EVALUATION OF ANTIOXIDANT ACTIVITY, QUANTITATIVE ESTIMATION OF PHENOLS AND FLAVONOIDS OF OAK ACORN FROM ERBIL/KURDISTAN

GULZAR ISMEAL IBRAHIM
Dept. of chemistry, College of Education, University of Salahaddin, Kurdistan Region-Iraq.
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ABSTRACT

In this study, antioxidant activity and total phenolic content of aqueous extract of oak acorn Quercus *robur (*belongs to *fagaceae* family*)* was investigated by using *in vitro* antioxidant models including total antioxidant capacity, reducing power and metal chelating activity. The total contents of phenol, flavonoid and alkaloid were quantitatively estimated in oak's acorn and its shell. Result showed that total phenolic, flavonoid, and alkaloid contents were higher in acorn than in its shell. The antioxidant activity of aqueous extract of acorn and its shell were determined according to the phosphomolybdenum method, reducing power and metal chelating was determined by using spectrophotometer. The results were compared to standard compounds such as ascorbic acid, rutin, gallic acid and colchicine. The overall result showed that oak acorn can serve as good sources of bioactive poly phenol in human diet and can be considered as a good candidate for nutritional supplement formulation due to their high concentrations of phenolic compounds, flavonoids, as well as their strong antioxidant activity.

Keywords: Acorn; *Quercusrobur*, *poly phenols, flavonoids, alkaloid, antioxidant activity, reducing power, metal chelating power.*

INTRODUCTION

The commercial development of plants as sources of antioxidants to enhance health and food preservation is of current interest (Rice-Evans, et al., 1997). Epidemiological studies have indicated the presence of a positive association between the consumption of phenolic-rich foods or beverages and the prevention of disease (Salbert, et al., 2000). Phenolic compounds are often related to the antioxidants activity of plant due to their ability to absorb and / or neutralize free radical by means of quenching unpaired oxygen radical or decomposing peroxides. In the present times, there is an increasing interest in natural antioxidants presented in dietary plants, which may contribute to prevent oxidative disorder in human by replacing the use of synthetic antioxidants which being also recognized as carcinogenic agents (Ito, et al., 1983). Synthetic antioxidants like butylated hydroxyl anisol (BHA), butylated hydroxyl toluene (BHT), gallic acid esters have been suspected to be carcinogenic. Hence, strong limitation has been placed on their use and there is a trend to replace them with naturally occurring antioxidants (Barlow, 1990). Sources of natural antioxidants are primarily phenolics that may occur in all products and parts of plant such as fruits, vegetables, nuts, seeds, leaves and bark. Due to their potential antioxidant action, plant phenols and poly phenols, with their potential to act as antioxidants, play a major role in the prevention of various pathological conditions such as cancer, cardiovascular and neurodegenerative diseases like Alzheimer disease which is believed to be associated with oxidative stress (Losso, *et al*., 2007). It have been reported that many crude extract and pure natural compounds exhibit potent antioxidants potential (Chu, 2000. Mantl, *et al*., 2000).

Acorn has been used as food by human for thousands of years virtually it is found everywhere (Baindrige, 1986). They are consumed in the form of bread cake and as coffee substitute (Fernald, et al., 1943).The preparation of drinks based on thermally treated acorn (dry roasting) was recommended for children. There are data in the current literatures on the antioxidant action of some acorn components (Rakia, *et al*., 2005).Beside acorn's nutritive components; it also possesses biologically active ingredients that enable the utilization of acorn in the preparation of functional food (lee, *et al*., 1992). Water extract of acorn can be used as a new natural antioxidant and functional food. The term functional food mean any food or food ingredient that might provide health benefits beyond the traditional nutrients content (Farrari, *et al*., 2003) The word-wide nutritional health products (NHPs) market, which include organic food,

nutraceutical, functional food and dietary supplements has become an extremely fast growing market. Consumer's confidents and raised attitudes towards herbal supplements indicate the presence of important growth demand (Molyneaux, 2002).

In the light of previous comments, the present investigation was undertaken to determine the phenols and flavonoids of Oak's acron and its shell and evaluate its antioxidant activity.

MATERIALS AND METHODS:

Plant materials

The samples of *Quercu srobur* acorn (Family*:Fagaceae*) used in the current investigation were collected during November 2011. The ripe nuts that had fallen to the ground, without damage were first washed and dried in an oven at 60ºC for 10 hours to prevent spoilage during storage; the same procedure was also applied to the oak's shell.

Preparation of oak extract

The extract was prepared by mixing 0.75 g of milled sample with 150 ml of distilled water, incubated at 50 ^{o}C in the water bath for 30 min. The mixture then cooled, the volume was completed to 250 ml, filtered and stored at 4ºC until use for assays.

Determination of the total phenolic compounds

The total phenolic compound contents were determined using the Folin-Ciocalteu reagent (Singleton, *et al*., 1965). To 0.1 ml of the extract (contain 100µg/ml), 1 ml of Folin-Ciocalteu reagent, and 3 ml of NaCO3 (2%) were added, mixed well and incubated in a bath for 2 hour. The total phenols were determined by spectrophotometer at 760 nm. The calibration curve was prepared with gallic acid solution ranging from 20 -100 µg/ml. The concentration of total phenolic compounds in the extract was determined as µg of gallic acid equivalent using an equation obtained from the standard gallic acid curve expressed as µg gallic acid /g dry weight of the plant material. The data were presented as the average of three analyses.

Determination of flavonoids

The total flavoniod contents of sample plants were determined according to the colorimetric method (Bushra, *et al*., 2002). Briefly, 3 ml solution of extract (contain 100 µg/ml) was mixed with 5 ml of H_2O, 0.3 ml of (5%) sodium nitrite, 0.7 ml of (10%) aluminum chloride and 1ml of 1 M sodium hydroxide , left at room temperature for 30 min. The absorbance of the reaction mixture was measured at 515 nm. Rutin was chosen as a standard. Using the standard curve (20 -100mg/ml), the level of the total flavonoid contents in sample extract was determined in triplicate, respectively. The results were expressed as µg rutin equivalents/g dried plant materials.

Determination of total alkaloids

The total alkaloids contents in the extract were measured using 1, 10-phenanthroline methods described by (Sing, *et al*. (2004). 1 ml of extract (contain 100µg/ml) was mixed with 1ml of 0.025 M $FeCl_3$ in 0.5M HCl and 1 ml of 0.05 M of 1, 10-phenanthroline in ethanol. The mixture was incubated for 30 minutes in hot water bath with maintained temperature at 70ºC. The absorbance of red colored complex was measured at 510 nm against reagent blank. Alkaloid contents estimated and it was calculated with the help of standard curve of Colchicines. The value was expressed as µg Colchicines equivalents/ g dried plant materials.

Determination of total antioxidant activity

The total antioxidant capacity of the extract was evaluated by the phosphomolybdenum method according to the procedure described by (Prieto, *et al*. (1999). The assay is based on the reduction of Mo (VI) to Mo (V) by the extract and subsequent formation of a green phosphate /Mo (V) complex at acid pH. 0.3 ml of extract (contain 100µg/ml) was mixed with 3 ml of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). In case of blank 0.3 ml of water was used in place of extract. The tube containing the reaction solution was capped and incubated in a boiling water bath at 95ºC for 90 min. After cooling to room temperature, the absorbance of the solution was measured at 695 nm using a spectrophotometer. The antioxidant capacity of extracts was expressed as equivalents of ascorbic acid using standard: [A=0.004 xs +0.275; r^2= 0.998] where A is the absorbance at 695 nm and x the concentration as ascorbic acid equivalent (µg/ml). The value was expressed as µg ascorbic acid equivalents/ g dried plant materials. The data were presented as average of three analyses

Reducing Power determination

The reducing power of the extract was determined according to the method of Oyaizu (Oyaizu, 1986). 2.5 ml of the aqueous sample (20-100 µg/ml) was mixed with phosphate buffer (2.5 ml, 0.2 M, PH 6.6) and potassium ferricyanide (2.5ml, 1%).The mixture was

incubated at 50ºC for 20 min. A portion (2.5 ml, 10%) of trichloroacetic acid was added to the mixture to stop the reaction. The extract was centrifuged at 1000 ×g for 10 min. The upper layer of solution (2.5ml) was mixed with distilled water (2.5 ml) and $FeCl_3$ (0.5 ml, 0.1%), the absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. Vitamin C was used as positive control. Phosphate buffer (PH 6.6) was used as blank solution. All analyses were run in triplicate and result averaged.

Metal chelating activity

The chelating of ferrous ions by the extract was estimated by the method of Dinis, *et al.* (1994). 1 ml of sample (20-100 µg/ml) was added to a solution of 2 mM $FeCl_2$ (0.05 ml). The reaction was initiated by the addition of 5 m Mferrozine (0.2 ml), the mixture was shaken vigorously and left standing at room temperature for 10 min. Absorbance of the solution was then measured at 562 nm. The percentage of inhibition of ferrozine –Fe^{+2} complex formations was calculated by the given formula.

Metal chelating effect (%) = $[(A_0-A_1)/A_0\times100]$

Where A_0 was the absorbance of the control and A_1 was the absorbance of the extract or standards. Na_2 EDTA was used as positive control. The control contains $FeCl_2$ and ferrozine.

Results and Discussion

1- Total phenolic, flavonoids and alkaloid contents in the extract of oak acorn and its shell.

The total phenolic compound, falvonoid and alkaloid were measured for oak acorn and its shell. The results are shown in Table (1). The total phenolic content of oak acorn and its shell were 63.16± 1.13, 36.33±0.92 µg gallic /g dry weight. The content of extractable phenolic compound in extract, determined from the regression equation of the calibration curve (y=0.006x-0.097, r^2 =0.995), Fig (1).These amount were comparable with results describe in the literature (Rakic, *et al*., 2005). The total flavonoids content of oak acorn and its shell were 145 ±1.12, 205±1.31 µg rutin equivalent g^{-1} of extract powder by reference to standard curve (y=0.001x+0.002 r^2= 0.998), Fig (2). Flavonoids are very important plant constituent because of active hydroxal groups and show antioxidant activity (Kumar, *et al*., 2008).The total alkaloid of oak acorn and it its shell were 13.44 ±0.94, 5.33±.52 µg colchicines equivalent g^{-1} of extract powder, by reference to standard curve (y=0.009x+0.185 r^2=0.998), Fig(3). It is expected that several activities might be related to possible antioxidant action from alkaloids like poly phenol compound (Einbonda, *et al*., 2004).The result indicated that acorn and its shell are a raw material rich in poly phenol, flavonoids. Phenol and poly phenolic compounds such as flavonoids are widely found in food product derived from plant sources (Cook, *et al*., 1996), they have shown that it possess significant antioxidant action on human health and fitness. The flavonoids act through scavenging or chelating process (Kessler, *et al.,* 2003).The high potential of phenolic to scavenge free radical may be due to many phenolic they posses (Sawa, *et al*., 1999).

Table 1: Total phenolic content (expressed as µg gallic acid equivalent), flavonoids content (express as µg rutin equivalent) and alkaloids (expressed as µg colchicines equivalent) from extract of oak acorn.

Extract	Total phenol content µg/g	Total flavonoids content µg/g	Total alkaloids content µg/g
Acorn	**63.16±1.03**	**145±1.23**	**13.44±0.54**
Shell	**36.33±0.93**	**205±1.39**	**5.33±0.66**

Valued are means of triplicates ±SD

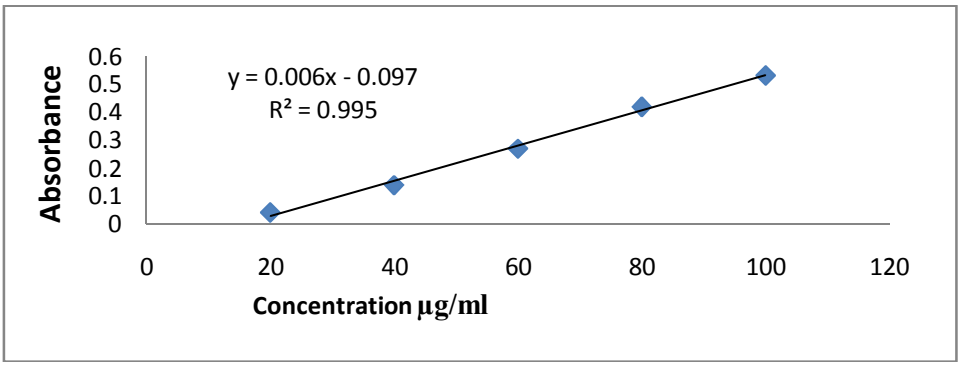

Fig (1): Calibration curve for gallic acid (µg/ml)

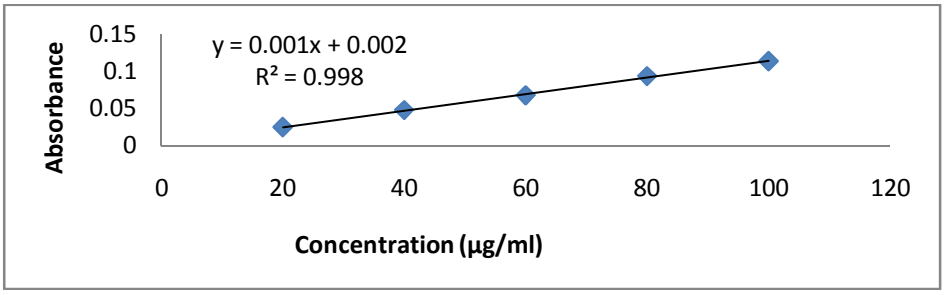

Fig (2): Calibration curve of rutin(µg/ml)

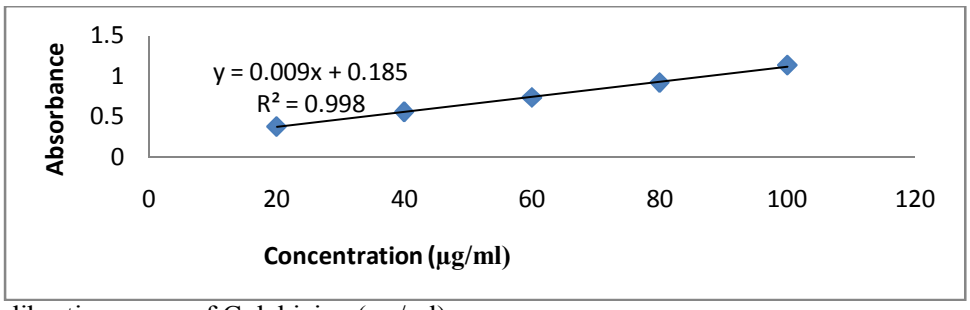

Fig (3): Calibration curve of Colchicine (µg/ml)

2- Total antioxidant activity

The phosphmolybdenum method usually detects antioxidant such as ascorbic acid, some phenolics, α-tocopherol and carotenoids (Prieto, et al., 1999).Ascorbic acid, glutathione, poly phenol and aromatic amines have the ability to donate hydrogen and electrons and that can be detected by the three assay models. The antioxidant activity of aqueous extract of oak acorn and it is shell presented in Table 2.Total antioxidant capacity of the extracts, expressed as equivalents of ascorbic acid (µg/g of dry material). The total antioxidant of oak acorn and it its shell were 44.7±1.13, 10.0 ± .69 µg ascorbic acid equivalent g^{-1} of extract powder, by reference to standard curve (y=0.004x+0.270 r^2=0.998), Fig (4).The result presented above indicated that the antioxidant activity of oak may be due to the presence of poly phenols, flavonoids that may act by donating electrons and free radicals. The antioxidant breaks the free radical chain by donating a hydrogen atom (Dorman, *et al*., 2003).

Table 2: Total antioxidant content (expressed as mg ascorbic acid equivalent), from extract of oak acorn

Extract	Total antioxident content µg/g
Acorn	**44.7±1.13**
Shell	**10±0.69**

Valued are means of triplicates ± SD

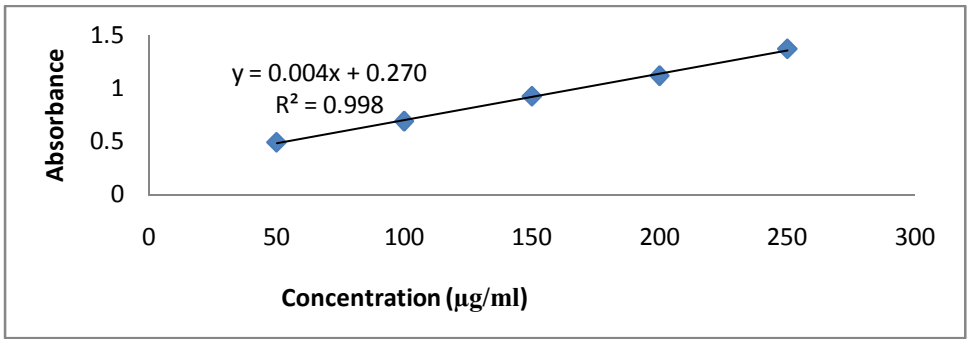

Fig (4): Calibration curve of Ascorbic acid (µg/ml)

3-Reducing Power

Another assay that is the mechanism related to electron denoting ability of the extract is reducing power assy. In this assay the presence of electron donor in the sample would result in reduction of Fe^{+3} /ferricyanide complex to Fe^{+2}. An electron-donating reducing material, deals with antioxidant activity to donate an electron to free radical from the solution. An increasing in the reactive ability. (Fig 5), shows the dose-response curves for the reduction power of the extract. It was found that reducing power of the extract also increase with increasing of its concentration. Vitamin C was used as standard. Similar relation between Fe^{+2} reducing activity and total phenol contents have been reported in the literature (Goa, *et al*., 2000).

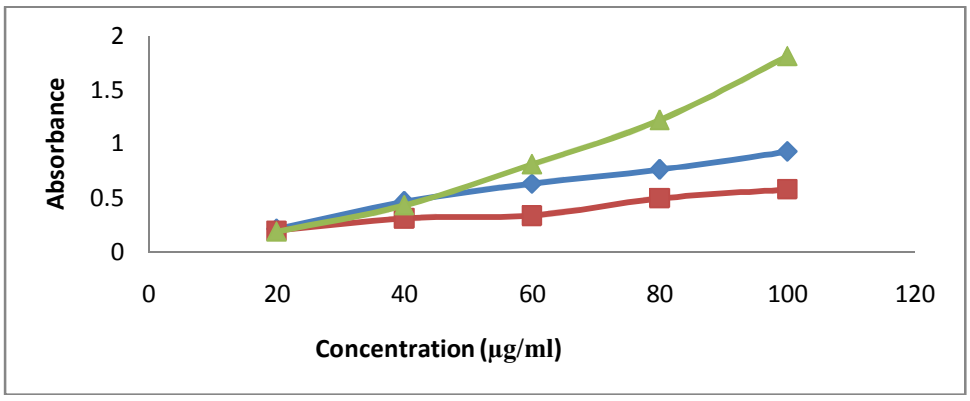

Fig (5): Reducing power of Ascorbic acid, oak acorn and oak shell

4-**Metal chelating activity**

Iron chelators mobilize tissue iron by forming soluble stable complexes that then excreted in the feces and/ or urine. It also accelerates lipid peroxidation by breaking down hydrogen and lipid peroxide forms by Fenton free radically reaction; $Fe^{+3} + H_2O_2 \rightarrow Fe^{+2} + OH^- + OH^-$ (Halliwell, *et al*., 1990). Chelating therapy reduces iron-related complication in human and thereby improves quality of life and overall survival in some disease such as Thalassemia major. (Mohammed, *et al*., 2008). The Fe^{+2} ion can form complex with ferrozine .In the presence of chelating agents the complex formation is prevented, resulting in decrease in the red color of complex. A measurement of color reduction allows for the determination of metal chelating activity. (Yamaguchi, *et al*., 2000) .In this assay, extract of oak acorn and standard antioxidants interfered with formation of a ferrous-ferrozine complex, suggesting that it has chelating activity and captures ferrous ion before ferrozine (Fig. 6) with IC50 of 60.1 µg/ml for acorn, 48.7 µg/ml for Na_2 EDTA and 39.9 µg/ml for the shell respectively. Chelating agents that form σ bonds with a metal are effective as secondary antioxidant because they reduce the reduce potential, and there by stabilize the oxidized form of the metal ion(Gordon, 1997) .This study shows that oak acorn has a marked capacity for iron binding ,suggesting the presence of poly phenol that has potent iron chelating capacity.

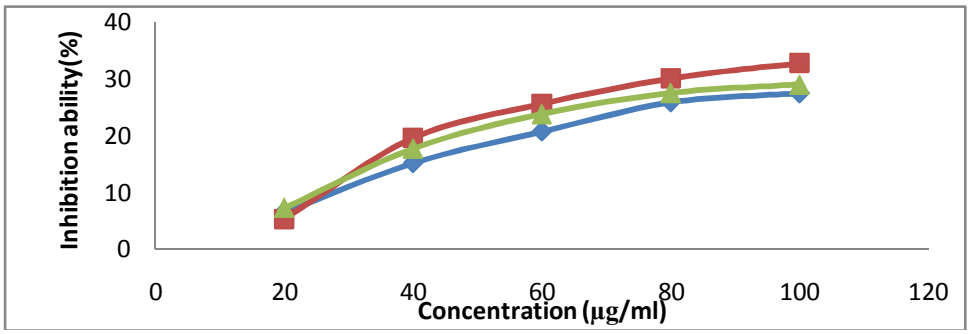

Fig (6): Ferrous ion chelating activity of acorn, shell, and Na $_2$EDTA

Conclusion

Overall, it has been verified that Oak acorn *Quercus robur* and its shell is characterized by its functional-protecting materials. The results obtained in the present study indicate that aqueous extract of oak acorn exhibit a wide variety of phenolic, antioxidant, reducing and metal chelating activity. That might be attributed to its poly phenolic capacity .The finding of the present study suggest that oak acorn, beside its nutritional component could be a source of natural antioxidant that have a great important as therapeutic agent in preventing or slowing disease. Therefore, further investigation focused on evaluating the mechanism of this activity is very promising.

REFERENCES

Baindrig, D. A., (1986). Use of Acorn food in California:Past , Presnet , Future. Presented at the Symosium on Mulitple –use management of California's hard woods. Nov 12-14.

Barlow, S.M., (1990). Toxicological aspects of antioxidants used as food additive. In food antioxidants, Hudson BJF (ed), Elsevier, London, pp 253-307.

Bushra, S., Farooq, A., and Muhammd, A., (2009).Effect of extraction solvent /technique on the antioxidant activity of selected medicinal plant extraction. *Molecules,* **14**, 2167-2180.

Chu, Y., (2000). Flavonoid content of several vegetables and their antioxidant activity. *J Sci. Food Agric*., **80**, 561-566.

Cook,N.C., and Samman, s., (1996). Flavonoid-chemistry,metabolism, cardi-oprotective effects and dietary source. *Nutr.Biochem*.**7,** 66-76.

Dinis,T.C.P., Madeira,V.M.C., and Almeida,L.M. (1994). Action of phenolic derivatives (acetaminophen, salicylate and 5-aminosalicylate) as inhibitor of membrane lipid peroxidation as peroxyl radical scavenging effects.*Chem.Pham. Bull.* **36**, 2090-2097.

Dorman, H.J.D., Kosar, M. Kahols, K. Holm, Y., and Hiltunen, R., (2003). Antioxident properties of composition of aqueous extracts from Mentha species, hybrid, varieties and cultivars.*J.Agr.Foodchem*. **51**, 4563-4569.

Einbonda,L.S.,Reynertsona,K.A., Luo,X.D., Besileb,M.J., and Kennellya,E.J., (2004).Anthocyanin antioxidant from edbilfruit.*Food Chem*.**84**,23-28

Fernald, H., and Kinsy, A., (1943). Edible wild plant of eastern north America .Cornwall-on-hudson,NY:Acedmic press.

Ferrari,C.K.B., and Torres, E.A.F.S., (2003). Biochemical pharmacology of functional foods and preventation of chronic diseases of aging .*Biomedicine &pharmacotherapy*.**57**, 271-260.

Goa, X., Brork, L., Trajkovski, V., and Uggla, M., (2000).Evalution of antioxidant activity of rosehip ethanol extract in different test system.*J.Agr.foodchem*. **80**, 2021-2027.

Gordon, M. H., (1997).Food antioxidant .In:Hudson,B.J.F., (Ed)., London, Elsevier,1.

Halliwell, B., Aeschbech. R., Loliger, J., and Aruma, Ol., (1995).The characterization of antioxidant .*Food Chem.Toxicol.,* **33**, 601-617.

Ito, N., Fukushima, S., Hasegawa, A., Shibata, M., and Ogiso, T., (1983). Carcinogenicity of butylatedhydroxalanisol in F344 rats.*J.Natl.Cancer Inst*., 70, 343-34

Kessler, M., Ubead, G.,,and Jung , L., (2003). Anti-and pro-oxidant activity of rutin and quercetin derivative.*J.Pharm.Pharmacol*., **55**, 131-142.

Kumar,P.S., Sucheta,S.,Deepa,V.S.,Selvamani,P., and Letha,S.,(2008).Antioxident activity in the in the some selected Indian medical plant.*Afr.J.Biotechnol*.,**7**(12),1826-1928

Lee, M.H., Jeang, J.H., and Man-Jinoh., (1992).Antioxident activity of gallic acid in acorn extract.*JKorSoc.Food Nutrition,* **21**(6), 639-700.

Losso, J.N., Shahidi, F., and Bagchi, D., (2007).Anti-angiogenic functional and medical foods.Boca Raton FL; Taylor&Francis.

Mantle, D., Eddeb, F., and Pickering, A. T., (2000).Comparsion of relative antioxidant activities of British medicinal plant species in vitro *J.Ethnopharmacol*., **72**, 47-51.

Molyneaux, M., (2002). Consumer attitudes predict upward trends for the herbal market place. *HerbalGram*., **54**, 64-65.

Mohammad, A.E., Fereshtch, P., and Ahmad, R.B., (2008). Iron chelating activity, phenol and flavonoid content of some medical plants from Iran.*Afr.J.Biotechnol*., **7**(18), 3188-3192.

Oyaizu, M., (1986). Studies on products of browingreaction:antioxidant activity of product of browing reaction prepared from glucosamine .*J.Natr*.**44**, 307-315.

Prieto.P., Pineda, M., and Aguilar, M., (1999) .Spectrophotometer quantitation antioxidant capacity through the formation of a phosphomolybdenum complex: application to the determination of vitamins E, *Anal.Biochem*.**, 269,** 337-341**.**

Rakic, s., Povrenovic, D., Maletic, R., and Zivkoviae, M., (2005).Drying of the aqueous extract of acorn *Quercusrobur* in a Spout- fluid Bed.*J .of Agric.Sci.,* **50**(2), 173-182.

Rice-Evans, C.A., Miller, N.G., and Paganga, G., (1996).Structure-antioxidant activity relationship of flavonoids and phenolic acid, *free radical Biol.med*.,**20**,933-956.

Rice-Evans, C.A., Miller, N.G., and Paganga, G., (1997).Antioxidents properties of phenolic compounds,*Trends. plant sci.,***2**, 152-159.

Sawa, T., Nako, M., Akaike, T. Ono, k., and Maeda, H., (1999).Alkylperoxyl radical scavenging activity of various flavonoids and other phenolic compounds: implimantation for the antitumor promoter effect of vegetables.*J.Agric.food.chem*., **47**, 397-492.

Scalbert, A., and Williamson, G., (2000). Dietary intake and bioavailability of poly phenol*, J. Nutr*.,**130**, 2073S-2085S.

Singh, D.K., Srivastva,B., and Sahu, A., (2004). Spectrophotometer, determination of Rauwolfiaalkaloids,estimation of reserpine in pharmaceuticals. *Analytical Sc,i*. **20,** 571-573.

Singleton, V.L., and Rossi, J.A., (1995).Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents.*Am.J.Enol.Viticult*.,**16**, 144-158.

Yamaguchi, F., Ariga, T., Yoshimira, Y., and Nakazawa H., (2000).Antioxident and antilycationofcarcinol from Garciniaindica fruit rind.*J.Agr.Foodchem*.,, 180-185.

دەست نیشانکردنی چالاکی دژەئۆکسان و دیاریکردنی بڕی ئاوێتە فینۆڵی و فلاڤینۆیدیەکانی بەڕوو - هەولێر/کوردستان

لەم لێکۆڵینەوەیەدا، چالاکی دژەئۆکسان و تێکڕای بڕی ئاوێتە فینۆلییەکان لە دەرهێنراوی ئاوی بەڕوو دیاریکرا لە تاقیگەدا بە بەکارهێنانی مۆدێلی دژەئۆکسانەکان، وەك: تێکڕای توانای دژەئۆکسان و توانای لێکردنەوە و توانای گرتنی کانزاکان. هەروەها تێکڕای بڕی ئاوێتە فینۆڵی و فلاڤینۆیدیەکان و تفتەکان دیاریکرا لە دوو بەشی بەڕوودا : تۆوەکەی و بەرگەکەی. ئەنجامەکان ئەوەیان دەرخست کە بڕی ئاوێتە فینۆڵی و فلاڤینۆیدیەکان و تفتەکان لە کاکڵی تۆوەکەی زیاترە وەك لە بەرگەکەی. هەروەها بەگوێرەی ڕێگای فۆسفۆمۆڵبیدیۆم ، چالاکی دژەئۆکسانی دەرهێنراوە ئاوییەکەی کاکڵ و بەرگەکەی دیاریکرا، وە بە بەکارهێنانی پێوەری شەبەنگی ڕووناکیش توانای لێکردنەوە و توانای گرتنی کانزاکانیش دیاریکرا. پاشان ئەنجامەکان بەراوردکران بە ڕێژەی پێوانەیی هەندێ لە ئاوێتەکانی وەك ترشی ئەسکۆربیك و ڕووتین و ترشی گالیك و کۆلچین.

هەموو ئەنجامەکان ئەوەیان دەرخست کە بەڕوو دەتوانرێ بە سەرچاوەیەکی باشی ئاوێتە فرە-فینۆڵە چالاکەکان دابنرێت لە سیستەمی خۆراکی مرۆڤدا ، وە دەتوانرێ پاڵپشتێکی خۆراکی باش بێت بۆ مرۆڤ چونکە ئاوێتە فینۆڵی و فلاڤینۆیدیەکانی بە خەستی یەکی بەرز تێدایە و هەروەها ئەو توانا بەهێزەش کە هەیەتی وەك دژە ئۆکسانێك..

نشاط مضادات الأكسدة وتقديرالفينولات والفلافونويدمنسنديات البلوط في كردستان/اربيل

الخلاصة

في هذه الدراسة ، تم التحقيق من نشاط مضادات الأكسدة و المحتوى الكلي الفينولات من المستخلص المائي للبلوط (*quercus robur*)والتى تنتمي إلى عائلة (fagaceae) باستخدام نماذج في المختبر ، بما في ذلك مضادات الأكسدة إجمالي القدرة ، القوى الاختزالة ونشاط المعدنية المخلبية . قدرت إجمالي محتويات الفينول ، الفلافونويد و قلويد كميا في جزئين من البلوط (الجوزة و قوقعته). وأظهرت النتيجة أن مجموع الفينولية ، الفلافونويد ، و محتويات قلويد كانت أعلى من تلك التي في الجوزة الى قوقعته . تم تحديد النشاط المضادة للأكسدة من المستخلص المائي من الجوزة و غلافه وفقا لطريقة phosphomolybdenum، القوى الاختزالة ، و المعادن مخلبية تحدد باستخدام مقياس الطيف الضوئي ، نتائجها قورنت مع المركبات القياسية مثل حمض الاسكوربيك ، وروتين ، حمض الغال و الكولشيسين . وأظهرت النتيجة الإجمالية أن البلوط يمكن أن تكون بمثابة مصادر جيدة للفينولات الكلية النشطة بيولوجيا في النظام الغذائي للإنسان و يمكن اعتباره مرشحا جيدا لصياغة المكملات الغذائية بسبب تركيزها العالى من المركبات الفينولية ، الفلافونويد ، فضلا عن نشاطها المضادة للأكسدة قوية.