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PHYTOCHEMICAL CONSTITUENTS AND HYPOGLYCEMIC EFFECTOF METHANOL EXTRACT OF *Vitellaria paradoxa* STEM BARK IN ALLOXAN INDUCED DIABETIC RATS

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Abstract
Vitellaria paradoxa, commonly known as Shea butter tree, is a highly medicinal plant which is locally abundant in Nigeria. This study aims to investigate the phytochemical constituents and hypoglycemic activity of methanol extract of *Vitellaria paradoxa* stem bark in alloxan induced diabetic rats. The phytochemical screening was carried out using standard procedures and diabetes was induced by a single intraperitoneal dose of 120mg/kg body weight of alloxan and 6hrs later, the animals were kept on 20% glucose for 24hrs.The result showed these phytoconstituents; alkaloids, flavonoids, anthraquinones, saponins, tannins, phlobatanins and cardiac glycosides were present. The oral administration of extract at a dose of 500mg/kg body weight showed a significant reduction ($P<0.05$) in the blood glucose level of diabetic rats from 168.75±10.61 - 95.25±1.65 while the standard drug (Glibenclamide 0.5mg/kg body weight) decreased Fasting blood glucose levels from 162.25±5.36 - 86.25±3.40. There was a significant increased ($P>0.05$) in blood glucose level of induced diabetic but not treated rats from 192.50±8.86 to 195.25±12.96 compared to non-induced but treated with extracts from 101.25±3.73 to 81.00±5.98. These results suggest that the extract may possess some anti-hyperglycemic potentials.

Keywords: *Vitellaria paradoxa,* alloxan, phytochemical constituents, hypoglycemic

1.0 Introduction

World Health Organization (WHO) defined diabetes mellitus as a health disorder resulting from metabolic disorders of numerous etiology which is associated with chronic hyperglycemia with disturbances in the metabolism of carbohydrates, fats and proteins owing to the defects in insulin secretion or action, or combination of the two. Long-term damage, dysfunction and failure of several organs are effects associated with diabetes mellitus. Symptoms of diabetes mellitus may include thirst, polyuria, blear vision and loss of body weight (Rakesh *et al.,* 2015). Diabetes mellitus (DM) is the most common endocrine disorder affecting approximately 5% of the world population. In the year 2000, the estimated occurrence of diabetes in Nigeria was 1.707 million. It is projected by WHO that by 2030, the number of diabetics would be 4.835 million. Hyperglycemia is the principal cause of complications such as retinopathy, neuropathy, nephropathy, cardiovascular complications and ulceration. The effective blood glucose control is the key to improving the quality of life in patients with diabetes (Defronzo *et al.,* 1992; Laha and Paul, 201). Insulin therapy and oral hypoglycemic drugs have been used for the treatment of

diabetes mellitus. The drawbacks in the use of the insulin therapy and hypoglycemic drugs are the adverse side effects exerted by these synthetic anti-diabetic agents on human body (Cinmay *et al.,* 2015). Since time immemorial, varieties of plants and their produce have been used by human as foods or drugs in treatment of disease (Sodipo *et al.,* 2001).Medicinal plant extracts have been valuable antidiabetic agents and may involve one or more active component responsible for blood glucose reduction (Grover, 2002). Based on the WHO recommendations hypoglycemic agents of plants origin used in traditional medicine are important. There are many literature reviews by different authors on the plants with hypoglycemic effect, but most informative is the review of Atta-Ar-Rahman who has documented over 300 plant species for their antihyperglycemia activity. WHO listed 21,000 plants which are used for pharmacological purposes (Cinmay *et al.,* 2015). Abdel *et al.,* (2014) reported that natural products are the major source of raw materials for the development of modern drugs. Herbal preparations are used for managing health conditions mostly by rural area dwellers due to its availability, low cost and lesser side effects.

Vitellaria paradoxa belongs to the family Sapotaceae. It is the only species in the genus which is of Africa origin (Lovett and Hag, 2000). It is a plant that is locally abundant in Nigeria in the derived savannah Zones, particularly near thousands of villages (Adegoke *et al.,* 2007). It is rich in oil and replaces oil palm as source of edible oil in Northern Nigeria. Shea butter is the fat extracted from the kernel of Shea fruits. It is becoming increasingly popular as a component of cosmetic formulation or addition to its long standing use as cocoa butter substitute in the chocolate industry (Adgidizi *et al.,* 2008). Shea butter contains high level of uv-absorbing triterpenes esters which include cinnamic acid, tocopherols (vitamin A), and phytosterols (Wiesman *et al.,* 2003). Shea butter oil is thoroughly used as cooking fat in Africa and serves as a suitable base for tropicalmedicine used locally to relieve inflammation of the nostril, skin burn, dermatitis, rheumatic andjoint pains. There are no reports of allergic reaction owing to consumption of shea butter nuts or its produce (Watson, 2005). The consumption of plants and their produce have been associated with reduced risk of disease like Alzheimer disease, cancer, lipid peroxidation and other free radical driven disorder as well as malaria. Medicinal potentials of locally available plants and their produce was from the claim of some herbalists, native and ancient that some plants and their produce have medical value but they lack enough empirical proofs to justify these claims. Scientific evaluation of medicinal plant is important in the discovery of novel drugs and also helps to assess risk associated with the use of conventional drugs of herbal origin (Mroeh, 2008).

2.0 Materials and Methods

The fresh stem bark of *vitellaria paradoxa* were collected in August 2018 from bosso secondary school Minna- Niger state, Nigeria. Authentication and identification was done in the department of Biological science, Federal University of Technology, Minna, Niger state. All chemical and reagents used were of analytical grade.

Sample Preparation and Extraction

Fresh stem bark of *Vitellaria paradoxa* was washed, rinsed and air-dried in the laboratory room temperature for a period of two weeks and pre-crushed manually using mortar and pestle. It was further pounded into powder and kept in polythene bag. 200g of powdered stem bark of *vitellaria paradoxa* were divided into a four portion and each 50g was dissolved in 400ml of 70% methanol and was dispensed into a reflux apparatus for extraction at 60°C for 3hrs. The extract

was filtered using filtered paper. The filtrate was concentrated using rotary evaporator and water bath. The extract was collected, weighed and stored in avail, and then kept in the refrigerator until required for used.

Phytochemical Screening

The methanol extract of *Vitellaria paradoxa* stem bark was screened for qualitative phytochemical properties by standard methods of (Trease and Evans, 2002; Sofowora *et al.,* 1982)

Saponins

To 0.5 mL of extract, 10mL of distilled water was added. Frothing which persist on warming of test confirms the preliminary evidence for the presence of Saponins (Trease and Evans, 2002).

Tannins

About 3 mL of the extract was added to 5ml of distilled water and then heated in water bath. To this solution, iron (III) chloride (FeCL3) solution was added. A blue black or green precipitate indicated the presence of tannins (Trease and Evans, 2002).

Alkaloids

To 2mL of the extract, an aliquot of 10 mL of 1%HCL in water bath for 30minutes. The solution was then treated with few drops of Wagner's reagent and colour change was observed. Presence of alkaloids was indicated with precipitate formation (Sofowora, 1982).

Cardiac Glycosides

An aliquot (2 mL) of chloroform was used to dilute 0.5mL of extract. Sulfuric acid was carefully added to the solution drop wise. At the sulfuric acid interphase, a reddish coloration indicated the presence of cardiac glycosides (Trease and Evans, 2002).

Anthraquinones

To 5 mL of the extract, 10 mL of benzene, 5mL of 10% aqueous ammonia was added and shaken. Presence of anthraquinones was indicated by a pink/violet colour in the ammonia phase at the bottom of test tube (Sofowora, 1981)

Phlobatanins

About 0.5g of the extract was left boiling with 5ml of 1% HCL in a test-tube. A red precipitate indicated the presence of phlobatanins (Aktar *et al.,* 2015*).*

Flavonoids

To 4ml of extracts, 1.5ml of 50%% methanol was added. The solution was slightly heated and metal magnesium was added. Then 5 drops of concentrated hydrochloric acid to the solution and observed for red coloration.

Reducing Sugars

To 0.5ml of extract solution, 1ml of distilled water is added and about 6 drops of Fehling's solution was prepared by mixing equal volumes of equal molar concentrations of A and B. The formation of brick red precipitates is an indication of the presence of reducing sugars.

Preparation of Stock Solution

The concentrated methanol extract of *Vitellariaparadoxa* stem bark(1 g) was dissolved in 10ml of distilled water and the prepared solution was stored in the refrigerator.

Experimental Animals

ThirtyWistar rats of both sexes (8-11 week) old obtained from the animal house of Ahmadu Bello University, Zariaweighing between 100 -150g were used for the study. The rats were housedin well ventilated plastic cages, allowed free access to food, water and acclimatizedfor two weeks.

Induction of Diabetes

Prior to diabetic induction, the blood glucose concentration of the rats was taken. The rats were fasted for 12hrs and diabetes was induced by intraperitoneal injection of alloxan at a dose of 120mg/kg body weight and 6hrs later the animals were kept on 20% glucose for next 24hrs. After 72hrs(3days) of induction, the animals with blood glucose level above 150mg/dl were selected for the experiment.

Experimental Design

The rats were divided into the groups belowwith each group consisting of 6 rats.

GROUP A; Alloxan induced diabetic rats treated with 500mg/kg body weight of extract

GROUP B; Alloxan induced diabetic rats treated with 0.5mg/kg body weight of standard

GROUP C; Alloxan induced diabetic rats but not treated

GROUP D; Non-induced rats but treated with 500mg/kg body weight of extract

GROUP E; Non- induced not treated (normal).

Determination of Blood Glucose Level

The blood glucose level was determined using Accu-check glucometer and glucose strips. The rats were fasted for 12 hours before the tail vein was punctured and blood from the tail was dropped on the tip of glucose strip placed in a glucometer, and blood glucose concentration was recorded in mg/dl

Determination of Bodyweight

The bodyweight of rats was determined using dry cell battery weighing balance.The weight was monitored before induction, after induction and during period of treatment weekly.

Oral Administration Of Extracts

Methanol crude extract (500 mg/kg bodyweight) was administered orally to group A and D while 0.5mg/kg body weight of standard drug (glibenclamide) was administered to group B

Stastical Analysis

The data obtained were subjected to Analysis of Variance (one- way Anova) using SPSS statistical package. Means were separated using Duncan's Multiple Range Test (DMRT). The data was given as mean ±SEM (Standard error mean). Differences were considered significant at $P < 0.05$.

3.0 Results and Discussion

Table 1: Phytoconstituents of the methanol extract of *Vitellaria paradoxa* stem bark

Phytochemical	Inference
Alkaloids	+
Saponins	+
Reducing sugar	-
Flavonoids	+
Anthraquinones	+
Cardiac glycosides	+
Phlobatannins	+
Tannins	+

Key: + = present, - = absent

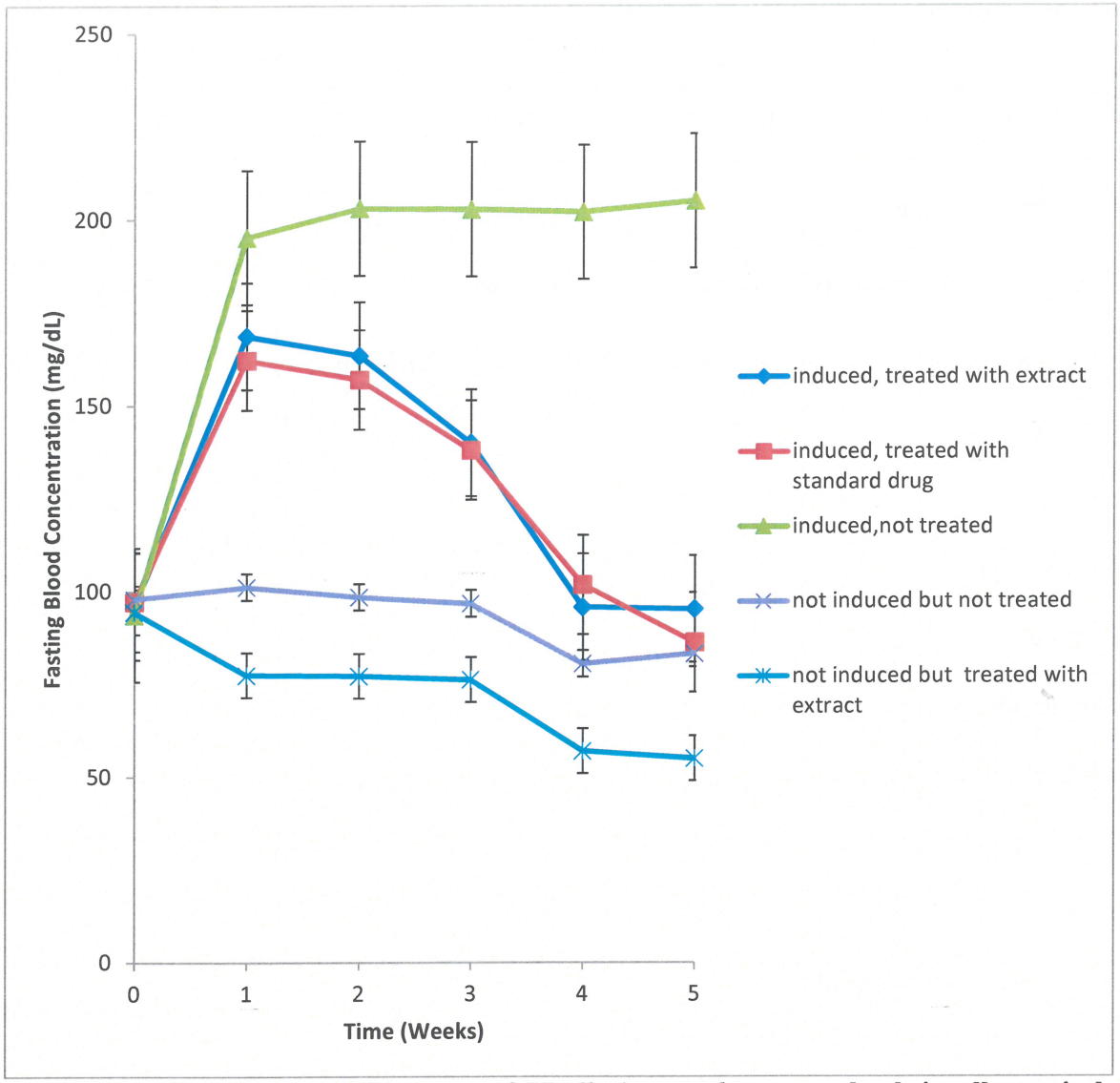

Figure 1: Effect of methanol extract of *Vitellaria paradoxa* stem bark in alloxan induced diabetic rats

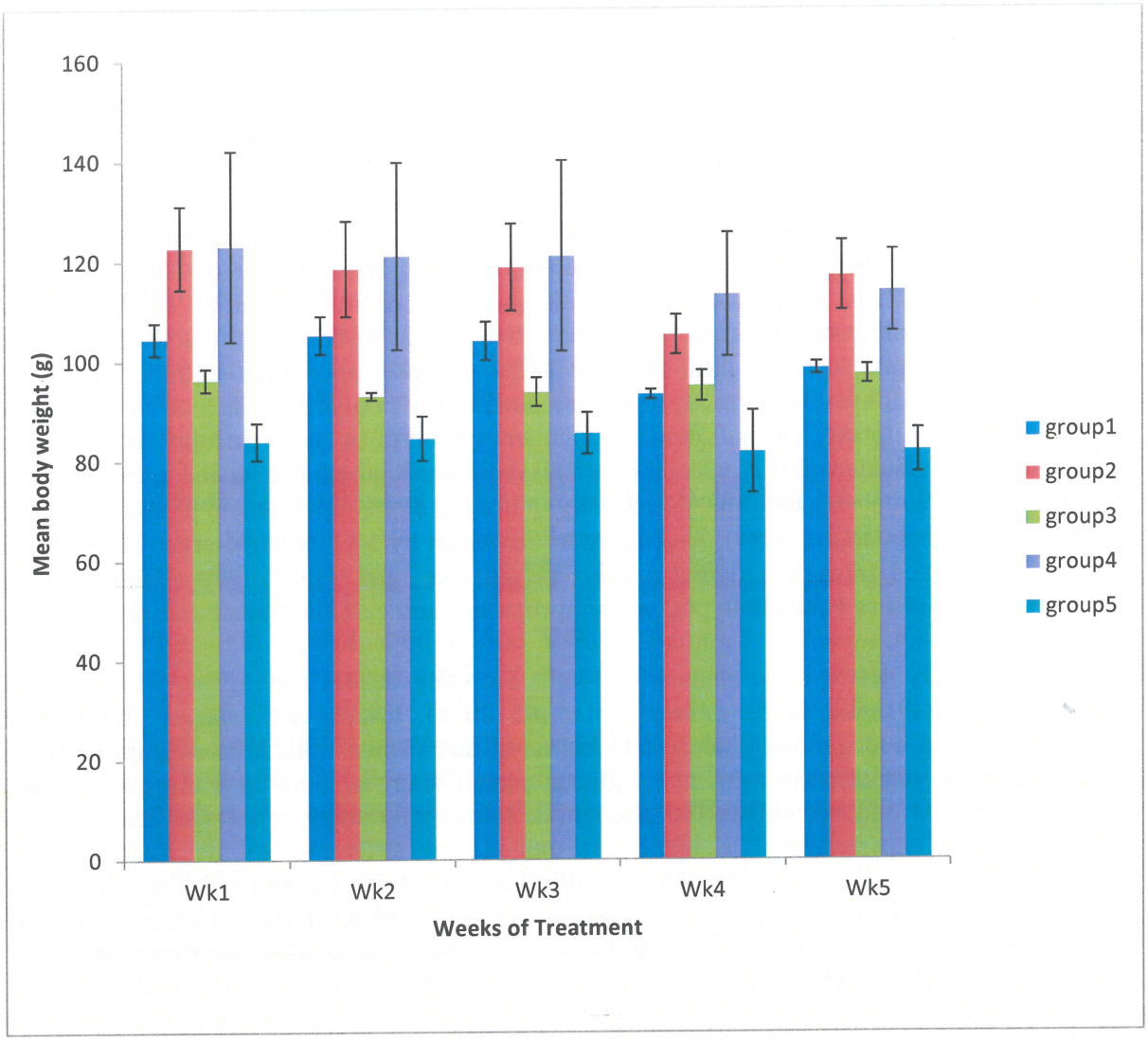

Figure 2: Effect of methanol extract of *Vitellaria paradoxa* stem bark on body weight of alloxan induced diabetic rats.

Key:
Grp1-induced treated with 500mg/kg body weight of extract
Grp2-induced treated with 5mg/kg body weight of standard drug (glibenclamide)
Grp3-induced not treated
Grp4-no- induced but treated with extract
Grp5-non- induced not treated (normal

This study has revealed the presence of seven phytoconstituents in the methanol extract of the stem bark of *vitellaria paradoxa*(Table 1). The phytocomposition includes; alkaloids, flavonoids, anthraquinones, saponins, tannins, phlobatanins and cardiac glycosides. This result has been similarly reported by Onwuliri, 2004 and Falana *et al.,* 2016. The insignificant difference ($P>0.05$) hypoglycemic potentials observed in alloxan-induced diabetic rats treated with

500mg/kg body of methanol extract of *vitellaria paradoxa* stem bark when compared with the standard drug glibenclamide administered at 0.5mg/kg bw (Figure 1) showed some promising hypoglycemic activity. This result correlates with the work of Abubakar et al., 2019 that the Crude Methanol extract of *Vitellaria paradoxa* leaf had significant hypoglycemic and antioxidant activity. The hypoglycemic activity of the plant under study may be linked to the individual or synergistic action of of phytoconstituents present. Flavonoids have been reported by many authors to mediate it antihyperglycemic activity by potentiating the insulin effect either by increasing the pancreatic insulin secretion by the cells of islet of langerhan or it enhancing it release from bound insulin (Pari and Amarnath, 2004; Saidu et al., 2014: Abubakar et al., 2014). Alkaloids are also known to exert a wide range of antidiabetic activities through different mechanisms such as inhibiting intestinal glucose uptake or stimulating insulin secreation (Chattopadhyay, 1999). Saponins are potential hypoglycemic agents which acts by increasing glucose uptake and subsequent glycogen synthesis or as an insulin secretaguogue (Naik et al., 199; Santos, 2012). Tannins displayed beneficial effects in the prevention and treatment of type 2 diabetes mellitus, possibly by inhibiting alpha-amylase and alpha-glucosidase activities (Palanisamy *et al*., 2011). According to the report by Jouzier and Berké (2012), tannic substances extracted from *Eucalyptusglobulus* (*Myrtaceae*) and fruits of *Terminalia catappa* (Combretaceae) possess valuable hypoglycemic properties and exhibit antidiabetic activity

Alloxan selectively destroys the pancreatic β-cells secreting insulin leaving the less active cell thus resulting in diabetic state. (Szkudelski *et al.,* 2001). Insulin deficiency or insensitivity causes increase in endogenous glucose concentration due to non-utilization of glucose by the cells. This causes increased in demand for gluconeogenic sources, there by leading to excessive breakdown of fats and protein as the alternative source of energy. The significant increase ($P<0.05$) in the body weight of diabetic rats treated with 500mg/kg of methanol extract of *vitellaria paradoxa* stem bark compared with the untreated rats showed that the weight improvement might be due to proper maintenance of glucose homeostasis while the decreased in body weight of the untreated rats might be due to excessive breaking down of tissue protein and lipid cause by insulin insufficiency (Ramachandran *et al.,* 2012).

4.0 Conclussion

This study reveals that methanol extract of*Vitellaria paradoxa* stem bark possess some important phytochemical constituent that has hypoglycemic potentials

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References

Abdel N. S., Fadia, S., Youssef and Mohamed Ashour, L. (2014).Medicinal Plants with Potential Antidiabetic activity & their Assesment. *Medicinal & Aromatic Plants,*3(151) 2167-2412.

Abubakar, N. A.,Oise, A.E. and Saidu, A. N. (2014). Phytochemical Constituents and Hypoglycemic Effect of Aqueous and Ethanolic Extracts of *Murraya Koenigii* in Alloxan Induced Diabetic Rats. *Journal of Dental and Medical Sciences*, 13 (9) 8-12

Abubakar, A.N, Saidu, A.N., Akanya, H. O. Egwim, E.C. (2019). Antioxidants and hypoglycemic effect of some medicinal plants, GSC *Biological and Pharmaceutical Sciences,* 08 (02) 70-90

Adegoke, K. S., Ndukwe, I. G., Amupitan, J. O., and Isah, Y. (2007).Phytochemical screening of the crude extract of the roots, stem bark and leaves of *vitellaria paradoxa* (GAERTN. F). *Africanjournal of Biotechnology*, 16, 1905-1909.

Adgidizi, P. P., and Ogbonnaya, C. (2008), Evaluation of some Physico-chemical properties of shea butter related to its value for food and industrial utilization. *International Journal of Post-Harvest*, 3, 9-17

Akhtar, S., Ismail, T., Fraternale, D., Sestili, P. (2015). Pomegranate peel and peel extracts: Chemistry and food features. *Food Chemistry*, 174, 417-425.

Chattopadhyay, R.R. (1999). A comparative evaluation of some blood sugar lowering agents of plant origin. *Journal. Ethnopharmacoly, 67*, 367–372.

Cinmay, D., Deshumukh and Anurekha J. (2015). Diabetes Mellitus: A Review, *International Journal of Pure and Applied Bioscience*, 2015, 3(3):227-228.

Defronzo, R. A., Bonadonna, R. C., and Ferrannini, E. (1992). Pathogenesis of NIDDM.A balance overview. *Diabetes care,*15, 318-368.

Falana, M. B., Bankole, M. O., Ojo, D. A., & Omemu, A. M. (2016). Comparative Phytochemical Investigation of the Various Parts of *Vitellaria Paradoxa*. *Journal of Natural Sciences Research*, (6) 11.

Grover, J. K., Yadav, S., and Vats, V. (2002). Medicinal plants of India with anti-diabetic potential. *Journal of Ethnopharmacology*, 81, 81-100

Jouzier E., & Berké B., (2012). Diabète et Philatélie II – Plantes hypoglycémiantes. *Bulletin de la Société de Pharmacologie deBordeaux*, 151(1-4), 141-170.

Laha S, Paul S *Gymnema sylvestre* Gurmar (2019). A Potent Herb with Anti-Diabetic Antioxidant Potential *Pharmacology journal*, 11(2):201-6

Lovett, P., and Hag, N.(2000). Evidence for a tropic selection of the Shea nut tree (*Vitellariaparadoxa*). *Agro-forestry Systems*, 48, 293-304.

Mroeh, Mohammed. (2008).The effect of oil Enriched Diets on plasma lipid profile, Glucose and coagulation. *PAN Arabic Medical Journal,* 34, 7-54.

Naik SR, Barbosa Filho JM, Dhuley JN, Deshmuleh V (1991). Probable mechanism of hypoglycaemic activity of basic acid, a natural product isolated from *Bumelia sartorum. Journal Ethnopharmacol.* 33: 37-44.

Onwuliri, F. C. (2004). Antimicrobial studies of the extracts of *Acalypha wilkesiana L.* on microorganisms associated with wound and skin infections. *West African Journal of Biological Science*, vol.15, pp. 15-17

Palanisamy,D., Ling,L. T.,Manaharan T., andAppleton, D. (2011). "Rapid isolation of geraniin from *Nephelium lappaceum* rind waste and its anti-hyperglycemic activity," *Food. Chem.*, vol. 127, pp. 21–27,

Pari, l., and Amarnath, S. M. (2004). Antidiabetic activity of Boerhaavia diffusa L.: Effect on hepatic key enzymes in experimental diabetes. *Journal of Ethnopharmacology*, 91,109–113. [Crossref], [PubMed], [Web of Science ®], [Google Scholar].

Rakesh, Joshi, N. William, Setzer, F. Valdir, Da Veiga Junior (2015). Aromatic and Medicinal Plants with Anti-Diabetic Potential from India: A Review, *American Journal of Essential Oils and Natural Products,* 2(4), 24.

Ramachandran, S., Rajasekaran., Manisenthilkumar.(2012). Investigation of hypoglycemic, hypolipidemic and antioxidant activities of aqueous extract of *Terminalia paniculata* bark indiabetic rats. *Asian Pacific Journal of Tropical Biomedicine,*2(4), 262–268. [PMC free article] [PubMed]

Saidu, AN, Abubakar, AN, Daniel, MU Kabiru, AY (2014) Phytochemical Screening Effects of MethanolicExtract of*AzadirachtaIndica* Leaf in Alloxan Induced Diabetic Rats *IOSR Journal of Pharmacy Biological Sciences (IOSR-JPBS) e-ISSN: 2278-3008, p-ISSN:2319-7676 Volume 9, Issue 1 Ver VI (Feb 2014), PP 16-20 wwwiosrjournalsorg* wwwiosrjournalsorg 16 | Page

Santos,F.A.,Frota,J.T.,Arruda,B.R.,Melo,T.S.,Silva,A.A.C.A.,Brito,G.A.C.,Chaves,M.H., Rao,V.S., (2012). Antihyperglycemic and hypolipidemic effects of α,β-amyrin, a triterpenoid mixture from Protium heptaphyllum in mice..*LipidsHealthDis*.11, 98–105

Sodipo, O. A., Modu, S., and Ene, A. C. (2001). Studies on the effect of administration of various concentration of black caraway oil on some biochemical and histopathological parameters in alloxan induced diabetic rats. *Bioscience Research communication*. 13, 263-266.

Sofowora, A. (1982). Medicinal plant and traditional medicine in Africa. Published in Association with Spectrum Books Limited Ibadan, by John Wiley and Sons, Chinchester, *New York*, pp 66-91

Szkudelski, T. (2001). The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. *Physiological Research*, 2001, 50, 537–546.

Trease G. E., and Evans, W. C. (2002). Text book of pharmacognosy.14th edition, Balliere Tindall, London, pp-150- 153

Watson, Shea butter institute online hand book, 2005, www.americansheainstitute.com

Wiesman, Z., Maranz, S., Bisgaard J., and Bianchi, G. (2003). Germplasm resources of *Vitellariaparadoxa* based on variations in fat composition across the species distribution range. *Agroforestry Systems*. 60, 71-76.