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Inhibitory effects of methanolic extracts of two eggplant species from South-western Nigeria on starch hydrolysing enzymes linked to type-2 diabetes

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This study sought to investigate the inhibitory effect of methanolic extract of different species of African eggplant (*Solanum melongena* L.) and (*Solanum macrocarpon*) on starch hydrolysing enzymes relevant to type-2 diabetes (α-amylase and α-glucosidase). The phenolic content and antioxidant properties of the eggplant varieties were also assessed. The results revealed that both extracts exhibited mild α-amylase and stronger α-glucosidase inhibitory activities in a dose dependent manner. Furthermore, *S. macrocarpon* exhibited stronger radicals (1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging ability, nitric-oxide ($NO^{\cdot}$) scavenging ability and ferric reducing antioxidant property (FRAP) than *S. melongena*, while *S. melongena* had stronger hydroxyl ($OH^{\cdot}$) radical ability. The inhibition of starch hydrolysing enzymes and antioxidant activities suggested the potential use of eggplants in the dietary management or control of postprandial hyperglycemia associated with type-2 diabetes.

Key words: Antioxidant activity, diabetes, α-amylase, α-glucosidase, *Solanum macrocarpon, Solanum melongena.*

INTRODUCTION

Non insulin dependent diabetes mellitus (NIDDM) is the commonest form of diabetes which accounts for 90% of all cases. The prevalence of this disease is increasing annually and the number of diabetics is projected to rise above 300 million worldwide (Bailey and Day, 2004; Li et al., 2004). Hyperglycemia is a metabolic disorder primarily characterized by β-cells disorder, relative insulin deficiency, and an abnormal rise in blood sugar right after a meal (Kwon et al., 2007). Pancreatic α-amylase breaks down large polysaccharides (starch) into disaccharides and oligosaccharides, before the action of α-glucosidases which break down disaccharides into monosaccharides (glucose) which is readily absorbed into the blood stream.

Inhibition of pancreatic α-amylase and α-glucosidase is the mechanism adopted by many commercially available drugs for the management of non insulin dependent diabetes mellitus (Krentz and Bailey, 2005). Hence, inhibition of intestinal α-glucosidase which delays the absorption of glucose after starch conversion moderates the postprandial blood glucose elevation and thus mimics the effects of dieting on hyperglycemia (Bischoff, 1994). Chronic amylase inhibition may also be useful for treating type 2 diabetes and obesity (Koike, 2005). Increasing evidence in both experimental and clinical studies have shown the participation of oxidative stress in the development and progression of diabetes mellitus (Baynes and Thorpe, 1999; Ceriello 2000). This is usually accompanied by increased production of free radicals or impaired antioxidant defences (Halliwell et al., 1990; Saxena et al., 1993).

Free radicals are formed disproportionately in diabetes by glucose oxidation, non-enzymatic glycation of

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proteins, and the subsequent oxidative degradation of glycated proteins. Abnormally high levels of free radicals and the simultaneous decline of antioxidant defense mechanisms can lead to damage of cellular organelles and enzymes, increased lipid peroxidation, and development of insulin resistance (Maritim et al., 2003).

The growing number of diabetics, coupled with the harsh side effects of some synthetic drugs has led to the increasing search for alternatives, which are relatively cheap with minimal side effect (Chakraborty et al., 2002). One therapeutic approach for treating diabetes is to decrease the post-prandial hyperglycemia (Kwon et al., 2006, 2007).

Phenolic compounds are widely distributed in edible plants and have been suggested to protect against a variety of diseases (Johnson et al., 2005). Recent investigations suggest that phenolic components of higher plants may act as antioxidants or via other mechanisms prevent disease processes (Wang et al., 2000). Recent findings have also demonstrated that phenolics cross intestinal barriers and are sufficiently absorbed to have the potential to exert biological effects (Williamson and Monach, 2005).

Phenols are the most abundant antioxidants in human diets. They exhibit a wide range of biological effects including antibacterial, anti-inflammatory, antiallergic, hepatoprotective, antithrombotic, antiviral, anticarcinogenic and vasodilatory actions (Scalbert and Williamson, 2000; Middleton et al., 2000). They are capable of removing free radicals, chelate metal catalysts, activating antioxidant enzymes, reducing α-tocopherols and inhibiting oxidases (Amic et al., 2003). They are also important for improving the sensory and nutritional qualities, in that they impart colors, flavors and tastes (Kim et al., 2002).

Eggplant is an economic flowering plant belonging to the family Solanaceae and widely distributed throughout the temperate and tropical regions (Eun-Ju et al., 2011). Members are mostly herbaceous plants, and the fruit is berry and the seeds have large endosperm and are grown mainly for food and medicinal purposes (Kwon et al., 2008). However, there are different types of species and selection in the genus that have diverse shape, size and colour. While information is available on phenolic contents, flavonoids, anthocyanins, antioxidants, and anti-diabetic activity of *Solanum melongena* eggplant fruit extract (kwon et al., 2008; Scalzo et al., 2010), few information are reported for *Solanum macrocarpon* which is commonly found in southwestern Nigeria known as “igbagba pupa”, and used as sauce, stew and part of salad, but there are information on the leaf part which is been used as soup condiment in Nigeria (Oboh et al., 2005; Fasuyi, 2006; Ijarotimi et al., 2010), while the aqueous extract of the fruit had been shown to be nutritious, to lower high blood pressure, to treat constipation and lower hyperlipidaemia (Chinedu et al., 2011; Dougnon et al., 2012; Sodipo et al., 2013).

Both of these species are domesticated and differ from their wild ancestors in that they underwent artificial selection for larger fruit with less bitter flavour (Knapp et al., 2013; Grubben and Denton, 2004). This present study sought to explore further and to compare the ability of these African eggplant varieties *in vitro* ability using different models of oxidative stress. It is also expedient to investigate the mechanism of anti-diabetic effect of the methanolic extract of these species since there are dearth of information on the *S. macrocarpon*.

MATERIALS AND METHODS

Sample collection

Fresh eggplants (*S. melongena* L.) and (*S. macrocarpon*) (200 g) each were purchased from Erekesan main market at Akure, Ondo State, Nigeria. The identification and authentication of the samples was carried out at the Crop, Soil, and Pest management (CSP) Department of the Federal University of Technology, Akure, Nigeria. The eggplant samples were sliced into pieces, sun-dried for 7 days and milled into powder using a Waring heavy duty blender (Waring Products Division, New Hartford, Connecticut, USA), and the powder was stored in an airtight plastic container. All the chemicals used were of analytical grade, and distilled water was used for the analyses.

Chemicals and equipment

Folin-Ciocalteu’s phenol reagent, gallic acid and anhydrous sodium carbonate used were products of Fluka (Buchs, Switzerland). Quercetin and DPPH, ascorbic acid and starch were products of Merck (Darmstadt, Germany), H_2O_2, and Iron (II) sulphate were products of Sigma (Aldrich, USA). Iron (III) chloride 6-hydrate and trichloroacetic acid are Fisher products. All other chemicals used were purchased from Rovet Scientific Limited, Benin City, Edo State, Nigeria. The distilled water used was obtained from the Chemistry Department at Federal University of Technology, Akure. Optical absorbance was measured with a ultraviolet (UV)-Visible spectrophotometer (Model 6305; Jenway, Barloworld Scientific, Dunmow, United Kingdom).

Sample preparation

Extraction of phenolics compounds was carried out using the modified method of Chu et al. (2002). About 20 g each of the milled samples was soaked in 80% methanolic (1:20 w/v) for 24 h. Thereafter, they were filtered through Whatman No. 2 filter paper on a Bucher funnel under vacuum. The filtrate was evaporated using a rotary evaporator under vacuum at 40°C to dryness. All samples extract were stored at -4°C.

Extract preparation

A 0.5 g of the methanol extract of each sample was reconstituted in 50 ml distilled water, which was subsequently used for the various assays. All antioxidant tests and analyses were performed in triplicate, and results were averaged.

Total phenol determination

The total phenol content of the sample was determined by adding

0.5 ml of the sample extract to an equal volume of water, and 2.5 ml 10% Folin-Ciocalteau reagent (v/v) and 2.0 ml of 7.5% sodium carbonate was subsequently added. The reaction mixture was incubated at 45°C for 40 min, and absorbance was measured at 726 nm (JENWAY 6305), gallic acid was used as the standard phenol (Singleton et al., 1999).

Determination of total flavonoid content

The total flavonoid content of the extracts of the sample was determined by the method reported by Meda et al. (2005). The volume 0.5 ml of samples and the standard quercetin was mixed with 0.5 ml methanol, 50 µl of 10% $AlCl_3$, 50 µl of 1 mol/L potassium acetate and 1.4 ml water. The reaction mixture was incubated at room temperature for 30 min. Thereafter, the absorbance of the reaction mixture was subsequently measured at 415 nm. The total flavonoid content was calculated using quercetin as standard.

Ferric reducing power (FRAP)

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 mM 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, and 5 ml of the supernatant was mixed with an equal volume of water and 1 ml of 0.1% ferric chloride. The absorbance was measured at 700 nm against a reagent blank. A higher absorbance indicates a higher reducing power.

α-Amylase inhibition assay

The methanolic extracts volume (500 µl) and 500 µl of 0.02 M sodium phosphate buffer (pH 6.9 with 0.006 M NaCl) containing Hog pancreatic α-amylase (EC 3.2.1.1) (0.5 mg/ml) were incubated at 25°C for 10 min. Then, 500 µl of 1% starch solution in 0.02 M sodium phosphate buffer (pH 6.9 with 0.006 M NaCl) was added to each tube. The reaction mixtures was incubated at 25°C for 10 min and stopped with 1.0 ml of dinitrosalicylic acid colour reagent. Thereafter, the mixture was incubated in a boiling water bath for 5 min and cooled to room temperature. The reaction mixture was then diluted by adding 10 ml of distilled water, and absorbance measured at 540 nm using the spectrophotometer (JENWAY 6305). The percentage (%) enzyme inhibitory activity of the aqueous extracts was calculated (Worthington, 1993).

α-Glucosidase inhibition assay

The volume of the methanolic extracts (50 µl) and 100 µl of α-glucosidase solution (1.0 U/ml) in 0.1 M phosphate buffer (pH 6.9) was incubated at 25°C for 10 min. Then, 50 µl of 5 mM p-nitrophenyl-α-D-glucopyranoside solution in 0.1 M phosphate buffer (pH 6.9) was added. The mixtures were incubated at 25°C for 5 min. Then, 2 ml of Na_2CO_3 was added to terminate the reaction before reading the absorbance at 405 nm in the spectrophotometer (JENWAY 6305). The α-glucosidase inhibitory activity was expressed as percentage inhibition. The percentage (%) enzyme inhibitory activity of the aqueous extracts was calculated (Apostolidis et al., 2007).

$$\% \text{ inhibition} = \frac{(\text{Absorbance of control} - \text{Absorbance. of samples})}{\text{Absorbance of control}} \times 100$$

1,1-Diphenyl-2-picrylhydrazyl (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 was mixed with 1 ml of 0.4 mM 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 contained all the reagents without the test sample.

Fenton reaction (degradation of deoxyribose)

The ability 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). Methanolic extract (0 to 100 µl) was added to a reaction mixture containing 120 ml 20 mM deoxyribose, 400 ml 0.1 M phosphate buffer, 40 ml 20 mM hydrogen peroxide, and 40 ml 500 mM $FeSO_4$,, and the volume were made to 800 ml 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 (JENWAY6305).

$$\% \text{ inhibition} = \frac{(\text{Absorbance of control} - \text{Absorbance. of samples})}{\text{Absorbance of control}} \times 100$$

Determination of nitric-oxide radical scavenging ability ($NO^{\cdot}$)

The scavenging effect of the extracts against nitric-oxide radical was measured according to the method of Mercocci et al. (1994). The reaction mixture containing 1 ml (25 mM) sodium nitroprusside in phosphate buffer saline and samples for 100, 200, 300, and 400µl, respectively were incubated at 37°C for 2 h. An aliquot after (0.5 ml) of the incubation was removed and diluted with 0.3 ml Greiss reagent (1% suphanilamide in 5% H_3P0_4 and 0.1% Napthlethylenediaminedihydrochoride). The absorbance of the chromophore formed was immediately read at 570 nm against distilled water as blank.

Statistical analysis

The results of the replicate readings were pooled and expressed as mean ± standard deviation. Student *t* test was performed and significance was accepted at $P \leq 0.05$ (Zar, 1984). IC_{50} (concentration of extract that will cause 50% inhibitory activity) was determined using linear regression analysis

RESULTS

Results of this study (Table 1) showed that the phenolic compound *S. macrocarpon* eggplant extract (50.01

Table 1. Total phenol, Total flavonoids,and Ferric Reducing power (FRAP) of methanolic extracts of eggplant species.

Samples	Total phenols(mgGAE/100g)	Total flavonoids (mgQE/100g)	Reducing power (mg/100g)
S.macrocarpon	50.01±1.85[a]	15.48±1.75[a]	56.67±1.43[a]
S.melongena	34.57±1.07[b]	9.34±1.34[b]	48.78±0.55[b]

Data represent means of triplicate determinations.Values with the same letter along the same column are not significantly different ($P < 0.05$).mgGAE:Milligram gallic acid equivalent; mgQE:Milligram Quercetin equivalent.

Table 2. IC_{50} µg/mL of antioxidant activities and enzymes inhibition methanolic extracts of eggplant species.

Samples	DPPH-Free Radical scavenging ability	OH-Radical scavenging ability	No-Radical Scavenging ability	α-amylase inhibition	α-glucosidase inhibition
S.macrocarpon	727.80±0.19[a]	163.08±0.28[b]	551.26±0.89[a]	42.66±0.24[a]	71.77±0.50[a]
S.melongena	965.25±0.02[b]	91.99±0.27[a]	874.12±0.42[b]	40.11±0.15[a]	63.24±0.30[b]

Data represent means of triplicate determinations.Values with the same letter along the same column are not significantly different ($P < 0.05$).

mg/100 g) is significantly ($P < 0.05$) higher than *S. melongena* eggplant extract (34.57 mg/100 g). In this study, total flavonoids were evaluated in the two species. Table 1 results revealed that *S. macrocarpon* had higher total flavonoids content (15.48 mgQE/100 g) than *S. melongena* (9.34 mgQE/100 g). The ferric reducing antioxidant capacity (FRAP) of the extract reported as ascorbic acid equivalent which is the ability of the phenolic extracts to reduce Fe (III) to Fe (II); a measure of their antioxidant properties. Both species had high reducing power as shown in Table 1. It showed that *S. macrocarpon* had a significantly higher ($P < 0.05$) reducing power (56.67 mg/100 g) than *S. melongena* (48.78 mg/100 g).

The interaction of eggplant extracts with α-amylase, as shown in Figure 1, revealed that both extracts had a marked inhibition of pancreatic α-amylase activity in a dose dependent manner in the range of 0-100 µg/ml. However, judging by the IC_{50} (extract concentration causing 50% enzyme inhibition) value (Table 2), there was no significant change ($P > 0.05$) in the inhibitory ability of *S. macrocarpon and S. melongena* extract. In the same vein, the ability of the eggplant extracts to inhibit α-glucosidase activity *in vitro* was investigated and the result is presented in Figure 2. The results revealed that both eggplant extract inhibited α-glucosidase in a dose-dependent manner (0 to 100 µg/ml). However, there was higher inhibitory ability of *S. melongena* extract on α-glucosidase than *S. macrocarpon* extract. It was observed that the extracts showed a stronger inhibition of pancreatic α-glucosidase and a mild inhibition of α-amylase as shown in Figures 1 and 2.

The IC_{50} of the ability of the 80% methanolic extracts to scavenge 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical is shown in Figure 3. DPPH had been used to test the free radical scavenging ability of various natural products (Brand et al., 1995). In this study, the extracts obtained from *Solanum* species exhibit lower IC_{50} values, indicating the higher potential as free radical scavengers. However, *S. macrocarpon* had more lower IC_{50} value of (727.80 µg/ml) while *S. melongena* had (965.25 µg/ml). Both extract scavenged DPPH in a dose-dependent manner at all tested concentration (0 to 330 µg/ml) (Figure 3). However, the results revealed that *S. macrocarpon* had significantly higher ($P < 0.05$) free radical scavenging ability from the IC_{50} than *S. melongena*.

IC_{50} values represent eggplant concentrations required to scavenge 50% free radicals as presented in Table 2; a low IC_{50} translates to a higher antioxidant activity. Table 2 and Figure 4 showed the inhibitory effect of *S. macrocarpon* and *S. melongena* eggplant on deoxyribose degradation under different Fe^{2+}/H_2O_2 oxidative assault. It is noteworthy that when Fe^{2+} or H_2O_2 was used as the oxidants, *S. melongena* eggplant exerted a marked inhibitory effect (91.99 µg/ml) on deoxyribose degradation when compared to *S. macrocarpon* (163.10 µg/ml). This showed that individual components of the eggplants exhibited inhibitory effect on $OH^{\bullet}$ radical-induced oxidative assault on deoxy-ribose degradation. The nitric oxide ($NO^{\bullet}$) radical scavenging ability of the methanolic extracts of *S. macrocarpon* and *S. melongena* is presented in Figure 5 and the IC_{50} in Table 2. The results revealed that both extracts were able to scavenge $NO^{\bullet}$ radical produced in a dose dependent manner (0 to 250 µg/ml). However, as revealed from the Figure 5, the $NO^{\bullet}$ radical scavenging ability of *S. macrocarpon* at the maximum concentration exhibited inhibition at (17.38%), while *S. melongena* exhibited a 12.49% maximum tested concentration.

DISCUSSION

Phenolic compound embraces a wide range of plant substances which possess common aromatic ring bearing one or more hydroxyl substituents. They tend to

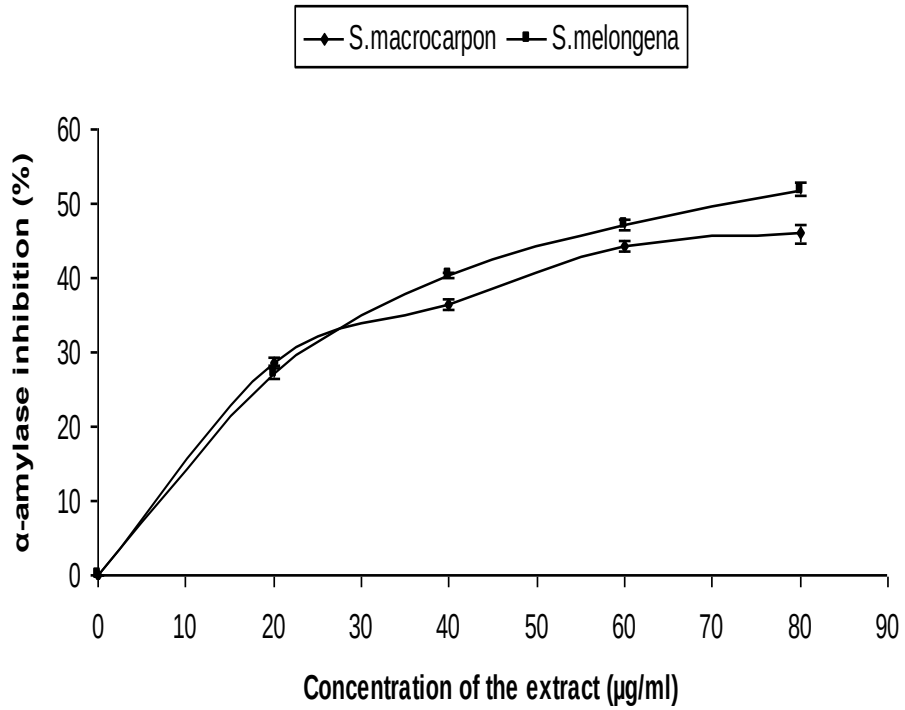

Figure 1. α-Amylase inhibitory activity of methanolic extract of eggplant species.Values represent mean ± standard deviation, *n* = 3. (P<0.05)

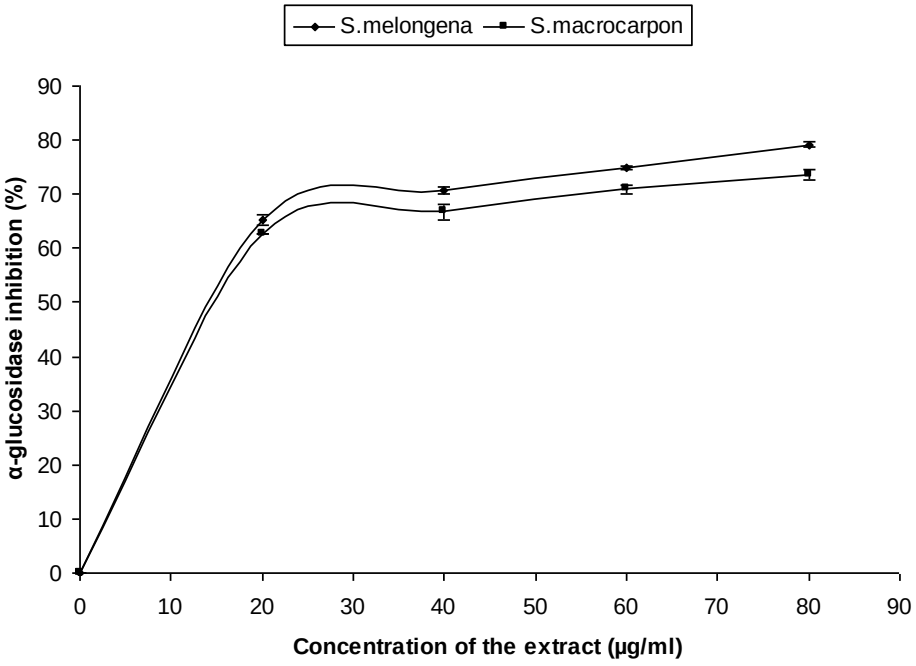

Figure 2. α-Glucosidase inhibitory activity of methanolic extract of eggplant species. Values represent mean ± standard deviation, *n* = 3 (P<0.05)

be water soluble since they most frequently occur combined with sugar as glycosides and are usually located in the cell vacuole (Galston., 1969; Materska and Perucka, 2005). Phenolic compounds are plant-derived antioxidants that possess metal-chelating capabilities and radical-scavenging properties (Bors and Saran, 1987;

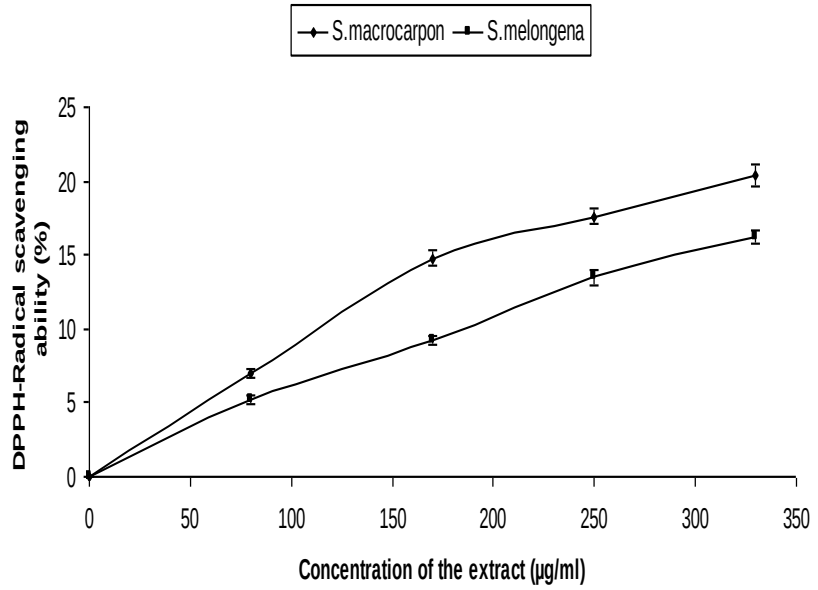

Figure 3. DPPH free radical-scavenging ability of methanolic extract of eggplant species.Values represent mean ± standard deviation, *n* = 3.(P<0.05)

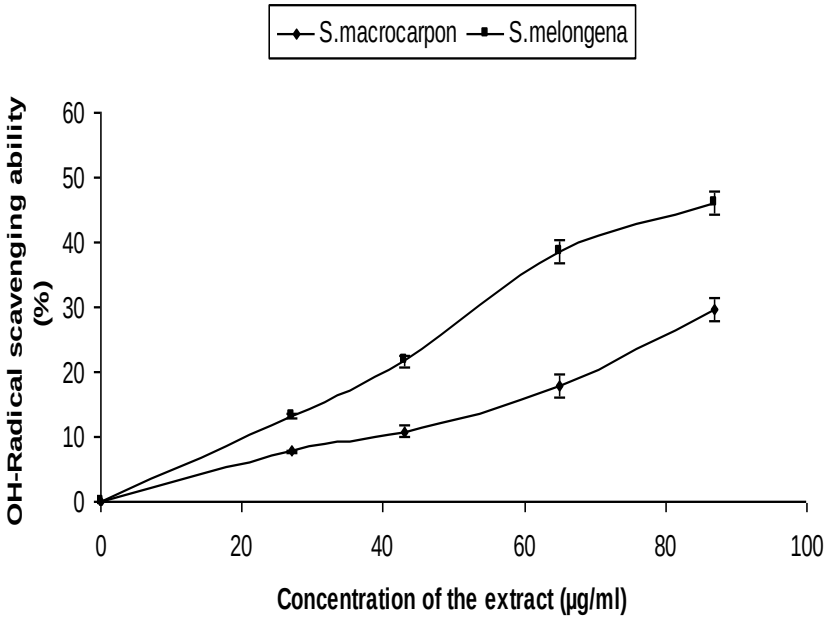

Figure 4. Hydroxyl radical-scavenging ability of methanolic extract of eggplant species.Values represent mean ± standard deviation, *n* = 3.(P<0.05)

Lopes et al., 1999). Phenols display a vast variety of structure which can be divided into three main classes, which are flavonoids, tannin and phenolic acids (Strube et al., 1993). Phenolic compounds known to possess high antioxidant activity are commonly found in fruits, vegetables, and herbs. The results of this study is lower than the methanolic extracts of Thailand varieties of eggplant (Akanitapichat et al., 2010), and methanol extract of the bark and leaves of southern African medicinal plants (Steenkamp et al., 2013).

However, this results is higher than that of aqueous extracts of seed and flesh of riped and unripe *S. melongena* (Fetegbe et al., 2012), and aqueous extract of *S. aethiopicum* cultivars (Nwanna et al., 2013). These differences may be due to the complexity of these groups of compounds, method

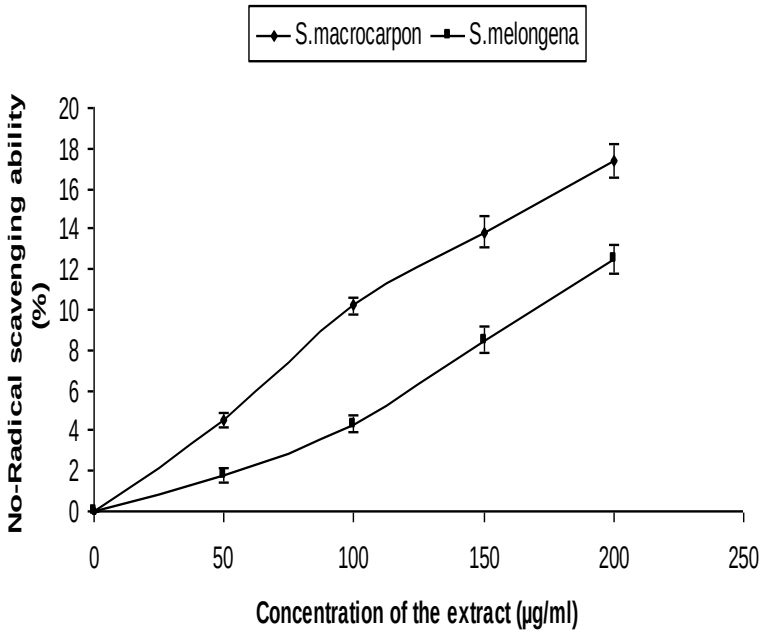

Figure 5. Nitric-oxide radical-scavenging ability of methanolic extract of eggplant species.Values represent mean ± standard deviation, *n* = 3.(P<0.05)

of extraction, climatic condition and cultural practices, maturity at harvest, and storage condition (Podsedek, 2007). Singleton et al. (1999) reported that different phenolic compounds in matrix samples have different responses in the Folin–Ciocalteu method, depending on the number of phenolic groups they have. Some of the inorganic substances (hydrazine, hydroxyammonium chloride, iron ammonium sulfate, iron sulfate, manganese sulfate, and so on) may react with the Folin–Ciocalteureagent to give elevated apparent phenolic concentrations (Prior et al., 2005). Flavonoids and other plant phenolics are especially common in leaves, flowering tissues and woody parts such as the stem and bark (Larson, 1988). Flavonoids are a class of widely distributed phytochemicals with antioxidant and biological activity. The results gotten is higher than that of (Chaira et al., 2009) on water-methanolic extracts on different common Tunisia date cultivars, also on *S. aethiopicum* cultivars (Nwanna et al., 2013) but lower than that reported by Sariburun et al. (2010) on different varieties of blackberry and ethanolic extract of *Zanthoxylum armatum* fruit (Barkatullah et al., 2013). However, the results are within the same range of flavonoids extracts of selected tropical plants (Mustafa et al., 2010). The trend in the total flavonoid contents agreed with the total phenolic contents results. The antioxidant activity of flavonoids has been linked to the number of OH groups and their arrangement on the molecule. They are constituents of plant foods that have been implicated in the reduction of cancer risk (Sies, 1986). Phenolics and flavonoids of plant origin are reported to have potent antioxidants, and homeostatic balance between pro-oxidant and anti-oxidants is known to be important for maintenance of health as well as prevention from various degenerative diseases (Rawat et al., 2011).

Antioxidants are strong reducing agents and this is principally because of the redox properties of their hydroxyl groups and the structural relationships of any parts of their chemical structure (Eleazu et al., 2011). The ability of the extracts of both eggplants to reduced Iron (III) to Iron (II) is higher than that reported by Eun-Ju et al. (2011) on antioxidant activity of different parts of eggplant (*S. melongena* L.), on riped and unriped *S. melongena* L. (Fetegbe et al., 2012), and on plantain extracts (Shodehinde and Oboh, 2012). It is also well known that iron and iron complexes stimulate lipid peroxidation in cells (Gogvadze et al., 2003). The mechanism(s) that underlies the antioxidant activity of both eggplant extracts measured in the presence of Fe^{2+} may be credited to the ability of the extracts to scavenge radical, reduce Fe^{3+}. This showed that eggplants phytochemicals such as phenolic compounds are strong reducing agents that could act by readily neutralizing free radicals through electron donation or hydrogen atom transfer.

This study confirm work done by Know et al. (2008) also, Das et al. (2011) showed that there was a direct relationship between eggplants and their cardioprotective ability. In addition, the ripe fruits of these eggplant species may contain other compounds such as anthocyanins that give the fruits its characteristic yellow

and purple pigments and possibly additional antioxidant effect. In addition, it was found that extracts from *S. melongena* (purple colour) small size eggplant fruits with potent antioxidant activities have been shown to contain high phenolic and anthocyanin content (Nisha et al., 2009). Nevertheless, this *S. macrocarpon* eggplant consumed popularly in the southeastern Nigeria have also shown to have high antioxidant activities which must have resulted from its high phenolics and flavonoids content.

The results of enzymes inhibition is in agreement with earlier reports that plant phytochemicals (Adefegha and Oboh, 2013) are mild inhibitors of amylase and strong inhibitors of glucosidase activity (Kwon et al., 2007). Higher inhibition of glucosidase activity and mild inhibition of amylase activity of the eggplant extracts could address the major drawback of currently used glucosidase and amylase inhibitor drugs with side effects such as abdominal distention, flatulence, meteorism and possibly diarrhoea (Pinto et al., 2009). It has been suggested that such adverse effects might be caused by the excessive pancreatic amylase inhibition resulting in the abnormal bacterial fermentation of undigested carbohydrates in the colon (Kwon et al., 2007). This agrees with the findings on *S. melongena* phenolics which have been recommended as a choice diet for the management of type 2 diabetes (Pinto et al., 2009; Nickavar and Yousefian, 2009). The results of the enzymes (α-amylase and α-glucosidase) inhibitory assays agreed with the phenolic contents, flavonoids contents and reducing power activity of both extracts. Nevertheless, these extracts had higher inhibitory effect than that reported by (Oboh et al., 2012) on ethanolic extract of fluted pumpkin, also to what was reported on 80% acetone and ethyl acetate phenolics extract on bitterleaf (Saliu et al., 2011).

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 that involves free radical scavenging (Hu et al., 2000). In addition, the trend in the results agree with the flavonoid distribution in the extracts and many earlier research articles, where correlations were reported between flavonoids content and antioxidant capacity of some plant foods (Chen et al., 2007; Shodehinde and Oboh, 2012). The inhibitory properties of the methanolic extracts to scavenge DPPH is higher than the ability of three Brazilian plants using three different extractant (Pereira et al., 2009) but lower than that gotten by Sariburun et al. (2010) on raspberry and blackberry cultivars. However this results correlate with that of Pereira et al. (2009) in which the inhibitory potency of DPPH radical was higher in methanolic extractant than aqueous extract. The oxygen molecule might produce a highly reactive oxygen species (ROS) by some exogenous factors and endogenous metabolic processes in human body.

ROS include a number of chemically reactive molecules such as hydrogen peroxide (H_2O_2), superoxide (O^{2-}), and the hydroxyl radical ($^{\bullet}OH$). The $^{\bullet}OH$ in the cells can easily cross cell membranes at specific sites, react with most biomolecules and furthermore cause tissue damage and cell death. Thus, removing $^{\bullet}OH$ is very important for the protection of living systems. As shown, the $^{\bullet}OH$ scavenging effect by the *S. macrocarpon* and *S. melongena* eggplants extracts, and apparently the *S. macrocarpon* eggplant extracts exhibit higher potent •OH scavenging ability with consequent protection of deoxyribose damage than *S. melongena* from the IC_{50}. We may attribute this observed •OH scavenging ability to possible total phenolic acids, tannin, flavonoids contents such as anthocyanins, flavonols, flavones, flavonols, flavanones (Noda et al., 2000; Kwon et al., 2008). More so, these differences could be due to the complexity of individual phenolic constituents and their concentrations (Wahle et al., 2009).

The oxidative destruction of the beta cells in the pancreas is crucial in the development of diabetes, thus radical have been shown to be effective in the management of diabetes in animal models. Diabetes can also be initiated in animals by synthetic drugs such as alloxan and streptozotocin through the production of reactive oxygen species (Oberley, 1988). Single nucleotide polymorphisms (SNP) is a component of antihypertensive drugs; it causes cytotoxicity through the release of cyanide or $NO^{\bullet}$ which acts in cooperation with other ROS to cause neuronal damage (Oboh and Rocha, 2007). Moreso, SNP has been shown to undergo photodegradation ultimately producing nitrogen oxide ($NO^{\bullet}$) $[(CN)_5\text{-}Fe]^{3+}$ and $[(CN)_4\text{-}Fe]^{2+}$ species (Arnold et al., 1984). Likewise, it has been observed that the iron moiety of SNP may have a free iron coordination site for H_2O_2, which could trigger the generation of highly reactive oxygen species such as hydroxyl radicals ($^{\bullet}OH$) via the Fenton reaction (Graf et al., 1984). Therefore, following a short-lasting release of $NO^{\bullet}$, iron moiety of SNP could cause a long-lasting generation of $^{\bullet}OH$ radicals and oxidant stress/injury similar to that of ferrous citrate iron complexes, which may initiate a lipid peroxidation chain reaction and oxidative injury (Mohanakumar et al., 1994).

S. melongena extract exhibited higher inhibition of $NO^{\bullet}$ radical produced than *S. macrocarpon*, the reason may be as a result of the synergistic activity of the phenols constituents, however this is contrary to the ability of this extract on the produced $^{\bullet}OH$. Furthermore, since nitric oxide has been proposed to act as a pro-oxidant at high concentrations, or when it reacts with superoxide, forming the highly reactive peroxynitrite ($ONOO^{\bullet}$) (Radi et al., 2001), we can speculate that the eggplant extracts may scavenge superoxide and peroxynitrite, and may further interact with the cyanide moiety or even the release of iron from the ferrocyanide moiety of SNP, thereby exerting a synergistic-like anti-oxidant effect. The results of this study could eventually explain the mechanism of action of eggplants in the management of postprandial

hyperglycemia and one of its long term complications.

Conclusion

This study revealed that the antioxidative properties and inhibition of key enzymes linked to non-insulin diabetes mellitus (α-amylase and α-glucosidase) could be part of the mechanism through which these eggplants could manage/prevent type-2 diabetes.

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