Anti-diabetic Activity of Aqueous Extract of *Curcuma longa* (Linn) Rhizome in Normal and Alloxan-Induced Diabetic Rats

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Abstract: The present study was carried out to investigate the anti-diabetic activity of aqueous extract of *Curcuma longa* rhizome in both normal and alloxan induced diabetic rats. Alloxan-induced diabetic and non-diabetic rats were administered orally with aqueous extract of *Curcuma longa* rhizome at 200 mg/kg for 28 days, after which the blood glucose, protein, albumin and lipid profile were determined and compared with the normal control. There was a significant ($p<0.05$) increase in the level of blood glucose, total cholesterol, Low Density Lipoprotein (LDL), triglyceride (TG) and a significant ($p<0.05$) decrease in the level of High Density Lipoprotein (HDL), total protein, albumin and body weight of the diabetic untreated rats. Oral administration of aqueous extract of *Curcuma longa* rhizome at a dose of 200 mg/kg body weight for 28 days to diabetic rats resulted in a reversal of the above diabetic conditions. Phytochemical screening of the aqueous, acetone and ethanolic extract of *Curcuma longa* rhizome revealed the presence of alkaloids, cardiac glycoside and resins while flavonoids, tannins, saponins, balsams and phenols were not detected in all solvent extracts. The data from this study suggest that the aqueous extract of *Curcuma longa* rhizome used in the study possesses anti-diabetic activities and could be used for the management of diabetes and associated metabolic alterations.

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1.0 Introduction

Diabetes mellitus (DM) is a group of syndromes characterized by hyperglycemia, altered metabolism of lipids, carbohydrates and proteins and an increased risk of complications from vascular diseases [1] which are attributed to an insufficient supply of insulin [2]. It could also occur when the insulin receptors are resistant to the functions of circulating insulin [3]. Currently, there are over 150 million diabetic patients worldwide and this likely tends to increase to 300 million or more by the year 2025 [4]. DM is characterized by increases in glucose levels build up in the blood and urine, causing excessive urination, thirst, hunger and problems with carbohydrate, fat and protein metabolism [5, 6, 7]. It has already been established that chronic hyperglycemia of diabetes is associated with long term damage, dysfunction and eventually the failure of organs, especially the eyes, kidneys, nerves, heart and blood vessels [8].

Management of diabetes without side effect is still a challenge to the medical system. Currently available synthetic antidiabetic agents produce serious side effects such as hypoglycemic coma and hepatorenal disturbances [9]. Moreover, they are not safe for use during pregnancy [10]. Hence, the search for safer and more effective anti-diabetic agents is still ongoing. At present, thousands of plant metabolites are being successfully used for the treatment of a variety of diseases. According to an estimate, 80% of the world's population relied upon plants for their medication. The use of the medicinal plants is increasing in many countries where 35% of drugs contain natural products and ethno botanical information indicates that plant species are used in the traditional management of diabetes [11, 12, 13, 14].

Curcuma longa (Linn) commonly known as turmeric; is an erect perennial herb [15] with pulpy, orange, tuberous roots that grows to about 2 feet in length [16] and belongs to the family *Zingiberaceae.* Its rhizome is pungent and bitter [15] and widely used in indigenous medicine and as household remedies [17]. It is also recommended for treating diabetes, high cholesterol, abdominal pains, menstrual disorder, wounds, eczema, jaundice, inflammations, cancerous symptoms and as a blood purifying activity [16]. The herb contains curcumin as the active ingredient, which is a yellow coloured phenolic pigment obtained from the powdered rhizome of *C. longa* Linn. In the crude extracts of *C. longa* about 70 to 76 % curcumin is present, along with about 16% demethoxycurcumin and 8% bisdemethoxycurcumin[18]. Studies have also demonstrated a wide spectrum of therapeutic effects,

such as anti-inflammatory [19, 20, 21], antioxidant [22], antitumor [23, 24], antibacterial [25], antifungal [26], antiviral [27], anti-spasmodic [28], immunomodulation [29] and hepatoprotective [30] activities. The present study was designed to investigate anti-diabetic activity of aqueous extract of *Curcuma longa* rhizome in normal and alloxan induced diabetic rats.

2.0 Materials and Methods

2.1 Plant Material

The rhizomes of *Curcuma longa* used for this study were obtained from Federal College of Forestry, Jos, Nigeria and were authenticated at the Department of Plant Science and Technology, University of Jos, Jos, Nigeria, where a voucher specimen was deposited at the Herbarium of the Institute.

2.2 Chemicals

Alloxan monohydrate was obtained from Sigma-Aldrich Chemical Company, St Louis, U.S.A. All the other chemicals used were of analytical grade and prepared in glass distilled water.

2.3 Experimental Animals

Adult Wistar (male and female) albino rats (20) weighing between 180-250g were obtained from the National Veterinary Research Institute, Vom, Jos, Nigeria. The animals were housed in aluminum cages under standard conditions. They were maintained on standard animal pellets (purchased from Grand Cereal and Oil Mills Limited Jos, Nigeria) and water *ad libitum*. The animals were acclimatized for two weeks before the commencement of the experiment.

2.4 Preparation of Plant Extract

The *Curcuma longa* rhizomes were oven dried at 40°C for 72 hours to a constant weight. The dried rhizomes were then pulverized using Beltone Luinohun Blender (model MS-223, Taipei, Taiwan). The powdered form was stored in airtight plastic container until required for use. 100g of the fine powder was percolated in one liter of distilled water for 48 hours at 37^0C (to ensure maximum extractions of phytochemicals). This was then filtered with Whatman No. 1 to remove all un-extractable matter. The same procedure was employed for acetone and ethanolic extracts except that the aqueous filtrate was concentrated on a steam bath and reconstituted in distilled water to give the required dose of 200 mg/kg body weight as used in this study. (A value arrived at from information obtained during ethnobotanical survey). The reconstituted aqueous extract was administered orally using cannula to normal and diabetic rats.

2.5 Phytochemical Screening

The aqueous, acetone and ethanolic extracts of *Curcuma longa* rhizome were screened for some phytochemical constituents using standard procedures [31, 32].

2.6 Induction of Diabetes

The animals were fasted overnight and diabetes was induced by a single intraperitoneal injection of a freshly prepared Alloxan monohydrate (150 mg/kg body weight) in ice cold 0.9% NaCl solution. The animals were allowed 5% glucose solution overnight *ad libitum* to overcome the drug-induced hypoglycemia. Control (normal) rats were not injected with alloxan and were placed on normal saline alone. After 24 hours, rats with blood glucose level >7.0 mmol/L were considered as diabetic and used for the experiment.

2.7 Experimental Design

After randomizing into various groups and before initiation of the experiment, the rats were acclimatized to the animal house conditions. The rats were maintained during the study on standard rat feed consisting of 70% carbohydrate, 14.50% protein, 7% fat, 7.20% fibre and 1.20% minerals. The animals were randomized into 4 groups of 5 animals each for the evaluation of anti-diabetic activity.

GROUP A: Normal control rats on 0.5 ml normal distilled water per day.

GROUP B: Diabetic control rats on 0.5 ml normal distilled water per day.

GROUP C: Diabetic treated rats on 0.5 ml of extract (equivalent to 200mg/kg aqueous *Curcuma longa* rhizome extract).

GROUP D: Normal treated rats on 0.5 ml of extract (equivalent to 200mg/kg aqueous *Curcuma longa* rhizome extract).

All administered were done orally per day using cannula for 28 days.

2.8 Body Weight Determination

Total body weight of diabetic and non-diabetic Wistar rats were determined using digital balance, before and after the experimental period and recorded as initial body weight (IBW) and final body weight (FBW) respectively.

2.9 Collection of Blood Sample

The methods described by Yakubu *et al.* [33] were used for the collection of blood samples and preparation of serum. In brief, with the animal under diethyl ether anaesthesia, the neck area was quickly shaved to expose the jugular veins. The veins after being slightly displaced (to avoid contamination with interstitial fluid) were then sharply cut with a sterile

scalpel blade. Blood was collected into EDTA sample bottles for haematological assay and also collected into clean, sterile sample bottles which were allowed to clot for 30 minutes. This was then centrifuged at 33.5 g for 15 minutes using a Uniscope Laboratory Centrifuge. The sera were aspirated with Pasteur pipettes and stored frozen until required for the biochemical analyses.

2.10 Assay of Biochemical Parameters

The total protein content of the serum was determined using the Biuret method [34]. Albumin (ALB) level was determined as described by Grant and Kacchman [35]. Serum glucose [36], total cholesterol [37], Low Density Lipoprotein (LDL) [38], High Density Lipoprotein (HDL) [39] and triglyceride (TG) [40] were also determined. All measurements were done using Spectronic 21 spectrophotometer (Bausch and Lomb, NY).

2.11 Statistical Analysis

Data are expressed as mean ± standard deviation (SD). Comparison of the data from test control groups of animals were analyzed by One Way Analysis of Variance (ANOVA) at the confidence limit of 95% and where applicable, Least Significant Difference (LSD) was used to determine significant results, differences between groups were considered statistically significant at $p<0.05$.

3.0 Results

Table 1 shows the phytochemicals present in the aqueous, acetone and ethanolic extract of *Curcuma longa* rhizome. The presence of alkaloids, cardiac glycoside and resins were detected in the three solvent. Carbohydrate was present only in the acetone extract of the rhizome while terpenes and steroid were present in the aqueous and ethanolic extract of the rhizome. Flavonoids, tannins, saponins, balsams and phenols were not detected in all solvent extracts.

Table 2 shows the effect of aqueous extract of *Curcuma longa* rhizome on body weight of both normal and alloxan induced diabetic rats. There was a significant ($p<0.05$) decrease in the body weight of rats in the diabetic control group when compared with the normal. The administration of the aqueous extract of *Curcuma longa* rhizome at the dose of 200 mg/kg body weight increase body weight of rats in the diabetic treated group.

Table 3 shows the effect of aqueous extract of *Curcuma longa* rhizome on blood glucose, protein and albumin of both normal and alloxan induced diabetic rats. There was a significant ($p<0.05$) increase in the blood glucose level of rats in the diabetic untreated group when compared with the normal control, normal treated and diabetic treated groups. Also, there was significant ($p<0.05$) decrease in the level of total protein and albumin of rats in the diabetic untreated group compared to normal rats at the 28th day. The administration of the aqueous extract of *Curcuma longa* rhizome at the dose of 200 mg/kg body weight significantly ($p<0.05$) decrease the blood glucose and significantly ($p<0.05$) increase the total protein and albumin of rats in the diabetic treated group.

Table 4 shows the effect of aqueous extract of *Curcuma longa* rhizome on High Density Lipoprotein (HDL), total cholesterol (TC), triglyceride (TG), and Low Density Lipoprotein (LDL) of normal rats and alloxan induced diabetic rats. There was a significant ($p<0.05$) increase in the level of TC, TG and LDL and a significant ($p<0.05$) decrease in the level of HDL of rats in the diabetic untreated group when compared with the control and other treated groups. The administration of the aqueous extract of *Curcuma longa* rhizome at the dose of 200 mg/kg body weight significantly ($p<0.05$) decrease the level of TC, TG and LDL and significantly ($p<0.05$) increase the level of HDL of diabetic treated group at the 28th day of the study.

Table 1: Phytochemical screening of aqueous, acetone and ethanolic extract of *Curcuma longa* rhizome

Phytochemicals	Aqueous	Acetone	Ethanol
Alkaloids	+	+	+
Flavonoids	–	–	–
Tannins	–	–	–
Saponin	–	–	–
Cardiac glycosides	+	+	+
Terpenes and steroids	+	–	+
Balsams	–	–	–
Phenols	–	–	–
Resins	+	+	+
Carbohydrates	–	+	–

Key= + present; - absent.

Table 2: Effect of aqueous extract of *Curcuma longa* rhizome on body weight of alloxan induced diabetic rats

	Weight Variation (g)		
Group	Initial (IBW)	Final (FBW)	
Control	248±3.80	279±2.80	
Diabetic control	250±4.00[ab]	190±4.98[a]	
Diabetic treated	255±3.80[ab]	277±2.78[ab]	
Normal treated	250±3.78[ab]	280±2.77[ab]	

Values are expressed as mean ± SD, n= 5 for each group
[a] values are significantly different from normal control ($p<0.05$)
[b] values are significantly different from the diabetic control group ($p<0.05$)

Table 3: Effect of aqueous extract of *Curcuma longa* rhizome on blood glucose, protein, and albumin of alloxan induced diabetic rats

Group	Glucose (mmol/L)	Protein (g/L)	Albumin (g/L)
Control	5.03±0.01	72.00±1.41	33.25±0.96
Diabetic control	18.45±0.01[a]	66.00±0.82[a]	27.25±0.96[a]
Diabetic treated	7.83±0.01[ab]	68.25±0.96[ab]	35.25±0.96[b]
Normal treated	4.94±0.01[ab]	66.25±0.96[ab]	27.25±0.96[a]

Values are expressed as mean ± SD, n= 5 for each group
[a] values are significantly different from normal control ($p<0.05$)
[b] values are significantly different from the diabetic control group ($p<0.05$)

Table 4: Effect of aqueous extract of *Curcuma longa* rhizome on serum lipid profile of alloxan induced diabetic rats

	Lipid profile (mmol/L)			
Group	TC	TG	LDL	HDL
Control	3.10±0.08	1.18±0.05	2.15±0.10	2.73±0.13
Diabetic control	5.68±0.06[a]	2.17±0.01[a]	3.13±0.09[a]	0.43±0.10[a]
Diabetic treated	4.69±0.02[ab]	1.71±0.02[ab]	2.03±0.09[b]	0.98±0.13[ab]
Normal treated	2.95±0.10[b]	0.85±0.06[ab]	1.90±0.08[ab]	2.93±0.10[b]

Values are expressed as mean ± SD, n= 5 for each group
[a] values are significantly different from normal control ($p<0.05$)
[b] values are significantly different from the diabetic control group ($p<0.05$)

4.0 Discussion

Prolonged exposure to hyperglycemia is now recognized as the primary causal factor in the pathogenesis of diabetic complications as well as induces a large number of alterations in vascular tissue that potentially promote or accelerated atherosclerosis [41]. In this study, there was a significant increase in the level of blood glucose of rats in the diabetic group. Oral administration of aqueous extract of *Curcuma longa* rhizome to the diabetic rats significantly reduced the blood glucose level compared with the control. Previous reports has indicated that plant extracts possess hypoglycemic properties, possible insulin release stimulatory effects and uptake of peripheral glucose, which in turn reversed alloxan induced hyperglycemia [42,43,44,45]. Mafulul *et al.* [46] reported that the possible mechanism by which aqueous extract from plants brings about their hypoglycemic action may be by induction of pancreatic insulin secretion from β cells of islets of Langerhans or due to enhanced transport of blood glucose to peripheral tissue. Reducing insulin resistance and inhibition of intestinal glucose absorption are also possible mechanisms as reported by Xi *et al.* [47] and Youn *et al.* [48] respectively. They may contain biomolecules that can modify or stimulate insulin receptors, modify the structure of glucose transport protein (GLUT 4) and may inhibit insulin antagonist within the body as reported by Igbakin and Oloyede [49].

In the present study, alloxan induced diabetes rats produced a significant decrease in body weight. The

loss in weight in the diabetic groups is attributed to the alloxan that was injected into the animals. Alloxan is known to partially destroy the beta-cells of the islets of the Langerhans of the pancreas that function in insulin regulation, producing type 1 diabetes. The destruction of the pancreas results in the utilization of non-carbohydrate moieties such as protein for the synthesis of glucose. The loss of structural proteins in increased gluconeogenesis together with increased lipolysis and increased synthesis of ketone bodies results in severe weight loss [50]. Furthermore, the weight loss observed in alloxan-diabetic rats can be due to a reduction of food intake [51]. Administration of aqueous extract of *Curcuma longa* rhizome was found to be effective in ameliorating the weight loss observed in the diabetic rats compared with the control.

Albumin is a major protein of human plasma and represents about 25% of total hepatic protein synthesis and half its secreted proteins. Its synthesis is depressed in a variety of diseases, particularly those of the liver [52]. Table 3 shows that there was a significant decrease in the concentration of albumin and total serum protein of the untreated alloxan induced diabetic rats when compared with the control and the treated groups. This observation may be attributed to numerous effects of hyperglycemia in alloxan-induced diabetes. Hyperglycemia increases gluconeogenesis and as such leads to excess protein breakdown as well as excess loss of nitrogen resulting in negative nitrogen balance [53]. A decline in the total protein level in diabetic rats has been attributed to inhibition of oxidative phosphorylation, which leads to decrease in protein synthesis, increase in catabolic processes and reduction in protein absorption [54]. Also, decrease in the total protein of alloxan induced rat, might be due to decrease in protein content of the urine, leading to microproteinurea which is an important clinical marker of diabetic nephropathy [55], it may also be due to increased protein catabolism [56] as a result of insulin deficiency from free radical generated due to alloxan induction, since it has been established that insulin stimulates the incorporation of amino acids into protein [56]. These results imply that administration of the extracts caused a remarkable increase in the serum total protein and albumin levels in the diabetic rats.

Diabetes affects both glucose and lipid metabolism [57]. In the postprandial state elevated serum insulin increases lipoprotein lipase activity in adipose tissue and promotes fuel storage as triglycerides in normal metabolism [58]. The deficiency of insulin depletes the activity level of lipoprotein lipase, thus leading to deranged lipoprotein metabolism during diabetes [59]. Alloxan induced diabetic untreated rats showed significantly increased serum lipid profiles except HDL compared with the control rats. The elevated TG, TC, LDL level and decreased HDL level in alloxan-induced diabetic rats observed in this study is in agreement with the previous reports regarding alteration of these parameters under diabetic condition [43, 60]. This may be due to the increase in the mobilization of free fatty acids (FFA) from the peripheral depots, since insulin inhibits the hormone sensitive lipase [61]. Serum FFA concentration is a result of the balance between the release from lipolysis, neosythesis and disposal and represent the major determinant of insulin effect on FFA oxidation and non-oxidative metabolism [62]. Oral administration of the aqueous rhizome extract of *Curcuma longa* to the diabetic rats significantly reduced the level of TG, TC, and LDL and significantly increases the level of HDL. The results suggest that aqueous extract of *Curcuma longa* rhizome possesses potential therapeutic value in combating atherosclerosis, which is one of the major complications of diabetes by lowering serum lipids particularly total cholesterol, triglyceride and low density lipoprotein level.

Phytochemicals are a natural bioactive compound found in plants, such as vegetables, fruits, medicinal plants, flowers, leaves and roots that work with nutrients and fibers to act as a defense system against disease or more accurately, to protect against disease [63]. The performance of *Curcuma longa* rhizome extract in reversing the negative effects of alloxan on diabetic rats may due to the presence of phytochemicals shown in table 1. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds [64]. Previous reports have demonstrated the anti-diabetic activity of triterpenoid and glycosides [65, 66]. In this study, the phytochemical investigation of *Curcuma longa* rhizome indicated the presence of alkaloids, terpenes and steroid, cardiac glycoside, resins and carbohydrates in different solvents, indicating their preferential solubility in these solvents.

5.0 Conclusion

This study has demonstrated that aqueous extract of *Curcuma longa* rhizome significantly reduces blood glucose, LDL, TG, TC and increase body weight, total protein and albumin in experimentally induced diabetic rats. Therefore, the plant has hypoglycaemic, hypolipidaemic effects at the dosage and duration of study. Further studies are needed to be carried out to isolate and identify the active principle(s) in the extract as well as elucidate its mode of action and toxicity for enhanced Phytotherapy.

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References

1. Davis SN. Insulin, oral hypoglycemic agents and the pharmacology of the endocrine pancreas. In: Goodman and Gilmans the Pharmacological Basis of Therapeutics. Brunton, L. L. (Ed.). McGraw-Hill, New York, 2006; 1613 - 1645.
2. Khalil EAM. Biochemical and histopathological studies on the influence of aqueous extract of fenugreek seed (*Trigonella foenum graecum*) on alloxan diabetic male rats. The Egyptian Journal of Hospital Medicine, 2004; *15:*83 – 94.
3. ADA. America Diabetes Association Diabetes care, 2010; 33, S1 S62-69
4. Zeggwagh NA, Ouahidi ML, Lemhadri A, Eddouks M. Study of hypoglycaemic and hypolipidemic effects of *Inula viscose* L. aqueous extract in normal and diabetic rats. Journal of Ethnopharmacology, 2006; 108: 223-227.
5. Kathleen AH. Type 1-diabetes: Prevention of the disease and its complications. Alternative Med Rev, 1996; 2: 256-281.
6. Adesokan AA1, Oyewole OI, Turay BMS. Kidney and Liver Function Parameters in Alloxan-Induced Diabetic Rats Treated with Aloe Barbadensis Juice Extract. Sierra Leone Journal of Biomedical Research, 2009; Vol. 1 (1) pp. 33-37,
7. Aloulou A, Hamden K, Elloumi D, Ali MB, Hargafi K, Jaouadi B, Ayadi F, Elfeki A, Ammar E. Hypoglycemic and antilipidemic properties of kombucha tea in alloxan-induced diabetic rats. BMC Complementary and Alternative Medicine, 2012; 12:63
8. Sharma B, Balomajumder C, Roy P. Hypoglycemic and hypolipidemic effects of flavonoid rich extract from *Eugenia jambolana* seeds on streptozotocin induced diabetic rats. Food and Chemical Toxicology, 2008; 46: 2376-2383.
9. Gupta R, Bajpai GK, Johri S, Saxena AM. An Overview of Indian Novel Traditional Medicinal Plants with Antidiabetic Potentials. Afr. J. Trad. CAM, 2008; 5 (1): 1 – 17.
10. Rahman AU, Zaman K. Medicinal plants with hypoglycemic Activity. J. Ethnopharmacol, 1989; 26: 1-33.
11. Abo KA, Fred-Jaiyesimi AA, Jaiyesimi AEA. Ethnobotanical studies of medicinal plants used in the management of diabetes mellitus in South Western Nigeria. J. Ethnopharmacol, 2008; 115: 67–71.
12. Li WL, Zheng HC, Bukuru J, Kimpe De N. Natural medicines used in the traditional Chinese medical system for therapy of diabetes mellitus. J. Ethnophamacol, 2004; 92: 1-21.
13. Oubre AY, Carlson TJ, King SR, Reaven GM. From plant to patient: an ethnomedical approach to the identification of new drugs for the treatment of NIDDM. Diabetologia, 1997; 40: 614–617.
14. Van Huyssteen M, Milne PJ, Campbell EE, Van de Venter M. Antidiabetic and Cytotoxicity Screening of Five Medicinal Plants Used by Traditional African Health Practitioners in the Nelson Mandela Metropole, South Africa. Afr. J.Trad., Complement. and Alt. Med. (AJTCAM), 2011; 8(2): 150-158.
15. Gopinathan N, M Sai Harish Singh, Chitra K, Uma Maheswara Reddy C. In Vitro Antiplatlet Activity- Ethanolic Extract of Rhizome of *Curcuma Longa* Linn. Ijbr, 2011; 2 (2)138-142
16. Sawant RS, Godghate AG. Qualitative Phytochemical Screening of Rhizomes of Curcuma Longa Linn. International Journal of Science, Environment and Technology, 2013; Vol. 2, No 4, 634 – 641
17. Aggarwal BB, Kumar A, Aggarwal MS, Shishodia S. In: Bagchi D, Preus HG. (Eds.) Phytopharmaceuticals in Cancer Chemoprevention, CRC Press: Boca Raton, FL, 2004; 349–387.
18. Cooper TH, Clark JG, Guzinski JA. In: Ho CT, Osawa T, Rosen T (Eds.) Food Phytochemicals for Cancer Prevention IIteas, spices & herbs. American Chemical Soeity: Washington DC, 1994; 23: 231-6.
19. Arora RB, Kapoor V, Basu N, Jain AP. *Ind. J. Med. Res*, 1971; 59(8) :1289-1295
20. Menon VP, Sudheer AR. *Adv Exp Med Biol*, 2007; 595:105-25.
21. Jacob A, Wu R, Zhou M, Wang P. *PPAR Res*, 2007; 89369.
22. Adhikari S, Indira-Priyadarsini K, Mukherjee T. J. Clin. Biochem. Nutr, 2007; 40(3): 174–183.
23. Kawamori T, Lubet R, Steele VE, Kelloff GJ, Kaskey RB, Rao CV, Reddy BS. Cancer Res., 1999; 59:597-601
24. Bisht S, Feldmann G, Soni S, Ravi R, Karikar C, Maitra A, Maitra A. *J. Nanobiotechnology*, 2007; 5: 3.

25. Negi PS, Jayaprakasha GK, Jagan Mohan Rao L, Sakkariah KK. *J. Agric. Food Chem*, 1999; 47:4297-4300
26. Apisariyakul A, Vanittanakom N, Budddhasukh D. *J. Ethnopharmacol*, 1995; 49:163-9
27. Bourne KZ, Bourne N, Reising SF, Stanberry LR. *Antiviral Res*, 1999; 42:219-226
28. Itthipanichpong C, Ruangrungsi N, Kemsri W, Sawasdipanich *J. Med. Assoc. Thai*, 2003; 86:299-309
29. Gautam SC, Gao X, Dulchavsky S. *Adv. Exp. Med. Biol*, 2007; 595:321-41.
30. Park EJ, Jeon CH, Ko G, Kim J, Sohn DH. J. Pharm. Pharmacol, 2000; 52:437-440
31. Trease GE, Evans WC. Pharmacognosy, 11th edn., Bailliere *Tindall*, London, 1989; pp. 45-50.
32. Sofowora A. Medicinal Plants and Traditional Medicine in Africa (2nd Edition). Spectrum Books Ltd, Ibadan, Nigeria. 1993; p. 289
33. Yakubu MT, Akanji MA, Oladiji AT. Aphrodisiac potentials of aqueous extract of Fadogia agrestis (Schweinf. Ex Heirn) stem in male albino rats. Asian J Androl, 2005; 7:399–404.
34. Henry RJ, Cannon DC, Winkleman JW. Clinical Chemistry, Principles and Techniques. Harper and Row, 2nd ed. 1974
35. Grant GH, Kacchman JF. In: "Fundamental of Clinical Chemistry" (Tietz, N.W), 3rd edition. W.B. Sauders Company, Philadelphia, 1987; Pp.298-320.
36. Trinder P. Mono reagent enzymatic glucose in clinical chemistry. (W.B. Saunders, Philandelphia/London), 1969; pp24-27.
37. Fredrickson DS, Levy RI, Lees RS. Fat transport in lipoproteins-An integrated approach to mechanisms and disorders, New England Journal of Medicine, 1967; 276 148–156.
38. Albers JJ, Warmick GR, Cheung MC. Quantitation of high density lipoproteins. Lipids, 13, 1978, 926-932.
39. Assman G, Jabs HU, Kohnert U, Nolte W, Schriewer H. Clin. Chim Acta, 1984 140, 77
40. Jacobs NJ, Van Demark PJ. Arch Biochem Biophys, 1960; 88, 250
41. Aronson D, Rayfield EJ. How hyperglycemia promotes atherosclerosis: molecular mechanisms *Cardiovascular Diabetology*, 2002; 1(1)
42. Okonkwo PO, Okoye ZSC. Hypoglycaemic effects of the aqueous extract of *Newbouldia laevis* root in rats, *Int. J. Biol. Chem. Sci*, 2009; *3(5)*, 998-1004.
43. Luka CD, Tijjani H. Comparative studies of the aqueous extracts of O*cimum gratissimum, aloe vera, brassica oleracea*and *ipomoea batatas* on some biochemical parameters in diabetic rats, *IOSR Journal of Pharmacy and Biological Sciences*, 2013; 6(3), 23-29.
44. Bajaj S, Srinivasan BP. Investigations in to the anti-diabetic activity of Azadirachtsindica, India J. Pharmacol, 1999; 31, 138-141.
45. Pari, L, Ramakrishnan R, Venkateshwaran S. Antihyperglycaemic effect of Diamed, a herbal formulation, in experimrntal diabetes in rats, J. Pharm. Pharmacol, 2001; 53, 1139-1143.
46. Mafulul SG, Luka CD, Kutshik RJ and Ojiakobi UP. Protective Effect of Ethanolic Extract of *Gongronema Latifolium* Leaves in Alloxan-Induced Diabetic Rats. IOSR Journal of Pharmacy and Biological Sciences, 2013; 2319-7676. Volume 7, Issue 6, PP 63-68
47. Xi L, Qian Z, Xu G, Zhou C, Sun S. Crocetin attenuates palmitate-induced insulin insensitivity and disordered tumor necrosis factorand adiponectin expression in rat adipocytes. Br. J. Pharmacol, 2007; 151: 610-617.
48. Youn JY, Park HY, Cho KH. Anti-hyperglycemic activity of *Commelina Communis* L. inhibition of a-glucosidase. Diabetes Res. Clin. Pract, 2004; 66: S149-S155.
49. Igbakin AP, Oloyede OB. Comparative Studies on the Hypoglycaemic, Hypoproteinaemic, Hypocholesterolaemic and Hypolipidaemic Properties of Ethanolic and Normal Saline Extracts of the Root of *Vernonia Amygdalina* in Diabetic Rats. Advances in Environmental Biology, 2009; 3(1): 33-38
50. Onunogbo CC, Ohaeri OC, Eleazu CO. Effect of Mistletoe (Viscum album) Extract on the Blood Glucose, Liver Enzymes and Electrolytes Balance in Alloxan Induced Diabetic Rats. American Journal of Biochemistry and Molecular Biology, 2012; ISSN 2150-4210. Pp1-8
51. Hfaiedh N, Murat JC, Elfeki A. Diabetes-Induced Damages in Rat Kidney and Brain and Protective Effects of Natural Antioxidants. J Nutr Food Sci, 2013; 3: 209. doi:10.4172/2155-9600.1000209
52. Murray, R.K., Granner, D.K., Mayes, P.A. and Rodwell, V.W. (2003). Harper's Illustrated Biochemistry. A Lange Medical Book, 20th Edition. McGraw-Hill, pp. 102-583
53. Murray, R.K., Granner, D.K., Mayes, P.A. and Rodwell, V.W. (2000). Harper's Biochemistry. 25th Edn., Appleton and Lange, Philadelphia, pp: 179-203, 621-630
54. Yassin, M.M., Ashour, A.R.A. and Elyazji, N.R. (2004). Alterations in body weight, protein

profile, non-protein nitrogen constituents and kidney structure in diabetic rats under glibenclamide treatment. J. Islamic Univ. Gaza, 12: p37-54
55. Mauer, S.M., Steffes, M.W. and Brown, D.M. (1981). The kidney in diabetes. Am. J. Med.70, 603.
56. Almdal, T.P., Vilstrup, H. (1988). Strict insulin treatment normalizes the organic nitrogen contents and the capacity of urea–N synthesis in experimental diabetes in rats. Diabetologica 31, 114–118
57. Sperling MA, Saunders PA. Diabetes mellitus in: Behrman RM, sKliegman HB. Jenson (Eds) Nelson text book of pediatrics, 2000, pp. 1767-1791.
58. Bhagavan NV. Metabolic homeostasis in: N.V. Bhagavan (Eds) Medical biochem Harcourt/Academic, Florida, 2002; 485-519.
59. Ranganathan G, Li C, Kern PA. The translational regulation of lipoprotein lipase indiabetic rats involves the 3'-untransalted region of lipoprotein lipase mRNA. J Biol Chem. 2000; 275(40): 986-40991.
60. Luka CD, Tijjani H, Joel EB, Ezejiofor UL and Onwukike P. Hypoglycaemic Properties of Aqueous Extracts of *Anacardium occidentale*, *Moringa oleifera*, *Vernonia amygdalina* and *Helianthus annuus:* A Comparative Study on Some Biochemical Parameters in Diabetic Rats, International Journal of Pharmaceutical Science Invention (IJPSI), 2013; Volume 2, Issue 7, PP 16-22.
61. Bopanna KN, Kannan J, Sushma G, Balaraman R, Rathod SP. Antidiabetic and antihyperlipemic effects of neem seed kernel powder on alloxan diabetic rabbits. Indian Journal of Pharmacology, 1997; 29: 162–167.
62. Bonadonna RC, Groop LC, Zych K, Shank M, DeFronzo RA. Dose-dependent effect of insulin on plasma free fatty acid turnover and oxidation in humans American Journal of Physiology, 1990; 259: E736–E750.
63. Krishnaiah D., Devi T., Bono A. and Sarbatly R. Studies on phytochemical constituents of six Malaysian medicinal plants. Journal of Medicinal Plants Research, 2009; Vol. 3(2), pp. 067-072 ISSN 1996-0875©
64. Okwu DE. Evaluation of the chemical composition of indigenous spices and flavoring agents Global J. Pure and Appl. Sci, 2001; 7(3): 455-459
65. Reher G, Slijepcevic M, Krans L. Hypoglycaemic activity of triterpenes and tannins from Sarcopoterium spinosum and two Sanguisorba Species. Planta Med, 1991; 57: A57-A58.
66. Kako M, Miura T, Nishiyama Y, Ichimaru M, Mariyasu M, Kato A. Hypoglycemic activity of some Triterpenoid Glycosides. J. Nat. Prod, 1997; 60: 604-605.

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