Antidiabetic Effect of Aqueous Root Extract of Strophanthus hispidus in Fructose-induced Type 2 Diabetic Sprague Dawley Rats

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ABSTRACT

Background: Strophanthus hispidus DC. (Apocynaceae) (SHP) is used in African traditional medicine in the treatment of several diseases including diabetes.

Objective: The study sought to explore and scientifically validate the antidiabetic activity of Strophanthus hispidus in fructose-induced Type 2 diabetic Sprague Dawley rats.

Methods: S. hispidus (50, 100 and 200 mg/kg p.o.), glibenclamide (5 mg/kg p.o.; diabetic control) and distilled water (10 mL/kg p.o.; normal control) were administered once daily for 28 days to 12 weeks fed fructose rats with homeostatic model assessment for insulin resistance (HOMA-IR) ≥1.5 and fasting blood glucose (FBG) level ≥200 mg/dL. Measurement of FBG and body weight of the fructose-induced Type 2 diabetic rats was carried out at 7 days interval. On day 28, blood samples were collected for assessment of serum biochemical parameters, including albumin (ALB), total protein (TP), creatinine, alanine transaminase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP), triglycerides (TG), total cholesterol (TC), high density lipoprotein (HDL), low density lipoprotein (LDL), bilirubin and urea; serum insulin; and haematological (haemoglobin, Hb and glycated haemoglobin, HbA1c) parameters. The liver of rats was harvested for glycogen assay.

Results: S. hispidus-treated fructose-induced Type 2 diabetic rats produced significant (p<0.05) day-dependent reduction in FBG level and significant (p<0.05) increase in HDL and TP levels compared with diabetic control rats. In addition, significant (p<0.05) decrease in TG, LDL, TC, AST, ALT, ALP, bilirubin, creatinine and urea levels was produced compared with diabetic control rats. Furthermore, significant (p<0.05) increase in serum insulin and liver glycogen levels was also observed. S. hispidus-treated diabetic rats displayed significant (p<0.05) increase in Hb and decrease in HbA1c levels.

Conclusion: The findings in this study showed that S. hispidus possesses beneficial antidiabetic property in Type 2 diabetes. This validates its use in African traditional medicine in the treatment of diabetes.

Keywords: Strophanthus hispidus, antidiabetic, biochemical, haematological, fructose.

INTRODUCTION

Diabetes mellitus (DM) Type 2 is a long-term metabolic syndrome characterised by high blood glucose level, corresponding lack of insulin and insulin resistance (1). Recurrent signs and symptoms include polydipsia, persistent urination, and inexplicable weight loss (2). Symptoms may also comprise polyphagia, tiredness, and impaired wound healing (2). Complications that occur over long period of time as a result of uncontrolled elevated blood glucose level include microvascular (retinopathy, nephropathy and neuropathy) and macrovascular (ischaemic heart disease, stroke and peripheral vascular disease) damage (3). Diabetes mellitus Type 2 comprises approximately 90% of cases of diabetes, with the other 10% mainly due to diabetes mellitus Type 1 and gestational diabetes (2).

Due to various side effects, such as stomach upset, skin rash or itching, weight gain, tiredness or dizziness, metal taste, bloating, diarrhoea, liver disease and anaemia risk, swelling of legs or ankles, associated with the available oral hypoglycaemic agents (4), there is the need to develop new antidiabetic agents in which medicinal plants have been shown to be effective in this regard with minimal side effects (4).

Strophanthus hispidus DC. (Apocynaceae) is used in African traditional medicine in the treatment of several ailments such as skin diseases, diabetes, leprosy, ulcers, malaria, dysentery and gonorrhoea (5, 6). Its potent anti-inflammatory property has been reported (7). This study, for the first time, was designed to investigate the antidiabetic activity of the aqueous root extract of S. hispidus in fructose-induced Type 2 diabetic rats.
MATERIALS AND METHODS

Plant Material and Extraction

Fresh root and leaves of *S. hispidus* were collected from Ile-Oluji village, Oke-Igbo Local Government Area, Ondo State, Southwest, Nigeria. The roots were used in this study but the leaves were collected for identification and authentication carried out by Prof JD Olowokudejo of the Department of Botany, University of Lagos, Lagos, Nigeria, where a voucher specimen (LUH 2618) was deposited. The roots were cut into smaller pieces and air-dried in the laboratory at room temperature for 7 days, pulverized using Christy and Norris 8' Lab Milling Machine (serial No. 50158, Knightdale Road, Ipswich, Suffolk, United Kingdom). The pulverized samples (250 g) were macerated with 1.5 L of distilled water and refrigerated for 72 h. The resulting extract was sieved with muslin cloth and then filtered using Whatman No. 1 paper (150 mm). The filtrate was evaporated to dryness in an oven (Gallenhamp®, Leicestershire, UK) at 40°C.

Chemicals and Reagents

Glibenclamide (Diatab®, May and Baker Nigeria Plc., Ikeja, Lagos, Nigeria), D-Fructose (Surechem Ltd., Suffolk, England), glucose D (Havit Remedies Pvt Ltd., Chhatral, India), insulin (Wockhardt Ltd., Mumbai, India), and Drabkin reagent (Sigma-Aldrich, Schnelldorf, Germany).

Experimental Animals

Male and female Sprague Dawley rats (90–120 g) were obtained from the Laboratory Animal Centre of the College of Medicine, University of Lagos, Lagos, Nigeria. The rats were fed with standard rodent diet (Livestock Feeds Plc., Ikeja, Lagos, Nigeria) and allowed access to water *ad libitum*. Before the commencement of experiments, the rats were acclimatized for two weeks under standard environmental conditions of temperature with 12 h dark/light cycle. The rats were fasted overnight (12 h) before the experiment. The experimental protocols were approved by the Health Research Ethics Committee of the College of Medicine of the University of Lagos, Lagos, Nigeria (CMUL/HREC/11/17/283).

Oral Glucose Tolerance Test OGGT

Sprague Dawley rats fasted overnight were randomly selected into 6 groups of 8 rats/group. After the determination of fasting blood glucose (FBG) levels, administration of distilled water (10 mL/kg p.o.), insulin (4 I.U./kg s.c.), glibenclamide (5 mg/kg p.o.) and *S. hispidus* at 50, 100 and 200 mg/kg to different groups of animals were carried out. The blood glucose levels of the rats were determined at 30 and 60 min. post-treatments. The doses of SHP were chosen based on the previous study of the rats were determined at 30 and 60 min. post-treatments.

Determination of Fasting Blood Glucose Level

Fasting blood glucose (FBG) was measured by placing 2 drops of venous blood drawn from the tail of each rat on glucose strip inserted into the Glucometer (Accu-Chek®) and reading values on the digital display.

Induction of Fructose-Induced Type 2 Diabetes Mellitus

The rats were given freshly prepared 20% fructose solution as drinking water daily for 12 weeks. The animals had daily access to the 20% fructose solution as the only source of drinking water for 12 weeks. After 12 weeks, 0.5 mL blood samples were collected from the rats through retro-orbital sinus for blood glucose and serum insulin levels determination without causing injury to the rats for homeostatic model assessment for insulin resistance (HOMA-IR). The rats with HOMA-IR value ≥1.5 were considered insulin resistant (9). Similarly, estimation of FBG level was done after 12 weeks and animals with blood glucose level ≥200 mg/dL were considered to be diabetic and were used in this study (10–13).

\[
\text{HOMA-IR} = \frac{\text{Glucose} \times \text{Insulin}}{22.5}
\]

Glucose level in mmol/L, Insulin level in mg/dL.

Treatment Protocols

Diabetic rats (n=40) were allocated into 5 groups of 8 rats each including diabetic control (0.9% normal saline; 10 mL/kg), glibenclamide (5 mg/kg) and SHP (50, 100, and 200 mg/kg). Additional non-diabetic group of 8 normal rats were given distilled water (normal control; 10 mL/kg). All treatments were given orally once daily for four weeks. Fasting blood glucose level and body weight of each rat was measured on days 7, 14, 21 and 28. On day 28, blood was collected from each rat through the retro-orbital sinus into lithium heparinized bottle for biochemical assay and ethylenediaminetetraacetic acid (EDTA) bottle for haematological assay. The animals were subsequently sacrificed by cervical dislocation and the livers were harvested for liver glycogen assay (14).

Assays

Biochemical Parameters and Lipid Profile Determination

Serum samples were investigated for aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), triglycerides (TG), total cholesterol (TC), low density lipoprotein (LDL), high density lipoprotein (HDL), albumin (ALB), total protein (TP), creatinine, bilirubin, and urea using Roche and Cobas 8000 modular analyzer series (Roche, Indianapolis, USA).

Serum Insulin Estimation

The quantity of serum insulin was estimated by an enzyme-linked immunosorbent assay (ELISA) protocol using rat insulin ELISA kit (Merckodia Ultrasensitive Rat Insulin ELISA, Winston Salem, North Carolina, USA) (14). Hepatic Glycogen Estimation

The level of hepatic glycogen was estimated according to the method previously described (14). Frozen hepatic tissue (100 mg) was placed in a cold citrate buffer (0.1 M with pH 4.5) and was extracted with perchloric acid (HClO₄, 6% w/v 15 mL). The resulting mixture was centrifuged (2000 rpm for 15 min.) and the supernatant (0.5 mL) was neutralized with KOH (10% w/v). At room temperature, the glycogen present in the supernatant was hydrolysed by α-amylglucosidase (50 U/mL;
Sigma-Aldrich, Schnellndorf, Germany) in sodium acetate buffer (50 mM/L; pH 4.8) overnight. Assessment of the glucose released was carried out using glucose assay kit (Sigma-Aldrich, Schnellndorf, Germany). The glycogen content of the liver samples was calculated as the difference between the glucose with and without α-amylglucosidase incubation and expressed as mg/g wet tissue.

**Haemoglobin (Hb) and Glycated Haemoglobin (HbA1c) Estimation**

Drabkin and Austin (15) and Nayak and Pattabiraman (16) methods were adopted for the estimation of haemoglobin (Hb) and glycated haemoglobin (HbA1c) levels respectively.

**Statistical Analysis**

The results obtained are expressed as mean ± standard error of mean (SEM) and the data were analysed using two-way ANOVA followed by Tukey’s multiple comparison test using GraphPad Prism 6 (Graph-Pad Software Inc., CA, USA). Results were considered significant when p<0.05.

**RESULTS**

**Oral Glucose Tolerance Test (OGGT)**

The hypoglycaemic effects of SHP in the oral glucose tolerance test (OGTT) in normoglycaemic rats are presented in Table 1. The plasma glucose in the normal control group increased to its maximal level at the time point of 90 min. after glucose (2.0 g/kg) loading and then gradually declined to initial level at 340 min. The treatment with different doses of SHP (50 and 200 mg/kg) significantly (p<0.05) reduced blood glucose levels at 90 and 120 min. compared to normal control. Significant (p<0.05) reductions in blood glucose levels were observed at 90, 120, 240 and 360 min. with SHP 100 mg/kg, glibenclamide 5 mg/kg and insulin 4 I.U. after glucose (2.0 g/kg) loading compared to normal control.

**Effect of S. hispidus on Glucose Level in Fructose-Induced Diabetic Rats**

Significant (p<0.05) increase in HOMA-IR (≥1.5) suggests that insulin resistance has been observed in this study (Figure 1). *S. hispidus* produced significant (p<0.05) dose-independent but day-dependent reduction of blood glucose level in diabetic rats compared with the diabetic control. The peak percentage reduction in blood glucose level was achieved on day 28 of treatment at all the treatment doses including glibenclamide (5 mg/kg). The peak percentage reduction (82.3%) of blood glucose level was achieved at SHP 100 mg/kg, suggesting greater effectiveness than glibenclamide (5 mg/kg) with percentage reduction of glucose level of 70.2% (Table 2).

**Effect of S. hispidus on Body Weight of Fructose-Induced Diabetic Rats**

There was significant (p<0.05) increase in the body weight of the rats after 12 weeks of fructose administration compared with normal control and initial body weight. After 14 days treatment with SHP (50, 100 and 200 mg/kg) and glibenclamide (5 mg/kg), there was reduction in body weight. After 28 days,

#### Table 1: Effect of *Strophanthus hispidus* in Oral Glucose Tolerance Test

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Dose (mg/kg)</th>
<th>Glucose level (mg/dL)</th>
<th>Basal</th>
<th>30 min.</th>
<th>60 min.</th>
<th>90 min.</th>
<th>120 min.</th>
<th>240 min.</th>
<th>360 min.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10 mL/kg</td>
<td>83.00 ± 2.4</td>
<td>93.30 ± 6.3</td>
<td>82.30 ± 3.3</td>
<td>162.30 ± 9.2</td>
<td>179.50 ± 4.4</td>
<td>97.30 ± 2.1</td>
<td>81.60 ± 6.5</td>
<td></td>
</tr>
<tr>
<td><em>S. hispidus</em></td>
<td>50</td>
<td>79.30 ± 5.2</td>
<td>90.10 ± 5.0</td>
<td>72.80 ± 3.7</td>
<td>123.30 ± 3.6</td>
<td>79.00 ± 12.5</td>
<td>87.00 ± 1.3</td>
<td>52.10 ± 6.8</td>
<td></td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>68.10 ± 2.7</td>
<td>63.80 ± 2.9</td>
<td>68.30 ± 3.2</td>
<td>120.00 ± 17.2</td>
<td>61.60 ± 10.9</td>
<td>53.50 ± 5.7</td>
<td>46.30 ± 2.3</td>
<td></td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>65.00 ± 6.7</td>
<td>69.30 ± 4.7</td>
<td>70.50 ± 2.7</td>
<td>118.60 ± 3.4</td>
<td>68.80 ± 3.0</td>
<td>76.10 ± 2.1</td>
<td>53.00 ± 4.3</td>
<td></td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>5</td>
<td>80.50 ± 5.0</td>
<td>61.60 ± 10.1</td>
<td>59.00 ± 7.38</td>
<td>120.80 ± 19.3</td>
<td>74.00 ± 6.3</td>
<td>63.30 ± 7.0</td>
<td>61.60 ± 8.1</td>
<td></td>
</tr>
<tr>
<td>Insulin</td>
<td>4 I.U.</td>
<td>74.00 ± 5.3</td>
<td>59.00 ± 13.7</td>
<td>32.10 ± 14.2</td>
<td>82.50 ± 9.5</td>
<td>67.30 ± 9.8</td>
<td>56.50 ± 7.8</td>
<td>48.30 ± 5.7</td>
<td></td>
</tr>
</tbody>
</table>

*Control received distilled water; results are presented as mean ± SEM. (n=5); *p<0.05 statistically significant compared to control (vehicle) (Two-way ANOVA followed by Tukey’s multiple comparison test).*

**Fig 1:** HOMA-IR values in Fructose-Induced Diabetic Rats.  
**Fig 2:** Serum Insulin Level in Fructose-Induced Diabetic Rats.
the weight of SHP and glibenclamide treated rats were significantly (p<0.05) decreased compared with the diabetic control rats; these were however significantly (p<0.05) higher than normal control group (Table 3).

**Effect of S. hispidus on Biochemical and Lipid Profile of Fructose-Induced Type 2 Diabetic Rats**

The levels of LDL, TC, and TG in fructose-induced Type 2 diabetic rats significantly (p<0.05) increased while HDL level decreased significantly (p<0.05) in diabetic control when compared with normal control (Table 4). S. hispidus (50, 100 and 200 mg/kg) and glibenclamide (5 mg/kg) treated groups had significant (p<0.05) reductions in LDL, TC and TG levels and significant (p<0.05) increase in HDL level compared to diabetic control groups. Significant (p<0.05) decrease in creatinine, urea, ALT, AST and ALP level was also observed in all treated groups compared with diabetic control. A reduction in bilirubin and TP level was recorded but not significant (p>0.05) compared with the diabetic control.

**Effect of S. hispidus on Serum Insulin Level of Fructose-Induced Diabetic Rats**

Significant (p<0.05) decrease in serum insulin levels in all the SHP (50, 100 and 200 mg/kg) and glibenclamide (5 mg/kg) treated groups was observed compared with diabetic control (Figure 2). This effect was prominent with 100 mg/kg dose of S. hispidus. The normal control group also displayed a significant decrease in serum insulin level compared with the diabetic control.

**Effect of S. hispidus on Hepatic Glycogen Content of Fructose-Induced Diabetic Rats**

Hepatic glycogen level was significantly (p<0.05) reduced in diabetic control compared with normal control. However, SHP (50, 100 and 200 mg/kg) and glibenclamide elicited significant (p<0.05) increase in hepatic glycogen level. This effect was not dose-dependent with SHP 100 and 200 mg/kg and glibenclamide 5 mg/kg producing the same hepatic glycogen level as shown in Figure 3.

**Effect of S. hispidus on Haemoglobin and Glycated Haemoglobin Levels of Fructose-Induced Diabetic Rats**

Haemoglobin (Hb) level was significantly increased (p<0.05) and glycated haemoglobin (HbA1c) level was significantly decreased (p<0.05) in SHP (50, 100 and 200 mg/kg) and glibenclamide (5 mg/kg) treated groups compared with diabetic control (Figure 4). Significant (p<0.05) decrease in Hb and increase in HbA1c was observed in the diabetic control group. This effect produced by SHP was not dose-dependent with 100 mg/kg dose eliciting the most prominent effect.

### Table 2: Effect of S. hispidus on Glucose Level of Fructose-Induced Type 2 Diabetes Rats

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Dose (mg/kg)</th>
<th>Basal</th>
<th>Day 0</th>
<th>Day 1</th>
<th>Day 7</th>
<th>Day 14</th>
<th>Day 21</th>
<th>Day 28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>10 mL/kg</td>
<td>74.2 ± 2.28</td>
<td>75.4 ± 1.07</td>
<td>70.2 ± 0.86 *</td>
<td>71.0 ± 1.58 *</td>
<td>72.6 ± 0.97 *</td>
<td>74.0 ± 1.00 *</td>
<td>72.0 ± 0.83 *</td>
</tr>
<tr>
<td>S. hispidus</td>
<td>50</td>
<td>78.0 ± 2.02</td>
<td>334.6 ± 16.9</td>
<td>316.4 ± 12.7</td>
<td>270.2 ± 13.1 * (5.4)</td>
<td>192.9 ± 5.39 * (21.6)</td>
<td>104.8 ± 0.91 * (62.5)</td>
<td>66.2 ± 1.59 * (76.3)</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>82.8 ± 2.81</td>
<td>279.8 ± 18.3</td>
<td>272.2 ± 17.4 * (2.7)</td>
<td>219.2 ± 5.39 * (21.6)</td>
<td>104.8 ± 0.91 * (62.5)</td>
<td>66.2 ± 1.59 * (76.3)</td>
<td>49.4 ± 2.9 * (82.3)</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>80.4 ± 3.69</td>
<td>283.2 ± 16.3</td>
<td>273.6 ± 15.3 * (3.3)</td>
<td>214.6 ± 3.18 * (24.2)</td>
<td>156.4 ± 7.40 * (44.7)</td>
<td>88.6 ± 3.68 * (68.7)</td>
<td>65.4 ± 2.56 * (76.9)</td>
</tr>
<tr>
<td>Glibenclamide</td>
<td>5</td>
<td>86.0 ± 3.40</td>
<td>273.6 ± 20.5</td>
<td>267.6 ± 19.8 * (2.1)</td>
<td>227.8 ± 11.84 * (16.7)</td>
<td>149.0 ± 8.05 * (45.5)</td>
<td>92.8 ± 2.03 * (66.0)</td>
<td>81.4 ± 2.97 * (70.2)</td>
</tr>
<tr>
<td>Diabetic control</td>
<td></td>
<td>81.0 ± 2.30</td>
<td>307.6 ± 15.69</td>
<td>319.6 ± 18.15</td>
<td>328.0 ± 7.38</td>
<td>325.6 ± 8.03</td>
<td>327.8 ± 10.8</td>
<td>342.4 ± 17.08</td>
</tr>
</tbody>
</table>

Diabetic control received normal saline (10 mL/kg); normal control received distilled water; results are presented as mean SEM (n=5).

The values in parenthesis indicate percentage reduction in blood glucose level.

### Table 3: Effect of S. hispidus on Body Weight of Fructose-Induced Type 2 Diabetic Rats

<table>
<thead>
<tr>
<th>Treatments/ Dose (mg/kg)</th>
<th>Initial</th>
<th>Basal</th>
<th>Day 1</th>
<th>Day 7</th>
<th>Day 14</th>
<th>Day 21</th>
<th>Day 28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>0.112 ± 0.004</td>
<td>0.112 ± 0.003</td>
<td>0.120 ± 0.004</td>
<td>0.120 ± 0.004</td>
<td>0.115 ± 0.004 *</td>
<td>0.112 ± 0.003 *</td>
<td>0.111 ± 0.002 *</td>
</tr>
<tr>
<td>S. hispidus 50</td>
<td>0.090 ± 0.001</td>
<td>0.155 ± 0.003 *</td>
<td>0.155 ± 0.003 *</td>
<td>0.163 ± 0.003 *</td>
<td>0.164 ± 0.002 *</td>
<td>0.154 ± 0.003 *</td>
<td>0.144 ± 0.003 *</td>
</tr>
<tr>
<td></td>
<td>0.095 ± 0.002</td>
<td>0.165 ± 0.001 *</td>
<td>0.165 ± 0.001 *</td>
<td>0.173 ± 0.001 *</td>
<td>0.166 ± 0.001 *</td>
<td>0.165 ± 0.001 *</td>
<td>0.156 ± 0.002 *</td>
</tr>
<tr>
<td></td>
<td>0.090 ± 0.001</td>
<td>0.157 ± 0.002 *</td>
<td>0.157 ± 0.002 *</td>
<td>0.166 ± 0.002 *</td>
<td>0.159 ± 0.003 *</td>
<td>0.140 ± 0.003 *</td>
<td>0.139 ± 0.003 *</td>
</tr>
<tr>
<td>Glibenclamide 5</td>
<td>0.011 ± 0.002</td>
<td>0.154 ± 0.003</td>
<td>0.154 ± 0.003</td>
<td>0.164 ± 0.002 *</td>
<td>0.151 ± 0.002 *</td>
<td>0.136 ± 0.002 *</td>
<td>0.122 ± 0.001 *</td>
</tr>
<tr>
<td></td>
<td>0.094 ± 0.001</td>
<td>0.160 ± 0.001 *</td>
<td>0.160 ± 0.001 *</td>
<td>0.167 ± 0.001 *</td>
<td>0.169 ± 0.001 *</td>
<td>0.177 ± 0.003 *</td>
<td>0.180 ± 0.004 *</td>
</tr>
</tbody>
</table>

Diabetic control received normal saline (10 mL/kg); normal control received distilled water; results are presented as mean ± SEM (n=5).

*p<0.05 statistically significant compared to normal control and initial body weight; *p<0.05 statistically significant compared to diabetic control (Two-way ANOVA followed by Tukey’s multiple comparison test).
Diabetic control received normal saline (10 mL/kg); normal control received distilled water (10 mL/kg); results are presented as mean ± SEM (n=5). *p<0.05 statistically significant compared to diabetic control (Two-way ANOVA followed by Tukey’s multiple comparison test). Gliben: Glibenclamide; CREA: creatinine; ALT: alanine amino transaminase; BIL: bilirubin; AST: aspartate aminotransferase; ALB: albumin; TP: total protein; TC: total cholesterol; TG: triglyceride; ALP: alkaline phosphatase; HDL: high density lipoprotein; LDL: low density lipoprotein.

**Table 4: Biochemical Parameters and Lipid Profile of Fructose-Induced Type 2 Diabetic Rats Treated with S. hispidus**

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Dose (mg/kg)</th>
<th>CREA (µmol/L)</th>
<th>UREA (µmol/L)</th>
<th>BIL (µmol/L)</th>
<th>ALT (IU/L)</th>
<th>AST (IU/L)</th>
<th>ALB (g/dL)</th>
<th>ALP (mg/dL)</th>
<th>TP (mg/dL)</th>
<th>TC (mmol/L)</th>
<th>TG (mmol/L)</th>
<th>HDL (mg/dL)</th>
<th>LDL (mg/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>2.55 ± 0.04a</td>
<td>21.24 ± 0.43a</td>
<td>0.56 ± 0.03a</td>
<td>2.68 ± 0.07a</td>
<td>17.10 ± 0.34a</td>
<td>4.01 ± 0.06a</td>
<td>31.18 ± 2.17a</td>
<td>6.89 ± 0.18a</td>
<td>42.16 ± 1.63a</td>
<td>16.58 ± 1.09a</td>
<td>86.13 ± 1.54a</td>
<td>17.39 ± 0.30a</td>
<td></td>
</tr>
<tr>
<td>S. hispidus 50</td>
<td>2.96 ± 0.04a</td>
<td>30.50 ± 2.47a</td>
<td>0.50 ± 0.04a</td>
<td>2.82 ± 0.03a</td>
<td>22.35 ± 0.37a</td>
<td>3.08 ± 0.08a</td>
<td>25.70 ± 2.81a</td>
<td>9.52 ± 0.17a</td>
<td>55.13 ± 2.01a</td>
<td>18.18 ± 0.23a</td>
<td>88.75 ± 2.59a</td>
<td>22.15 ± 0.39a</td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>2.08 ± 0.06a</td>
<td>15.34 ± 0.22a</td>
<td>0.29 ± 0.01a</td>
<td>2.64 ± 0.42a</td>
<td>18.87 ± 1.80a</td>
<td>1.96 ± 0.37a</td>
<td>13.55 ± 0.81a</td>
<td>7.04 ± 0.21a</td>
<td>26.24 ± 1.35a</td>
<td>15.49 ± 0.20a</td>
<td>98.48 ± 0.78a</td>
<td>14.51 ± 1.62a</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>2.08 ± 0.03a</td>
<td>16.65 ± 0.59a</td>
<td>0.32 ± 0.03a</td>
<td>3.31 ± 0.29a</td>
<td>25.28 ± 0.95a</td>
<td>2.22 ± 0.18a</td>
<td>17.01 ± 0.78a</td>
<td>6.91 ± 0.11a</td>
<td>35.46 ± 0.86a</td>
<td>18.44 ± 0.85a</td>
<td>81.28 ± 0.79a</td>
<td>13.74 ± 1.40a</td>
<td></td>
</tr>
<tr>
<td>Gliben</td>
<td>5</td>
<td>2.87 ± 0.11a</td>
<td>18.54 ± 1.88a</td>
<td>0.39 ± 0.01a</td>
<td>3.14 ± 0.18a</td>
<td>21.83 ± 0.65a</td>
<td>2.67 ± 0.18a</td>
<td>27.37 ± 1.98a</td>
<td>7.20 ± 0.24a</td>
<td>36.49 ± 1.32a</td>
<td>19.27 ± 0.20a</td>
<td>61.24 ± 2.28a</td>
<td>23.10 ± 0.65a</td>
</tr>
<tr>
<td>Diabetic control</td>
<td>21.48 ± 1.86</td>
<td>94.40 ± 2.64</td>
<td>0.89 ± 0.20a</td>
<td>18.24 ± 0.55</td>
<td>88.70 ± 1.15</td>
<td>4.04 ± 0.15a</td>
<td>109.94 ± 6.69</td>
<td>10.35 ± 0.26</td>
<td>194.41 ± 9.24</td>
<td>111.17 ± 6.71</td>
<td>9.94 ± 0.42</td>
<td>162.53 ± 22.65</td>
<td></td>
</tr>
</tbody>
</table>

**DISCUSSION**

It is an established fact that fructose feeding causes insulin resistance in experimental animals (17). Low HOMA-IR values suggest high insulin sensitivity, whereas high HOMA-IR values indicate low insulin sensitivity (insulin resistance) (18). There is therefore no doubt that glucose intolerance observed in this study is a consequence of insulin resistance induced by excess fructose feeding in the rats (high HOMA-IR values; ≥1.5). Metabolic disorder appeared after 12 weeks of 20% fructose intake in drinking water as only source of water. This was characterised by a reduction in insulin sensitivity and weight gain, which may be due to an expansion of adipose cells.

Treatment of the fructose-induced diabetic rats with *S. hispidus* significantly reduced blood glucose level. This activity might be attributed to the insulin sensitizing effects of *S. hispidus*. The blood glucose lowering effect, which indicates insulin sensitivity, was more prominent with the extract at the dose of 100 mg/kg which was more effective than glibenclamide. In the oral glucose tolerance test, *S. hispidus* suppressed the postprandial rise in blood glucose in normal rats following a heavy (2 g) glucose meal with maximum suppressive effect corresponding with the time of peak blood glucose level after meal. Oral glucose tolerance test (OGT) is broadly used to assess insulin release and insulin resistance in various clinical settings and to determine the potential of the body to use glucose which is the body’s key source of energy (19). The time-course of activity also showed that *S. hispidus* (50, 100 and 200 mg/kg), insulin (4 I.U.) and glibenclamide (5 mg/kg) reduced glucose level below basal in 2 h. The blood glucose lowering effect demonstrated by *S. hispidus* (100 mg/kg) was greater than that of glibenclamide (5 mg/kg) but comparable with the effect of insulin (4 I.U.).

Diabetes dyslipidaemia has long been revealed to have a resilient relation with coronary heart disease, which is the most treacherous and life threatening complication of diabetes (20, 21). The present study revealed significant increase in TC, LDL, TG and significant reduction in HDL in diabetic control group. The alterations in lipid and lipoprotein profiles of the diabetic rats as a result of insulin resistance were significantly reversed after 28 days of *S. hispidus* treatment, resulting in significant decrease in TC, LDL, TG and significant increase in HDL levels. The significant improvement of the serum lipid levels in *S. hispidus* treated diabetic rats might have been as a result of insulin sensitizing property of *S. hispidus*. In addition, the antiadipogenic and insulin sensitizing effects of *S. hispidus* demonstrated in this study might be credited, at least in part, to its anti-inflammatory properties (7), because study has shown that there is an increase in serum TNF-α level, pro-inflammatory
cytokine in fructose-induced diabetic rats (22). TNF-α has also been reported to impair the stimulatory effect of insulin on peripheral glucose uptake and its suppressive action on hepatic glucose production (23). Similarly, TNF-α circulate free fatty acids and thus contributes to the pathogenesis of insulin resistance (24). Serum creatinine, ALT, AST, bilirubin, urea and ALP levels were significantly reduced in S. hispidus and glibenclamide treated groups compared with the diabetic control group. This indicates potential hepatoprotective effect of S. hispidus and also protection against nephropathy in diabetes.

It has been reported that excessive fructose intake promote obesity and it’s cardiovascular and metabolic complications (25). Significant excessive increase in body weight of the rats was observed after 12 weeks of fructose intake compared with the normal control. After 14 days treatment with S. hispidus, the body weight reduced significantly compared with the diabetic control. This suggests adequate control of serum glucose level as a result of increase insulin sensitivity, glucose uptake and utilization by the body. Insulin sensitizing effects of S. hispidus results in utilization of serum insulin which might account for significant increase in hepatic glycogen level as demonstrated by S. hispidus in this study.

Glycated haemoglobin (HbA1c) significantly decreased in S. hispidus treated groups with significant increase in haemoglobin (Hb) level, in contrast with significant increase in HbA1c and decrease in Hb levels observed with diabetic control rats. It has been shown that protein glycation during hyperglycaemia customarily results to production of HbA1c. Therefore, HbA1c level is used as the most dependable measure for evaluation of glycaemic control in the management of diabetes (26). The decrease HbA1c levels in S. hispidus treated diabetic rats indicate less protein glycation, perhaps resulting from the reduction in blood glucose levels observed in these animals. The beneficial antidiabetic effect demonstrated by S. hispidus compared effectively with glibenclamide used as standard drug in this study.

CONCLUSION
Conclusively, S. hispidus demonstrated beneficial antidiabetic activity in fructose-induced Type 2 diabetic rats. This supports the claim in African traditional medicine of the usefulness of the plant extract in the treatment of diabetic mellitus.

CONFLICT OF INTEREST
The authors declare no conflict of interest.

REFERENCES


