African Journal of Food Science Vol 4(6), pp. 403 - 409, June 2010
Available online http://www.academicjournals.org/AJFS
ISSN 1996-0794 ©2010 Academic Journals

Full Length Research paper

Changes in the total phenol content and antioxidant properties of pepperfruit (*Dennettia tripetala*) with ripening

B. C. Adedayo*, G. Oboh and A. A. Akindahunsi

Department of Biochemistry, Federal University of Technology, P. M. B. 704, Akure, Nigeria.

Accepted 5 January, 2010

Pepperfruit (*Dennettia tripetala* G. Baker (Annonaceae) is a well-known Nigerian spicy medicinal plant normally used in folk medicine to cure fever, cough, toothache, as well as a stimulant and in the preparation of some special dishes for pregnant and postpartum women. The present study sought to assess the antioxidant properties of the aqueous extract of ripe (RPF) and unripe pepperfruit (UPF) extract. The aqueous extract of the pepperfruits were prepared, and their antioxidant phytoconstituents (total phenol,total flavonoid and ascorbic acid) and activities (Fe^{2+} chelation ability, OH* scavenging ability, reducing power, DPPH and ABTS radical scavenging abilities) were subsequently determined. The total phenol (1.4 mg/g) content of RPF was significantly higher ($P < 0.05$) than that of UPF (1.0 mg/g), while there was no significant difference in the vitamin C and total flavonoid content. Conversely, UPF had higher antioxidant activities, as typified by higher reducing power, DPPH*, ABTS* and OH* scavenging abilities, and Fe chelating ability. Therefore, the physiological changes that accompanies ripening of pepperfruit that brings about changes in pigment would increase the total phenol content, but decreases the antioxidant properties of pepperfruit.

Key words: Pepperfruit, phenol, flavonoid, vitamin C, antioxidant.

INTRODUCTION

Various reactive oxygen species(ROS) are generated in living organisms through different ways. Reactive oxygen species (ROS), which include free radicals such as superoxide anion radicals, hydroxyl radicals and nonfree-radical species such as H_2O_2 and singled oxygen, are various forms of activated oxygen(Gulcin et al., 2002). These molecules are exacerbating factors in cellular injury and aging process (Lai et al., 2001). Iron, an essential metal for normal cellular physiology, can result in cell injury when in excess. This is because it plays a catalytic role in the initiation of free radical reactions. Fe (II) can react with hydrogen peroxide (H_2O_2) to produce the hydroxyl radical (OH•) via the Fenton reaction, whereas superoxide can react with iron (III) to regenerates iron (II) that can participate in the Fenton reaction (Harris, 1996).

The human body is equipped with an antioxidant defense system that deactivates these highly reactive free radicals; this includes antioxidant enzymes (made in the body) and antioxidant nutrients (found in foods) that soak up all the excess reactivity that these free radicals have, turning them to harmless particles that can be get rid of (Oboh, 2006; Oboh and Akindahunsi, 2004; Oboh, 2005). However, recent studies have revealed that one of the practical ways through which the activity of free radicals could be managed in the body is through dietary means (Oboh and Rocha, 2007). Dietary antioxidants may play an important role in protecting the cell against damage caused by free radicals by acting as radical scavengers, reducing agents, forming complexes with pro-oxidant metals, and quenchers of singlet oxygen formation(Oboh, 2005; Hochstein and Atallah, 1988).

Dennettia tripetala G. Baker (Annonaceae) also known as pepperfruit tree is a woody plant of at least 3meters height with simple leaves and abundant fruits widely consumed in Southern Nigeria. It is found in the tropical rainforest region of Nigeria and sometimes in Savana areas (Okwu et al., 2005). It is locally called “Nkarika” by the Efiks of Calabar,”Nmimi” by the Igbos and “Igbere” by the Yorubas. The young leaves and fruits have instinctive

*Corresponding author. E-mail: chrisadeb2005@yahoo.com.

spicy taste (Achinewhu et al., 1995). The mature fruits constitute the main edible portions. Some communities in parts of Southern Nigeria also utilized the leaves and roots, in addition to the fruits for medicinal purpose (Iwu, 1989). *Dennettia tripetala* is used as masticators, which when chewed produces unique peppery effect (Keay, 1989). The peppery spicy taste of mature *D. tripetala* fruits usually serves as a mild stimulant to the consumer. The fruits are sometimes taken with kolanut, garden egg and palm wine in parts of Nigeria, especially in Southern part of Nigeria where it serves also for cultural entertainment of guests, particularly during coronation, new yam festivals, weddings and marriage festivals (Enwere, 1998; Keay, 1989). *D. tripetala* fruit has also been reported to be used as spice in flavouring food, and as seasoning which are added to prepared food such as meat, sausages, soups and vegetable (Lebouef and Caver, 1972). The peppery fruits of *D. tripetala* are applied to the food meant for pregnant women and are important in the diets of postpartum women, during which time it is claimed that spices and herbs aid uterine contraction (Okwu and Morah, 2004; Achinewhu et al., 1995). Okwu et al. (2005) also reported that *D. tripetala* fruits contain important nutritive substances such as vitamins, minerals and fibre. However, there is limited information on the antioxidant activity of *D. tripetala* fruits as a dietary source of antioxidant. The present study therefore tend to assess the potentials of this pepperfruits as a dietary source of antioxidant and to determine the state (ripe or unripe) in which the antioxidant activities is higher.

MATERIALS AND METHODS

Materials

Fresh samples of ripe and unripe *D. tripetala* fruits were purchased from Oba's market in Akure, Ondo State, Nigeria.The identification and authentication was carried out at the Crop, Soil, and Pest management (CSP) Department of the Federal University of Technology, Akure, Nigeria. All the chemicals used were of analytical grade, while the water was glass distilled.

Methods

Aqueous extract preparation

Matured ripe and unripe pepperfruits were washed in distilled water and chopped into small pieces by table knife. The aqueous extract of the pepperfruit was subsequently prepared by homogenizing the pepperfruits in water (1:20 w/v); the homogenates were centrifuged at 2,000 rpm for 10 min. The supernatant was used for the assay (Oboh et al., 2007).

Determination of total phenol content

The total phenol content was determined on the extracts using the method reported by Singleton et al. (1999). Appropriate dilutions of the extracts were oxidized with 2.5 ml of 10% Folin–Ciocalteau's reagent (v/v) and neutralized by 2.0 ml of 7.5% sodium carbonate. The reaction mixture was incubated for 40 min at 45°C and the absorbance was measured at 765 nm. The total phenol content was subsequently calculated using Gallic acid as standard.

Determination of total flavonoid content

The total flavonoid content of the extracts was determined using a slightly modified method reported by Meda et al. (2005). Briefly, 0.5 ml of appropriately diluted sample was mixed with 0.5 ml methanol, 50 µl of 10% $AlCl_3$, 50 µl of 1 mol L^{-1} potassium acetate and 1.4 ml water, and allowed to incubate at room temperature for 30 min. Thereafter, the absorbance of the reaction mixture was subsequently measured at 415 nm. The total flavonoid was calculated using Quercetin as standard.

Vitamin C determination

The vitamin C was determined using the method of Benderitter et al. (1998). Briefly 75 µl of DNPH (2 g of dinitrophenyl hydrazine, 230 mg thiourea and 270 mg $CuSO_4$. $5H_2O$ in 100 ml 5 M H_2SO_4) was added to 500 µl reaction mixture (300 µl of the pepperfruit extracts with 100 µl 13.3% TCA and water, respectively). The reaction mixture was subsequently incubated for 3 h at 37°C, then 0.5 ml H_2SO_4 65% (v/v) was added to the medium, and the absorbance was measured at 520 nm, and the Vitamin C content of the sample was subsequently calculated, using a vitamin C standard curve.

Determination of reducing property

The reducing property of the extracts was determined by assessing the ability of the extract to reduce $FeCl_3$ solution as described by Oyaizu (1986). A 2.5 ml aliquot was mixed with 2.5 ml of 200 mmol l^{-1} sodium phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min and then 2.5 ml of 10% trichloroacetic acid was added. This mixture was centrifuged at 650 rpm for 10 min. 5 ml of the supernatant was mixed with an equal volume of water and 1ml of 0.1% ferric chloride. The absorbance was measured at 700 nm and ferric reducing antioxidant property was subsequently calculated using ascorbic acid as standard.

DPPH free radical scavenging ability

The free radical scavenging ability of the extracts against DPPH (1,1-diphenyl–2 picrylhydrazyl) free radical was evaluated as described by Gyamfi et al. (1999). Briefly, an appropriate dilution of the extracts (1 ml) was mixed with 1 mL of 0.4 mmol l^{-1} methanolic solution containing DPPH radicals. The mixture was left in the dark for 30 min and the absorbance was measured at 516 nm. The DPPH free radical scavenging ability was subsequently calculated with respect to the reference (which contains all the reagents without the test sample).

2,2-azinobis(3-ethylbenzo-thiazoline-6-sulfonate) $ABTS^*$ scavenging ability

The $ABTS^*$ scavenging ability of both extracts was determined according to the method described by Re et al. (1999). $ABTS^*$ was generated by reacting an ABTS aqueous solution (7 mmol l^{-1}) with $K_2S_2O_8$ (2.45 mmol l^{-1}, final concentration) in the dark for 16 h and adjusting the Abs 734 - 0.700 nm with ethanol. 0.2 ml of appropriate dilution of the extract was added to 2.0 ml $ABTS^*$ solution and the

Table 1. Total phenol, Total flavonoid and Vitamin C content of Pepperfruits (mg/g).

Sample	Total phenol	Total flavonoid	Vitamin C
Ripe	1.4 ± 0.0	0.1 ± 0.0	6.3 ± 0.2
Unripe	1.0 ± 0.0	0.1 ± 0.0	5.9 ± 0.7

absorbance were measured at 734 nm after 15 min. The trolox equivalent antioxidant capacity was subsequently calculated.

OH radical scavenging ability

The ability of the extract of the extract to prevent Fe^{2+}/H_2O_2-induced decomposition of deoxyribose was carried out using the method of Halliwell and Gutteridge,1981.Briefly, freshly prepared aqueous extract (0 - 100 µl) was added to a reaction mixture containing 120 µl 20 mM deoxyribose, 400 µl 0.1 M phosphate buffer, 40 µl 20 mM hydrogen peroxide, and 40 µl 500 µM FeSO4, and the volume were made to 800 µl with distilled water. The reaction mixture was incubated at 37°C for 30 min and the reaction was stopped by the addition of 0.5 ml of 2.8% trichloroacetic acid; this was followed by the addition of 0.4 ml of 0.6% thiobarbituric acid (TBA) solution. The tubes were subsequently incubated in boiling water for 20 min. The absorbance was measured at 532 nm in a spectrophotometer.

Fe^{2+} chelation assay

The Fe^{2+} chelating ability of both extracts were determined using a modified method of Minotti and Aust (1987) with a slight modification by Puntel et al. (2005). Freshly prepared 500 µmol l^{-1} $FeSO_4$ (150 µl) was added to a reaction mixture containing 168 µl of 0.1mol l^{-1} Tris-HCl (pH 7.4), 218 µl saline and the extracts (0 - 25 µl). The reaction mixture was incubated for 5 min, before the addition of 13 µl of 0.25% 1,10-phenanthroline(w/v).The absorbance was subsequently measured at 510 nm in a spectrophotometer. The Fe^{2+} chelating ability was subsequently calculated.

Data analysis

The results of the replicates were pooled and expressed as mean±standard deviation. Analysis of variance and Student's *t*-test were carried out (Zar, 1984). Significance was accepted at $p \leq 0.05$.

RESULTS AND DISCUSSION

The results of thet total phenol content of ripe (RPF) and unripe (UPF) Dennettia tripetala (pepperfruits) are presented in Table 1. The result of the study revealed that RPF (1.4 mg/g) pepperfruit had a significantly higher ($P < 0.05$) total phenol content than UPF (1.0 mg/g). This value is higher than that of ripe and unripe *Capsicum pubescens* (Oboh and Rocha,2007) and *Capsicum chinese, Habanero* but lower than that of *Capsicum annum, Tepin* (Oboh et al., 2007) and some commonly consumed fruit (Chu et al., 2002; Sun et al., 2002) and green leafy vegetables (Oboh, 2005; Oboh and Akindahunsi, 2004). The basis for the significant difference in the total phenol content of ripe and unripe peppefruit cannot be categorically stated, however, it may be due to the physiological changes that accompanied ripening that brings about changes in pigments, may have caused an increase in the total phenol content (Oboh et al., 2007; Materska and Perucka, 2005). The trend in the changes of phenol content with ripening agrees with that of *Capsicum chinese, Habanero* (Oboh et al.,2007; Materska and Perucka, 2005) in that ripe pepper had higher total phenol than the unripe.

The total flavonoid contents of the ripe and unripe pepperfruit is presented in Table1, the result revealed that there was no significant difference ($P > 0.05$) in the total flavonoid content of UPF and RPF, this possibly implies that the increase in the total phenol content in RPF may have occurred in the non-flavonoid constituents of the pepperfruit. However, the total flavonoid content of pepperfruits were lower than that of some common tropical green leafy leafy vegetables (Oboh et al., 2008). Flavonoids are a class of widely distributed phytochemicals with antioxidant and biological activity. They are constituents of plant foods that have been implicated in the reduction of cancer risk (Wolfe and Liu, 2008). In the Zutphen Elderly Study, flavonoid intake from fruits and vegetables was inversely associated with all-cause cancer risk and cancer of the alimentary and respiratory tracts (Hertog et al., 1994). Lung cancer risk has also been inversely associated with flavonoid (Knekt et al., 1997) and quercetin intake (Le Marchand et al., 2000). Although not definitive, many other epidemiological studies have shown a trend for decreased cancer risk with higher flavonoid consumption (Neuhouser, 2004; Graf et al., 2005).

The results of the ascobic acid content of the pepperfruit is also presented in Table 1. The result revealed that there was no significant difference ($P > 0.05$) between the vitamin C content of RPF (6.3 mg/g) and UPF (5.9 mg/g), unlike earlier reports on *Capsicum annum, Tepin, Capsicum chinese, Habanero and Capsicum pubescens* (Oboh et al., 2007; Oboh and Rocha, 2008), where ripening caused a significant change in the vitamin C content of the pepper. However, the vitamin C content of the pepperfruit (ripe and unripe) was higher than that of *Capsicum annum*, *Tepin* (ripe and unripe) and *Capsicum chinese, Habanero* (ripe and unripe) (Oboh et al., 2007) and what Chu et al. (2002) reported for red pepper and also that of some common vegetables (Oboh, 2005; Sun et al., 2002).

The antioxidant activities of plant phytochemicals occurs by preventing the production of free radicals or by neutralizing/scavenging free radicals produced in the body or reducing/chelating the transition metal composition of foods (Amic, 2003; Oboh et al., 2007). Prevention of the chain initiation step by scavenging various reactive species such as free radicals is considered an important antioxidant mode of action (Dastmalchi et al., 2007). DPPH is a free radical donor that accepts an electron or hydrogen to become a stable diamagnetic molecule (Je et al., 2009). The tendencies of electron or hydrogen donation are critical factors in characterizing antioxidant activity

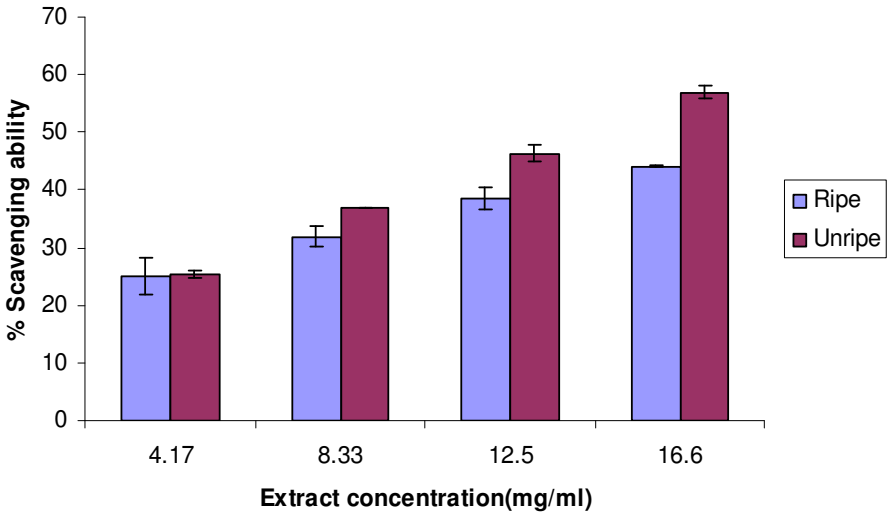

Figure 1. DPPH* scavenging ability of aquous extract of pepperfruit.

Figure 2. Antioxidant capacity of aquous extract of pepperfruit.

that involves free radical scavenging (Hu et al., 2000). The DPPH* scavenging ability of aqueous extract of ripe and unripe pepperfruit is presented in Figure 1, both extracts scavenged DPPH* in a dose dependent manner; however, the result revealed that UPF had higher free radical scaveging ability than RPF.

DPPH is frequently used in the determination of free radical scavenging ability of various food components; however, it has the limitation of colour interference and sample solubility (Dorman and Hiltunen, 2004; Oboh et al., 2008). Therefore, the free radical scavenging ability of the extracts was further studied using a moderately stable nitrogen-centered radical species; ABTS radicals (Re et al., 1999). The ABTS radical based model of free radical scavenging ability has the advantage of being more versatile due to the minimal spectral interference as the absorption maximum used is 760 nm, a wavelength not normally encountered with natural products (Re et al., 1999). ABTS* scavenging ability reported as trolox equivalent antioxidant capacity (TEAC), is presented in Figure 2. The result showed that the ABTS* scavenging ability of the RPF (0.05 mmol/g) was significantly lower than that of unripe (0.07 mmol/g). The trend in the ABTS* scavenging ability of both aqueous extract ripe and unripe pepperfruit agrees with that of DPPH free radical scavenging ability, in that despite the fact that ripening increase the total phenol content of the pepperfruit, there was a decrease in the radical scavenging abilities of the

Figure 3. Ferric reducing antioxidant properties of aquous extract of pepperfruit.

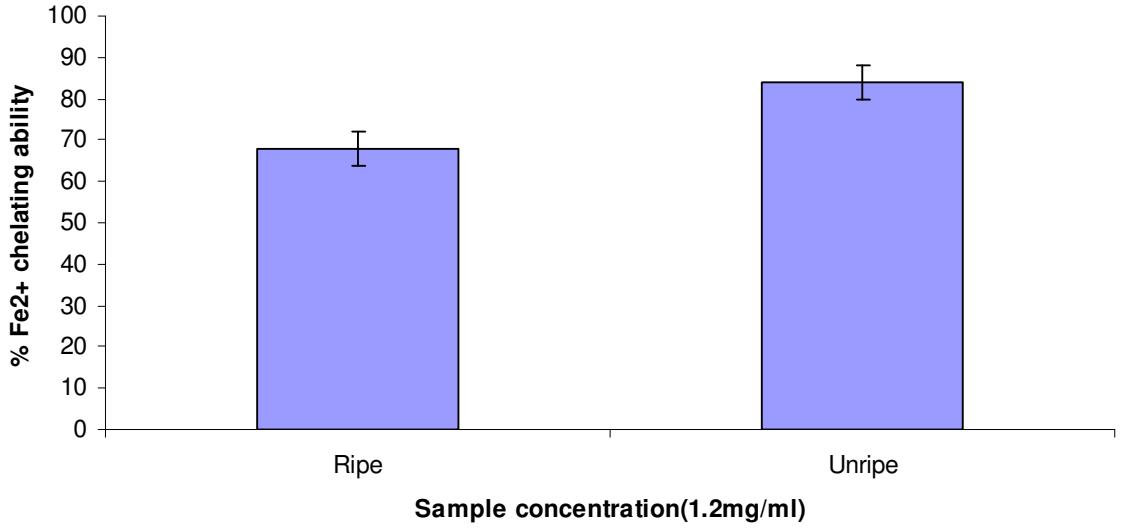

Figure 4. Fe^{2+} chelating ability of aquous extract of pepperfruit.

pepperfruits.

Reducing power is a novel antioxidation defence mechanism; the two mechanisms that are available to affect this property are by electron transfer and hydrogen atom transfer (Dastmalchi et al., 2007). Both extracts were able to reduce Fe^{3+} to Fe^{2+} and the results are presented in Figure 3 as ascorbic acid equivalent. How-ever, UPF had a higher reducing power than the RPF. This trend agrees with the earlier trend by DPPH* and ABTS* scavenging abilities (Figures 1 and 2) earlier discussed, this goes a long way confirm that the physio-logical changes that accompanied ripening that brings about change in pigments would decrease the antioxidant activities of pepperfruit, despite the increase in the water soluble phenolics. The basis for this findings cannot be categorically stated, however, it will not far fetch from the possibility that the process of ripening may have led to loss of some key antioxidant constituents in the pepperfruits, which were not analyzed for in this study.

Antioxidants chelate and deactivate transition metals, thereby preventing such metals from participating in the initiation of lipid peroxidation and oxidative stress through metal catalysed reaction (Oboh and Rocha, 2007). Fe^{2+} chelating ability of the extracts is presented in Figure 4. The results revealed that both extracts chelate Fe^{2+}; however, UPF had higher Fe^{2+} chelating ability than the RPF. Furthermore, Fe^{2+} can catalyze one-electron transfer reactions that generate reactive oxygen species such as the very reactive OH^{*}, which is formed from H_2O_2 through the Fenton's reaction (Oboh, 2008), and OH^{*} has

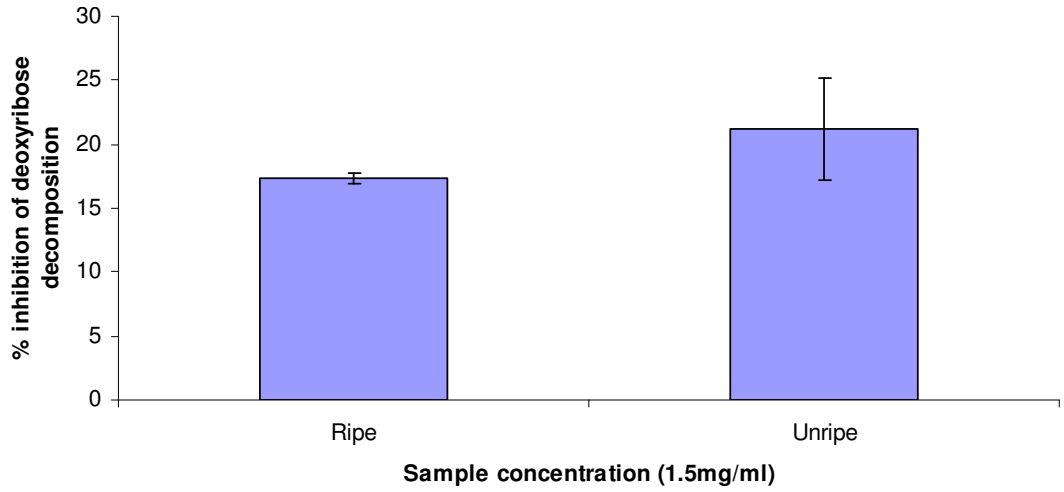

Figure 5. OH* Scavenging ability of aquous extract of pepperfruit.

been recognized to date as the most reactive oxygen specie (ROS). The overproduction of ROS can directly attack the polyunsaturated fatty acids of the cell membranes and induce lipid peroxidation (Elmegeed et al., 2005). As shown in Figure 5, both extracts scavenged OH radical produced in Fe^{2+}/H_2O_2 induced decomposition of deoxyribose in Fenton's reaction. This results also revealed that UPF had significantly higher ($P < 0.05$) OH* scavenging ability than RPF. It is worth noting that pepperfruit (ripe and unripe) had higher OH* radical scavenging ability than the various varieties of pepper earlier reported (Oboh et al., 2007; Oboh and Rocha, 2008).

Conclusion

The present study shows that unripe pepperfruit has higher antioxidant activities than the ripe pepperfruit, despite its lower total phenol content. Therefore, the physiological changes that accompanies ripening of pepperfruits that brings about changes in pigment would increase the total phenol, but decreases the antioxidant properties of pepperfruit.

REFERENCES

Achinewhu SG, Ogbonna C, Hard AD (1995). Chemical composition of indigenous wild herbs, spices fruits, nuts and leafy vegetables used as food. Plants Food for Human Nutrition. Kluwer Publishers, Netherlands, 48: 341-388.

Amic D, Davidovic-Amic D, Beslo D, Trinajstic N (2003). Structure–radical scavenging activity relationship of flavonoids. *Croatia* Chem Act, 76: 55-61.

Benderitter M, Maupoil V, Vergely C, Dalloz F, Briot F, Rochette L (1998). Fundam Clin. Pharm., 12: 510-516.

Chu Y, Sun J , Wu X , Liu RH, (2002). Antioxidant and antiproferative activity of common vegetables. J. Agric. Food Chem., 50: 6910-6916.

Dastmalchi K, Dorman HJD, Kosar M, Hiltunen R (2007). Chemical composition and *in vitro* antioxidant evaluation of a water soluble Moldavian balm (*Dracocephalum moldavica* L.) extract. Lebensm Wiss Technol., 40: 239-248.

Dorman HJD, Hiltunen R (2004). Fe (II) reductive and free radical scavenging properties of summer savory (*Satureja hortensis* L.) extract and subfractions. Food Chem., 88: 193-199.

Elmegeed GA, Ahmed HA, Hussein JS (2005). Novel synthesized amino steroidal heterocycles intervention for inhibiting iron induced oxidative stress. Eur. J. Med. Chem., 40: 1283-1294.

Enwere NJ (1998). Foods of plant origin. Afro-Orbis Publications Ltd., University of Nigeria, Nsukka. fractions. Cereal Chem., 48: 312-316.

Graf BA, Milbury PE, Blumberg JB (2005). Flavonols, flavones, flavanones, and human health: epidemiological evidence. J. Med. Food, 8(3): 281-290.

Gulcin I, Oktay M, Kufrevioglu OI, Aslan A (2002). Determination of antioxidant activity of lichen Cetraria islandica (L) Ach. J. Ethnopharm., 79(3): 325-329

Gyamfi MA, Yonamine M, Aniya Y (1999). Free-radical scavenging action of medicinal herbs from Ghana: *Thonningia sanguinea* on experimentally-induced liver injuries. Gen Pharmacol., 32: 661-667.

Halliwell B, Gutterdge JMC (1981). Fomation of a thiobarbituric-acid-reative substance from deoxyribose in the presence of iron salts:The role of superoxide and hydroxyl radicals. FEBS Lett., 128: 347-352.

Harris JR (1996). Biochemistry and biomedical cell biology, Plenum, New York. p. 25.

Hertog MGL, Feskens EJM, Hollman PCH, Katan MB, Kromhout D (1994). Dietary flavonoids and cancer risk in the Zutphen Elderly Study. Nutr. Cancer, 22(2): 175-184.

Hochstein P, Atallah AH (1988). The nature of oxidants and antioxidant systems in the inhibition of mutation and cancer.Mutatation Res., 202: 363-375.

Hu C, Zhang Y, Kitts DD (2000). Evaluation of antioxidant and prooxidant activities of bamboo *Phyllostachys niger* var. Henonis leaf extract *in vitro*. J. Agric. Food Chem., 48: 3170-3176.

Iwu MM (1989). Food for medicine, in Dietary plants and masticastors as sources of biologically active substances. University of Ife Press, pp. 303-310.

Je JY, Park PJ, Kim EK, Ahn CB (2009). Antioxidant and angiotensin I converting enzyme inhibitory activity of *Bambusae caulis* in Liquamen. Food Chem., 113: 932-935.

Keay RWJ (1989). Trees of Nigeria, Clarendon Press Oxford, UK, pp. 19-30.

Knekt P, Ja¨rvinen R, Seppa¨nen R, Helio¨vaara M, Teppo L, Pukkala E, Aromaa A (1997). Dietary flavonoids and the risk of lung cancer and other malignant neoplasms. Am. J. Epidemiol., 146(3): 223-230.

Lai LS, Chou ST, Chao WW (2001). Studies on the antioxidative of Hsian-tsao (Mesona procumbens Heml) leaf gum. J. Agric. Food Chem. 49: 963-968

Le Marchand L, Murphy SP, Hankin JH, Wilkens LR, Kolonel LN (2000). Intake of flavonoids and lung cancer. J. Natl. Cancer Inst., 92(2): 154-160.

Lebouef M Caver A (1972). Alkaloids desecoces I; Uvariopsine guineensis. Phytochemistry, 11: 28-33.

Materska M, Perucka I (2005). Antioxidant activity of the main phenolic compounds Isolated from Hot pepperfruit (*Capsicum annuum* L.). J. Agric. Food Chem. 53: 1750-1756.

Meda A, Lamien CE, Romito M, Millogo J, Nacoulma OG (2005). Determination of the total phenolic, flavonoid and proline contents in Burkina Fasan honey, as well as their radical scavenging activity. Food Chem., 91: 571- 577.

Minotti G, Aust SD (1987). An investigation into the mechanism of citrate-Fe^{2+}-dependent lipid peroxidation. Free Radical Biol. Med. 3: 379- 387.

Neuhouser ML, (2004). Dietary flavonoids and cancer risk: evidence from human population studies. Nutr. Cancer, 50(1): 1- 7

Oboh G, Akindahunsi AA (2004). Change in the ascorbic acid, total phenol and antioxidant activity of sun-dried commonly consumed green leafy vegetables in Nigeria. Nutr. Health, 18: 29-36.

Oboh G, (2005). Effect of blanching on the antioxidant property of some tropical green leafy vegetables. Lebensmittel Wissenschaft und Technologie. 38: 513-517

Oboh G, Puntel RL, Rocha JBT (2007a). Hot pepper (*Capsicum annuum,* Tepin and *Capsicum chinese,* Habanero) prevents Fe^{2+}-induced lipid peroxidation in Brain: *in vitro*. Food Chem. 102: 178-185.

Oboh G, Rocha JBT (2007b). Polyphenols in red pepper [*Capsicum annuum var. aviculare* (Tepin)] and their protective effect on some pro-oxidants induced lipid peroxidation in brain and liver- *In vitro.* Eur. Food Res. Technol., 225: 2

Oboh G, Raddatz H, Henle T (2008a). Antioxidant properties of polar and non-polar extracts of some tropical green leafy vegetables. J. Sci. Food Agric. 88: 2486-2492.

Oboh G, Rocha JBT (2008b).Water extractable phytochemicals from *Capsicum pubescens* (tree pepper) inhibit lipid peroxidation induced by different pro-oxidant agents in brain: *In vitro* European Food Res. Technol. 226: 707-713

Okwu DE, Morah FNI (2004). Mineral and nutritive value of *Dennettia tripetala* fruits. Fruits, 59: 437-442.

Okwu DE, Morah FNI, Anam EM (2005). Isolation and characterization of phenanthrenic alkaloid uvariopsine from Dennettia tripetala fruits. J. Med. Aromatic Plant Sci. 27: 496-498.

Oyaizu M (1986). Studies on products of browning reaction: antioxidative activity of products of browning reaction prepared from glucosamine. Jpn. J. Nutr. 44: 307-315.

Puntel RL, Nogueira CW, Rocha JBT (2005). Krebs cycle intermediates modulate thiobarbituric reactive species (TBARS) production in Rat Brain *In vitro*. Neurochem. Res. 30(2): 225-235.

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C (1999). Antioxidant activity applying an improved ABTS radical cation decolorisation assay. Free Rad. Biol. Med. 26: 1231-1237.

Singleton VL, Orthofer R, Lamuela-Raventos RM (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Cioalteau Reagent. Methods Enzymol. 299: 152-178.

Sun J, Chu Y, Wu X, Liu R (2002). Antioxidant and antiproliferative activities of common fruits. J. Agric. Food Chem. 50: 7449-7454 .

Wolfe KL, Liu RH (2008). Structure-Activity Relationships of Flavonoids in the Cellular Antioxidant Activity Assay. J. Agric. Food Chem. 56: 8404-8411.

Zar JH (1984). Biostatistical analysis. USA: Prentice-Hall, Inc., p. 620.