

Journal of Taibah University Medical Sciences Volume 11, Issue 3, June 2016, Pages 236-242

Original Article

# In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the $\alpha$ -amylase and $\alpha$ -glucosidase inhibitory properties of acarbose

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#### Abstract

#### Objective

This study sought to determine the antioxidant activities of African birch leaf, to assess its interaction with key enzymes relevant to type 2 diabetes ( $\alpha$ -amylase and  $\alpha$ -glucosidase) and to evaluate its effect on acarbose *in vitro*.

#### Methods

One milligram per milliliter of aqueous extract of African birch and acarbose were separately prepared. At the same time, both the African extract and acarbose solution (50:50 v/v) were thoroughly mixed until homogeneity was attained. The phenolic phytoconstituents and antioxidant properties of African birch leaf were subsequently determined. Finally, the effects of African birch extract, acarbose solution and a mixture of acarbose and African birch extract on  $\alpha$ -amylase and  $\alpha$ -glucosidase activities were assessed *in vitro*.

Results

The results showed that Afric exemplified by its radical scavenging abilities,  $Fe^{2+}$  chelating ability and prevention of lipid peroxidation. Acarbose had significantly (p < 0.05) higher  $\alpha$ -amylase (IC<sub>50</sub> = 11.77 µg/ml) and  $\alpha$ glucosidase (IC<sub>50</sub> = 9.05 µg/ml) activities compared to African birch extract [ $\alpha$ -amylase (IC<sub>50</sub> = 242.17 µg/ml);  $\alpha$ -glucosidase (IC<sub>50</sub> = 196.35 µg/ml)]. However, the combination of acarbose and African birch extract showed an additive effect on  $\alpha$ -amylase inhibition, while a resultant synergistic action was observed against  $\alpha$ -glucosidase inhibition.

## Conclusion

The additive and synergistic actions of the combination of African birch extract and acarbose solution suggest effective, complementary and alternative strategies towards the management/treatment of hyperglycaemia associated with type 2 diabetes.



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# Keywords

African birch; Acarbose;  $\alpha$ -Amylase;  $\alpha$ -Glucosidase; Leaf; Type 2 diabetes

# Introduction

Diabetes is a group of metabolic diseases characterized by hyperglycaemia, which results from defects in insulin resistance, insulin secretion and beta cell dysfunction.<sup>1</sup> Of all of the diagnosed cases of diabetes, NIDDM accounts for approximately 90–95%.<sup>2</sup> It has been linked to several risk factors involving lifestyle, such as smoking, obesity, poor diet and physical inactivity.<sup>3</sup>  $\alpha$ -Amylase and  $\alpha$ -glucosidase are critical enzymes involved in starch hydrolysis and are implicated in the onset of type 2 diabetes (T2D).<sup>4</sup> Pancreatic  $\alpha$ -amylase catalyzes the hydrolysis of starch to a mixture of oligosaccharides and disaccharides, while intestinal  $\alpha$ -glucosidase breaks down disaccharides and oligosaccharides into glucose which, upon absorption, enters the bloodstream.<sup>5</sup> Degradation of these complex carbohydrates is very fast and may result in an elevated blood glucose level after a meal.<sup>6</sup> Synthetic oral antidiabetic agents such as acarbose and miglitol elicit their therapeutic effects through the inhibition of these enzymes, but their administration is associated with serious side effects and a failure to alleviate diabetic complications.<sup>7</sup> Recently, interest in the use of medicinal plants in the form of traditional medicine has increased all over the world, especially in Africa, due to their cheap cost, availability and effectiveness.<sup>8</sup>

Nigeria has a wide range of p

pharmacological relevance. A  $\equiv$  Outline  $\bigotimes$  Download Share Export tropical plant with large ecological abstruction across ratios. The plant parts (real, etc.), each and root) are used in traditional medicine for the treatment of some ailments because they have been reported to possess antimicrobial, hepatoprotective and antioxidant properties.10, 11 A previous study by Etuk and Mohammed<sup>12</sup> revealed a spontaneous decrease in blood glucose levels in alloxan-induced diabetic rats administered African birch extract. However, there is a dearth of information on the possible mechanism by which the plant elicits its antidiabetic effects.

Consequently, we aimed to assess the antioxidant activities and interaction between aqueous African birch leaf extract with key enzymes linked to starch hydrolysis. Additionally, considering the combinatorial usage of traditional plants and synthetic drugs in Nigeria, it is pertinent to evaluate and compare the effects of this plant to acarbose (a known antidiabetic drug) *in vitro*.

## Materials and Methods

#### Materials

#### Sample collection

African birch leaf was purchased from local traditional healers at the Erekasan Market in Akure Metropolis of Ondo State, Nigeria. Authentication of the plant was performed by the Crop, Soil and Pest Management (CSP) Department at the Federal University of Technology, Akure (FUTA), Nigeria (Voucher specimen no: FUTA/HB/0112AL1).

#### Chemicals and reagents

All chemicals and reagents used in this study were of analytical grade, and glass-distilled water was used.

#### Aqueous extraction of African birch leaf and acarbose

African birch leaf was thoroughly washed under running distilled water, air dried, ground into powder and kept dry in an air-tight container prior to extraction according to the method of Adefegha and Oboh.<sup>13</sup> The sample (5 g) was weighed, soaked in 100 ml of distilled water and left for 24 h. The mixture was filtered after 24 h and the filtrate kept in a refrigerator below 0 °C. The frozen extract solution was recovered as dried extract under pressure of approximately 200–450 mbar at 40 °C. The dried extract was reconstituted in distilled water (1 mg/ml) and used for subsequent analysis. Similarly, a stock concentration of 1 mg/ml acarbose was prepared and later diluted for subsequent use.

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Phenolic characterization

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## Determination of total phenome content

Briefly, 1 mg/ml of extract was oxidized with 2.5 ml of 10% Folin–Ciocalteu's reagent (v/v) and neutralized with 2.0 ml of 7.5% sodium carbonate. The reaction mixture was incubated for 40 min at 45 °C and absorbance was measured at 765 nm using a UV/Visible Spectrophotometer, 6315 Series (Jenway, Chelmsford, Essex CM1 3UP, England). Total phenolic content was subsequently calculated as the gallic acid equivalent.<sup>14</sup>

## Determination of total flavonoid content

Briefly, 0.5 ml of extract (1 mg/ml) was mixed with 0.5 ml of methanol, 50 µl of 10% AlCl<sub>3</sub>, 50 µl of 1 M potassium acetate and 1.4 ml of water and incubated at room temperature for 30 min. Absorbance of the reaction mixture was subsequently measured at 415 nm, and total flavonoid content was calculated and expressed as the quercetin equivalent.<sup>15</sup>

# Quantification of phenolic compounds by HPLC-DAD

Reverse phase chromatographic analyses were carried out under gradient conditions using a  $C_{18}$ column (4.6 mm  $\times$  150 mm) packed with 5-µm diameter particles; the mobile phase was water containing 1% formic acid (A) and acetonitrile (B), and the composition gradient was as follows: 13% of B until10 min and then changed to obtain 20%, 30%, 50%, 60%, 70%, 20% and10% B at 20, 30, 40, 50, 60, 70 and 80 min, respectively, following the method described by Adefegha et al.,<sup>16</sup> with slight modifications. African birch extract and the mobile phase were filtered through a 0.45-µm membrane filter (Millipore) and then degassed in an ultrasonic bath prior to use. African birch extract was analyzed at a concentration of 20 mg/ml. The flow rate was 0.7 ml/min, and the injection volume was 40 µl. The wavelengths were 254 nm for gallic acid; 280 nm for catechin and epicatechin; 325 nm for chlorogenic, caffeic and ellagic acids; and 365 nm for quercetin, quercitrin, rutin and kaempferol. All chromatography operations were carried out at ambient temperature and in triplicate.

## In vitro antioxidant studies

# 1,1-Diphenyl-2 picrylhydrazyl free radical scavenging ability

The free radical scavenging ability of the plant extract against DPPH (1,1-diphenyl-2 picrylhydrazyl) was evaluated as described by Gyamfi et al.  $^{17}$  Briefly, the extracts (0–500  $\mu g/ml)$ were mixed with 1 ml of 0.4 mM methanolic solution containing DPPH radicals. The mixture was left in the dark for 30 min and absorbance was read at 516 nm. DPPH free radical scavenging ability was subsequently calculated and expressed as a percentage (%).

## Fenton reaction (degradation of deoxyribose)

The method of Halliwell and

to prevent the Fe<sup>2+</sup>/H<sub>2</sub>O<sub>2</sub>-ind  $\stackrel{:}{\models}$  Outline  $\stackrel{!}{\triangleright}$  Download Share Export added to a reaction mixture communication of the provide and the provide of 800 µl with distilled water. The reaction mixture was incubated at 37 °C for 30 min and the reaction 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 solution. The tubes were subsequently incubated in boiling water for 20 min and absorbance was measured at 532 nm. The OH radical scavenging ability was subsequently calculated and expressed as a percentage (%).

## Fe<sup>2+</sup> chelation assay

The Fe<sup>2+</sup> chelating ability of the extract was determined using a modified method of Minotti and Aust.<sup>19</sup> One hundred fifty microliters of freshly prepared FeSO<sub>4</sub> (500  $\mu$ M) was added to a reaction mixture containing 168  $\mu$ l of 0.1 M Tris–HCl (pH 7.4), 218  $\mu$ l of saline and the extracts (0–200  $\mu$ g/ml). The reaction mixture was incubated for 5 min before the addition of 13  $\mu$ l of 0.25% 1,10-phenanthroline (w/v). Absorbance was subsequently measured at 510 nm in a Jenway UV/Visible spectrophotometer, 6315 series. The Fe<sup>2+</sup> chelating ability was calculated and expressed as a percentage (%).

#### Lipid peroxidation and thiobarbituric acid reactions

Wistar rats were anaesthetized in mild diethyl ether, and the pancreases were excised. Pancreatic tissue was rapidly rinsed in saline, placed on ice and weighed. The tissue was subsequently homogenized in cold saline (1/10 w/v) with approximately 10 up-and-down strokes in a Teflon glass homogenizer. The homogenate was centrifuged for 10 min at  $3000 \times g$  to yield pellets that were discarded, and low-speed supernatant (Sl) was obtained. The lipid peroxidation assay was carried out using the modified method of Ohkawa et al.<sup>20</sup> Briefly 100 µl of the S1 fraction was mixed with a reaction mixture containing 30 µl of 0.1 M Tris–HCl buffer (pH 7.4), extracts (0–312.50 µg/ml) and 30 µl of 250 µM freshly prepared FeSO<sub>4</sub>. The volume was brought up to 300 µl with distilled water before incubation at 37 °C for 1 h. The color reaction was developed by adding 300 µl of 8.1% sodium dodecyl sulfate to the reaction mixture containing S1. This was subsequently followed by the addition of 600 µl acetic acid/HCl (pH 3.4) and 600 µl of 0.8% thiobarbituric acid. This mixture was incubated at 100 °C for 1 h and absorbance was read at 532 nm. Lipid peroxidation was subsequently calculated as the amount of malondialdehyde (MDA) produced (% of control).

#### Enzyme inhibition assay

#### $\alpha$ -Amylase assay

 $\alpha$ -Amylase activity was determ

birch extract (0–0.8 mg/ml) of E Outline Download Share Export (pH 6.9 with 0.006 M NaCl) community roop particulate a unique (10 0.001 mg/ml) were incubated at 25 °C for 10 min. Then, 50 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 were incubated at 25 °C for 10 min and stopped with 100  $\mu$ l dinitrosalicylic acid color 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 1.0 ml distilled water, and absorbance was measured at 540 nm.  $\alpha$ -Amylase inhibitory activity was subsequently calculated and expressed as a percentage (%).

#### $\alpha$ -Glucosidase inhibition assay

α-Glucosidase activity was determined by the modified method described by Oboh et al.<sup>4</sup> Fifty microliters of African birch extract (0–0.8 mg/ml) or acarbose (0–80 µg/ml) and 100 µl of α-glucosidase solution (1.0 U/ml) in 0.1 M phosphate buffer (pH 6.9) were incubated at 25 °C for 10 min. Then, 50 µl of 5 mM p-nitrophenyl-α-p-glucopyranoside solution in 0.1 M phosphate buffer (pH 6.9) was added. The mixtures were incubated at 25 °C for 5 min before reading the absorbance at 405 nm. α-Glucosidase inhibitory activity was calculated and expressed as percentage (%) inhibition.

#### Statistical analysis

The mean values from triplicate experiments were pooled and expressed as the mean  $\pm$  standard deviation (STD). One-way analysis of variance was used to analyze the results. Tukey's test was used for the post hoc analysis, and the least significance difference (LSD) was carried out. GraphPad Prism 6 software was used for the analysis. The IC<sub>50</sub> (extract concentration causing 50% enzyme inhibitory/antioxidant activity) values were determined using non-linear regression analysis.

#### Results

## Spectrophotometric and HPLC-DAD analysis of phenolics in African birch extract

Table 1 lists the total phenolic contents of African birch leaf. The results show a total phenolic content of 6.87 mg GAE/g and a total flavonoid content of 4.38 mg QUE/g. Furthermore, HPLC-DAD analysis was performed on African birch leaf extract to identify and quantify the presence of phenolic acids and flavonoids (Figure 5). The results indicate that the plant extract contains chlorogenic acid (27.19  $\pm$  0.04 mg/g) and quercetin (14.69  $\pm$  0.03 mg/g) as the predominant phenolic compounds. Other phenolic compounds found in African birch include catechin (8.45  $\pm$  0.08 mg/g), kaempferol (5.72  $\pm$  0.11 mg/g), rutin (5.43  $\pm$  0.10 mg/g), epicatechin

 $(3.17 \pm 0.06 \text{ mg/g})$ , ellagic acid  $(1.36 \pm 0.04 \text{ mg/g})$  and caffeic  $\blacksquare$  Outline  $\square$  Download Share Export

#### Table 1. The total phenol and flavonoid content of African birch leaf.

Phenolic contents	African birch
Total phenolic (mg/GAE g)	6.87 ± 0.45
Total flavonoid (mg/QUE g)	4.38 ± 0.33

Table 2. Quantification of phenolic compounds by HPLC-DAD in African birch leaf extract.

Compounds	African birch		LOD	LOQ
	mg/g	%	µg/mL	µg/mL
Gallic acid	$2.97 \pm 0.07^{e}$	0.29	0.019	0.067
Catechin	$8.45 \pm 0.08^{\circ}$	0.84	0.024	0.081
Chlorogenic acid	$27.19 \pm 0.04^{a}$	2.71	0.032	0.039
Caffeic acid	$1.14 \pm 0.05^{g}$	0.11	0.015	0.049
Ellagic acid	$3.08 \pm 0.08^{e}$	0.30	0.025	0.083
Epicatechin	$3.17 \pm 0.06^{e}$	0.31	0.016	0.057
Rutin	$5.43 \pm 0.10^{d}$	0.54	0.028	0.096
Quercitrin	$1.36\pm0.04^{f}$	0.13	0.013	0.042
Quercetin	$14.69 \pm 0.03^{b}$	1.46	0.009	0.027
Kaempferol	$5.72 \pm 0.11^{d}$	0.57	0.034	0.112

Results are expressed as mean ± standard deviations (SD) of three determinations.

Values with different superscript alphabets (a–g) along the same column are significantly (p < 0.01) different using the Tukey test.

In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the α-amylase and α-glucosidase inhibitory pro...

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The free radical scavenging a  $\bigcirc$  Outline  $\bigcirc$  Download Share Export Table 3. The results showed that the leaf extract (IC<sub>50</sub> = 348.25 µg/ml) scavenged DPPH. Additionally, the OH\* scavenging ability of the plant sample was determined; the plant extract (IC<sub>50</sub> = 183.14 µg/ml) scavenged OH•. In a similar manner, the plant chelated Fe<sup>2+</sup> with an IC<sub>50</sub> value of 187.45 µg/ml. Incubation of pancreas homogenates in the presence of Fe<sup>2+</sup> caused a significant increase (p < 0.05) in MDA content. However, African birch extract inhibited MDA production with an IC<sub>50</sub> value of 174.00 µg/ml, as presented in Table 3.

Table 3. IC<sub>50</sub> Values for the DPPH and OH radical scavenging ability, Fe<sup>2+</sup> chelating ability, inhibition of FeSO<sub>4</sub> induced MDA production in rat's pancreas *in vitro*,  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activities of African birch and acarbose.

Assays	Samples		
	African birch (µg/ml)	Acarbose (µg/ml)	
DPPH•	348.25 ± 3.83	N.D	
OH•	183.14 ± 4.16	N.D	
Iron chelation	187.45 ± 3.82	N.D	
Lipid peroxidation	$174.00 \pm 2.78$	N.D	
α-Amylase	242.17 ± 4.20*	11.77 ± 1.25**	
α-Glucosidase	196.35 ± 3.30*	9.05 ± 1.33**	

Values represent means ± standard deviation of triplicate readings.

Values with asterisk (\*) and (\*\*) along the same row are significantly different (p < 0.05).

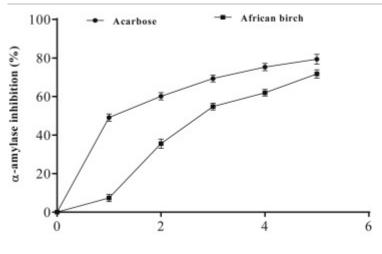
N.D; Not determined.

## $\alpha\text{-}Amylase$ and $\alpha\text{-}glucosidase$ inhibitory activities

The  $\alpha$ -amylase inhibitory properties of acarbose and African birch extract were assessed and are presented in Figure 1. The results revealed that both acarbose and the leaf extract inhibited  $\alpha$ -amylase activity in a concentration-dependent manner. Acarbose (IC<sub>50</sub> = 11.77 µg/ml) had a significantly (p < 0.05) higher  $\alpha$ -amylase inhibitory effect than African birch extract (IC<sub>50</sub> = 242.17 µg/ml). The *in vitro* effect of the combination of African birch leaf extract with

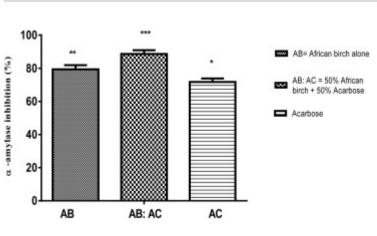
acarbose on  $\alpha$ -amylase inhibi

synthetic drug (acarbose) and  $\equiv$  Outline  $\square$  Download Share Export (p < 0.05) increase in  $\alpha$ -amylate matter q accuracy compared with the plane entraction, and synthetic drug (acarbose) only. Likewise, the  $\alpha$ -glucosidase inhibitory abilities of the plant, acarbose and their combination are presented in Figure 3, Figure 4, and IC<sub>50</sub> values are listed in Table 3. The results revealed that acarbose (IC<sub>50</sub> = 9.05 µg/ml) had significantly (p < 0.05) higher  $\alpha$ glucosidase inhibitory activity than African birch extract (IC<sub>50</sub> = 196.35 µg/ml). Additionally, the combination of African birch extract and acarbose (50:50 v/v) did not cause a significant (p > 0.05) change in  $\alpha$ -glucosidase inhibitory activity compared with African birch only and acarbose only.



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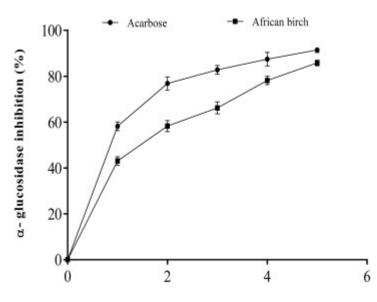
Figure 1. Inhibition of  $\alpha$ -amylase activities (%) by African birch leaf extract and acarbose. The concentrations of the extract used for the plot are 0.00, 0.20, 0.40, 0.6 and 0.80 mg/mL for the values 0, 1, 2, 3 and 4 respectively as indicated on the x-axis of the graph. The concentrations of the acarbose used for the plot are 0.00, 20.00, 40.00, 60.00 and 80.00 µg/mL for the values 0, 1, 2, 3 and 4 as shown on the x-axis of the graph. Values represent mean of standard deviation of triplicate readings.



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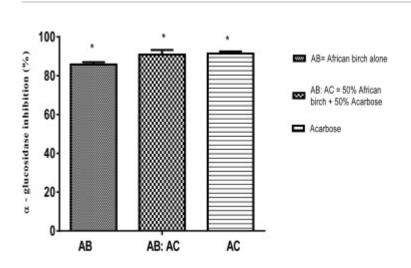
Figure 2. Inhibition of  $\alpha$ -amy  $\equiv$  Outline  $\square$  Download Share Export African birch leaf extract (509

(\*\*\*)] on bar chart indicate significant (p < 0.05) difference between African birch leaves, acarbose and mixture of acarbose and African birch.



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Figure 3. Inhibition of  $\alpha$ -glucosidase activities (%) by African birch leaf extract and acarbose. The concentrations of the extract used for the plot are 0.00, 0.20, 0.40, 0.6 and 0.80 mg/mL for the values 0, 1, 2, 3 and 4 respectively as indicated on the x-axis of the graph. The concentrations of the acarbose used for the plot are 0.00, 20.00, 40.00, 60.00 and 80.00 µg/mL for the values 0, 1, 2, 3 and 4 as shown on the x-axis of the graph. Values represent mean of standard deviation of triplicate readings.



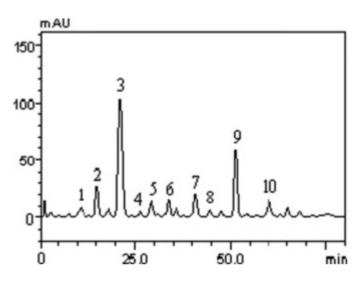
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Figure 4. Inhibition of  $\alpha$ -glucosidase activities (%) by African birch leaf extract, combination of African birch leaf extract (50%) and acarbose (50%) and Acarbose alone. (\*) on the bar chart indicates non significant (p > 0.05) difference.



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Figure 5. Representative high performance liquid chromatography profile of African birch leaf. Gallic acid (peak 1), catechin (peak 2), chlorogenic acid (peak 3), caffeic acid (peak 3), ellagic acid (peak 5), epicatechin (peak 6), rutin (peak 7), quercitrin (peak 8), quercetin (peak 9) and kaempferol (peak 10).

#### Discussion

#### Total phenolic and flavonoid contents

One of the most abundant antioxidants in plants is polyphenols, which are widely distributed across several food products such as herbs, vegetables, cereals, olive, dry legumes, chocolate and beverages.<sup>22</sup> In this study, the total phenol content of African birch was lower than that reported in a previous study from our laboratory on *Crassocephalum crepidioides* leaf, but it had a higher total flavonoid content than that of *C. crepidioides*.<sup>23</sup> Additionally, the phenolic content of African birch was significantly (p < 0.05) higher than that of *Celosia* spp, as reported by Molehin et al.<sup>24</sup>

Antioxidant properties of African birch leaf

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hyperglycaemia, causes oxida 🗮 Outline 🔀 Download Share Export of diabetic complications.<sup>25</sup> 7 ... use or antionical suppression in reducing are revered.

oxidative stress and in slowing or preventing the development of complications associated with diseases have shown promising therapeutic effects.4, 5, 16, 26 Many synthetic antioxidant components have shown toxic and/or mutagenic effects. Hence, much attention has been given to naturally occurring antioxidants.<sup>26</sup> Numerous plant constituents have shown free radical scavenging or antioxidant activity.14, 23, 24, 26 The free radical scavenging abilities, prevention of iron chelating ability and lipid peroxidation (Table 3) exhibited by African birch may be part of the mechanism through which African birch leaf exerts its biological activities, which may be responsible for its use in folkloric medicine as an antidiabetic agent.

Effect of African birch leaf extract on the inhibitory properties of a carbose against  $\alpha$ -amylase and  $\alpha$ -glucosidase activities

The inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase activities by African birch extract suggests the scientific basis for its folkloric use in the management/treatment of diabetes and could be of great importance in the development of a modern therapeutic in the management of T2D.4, 16 Furthermore, chlorogenic acids and quercetin were abundant compared with other phenolics found in African birch leaf. A recent report showed that caffeic and chlorogenic acids are good inhibitors of  $\alpha$ -amylase and  $\alpha$ -glucosidase activities.<sup>27</sup> Therefore, we suggest that the inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase activities by plants and plant foods may be attributed to their phenolic constituents.23, 27 This agrees with some studies where the inhibition of key enzymes ( $\alpha$ -amylase and  $\alpha$ -glucosidase) relevant to T2D by plants has been suggested as a therapeutic approach in the management of this disease, and some vital bioactive compounds, including polyphenols, may possess interesting structure-function benefits and promising potential.5, 16 The increased inhibition in  $\alpha$ -amylase activity by acarbose and African birch extract when combined (50:50) resulted in an additive effect (Figure 2), whereas the combination of both acarbose and leaf extract caused a synergistic effect on their inhibition of  $\alpha$ -glucosidase *in vitro* (Figure 4). The herb–food interaction may have a beneficial effect by increasing drug efficacy or diminishing potential side effects, as this was validated in our results (Figure 2, Figure 4). Findings from this study correlate with those from an earlier study by Boath et al.,<sup>22</sup> who affirmed that synergies between acarbose and berry extracts in diabetes treatment/management may enhance the hypoglycaemic effect and the inhibition of  $\alpha$ -glucosidase activity.

#### Conclusion

This study reports the antioxidant activities of African birch leaf and the additive actions and synergistic interaction between African birch leaf and acarbose in inhibiting  $\alpha$ -amylase and  $\alpha$ -glucosidase activities, respectively. However, these effects could be attributed to some phenolic components in African birch leaf.

Conflict of interest

The authors have no conflict  $\Xi$  Outline

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# Funding

This study was funded by the Education Trust Fund Academic Staff Training and Development (AST & D) Intervention (Ref: FUTA/VCPU/ETF/155).

# Ethical approval

The ethics committee of the School of Sciences, Federal University of Technology, Akure, Nigeria gave approval for the use of laboratory animals (Ref: FUTA/SOS/1399). The ethical regulations are in accordance with national and institutional guidelines for the protection of animal welfare during experiments.

# Authors' contributions

SAA and GO conceived the idea and designed the work, interpreted the data, performed the statistical analysis, purchased chemicals and reagents needed for the experiments and addressed the reviewers' comments. OSO, TOJ and SIO were students who participated in all experimental analyses and made the initial draft of the paper.

Recommended articles Citing articles (6)

## References

World Health Organization
 Definition, diagnosis and classification of diabetes mellitus and its complications
 Report of WHO Consultation
 WHO, Geneva (1999), p. 66
 Google Scholar

- C. Willi, P. Bodenmann, W.A. Ghali, F.D. Faris, J. Cornuz
  Active smoking and the risk of type 2 diabetes: a systematic review and meta-analysis
  J Am Med Assoc, 298 (2007), pp. 2654-2664
  CrossRef View Record in Scopus Google Scholar
- S. Wild, G. Roglie, A. Gracia, R. Sacre, H. King
  Global prevalence of diabetes
  Diabetes Care, 27 (2004), pp. 1047-1053
  CrossRef View Record in Scopus Google Scholar

1/30/2020	In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the α-amylase and α-glucosidase inhibitory pro			
4	G. Oboh, A.J. Akinyem			
	Inhibitory effect of aq \Xi Outline  Download Share Export			
	type-2 diabetes			
	J Food Nutr Res, 49 (2010), pp. 14-20			
	View Record in Scopus Google Scholar			
5	Y.I. Kwon, E. Apostolidis, Y.C. Kim, K. Shetty			
	Health benefits of traditional corn, beans and pumpkin: <i>in vitro</i> studies for hyperglycemia			
	and hypertension management			
	J Med Food, 10 (2) (2007), pp. 266-275			
	CrossRef View Record in Scopus Google Scholar			
6	S. Kumar, S. Narwal, V. Kumar, O. Prakash			
	Alpha-glucosidase inhibitors from plants: a natural approach to treat diabetes			
	Pharmacog Rev, 5 (2011), pp. 19-29			
	View Record in Scopus Google Scholar			
7	A.Y. Cheng, I.G. Fantus			
	Oral antihyperglycemic therapy for type 2 diabetes mellitus			
	Can Med Assoc J, 172 (2005), pp. 213-226			
	CrossRef View Record in Scopus Google Scholar			
8	A.A. Elujoba, O.M. Odeleye, C.M. Ogunyemi			
	Traditional medicine development for medical and dental primary health care delivery			
	system in Africa			
	Afr J Tradit Complement Altern Med, 2 (1) (2005), pp. 46-61			
	View Record in Scopus Google Scholar			
9	M. Sacande, S. Sanogo			
	Anogeissus leiocarpus (DC.) Guill and Perr			
	Seed Leafl (2007), p. 119			
	Google Scholar			
10	K. Batawila			
	Antifungal activities of five Combretaceae used in Togolese traditional medicine			
	Fitoterap, 76 (2005), pp. 264-268			
	Article 🔀 Download PDF View Record in Scopus Google Scholar			
11	S.E. Atawodi, O.O. Adekunle, I. Bala			
	Antioxidant, organ protective and ameliorative properties of methanol extract of			
	Anogeissus leiocarpus leaf against carbon tetrachloride-induced liver injury			

Int J Pharm Sci Res, 2 (6) (2011), pp. 1443-1448

View Record in Scopus Google Scholar

1/30/2020	In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the α-amylase and α-glucosidase inhibitory pro
12	E.U. Etuk, B.J. Moham
12	Informant consensus 🗮 Outline  Download Share Export
	in north western Nigera for the treatment of anyotes mentus
	Afr J Pharm Pharmacol, 3 (2009), pp. 496-500
	View Record in Scopus Google Scholar
13	S.A. Adefegha, G. Oboh
	Enhancement of total phenolics and antioxidant properties of some tropical green leafy
	vegetables by steam cooking
	J Food Process Preserv, 35 (2011), pp. 615-622
	CrossRef View Record in Scopus Google Scholar
14	V.L. Singleton, R. Orthofor, R.M. Lamuela-Raventos
	Analysis of total phenols and other oxidation substrates and antioxidants by means of
	Folin-Ciocalteu reagent
	Method Enzymol, 299 (1999), pp. 152-178
	Article 🐒 Download PDF View Record in Scopus Google Scholar
15	A. Meda, C.E. Lamien, M. Romito, J. Millogo, O.G. Nacoulma
	Determination of the total phenolic, flavonoid and proline contents in Burkina Faso
	honey, as well as their radical scavenging activity
	Food Chem, 91 (2005), pp. 571-587
	View Record in Scopus Google Scholar
16	S.A. Adefegha, G. Oboh, O.M. Adefegha, A.A. Boligon, M.L. Athayde
	Antihyperglycemic, hypolipidemic, hepatoprotective and antioxidative effects of dietary
	clove (Szyzgium aromaticum) bud powder in a high-fat diet/streptozotocin-induced
	diabetes rat model
	J Sci Food Agric, 94 (2014), pp. 2726-2737
	CrossRef View Record in Scopus Google Scholar
17	M.A. Gyamfi, M. Yonamine, Y. Aniya
	Free-radical scavenging action of medicinal herbs from Ghana: Thonningia sanguinea on
	experimentally-induced liver injuries
	Gen Pharmacol, 32 (1999), pp. 661-667
	Article 📆 Download PDF View Record in Scopus Google Scholar
18	B. Halliwell, J.M.C. Gutteridge
	Formation of a thiobarbituric-acid- reactive substance from deoxyribose in the presence
	of iron salts: the role of superoxide and hydroxyl radicals
	FEBS Lett, 128 (1981), pp. 347-352
	Article 📆 Download PDF CrossRef View Record in Scopus Google Scholar

1/30/2020	In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the α-amylase and α-glucosidase inhibitory pro
19	G. Minnoti, S.D. Aust
	An investigation into 📜 Outline  Download Share Export
	Free Rad Biol Med, 3 (1997), pp. 379-309
	Google Scholar
20	H. Ohkawa, N. Ohishi, K. Yagi
	Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction
	Anal Biochem, 95 (1979), pp. 351-358
	Article 📆 Download PDF View Record in Scopus Google Scholar
21	V. Worthington
	Alpha amylase
	K. Worthington, V. Worthington (Eds.), Worthington enzyme manual, Worthington
	Biochemical Corp, Freehold NJ (1993), pp. 36-41
	View Record in Scopus Google Scholar
22	A.S. Boath, D. Stewart, G.J. McDougall
	Berry components inhibit $\alpha$ -glucosidase in vitro: synergies between acarbose and
	polyphenols from black currant and rowanberry
	Food Chem, 132 (2012), pp. 929-936
	Article 📆 Download PDF View Record in Scopus Google Scholar
23	S.A. Adefegha, G. Oboh, O.R. Molehin, J.A. Saliu, M.L. Athayde, A.A. Boligon
	Chromatographic fingerprint analysis, acetylcholinesterase inhibitory properties and
	antioxidant activities of redflower ragleaf (Crassocephalum crepidioides) extract
	J Food Biochem, 40 (2016), pp. 109-119
	CrossRef View Record in Scopus Google Scholar
24	O.R. Molehin, S.A. Adefegha, G. Oboh, J.A. Saliu, M.L. Athayde, A.A. Boligon
	Comparative study on the phenolic content, antioxidant properties and HPLC
	fingerprinting of three varieties of Celosia species
	J Food Biochem, 38 (2014), pp. 575-583
	CrossRef View Record in Scopus Google Scholar
25	J.S. Johansen, A.K. Harris, D.J. Rychly, A. Ergul
	Oxidative stress and the use of antioxidants in diabetes
	Link Diabetol, 4 (2005), pp. 5-6
	CrossRef Google Scholar
26	G. Oboh, S.A. Adefegha, A.O. Ademosun, D. Unu
	Effects of hot water treatment on the phenolic phytochemicals and antioxidant activities
	of Lemon grass (Cymbopogon citratus)

Elect J Agric Food Chem, 9 (3) (2010), pp. 503-513

30/2020	In vitro antioxidant activities of African birch (Anogeissus leiocarpus) leaf and its effect on the α-amylase and α-glucosidase inhibitory pro
	View Record in Scopus
	📜 Outline  Download Share Export
27	G. Oboh, O.M. Agunlo
	Caffeic and chlorogenic acids inhibit key enzymes linked to type-2 diabetes (in vitro): a
	comparative study
	J Basic Clin Physiol Pharmacol, 26 (2) (2015), pp. 165-170
	View Record in Scopus Google Scholar
	Peer review under responsibility of Taibah University.
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