INTERNATIONAL RESEARCH JOURNAL OF PHARMACY
www.irjponline.com ISSN 2230 – 8407
Research Article

IN VITRO FREE RADICAL SCAVENGING ACTIVITY OF *AMORPHOPHALLUS COMMUTATUS* AN ENDEMIC AROID OF WESTERN GHATS, SOUTH INDIA

R. Kavitha Krishna[1]*, S. Krishna Kumari[2], M. Vinodha[1], Anu Sebastin[1], R. Gomathi[1]
[1]Department of Biotechnology, Dr.Mahalingam Centre for Research and Development, Nalla Muthu Gounder Mahalingam College, Pollachi – 642001, Tamilnadu, India
[2]Department of Biochemistry, Kongunadu arts and science college, Coimbatore, Tamilnadu, India

Article Received on: 03/12/11 Revised on: 18/01/12 Approved for publication: 10/02/12

*Email: r.kavithakrishna@gmail.com

ABSTRACT
The aim of this study is to evaluate *In vitro,* antioxidant potential of five different solvent fractions of *Amorphophallus commutatus* tuber using multiple assays. The synthetic radicals $DPPH^{o}$ and $ABTS^{o+}$ were scavenged effectively by the ethyl acetate fraction with IC_{50} value of 61.32±1.39µg and 69.6±1.1 µg respectively. Significant ($p<0.05$) radical scavenging was observed in the ethyl acetate fraction against radicals like super oxide, Nitric oxide, and Hydroxy with the IC_{50} values of 92.5±1.1 µg, 139.6±2.2 µg, 66.3±0.9 µg and 62.4±1.1 µg respectively. $ABTS^{o+}$ scavenging of ethyl acetate was significant than BHT and other radicals were also scavenged significantly by ethyl acetate fraction. The results assure the presence of antioxidant principles in various fractions and among the fractions ethyl acetate showed significant activity.
Keywords: *Amorphophallus*, Synthetic radicals, Radical scavenging, Free Radicals, Super oxide, Nitric oxide.

INTRODUCTION

Free radicals, Reactive oxygen species (ROS) and Reactive Nitrogen species (RNS) play a vital role in various biological processes especially in host defense mechanisms. But over production of these contribute to oxidative stress related pathological phenomenon. The excess free radicals are removed by various reactive oxygen species scavenging enzymes and antioxidant chemical species[1] . The involvement of active oxygen and free radicals in the pathogenesis of human disease such as cancer, aging, artherosclerosis, malaria, rheumatoid arthritis, neurogenerative disorders[2] has been recently understood more and more. The mechanisms of action of antioxidants include complexation of redox-catalytic metal ions, Scavenging of free radicals and decomposition of peroxides. The antioxidant extracts have been found to involve lot of mechanism and mechanistic synergism. Therefore a multiple experimental approaches are recommended for identification of complete putative chain- breaking capacity[3].

Growing knowledge about the health promoting impact of antioxidants and its additives in everyday food, combined with the hazardous effect of synthetic additive has led to increased interest in identification of natural antioxidants[4] like flavonoids, anthocyanins, carotenoids, dietary glutathionine, vitamins and endogenous metabolites.

It is therefore evident about the need for developing functional food from phytochemicals of plant origin as they are viewed to be promising as therapeutic agent , nutraceutical and food preservative to improve the status of human health and disease prevention[5].

Plants of the genus *Amorphophallus* have a long history of use in tropical and subtropical Asia as a food source and as a traditional Chinese medicine (TCM). *Amorphophallus* species, have been used as food, medicine, fodder and for wine production[6]. *Amorphophallus companulatus* has been reported to be used as antimycobacterial agent in aurveda[7]. Khan , Rhaman & Islam (2008) has reported the presence of antibacterial, antiproliferative and antifungal properties of *Amorphophallus companulatus*[8].

Western ghats in south India is one of the Biodiversity hotspots of the world. *Amorphophallus commutatus* (Schott) Engl, a member of the Araceae, is a tuber depressed globose, is a rare cormous herb that is found in evergreen and semi-evergreen forest of south western India (endemic to Western Ghats). Tuberous corms of *A.commutatus* were used for treatment of piles, tumours and cysts[9]. Tubers of *Amorphophallus commutatus* has also been used as antidote for snake bite by tribal's living in fifty villages of Sitamata wildlife, sanctuary, Rajasthan, India[10].

There are no reports on the bioactivity of the plant *Amorphophallus commutatus*, an endemic aroid of Westen ghats but the cormous herb has been reported to be used by the tribes for various ailments. Hence, this work is done with the aim of validating the antioxidant and free radical scavenging properties of different extracts of the herb. As the presence of antioxidant activity is one of the primary property for identification of other bioactivities in the plant.

MATERIALS AND METHODS

Plant sample

The whole plants of *Amorphophallus commutatus* wsa collected from All India Coordinated Research Project on Medicinal and Aromatic plants, College of Horticulture, Kerala Agricultural University, Vellanikkara, Thrissur, Kerala.

Extraction

Organic solvents in the increasing order of polarity (Petroleum ether, chloroform, Ethyl acetate, methanol) and aqueous extract (hot water) of the plant materials were prepared according to the method described by (Harbone,1998)[11].The samples were sequentially extracted using a soxhelet apparatus.

Preparation of sample

Each of the five extracts were evaporated to dryness after extraction under reduced pressure using rotary evaporator. The concentrated extracts were lyophilized and were solubilised with DMSO. The hot water extract was solubilised using sterile distilled water. All the five extracts were diluted and concentration ranging between 25µg to 100

µg was used for all the assays. Standard BHT in the same concentration was used as positive control.

***IN VITRO* RADICAL SCAVENGING ASSAYS**

Scavenging activity on synthetic radicals

DPPH radical scavenging activity[12]

The free radical scavenging activity of different solvent fractions of *Amorphophallus commutatus* was measured according to the method of Mensor *et al*., 2001 using DPPH as free radical. The reaction mixture contained 0.1 ml of different concentration of solvent fractions with 1.9 ml of 0.1 mM DPPH in methanol. The control was devoid of the sample. BHT was used as a positive control. The reaction mixture was shaken well and read at 517 nm after incubation at 25° C for 30 min. The percentage DPPH radical scavenging activity was calculated from the following equation.

DPPH Scavenging activity (%) = $1 - \frac{A_s}{A_c} x100$

Where A_c was the absorbance of control reaction and A_s the absorbance in the presence of *A.commutatus* tuber extracts.

ABTS radical cation decolorisation assay [13]

The plant extract with antioxidant property was added to a pre-formed ABTS radical solution and after a fixed time period the remaining $ABTS^{\bullet+}$ is quantified spectrophotometrically at 734nm (Gulcin, 2006). $ABTS^{\bullet+}$ was produced by reacting 2mM ABTS in H_2O with 2.45mM potassium persulfate ($K_2S_2O_8$), stored in the dark at room temperature for 4 h. The $ABTS^{\bullet+}$ solution was diluted to give an absorbance of 0.750±0.025 at 734 nm in 0.1M sodium phosphate buffer (pH 7.4). Then, 1mL of $ABTS^{\bullet+}$ solution was added to different concentration of various solvent fractions. The absorbance was recorded after 30min. The percentage of radical scavenging was calculated for each concentration relative to a control without extract. The scavenging capabilities of test compounds were calculated using the following equation.

$ABTS^{\bullet+}$ scavenging (%) = $1 - \frac{A_s}{A_c} x100$

Where A_c was the absorbance of control reaction and A_s the absorbance of remaining $ABTS^{\bullet+}$ in the presence of *A.commutatus* tuber extracts.

Superoxide anion radical scavenging activity[14]

The assay depends on the capacity of different solvent fractions of *A.commutatus* tuber extracts to inhibit formazan formation by scavenging the superoxide radicals generated in a riboflavin-light- NBT system (Beauchamp & Fridovich, 1971). The reaction mixture contained 50 mM phosphate buffer, pH 7.6, 20µg riboflavin, 12 mM EDTA, and NBT 0.1 mg/3 ml, added in sequence. Different concentrations of *A.commutatus* tuber extracts were added to the reaction mixture and the reaction was initiated by illuminating the tubes under fluorescent lamp (20w) for 15minutes. Immediately after illumination, the absorbance was measured at 560 nm. Decreased absorbance of the reaction mixture indicates increased superoxide anion scavenging activity. Identical tubes, with reaction mixture, were kept in the dark and served as blanks. The percentage inhibition of superoxide anion generation was calculated using the following equation.

Super oxide radical scavenging capacity (%) = $1 - \frac{A_s}{A_c} x100$

Where A_c was the absorbance of control reaction and A_s the absorbance in the presence of *A.commutatus* tuber extracts[15].

Nitric oxide radical scavenging activity[16]

Sodium nitroprusside in aqueous solution at physiological p^H spontaneously generates nitric oxide (Marcocci, Maguire & Droy, 1994) which interacts with oxygen to produce nitric ions that can be estimated by using Greiss reagent. Scavengers of nitric oxide compete with oxygen leading to reduced production of nitric oxide. Various concentrations of different solvent fractions of *A. commutatus* were mixed with 0.4 ml of Sodium nitroprusside (10 mM) in phosphate buffer saline (PBS) pH 7.4. The volume was made up to 1.5 ml with distilled water and incubated at room temperature for 30 min. After incubation 0.5 ml of Greiss reagent (1% sulfanilamide, 2% H3PO4 and 0.1% napthylethylenediamine dihydrochloride) was added. The absorbance of the chromaphore formed during the diazotization of nitrite with sulfanilamide and subsequent coupling with napthyl ethylene diamine was read at 540 nm. BHT treated in the same way as the samples and served as a positive control. The same reaction mixture without extract served as the control. The percentage inhibition of Nitric oxide formation by the extracts was measured by the following formula.

Nitric oxide radical scavenging capacity (%)

$= 1 - \frac{A_s}{A_c} x100$

Where A_c was the absorbance of control reaction and A_s the absorbance in the presence of *A.commutatus* tuber extracts.

Prevention of deoxyribose degradation[17]

The ability of test compound to prevent Fe^{2+}/H_2O induced decomposition of deoxyribose was carried out using the method of Halliwell and Gutteridge (1981). Test compounds of different fractions of *Amorphophallus commutatus* tuber was added to a reaction mixture containing 120µl of 20mM deoxyribose, 400µl 0.1M phosphate buffer, 40µl of 20mM hydrogen peroxide and 40µl of 500µM FeSo4, and the volume was made up to 800µl with distilled water. The reaction mixture was incubated at 37ºC for 30 min, and the reaction was then arrested by adding 0.5 ml of 2.8% TCA. This was followed by the addition of 0.4ml of 0.6% TBA solution. The tubes were subsequently incubated in boiling water for 20 min. The absorbance was measured at 532 nm in spectrophotometer. The percentage inhibition of hydroxyl radical production by the extracts was calculated using the following equation.

Hydroxy radical scavenging capacity (%) = $1 - \frac{A_s}{A_c} x100$

Where A_c was the absorbance of control reaction and A_s the absorbance in the presence of *A.commutatus* tuber extracts.

Calculation of IC_{50}

Various concentrations (25 -100 µg) of *A.commutatus* tuber extracts were taken for the study and IC_{50} value (which shows 50% inhibition) was calculated using Regression analysis in MS Excel.

Statistical analysis

Experimental results concerning this study were represented mean ±SD of three parallel measurements. Analysis of variance was performed by Two - way ANOVA employing Agres stat version 7.01. The p value < 0.05 were regarded as significant.

RESULTS

Antioxidant capacity is widely used as a parameter for identification of bioactive medicinal components. It is well understood that termination of free radical chain propagation by antioxidants results in reduction of chronic diseases, DNA damage, Mutagenesis, carcinogenesis and inhibition of pathogenic bacterial growth[18].

IN VITRO RADICAL SCAVENGING ASSAYS

Scavenging activity on synthetic radicals

DPPH Radical scavenging activity

The Table .1 indicates the free radical scavenging ability of different solvent fractions of *Amorophophallu commutatus* and standard BHT. Ethyl acetate fraction exhibited significant radical scavenging effect of 80.10 ± 2.07 % at 100µg concentration. This value was significant ($p<0.05$) than standard BHT which showed 76.79 ± 1.10 % at 100µg concentration. The methanol fraction showed 60.68 ± 1.77 % followed by chloroform 38.46 ± 1.30 and Petroleum ether corresponding to 30.59 ± 1.40 %. The IC_{50} value are shown in Table.1. The IC_{50} value of BHT was 53.9 ± 0.6 µg followed by Ethyl acetate, Methanol, Chloroform and Petroleum ether corresponding to 61.32 ± 1.39 µg, 77.01 ± 2.4 µg, 120.80 ± 4.77 µg and 209.62 ± 14.83 µg respectively. The results indicate that Ethyl acetate fraction at 100 µg concentration exhibited significant ($p<0.05$) radical scavenging ability when compared to standard BHT. But BHT even at lower concentration has shown 59.35±2.89% that was almost 46% higher than the ethyl acetate fraction and that is the reason that the IC_{50} value of BHT is better than Ethyl acetate fraction.

ABTS radical cation scavenging activity

Bleaching of blue – green $ABTS^{o+}$ at 734 nm can be monitored by decrease in absorbance similar to DPPH assay. Among the tested solvent fractions ethyl acetate fraction exhibited effective cation scavenging activity. As shown in Table.2. Ethyl acetate fraction was an effective ABTS•+ radical scavenger with % radical scavenging corresponding to 76.33±1.50 at 100µg and the corresponding IC_{50} value are shown in Table.2 and was 69.6±1.1µg. The values of BHT the positive standard at 100µg was 69.53±0.49% with an IC50 value of 80.8±1.1 µg. The next better activity was observed by chloroform extract with an IC 50 value of 83.3±0.9µg. Moderate radical scavenging activity was produced by methanol and Hotwater with their IC50 values corresponding to 104.8±0.5µg and 124.4±0.8µg respectively. No significant ($p<0.05$) activity was observed in Petroleum ether extract with the IC 50 value of 219.1±6.8µg. $ABTS^{o+}$ scavenging activity was in correlation with $DPPH^{o}$ scavenging activity.

Superoxide anion radical scavenging activity

The percentage scavenging effect of superoxide anion (Table.3) was significant ($p<0.05$) in BHT at 100 µg corresponding to 72.51 ±0.89 %. The effect was similar between ethyl acetate and petroleum ether fractions with their effect at 100 µg corresponding to 62.88 ± 0.99 % and 61.65 ± 0.88 % respectively. The order of scavenging of other fractions at 100 µg were as follows chloroform 45.39 ± 1.08 %, Hot water 31.20 ±1.77 % and methanol 26.06 ±1.88 %. The IC_{50} values are as shown in the table. 3.

Nitric oxide radical scavenging activity

In the present study nitric oxide radical (NO) generated from sodium nitro prusside at physiological p^{H} was found to be inhibited by different fractions in a dose dependent manner as shown in Table.4. Standard BHT exhibited significant ($p<0.05$) Nitric Oxide scavenging activity corresponding to 51.68±1.13% at 100 µg. The % scavenging of solvent fractions was in the following order Ethylacetate > Petroleum ether > Methanol > Chloroform > Hotwater. The values were 39.87±0.51%, 34.46±1.27%, 31.56±0.92, 24.56±1.18% and 23.26±1.51% respectively. Their corresponding IC_{50} values as shown in Table.4 were 139.6±2.2 µg, 142.4±5.2 µg, 158.6±1.9 µg, 176.4±4.9 µg and 236.2±10.4 µg. Standard BHT exhibited IC50 value of 82.4±1.0 µg which was nearly 40% greater than Ethylacetate fraction which showed significant ($p<0.05$) activity among the extracts.

Prevention of deoxy Ribose Degradation

The OH^{o} is known to cause DNA damage by degradation of the deoxyribose moiety. The effect of different solvent fractions on degradation of deoxy ribose by generation of hydroxyl radicals through fentons reactions is shown in Table.5. Ethyl acetate fraction exhibited significant ($p<0.05$) percentage of OH^{o} scavenging next to standard BHT. The values at 100 µg were 64.8 ± 1.86 % and 87.42 ± 1.99 % respectively. The order of inhibition of hydroxyl radical was chloroform > Petroleum ether > Methanol > Hot water with their values at 100ug corresponding to 62.46 ± 3.04 %, 56.07 ± 1.77 %, 38.74 ± 4.10 % and 33.43 ± 2.19 % respectively. The IC50 value (Table.1) of BHT was 49.6 ± 0.8 µg followed by ethyl acetate 62.4 ± 1.1 µg, chloroform 65.0 ± 2.6 µg, petroleum ether 98.2 ± 0.9 µg, Methanol 111.1 ± 5.4 µg and Hot water 167.2 ± 11.2 µg.

DISCUSSION

There are several methods of determination of antioxidant properties. The extracts with mixture of chemical compounds with different functional group, polarity and chemical behavior render chemical complexity to the extracts that scattered the results of the test employed. This study is approached with multiple assays to evaluate the antioxidant potential of different extracts of *Amorphophallus commutatus* tuber and compared against standard BHT as it would be more informative and even necessary.

$DPPH^{o}$ scavenging activity of aqueous extract of potato peel which exhibited 90% scavenging at 5mg concentration[19]. The methanolic extract of the tuber of *Amorphophallus companulatus* the species closer to *Amorphophallus commutatus* was examined for the DPPH radical scavenging activity and was reported to have an IC50 value of 3.39mg/ml[20]. The whole plant of *Coronopus didymus* was fractionated on the basis of polarity to evaluate the radical scavenging ability and reported the presence of $ABTS^{o+}$ scavenging activity in the non polar fractions[21].

Super oxide anion is oxygen centered weak oxidant play a vital role in the formation of other reactive oxygen species like singlet oxygen and hydroxyl radicals leading to lipid peroxidation. SO^{o-} is known to involve in the accumulation of ROS/ RNS in cells leading to redox imbalance and associated physiological consequences[22]. The potato peel extract was identified to scavenge SO^{o} effectively in a dose dependent manner[19].

Nitric oxide radical (NO) act as a chemical mediator and is involved in the regulation of physiological activities. It is found to be generated by endothelial cells, macrophages and neurons[23]. The ethyl acetate extract of *Stachytarpheta jamaicensis* has also been shown to possess antioxidant effects, inhibiting super oxide radical and NO radical in peritoneal macrophages[24]. Nitric oxide radicals play a vital role in vascularisation and metastasis of tumor[25] and extracts with higher NO scavenging activity may be utilized in reducing the pathogenesis caused by cancer.

Hydroxy radical (OH^o) is an extremely reactive free radical produced by the biological system. It quickly initiates the process of free radical chain reaction by abstracting hydrogen atom from unsaturated fatty acids[26]. Thereby, leading to membrane damage, DNA strand breakage, and finally inducing cytotoxicity, carcinogenesis or mutagenesis[27]. The hydroxyl radical scavenging activity of the peanut skin extract was noted to be higher than BHT[28]. The potato peel extract exhibited a strong concentration dependent hydroxyl radical scavenging activity in deoxy ribose degradation system[19]. There are no specific enzymes to defend against OH^o radicals[29] and discovery of some compounds with hydroxyl radical scavenging ability would gain significant role by providing ailment to the damage caused by OH^o.

CONCLUSION

The present study reports for the first time *in vitro* free radical scavenging activity of *Amorphophallus commutatus* an endemic plant of western ghats. The antioxidant and free radical scavenging potency of all the five extracts at five different concentration was investigated employing various established *in vitro* systems and the IC_{50} values calculated indicate that the ethyl acetate fraction was identified to possess significant radical scavenging activity among the five extracts. The data obtained in the *in vitro* study has established that the Ethyl acetate fraction has potential free radical scavenging properties followed by petroleum ether and methanol extracts. These findings are very useful for the future research to identify the specific phytochemical responsible for the antioxidant activities. Characterization of the Phytochemical and structural elucidation of the responsible compound await a major breakthrough as they have varied applications in the food, medicine and nutraceutical industries. Data mining of the elucidated structure for structure activity relationship could be carried out. This might lead to enhanced predictability of specific changes in the molecule to increase its activity. However an elaborate study needs to be carried out to ascertain the in vivo safety of the extracts in experimental animal models.

ACKNOWLEDGMENT

The authors wish to thank the Department of Science and Technology, Government of India for having funded this project under the Women scientist scheme (SR/WOS-A/LS-17/2009).

REFERENCES

1. Halliwell B, Gutteridge JMC. Role of free radicals and catalytic metal ions in human disease: an overview. Methods in Enzymology 1998; 186: 1–85.
2. Moskovitz J, Yim MB, Chock PB. Free radicals and disease. Archives of Biochemistry and Biophysics 2002; 397: 354–359.
3. Mello LD, Kubota LT. Biosensors as a tool for the antioxidant status evaluation. Talanta 2007; 72: 335–348.
4. Moure A, Cruz JM, Franco D, Dominguez JM, Sineiro J, Dominguez H et al.,. Natural antioxidants from residual sources. Food Chemistry 2001; 72: 145–171.
5. Nogochi C, Nikki E. Phenolic antioxidants: A rationale for design and evaluation of novel antioxidant drugs for atherosclerosis. Free Radicals in Biology and Medicine 2000; 28: 1538–1546.
6. Hetterscheid W L A, Ittenbach S. Everything you always wanted to know about Amorphophallus, but were afraid to stick your nose into. Aroideana 1996; 19: 7–131.
7. Raju Gautam, Arvind Saklani, Jachak SM. Indian medicinal plants as a source of antimycobacterial agents. Journal of Ethnopharmacology 2007; 110: 200–234.
8. Khan A, Rhaman M, Islam MS. Antibacterial, antifungal and cytotoxic activities of amblyone isolated from Amorphophallus campanulatus. Indian Journal of Pharmacology 2008; 40: 41-44.
9. Ravikumar K, Ved DK. (2004). Illustrated Field Guide – 100 Red listed medicinal plants of conservation concern in S.India, FRLTH, Bangalore.
10. Jain A, Katewa SS, Galav PK, Sharma P. Medicinal plant diversity of Sitamata wildlife sanctuary, Rajasthan, India. Journal of ethnopharmacology 2005; 102: 143–157.
11. Harbone JB. (1998). Phytochemicals methods: Aguide to modern technique of plant analysis. Londone: Chapman and Hall ltd.
12. Mensor LL, Menezes FS, Leitao GG, Reis AS, dos Santos TC, Coube, CS, Leitao SG. Screening of Brazilian plant extracts for antioxidant activity by the use of DPPH free radical method. Phytotheraphy Research 2001; 15: 127-130
13. Gulcin I. Antioxidant activity of caffeic acid (3,4-dihydroxycinnamic acid). Toxicology 2006; 217: 213–220.
14. Beauchamp C, Fridovich I. Superoxide dismutase: improved assays and an assay applicable to acrylamide gels. Analytical Biochemistry 1971; 44: 276–277.
15. Gulcin I, Oktay, Kirecci ME.Screening of antioxidant and antimicrobial activities of anise (*Pimpinella anisum* L.) seed extracts, Food Chemistry 2003; 83: 371–382.
16. Marcocci L, Maguire JJ, Droy MT. The nitric oxide scavenging properties of Gingo biloba extract EGb 761. Biochemical and Biophysical Research Communications 1994;15: 748–755.
17. Halliwell B, Gutteridge JMC. Formation of thiobarbituric acid reactive substances from deoxyribose in the presence of iron salts: The role of superoxide and hydroxyl radicals. FEBS Letters 1981; 128: 347-352.
18. Zhu QY, Hackman RM, Ensunsa JL. Antioxidative activities of oolong tea. Journal of Agricultural and Food Chemistry 2002; 50: 6929–6934.
19. Singh N, Rajini PS. Free radical scavenging activity of an aqueous extract of potato peel. Food. Chemistry 2004; 85: 611-616.
20. Ramesh CK, Raghu KL, Jamuna KS, Joyce GS, Mala RSV, Vijay avin BR. Compaitive evaluation of antioxidant property in methanol extracts of some common vegetables in India. Annals of Biological Research 2011; 2(2): 86-94.
21. Prabhakar KR, Veeresha VP, Vipana K, Sudheera M, Priyadarsini KI, Satish, RBSS, Unnikrishnana MK. Bioactivity-guided fractionation of Coronopus didymus: A free radical scavenging perspective. Phytomedicine 2006; 13: 591–595
22. Pervaiz S, Clement M. Superoxide anion: oncogenic reactive oxygen species? The International Journal of Biochemistry and Cell Biology 2007; 39: 1297–1304.
23. Förstermann U . Nitric oxide and oxidative stress in vascular disease. Pflügers Archiv: European Journal of Physiology 2010; 459: 923–939.
24. Alvarez E, Leiro JM, Rodriguez M, Orallo F. Inhibitory effect of leaf extracts of *Stachytarpheta jamaicensis* (Verbenaceae) on the respiratory burst of rat macrophages. Phytotherapy Research 2004; 18(6): 457–462.
25. Jayakumar R, Kanthimathi MS. Inhibitory effects of fruit extracts on nitric oxide induced proliferation in MCF-7 cells. Food Chemistry 2011; 126: 956–960.
26. Gordon M H (1990). The mechanism of antioxidant action in vitro. In B. J. F. Hudson (Ed.), Food antioxidants. London: Elsevier Applied Science. 1–18.
27. Babu BH, Shylesh BS, Padikkala J. Antioxidant and hepatoprotective effect of Alanthus icicifocus. Fitoterapia 2001; 72: 272–277.
28. Gulcin I, Dastan A. Synthesis of dimeric phenol derivatives and determination of in vitro antioxidant and radical scavenging activities. Journal of Enzyme Inhibition and Medicinal Chemistry 2007; 22: 685–695.
29. Zhou D, Ruan J, Cai Y, Xiong Z, Fu W, Wei A. Antioxidant and hepato protective activity of ethanol extract of Arachniodes exilis (Hance) Ching. Journal of Ethnopharmacoiogy 2010; 129: 232–237.

Table 1: DPPH radical scavenging activity of *Amorphophallus commutatus*

Concentration in µg	% radical scavenging activity of extracts					
	Petroleum ether	Chloroform	Ethyl acetate	Methanol	Hot water	BHT
25	3.72 ± 0.86	12.02 ± 0.55	27.74 ± 1.55	18.31 ± 1.03	2.26 ± 0.71	59.35 ± 2.89
50	7.77 ± 1.36	22.22 ± 0.80	37.99 ± 1.90	33.40 ± 1.48	4.91 ± 1.40	62.18 ± 1.61
75	12.75 ± 1.29	33.54 ± 1.88	62.79 ± .069	53.34 ± 1.56	6.30 ± 0.73	68.27 ± 1.07
100	30.59 ± 1.40	38.46 ± 1.36	80.10 ± 2.07	60.68 ± 1.77	11.73 ± 1.21	76.79 ± 1.10
IC_{50}	209.62 ± 14.83	120.80 ± 4.77	61.32 ± 1.39	77.0 ± 2.4	488.8 ± 58.7	53.9 ± 0.6
CD ($p<0.05$)	3.124					

Table 2: ABTS radical scavenging activity of *Amorphophallus commutatus*

Concentration in µg	% radical scavenging activity of extracts					
	Petroleum ether	Chloroform	Ethyl acetate	Methanol	Hot water	BHT
25	8.50 ± 1.06	13.20 ± 0.95	20.47 ± 0.74	14.44 ± 1.50	10.30 ± 1.21	9.23 ± 0.55
50	10.07 ± 1.01	20.30 ± 0.80	26.33 ± 0.85	15.50 ± 1.35	15.47 ± 1.36	20.30 ± 0.98
75	14.40 ± 0.62	41.37 ± 0.86	53.57 ± 0.91	25.50 ± 1.31	26.13 ± 0.21	45.57 ± 1.21
100	24.93 ± 1.42	68.17 ± 1.05	76.33 ± 1.50	59.10 ± 1.05	45.60 ± 0.70	69.53 ± 0.49
IC_{50}	209.1 ± 6.8	83.3 ± 0.9	69.6 ± 1.1	1004.8 ± 0.5	124.4 ± 0.8	80.8 ± 1.1
CD ($p<0.05$)	1.712					

Table 3: Superoxide radical scavenging activity of *Amorphophallus commutatus*

Concentration in µg	% radical scavenging activity of extracts					
	Petroleum ether	Chloroform	Ethyl acetate	Methanol	Hot water	BHT
25	10.29 ± 0.96	14.08 ± 1.73	12.69 ± 0.70	7.56 ± 1.47	6.74 ± 1.58	16.18 ± 1.01
50	16.71 ± 1.41	60.66 ± 0.83	16.05 ± 1.25	11.08 ± 1.71	9.66 ± 0.93	33.97 ± 1.30
75	32.71 ± 1.23	32.22 ± 1.36	36.45 ± 0.82	18.77 ± 1.59	11.27 ± 1.22	64.52 ± 0.64
100	61.65 ± 0.88	45.39 ± 1.08	62.88 ± 0.99	26.06 ± 1.88	31.20 ± 1.77	72.51 ± 0.89
IC_{50}	95.5 ± 2.7	115.4 ± 3.4	92.5 ± 1.1	198.1 ± 14.8	204.0 ± 15.8	66.1 ± 1.1
CD ($p<0.05$)	2.132					

Table 4: Nitric oxide radical scavenging activity of *Amorphophallus commutatus*

Concentration in µg	% radical scavenging activity of extracts					
	Petroleum ether	Chloroform	Ethyl acetate	Methanol	Hot water	BHT
25	5.10 ± 0.62	16.54 ± 2.31	9.90 ± 0.83	10.59 ±0.67	3.17 ± 0.25	38.17 ± 1.48
50	14.72 ± 1.05	17.97 ± 1.86	17.4 ± 1.57	16.44 ± 0.59	6.23 ± 1.03	40.57 ± 1.65
75	30.49 ± 1.42	20.77 ± 1.58	21.71 ± 1.93	22.47 ± 0.75	17.01 ± 0.84	43.22 ± 1.76
100	34.46 ± 1.27	24.56 ± 1.18	39.87 ± 0.51	31.56 ± 0.92	23.26 ± 1.51	51.68 ± 1.13
IC_{50}	142.4 ± 5.2	176.4 ± 4.9	139.6 ± 2.2	158.6 ± 1.9	236.2 ± 16.4	82.4 ± 1.0
CD ($p<0.05$)	2.134					

Table 5: Deoxyribose radical scavenging activity of *Amorphophallus commutatus*

Concentration in µg	% radical scavenging activity of extracts					
	Petroleum ether	Chloroform	Ethyl acetate	Methanol	Hot water	BHT
25	11.21 ± 1.82	51.53 ± 4.10	53.12 ± 4.90	20.32 ± 2.36	10.49 ± 1.51	51.44 ± 1.63
50	25.66 ± 2.08	52.29 ± 3.76	56.28 ± 3.55	28.66 ± 2.06	12.71 ± 1.05	64.45 ± 1.21
75	31.81 ± 2.33	57.22 ± 0.86	59.00 ± 2.05	35.26 ± 1.97	18.59 ± 1.17	75.52 ± 1.51
100	56.07 ± 1.77	62.46 ± 3.04	64.80 ± 1.86	38.74 ± 4.10	33.43 ± 2.19	87.42 ± 1.99
IC_{50}	98.2 ± 0.9	65.0 ± 2.6	62.4 ± 1.1	111.1 ± 5.4	167.2 ± 11.2	49.6 ± 0.8
CD ($p<0.05$)	4.129					

Source of support: Department of Science and technology, Conflict of interest: None Declared