cm^{-1}$) .Protein level was determined according to the method of Smith et al [45].

2.7.3. Organs Preparation before Glutathione Peroxidase Analysis

Organs were preparation according to the method of Paglia and Valentine, [3] as follows

1- Homogenize the sample in 4-8 volumes (per weight tissue) of cold buffer (50 mM phosphate buffer, pH 7.0, containing 5 mM EDTA and 1 mM 2-mercaptoethanol)

2- Centrifuge at 4000 rpm for 10-20 minutes at 2-8°C

3- Remove the supernatant fluid containing the enzyme.

2.7.4. Organs Preparation Before Malondialdehyde Analysis

Organs were preparation according to the method of Ohkawa et al. [36] as follows

1- Prior to dissection, perfuse tissue with a phosphate buffered saline solution, pH 7.4 containing 0.16 mg/ml heparin to remove any red blood cells and clots

2- Homogenize the tissue in 5-10 ml cold buffer (50mM potassium phosphate, pH 7.5) per gram tissue

3- Centrifuge at 4000 rpm for 15 minutes

4- Remove the supernatant for assay.

3. Statistical Analysis

Results were statistically analyzed by the least significant differences (L.S.D) at the level of probability procedure, according to Snedecor and Cochran, [46].

4. Result and Discussion

4.1. Proximate Composition of Artichoke

The chemical composition of the two varieties of artichoke "Green globe and Violet" is shown in Table 3. Results show that the moisture content of artichoke leaves "Violet" was significantly higher ($P \geq 0.05$) than that of the green globe leaves. Moreover significance could be also showed for the moisture of artichoke heads "Green Globe and Violet" that they recorded 74.54% and 77.70% respectively. Our results are in accordance with the results of Lutz, et al. [33]; they found that the moisture content of artichoke heads was 76.6%. Basay and Tokusoglu [5] reported that the moisture content of artichoke leaves was in the range of 79.86 to 83.03%.

Results in Table 3 show also the protein content of the two varieties of artichoke leaves and heads. Results show significance throughout the two varieties for both leaves and heads. The highest amount of crude protein was for the heads of green globe that recorded 18.25% followed by the heads of violet variety "17.74%". The protein content of the leaves for the two varieties violet and green globe recorded 12.87% and 8.74% successively. Our results are in agreement with the results of Hosseinzadeh et al. [21] who found that, the protein content of artichoke leaves (d.w) was in the range of 8.05% to 12.35%. However, our results was in disagreement with Lutz et al. [33] who found that the protein content of artichoke heads was only 15.96% which is less than our finding (17.74 to 18.25%).

Table 3. Proximate composition of the two varieties Green Globe and Violet of artichokes (on dry weight basis).

Component	Green Globe		Violet		LSD (P≥0.05)
	leaves	heads	leaves	heads	
Crude protein	8.74±0.21^{d}	18.25±0.22^{a}	12.87±0.07^{c}	17.74±0.21^{b}	0.3609
Crude Fat	2.34±0.06^{a}	2.06±0.05^{b}	2.36±0.14^{a}	1.93±0.17^{b}	0.2297
Crude fiber	31.47±0.24^{a}	28.63±0.11^{b}	28.97±0.16^{b}	26.96±0.19^{c}	0.3487
Ash	8.82±0.18^{a}	6.88±0.14^{b}	8.76±0.14^{a}	6.93±0.19^{b}	0.3163
Total Carbohydrates	48.63±0.44^{a}	44.18±0.13^{c}	47.04±0.11^{b}	46.44±0.76^{b}	0.8480
Moisture	78.67±0.26^{b}	74.54±0.21^{d}	80.55±0.24^{a}	77.70±0.17^{c}	0.4283

Different letters within each raw indicate significant difference
The data are expressed in mean ± S. D.

The fat content of the two varieties of artichoke is also shown in Table 3. No significant difference was shown for the fat content of the leaves of green globe or violet and also for the heads of the two varieties under investigation. However, significant difference was observed between the heads and the leaves of the two varieties. These results are in accordance with the results of Hosseinzadeh et al. [21] who found that, the fat content of artichoke leaves was in the range of 1.6 to 2.3% on dry wt. basis. Meanwhile, Jimenez-Escrig et al. [23] reported that the fat content of artichoke heads was only 1.69% (d.w).

Results in Table 3 show also the crude fiber content for the two varieties of artichoke. Significant difference could be shown among all the samples under investigation. The highest amount of fiber was for the leaves of green globe variety (31.47%). The lowest amount was recorded for the heads of violet variety (26.96%). So, the content of crude fiber of leaves for both varieties was higher than the corresponding heads.

Results of the ash content of the leaves for the two varieties of artichoke was non significant as shown in Table 3, that recorded 8.82 and 8.76% for green globe and violet respectively. The ash content for the heads of the two varieties was also non significant. However, the ash content of the leaves was higher than that of heads. Our results are in accordance with the results of Lutz et al. [33] who reported that, the content of artichoke heads was 7.04%. In addition, Hosseinzadeh et al. [21] reported that the ash content of artichoke leaves was 9.01%. Percentage of total carbohydrates (d.w) of the two varieties of

artichoke was significant difference as shown in Table 3. The highest amount of total carbohydrates was recorded for the leaves of green globe followed by the leaves of violet variety. The lowest percentage of total carbohydrates was for the heads of green globe variety.

Table 4. Total phenolic content and antioxidant activity for the two varieties of dried artichoke (Green Globe and Violet)

Plant part	Green Globe		Violet		LSD (P≥0.05)
	leaves	**heads**	**leaves**	**heads**	
Total phenolic content (g chlorogenic acid/ 100g dried sample)	8.603±0.265[a]	2.697±0.116[c]	5.666±0.257[b]	1.818±0.243[d]	0.4303
Antioxidant activity %	85.35±0.599[a]	67.83±0.930[c]	80.49±0.911[b]	62.48±0.920[d]	1.701

Different letters within each raw indicate significant difference
The data are expressed in mean ± S. D.

4.2. Total Phenolic Content and Antioxidant Activity of Different Varieties of Artichoke

Results in Table 4 show the total phenolic content (as g chlorogenic acid/ 100g dried artichoke) for the dried leaves and heads for the two varieties "Green Globe and Violet" artichoke. The highest concentration of total phenolic content was for the leaves of the two varieties that were significantly higher than the corresponding heads. Leaves of Green Globe variety had the highest concentration of total phenolic content (8.603%), followed by Violet leaves that recorded (5.666%). However, the edible part (heads) of artichoke was significantly lower than the leaves. Our results are in accordance with the results of Wang et al. [48] who reported that the total phenolic content of leaves of Green Globe was in the range of 8.760 to 9.561% (as chlorogenic acid). Meanwhile, they found that, the total phenolic content of leaves of violet variety was (6.806%), which is slightly higher than our results. On the other hand, Wang et al. [48] reported that the phenolic content of Green Globe heads was in the range of 1.600 to 2.238%. They found that the phenolic content of Violet heads was 2.893% that was higher than finding.

Results in Table 4 show also the antioxidant activity (%) of the two dried "Green Globe and Violet" varieties artichoke. Results of Green Globe leaves recorded the highest percentage of antioxidant activity (85.35%) that was superior to the other samples under investigation. Meanwhile, the lowest antioxidant activity was with the Violet heads (62.48%) that was inferior to the other samples. Results show that the total phenolic content was in direct proportion with the increasing of antioxidant activity. Wang et al. [48] found that, samples that had low phenolic compound content also had lower antioxidant activity. They also found that, the scavenging DPPH free radical activities of the different artichoke samples were highly correlated to the total phenols.

Table 5. Phenolic compounds (g/100g) for the two varieties (Green Globe and Violet) of dried artichoke heads and leaves.

Phenolic acids	**Green Globe**		**Violet**	
	leaves	**heads**	**leaves**	**heads**
5-o-Caffeoylquinic acid (Chlorogenic)	4.0012	0.2141	1.2910	0.1245
1,3-di-o-Caffeoylquinic acid (Cynarin)	1.5823	0.2792	1.4952	0.1936
1,5-di-o-Caffeoylquinic acid	0.0036	0.0516	0.0034	0.0491
Apignin	0.0046	0.0101	0.0037	0.0071
Luteolin	0.0384	0.0224	0.0327	0.0203
Caffeic acid	0.0102	0.0086	0.0064	0.0059
Salicylic acid	0.0202	0.0297	0.0239	0.0086
Gallic acid	0.0015	0.0057	0.0175	0.0034
Coumarin	0.0018	0.0028	0.0046	0.0024
Ferulic acid	-	0.0051	0.0093	0.0036
Narirutin	-	0.175	-	0.238
Total	5.6638	0.8043	2.8877	0.6565

4.3. Identification of Phenolic Compounds for the Two Varieties of Dried Artichoke.

The phenolic compounds of the two varieties of artichoke "Green Globe and Violet" extracts were identified by HPLC, results are shown in Table 5. Phenolic compound 5-o-Caffeoylquinic acid (Chlorogenic acid) was found to be in high concentration (g/100g dried sample) for the leaves of Green Globe variety that recorded 4.0012%, meanwhile it was 1.2910% for the leaves of Violet variety. In contrast, the phenolic compound5-o-Caffeoylquinic acid (Chlorogenic acid) was in low percentage for Green Globe and Violet heads that recorded 0.2792 and 0.1245% respectively.

The phenolic compound 1,3-di-o-Caffeoylquinic acid (Cynarin) was found in little amounts than that Chlorogenic acid, but still in higher amount than 1,5-di-o-Caffeoylquinic acid into the samples of artichoke under investigation. Shen et al. [43] reported that, the three compounds; (5-o-Caffeoylquinic acid (Chlorogenic acid), 1,3-di-o-Caffeoylquinic acid (Cynarin) and 1,5-di-o-Caffeoylquinic acid are the major active compounds in artichoke, they are considered to be responsible for their antiatherogenic action. The Caffeoylquinic acids are natural antioxidant with potential health benefits in the

context of inhibiting the development of cancers, exacerbated by the presence of reactive oxygen specie [14]. The phenolic compound Apignin was found in small amount for the two varieties of artichoke either leaves or heads. Pandino et al. [38] and Justesen and Knuthsen [25] reported that, Apignin is a compound which is seldom encountered in the plant kingdom, being found only in some herbs and vegetable.

Luteolin phenolic compound was also found in leaves and heads for two varieties of artichoke. Pandino et al. [38] and Kukic et al. [29] reported that, Luteolins are of interest since they show antimicrobial activity and inhibit cholesterol synthesis.

Table 6. Influence of leaves and heads artichoke extracts on the level of serum glucose, total cholesterol, LDL, HDL cholesterol and triglycerides in diabetic rats

Groups	Glucose (mg/dl)	Total cholesterol (mg/dl)	LDL (mg/dl)	HDL (mg/dl)	Triglycerides (mg/dl)
G1: Control (basal diet)	98.31± 0.95^{i}	88.95± 1.04^{g}	29.57± 0.93^{h}	48.39± 0.91^{a}	48.39±1.06^{g}
G2: Diabetic	328.36± 1.05^{a}	128.51± 1.06^{a}	80.95± 0.95^{a}	30.84± 0.93^{g}	83.54±1.07^{a}
G3: Diabetic injected with 20 units insulin /kg/day	120.90± 1.42^{h}	92.25± 1.05^{f}	29.11± 1.06^{h}	47.85± 0.85^{a}	48.10±1.09^{g}
G4: Diabetic +1.5g LEG/kg/day	127.04± 0.50^{g}	92.47± 1.07^{f}	31.73± 0.91^{g}	45.08± 1.06^{b}	50.33±0.87^{f}
G5: Diabetic +1.5g HEG/kg/day	177.98± 0.97^{c}	104.43± 1.07^{c}	50.02± 1.07^{c}	35.18± 1.09^{e}	60.42±1.06^{c}
G6: Diabetic +3g HEG/kg/day	148.58± 0.93^{e}	96.20± 0.92^{e}	38.47± 1.06^{e}	42.30± 0.91^{c}	54.15±0.88^{e}
G7: Diabetic +1.5g LEV/kg/day	145.42± 1.07^{f}	96.13± 0.91^{e}	36.03± 1.05^{f}	43.18± 0.89^{c}	53.28±0.91^{e}
G8: Diabetic +1.5g HEV/kg/day	186.61± 1.08^{b}	108.78± 0.91^{b}	54.25± 1.09^{b}	33.03± 1.06^{f}	63.95±0.90^{b}
G9: Diabetic +3g HEV/kg/day	159.90± 1.08^{d}	100.32± 1.07^{d}	43.28± 1.05^{d}	39.92± 1.07^{d}	57.13±1.07^{d}
LSD(P≥0.05)	1.771	1.742	1.759	1.683	1.680

LEG (leaves extract of Green Globe). HEG (head extract of Green Globe) LEV (leaves extract of Violet). HEV (head extract of Violet)
Different letters within each column indicate significant difference
The data are expressed in mean ± S. D.

4.4. Animal studies of Artichoke Leaves and Heads Aqueous Extracts.

4.4.1. Influence of Leaves and Head Extracts on the Serum Levels of Glucose, Total Cholesterol, LDL, HDL and Triglycerides in Diabetic Rats

Results in Table 6 show the glucose level, as well as total cholesterol, LDL, HDL and triglycerides of different groups of rats. Results show that the glucose level (mg/dl) for G2 group of rats was the highest level that recorded 328.36, it reduced up to 120.90 mg/dl for G3 of rats that injected with 20 units insulin, they were in significant difference. Moreover, the other groups of rats G4 up to G9 were in significant difference. This indicates about the effect of artichoke leaves or heads extracts on the glucose level of diabetic rats. The superior effect was with G4 "rats had 1.5 g LEG/kg/day" that recorded 127.04 mg/dl. The group of rats G7 those had 1.5 g LEV/kg/day recorded only 145.42 mg/dl glucose level. Results indicate about the powerful of phenolic compounds of Green globe.

Results in Table 6 also show the changes of total cholesterol, LDL, HDL and triglycerides (mg/dl) in the different experimental rats. The highest reduction in total cholesterol, LDL, HDL and triglycerides could be observed for G3 group of rats that injected with insulin. Moreover, results show some reduction for these parameters especially for G4 "Diabetic + 1.5g LEG/kg/day". It could be attributed to the effect of the amount of phenolic compounds in LEG compared with LEV. Results show also that G6 "group of diabetic rats +3g HEG" recorded higher effect on total cholesterol, LDL and triglycerides than G9 "group of diabetic rats + 3g HEV/kg/day". On the other hand, the HDL parameter show decrease for G2 "Diabetic group of rats". G3 followed by G4 had higher increase for HDL comparing with the other groups of rats under investigation. Gylling et al. [19] reported that, the hypertriglyceridemia and hypercholesterolemia are associated with abnormalities of lipoprotein levels in the blood, and streptozotocin increased the levels of LDL in the blood, and decreased the HDL level. They also added that, dyslipidemia is one of the major cardiovascular risk factors. It has been suggested that insulin deficiency in diabetes mellitus is associated with a variety of abnormalities in metabolic and regulatory processes, which cause the accumulation of lipids such as TC and TG in diabetic patients [17]. In addition, our results are in accordance with the results of Heidarian and Soofiniya [20] who reported that, the levels of serum glucose, total cholesterol and triglycerides in the streptozotocin-treated rats markedly increased, compared to normal control rats. However, the elevated serum glucose, triglyceride and total cholesterol levels significantly reduced by the oral administration of artichoke (1000and2000 mg/kg) in a dose-dependent manner.

Artichoke leave extract (ALE) contains bioactive and flavonoid compounds such as caffeoylquinic acids and luteolin glucosides. As it is known, cynarin is a major dicaffeoylquinic acid and chlorogenic acid is the main monocaffeoylquinic acid, whereas luteolin-7-O glucoside is the major flavonoid [32,48]. Their results indicated that ALE had a lipid lowering effect on the diabetic rats.

4.4.2. Influence of Leaves and Head of the Two Varieties Artichoke Extracts on the Levels of Total Cholesterol, LDL, HDL, Triglycerides, Glutathione Peroxidase and Malondialdehyde in Organs, Blood and Serum of Hypercholesterolemic Rats

Results in Table 7 show that the group G3 (HFD+1%cholesterol+1.5g LEG/kg/day) had the lowest level of total cholesterol, LDL, and triglycerides, these results were significantly difference. The group G6 (HFD+1%cholesterol+1.5g LEV/kg/day) had also low level of these parameters; this could be attributed to the

phenolic content of the two varieties. It could be noticed that, the artichoke leaves had better effect on the lowering of total cholesterol, LDL, and triglycerides than the artichoke heads for the two varieties even when the concentration of given dose increased up to 3g HE/kg/day. Results in Table 7 show that, the highest level of HDL was with G3 followed by G6. Our results are in accordance with the results of Kucukgergin et al. [28] who reported that, artichoke leaf extract treatment for hypercholesterolemic rats was useful for decreasing serum cholesterol and triglycerides levels of rats. Moreover, Esterbauer et al. [9], Itabe [22]; Nakajima et al. [35] reported that conditions leading to increased LDL oxidation have been considered to be a probable atherosclerotic risk factor.

Table 7. Influence of artichoke leaves and heads extracts on the levels of total cholesterol, LDL, HDL, triglycerides, glutathione peroxidase (GSH- PX) and malondialdehyde (MDA) in organs, blood serum of hypercholesterolemic rats

Groups	Total cholesterol (mg/dl serum)	LDL (mg/dl serum)	HDL (mg/dl serum)	Triglycerides (mg/dl serum)	GSH-PX			MDA		
					Liver (nmol/mg protein)	Heart (nmol/mg protein)	Blood (nmol/ml)	Liver (nmol/gT)	Heart (nmol/gT*)	Serum (nmol/ml)
G1	86.18±1.33^{g}	25.57± 0.80^{g}	50.39± 0.47^{a}	42.89±1.49^{t}	853.41± 3.58^{a}	348.71± 2.04^{a}	46.30± 0.65^{a}	205.27± 1.31^{g}	121.43± 1.13^{t}	5.16±0.45^{e}
G2	148.48±1.91^{a}	99.90± 1.29^{a}	30.60± 0.64^{h}	89.87±3.11^{a}	659.39± 2.83^{g}	231.49± 2.75^{g}	24.85± 0.29^{t}	240.88± 2.23^{a}	166.95± 1.13^{a}	10.74±0.45^{a}
G3	95.33±1.46^{t}	38.74± 3.00^{t}	48.68± 0.63^{b}	43.37±1.27^{t}	834.12± 3.37^{b}	332.77± 2.92^{b}	47.25± 0.56^{a}	208.07± 1.32^{fg}	122.53± 1.27^{et}	5.29 ±0.22^{e}
G4	120.93±0.55^{c}	67.34± 1.48^{c}	41.06± 1.11^{t}	56.01±2.22^{c}	788.73± 0.86^{e}	270.40± 2.75^{e}	34.37± 0.36^{d}	219.76± 1.34^{c}	132.56± 1.13^{c}	8.61±0.22^{b}
G5	100.91±0.67^{e}	45.92± 0.61^{e}	44.96± 1.01^{d}	49.28±0.39^{e}	795.60± 2.80^{d}	293.55± 2.48^{d}	38.20± 0.32^{c}	212.11± 1.50^{e}	125.97± 1.13^{d}	6.32±0.68^{d}
G6	99.18±0.88^{e}	43.80± 1.42^{e}	46.62± 0.94^{c}	47.78±1.76^{e}	815.69± 1.93^{c}	309.05± 3.10^{c}	44.28± 0.53^{b}	211.30± 1.24^{et}	124.42± 1.13^{de}	6.07±0.36^{de}
G7	125.18±0.37^{b}	72.70± 0.79^{b}	39.04± 0.71^{g}	61.75±1.11^{b}	755.88± 3.95^{t}	254.62± 2.44^{t}	29.33± 0.87^{e}	224.39± 1.27^{b}	136.16± 1.31^{b}	9.49±0.45^{b}
G8	104.10±1.74^{d}	50.64± 0.76^{d}	43.35± 0.73^{e}	52.37±1.27^{d}	785.71± 1.22^{e}	269.26± 0.73^{e}	35.36± 0.21^{d}	215.82± 1.28^{d}	130.83± 1.27^{c}	7.29±0.22^{c}
LSD (P≥0.05)	2.146	2.536	1.403	3.086	6.419	5.779	1.199	3.39	2.749	0.955

G1: Control (basal diet), G2: HFD+1%cholesterol, G3: HFD+1%cholesterol+1.5g LEG/kg/day, G4: HFD+1%cholesterol+1.5g HEG/kg/day, G5: HFD+1%cholesterol+3g HEG/kg/day, G6:HFD+1%cholesterol+1.5g LEV/kg/day, G7: HFD+1%cholesterol+1.5g HEV/kg/day and G8: HFD+1%cholesterol+3g HEV/kg/day

* gT: g tissue

Different letters within each column indicate significant difference

The data are expressed in mean ± S. D.

On the other hand, artichoke leaves extract has been proposed to be antiathergenic, due to its lipid-reducing and antioxidant effects [24,48]. It is reported to inhibit cholesterol biosynthesis in hepatocytes [15] decrease the oxidation of LDL [6,23,51]. Artichoke leaves extract (ALE) is also considered as choleretic, enhancing biliary excretion of cholesterol and increasing its conversion to bile acids [24]. In addition, ALE is known to have antimicrobial properties in gut, disrupting the intestinal microflora, thus affecting the absorption of various compounds including cholesterol [53]. Thus, ALE may influence the intestinal absorption and excretion of cholesterol from organism, besides suppression of endogenous cholesterol synthesis. The inhibition of cholesterol biosynthesis is due to luteolin, which modulates the HMG-CoA reductase activity (the key enzyme in the cholesterol biosynthesis pathway) [16,27]. Moreover, chlorogenic acid and luteolin may prevent atherosclerosis by inhibiting low-density lipoproteins (LDL) oxidation [6]. Therefore, artichoke leaf extracts show hypocholesterolemic activity, due to two parallel mechanisms: reduction of cholesterol biosynthesis and inhibition of LDL oxidation [8,15]. Artichoke extracts are well tolerated, and may be useful for the preventive treatments of mild hypercholesterolemia.

Results in Table 7 also show the changer in GSH-PX for the liver and heart as well as blood of different groups of rats. Significant difference could be shown for all tested groups of rats. The highest level of GSH-PX (nmol/mg protein) for rats liver was recorded for G1 "Control group" followed by G3 "rats fed with HFD+ 1% cholesterol+ 1.5g LEG/kg/day. G6" HFD+ 1% cholesterol+ 1.5g LEV/kg/day" had high level of GSH-PX. Results show that the artichoke leave extract had good effect on GSH-PX compared with artichoke head extract for the two varieties under investigation. Results of GSH-PX of heart for different groups of rats was in the same pass way as for the corresponding liver as mentioned before. Moreover, GSH-PX in blood of different groups of rats was significantly different. However, no significant difference could be noticed for G1 and G3 they recorded the highest values of GSH-PX. In contrast G2 "HFD+ 1% cholesterol" recorded the lowest value of GSH-PX.

It has also been reported that artichoke leave extract treatment caused significant increases in GSH-PX activities in liver [34]. Therefore, Kuçükgergin, et al. [28] observed that induction of GSH-PX by artichoke leaves extract may also be assumed to contribute to its antioxidant properties.

Results in Table 7 show the MDA (nmol/ gT or nmol/ ml serum) level in different groups of rats. The decrement in the levels of MDA from 240.88 for G2 to 208.07 for G3 could be attributed to the influence of artichoke leave extract in liver and heart tissues they were significantly different, G1 was in the normal range of MDA. Reduction of MDA in serum of rats was noticed after having artichoke leaves or head extracts in their diets. Kuçükgergin, et al. [28] reported that artichoke leaf extract caused significant decreases in MDA levels in the liver and heart tissues with increase in hepatic GSH-PX activities in hypercholesterolemic rats.

5. Conclusion

From obvious results, it could be concluded that Chlorogenic acid, Cynarin, 1,5-di-o-Caffeoylquinic, luteolin and apigenin were identified from the aqueous methanolic extracts of two artichoke (Green Globe and Violet). Regarding to the effect of artichokes extract on diabetic

and hypercholesterolemic rats, results indicated that G4 which administrated 1.5 g LEG/kg/day showed the lowest value of glucose level of diabetic rats. While G3 which administrated 1.5 g LEG/kg/day showed higher reduction on total cholesterol level of hypercholesterolemic rats and increased the GSH-PX and HDL level. So, the results recommended that artichoke had important biological benefits.

References

[1] Association of Official Agricultural Chemists, "Official Method of Analysis," 17 th Edition, AOAC, USA: Washington, 1995.

[2] Association of Official Agricultural Chemists, "Official Method of Analysis," 17 th Edition, AOAC, USA: Washington, 2000.

[3] Association of Official Agricultural Chemists, "Official Method of Analysis," 2nd Edition, AOAC, USA: Washington, 2005

[4] Bagri, P. Ali, M. Aeri, V. Bhowmik, M. and Sultana, S. Antidiabetic effect of Punica granatum flowers: effect on hyperlipidemia pancreatic cells lipid peroxidation and antioxidant enzymes in experimental diabetes. Food Chemistry Toxicology 47: 50-54. 2009.

[5] Basay, S. and Tokusoglu, O. The proximate composition and quality characteristics in artichoke (cynara scolymus L.) varieties developed with clonal selection. Journal of Food Agriculture and Environment 11: 584-587. 2013.

[6] Brown, J.E. Rice-Evans, C.A. Luteolin-rich artichoke extracts protect low density lipoprotein from oxidation in vitro. Free Radic Res 29:247-255. 1998.

[7] Burstein, M.; Scholnick, H. and Morfin, R. Rapid method for the isolation of lipoproteins from human serum by precipitation with polyanions. Journal Lipid Research, 11(6):583-595. 1970.

[8] Bundy, R. Walker, A.F. Middleton, R.W. Wallis, C. and Simpson, H.C.R. Artichoke leaf extract (Cynara scolymus L.) reduces plasma cholesterol in otherwise healthy hypercholesterolemic adults: a randomized, double blind placebo controlled trial. Phytomedicine 15:668-675. 2008.

[9] Esterbauer, H. Gebicki, J. Puhl, H. and Jurgens, G. The role of lipid peroxidation and antioxidant in oxidative modification of LDL. Free Radic Biol Med 13: 341-390. 1992.

[10] FAO, Food Agriculture Organization. The total world production of artichoke. www.fao.org. 2009.

[11] Fantini, N. Colombo, G. Giori, A. Riva, A. Morazzoni, P. Bombardelli, E. and Carai, M. Evidence of glycemia-lowering effect by a cynara scolymus L. extract in normal and obese rats. Phytotherapy Research 25: 463-466. 2011.

[12] Fassati, P. and Prencipe, L. Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. Clinical Chemistry. 28: 2077-2080. 1982.

[13] Fratianni, F. Tucci, M. De-Palma, M. Pepe, R. and Nazzaro, F. Polyphenolic composition in different parts of some cultivars of globe artichoke (Cynara Cardunculus L. var. scolymus (L) Fiori). Food Chemistry 104:1282-1286. 2007.

[14] Fukumoto, L. R. and Mazza, G. Assessing antioxidant and proxidant activities of phenolic compounds. Journal of Agricultural and Food Chemistry, 48:3597-3604. 2000.

[15] Gebhardt, R. Antioxidative and protective properties of extracts from leaves of the artichoke (Cynara scolymus L.) against hydroperoxide-induced oxidative stress in cultured rat hepatocytes. Toxicol Appl Pharmacol 144:279-286. 1997.

[16] Gebhardt, R. Inhibition of cholesterol biosynthesis in HepG2 cells by artichoke extracts is reinforced by glucosidase pretreatment. Phytother Res 16:368-372. 2002.

[17] Goldberg, R.B. Lipid disorders in diabetes. Diabetes Care, 4: 561-572. 1981.

[18] Guida, V. Ferrari, G. Pataro, G. Chambery, A. Di-Maro, A. and Parente, A. The effects of ohmic and conventional blanching on the nutritional bioactive compounds and quality parameters of artichoke heads. LWT-Food Science and Technology. 53: 569-579. 2013.

[19] Gylling, H. Tuominen, J.A. Koivisto, V.A. and Miettinen, T.A. Cholesterol metabolism in type 1 diabetes. Diabetes, 53: 2217-2222. 2004.

[20] Heidarian, E. and Soofiniya,Y. Hypolipidemic and hypoglycemic effects of aerial part of Cynara scolymus in streptozotocin-induced diabetic rats. Journal of Medicinal Plants Research 5(13) 2717-2723. 2011

[21] Hosseinzadeh, M. Shekari, F. Janmohammadi, M. and Sabaghnia, N. Effect of sowing date and fliar application of salicylia acid on forage yields and quality of globe artichoke (cynara scolymus L.) Annales 8: 50-59. 2013.

[22] Itabe, H. Oxidized low density lipoproteins: what is understood and what remains to be clarified. Biol Pharm Bull 26: 1-9. 2003.

[23] Jimenez-Escrig, A. Dragsted, L. Daneshvar, B. Pulido, R. and Saura-Calixto, F. In vitro antioxidant of edible artichoke(cynara scolymus L.) and effect of biomarkers of antioxidant in rats. Journal of Agriculture and Food Chemistry 51:5540-5545. 2003.

[24] Joy, J. F. and Haber S. L. Clinical uses of artichoke leaf extract. Am. J. Health-Syst. Pharm., 64:1906-1909. 2007.

[25] Justesen, U. and Knuthsen, P. Composition of flavonoids in frsh herbs and calculation of flavonoid intake by use of herbs in traditional Danish dishes. Food Chemistry, 73:245-250. 2001.

[26] Juzyszyn, Z. Czerny, B. Pawlik, A. and Drozdzik, M. The effect of Artichoke (Cynara scolymus L.) extract on ROS generation in HUVEC cells. Phytother. Res. 22: 1159-1161. 2008.

[27] Kraft, K. Artichoke leaf extracts: recent findings reflecting effects on lipid metabolism, liver and gastrointestinal tracts. Phytomedicine 4:369. 1997.

[28] Kuçukgergin, C. A. Aydın, A. F. Özdemirler-Erata, G. Mehmetçik, G. Koçak-Toke, N. and Uysal, M. Effect of artichoke leaf extract on hepatic and cardiac oxidative stress in rats fed on high cholesterol diet. Biol Trace Elem Res, 135:264-274. 2010.

[29] Kukic, J. Popovic, V. Petrovic, S. Mucaji, P. Ciric, A. Stojkovic, D. and Sokovic, M. Antioxidant and antimicrobial activity of Cynara cardunculus extracts. Food Chemistry, 107:861-868. 2008.

[30] Kusku-Kiraz, Z. Mehmetcik, G. Dogru-Abbasglu, S. and Uysal, M. Artichoke leaf extract reduces oxidative stress and lipoprotein dyshomeostasis in rats fed on high cholesterol diet. Phytotherapy Research 24: 565-570. 2010.

[31] Lattanzio, V. Kroon, P. A. Linsalata, V. and Cardinali, A. Globe artichoke: A functional food and source of nutraceutical ingredients. Journal of Functional Foods 1:131-144. 2009.

[32] Llorach, R. Espin, J.C. Tomas-Barberan, F.A. and Ferreres, F. Artichoke (Cynara scolymus L.) byproducts as a potential source of health-promoting antioxidant phenolics. J Agric Food Chem 50:3458-3464. 2002.

[33] Lutz, M. Henriquez, C. and Escobar, M. Chemical composition and antioxidant properties of mature and baby artichokes (cynara scolymus L.) raw and cooked. Journal of Food Composition and Analysis 24: 49-54. 2011.

[34] Mehmetçik, G. Özdemirler, G. Koçak-Toker, N. Cevikbaş, U. and Uysal, M. Effect of pretreatment with artichoke extract on carbon tetrachloride-induced liver injury and oxidative stres. Exp Toxicol Pathol 60:475-480. 2008.

[35] Nakajima, K. Nakano, T. and Tanaka, A. The oxidative modification hypothesis of atherosclerosis: The comparison of atherogenic effects on oxidized LDL and remnant liopoproteins in plasma. Clin Chim Acta 367:36-47. 2006.

[36] Ohkawa, H. Ohishi, N and Yagi, K. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal. Biochem., 95: 351-358. 1979.

[37] Paglia, D.E. and Valentine, W.N. Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. J Lab Clin Med 70:158-169.1967.

[38] Pandino, G. Lombardo, S. and Mauromicale, G. Globe artichoke leaves and floral stems as a source of bioactive compounds. Industrial Crops and Products, 44:44-49. 2013.

[39] Pinto, M. S. Lajalo, F. M. and Genovese, M. I. Bioactive compounds and quantification of total ellagic acid in strawberries (Fragaria×ananassa Duch) Food Chemistry 107: 1629-1635. 2008.

[40] Richmond, W. Preparation of cholesterol oxidase from Nocardia sp. And its application to the enzymatic assay of total cholesterol in serum. Clinical Chemistry, 19(12):1350-1356.1973.

[41] Sanchez-Rabaneda, F. Jauregui, O. Lamuela-Raventos, R. M. Bastida, J. Viladomat, F. and Codina, C. Identification of phenolic compounds in artichoke waste by high performance liquid chromatography-tandem spectrometry. J. of Chromatography A 1008: 57-72. 2003.

[42] Schutz, K. Kammerer, D. Carle, R. and Schieber, A. Identification and quantification of caffeoylquinic acids and flavonoids from

artichoke (cynara scolymus L) heads, juice and pomace by HPLC-DAD-ESI/MS. J. Agric. Food Chem. 52: 4090-4096. 2004.

[43] Shen, Q. Dai, Z. and Yanbin, L. Rapid determination of caffeoylquinic acid derivatives in cynara scolymus L by ultrafast liquid chromatography/ tandem mass spectrometry based on a fused core C18 column. Journal of separation science, 33:3152-3158. 2010.

[44] Silva, C.G. Herdeiro, R.S. Mathias, C.J. Panek, A.D. Silveira, C.S. Rodrigues, V.P. Renno, M.N. Falcao, D.Q. Cerqueira, D.M. Minto, A.B. Nogueira, F.L. Quaresma, C.H. Silva, J.F. Menezes, F.S. and Eleutherio, E.C. Evaluation of antioxidant activity of Brazilian plants. Pharmacol. Res. 52(3):229-233. 2005.

[45] Smith, P.K. Krohn, R.I. Hermanson, G.T. Mallia, A.K. Gartner, F.H. Provenzano, M. Fujimoto, E.K. Goeke, N.M. Olson, B.J. and Klenk, D.C. Measurement of protein using bicinchoninic acid. Anal Biochem 150:76-85.1985.

[46] Snedecor, G. W. and Cochran, W. G. Statistical methods. Oxford and J. B. H Publishing Com. 7th edition. 1980.

[47] Trinders, P. Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. Annala of Clin. Biochem. 6: 24-33.1969.

[48] Wang, M. F. Simon, J. E. Aviles, I. F. He, K. Zheng, Q. Y. and Tadmor, Y. Analysis of antioxidative phenolic compounds in artichoke (Cynara scolymus L.). Journal of Agricultural and Food Chemistry, 51, 601-608. 2003.

[49] Wider, B. Pittler, M. H. Thompson-Coon, J. and Ernst, E. Artichoke leaf extract for treating hypercholesterolaemia. Cochane Database Syst Rev. 4: CD003335. 2009.

[50] Wieland, H. and Seidel, D. A. simple specific method for precipitation of low density lipoproteins. Journal of Lipid Research, 24(7):904.1983.

[51] Zapolska-Downar, D. Zapolska-Downar, A. Naruszewicz, M. Siennicka, A. Krasnodebska, B. and Kolodziej, B. Protective properties of artichoke (*Cynara scolymus* L.) against oxidative stress induced in cultered endothelial cells and monocytes. Life Sci 71: 2897-2908. 2002.

[52] Zheng, W. and Wang, S. Y. Antioxidant activity and phenolic compounds in selected herbs. Journal of Agricultural and Food Chemistry, 49: 5165-5170. 2001.

[53] Zhu, X. Zhang, H. and Lo, R. Phenolic compounds from the leaf extract of artichoke (*Cynara scolymus* L.). and their antimicrobial activities. J Agric Food Chem 52: 7272-7278. 2004.