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Research Article

ASSAY PLANT AS NATURAL ANTIOXIDANT OF *Capparis spinosa* ROOT BARK

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Abstract

Root bark of *Capparis spinosa* L. (CS) was used in this study, CS is one of the most aromatic plants in wild in the dry region around the west, central Asia and the Mediterranean basin. It contains a wide range of phytochemical constituents. The present study detection active compounds in root bark, flanonoids, glycosides, tannins and alkaloids, all these gave positive test. As well as determination of total phenols in aqueous and ethanolic extracts (RBAA),(RBEA),8.712 and s5.781%,also flavonoids 8.154 and 5.385% respectively. Evaluated antioxidant activity of RBAE and RBEE in vitro by two applications (1) reducing power of RBAE that, 74.289% more than of RBEE, 67.331% according to reference BHT,93% at 10 mg/ml. While RBAE and RBEE were exhibited less chelating ability (2 applications) less than reducing power for two extracts. RBAE showed higher chelating ability 51.504% compared to RBEE 35.210% at 10 mg/ml concentration depending on EDTA as reference.

Keywords: *Capparis spinosa*,antioxidant activity ,active compounds,phenolic constituent

Introduction

The use of medicinal herbs is getting more popular day by day with gradual increase in the percentage of the people using herbal medicines, it is also worth mentioning that the use of wild edible herbs culinary plants is increasing in the global food market for diverse item ranging from salad to desserts (1).*Capparis spp.* is one such plant established to have highly diverse economic and medicinal value in different system of medicines (2). *Capparis spp.* are growing in a broad range of climatic conditions, such as dry desert to cooler terrains either as shrub, trees or creeper (3). *Capparis spinosa* is one of the most aromatic plants in wild in the dry regions around the west central Asia and the Mediterranean basin, and reports to contain a wide range of phytochemical constituents (4). Capparis, Fam: Capparidaceae. Arabic name, Kabar, a large genus,convering about 250 species, two species of capparis (*C.cartilaginea and C.spinosa*) have been reported from Iraq. *C.spinosa* English name (Caper Bush).It is found practically everywhere in Iraq, especially in the deserts, in waste lands, in fields and ascemels in the hills where it grows on the sloping rocks revices. The bark is bitter, aperient, diuretic, tonic besides another uses. The fruits is eaten by many people (5,6). *Capparis spinosa* (CS) has many active constituents such as flavonols like: kaempferol and quercetin (7), also alkaloids, lipids and glucosinolates (8,9).Various biochemical compounds present in *Capparis sp*. Might be medicinally important and/or nutritionally valuable(10,11).The bark of plant root is also used in liver, infusion of stem and root bark is used for diarrhea and febrifuge. Also the flower buds and roots are used as renal disinfectants, diuretic, tonic and for arterioselerosis and as compresses for the eyes(12). Different parts of *Capparis* spp have been reported to cure asthma, cough, gout ear infection, rheumatism and ulcer(13).The plant is environmentally cheap and without side effects(14,15).Raw flower buds contain a number of antioxidant phytochemicals such as flavonoids and other polyphenols(16,17,18).The main constituents of CS have been demonstrated to be flavonoids alkaloids,lipids and glucosinolates(8,9).Most sources of natural antioxidants originate from plant materials(19).The present study was under taken to evaluate(in vitro) the antioxidant activity

of aqueous and ethanolic extract of root bark for *C.spinosa* which has flavonoids and glycosides.

Materials and Methods

Plant collection

Capparis spinosa was collected from Abu-Graib/Iraq in August 2010.The whole plant was deposited to be identified, the identification done by state board for seed testing and certification- Department of plants - International Iraqi herbarium.The collected roots were washed and the bark was peeled off and air-dried at shade at room temperature. Then grounded into powder(Harbone,1984 --1cap).Powdered parts were kept in plastic tubes in deep freeze(-20^{o}c)until the time of use.

Chemical quality detection

The methods application according to (21);(22);(23).To determination PH and testing tannin, glycosides, alkaloid and flavonoids.

Extraction

Hot water extraction of CS grounded was done according to the method described (24).Briefly fifty grams of CP materials were cut into small pieces, melded and placed in a flask(2L) with 1000ml of distilled water and boiled for 15min,the mixture was filtered twice, first through cheese-cloth and then through filter paper(what man No.1). The obtained CP extract was preserved in sterile dark bottles in a cool environment (4^{o}c) until further use. Ethanolic extraction: fine powder extracted by maceration method two times with 80% ethanol(25).After filtration of total extract, the solvent was removed under reduced pressure .

Determination of total phenolic compounds

A Folin-ciocalteu's colorimetric method was used as described by Ayoola et al. (2008). To a 0.5 ml of (1 mg/ml)extract a 2.5 ml of a ten-fold diluted Folin-ciocalteu's reagent and 2ml of 7.5%.sodium carbonate solution were addedbefore the reaction allowed standing for 30 min at room temperature. The absorbance was recorded at 760 nm byusing UV/VIS Spectroscan 80 D spectrophotometer. The total phenolic compounds were determined according to gallic acid standard curve (0.01 to 1 mg/ml).

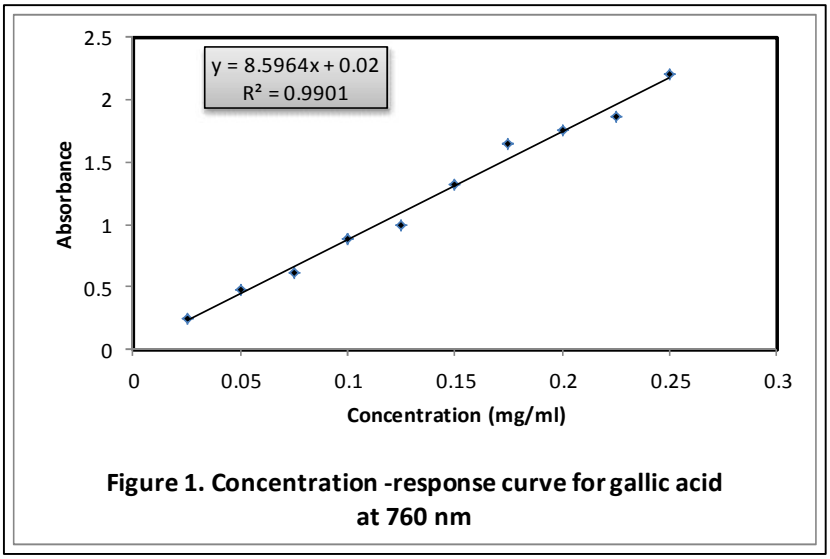

Figure 1. Concentration -response curve for gallic acid at 760 nm

Determination of total flavonoids compounds

The total flavonoids in aqueous and ethanolic extracts were determined according to Rao et al, (2012). 1 ml extract solution (1mg/ml) was placed in 10 ml volumetric flask. 5 ml of distilled water and 0.3 ml of 5% NaNo2 solution were added. After 5 min 0.6 ml of 10% AlCl3 was added. 2 ml of 1M NaOH solution was added after another 5 min, and the volume was made up to 10 ml with distilled water. The mixture was mixed thoroughly and the absorbance was measured at 510 nm. The total flavonoids were expressed as µg catechin equivalents per gram dry matter according to catechin.

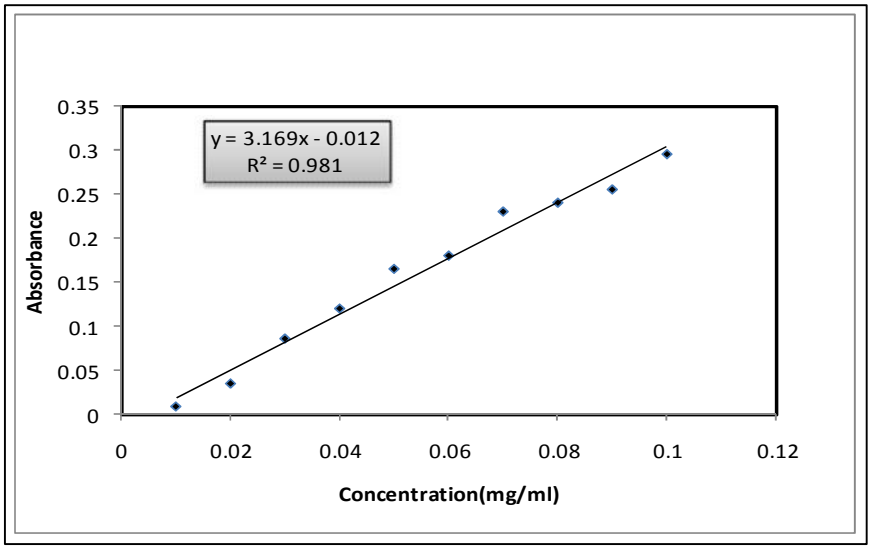

Figure 2. Concentration-response curve for catechin at 510 nm

The assay of antioxidant activity:

The reducing power: The reducing power was estimated as described by (28) .1ml extract of (2-10mg/ml) was mixed with 2.5ml of 1% potassium ferric cyanide and 2.5ml of 0.2M(pH,6.6) 0f sodium phosphate buffer and incubated at 50^{o}c for 20 min. To stop the reaction,2.5ml of 1% trichloroaceticacide(TCA) was added to the mixture and centrifuge for 10 min at 3000 rpm.0.5ml of the supernatant was mixed with 1ml of 1% ferric chloride and stand for 10min.The absorbance was measured at 700nm.0.02% of BHT used as reference.

The chelating ability: Chelating ability was determined according to (29) with some modification. 1ml of (2-10mg/ml) extract was mixed with 0.2ml ferric chloride of 2mM and 0.2ml 8-Hydroxyquinoline (5mM). After 10min at room temperature, the absorbance was determined at 562nm.The EDTA-Na2 was used as reference.

Results and Discussion

Capparis spinosa is one of the most important species a among the medicinal plants of the kingdom and possess high pharmaceutical economic and ecological values(30).The root bark of CS was studied for their flavonoids, alkaloids, glycosides and tannins all tests for these compounds were positive(Table 1).

Table (1) Chemical quality detection of *Capparis spinosa* root bark active compounds

compound	detection	Detected indicator	Result of detection
Flavonoids	Ethyl alcohol 95% KOH	Yellow ring	+
Glycosides	Benedict indicator Fehling indicator A,B	Red color Red residue	+
Tannins	A-lead acetate 1% B-Ferric chloride 1%	White gel residue Blue greenish color	+
Alkaloides	A-Meyer's test B-Picric acid	White residue Yellow color	+

The main constituents of CS have been demonstrated to be flavonoids, alkaloids, lipids and glucosinolates(8,9),that aggrement with results in table(1).Previous studies have shown the presence of indole,aliphatic glucosinolates, polyphenols, flavonoids and alkaloids in CS (31,32).Water and ethanol were selected as the extraction solvents since both are commonly used in the food industry in a variety of ways and are more stable in the human body than any other solvents, also the cost of both low. Root barka aqueous extract of CS had the highest of total phenolic and flavonoids contents 8.154, 7.812% respectively compared to those of ethanolic extract 5.781, 5.385% respectively (Table 2).

Table (2). Phenolic contents *Capparis spinosa* extracts

Extract	plant	Phenolic contents %	flavonoide contents %
Aqueous	*Capparis spinosa*	8.154	7.812
Ethanolic		5.781	5.385

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Colorimetric assay utilize reagents that react with phenols to form colored products that are readily quantifiable by absorption measurements, these methods are of great utility for screening of plant materials for phenols and as a way to measure gross phenolic content (33).The results in the present study reveals that the content of total phenols and flavonoids in varied according to the type of solvents those used in extraction method and method of extraction. The results agrees with that obtained by (34).The high percentages of the total phenolic and flavonoids in aqueous extract mean that,the water as extracting solvent and according to the chemical composition of phenolic compounds are more effective than ethanol. Fig (3)which are refer to the reducing power method for the determination of the antioxidative abilities of aqueous(RBAE) and ethanolic extract(RBEE) of CS bark root as compared with BHT as a references. At concentration 10mg/ml BHT, RBAE and RBEE extracts showed reducing activity 93%,74.289% and 67.331% respectively (Fig 3).

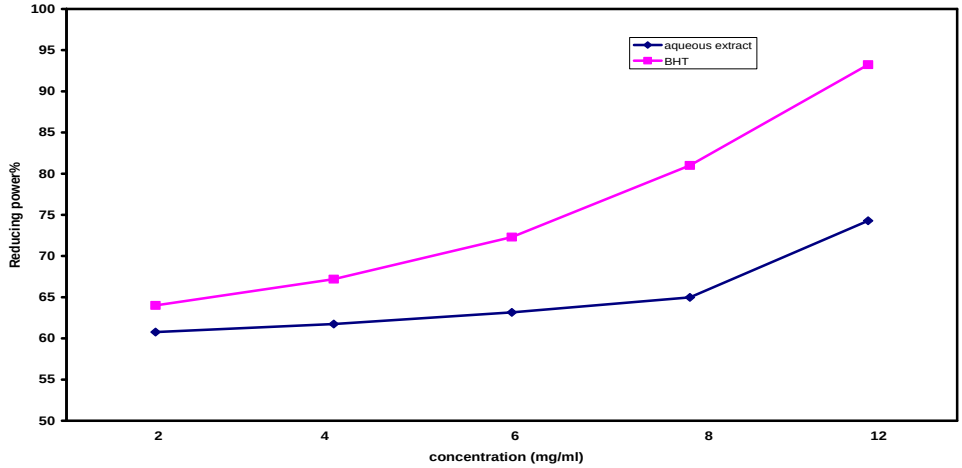

Figure 3. Reducing power of aqueous extract of *Capparis spinosa*

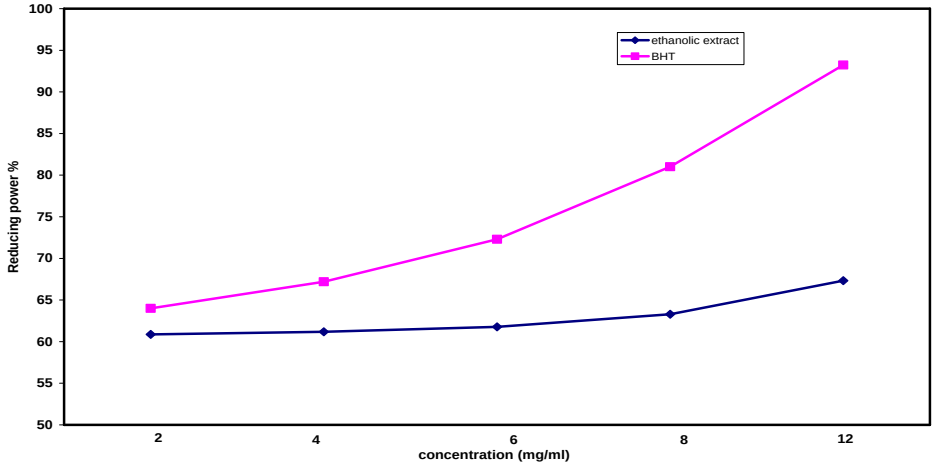

Figure 4. Reducing power of ethanolic extract of *Capparis spinosa*

The reducing power might be due to hydrogen donating ability. Reducing power indicates compounds that are electron donors, which can act as primary and secondary antioxidants(35).The reducing properties are generally associated with the presence of reductones, which have been shown to exert antioxidant action by breaking the free radical chain through the donation of a hydrogen atom(36).Antioxidant properties of methanolic extracts of raw floral buds of CS have been shown in various in vitro model(37,38).On other hand, the antioxidant potential of the edible caper is less known(39).It is a worthy mentioning that the aqueous extract of CS bark root exhibited a notable antioxidant activity as reducing power. The chelating ability was enhanced by increasing concentration of samples. The chelating activity of RBAE was 11.348% and 51.504% at 2mg/ml and 10mg/ml. (Fig 5), where RBEE had 9.027 and 35.210% at the same concentrations (Fig 6).

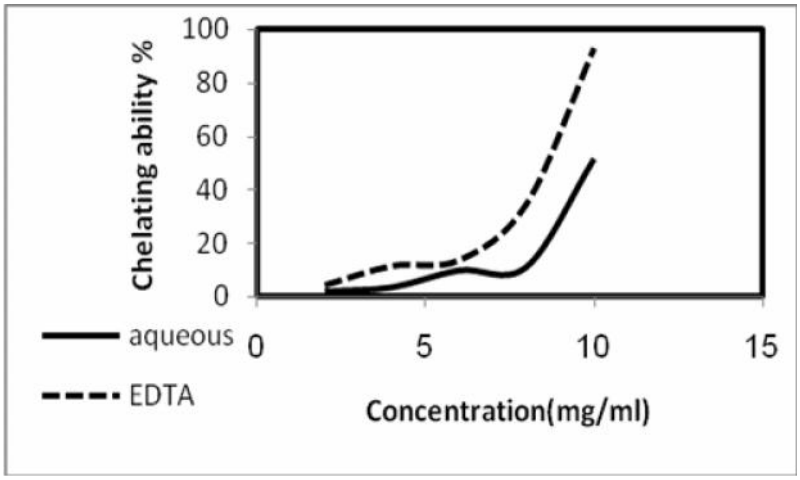

Figure 5: Chelating ability of aqueous extract of *Capparis spinosa* as compared with EDTA at the same concentration

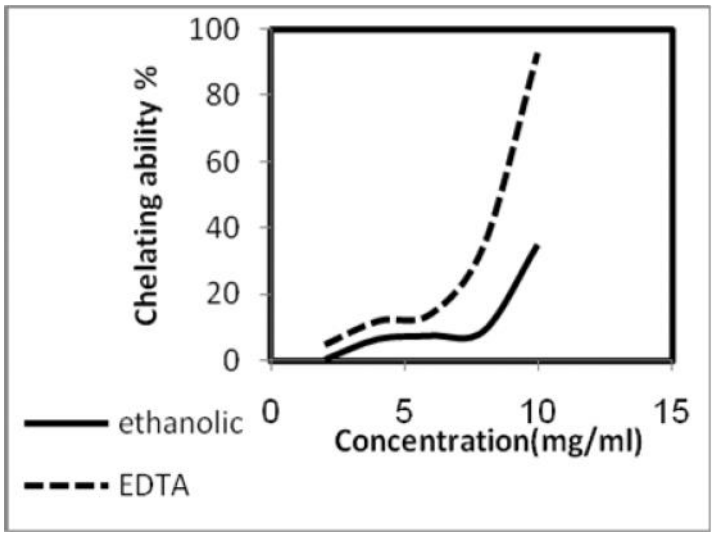

Figure 6: Chelating ability of ethanolic extract of *Capparis spinosa* as compared with EDTA at the same concentration

The same mode of action is also associated with the second method of determination the antioxidant activity(chelating ability).As shown in figs(5) and 6,the abilities of RBAE and RBEE as chelating agents(comparing with EDTA as reference) are less than their as reducing power, also RBAE had chelating ability more than RBEE but less of reference. Therefore, the usage of CS extracts in foods, which may act as natural antioxidant preservatives, may prolong the shelf-life of relevant food products, as well as influence on health of consumers.

Conclusion

Medicinal plants are the nature gift to human being to preserve our health and food. Capparidaceae family comprises various important. *Capparis spinosa* was found to have food value, potential culinary value and possessed cosmetic ingredients. It is worthy to conclude, that CS provide for raw materials for pharmaceutical aromatic and food industries. CS can be used it to elongation shelf life of food as preservative agent through antioxidant, antimicrobial activity. CS has many active constituents such as glucosinolates, flavonoids which are well known about bioactivities. It is worth to utilize from main constituents from all parts of CS, in additional to nutritive value. Also this plant is environmentally available without side effects. May be CS is a promise plant to remedy from epidemic influence.

References

1. Hassan, S and Mohammad, N.A.(2010) Ethanobatanical and pharmaceutical evaluation of *Capparisspinosa* L, validity of total folk and Unani system of medicine. Jounal of Medicinal Plants Research,4(17):1751-1756.
2. Azaizeh, H; Fulder, S; Khalil,K and Said,O.(2003). Ethromedicinal knowledge of local Arab practitioners in the Middle East Region.Fitoterapia,74:98-108.
3. Mishra, S.N; Tomar, P.C and Lakra,N. (2007).Medicinal and food value of capparis----a harsh terrain plant. Indian Journal of Traditional Knowledge,6(1):230-238.
4. Assad, A. B; khesar, H. Kh and Sa'ad,S. M.B.(2012).Cytotoxic and cytogenetics effects of aqueous, methanolic and secondary metabolites extracts of *Capparis spinosa* on tumor cell lines *in vitro.* Jordan.J of Biological Sciences,5(1):15-30.
5. Chakravarty, H.L.(1976).Plant wealth of IRAQ.A: Dictionary of economic plants.vol.one. Ministry of Agriculture & Agrarian Reform, Baghdad, Iraq.
6. Sahira, Abd-Rahman; Ikhlas, H. Alwan.(2012). Atlas of Iraqi medicinal plants: part one.Ministry of Agriculture/state board for seed testing and certification- Department of plants -International Iraqi herbarium.
7. Afsharypuor, S; Jeiran,K and Arefian,A. (1998).First inestigation of the flavor profiles of the leaf, ripe fruit and root of *Capparis spinosa* var.*mucronifolaIran*, J. Pharmaceutica Acta Helvetiae, 72:307-308.
8. Calis, I.K; Lorenzetto, P.A and Ruedi, P.(2002).6S-Hydroxy-3-oxo-alpha-ionol glucosides from *Capparis spinosa.*Phytochemistry,59(4):451-457.
9. Brevard,H; Brombilla,M; Chaintreau, A and Marison, J.P.(1992). Occurrence of elemental sulphur in caper (*Capparis spinosa* L.) and first investigation of the flavor profile. Flavour and Fragrane Journal,7(6):313-321.
10. Juneja TR; Gaind, KN and Gain, PC.(1970). Preparation and study of hypnotic activity and acute toxicity of stachychlor,Indian.J.Pharm,32:4-5.
11. Mitchell, JC.(1974).Contact dermatitis from plants of the caper family, Capparidaceae. Effect on the skin of some plants which yield isothiocyanate,Br.J.Dermatol,91:13-20.
12. Batanouny, KH (with contribution of:Aboutabl, E;Shabana, M;Soliman,F.). (1999).Egypt, International union for conservation (IUCN): Switzerland: Academy of Scientific Research and Technology, wild medicinal plants in Egypt;pp:130-131.
13. Kirtikar, KR and Basu, BD.(1933).Indian medicinal plants.vol,1,(Lalit Mohan Publication Allahabad), 197-198.
14. Montbriand, MI. (1997).Empowerment of seniors through improved communication about medication In: Proceedings of the sixth science in health. Social services for the Elderly and the Disabled, Heumann, L.F. (ed).University of II1inasis of Urbana-Champaign,Chicago.11.,258-264.
15. Montbriand, MI.(2000).Senior and health-professionals mismatched perceptions and communication about prescription and non-prescription medication.Cancer.J.Aging,19:35-56.
16. Rodrigo ,M; Lazaro, M.J; Alvarruiz, A and Giver,V.(1992).Composition of capers (*Capparis spinosa*): influence of cultivar, size and harvest date. Journal of Food Science,57:1152-1154.
17. Sharaf, M; EL-Ansari, M.A and Saleh,N.A.M. (2000). Quercetintriglycoside from Capparis spinosa.Fitoterapia,71:46-49.
18. Germano,M.P;Pasquale,R;Angelo,V;Catania,S;Silvari,V;Costa,C.(2002).Evaluation of extracts and isolated fraction from CapparisspinosaL. As an antioxidant source.Journal of Agriculture and Food Chemistry,27:1168-1171.
19. Panda,H. (2004).Hand book on medicinal herbs with uses.) Asia Business, India),pp:258-259.
20. Harbone, J.B. (1984). Phytochemical methods. 2nd ed.Chapman and Hall, London.
21. Harbone, J.B. (1973). Phytochemical methods. Champman and Hall,London, New York.
22. Brain, K.R and Turner, T.D.(1975).The practical evaluation of phytopharma ceuticals.Wright-Scientica,Bristol,57-58.
23. Sofowora, A.(1993).Medicinal plants and traditional medicine in Africa. Spectrum book limited,Ibadan,Nigeria.,151-153.

24. Abdel-Salam, A.M; Ammar, A. S and Galal.W.K. (2009).Evaluation and properties of formulated low calories functional yoghurt cake. J.Food. Agric.Environ.,7(2):218-221.
25. Markham ,K R.(1982). Techniques of flavonoid identification. Academic Press, London 16.
26. Ayoola ,G.A; Ipav, S.S; Sofidiya,M.O;Adepoju-Bello, A.A; Coker, H.A and Odugbemi, T.O.(2008). Phytochemical screening and free radical scavenging activities of the fruits and leaves of *Allanblackia floribunda* Oliv (Guttiferae) .International Journal of Health Research,1(2):87-93.
27. Rao, K.S; Keshar, N.K and Ravi,K.B.V.V. (2012).Microwave assisted extraction and evaluation of in vitro antioxidant activity of *Cinnamomum aromaticum*. J.Medicinal Plants Research,6(3):439-448.
28. Chou, HJ; Kuo, JT; Lin, ES. (2009). Comparative antioxidant properties of water extracts from different parts of beefsteak plant (*Perilla frutescens*). J. Food Drug Anal., 17: 489-496.
29. Su, M.S.; Shyu, Y.T. and Chein, P.J. (2008).Antioxidant activities of citrus herbal product extracts, Food Chem., 111:892-896.
30. Hassan,S and Mohammed, N.ALyemeni.(2010). Ethnobotanical and pharmaceutical evaluation of *Capparis spinosa* L, validity of local folk and Unani system of medicine. J. Medicinal Plant Research,4(17):1751-1756.
31. Ahmed,Z.F; Rizk,A. M; Hammouda, F.M and Seif EL-Nasr, M. M .(1972).Phytochemistry,11,251.In: 1H-Indole-3 acetonitrile glycosides from *Capparis spinosa* fruits .Ihsan, C; Ayse, K and Peter, R. Phytochemistry,50(1999) 1205-1208.
32. AL-Said, M.S; Abdelsattar,E.A; Khalifa, S.I and EL-Feraly, F.S.(1988).Pharmazie,43,60. In: 1H-sIndole-3 acetonitrile glycosides from *Capparis spinosa* fruits .Ihsan, C; Ayse,K and Peter, R. Phytochemistry,50(1999) 1205-1208.
33. Hagerman, A.(2002).Tanninchemistry. http:/www. users.muohio.edu/hagermae/tannin.pdf(Accessed November).
34. Henning, SM; Fajardo,C;Lee, HW;Youssefian, AA;Go VLW and Herber,D.(2003).Catechine content of 18 teas and a green tea extract supplement correlates with the oxidant capacity.Nutr.Cancer,45:226-235.
35. Yen,GC;Chen, HY.(1995).Antioxidant activity of various tea extracts in relation to their antimutagenicity. J. Agric. Food Chem.,43:27-32.
36. Shimada,K; Fujikawa,K; Yahara,K; Nakamura, T.(1992). Antioxidative properties of xanthan on the autoxidation of soybean oil in cyclodextrin emulsion.J. Agric. Food Chem.,40:945-948.
37. Bonina,F;Puglia,C;Ventura,D;Aquino,R;Tortora,S; Sacchi,A;Saija,A;Tomanio,A;Pellegrino,M.L and DE Carparis, P.(2002).In vitro antioxidant and in vivo photoprotective effects of a lyophilized extract of *Capparis spinosa* L. buds. Journal of Cosmetic Science,53:321-335.
38. Germano,M.P; DEPasquale, R;D'Angelo, V;Catania, S;Silvari,V; and Costa, C.(2002).Evaluation of extracts and isolated fraction from *Capparis spionsa* L. buds as an antioxidant source. Journal of Agriculture and Food Chemistry,27:1168-1171.
39. Aslantürk,Ö S and Tülay,A C.(2009). Genotoxic and antimutagenic effects of *Capparis spinosa* L. on the *Allium cepa* L. root tip meristem cells.Caryologia,62(2):114-123.

