Pak. J. Sci. Ind. Res. 2010 **53** (2) 76-80

Antioxidant Properties of *Telfairia occidentalis* as Affected by the Market Storage Method in Nigeria

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(received November 11, 2009; revised February 12, 2010; accepted February 15, 2010)

Abstract. The effect of market storage methods in Nigeria on the antioxidant properties of *Telfairia occidentalis* was assessed over a period of 96 hours with respect to the vitamin C, total phenol and phytate contents, as typified by their reducing power and free scavenging ability. *T. occidentalis* had a phytate content of 28.83 mg/100 g and there was no significant (P>0.05) difference in the phytate content in the first 24 h of storage but significantly (P>0.05) reduced at the end of the storage period of 96 h (26.74 mg/100 g) with 7.24% loss. Vitamin C content reduced significantly (P>0.05) as the storage period increased with a very high percentage loss (81.58%) at the end of the storage period. The vegetable had 2.78 mg GAE/100 g total phenol and was slightly reduced but not significant (P>0.05) during the first 24 h of storage. *T. occidentalis* had scavenging ability > 90%, which significantly (P>0.05) decreased as the storage period increased (57.47% loss at 100% conc. and 56.28 % loss at 50% conc.).

Keywords: *Telfairia occidentalis*, vitamin C, total phenol, phytate, storage reducing power, scavenging ability

Introduction

Free radicals are highly reactive chemical substances such as peroxide, hydroxyl radical, singlet oxygen etc. that travel around in the body and cause damage to the body cells (Alia *et al*., 2003). Antioxidants are powerful free radical scavengers in the body. Antioxidants are believed to play a very important role in the body defence system against reactive oxygen species (ROS), which are the harmful by-products generated during normal cell aerobic respiration (Gutteridge and Halliwell, 2000). Antioxidant nutrients (found in foods) soak up all the excess energy that these free radicals have, turning them into harmless particles or waste products that can be removed (Oboh, 2005). Increasing intake of dietary antioxidants may help to maintain an adequate antioxidant status, therefore, the normal physiological function of a living system (Kaur and Kapoor, 2001; Record *et al.*, 2001). Regular consumption of fruits and vegetables has always been associated with health benefits. Fruits and vegetables contain a wide variety of biologically active, non-nutritive phytochemicals which impart health benefits beyond basic nutrition (Gupta and Prakash, 2009; Oomah and Mazza, 2000). Researchers have estimated that every serving increase in fruit and vegetable consumption reduces the risk of cancer by 15%, cardiovascular disease by 30% and mortality by any cause by 20% (Gupta and Prakash, 2009). This is often attributed to different antioxidant components in fruits and vegetables such as ascorbic acid, vitamin E, carotenoids, lycopenes, polyphenols and other phytochemicals (Prior and Cao, 2000). Vegetables play significant role in human nutrition, specially as a source of vitamins (A, B, C, E), minerals and dietary fibre (Aletor and Adeogun, 1995).

Leafy vegetables are important items of diet in many Nigerian homes (Mepba *et al.*, 2007). They are valuable sources of nutrients specially in rural areas where they contribute substantially to protein, mineral, vitamins, fiber and other nutrients which are usually in short supply in daily diets (Mosha and Gaga, 1999). They also add flavour, variety, taste, colour and aesthetic appeal to what would otherwise be a monotonous diet (Mepba *et al.*, 2007). Vegetables are in abundance shortly after the rainy season but become scarce during the dry season during which cultivated types are used. Some eventually find their way to urban markets (Mepba *et al.*, 2007). In Nigeria, during the season of abundance of vegetables, the market women do not always sell all their vegetables on the day of harvest; it has to be preserved for 24 h or more in order for them to break even financially. The modern preservation method of refrigeration and controlled/modified atmosphere are not available to these market women in Nigeria, so they have to design their own methods of preserving their produce. Much work has been done on the phytochemical content of vegetables but there is paucity of information on the effect of storage on the phytochemical content of vegetables. The aim of this study is, therefore, to determine the effect of market

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vegetable storage method in Nigeria on the antioxidant-phyto-constituents of *Telfairia occidentalis*.

Materials and Methods

Freshly harvested *Telfairia occidentalis* (Ugwu) was obtained from a farm in Rufus Giwa Polytechnic, Owo. The vegetable was subjected to market condition and stored using market storage method. They were displayed inside a plastic basin; at 6 pm, they were sprinkled with water, placed inside polypropylene bag and left outside at room temperature of 27±1 °C. This process was carried out for 96 h; sampling and antioxidant determination were carried out after 24 h and 96 h of storage.

Sample preparation. The edible portion of the vegetable (50 g) was separated, washed, drained completely, chopped, sundried and analyzed for phytate, ascorbic acid, total phenols, and antioxidant activities. The extracts were prepared in duplicate and all analysis was carried out in triplicate.

Sample analysis. The method of Maga (1982) was used for phytate determination. To the sample (2.0 g), 100 mL of 2% concentrated hydrochloric acid was added. The sample was soaked for 3 h, and then filtered through Whatman # 43 filter paper. 50 mL of the filtrate was placed in 250 mL beaker and 107 mL of distilled water was added to give proper aci-dity. 10 mL of 0.3% ammonium thiocyanate solution was added as an indicator and the solution was titrated with standard iron (III) chloride solution which contained 0.00195 g iron/mL. The equivalence point was slightly brownish-yellow which persisted for 5 min. Phytate content was expressed as the percentage (%) phytate in the sample. Vitamin C content was determined by AOAC (1990) method. 5 g of the sample was extracted with 100 mL H_2O and 25 mL of 20% glacial acetic acid was added to 10 mL of the sample extract and titrated against standardized 2,6 dichloroindophenol (0.05 g / 100 mL) solution. Total phenol was determined by mixing 0.2 mL phenolic extract (0.2 g of *T. occidentalis* extracted with 20 mL 70% acetone) with 0.8 mL Folin-Ciocalteau reagent and 2 mL of 7.5% sodium carbonate. The mixture was diluted to 7 mL with distilled water and the absorbance was measured after 2 h at 765 nm; the result was calculated as gallic acid equivalent. (Iqbal *et al.,* 2005). The reducing property was determined by assessing the ability of the sample extract to reduce $FeCl_3$ solution as described by Pulido *et al.* (2000). Briefly, appropriate dilutions (0-1.0 mL) were mixed with 2.5 mL of 200 mm sodium phosphate buffer (pH 6.6) and 2.5 mL of 1% potassium ferricyanide. The mixtures were incubated at 50 ºC for 20 min. Thereafter, 2.5 mL of 10% trichloroacetic acid was added and subsequently centrifuged at 650 rpm for 10 min. Then 5 mL of the resulting supernatant was mixed with equal volume of water and 1 mL of 0.1% ferric chloride. The absorbance was taken at 700 nm against a reagent blank. Increased absorbance of the reaction mixture indicated increased reducing power. Free radical scavenging activity using 1, 1-diphenyl-2-picryl hydrazyl (DPPH) as described by Singh *et al.* (2002) Different concentrations of the methanolic extract were taken in different test tubes and the volume was made to 1 mL with methanol. 4 mL of 0.1 mM methanolic solution of DPPH was added. The tubes were shaken vigorously and allowed to stand for 20 min at room temperature. A control was prepared as above without the sample and methanol was used for base line correction. Changes in absorbance of samples were measured at 517 nm. Free radical scavenging activity was expressed as percentage inhibition and was calculated using the following formula:

Free radical scavenging activity (%) = (control OD – sample OD) / control OD × 100

Statistical analysis. The results were statistically verified on the basis of analysis of variance, using SAS (2002). Mean separation was carried out where there was significant differences using Duncan multiple range test procedure as described in the SAS soft ware. Significance was accepted at $P>0.05$.

Results and Discussion

Consumers usually purchase a fresh produce driven by their visual appearance, while other components of quality such as texture and aroma make the consumers to re-purchase the same produce the next time (Kader, 2001). Inadequate storage conditions may negatively affect the produce quality. A produce badly stored may have a good visual appearance but altered taste or aroma. The optimal temperature and relative humidity during storage may help to reduce the degenerative processes that occur in vegetables during the postharvest stages (Kader, 2001). This work was carried out to estimate the antioxidant properties of the vegetable, *Telfairia occidentalis* kept for 96 h under market storage conditions.

Phytate has been shown to have anticancer and antioxidant activity. It forms an iron chelate that suppresses lipid peroxidation by blocking iron-driven hydroxyl radical generation (Oboh, 2006). Phytate content of the vegetable is presented in Table 1; it shows that *T. occidentalis* had a phytate content of 28.83 mg/100 g. This value was lower when compared to the values reported for the same vegetable *T. occidentalis* as 48.8 mg/100 g for leaves harvested 12 weeks after planting and 84.4 mg/100 g for leaves harvested 50 weeks after planting (Akwaowo *et al.,* 2000). This variation may be due to the environmental factors. Phytate content is also low as com-

pared to other vegetables (Udosen and Ukpanah, 1993). There was no significant (P>0.05) difference in the phytate content of the vegetable in the first 24 h of storage but content significantly (P>0.05) reduced at the end of the storage period of 96 h (26.74 mg/100 g) with 7.24% loss. This reduction of phytate in storage is in agreement with the findings of Hernandez-UnZon and Ortega-Delgado (1989) of a decrease of 4% in phytic acid of stored common bean seeds (*Phaseolus vulgaris* L) under hermetic conditions. The reduction in phytate could be attributed to the action of phytase which hydrolytically cleaves and frees the bound phosphorus from the phytic acid molecule, liberating calcium and magnesium cations at the same time.

Vitamin C is required for the prevention of scurvy and maintenance of healthy skin, gums and blood vessels and acting as an antioxidant, it reduces the risk of arteriosclerosis, cardiovascular diseases and some forms of cancer (Lee and Kader, 2000). Vitamin C content of stored *T. occidentalis* as shown in Table 1 (58.1 mg/100 g), was considerably higher than the values reported for some commonly consumed sundried tropical green leafy vegetables in Nigeria (Oboh and Akindahunsi, 2004), but low in comparison to the value of 101.36 mg/100 g of *Trigonella foenum graecum* (Gupta and Prakash, 2009). Vitamin C content of the vegetable was significantly (P>0.05) reduced as the storage period increased with a percentage loss of 81.58% at the end of the storage period. The decrease in Vitamin C content during storage agrees with the observation of Nwufo (1994) of decrease in chlorophyll and ascorbic acid contents of leafy vegetables during storage. The loss in ascorbic acid has also been reported in the study of four different vegetables, with okra having the least and spinach the highest loss during storage (Giannakourou and Taoukis, 2003). Tulio *et al.* (2002), analyzing the effects of storage temperature on the postharvest quality of jute leaves reported that the ascorbic acid content declined at all storage temperatures with the increase of storage period. Oladele and Aborisade (2009) also reported decrease in vitamin C content of Indian spinach (*Basella rubra* L) during storage. However, it has been noted that when reporting vitamin C levels, many workers did not take into consideration dehydroascorbic acid (DHA). In many horticultural crops, DHA represents less than 10% of total vitamin C but DHA tends to increase during storage (Wills *et al.*, 1984). Vitamin C is the most sensitive to destruction when the commodity is subjected to adverse handling and storage conditions. Losses are enhanced by extended storage, higher temperature, low relative humidity, physical damage and chilling injury (Lee and Kader ,2000). The loss in vitamin C may be the result of the activity of the enzyme ascorbate oxidase, proposed to be the major enzyme responsible for enzymatic degradation of ascorbic acid.

Table 1. Effect of storage time on the antioxidant phytochemicals of *Telfairia occidentalis* (Ugwu)

Storage period (hours)	Vitamin C (mg/100 g)	Total phenol (mg GAE/100 g)	Phytate (mg/100 g)
0	58.1±0.3[a]	2.78±0.2[a]	28.83±0.1[a]
24	32.9±0.1[b]	2.56±0.2[a]	28.80±0.1[a]
96	10.7±0.1[c]	2.08±0.1[b]	26.74±0.1[b]
Loss (%)	81.58	25.17	7.24

Values represent mean of triplicate analysis; values with the same letter in a column are not significantly different (P> 0. 05).

The antioxidant activity of phenolic compounds is mainly due to redox properties, which allow them to act as reducing agents, hydrogen donors, singlet oxygen quenchers, heavy metal chelators and hydroxyl radical quenchers (Kaur and Kapoor 2002). Value of total phenolics (Table 1), in the vegetable (2.78 mg GAE/100 g) agrees with the value reported for the same vegetable but lower to the value reported for *Amaranthus cruentus* and *Ocimum gratissimum* (Oboh and Akindahunsi, 2004). There was slight reduction in the total phenol content of the vegetable but not statistically significant (P>0.05) in the first 24 h of storage; however, it reduced significantly (P>0.05) at the end of the storage period of 96 h with 25.17% loss. Ose *et al.* (1997) also reported that the total phenol contents of water convolvulus, *Calendula arvensis*, leaves decreased during storage at low temperature; this may be attributed to the fact that phenols are susceptible to oxidation by the enzyme phenolase which converts them to quinones. These compounds are often extremely reactive and therefore short lived (Kays, 1991).

The antioxidant effect exponentially increases as a function of the development of the reducing power, indicating that the antioxidant properties are concomitant with the development of reducing power (Oyaizu, 1986). Reductones are believed not only to react directly with peroxides but also prevent peroxide formation by reacting with certain precursors (Gupta and Prakash, 2009). The results revealed (Fig. 1) a significant (P>0.05) reduction in the reducing power of *Telfairia occidentalis* as the storage period increased, with 41.67% reducation at the end of the storage period of 96 h, agreeing with the decrease in the total phenol content, which could be attributed to the decrease in the proton donors necessary to react with the precursors as a result of the storage.

1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical is a stable, free radical and accepts an electron or hydrogen radical to become a stable diamagnetic molecule. The methodology involves reaction of specific compounds or extracts with DPPH in methanol solution. In the presence of hydrogen donors, DPPH

is reduced and a free radical is formed from the scavenger. The reaction of DPPH is monitored by the decrease in the absorbance of its radical at 517 nm, but upon reduction by an antioxidant, the absorption disappears (Gupta and Prakash, 2009). The effect of storage on the free radical scavenging ability of *T. occidentalis* (Fig. 2) shows that *T. occidentalis* had scavenging ability > 90% and that there was a significant (P>0.05) decrease in the free radical scavenging ability of *T. occidentalis* as the storage period increased (57.47% loss at 100% concentration and 56.28 % loss at 50% concentration). This reduction in the scavenging ability may be attributed to the decrease observed in the vitamin C and total phenol contents and reducing power as discussed earlier.

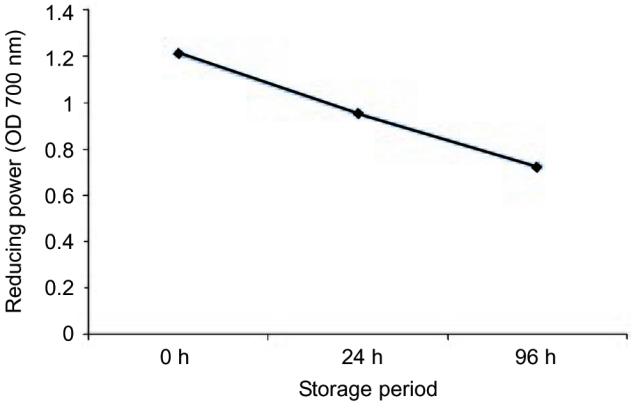

Fig. 1. Effect of storage time on reducing power of *Telfairia occidentalis* (Ugwu).

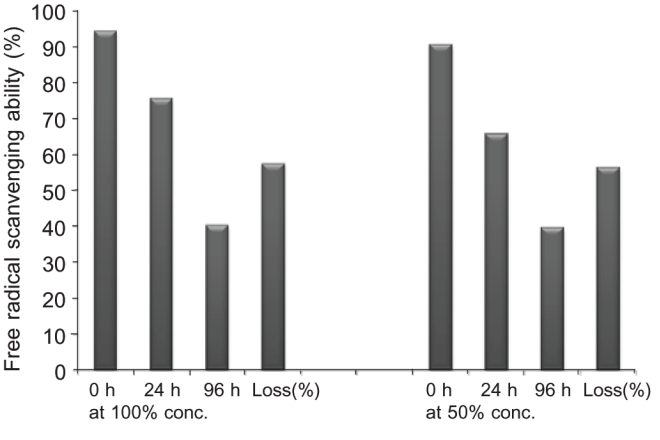

Fig. 2. Effect of storage period on the free radical scavenging ability of *Telfairia occidentalis* (Ugwu).

Conclusion

The results showed that the antioxidant contents of *T. occidentalis* reduced during storage though during the first 24 h of storage, there was no significant (P>0.05) difference in the total phenol and the phytate contents. Consequently, the antioxidant activities were also reduced. Considering this available storage method, vegetables should be sold within 24 h of harvesting in Nigeria.

Acknowledgement

The authors thank specially Dr. G. Oboh of the Federal University of Technology, Akure, for providing the facilities of his personal laboratory, materials and chemicals during the course of this work and also for his critical comments.

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