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Effects of Aqueous Extract of *Blighia sapida* Leaves on Alloxan-induced Diabetic Rats

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ABSTRACT: Aqueous extract of *Blighia sapida* leaves was investigated for anti-diabetic activity in alloxan-induced diabetic albino rats. Thirty six albino rats (186.50 ± 7.58 g) were completely randomized into six groups (A-F) such that animals in group A (non-diabetic rats) received orally 0.5 ml of distilled water while the diabetic animals (made diabetic by intraperitoneal administration of single dose of 150 mg/kg body weight) in groups B, C, D, E and F were treated with 0.5 ml of distilled water, 2.5 mg/kg body weight of glibenclamide, 25, 50 and 100 mg/kg body weight of the extract respectively. The levels of blood glucose, serum total cholesterol, triacylglycerol, albumin, creatinine, urea, malondialdehyde, and glycogen in the liver of the animals as well as haemoglobin (Hb) and packed cell volume (PCV) were evaluated. The results revealed that the levels of blood glucose, serum cholesterol, albumin, creatinine, urea, triacylglyceride and malondialdehyde increased significantly ($P<0.0.05$) in the diabetic animals whereas the liver glycogen, Hb, PCV, liver-, kidney- and pancreas-body weight ratios decreased significantly ($P<0.05$). These alterations were not significantly different in the diabetic rats administered 25, 50 and 100 mg/kg body weight of the extract whereas these parameters in the diabetic animals treated with glibenclamide compared well with the non-diabetic distilled water administered animal. Contrary to ethno-medicinal claim, this study has revealed that the aqueous extract of *Blighia sapida* leaves at the doses of 25, 50, 100 mg/kg body weight did not have anti-hyperglycemic activity and is not suitable for managing complications associated with diabetes.

KEYWORDS: Diabetes, *Blighia sapida*, Alloxan monohydrate, Glibenclamide, Diabetes complications

1.0 Introduction

Diabetes mellitus is a clinical syndrome characterized by inappropriate hyperglycemia caused by a relative or absolute deficiency of insulin or resistance to the action of the hormone at the cellular level (Wadkar *et al.*, 2008). The incidence of diabetes as at the year 2010 was estimated to be 285 million people and this is expected to double by the year 2030 (Wild *et al*., 2004). Although there is a paucity of data on the prevalence of diabetes in Nigeria and other African countries, available information suggest that diabetes is emerging as a major health problem in Africa, including Nigeria (Mbanya *et al*., 1996). Insulin, a pancreatic hormone, is essential in the regulation of carbohydrate, lipid and protein metabolism (WHO, 1999). Underproduction of, or insensitivity of cells to insulin or a combination of both represents the core aetiology of diabetes mellitus. The complications of diabetes are linked to oxidative stress induced by hyperglycemia which overcomes the body's natural anti-oxidant system (Kikkawa *et al*., 2003; Udoh *et al*., 2007). In the later stages of diabetes, lipid metabolism is affected and expressed as hyperlipidemia and hypercholesterolemia which are also risk factors in atherosclerosis (Schwartz, 2006). There is also the possibility of liver damage in diabetes due to increased gluconeogenesis and ketogenesis (Felig *et al*., 1970). Management of diabetes mellitus is based on mechanisms which increase insulin secretion and orthodox drugs employed include the secretagogues and sensitizers which sensitize cells to insulin as well as inhibit intestinal glucose absorption and gastric emptying respectively. These management options, although quite effective, may not only have undesirable side effects, but can also constitute

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an economic burden on the user (Grahame-Smith and Aronson, 2002).

In Nigeria, several plant species have been claimed to possess anti-diabetic properties with a few scientific studies to substantiate this claim. For example, methanolic, ethylacetate extracts of *Cnestis ferruginea* leaves, aqueous extract of *Cochlospermum planchonii* leaves and *Unteria umbellate* seed fractions have been evaluated for antidiabetic activities in streptozotocin- and alloxan-induced hyperglycaemic rats (Adisa *et al*., 2010; Yakubu *et al*., 2011; Adeneye *et al*., 2012). Notwithstanding these and many other pharmacological studies, there is still need for the continuous evaluation of these botanicals for anti-hyperglycemic activity.

Blighia Sapida (Sapindaceae) also known as the Akee apple or Akee (English), ''Isin'' (Yoruba; Western Nigeria), ''Gwanja kusa'' (Hausa; Northern Nigeria) and ''Okpu'' (Ibo; Eastern Nigeria) is native to tropical West Africa Gabon, Benin, Burkina Faso, Cote d'Ivore, Ghana, Guinea, Guinea Bissau, Mali, Nigeria, Senegal, Sierra Leone and Togo as well as Cameroon, Sao Tome and Principe, (Koenig, 2002). It is an evergreen tree that grows to about 10 metres high, with a short trunk and a dense crown. The pinnate, leathery, compound leaves are 15-30 centimetres long. The flowers are greenish-white and bloom during the warm months (Robert, 1998; Kristen, 2003) whereas the fruit is pear shaped. Various parts of *Blighia sapida* have been reported to be used as eye drop in ophthalmic and conjunctivitis (pulp and leaves), terminate unwanted pregnancy when the root is used in conjunction with *Xylopia aethiopica*, expel parasites (seed), stomachic (leaves and bark), food and soap (Irvine, 1965; Morton, 1987; Abolaji *et al*., 2007). It has also been claimed to manage dysentery, epilepsy, haemorrhage, dental decay, constipation, whitlow, yellow fever and diabetes (Kean and Hare, 1980; Gbolade, 2009).

The phytochemical constituent of aqueous extract of *B. sapida* root bark have been reported to contain alkaloids, saponins, cardiac glycosides, reducing sugar, carbohydrates, flavonoids, phenol and tannins (Saidu *et al*., 2012). The anti-diarrhoeal activity of its leaves, nutritional potential of the sun-dried *B. sapida* aril, physico-chemical properties and toxicity of the oil from *B. sapida* seeds have also been reported (Antwi *et al.*, 2009; Oladiji *et al*., 2009; Quattara *et al.*, 2010). Furthermore, the hypoglycaemic effect of aqueous extract of *B. sapida* root bark on normoglycemic albino rats have also been reported (Saidu *et al*., 2012). Hypoglycin A, an amino acid isolated from the fruit of *B. sapida* have been reported to be responsible for the antidiabetic activity in rats (Atolani *et al*., 2009). Furthermore, the aqueous extract of *B. sapida* aril have also been investigated for blood glucose lowering effect in mice with diabetes (Rodriguez *et al*., 2012). The inhibitory effects of *B. sapida* leaf extract at the concentrations of 1.25-10.00 mg/ml on the key enzymes (α-amylase and α-glucosidase) linked to diabetes have also been reported (Kazeem *et al*., 2013). Despite all the myriads of pharmacological studies on the antidiabetic activities of the various parts of *B. sapida* in laboratory animals, information appears not to exist on the effect of aqueous extract of *B. sapida* leaves in alloxan induced diabetic rats.

Therefore, the present study was undertaken to evaluate the anti-hyperglycemic activity of the aqueous extract of *B. sapida* leaves and its effects on biochemical markers of complications associated with diabetes in alloxan-induced diabetic rats.

2.0 Materials and Methods

2.1 Plant material

The plants were collected in October, 2010, within the premises of College of Education, Ilorin, Nigeria, and were identified at the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria.

2.2 Chemicals and assay kits

Alloxan monohydrate was a product of May and Baker Ltd., Dagenham, England, while Glibenclamide was from HOVID Bhd., Ipoh, Malaysia. The assay kits for cholesterol, triaclyglyceride and albumin were products of Randox Laboratories, Co-Atrim, UK, while those for urea and creatinine were from Quinica Clinical Aplicada, S.A Amosta, Spain. All other reagents used were of analytical grade and were obtained from British Drug Houses Laboratory Supplies, Poole, UK.

2.3 Laboratory animals

Male and female albino rats of Wistar strain, weighing 186.50 ± 7.58 g were obtained from the animal Holding Unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. The animals were housed in clean plastic cages, placed in well ventilated house conditions (temperature 23 ± 2°C, humidity 40-45%, photoperiod: 12 hours natural light and 12 hours dark). The animals were allowed free access to rat pellets (Bendel Feeds, and Flour Mills, Ewu, Edo State Nigeria) and tap water.

2.4 Preparation of extract

The procedure described by Yakubu (2012) was adopted for the preparation of the plant extract. Briefly, the leaves were rinsed under running tap water, dried at room temperature for 2 weeks and then pulverized with electric blender (Tamashi Model XHGL-301, China). A known weight (200 g) of the powder which was extracted in 1L of distilled water for 48 hours was later filtered (Whatman No. 1 filter paper). The filtrate was then concentrated on a steam bath to give brownish black slurry which was reconstituted in distilled water to give the desired doses of 25, 50 and 100 mg/kg body weight used in the present study.

2.5 Induction of diabetes

The rats were acclimatized for 14 days before the commencement of the experiment. The animals were deprived of food but given free access to water for 6 hours and their fasting blood glucose level determined by drawing blood samples (0.6 µL) from the sharply cut tail vein, placing it on the test strip that had been inserted into the Bayer Contour ™ TS blood glucose meter (Bayer Consumer Care AG, Postfach, Basel, Switzerland), before the induction of diabetes. A known volume (1 ml) of alloxan monohydrate corresponding to 150 mg/kg body weight was administered intraperitoneally to each of the 30 animals to induce hyperglycemia. An hour after the administration of alloxan, pelleted feeds was restored back to the animals in addition to giving 5% dextrose saline through a feeding bottle to overcome the early hypoglycaemic phase (Mundargi *et al*., 2011). Only animals with blood glucose level of 11.0 mmol/L and above were used for the study.

2.6 Animal treatment

A total of 36 rats of both sexes, housed in clean plastic cages were completely randomized into six groups (A-F) of six animals each. Rats in group A (non-diabetic) received 0.5 ml of distilled water while those in groups B, C, D, E and F which were made diabetic were in addition administered 0.5 ml of distilled water, same volume corresponding to 2.5 mg/kg body weight of glibenclamide, 25, 50 and 100 mg/kg body weight of the extract respectively. The administration was done orally once daily for 12 days using oropharyngeal cannula. Fasting blood glucose levels of the animals were determined on days 1, 2, 4, 6, 8, 10 and 12.

2.7 Preparation of serum and tissue homogenates

The method described by Yakubu *et al* (2013) was used for the preparation of serum and tissue homogenates.

2.8 Determination of biochemical parameters

The biochemical parameters were as determined for glycogen (Kemp *et al*., 1953), total cholesterol (Fredrickson *et al*., 1967), triacylglycerol (Fossati and Prencipe 1982), urea (Veniamin and Varkirtzi, 1970), creatinine (Blass *et al.*, 1974), albumin (Doumas *et al.*, 1971), malondialdehyde (Buege and Aust, 1978) and organ-body weight ratio (Yakubu *et al*., 2008) while the Automated Hematologic Analyser (Sysmex, Kx-21, Japan) was used to analyse the Haemoglobin (Hb) and packed cell volume (PCV).

2.9 Statistical analysis

Data were expressed as mean ± SEM of six replicates and statistically analyzed by Duncan Multiple Range Test complemented with Student's t-test. The analyses were done with SPSS (version 16.0), SPSS Chicago, IL, USA and values for $p<0.05$ were considered statistically significant.

3.0 Results

Administration of alloxan to normal rats significantly ($P<0.05$) and consistently elevated the fasting blood glucose level of the animals throughout the exposure period when compared with the normal rats that received distilled water only (Table 1). By the end of the exposure period (day 12), the blood glucose level had increased by 281.55% (Table 1). This pattern of increase in the blood glucose levels was obtained in the diabetic animals administered the various doses of the aqueous extract of *B. sapida* leaves as the glucose levels were consistently elevated throughout the experimental period. By the end of this period, the blood glucose content of the diabetic animals treated with the 25, 50 and 100 mg/kg body weight of the extract had increased by 170.76%, 224.4% and 145.87% respectively. In contrast, the administration of glibenclamide to the diabetic rats significantly reduced the blood glucose level up to the 4^{th} day and by the remaining days of the experimental period, the blood glucose levels had compared favourably ($P>0.05$) with the non-diabetic distilled water treated control animals (Table 1).

The administration of alloxan to the experimental animals significantly reduced the levels of glycogen in the liver as well as PCV and Hb of the animals (Table 2). These trend of reductions were also extended to the diabetic animals treated with all the doses of the extract of *B. sapida* leaves. In contrast, the glibenclamide when administered to the diabetic rats restored the levels of liver glycogen, PCV and Hb to those of non-diabetic distilled water treated animals (Table 2). Furthermore, the alloxan significantly increased the levels of total cholesterol, triacylglycerol, creatinine, urea, albumin and malondialdehyde in the serum of the animals. The levels of these metabolites were similarly elevated ($P<0.05$) in the diabetic animals that were also treated with all the doses of the aqueous extract of *B. sapida* leaves in a manner that was not dose related. Interestingly, the levels of these metabolites in the diabetic animals treated with glibenclamide compared favourably with the non-diabetic distilled water treated animals (Table 2).

The computed liver-, kidney- and pancreas-body weight ratios decreased significantly in the diabetic rats that were administered all the doses of the extract (Table 3). The reductions were however not dose-related. In contrast, the organ body weight ratios of diabetic rats treated with the glibenclamide were not significantly different from the non-diabetic distilled water treated control animals (Table 3).

4.0 Discussion

Several scientific studies that have lend credence to antidiabetic activity of the stem, root and aril parts of *B. sapida* prompted this study whether the whole plant including the leaves of the plant also have diabetic activity as claimed in some non-scientific literature or not.

The cytotoxic action of alloxan is mediated by reactive oxygen species, with a simultaneous massive increase in cytosolic calcium concentration, leading to a rapid destruction of β-cells (Szkudelski 2001). This histological alteration in the β-cells of Islet of Langerhans will adversely affect the availability of insulin which is necessary for mopping up excess glucose in the animals. Therefore, the determination of glucose concentration in the blood of animals among others is a useful quantitative index of diabetes.

Effective control of blood glucose level is a key step in preventing diabetes mellitus (both Type 1 and 2) or which eventually will reduce or reverse complications associated with the disease. The elevated blood glucose levels in the diabetic rats that received distilled water confirmed the selective cytotoxic effect of alloxan on the β-cells of the pancreas and consequentially impaired the bioavailability of insulin to the animals. This further indicates that diabetes was induced in the animals. The inability of the aqueous extract of *B. sapida* leaves to reduce/restore the blood glucose levels of the diabetic animals suggest that the extract of the plant part was not pharmacologically active as an anti-diabetic agent as erroneously claimed in some non-scientific literature. It is possible that the extract of the leaves did not contain at all or in sufficient quantity bioactive agent(s)

Table 1: Effect of administration of aqueous extract of *Blighia sapida* leaves on blood glucose levels of alloxan-induced diabetic rats

	Days of administration						
	1	2	4	6	8	10	12
Distilled water only	5.65±0.38[a]	5.35±0.33[a]	5.33±0.94[a]	4.89±0.66[a]	5.04±0.30[a]	5.43±0.58[a]	5.63±0.28[a]
Diabetes + distilled water	5.80±0.60[a]	19.64±1.05[b]	19.43±1.03[b]	19.33±1.09[cd]	20.87±2.15[d]	21.64±2.28[d]	22.13±2.70[d]
Diabetes + glibenclamide	6.09±0.08[a]	19.90±0.38[b]	7.04±1.04[b]	6.02±0.42[a]	6.04±0.31[a]	6.10±0.46[a]	6.08±0.82[a]
Diabetes + 25 mg/kg body weight	5.78±0.39[a]	19.50±2.47[b]	18.28±3.06[b]	18.23±2.90[c]	16.96±2.34[c]	16.22±1.93[c]	15.65±2.09[b]
Diabetes + 50 mg/kg body weight	6.13±0.13[a]	23.93±1.86[c]	23.79±1.84[c]	21.33±1.85[d]	20.63±1.44[d]	20.26±1.52[d]	19.89±1.54[c]
Diabetes + 100 mg/kg body weight	6.06±0.37[a]	20.33±1.35[b]	18.88±1.42[b]	18.42±1.22[c]	16.96±1.13[c]	15.66±1.41[c]	14.90±0.91[b]

Values are mean ± standard deviation of six determinations
Test values on each day of administration across the rows carrying superscripts different from the control are significantly different ($P<0.05$)

Table 2: Effect of administration of aqueous extract of *Blighia sapida* leaves on some biochemical parameters of alloxan-induced diabetic rats

	Liver glycogen (mg of glucose/g of wet tissue)	Total cholesterol (mmol/L)	Triacylglycerol (mmol/L)	Creatinine (mmol/L)	Urea (mmol/L)	Albumin (g/dL)	Malondialdehyde (nmoles/ml)	PCV (%)	Hb (g/dL)
Control	5.73±1.70^{a}	2.51±0.04^{a}	1.21±0.02^{a}	1.08±0.04^{a}	3.19±0.01^{a}	3.45±0.60^{a}	0.5606±0.02^{a}	12.8±1.18^{d}	38.3±1.79^{d}
Diabetes only	1.84±0.43^{c}	2.84±0.02^{c}	2.41±0.01^{c}	3.08±0.05^{c}	5.21±0.01^{b}	6.29±0.34^{b}	0.5852±0.70^{b}	4.1±0.13^{a}	13.7±2.62^{a}
Diabetes+ glibenclamide	5.55±1.59^{a}	2.52±0.03^{a}	1.29±0.01^{a}	1.28±0.05^{a}	3.28±0.01^{a}	3.87±0.70^{a}	0.5618±0.50^{a}	12.3±0.33^{d}	37±3.85^{d}
Diabetes+25 mg/kg body weight	2.14±3.29^{b}	2.74±0.02^{bc}	2.31±0.02^{c}	3.00±0.04^{c}	5.18±0.02^{b}	6.32±0.37^{b}	0.592±0.11^{d}	6.6±0.31^{b}	20.0±2.08^{b}
Diabetes+50 mg/kg body weight	1.77±0.10^{c}	2.70±0.01^{b}	1.86±0.01^{b}	2.67±0.03^{b}	5.19±0.02^{b}	5.61±1.23^{b}	0.5852±0.15^{c}	7.5±0.17^{b}	22.7±1.03^{b}
Diabetes+100 mg/kg body weight	2.73±0.64^{b}	2.68±0.04^{b}	1.84±0.02^{b}	2.56±0.02^{b}	5.19±0.01^{b}	5.88±0.65^{b}	0.578±0.30^{b}	8.9±0.12^{c}	26.0±1.41^{c}

Values are mean ± standard deviation of six determinations
Test values down the column for each parameter carrying superscripts different from the control are significantly different ($P<0.05$)

Table 3: Effect of administration of aqueous extract of *Blighia sapida* leaves on some organ-weight ratio of alloxanized diabetic rats

Groups	Liver	Kidney	Pancreas
Control	5.29±1.57[b]	0.54±0.17[b]	0.70±0.21[b]
Diabetes + distilled water	3.15±0.45[a]	0.31±0.06[a]	0.39±0.25[a]
Diabetes+ glibenclamide	5.07±0.52[b]	0.51±0.15[b]	0.67±0.05[b]
Diabetes+25 mg/kg body weight	3.94±0.23[a]	0.33±0.08[a]	0.40±0.46[a]
Diabetes+50 mg/kg body weight	3.52±1.11[a]	0.26±0.044[a]	0.41±0.20[a]
Diabetes+100 mg/kg body weight	3.91±0.22[a]	0.36±0.098[a]	0.42±0.15[a]

Values are mean ± standard deviation of six determinations
Test values for each organ down the column carrying superscripts different from the control are significantly different ($P<0.05$)

of diabetes such as alkaloids and flavonoids or hypoglycin A or B earlier reported for other parts of the plant. This pro-hyperglycemic effect may be due to the absence of one or combination of inhibition of these: inhibition of α-glucosidase activities which leads to reduction in the uptake of glucose, interference with gastrointestinal glucose absorption (Musabayane *et al*., 2006), absence of insulin-like substances (Gay and Flat, 1999), and impaired secretion of insulin from the ß cells of the pancreas i.e. pancreatotropic action (Khan *et al.,* 1990; Trevedi *et al*., 2004; Yadav *et al*., 2008). Additional possibility is the inability of the animals to regenerate β-cells in the pancreas. The findings in these study contrast the previous reports on the antihyperglycemic activity of the fruit and root bark of *B. sapida* (Atolani *et al*., 2009; Saidu *et al* (2012). The glibenclamide, a reference antidiabetic agent, on the other hand might have lowered the blood glucose level by binding to the ATP-sensitive potassium channel and thus acted in a manner different from the extract.

Elevated levels of serum creatinine, uric acid, urea, albumin and total cholesterol as well as decrease in the levels of liver glycogen, packed cell volume and haemoglobin are common metabolic disorders of diabetes and have been widely reported (Resmi *et al*., 2001; Sokeng *et al*., 2005; Odetola *et al.,* 2006; Kumarappan *et al*., 2007; Ezeigbo and Asuzu, 2012; Yakubu *et al* 2013). The levels of these metabolites were altered in the same manner as previously reported for the diabetic animals treated with extract and diabetic animals administered distilled water. The trend of alterations in the various metabolites associated with diabetes is quite understandable since the aqueous extract of *B. sapida* leaves was not effective as an antidiabetic agent in the present study, the disease will consequentially progress and manifest its complications by altering the levels of these biomolecules as evidenced in the present study. The elevated level of lipid could be due to increased mobilization of free fatty acids from the peripheral fat depots (Bopanna *et al*., 1997) and uninhibitory actions of the lipolytic hormones on the fat depots (Goodman *et al.,* 1985; Oyedemi *et al.,* 2011) while the reduction in the level of glycogen in the liver of diabetic animals and those treated with the extract may be due impairment in the glycogen synthase system (Robert *et al.,* 2003). Furthermore, the elevated levels of urea may also be due to continuous catabolism of amino acids leading to a higher production of urea by the urea cycle (Yakubu *et al*., 2013). It has also been reported that MDA content increases during diabetes because of hypoinsulinaemia which increases the activity of fatty acyl coenzyme A oxidase that initiates β - oxidation of fatty acids, resulting in lipid peroxidation (Horie *et al.,* 1981). The increase in the serum MDA content of diabetic animals and those treated with the extract may not only be adduced to this reason but consistently emphasizes the ineffectiveness of the extract in controlling the blood glucose level in the animals and its attendant complications. The decrease in haemoglobin and PCV of diabetic and those diabetic animals treated with the extract which are consistent with previous studies may be due to the formation of glycosylated haemoglobin (Pari and Latha, 2002) resulting from secondary disease related complication. The decrease in Hb and PCV of the animals are indication of reduction in the number of viable haemoglobin required for adequate transportation of oxygen. Although, histopahtological examination was not carried out in the present study, the reduction in the computed liver-, kidney- and pancreas-body weight ratio may suggest atrophy of the cells. The finding in the present study agrees with that of Szkudelski (2001) on the mechanism of action of alloxan and streptozotocin on the cells of rat pancreas. Interestingly, all these alterations were attenuated by glibenclamide and compared favourably with the non-diabetic animals treated with distilled water suggesting that diabetes mellitus and its associated complications were reversed and further confirms the effectiveness of the reference drug.

Overall, data from this study revealed that the aqueous extract of *B. sapida* leaves at the doses of 25, 50 and 100 mg/kg body weight have not exhibited anti-diabetic activity nor attenuated complications associated with the disease as revealed by the altered biochemical parameters investigated in the present study which were not reversed to the control values. Therefore, the absence of one or more of the anti-diabetic principle(s) which were hitherto detected in the stem, root and aril may be responsible for this

ineffectiveness as an anti-diabetic agent. Therefore, the aqueous extract of the leaves, unlike the stem, root and aril is not pharmacologically active in managing diabetes.

References

Abolaji, O. A., Adebayo, A. H. and Odesanmi, O. S. (2007). Nutritional qualities of three medicinal plant parts *Xylopia aethiopica*, *Blighia sapida* and *Parinari polyandra* commonly used by pregnant women in the western part of Nigeria. Pakistan Journal of Nutrition 6: 665-668.

Adeneye A. A., Adeyemi O. O., Agbaje E.O. and Sofidiya M. O. (2012). The novel antihyperglycaemic action of *Hunteria umbellata* seed fractions mediated via intestinal glucose uptake inhibition. African Journal of Traditional Complementary and Alternative Medicine 9(1):17-24.

Adisa, R. A., Choudhary, M. I., Adewoye, E. O. and Olorunsogo, O. O. (2010). Hypoglycaemic and biochemical properties of *Cnestis ferruginea*. African Journal of Traditional Complementary and Alternative Medicine 7 (3):185 – 194.

Antwi, S., Martey, O. N. K., Donkor, K. and Okine, L. K. N. A. (2009). Anti-diarrhoeal activity of *Blighia sapida* (Sapindaceae) in rats and mice. Journal of Pharmacology and Toxicology 4: 117-125.

Atolani, O., Olatunji, G. A. and Fabiyi, O. A. (2009). *Blighia Sapida*; The plants and its Hypoglycinsan-Overview. Journal of Scientific Research 39(2):15-25

Blass, K. G., Thierbert, R. J. and Lam, L. K. (1974). A study of the mechanism of the Jaffe reaction. Journal of Clinical Chemistry and Clinical Biochemistry, 12, 336-343.

Bopanna, K. N., Kannan, J., Sushma, G., Balaraman, R. and Ratho, S. P. (1997). Antidiabetic and antihyperlipidaemic effects of neem seed kernel power on alloxan diabetic rabbits. Indian Journal of Pharmacology 29: 162-167.

Buege, J. A. and Aust, S. D. (1978). Microsomal lipid peroxidation. Methods in Enzymology 52: 302-310.

Doumas, B. T., Watson, W. A. and Biggs, H. G. (1971). Albumin standards and the measurement of serum albumin with bromcresol green. Clinical Chemistry Acta 31: 87-96.

Ezeigbo, I. I. and Asuzu, I. U. (2012). Anti-diabetic activities of the methanol leaf extracts of *Hymenocardia acida* (Tul.) in alloxan-induced diabetic rats. African Journal of Traditional Complementary and Alternative Medicine 9(2):204-209.

Felig, P. E., Ohman, J. L. and Chall, G. F. (1970). Plasma amino acid levels in diabetic ketoacidosis. Diabetes 19: 727-734.

Fossati, P. and Precipe, L. (1982). Serum triglycerides determined colourimetrically with an Enzyme that produces hydrogen peroxide. Clinical Chemistry 28(10): 2077-2080.

Fredrickson, D. S., Levy, R. I. and Lees, R. S. (1967). Fat transport in lipoproteins-an integrated approach to mechanisms and disorders. New England Journal of Medicine 276:215-225.

Gbolade, A. A. (2009). Inventory of antidiabetic plants in selected districts of Lagos State, Nigeria. Journal of Ethnopharmacology 121, 135-139.

Goodman, L. S. and Gilman, A. (1985). The pharmacological basis of therapeutics, 7th Edition. Mac Millan, New York. pp. 1490-510.

Gray, A. A. and Flat, P. R. (1999). Insulin releasing like activity of the traditional Antidiabetic plant *Coriander sativum* (coriander). Journal of Nutrition 81: 203-208.

Horie, S., Ishii, H. and Suga, T. (1981). Changes in peroxisomal fatty acid oxidation in diabetic rat liver. Journal of Biochemistry 90: 1691-1696.

Irvine, F. R. (1965). Botany and Medicine in West Africa. Ibadan University Press, Ibadan Nigeria. pp. 102-130.

Kazeem, M. I., Raimi, O. G., Balogun, R. M. and Ogundajo, A. L. (2013). Comparative Study on the α-Amylase and α-Glucosidase Inhibitory Potential of Different Extracts of *Blighia Sapida* Koenig. American Journal of Research Communication (in press).

Kean, E. A. and Hare, E. R. (1980). Gamma glutamyl trans-peptidase of the ackee plant *Blighia sapida*. Phytochemistry 19:199-204.

Kemp, A., Adrienne, J. M. and Heijningen, K. V. (1954) A colorimetric micro-method for the determination of glycogen in tissues. Biochemistry Journal 56(4): 646-648.

Kikkawa, R., Koya, D. and Haneda, M. (2003). Progress of diabetic neuropathy American Journal of Kidney Disfunction 41: 19-21.

Koenig, K. O. (2002). Germplasm resource information network. United States Department of Agriculturre. Retrieved 2011-10-18.

Kristen, L. (2003). Tropical flowering plants: A Guide to identification and cultivation. Timber Press.

Kumarappan, C. T., Rao, T. N and Mandal, S. C. (2007). Polyphenolic extract of *Ichnocarpus frutescens* modifies hyperlipidemia status in diabetic rats. Journal of Cell and Molecular Biology 6(2): 175–187.

Mbanya, J. C., Bonicci, F. and Nagan, K. (1996). Guidelines for the management of NIDDM in Africa. A consensus document, Greece, Novo Nordisk A/s. pp. 1-35.

Morton, J. F. (1987). Ackee: Fruits of warm climates. In: Morton, J. F. and Miami, F. L. (eds). pp. 269 - 271.

Musbayane, C. T., Bwititi, P. T. and Ojewole, J. A. O. (2006). Effect of oral administration of some herbal extracts on food consumption and blood glucose levels in normal and streptozotocin treated diabetic rats. Methods and Findings in Experimental and Clinical Pharmacology 28(4): 223-228.

Odetola, A. A, Akinloye, O., Egunjobi, C., Adekunle, W. A. and Ayoola, A. O. (2006). Possible antidiabetic and antihyperlipidemic effect of fermented *Parkia Biglobosa* (JACQ) extract in alloxan-induced diabetic rats. Clinical and Experimental Pharmacology and Physiology 33: 808-812.

Oladiji, A. T., Shoremekun, K. L. and Yakubu, M. T. (2009). Physicochemical properties of the oil from the fruit of *Blighia sapida* and toxicological evaluation of the oil-based diet in Wistar rats. Journal of Medicinal Food 12(5): 1127-1135.

Oyedemi, S. O., Yakubu, M. T. and Afolayan A. J. (2011). Antidiabetic activities of aqueous leaves extract of *Leonotis leonurus* in streptozotocin-induced diabetic rats. Journal of Medicinal Plants Research 5(1):119-125.

Pari, L. and Amarnath, S. (2004). Antidiabetic activity of *Boerhavia diffusa L*. effect on hepatic key enzymes in experimental diabetes. Journal of Ethnopharmacology 91: 109-113.

Pari, L. and Latha, M. (2002). Effect of *Cassia auriculata* flowers on blood sugar levels, serum and tissue lipids in streptozotocin diabetic rats. Singapore Medical Journal 43(12): 617-621.

Quattara, H., Niamké, B., Dally, T. and Kati-Coulibaly, S. (2010). Nutritional composition studies of sun dried *Blighia sapida* (K. koenig) aril from Côte d'Ivoire, Journal of Applied Bioscience 32: 1989-1994.

Robert, K. M., Daryl, K. G., Peter, A. M. and Victor, W. R. (2003). Harper's Illustrated Biochemistry, a LANGE Medical Book. 26th edition.

Robert, R. (1998). The Tropical Look. Timber Press.

Rodríguez De La P., Gloria C., Hleap, Z., José I., Zuluaga, G. and Claudia, L. (2012). Evaluación Del Extracto Del Arilo Del Ackee (*Blighia Sapida* K) Con Propiedades Hipoglucemiantes, En Biomodelos Biotecnología en el Sector Agropecuario y Agroindustrial, 10(2): 8.

Saidu, A. N., Mann, A. and Onuegbu, C. D. (2012). Phytochemical screening and hypoglycaemic effect of aqueous *Blighia sapida* root bark extract on normoglycemic albino rats. British Journal of Pharmaceutical Research 2(2): 89-97.

Schwartz, S. L. (2006). Diabetes and dyslipidemias. Diabetes Obesity Metabolism 8: 355-364.

Sokeng, D. S., Rokeya, B., Mostafa, M., Nahar, N., Mosihuzzaman, N., Ali, L. and Kamtchouing, P. (2005). Antihyperglycemic effect of *Bridellia ndellernsis* ethanol extract and fractions in streptozotocin-induced diabetic rats. African Journal of Traditional Complementary and Alternative Medicine 2:94-102.

Szkudelski, T. (2001). The mechanism of alloxan and streptozotocin action-cells of the rat pancreas. Physiology Research 50: 536-546

Trivedi, N. A., Mazumder, B., Bhatt, J. D. and Hemavathi, K. G. (2004). Effect of Shilajit on blood glucose and lipid profile in alloxan-induced diabetic rats. Indian Journal of Pharmacology 36: 373-376.

Udoh, A. E., Iya, N., Okon, E. and Mary, N. (2007). Red cell catalase activity in diabetics. Pakistan Journal of Nutrition 6: 511-515.

Veniamin, M. P. and Varkirtzi, C. (1970). Chemical basis of the carbamidodi-acetyl micro-method for estimation of urea, citruline and carbamyl derivatives Clinical Chemistry 16: 3-6

Wadkar, R. R., Supale, R. S., Tiwari, K. M., Patil, K. S. and Jalalpuri, S. S. (2008). Screening of roots of *Baliospermum montanum* for hepatoprotective activity against paracetamol induced liver damage in albino rats. International Journal of Green Pharmacy 220- 223.

WHO (1999). WHO news: Traditional medicine strategy launched. Bullettin of the World health Organization 80: 610-610.

Wild, S., Roglic, G., Green, A., Sicree, R. and King, H. (2004). Global prevalence of diabetes: Estimates for the year 2000 and projections for 2030. Diabetes Care 27: 1047-1053.

Yadav, J. P., Kalia, A. N. and Dangi, A. S. (2008). Hypoglycemic activity of the extract of *Salvadora oleoides* in normal and alloxan induced diabetes rats. Indian Journal of Pharmacology 40:23-27.

Yakubu, M. T. (2012). Effect of a 60-day oral gavage of a crude alkaloid extracts from *Chromolaena odoratum* leaves on hormonal and spermatogenic indices of male rats. Journal of Andrology 33(6): 1199-1207.

Yakubu, M. T., Akanji, M. A. and Oladiji, A. T. (2008). Effect of oral administration of aqueous extract of *Fadogia agrestis* stem on some testicular function indices of male rats. Journal of Ethnopharmacology 111(2): 288–292.

Yakubu, M. T., Awotunde, S. O., Ajiboye, T. O., Oladiji, A. T. and Akanji, M. A. (2011). Pro-sexual effects of aqueous extracts of *Massularia acuminata* root in male Wistar rats. Andrologia 43 (5): 334-340.

Yakubu, M. T., Salau, A. K., Oloyede, O. B. and Akanji, M. A. (2013). Anti-diabetic activity of aqueous leaf extract of *Ficus exasperata* in

alloxan-induced diabetic Wistar rats. Cameroon Journal of Experimental Biology (in press).