

Aqueous Extracts from Unripe Plantain (*Musa paradisiaca*) Products Inhibit Key Enzymes Linked with Type 2 Diabetes and Hypertension *in vitro*

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Abstract

Mature green plantains bought at Oja-Oba in Akure were roasted into a local meal called 'boli' while another portion was boiled in water. Each of the samples was sun dried and milled into flour. The aqueous extracts of roasted and boiled were prepared (10g/100mL). The study was based on inhibition of α -amylase, α -glucosidase and angiotensin 1 converting enzyme (ACE). The antioxidant activities of the plantain extracts to reduce Fe^{3+} to Fe^{2+} and to bind to iron (II) ions were evaluated. The phenolic contents (total phenol and total flavonoid), vitamin C and ABTS (2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) were also determined. The results revealed that the boiled flour had higher ($P < 0.05$) phenolic contents and reducing power while the roasted flour had higher Vitamin C content but there was no significant difference between the ABTS• antioxidant activity of both the roasted and boiled. The higher enzyme inhibitory activities on α -amylase, α -glucosidase and angiotensin 1 converting enzyme (ACE) were exhibited by the boiled flour extract. The trend of this result followed what was observed in the phenolic contents. In view of these findings there is suggestion that plantain possesses compounds with anti-diabetes and anti-hypertension activities that may be beneficial eventually as functional foods.

Keywords: Anti-diabetes, anti-hypertension, angiotensin 1 converting enzyme, α -amylase, α -glucosidase, extract.

1. Introduction

Non insulin dependent diabetes mellitus (NIDDM) is the commonest form of diabetes which accounts for 90% of all cases. 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 NIDDM (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). Many available synthetic drugs, such as acarbose, voglibose, and miglitol, are widely used to

inhibit these enzymes in patients with type 2 diabetes. However, these inhibitors are reported to cause some discomforting side effects such as abdominal distention, flatulence, meteorism, and diarrhea because of excessive α -amylase inhibition (Chakrabarti and Rajagopalan, 2002). This necessitates the search for inhibitors from natural sources with strong α -glucosidase, but mild α -amylase activities. One of the long-term complications of NIDDM is hypertension. Angiotensin I converting enzyme (ACE) (EC 3.4.15.1) plays an important physiological role in regulating blood pressure (Skeggs and Khan, 1957). ACE belongs to the class of zinc proteases and is expressed in the vascular endothelial lining of human lungs. ACE is a dipeptidyl carboxypeptidase that catalyzes the conversion of angiotensin I (decapeptide) to angiotensinII (octapeptide), it inactivates the antihypertensive vasodilator (bradykinin) and increases blood pressure (Skeggs and Khan, 1957). In western medicine, drug development effort has been directed toward excluding unwanted side-effects (Dzau, 1988). Inhibition of the angiotensin 1 converting enzyme is established as one modern therapeutic principle in the treatment of hypertension. Screening for anti-hypertensive effects in traditional plants has been performed over many years and

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several animal studies have been carried out (Villar *et al.*, 1986). Millions of people in developing nations, including Nigerians, have resorted to the use of plants to treat their ailments; this could be due to the high cost of orthodox health care or as a result of the global shift towards the use of natural sources, rather than synthetic drugs (Omonkhua and Onoagbe, 2011). Plantain (*Musa paradisiaca*) is a major food crops in the humid and sub-humid parts of Africa and a major source of energy for millions of people in these regions (Asiedu *et al.*, 1992). The annual world production of plantain is estimated at 75 million tones (John and Marchal, 1995). Plantains have been reported to be an important source of provitamin A in parts of Asia, Africa and Latin America (Sommer, 1989).

Plantains are a good source of vitamin A (carotene), vitamin B complex (thiamin, niacin riboflavin and B6) and vitamin C (ascorbic acid). Plantains provide a better source of vitamin A than most other staples (Aurang, 1987). They are notably high in potassium and low in sodium (Marriott *et al.*, 1983). carotenoid-rich foods protect against certain chronic diseases, including diabetes, heart disease and cancer (Ford *et al.*, 1999). Carotenoids are one of the most important classes of plant pigments and play a crucial role in defining the quality parameters of fruit and vegetables (Dadzie and Orchard, 1997). Unripe plantain meal is usually consumed by Nigerian diabetics to reduce postprandial glucose level (Willett *et al.*, 2002). *Musa paradisiaca* belong to the Musaceae family which are evergreen tropical giant herbs whose fruits can be consumed either as ripe or unripe using different types of processing methods which can be cooked, roasted, steamed, baked or grilled. Other products such as flour and chips have been derived (Nwokocha and Williams, 2009). Plantain is employed in the folklore management of diseases such as diabetes, ulcer and wound healing due to its hypoglycaemic, anti-ulcerogenic and analgesic properties (Bischoff, 1994). Earlier report on the glycemic indices of green plantain products had been discussed (Obboh and Erema, 2010, Willet *et al.*, 2002). It is therefore expedient to further research on activities of some of its processed (roasted and boiled) forms vis- a-vis their interactions with α -amylase, α -glucosidase and angiotensin 1 converting enzyme and their antioxidant activities and to infer their benefits in the prevention and management of metabolic diseases such as diabetes mellitus and hypertension in vitro. This present study will be the first to present the interactions of plantain products extracts on the enzymes linked to type 2 diabetes and hypertension.

2. Materials and Methods

2.1. Plant materials

“False Horn” matured green plantain (*Musa paradisiaca*) were bought at Oja-Oba in Akure. Ondo state. Authentications of the unripe plantains were carried out at the Department of Biology, Federal University of Technology, Akure, Nigeria.

Chemicals and equipment Folin-Ciocalteu’s phenol reagent, gallic acid and anhydrous sodium carbonate used were products of Fluka (Buchs, Switzerland). Quercetin and DPPH (2,2-diphenyl-1-picrylhydrazyl), Ascorbic acid

and starch were products of Merck (Darmstadt, Germany), Iron chloride, ACE, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid (ABTS), porcine pancreatic α -amylase (EC 3.2.1.1) and α -glucosidase (EC 3.2.1.20) were products of Sigma-Aldrich (USA). Iron (III) chloride 6-hydrate and trichloroacetic acid 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 UV-Visible spectrophotometer (Model 6305; Jenway, Barloworld Scientific, Dunmow, United Kingdom).

2.2. Preparations of Samples

2.2.1. Boiled green plantain flour

The “False Horn” matured green plantain was washed and peeled to reveal the pulp. The pulp (8kg) was boiled in 5L of tap water, for twenty minutes at a temperature of 100°C. The boiled pulps were later sun-dried for about 3 weeks to a constant weight (3.2 kg), and ground into flour.

2.2.2. Roasted green plantain flour

The “False Horn” matured green plantain was roasted using the Nigerian traditional method. This is done by putting the pulps on wire gauze over red hot charcoal. The roasting was carried out by frequently turning the pulps to maintain even browning. Roasting was done for 10 minutes. The roasted plantains were also later sun-dried for about 3 weeks to a constant weight (4.1kg), and ground into flour. The samples were kept in air tight containers for future analysis.

2.3. Aqueous extract preparation

Ten g of each milled sample (boiled and roasted plantain flour) was soaked in 100 ml distilled water for about 24 h. The mixture was filtered. In a situation where the filtrate appeared to be very cloudy, the filtrate was centrifuged to obtain a clear supernatant liquid, which was subsequently used for the various assays (Obboh *et al.*, 2007). All antioxidant tests and analyses were performed in triplicate, and results were averaged.

2.4. Determination of total phenol content

The total phenol content was determined according to the method of Singleton *et al.* (1999). Briefly, 0.5ml of aqueous extracts were oxidized with 2.5ml 10% Folin-Ciocalteu’s reagent (v/v) and neutralized by 2.0ml of 7.5% sodium carbonate. The reaction mixture was incubated for 40min at 45°C and the absorbance was measured at 765nm in the spectrophotometer (JENWAY 6305). The total phenol content was subsequently calculated as gallic acid equivalent.

2.5. Determination of total flavonoid content

The total flavonoid content of the unripe plantain extracts was determined using method of Meda *et al.* (2005). The volume of 0.5 ml of sample/standard quercetin was mixed with 0.5 ml methanol, 50 μ l of 10% AlCl₃, 50 μ l 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 measured at 415 nm in the spectrophotometer (JENWAY 6305). Total flavonoid content was calculated using quercetin as a standard.

2.6. Determination of vitamin C content

Vitamin C content of the unripe plantain extracts was determined using the method of Benderitter *et al.* (1998). A volume of 75 µl DNPH (2g dinitrophenyl hydrazine, 230mg thiourea and 270mg CuSO₄·5H₂O in 100ml of 5M H₂SO₄) was added to 500 µl reaction mixture. The reaction mixture was subsequently incubated for 3 h at 37°C, then 0.5ml of 65% H₂SO₄ (v/v) was added to the medium and the absorbance was measured at 520nm using a spectrophotometer (JENWAY 6305).

The vitamin C content of the extracts was subsequently calculated; 2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid (ABTS) radical scavenging ability.

The ABTS• radical scavenging ability of both extracts was determined according to the method described by Re *et al.* (1999). ABTS• radical was generated by reacting an (7 mmol/l) ABTS• aqueous solution with K₂S₂O₈ (2.45 mmol/l, final concentration) in the dark for 16 h and adjusting the Abs 734nm to 0.700 with ethanol. 0.2ml of the sample extract was added to 2.0ml ABTS⁺ solution and the absorbance were measured at 734nm after 15mins using the spectrophotometer (JENWAY 6305). The trolox equivalent antioxidant capacity (TEAC) was subsequently calculated. During the ABTS⁺ reaction, antioxidants transfer an hydrogen atom to radical cation and causes discoloration of the solution Vincenzo *et al.*, 1999).

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

2.7. α-Amylase inhibition assay

The aqueous extracts volume (500 µl) and 500 µl of 0.02M sodium phosphate buffer (pH 6.9 with 0.006 M NaCl) containing Hog pancreatic α-amylase (EC 3.2.1.1) (0.5mg/ml) were incubated at 25°C for 10 minutes. Then, 500 µl of 1% starch solution in 0.02 M sodium phosphate buffer (pH 6.9 with 0.006M NaCl) was added to each tube. The reaction mixtures was incubated at 25°C for 10 minutes and stopped with 1.0 ml of dinitrosalicylic acid colour reagent. Thereafter, the mixture was incubated in a boiling water bath for 5 minutes, 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).

2.8. α-Glucosidase inhibition assay

The volume of the aqueous 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 2ml of Na₂CO₃ 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, 2007).

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

2.9. Reducing power

The reducing activity of the plantain extracts was determined by assessing the ability to reduce FeCl₃ solution as described by Pulido *et al.*, (2000). A volume of 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 then centrifuged at 805 g for 10 min. After centrifugation, 1ml of the supernatant was mixed with 1ml of distilled water and 0.1ml of (0.1%) ferric chloride. The absorbance was measured at 700 nm in the spectrophotometer (JENWAY 6305) after allowing the solution to stand for 30minutes. A graph of absorbance vs. concentration of extract was plotted to observe the reducing power where a higher absorbance values indicates a higher reducing power.

2.10. Angiotensin-I-converting enzyme (ACE) inhibition assay

The volume of the aqueous extract (50 µl) and ACE solution (50µl, 4 mU) were incubated at 37°C for 15 min. The enzymatic reaction was initiated by adding 150µl of 8.33 mM of the substrate Bz-Gly-His-Leu in 125 mM Tris-HCl buffer (pH 8.3) to the mixture. After incubation for 30 min at 37°C, the reaction was arrested by adding 250µl of 1M HCl. The cleaved Gly-His bond and the Bz-Gly produced by the reaction was extracted with 1.5 ml ethyl acetate. Thereafter the mixture was centrifuged to separate the ethyl acetate layer; then 1 ml of the ethyl acetate layer was transferred to a clean test tube and evaporated. The residue was redissolved in distilled water and its absorbance was measured at 228 nm using the spectrophotometer (JENWAY 6305). The percentage (%) enzyme inhibitory activity of the aqueous extracts was calculated (Cushman and Cheng, 1981).

2.11. Data analysis

The results of the three replicates were pooled and expressed as mean ± standard error. Student's t-test was carried out (Zar, 1984). Significance was accepted at P < 0.05. EC₅₀ (concentration of extract that will cause 50% reducing activity or enzyme inhibitory activity) was determined using linear regression analysis.

3. Results and Discussion

Polyphenols are considered to be strong antioxidants because of the redox properties of their hydroxyl groups (Materska and Perucka, 2005). The distribution of phenolic content in the green plantain products are presented in Table 1. The results revealed that the total phenol content of the boiled flour (93.12mg/100g) extract was significantly (P<0.05) higher than the roasted flour extract (89.06mg/100g). The difference in the total phenolics could be suggested to result from the difference

in their processing methods: Boiling (moist heat treatment) has been reported to enhance the quality of foods (Onwuliri *et al.*, 2004). Hence, this could have contributed to the release of more phenolics in boiled flour extract than in roasted flour extract (dry heat treatment).

Table 1. Total phenolic contents, flavonoid contents, ABTS and vitamin C contents of the aqueous extracts of roasted and boiled flour.

The test	Roasted	Boiled	P
Total phenol content mg/100g	89.06±0.03	93.12±0.01	<0.05
Total flavonoid content mg/100g	48.34±0.01	61.03±0.01	<0.05
ABTS mmol TEAC/100g	0.61±0.04	0.59±0.04	<0.05
Vitamin C mg/100g	379.21±0.30	247.04±0.04	<0.05

Values represent mean ± standard deviation, n = 3. P < 0.05

The results of the phenolic content of these plantain products were higher than that of some commonly consumed green leafy vegetables in Nigeria (Oboh, 2006). Antioxidant activity in higher plants has often been associated with phenolic compounds (Thabrew *et al.*, 1998). In addition to their roles in plants, phenolic compounds in our diet may provide health benefits associated with reduced risk of chronic diseases (Liu, 2004). Earlier report has also shown that phenolic compounds can protect the human body from free radicals, whose formation is associated with the normal metabolism of aerobic cells and that they are strong antioxidants capable of removing free radicals, they may chelate metallic catalysts, activate antioxidant enzymes, reduce α -tocopherol radicals and inhibit oxidases Amic *et al.*, 2003).

Furthermore, the flavonoid contents were significantly ($P < 0.05$) higher in the extract of boiled (61.03mg/100g) flour than in roasted extracts (Table 1). The trend in the total flavonoid contents agreed with the total phenolic contents result. Flavonoids are the largest group of phenolics. They have been identified in fruits, vegetables, and other plant parts and linked to reducing the risk of major degenerative diseases. Plantain is employed in the folklore management of diseases such as diabetes, ulcer and wound healing due to its hypoglycaemic, anti-ulcerogenic and analgesic properties (Ojewole and Adewunmi, 2003). In addition, recent report has revealed that the anti-ulcerogenic properties of plantain has been identified as the flavonoid leucocyanidin (Lewis and Shaw, 2001). Antioxidants fall into two mechanistic groups: those that inhibit or retard the formation of free radicals from their unstable precursors (initiation) and those that interrupt the radical chain reaction (propagation and branching). The former ones are called preventive antioxidants and the latter ones chain-breaking

antioxidants (Vinqvist and Barclay, 2000). The high antioxidant activity of the green plantain aqueous extracts followed the trend observed in the total phenol content (Table 1). This supports the arguments that antioxidant properties of plants food correlates with the phenolic content (Oboh and Shodehinde, 2009). Vitamin C has been reported to contribute to the antioxidant activities of plant food. Ascorbic acid is a good reducing agent and exhibits its antioxidant activities by electron donation (Oboh and Akindahunsi, 2004). The Vitamin C content of the unripe plantain extracts (Table 1) revealed that the roasted flour extract (379.21mg/100g) had higher vitamin C content than the cooked flour extract (247.04mg/100g). The Vitamin C content was higher when compared to that of green beans (*Phaseolus vulgaris* L) which had a Vitamin C content of 14.8 mg/100g (Jiratanan and Liu, (2004).

The antioxidant potential of a compound can be attributed to its radical scavenging and the ability of the plant extracts to serve as an antioxidant was measured by ABTS• radical scavenging ability (Re *et al.*, 1999). Table 1 depicts the ABTS• scavenging ability expressed as trolox equivalent antioxidant capacity (TEAC). The result revealed that there was no significant difference between the roasted and the boiled flour extract.

Ferric reducing antioxidant property assay has been reported to express the corresponding concentration of electron-donating antioxidants in which ferric-to-ferrous iron reduction occurs rapidly with all reductants with half reaction reduction potentials above that of Fe^{3+}/Fe^{2+} (Halvorsen *et al.*, 2002). The reducing power, shown by the ability of the green plantain extracts to reduce Fe^{3+} to Fe^{2+} , was determined. The ferric reducing power increases regularly with increasing amount of extract (Fig. 1) in a dose dependent manner in the range of 1.14–4.54mg/ml. For a given amount of extract, the higher the absorbance, the better is the reducing power. Both extracts exhibited a considerable reducing power.

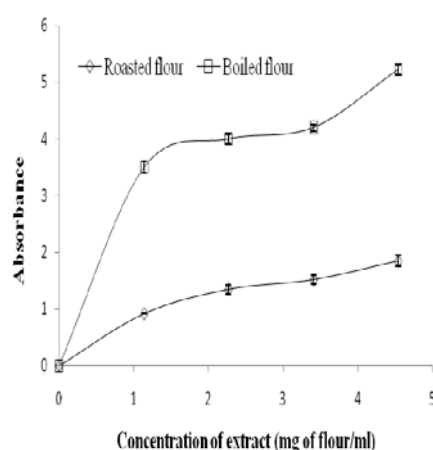


Figure 1. Reducing power of aqueous extract of roasted and boiled flour.

However, the boiled flour extract exhibited significantly higher ($P < 0.05$) reducing power than the roasted flour extract.

Correlating the reducing power with its EC_{50} (concentration of extract causing 50% reducing activity)

value (Table 2), it is observed that the boiled flour extract with lower EC_{50} had higher reducing activity. 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). Therefore, plantain phytochemicals such as phenolic compounds are strong reducing agents that could act by readily neutralizing free radicals through electron donation or hydrogen atom transfer.

Table 2. EC_{50} of enzyme inhibitory and antioxidant activities (mg/ml)

EC50 of enzyme inhibitory and antioxidant activities (mg/ml)		
α -amylase	α -glucosidase	Reducing power
7.34±0.12	7.27±0.07	104.98±6.27
5.91±0.11	5.78±0.13	35.84±2.89

Values represent mean \pm standard deviation, n = 3.

The interaction of plantain extracts with α -amylase, as shown in Fig. 2, revealed that both extracts caused a marked inhibition of pancreatic α -amylase activity in a dose dependent manner in the range of 2-8mg/ml. However, judging by the EC_{50} (extract concentration causing 50% enzyme inhibition) value (Table 2), the boiled flour extract had higher inhibitory activity than the roasted flour extract.

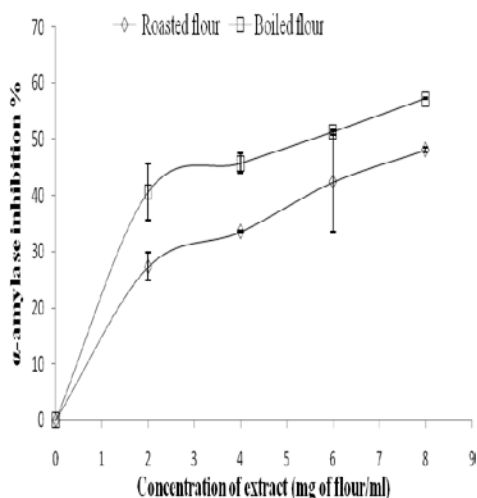


Figure 2. α -Amylase inhibitory activity of aqueous extract of roasted and boiled flour.

Postprandial hyperglycemia could induce the nonenzymatic glycosylation of various proteins and biomolecules; resulting in the development of chronic complications. Therefore, control of postprandial plasma glucose levels is critical in the early treatment of diabetes mellitus and in reducing chronic vascular complications (Ortiz *et al.*, 2007). Inhibition of enzymes involved in the metabolism of carbohydrates such as α -amylase and α -glucosidase is one of the therapeutic approaches for managing and controlling hyperglycemia (Shim *et al.*, 2003). The inhibition of α -amylase by the plantain extracts agreed with earlier reports on the inhibitory effect of

Allium species (Nickavar and Yousefian, 2009) and that of green and black tea on salivary α -amylase activity (Zhang and Kashket, 1998). Although the mechanisms underlying the mechanisms associated with the lowering effect of hyperglycemia in plant foods are presently not well understood, their phenolics might inhibit α -amylase activity *in vivo*. However, the effect of plantain phytochemicals plus its reducing property might be suggested to be responsible for the display of its higher α -amylase inhibitory activity.

Furthermore, the ability of the plantain extracts to inhibit α -glucosidase activity *in vitro* was investigated and the result is presented in Fig. 3.

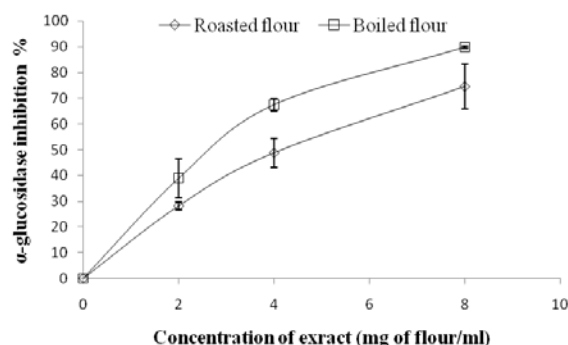


Figure 3. α -Glucosidase inhibitory activity of aqueous extract of roasted and boiled flour.

Both extracts exhibited a dose-dependent enzyme inhibitory activity in the range of 2-8 mg/ml of the extract tested. However, the boiled flour extract exhibited significantly ($P < 0.05$) higher inhibitory activity than the roasted flour extract, when taking into account the EC_{50} values of the aqueous extracts (Table 2). The results of the enzymes (α -amylase and α -glucosidase) inhibitory assays did agree with the phenolic contents and reducing power activity of both plantain extracts. The inhibition of α -glucosidase activity slows the breakdown of disaccharide to simple glucose, thereby reducing the rate at which glucose is absorbed into the blood stream (Kwon *et al.*, 2007). This is in agreement with earlier research revealing the inhibition of pancreatic α -amylase and α -glucosidase by different classes of phenolic compounds (Nickavar and Yousefian, 2009). Phenol-rich plant foods have been reported to exhibit pancreatic α -amylase and α -glucosidase inhibitory activities *in vitro* (Kwon *et al.*, 2007). On comparing the inhibition of α -amylase and α -glucosidase, both phenolic extracts were stronger inhibitors of α -glucosidase than α -amylase; this weak inhibition of α -amylase when compared to α -glucosidase is of great pharmaceutical importance, in addressing some of the side effects associated with the drugs (Acarbose and Voglibose) presently used for the management of diabetes. These drugs give some side effects which include abdominal distention, flatulence, meteorism, and possibly diarrhea (Bischoff, 1994). Such adverse effects might occur due to the excessive pancreatic α -amylase inhibition, which results in the abnormal bacterial fermentation of undigested carbohydrates in the colon (Hori *et al.*, 1987).

Angiotensin-1 converting enzyme (ACE) cleaves angiotensin I to produce angiotensin II, a powerful vasoconstrictor that has been identified as a major factor in hypertension (Villar *et al.*, 1986). ACE inhibitors was

discovered in snake venom and has since been widely developed to prevent angiotensin II production in cardiovascular diseases and are therefore utilized in clinics for treatments related to high blood pressure (Villar *et al.*, 1986). Much has not really been reported about the inhibitory activities of unripe plantain on enzymes linked to type 2 diabetes and hypertension but this present study has extended its research to investigate the effect of green plantain on angiotensin 1 converting enzyme (ACE) *in vitro*. Fig. 4 depicts angiotensin 1 converting enzyme inhibitory activity.

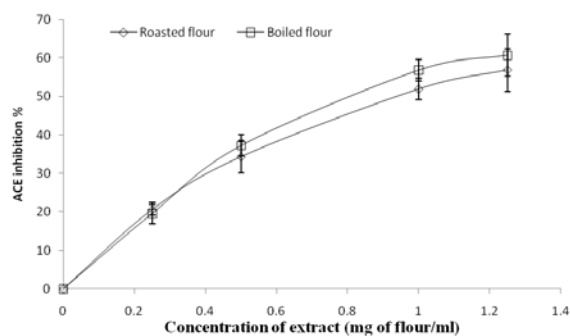


Figure 4. Angiotensin-i-converting enzyme (ACE) inhibitory activity of the aqueous extract of roasted and boiled flour.

The plantain products inhibited ACE activity in a dose-dependent manner in the range of 0.25-1.25mg/ml of the extract tested. However, the boiled flour extract showed higher ACE inhibitory activities than the roasted flour extract. The results of the ACE inhibition assay clearly indicate that the extracts from the green plantain inhibited ACE especially the boiled plantain flour extract which displayed higher total phenol, total flavonoid, reducing power and enzyme inhibitory activities. This higher activity could be suggested to be due to the moisture treatment it was subjected to thereby leading to the release of more phenolics than the roasted (dry treatment) flour extract. The results of this study could eventually explain the mechanism of action of plantain in the management of postprandial hyperglycemia and one of its long term complications.

4. Conclusion

This study revealed that the inhibition of key enzymes linked to non-insulin diabetes mellitus (α -amylase and α -glucosidase) and hypertension (Angiotensin I-converting enzymes) could be part of the mechanism through which the green plantain (which is widely taken by Nigerian diabetics) manage/prevents diabetes and hypertension. There is a high likelihood that plantain may provide the types of nutritional and health benefits associated with its consumption in general. Thereby making it likely candidate for bioactively useful phyto-molecule for the management of NIDDM with minimum side effects.

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