

Influence of Cooking Methods on Phenolic and Antioxidant Activity of Yams (*Dioscorea Spp.*) and Cocoyam (*Xanthoma Maffs [Scoth]*) Tuber Extracts

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ABSTRACT— The influence of cooking methods (boiling, roasting, deep-fat frying and oven-drying) on phenolic and antioxidant activity of yams (*Dioscorea cayenensis*, *Dioscorea dumetorum*, and *Dioscorea bulbifera*) and cocoyam (*Xanthosoma maffa* (Scoth)) were evaluated using Folin-Ciocalteu's reagent, Aluminum chloride, ABTS(2,2'-azinobis-(3-ethylbenzothiazoline-6-sulponic acid)diammonium salt, DPPH(2,2-diphenyl-1-picrylhydrazyl) and ORAC (oxygen radical absorbance capacity). The correlation coefficient analysis was also determined. Almost all the cooking methods significantly increased ($P < 0.05$) the polyphenols and flavonoids content of the yam (*Dioscorea*) tuber variety extracts, but with losses in *X. maffa* (Scoth) when compared to their fresh samples. High losses in ABTS radical scavenging activity was observed in *X. maffa* (Scoth) and *D. dumetorum* after all the cooking methods. Analysis of DPPH radical scavenging activity showed strong losses after different cooking methods in some of the tuber varieties. High ORAC value was observed in the boiled method in *D. cayenensis* and *D. bulbifera* while other methods showed significant ($P < 0.05$) losses. A high correlation coefficient was observed between total flavonoids (TF) and ABTS ($R^2 = 0.839$) in *D. cayenensis*, between total polyphenols (TP) and ORAC ($R^2 = -0.917$), TF and ORAC ($R^2 = -0.862$) in *D. dumetorum*, between TP and ORAC ($R^2 = -0.852$), TF and ORAC ($R^2 = -0.930$) in *D. bulbifera*, and between TF and ABTS ($R^2 = 0.957$), TF and DPPH ($R^2 = 0.825$) in *X. maffa* (Scoth). Significant correlation coefficient was also observed in the different cooking methods. Polyphenol and especially flavonoid were found to be major contributors to their antioxidant activities.

Keywords--- Phenolic, Antioxidant activity, Cooking methods, Yam (*Dioscorea spp.*), Cocoyam (*X.maffa* (Scoth)).

1. INTRODUCTION

Aerobic metabolism in the body generates oxidative molecules, free radicals and reactive oxygen species which are needed to maintain life processes. However, excess amount of these metabolites are harmful to health because they react readily with macromolecules and DNA, causing oxidative damage *in vivo* (Higuera-Clapara *et al*, 2006). Fruits and vegetables are regarded as the main contributors of free radical scavenging antioxidants (Jimenez-Monreal *et al*, 2009) for the prevention of oxidative damage to cells. Yams and cocoyam are mainly cultivated in the humid and sub-humid tropics and constitutes an important food source for local populations. They are grouped along with vegetables that are reported to have more than 70% antioxidant capacity against free radical actions (Kaur and Kapoor, 2002). Yam extracts contain hydrophilic antioxidant compounds like catechins, epicatechin, chlorogenic acid, leucoanthocyanins, anthocyanins and vitamin C (Ozo *et al*; 1984, Farombi *et al*; 2000, Bhanbari and Kawabata, 2004), and lipophilic antioxidants like carotenoids and α -tocopherol (Champagne *et al*, 2010). Yam extracts are reported to exert physiological effects and improve human health, acting as antioxidant to quench free radical actions through proton donation. Thus, yam phenolic were found to contain antioxidant activity, inhibits cancer, cardiovascular diseases, diabetes and aging related problems (Miyazawa *et al*, 1996; Chen *et al*, 2008; Adebayo *et al*, 2012). Apart from its health related benefits, yam and cocoyam can be a possible source of alternative food antioxidant to synthetic ones such as butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), and tertiary butylated hydroquinone (TBHQ) (Cornago *et al*, 2011), which can promote tumor apoptosis (Barlow, 1990). Yam tuber extracts also contain considerable amount of

organic acids especially succinic and citric acids which are contributors to sensory properties in vegetable products (Bhandari and Kawabata, 2004).

Traditional cooking methods of yams and cocoyam are mainly boiled, roasted, fried or pounded and eaten in association with vegetables and protein rich sauces (Udensi *et al.*, 2008). Cooking methods can greatly influence both the texture and nutritive value of yam and cocoyam tubers. Cooking softens the matrix and cellular tissues and facilitates the extraction of carotenoids and other non-nutrient phytochemicals with appropriate solvents (Rodriguez-Amaya, 1999). The gain or loss of phenolic compounds during cooking processes may vary with time and method, the cellular structure and granular sizes, isomerization and degradation caused by the action of polyphenol oxidase catalyzed reactions (Jung *et al.*, 2011). Phenolic compounds in yam and cocoyam undergo polyphenolic oxidase-catalyzed reactions to form a primary oxidation product called O-quinone, which combines with other components to form brown polymeric compounds (Ozo, 1985; Farombi *et al.*, 2000). Also, protein and amino acids in yam and cocoyam thermally reacts non-enzymatically with sugars to form brown-colored compound called the maillard reaction product. This product in association with extractible phenolic compounds is reported to increase antioxidant activity in yam tuber extracts (Adebayo *et al.*, 2012).

Antioxidant compounds acts in different ways in the oxidative sequence of lipid molecules. They can intercept singlet oxygen and prevent initiation reaction of oxidative degradation of lipids because of their ability to act against a wide range of free radical cations in multiple hydroxyl groups (Jimenez-Monreal *et al.*, 2009). Phenolic antioxidants from yam and cocoyam tuber extracts may therefore be important in the management of disease conditions attributed to free radicals, hence the steady supply of antioxidants from these sources can augment or boost the effect of enzymatic antioxidants (catalase, peroxidase, dismutase, glutathione) defense mechanisms in the body to prevent free radical mediated oxidative stress among yam and cocoyam consumers.

Because yam and cocoyam are consumed after being cooked, it is therefore important to know what happens to their phenolic and antioxidant activity during cooking processes like boiling, roasting, deep fat-frying, and oven-drying. The objective of this study are (a) to determine the phenolic content and antioxidant activity of cooked yams (*Dioscorea cayenensis*, *Dioscorea dumetorum*, *Dioscorea bulbifera*) and cocoyam (*Xanthosoma maffa* (Scoth), (b) to correlate phenolic compounds with antioxidant activities and (c) to evaluate the stability or bioavailability of phenolic in yams and cocoyam after thermal processing.

2. MATERIALS AND METHODS

Chemicals

Galic acid, Catechin hydrate, Folin-Ciocalteu's phenol reagent, AAPH (2,2'-azobis(2-amidinopropane)dihydrochloride), DPPH(2,2-diphenyl-1-picrylhydrazyl), ABTS (2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulponic acid) diammonium salt), and Fluorescein sodium salt were purchased from Sigma Chemical (MO, USA). Other chemicals used, namely, Acetone, Sodium nitrite, Aluminum chloride, Petroleum ether and Alcohol was of analytical grade.

Root and tuber samples

Three species of *Dioscorea* namely, *D. cayenensis* (yellow yam), *D. dumetorum* (trifoliate yam), *D. bulbifera* (aerial yam) and *Xanthosoma maffa* (Scoth) (yellow cocoyam) were collected from a yam farmer at Ututu, Abia State, Nigeria, after harvesting at full physiological maturity in January, 2012. They were without defects.

Sample preparation

Sample preparation and analysis was carried out at Nutrition Quality Laboratory, International Centre for Tropical Agriculture, Cali, Colombia (CIAT). Tuber samples were washed with deionized water and immediately peeled under water. After peeling, the samples were diced into 2cm cubes, filled with nitrogen gas and stored at – 70°C.

Processing/cooking methods

Each sample was divided into four portions of 100g each. Four final processing methods were chosen.

- i) **Boiling:** One hundred gram (100g) of *D. cayenensis*, *D. dumetorum*, *D. bulbifera*, *X. maffa* (Scoth) was boiled in deionized water (100mL) at 98°C for 15 minutes.
- ii) **Roasting:** One hundred grams (100g) of each sample was roasted in aluminum pot on top of electric oven at 150°C for 15 minutes.
- iii) **Frying:** One hundred gram (100g) of each sample was deep-fried in Oliosoya (vegetable oil), Team Foods, Colombia, for 10 minutes at 240°C.
- iv) **Oven – drying:** One hundred gram (100g) of each sample was oven- dried at 60°C for 24h (Thelco Precision Laboratory Oven, Thermo Scientific).

The cooked samples (4 x 4) were then lyophilized at -30°C for 48h (Labconco Corporation, Kansas City, Mo). After, the samples were pulverized into fine powder (Retsch Grindomix, USA), packaged with label in a cellophane bag and stored at -70°C (Ultra Freezer, Thermo Scientific) until use.

Extraction of phenolic compounds

A quantity of 0.5g of the ground powdered sample was mixed with 5 ml of the extracting solvent (acetone/deionized water 50:50 v/v), which was chosen from trials using different solvents in different proportions (water, ethanol, methanol, acetone). This was done to determine the extraction potency that was best and compatible with the solubility of phenolic compounds in the yams and cocoyam tuber matrix (result not shown). Mixing was done in a 50 ml BD Falcon tubes using Ultra Turax (1Ka T18 basic Staufen, Germany) for 10 seconds and then capped and re-mixed in a Vortex mixer (Fisher Scientific, USA) for 1 minute. Samples were then placed on a multi-purpose rotator (Barnstead International, USA) for 30 minutes at 600rpm and subsequently centrifuged at 4°C for 5 minutes, and at 6000rpm (Eppendorf Centrifuge 5804R Hamburg, Germany). Two (2) mL of sample extract was collected and stored in the dark at 4°C for the determination of total polyphenols, flavonoids and antioxidant activities.

Determination of total polyphenol

Total polyphenol (TP) content was determined following the method suggested by Jayaprakasha *et al*, (2001) using Folin-Ciocalteu reagent with a minor modification. In a 2ml Eppendorf tube, 780 μl deionized water, 20 μl sample extract, and 50 μl Folin-Ciocalteu reagent (1:1 v/v) with water were added and mixed. After 1 minute, 150 μl Sodium carbonate (0.2g/ml) was added, and the mixture was allowed to stand at room temperature in the dark for 1h. Then, 300 μl of the mixture was carefully introduced into a 96 well plate using Eppendorf micropipette. The absorbance was read at 750nm (μQuant , Biotech Instruments, USA). The total polyphenol concentration was calculated from a calibration curve, using Gallic acid (1mg/ml) as standard (200 – 1000mg/L).

Determination of total flavonoid

Total flavonoid (TF) content was determined using the method of Yong *et al*, (2008) with some modifications. In a 2ml Eppendorf tube, 660 μl deionized water, 80 μl sample and 30 μl Sodium nitrite (50mg/ml), and 30 μl Aluminum chloride (100mg/ml in methanol) were added and mixed. After 5 minutes, 200 μl of 1M Sodium hydroxide was added. Then, 300 μl of the mixture was transferred into the 96well plate and read at 500nm immediately. The total flavonoid concentration was calculated from a calibration curve using Catechin as standard (50 – 800mg/L).

Determination of antioxidant activity by DPPH

The ability to scavenge DPPH free radicals was determined according to the method of Brand-Williams *et al*, (1995) with some modifications. The stock solution was prepared by dissolving 24mg DPPH with 100mL methanol. It was stored at -20°C over-night. The working solution was obtained by mixing 10mL stock solution with 45mL methanol to obtain an absorbance of 1.1 ± 0.02 units at 517nm using the spectrophotometer (μQuant , Biotech Instruments, USA). Sample (yams and cocoyam) extracts (100 μL) were allowed to react with 1900 μL of the DPPH solution for 1h. Thereafter 300 μL of the reaction mixture was added into the 96well plate and read at 517nm (μQuant , Biotech Instruments, USA). The standard curve was linear between 150 – 500 μM Trolox. Results were expressed in mg Trolox Equivalent (TE/g) fresh weight. Additional dilution was needed if the DPPH value measured was over the linear range of the standard curve.

Determination of antioxidant activity by ABTS⁺ radical cation

For ABTS assay, the method of Arnao *et al*, (2001) was adopted with some modifications. ABTS radical cation (ABTS⁺) was produced by reacting 38.4mg ABTS and 6.6 mg potassium persulphate in 10mL of deionized water and allowing the mixture to stand in the dark at room temperature (about 29°C) for 12 – 16h before use. The ABTS⁺ stock solution was diluted with ethanol to obtain an absorbance of 1.1 ± 0.02 at 734nm. In the 96 well plate, 30 μL of sample extract or Trolox standard and 200 μL ABTS⁺ solution were added together and allowed to react for 6 minutes before taking absorbance at 734nm (μQuant , Biotech Instruments, USA). Data was expressed as Trolox Equivalent Fresh Weight (TE/gFW). The Trolox standard curve was linear between 50 – 400 μM Trolox.

Determination of antioxidant activity by oxygen radical absorbance capacity (ORAC)

The ORAC assay was performed as described by Huang *et al*, (2002). 130mg AAPH (2,2'-azobis(2-amidinopropane)dihydrochloride) was dissolved in 3mL PBS of 75mM (pH 7.4) to a final concentration of 153mM and made fresh daily. A fluorescein stock solution ($4 \times 10^{-3}\text{mM}$) was dissolved in 75mM PBS (pH 7.4) and stored. The fluorescein stock solution was diluted 1:1000 with PBS (pH 7.4). To the experimental 96 wells, 150 μL fluorescein diluted solution was added. In addition, blank wells received 25 μL of 75mM PBS (pH 7.4), while standards received 25 μL of trolox dilution and sample wells received 25 μL of sample extracts (diluted at 1:100). Reaction was initiated by adding 25 μL of AAPH reagent with shaking duration for 8 seconds. A Multi-Mode Micro-plate Reader, SynergyTM HT,

Biotech Instrument INC, USA, with injectors was used with 485/20nm excitation filter and a 530/25nm emission filter. The number of Kinetic cycles is 30 and Kinetic interval is 60 seconds.

ORAC values were calculated as described by Cao and Prior, (1999). The Area under the Curve (AUC) and the Net AUC of the standards and samples were determined using Equations (i) and (2), respectively. Results were expressed as Trolox Equivalent fresh weight (TE/g FW)

$$\text{AUC} = 0.5 + (R_2/R_1) + (R_3/R_1) + (R_4/R_1) \dots + 0.5 (R_n/R_1) \dots (1)$$

Where R_1 is the fluorescence reading at initiation of the reaction and R_n is the last measurement.

$$\text{Net AUC} = \text{AUC sample} - \text{AUC blank} \dots (2)$$

Statistical analysis

All tests were carried out in triplicate and means were separated using LSD at $P < 0.05$. Analysis of variance (ANOVA) and Pearson correlation coefficient analysis were performed by PROC.GLM in SAS (SAS, 2003).

3. RESULTS

3.1. Influence of cooking methods on polyphenol and flavonoid content of yams and cocoyam tuber extract.

Table 1A describes the influence of cooking methods on polyphenol content of the yams and cocoyam tuber extracts. Yam and cocoyam tuber varieties are usually consumed after being cooked.

Boiling. Boiled method increased the polyphenol content in *D. cayenensis* by 26 folds. *D. bulbifera* and *X. maffa* (Scoth) encountered significant losses ($P < 0.05$) of about 20 % and 17.8%, while *D. dumetorum* was unaffected by boiling with respect to their fresh samples.

Roasting. Roasted method produced significant increases ($P < 0.05$) in the total polyphenol content of the yam varieties but with decrease in the cocoyam tuber from their fresh samples (Table 1A). The increases ranged from 57 folds in *D. cayenensis*, 1.67 folds in *D. dumetorum*, 2.5 folds in *D. bulbifera* and a loss of 8.2% in *X. maffa* (Scoth) respectively.

Deep fat-frying. Similarly, there were significant increases in the polyphenol content after deep fat-fried method for the yam tubers and a loss in the cocoyam tuber with respect to their fresh samples. *D. cayenensis* increased by 38 folds, *D. dumetorum* increased by 1.6 folds, *D. bulbifera* increased by 2.3 while *X. maffa* (Scoth) decreased by 25%.

Oven-drying. This cooking method produced increased polyphenol values in the yam species and in *X. maffa* (Scoth). The increases were 29 folds in *D. cayenensis*, 3.8 folds in *D. dumetorum*, 3.6 folds in *D. bulbifera*, and 1.3 folds in *X. maffa* (Scoth) (Table 1A) when compared to their fresh values.

Boiled, roasted, and fried methods produced losses in the phenolic content of *X. maffa* (Scoth), but had significant increases in the yam *Dioscorea* species.

In Table 1B, cooking methods significantly increased ($P < 0.05$) the flavonoid content of the yam tuber extracts but incurred losses in *X. maffa* (Scoth) when compared to their fresh samples.

Boiling. Boiled method increased the flavonoid of *D. cayenensis* by 32.5 folds, *D. dumetorum* increased by 4.29 folds, *D. bulbifera* increased by 1.23 folds, and 69% loss in *X. maffa* (Scoth).

Roasting. Roasted method also increased the flavonoid content of the yams as follows; *D. cayenensis* (74.2 folds), *D. dumetorum* (4.49 folds), *D. bulbifera* (2.1 folds), and 48% loss in *X. maffa* (Scoth) respectively.

Deep fat-frying. In this method, flavonoid increased in *D. cayenensis* by 56.8 folds, in *D. dumetorum* by 7.85 folds, in *D. bulbifera* by 1.9 folds, and a loss of 65% in *X. maffa* (Scoth) when compared to their respective fresh samples.

Oven-drying. Oven-dried method increased the flavonoid of the yam tubers as follows; *D. cayenensis* (14.55 folds), *D. dumetorum* (14 folds), *D. bulbifera* (2.8 folds), and a loss of 38% in *X. maffa* (Scoth) (Table 1B). The best results were obtained in roasting, oven-drying, deep fat-frying, and lastly boiling methods in *D. cayenensis*, *D. dumetorum*, and in *D. bulbifera*. However, *D. dumetorum* had the lowest flavonoid content in almost all the processing methods.

The present study reveals that cooked yams and cocoyam tubers contain considerable amount of phenolic compounds in varying concentrations.

3.2. Antioxidant activity and reducing power of processed yam and cocoyam tuber extracts

Radical scavenging potentials of the yam extracts (*D.cayenensis*, *D.dumetorum*, *D.bulbifera*) and cocoyam (*X. maffa* (Scoth)) were evaluated by ABTS, DPPH and ORAC assays, and the results are expressed as Trolox Equivalent and are shown in Tables 2A, 2B and 2C

In Table 2A, the cooking methods produced significant increases ($P<0.05$) in ABTS radical scavenging activity for *D.cayenensis* and *D.bulbifera*. However, *X. maffa* (scoth) produced losses in all processing methods, while *D. dumetorum* decreased in other cooking methods with the exception of oven-dried cooking.

Boiling. The ABTS radical scavenging activity of boiled *D.cayenensis* increased by 5-folds, *D. dumetorum* and *X. maffa* (Scoth) lost by 2.8-folds and 7-folds.

Roasting. This cooking method produced significant ABTS antioxidant activity increases in *D.cayenensis* and *D.bulbifera* but incurred losses in *D.dumetorum* and *X.maffa* (Scoth). While *D. cayenensis* and *D. bulbifera* increased by 16.5 and 2.6-folds, *D. dumetorum* and *X. maffa* (Scoth) lost by 1.3 and 3.3folds respectively.

Deep fat-frying. In this process, the ABTS values for *D. cayenensis* and *D. bulbifera* significantly increased ($P<0.05$) while that for *D. dumetorum* and *X. maffa* (Scoth) incurred losses. *D. cayenensis* and *D. bulbifera* increased by 9.4 and 2.3 folds, while *D. dumetorum* and *X. maffa* (Scoth) decreased by about 1.25 and 2.98 folds respectively (Table 2A).

Oven-drying. In this cooking method, the ABTS scavenging activity of *X. maffa* (Scoth) incurred a loss of about 1.8 folds, while those of *D. cayenensis*, *D. dumetorum*, and *D. bulbifera* increased by 8.7, 1.9, and 1.65 folds respectively (Table 2A).

Roasted *D.cayenensis* and *D. bulbifera* had the strongest ABTS radical scavenging activity as against other cooking methods. The ABTS of *D.dumetorum* was strong in the oven-dried method while that of *X. maffa* (Scoth) was weakest in almost all the processed methods.

The result of the DPPH radical scavenging activity of *D. cayenensis*, *D. dumetorum*, *D. bulbifera* and *Xanthosoma maffa* (Scoth) is presented in Table 2B. *D. cayenensis* exhibited the highest DPPH free radical scavenging activity in almost all the cooking methods, especially in roasted and oven-dried methods. The processing methods indicated weak DPPH free radical scavenging activity in other tuber samples especially in *D. dumetorum* and *X. maffa* (Scoth).

Table 2C shows the effect of processing treatments of oxygen radical absorption capacity fluorescein (ORAC – FL) assay on the yams/cocoyam tuber extracts. The strongest ORAC-FL radical scavenging activity was observed in the boiled process. Other processing methods produced some degree of weakness with losses ranging from 40-65% in *D. cayenensis*, 23-78% in *D. dumetorum*, 32-63% in *D. bulbifera*, and 6-80% in *X. maffa* (Scoth) respectively. However, oven-dried method exhibited the weakest ORAC potential in all the processed methods.

4. DISCUSSIONS

Phenolic compounds (mainly polyphenol and flavonoid) are plant metabolites derived by ingestion of plant foods by animals. They are potent antioxidants that can scavenge free radicals in the body through the donation of hydrogen atom or proton from the aromatic ring of the phenolic compounds to stabilize the free radical action caused by reactive oxygen species. Reactive oxygen species (ROS) are known to cause not only oxidative stress and degenerative diseases in animals in situations of deficient antioxidant, but also causes oxidation of food products thereby diminishing organoleptic properties. To determine the antioxidant efficacy, antioxidant activity of the yams/cocoyam extracts was measured as the sum of the diverse antioxidants contained in the yams/cocoyam tuber samples using different antioxidant activity methods (Tabart *et al*, 2009). This is because the complex nature of phytochemicals antioxidants from plant sources and their multiple reaction mechanisms and characteristics cannot be performed accurately by one single assay (Chu *et al*, 2000; Du *et al*, 2009). This informs the use of DPPH, ABTS and ORAC assays to determine the antioxidant activity of the yams/cocoyam phenolic extracts in order to understand their free radical scavenging potentials *in vitro*.

From the results, there were wide variations in the phenolic content and antioxidant scavenging activity of the different varieties of the yams/cocoyam tuber extracts subjected to different cooking methods. This may be due to a number of factors which includes among others variety, cellular structures and granular sizes, phenolic content, temperature and retention after different cooking methods (Makris and Rossider, 2001; Pedraza – Charerri *et al*, 2006), extraction solvents and analytical methods. The cooking methods either increased or reduced the phenolic and antioxidant activity of the yams/cocoyam tuber extracts. Roasted method gave the highest polyphenol value in *D.cayenensis*, oven-dried method gave the highest phenolic result in *D. dumetorum*, *D.bulbifera* and *X. maffa* (Scoth) while boiled method produced the lowest phenolic values in all yam tuber samples (Table 1A). Roasting, oven-drying and frying methods produced strong ABTS radical scavenging activity in some yam extracts while boiling process showed weak strength. This result agrees with the high and remarkable ABTS antioxidant capacity reported in some processed yam species (Farombi *et al*, 2000; Hsu *et al*, 2003). Generally, the degree of losses/weak strength was greatest in the boiled method for both DPPH and

ABTS. Although DPPH and ABTS assays are indicators of free radical scavenging ability against hydroxyl radical through hydrogen atom transfer mechanism, the DPPH values was found to be lower than those obtained in ABTS assay.

There were the possibilities of the liberation of antioxidant components in the cooking methods due to the thermal destruction of cell walls and sub-cellular compartments of the yams/cocoyam varieties, the destruction of polyphenol oxidase or thermal inactivation of oxidative enzymes that can hinder the oxidation capacity of antioxidants, the production of non-nutrient antioxidant or the formation of compounds such as Maillard reaction products possessing antioxidant activity (Jimenez-Monreal *et al.*, 2009). These reactions can contribute to the increased potentials of phenolic and antioxidant activity obtained in the heat processed samples especially in the roasted, oven-dried and fried methods, as was the case in the thermal processing of sweet corn, tomato, and other vegetables (Nindo *et al.*, 2003), processed Chinese purple yam (Fang *et al.*, 2011), amala browned yam flour diet (Farombi *et al.*, 2000). Besides, heat can break the glycosidic bounds of some yam tubers, especially in *D. cayenensis* and *D. bulbifera*, which have large granular sizes ranging from 20-31 μ m (Amani *et al.*, 2005) and release the phenolic sugar glycosidic bounds, which may have reacted better with the assays to increase the antioxidant potentials in these yam tuber variety extracts. Most phenolic compounds are unstable during cooking processes due either to hydrolysis or oxidation. This factor may have resulted in the loss of phenolic and antioxidant activity in the cooked *X. maffa* (Scoth) due to the thermal degradation of the antioxidant compounds because of its small granular sizes (about 6-10 μ m) which enabled easy and fast heat and mass transfer mechanisms to cause the degradation of the antioxidant constituents.

Boiling process produced low values of antioxidant activity in the yams/cocoyam species except in the ORAC-FL assay. These losses may again be due to lixiviation phenomenon in which the phenolic compound and vitamin C that contributes to antioxidant activity was solubilized into the cooking water as was reported in cooked vegetables (Jimenez-Monreal *et al.*, 2009). In the case of high ORAC result from the boiled process, the cellular tissue and matrix of the yams/cocoyam tubers may have softened to enable the release of non-phenolic antioxidant compounds into the sample extract, thus yielding high ORAC values when compared to other processing methods as was evidenced in this study. This agrees with the findings of Teow *et al.* (2007) who opined that ORAC assay being more sensitive, can measure the combined lipophilic (carotenoids and α -tocopherol) and hydrophilic (phenolic and ascorbic acid) antioxidants in a food system for a higher result than other free radical scavenging measurement assays. Roasted, oven-dried and fried methods retained and/or increased active phenolic compounds in some yam tubers resulting to high antioxidant activity values. These methods can cause a non-enzymatic complex reaction mixture of the Maillard products that is believed to increase the phenolic and antioxidant activity potentials of yam tuber extracts (Adebayo *et al.*, 2012). Snacks from roasted and fried yam/cocoyam is not only a popular breakfast food in Nigeria but also the best known methods of processing yam and cocoyam products that offers good flavor and taste contributed by the volatiles during roasting and frying process.

In the frying process using vegetable oil, the yams/cocoyam tubers dehydrated but increased its content of α -tocopherol and polyphenol (Jimenez-Monreal *et al.*, 2009) which in turn increased the antioxidant potentials. Depending on the yam and cocoyam variety and assay, roasting, oven-drying, frying and boiling methods produced gains or losses in the phenolic content and antioxidant activity when compared with their fresh samples. The phenolic and antioxidant activity of these processed yams/cocoyam tuber extracts compared with the values of Gala apples (total phenolic 118mgGAE/100g) and flavonoids (62.0mgCE/100g), ABTS 205mgTEAC/100g) (Kim *et al.*, 2003).

This study identified the best cooking methods for yams and cocoyam focusing on the bioavailability of their phenolic compounds and radical scavenging activity potential with its health-related implications. Although there were losses in some processing/cooking methods as well as varietal differences, boiling produced the lowest phenolic content and weakest antioxidant activity except in the ORAC-FL assay. On the other hand, *X. maffa* (Scoth) suffered the greatest processing losses when compared to the fresh sample result. Roasted, fried and oven-dried methods obtained more bioavailable phenolic compound and stronger antioxidant activity than the boiled method.

Correlations between the phenolic compounds and antioxidant activity of the yam/cocoyam extracts

A high significant correlation coefficient between flavonoids and ABTS ($R^2=0.839$, $P<.0001$) was obtained in *D. cayenensis*, while total polyphenol fairly correlated with DPPH ($R^2 = 0.6502$, $P = <.05$) (Table 3A). ABTS⁺ assay correlated with DPPH assay ($R^2 = 0.896$, $P = <.0001$), and DPPH correlated with ORAC ($R^2 = -0.800$, $P = 0.0003$) (Table 3A).

In Table 3B, there was a high significant correlation between the phenolic and antioxidant activity for the extract of *D. dumetorum*. Total Polyphenol (TP) and total flavonoids (TF) correlated highly with ORAC ($R^2 = -0.919$, $P = <.0001$ and $R^2 = -0.8619$, $P = <.0001$). This result show fair correlation between phenolic potentials with ABTS and a high correlation with ORAC, which may have been contributed from the interference of non-phenolic compounds in the yams/cocoyam tubers extract with ORAC assay. Table 3C exhibited a high correlation coefficient between total phenolic and antioxidant activity for *D. bulbifera*. Total polyphenol and total flavonoids correlated highly with ORAC ($R^2 = -0.852$, $P = <.0001$) and ($R^2 = -0.930$, $P = <.0001$).

In table 3D (*X. maffa* (Scoth), the correlation coefficients between phenolic and antioxidant activity were high. Total flavonoids correlated highly with ABTS ($R^2 = 0.9572$, $P = <.0001$) and with DPPH ($R^2 = 0.8246$, $P = 0.0002$). On assay methods, correlation was between ABTS and DPPH ($R^2 = 0.8176$, $P = 0.0002$).

The implication of this result is that phenolic compounds contributed significantly to the antioxidant activity in the yams and cocoyam sample tuber extracts.

In other reports, a high correlation coefficient between total phenolic and antioxidant activities were established in *Actinidia* fruits (Du *et al.*, 2009); Guava fruit extracts (Thaipong *et al.*, 2006); Plums (Kim *et al.*, 2003); Amaranth, Quinoa, Buckwheat and Wheat (Alvarez – Jubete *et al.*, 2010) and Vegetables (Ciz *et al.*, 2009).

On processing/cooking methods, a significant linear correlation was observed in all the cooking methods, especially in roasted method between total polyphenol and ABTS ($R^2 = - 0.8581$, $P = 0.0004$) and between total flavonoid and DPPH ($R^2 = 0.9325$, $P = <.0001$).

Boiled method exhibited the weakest polyphenol, flavonoids, ABTS and DPPH radical scavenging activity but produced the strongest ORAC radical scavenging activity in all the processed yams/cocoyam tuber extracts. Oven-dried method showed the weakest ORAC radical scavenging activity in all the processed sample tubers.

5. CONCLUSION

Phenolic content reported in this study exhibited a high contribution to antioxidant activity in the different yams/cocoyam tuber extracts. The antioxidant activity results emphasized the importance of phenolic compounds in antioxidant behavior of yams and cocoyam tuber extracts from Nigeria to scavenge reactive oxygen species (ROS), inhibit oxidative stress and promote the health of yam and cocoyam consumers by stimulating the immune system. The best cooking methods for phenolic bioavailability and strong antioxidant activity are roasted, oven-dried and fried methods. Although these results are quantitative, the effect of *in vitro* trials from yams phenolic extracts on carcinogenicity and hypoglycemia are well documented in literature.

6. ACKNOWLEDGEMENT

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Table 1A: Influence of cooking methods on polyphenols content of yams and cocoyam tuber extracts (mgGAE/100gDW).

Processing methods	<i>D. Cayenensis</i> (Yellow Yam)	<i>D. dumetorum</i> (Trifoliate Yam)	<i>D. bulbifera</i> (Aerial Yam)	<i>Xanthosoma Maffa</i> (Scoth) (Yellow Cocoyam)
Flesh	7.02 ^l ±0.64	85.03 ^k ±3.46	107.62 ^{jk} ±8.78	163.37 ^{gh} ±6.95
Boiled	182.86 ^{fe} ±7.54	89.05 ^k ±3.34	85.86 ^k ±4.61	134.49 ^{hi} ±1.52
Roasted	399.56 ^a ±3.15	142.22 ^{hi} ±6.27	267.06 [±] 5.87	149.94 ^{hi} ±15.74
Fried	265.94 ^c ±3.53	139.46 ^{hi} ±1.47	245.29 ^{cd} ±10.01	122.60 ^{ij} ±18.74
Oven-dried	203.99 ^{ef} ±26.91	321.27 ^b ±7.67	393.43 ^a ±11.33	217.07 ^{de} ±6.54
LSD 0.05	30.12			

Values are means of 3 replicates. a-i Means in the cells with different superscripts are significantly different (P>0.05).

Table 2A: Influence of cooking methods of yam/cocoyam tuber extracts on ABTS radical scavenging activity (mgTE/100gFW)

Processing methods	<i>D. cayenensis</i> (yellow yam)	<i>D. dumetorum</i> (trifoliate yam)	<i>D. bulbifera</i> (aerial yam)	<i>X.maffa</i> (Scoth) (yellow cocoyam)
Raw	32.49 ^m ±0.26	436.87 ^{ef} ±7.90	300.26 ^{hi} ±28.66	756.01 ^b ±3.35
Boiled	163.69 ^{kl} ±10.14	155.92 ^l ±10.14	298.45 ^{hi} ±3.73	106.62 ^l ±0.43
Roasted	537.66 ^d ±14.66	319.94 ^{hi} ±23.80	786.10 ^{ab} ±33.68	226.39 ^{jk} ±5.24
Fried	310.50 ^{hi} ±51.68	348.91 ^{gh} ±6.33	684.98 [±] 41.97	253.11 ^{ij} ±26.05
Oven-dried	284.04 ^{hi} ±9.26	849.89 ^a ±34.42	496.17 ^{de} ±31.54	408.933 ^{ef} ±3.40
LSD (0.05)	68.55			

Values are means of 3 replicates. a-m Means in same cell with different superscripts are significantly different (P>0.05).

Table 1B. Influence of cooking methods on flavonoids (TF) content of yams and cocoyam tuber extracts (mg CE/100gfw)

Processing methods	<i>D. Cayenensis</i> (Yellow Yam)	<i>D. dumetorum</i> (Trifoliate Yam)	<i>D. bulbifera</i> (Aerial Yam)	<i>Xanthosoma maffa</i> (Scoth) (Yellow Cocoyam)
Flesh	3.14 ^l ±0.26	4.09 ^l ±0.38	91.76 ^{gh} ±1.59	155.38 [±] 1.82
Boiled	102.13 [±] 3.86	17.55 ^k ±1.74	113.19 [±] 2.28	48.58 [±] 0.46
Roasted	232.98 [±] 2.67	18.38 ^k ±0.79	193.44 [±] 1.81	80.15 [±] 4.39
Fried	178.48 [±] 7.85	32.12 [±] 2.70	176.42 [±] 4.70	54.05 [±] 0.94
Oven-dried	45.69 [±] 0.92	57.21 [±] 4.32	254.89 [±] 14.27	95.64 [±] 1.86
LSD 0.05	12.43			

Values are means of 3 replicates. a-l Means in same cell with different superscripts are significantly different (P>0.05).

Table 2B: Influence of cooking methods of yam/cocoyam tuber extracts on DPPH radical scavenging activity (mgTE/100gFW)

Processing methods	<i>D. Cayenensis</i> (yellow yam)	<i>D. dumetorum</i> (trifoliate yam)	<i>D. bulbifera</i> (aerial yam)	<i>X.maffa</i> (Scoth) (yellow cocoyam)
Raw	88.17 [±] 0.95	146.51 ^{hi} ±0.27	417.86 [±] 2.32	729.39 ^a ±4.61
Boiled	198.01 ^{fe} ±7.37	22.98 [±] 1.07	190.73 ^{fe} ±5.19	138.12 [±] 1.45
Roasted	525.68 ^b ±23.10	30.01 ^{kl} ±0.38	333.31 ^d ±47.75	242.34 ^{ef} ±5.65
Fried	335.99 ^d ±6.30	28.50 ^k ±3.69	371.90 ^d ±29.53	198.69 ^{fe} ±1.80
Oven-dried	481.49 [±] 2.81	45.17 ^{kl} ±11.74	244.91 [±] 28.49	69.49 ^{jk} ±11.45
LSD (0.05)	45.08			

Values are means of 3 replicates. a-l Means in same cell with different superscripts are significantly different (P>0.05).

Table 2C: Influence of cooking methods of yam/cocoyam tuber extracts on oxygen radical absorption capacity (ORAC) (mgTE/100gFW)

Processing methods	<i>D. cayenensis</i> (yellow yam)	<i>D. dumetorum</i> (trifoliolate yam)	<i>D. bulbifera</i> (aerial yam)	<i>X. maffa</i> (<i>Scoth</i>) (yellow cocoyam)
Raw	459.29 ^b ±35.68	680.59 ^a ±35.34	629.37 ^a ±17.43	699.45 ^a ±49.47
Boiled	527.52 ^b ±41.23	522.58 ^b ±43.05	641.65 ^a ±31.74	657.21 ^a ±51.61
Roasted	276.68 ^g ±2.48	337.85 ^d ±24.01	304.41 ^e ±3.30	357.84 ^d ±8.52
Fried	253.53 ^g ±2.76	268.96 ^g ±2.09	408.20 ^c ±2.04	272.65 ^g ±17.39
Oven-dried	161.71 ^h ±4.25	152.12 ^h ±1.29	222.12 ^h ±2.90	140.15 ^j ±0.55
LSD (0.05)	74.29	LSD (0.05)		

Values are means of 3 replicates. a-j Means in same cell with different superscripts are significantly different (P>0.05).

Table 3A: Pearson's correlation coefficients of antioxidant activity, total polyphenols and total flavonoids for *D. cayenensis* (yellow yam)

	TP	TF	ABTS	DPPH	ORAC
TP	1.0000				
TF	0.052 ^{ns}	1.0000			
ABTS	0.354 ^{ns}	0.839 ^{xx}	1.0000		
DPPH	0.650 ^x	0.599 ^{ns}	0.896 ^{xx}	1.0000	
ORAC	-0.566 ^{ns}	-0.282 ^{ns}	-0.593 ^{ns}	-0.800 ^x	1.0000

TP = total polyphenols, TF = total Flavonoids, ABTS = antioxidant capacity measured in ABTS assay, DPPH = antioxidant capacity measured in DPPH assay, ORAC = antioxidant capacity measured in ORAC assay, ns = non significant, ^x and ^{xx} = significant at P<0.05 or 0.001.

Table 3B: Pearson's correlation coefficients of antioxidant activity, total polyphenols and total flavonoids for *D. dumetorum* (trifoliolate yam)

	TP	TF	ABTS	DPPH	ORAC
TP	1.0000				
TF	0.876 ^{xx}	1.0000			
ABTS	0.725 ^x	0.722 ^x	1.0000		
DPPH	-0.496 ^{ns}	0.567 ^{ns}	0.099 ^{ns}	1.0000	

Table 3C: Pearson's correlation coefficients of antioxidant activity, total polyphenols and total flavonoids for *D. bulbifera* (aerial yam)

	TP	TF	ABTS	DPPH	ORAC
TP	1.0000				
TF	0.873 ^{xx}	1.0000			
ABTS	0.375 ^{ns}	0.598 ^{ns}	1.0000		
DPPH	-0.242 ^{ns}	-0.377 ^{ns}	0.172 ^{ns}	1.0000	
ORAC	-0.852 ^{xx}	-0.930 ^{xx}	-0.695 ^x	0.210 ^{ns}	1.0000

TP = total polyphenols, TF = total Flavonoids, ABTS = antioxidant capacity measured in ABTS assay, DPPH = antioxidant capacity measured in DPPH assay, ORAC = antioxidant capacity measured in ORAC assay, ns = non significant, ^x and ^{xx} = significant at P<0.05 or 0.001.

Table 3D: Pearson's correlation coefficients of antioxidant activity, total polyphenols and total flavonoids for *Xanthosoma maffa* (*Scoth*) (yellow cocoyam)

	TP	TF	ABTS	DPPH	ORAC
TP	1.0000				
TF	-0.613 [*]	1.0000			
ABTS	-0.691 [*]	0.957 ^{**}	1.0000		
DPPH	-0.293 ^{ns}	0.824 ^{**}	0.817 ^{**}	1.0000	
ORAC	-0.456 ^{ns}	0.252 ^{ns}	0.252 ^{ns}	0.636 [*]	1.0000

TP = total polyphenols, TF = total Flavonoids, ABTS = antioxidant capacity measured in ABTS assay, DPPH = antioxidant capacity measured in DPPH assay, ORAC = antioxidant capacity measured in ORAC assay, ns = non significant, ^{*} and ^{**} = significant at P<0.05 or 0.001.