

# HYPOGLYCEMIC AND HYPOLIPIDEMIC EFFECTS OF FLAVONOID EXTRACT FROM WONDERFUL KOLA (*BUCHHOLZIA CORIACEA*) SEED IN FRUCTOSE FED STREPTOZOTOCIN INDUCED TYPE 2 DIABETIC ALBINO RATS.

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## ABSTRACT

Wonderful kola (*Buchholzia coriacea*) is a plant use traditionally for treating diabetes, cold, cough and catarrh. The present study evaluates the possible hypoglycemic activity and ameliorating effects of oral administration of fructose-flavonoid rich seed extract of *Buchholzia coriacea* seeds, in Streptozotocin (STZ)-induced diabetic rats. A total of twenty five (25) rats were used for this study. They were divided into five (5) groups of five rats each; Normal control, negative control (Streptozotocin (STZ)-induced diabetic), positive control (Glibenclamide), 100mg/kg b.w. of the seed extract and 500mg/kg b.w. of the seed extract. Fasting blood glucose (FBG) levels were evaluated before and after extracts administration. The flavonoid extract significantly decreased ( $P<0.05$ ) FBG in hyperglycemic rats as the weeks progresses after extract administration. The administration of flavonoid and Glibenclamide (standard antidiabetic drug) for 6 weeks significantly lowered ( $P<0.05$ ) FBG level in STZ-induced diabetic rats. The extract significantly decreased ( $P<0.05$ ) the elevated levels of serum total cholesterol, triglyceride and thiobarbituric acid reactive species (TBARS) products in diabetic rats. In conclusion, the results suggest that *Buchholzia coriacea* seeds contain a potent hypoglycemic and antilipidemic effects which will could be a scientific merit in the traditional use of the extract in the management of diabetes and the improvement of its complications.

**Keywords:** *Buchholzia coriacea*, hypoglycemic, streptozotocin, flavonoid, cholesterol, antioxidant.

## INTRODUCTION:

Diabetes mellitus is a heterogeneous group of metabolic disorders characterized by hyperglycaemia and glucose intolerance resulting from inadequate insulin uptake or reduced effectiveness of insulin on its target site in stimulating glucose uptake. The presence of sugar signals the endocrine pancreas to secrete the hormone, insulin. Insulin facilitates the uptake and storage of sugar for the metabolic needs of the body tissues, particularly the liver, muscle and adipose. (Zheng, *et al.*, 2009). Persistent hyperglycaemia leads to other complications, including non-enzymatic protein glycation, hyperlipidaemia, and oxidative stress, all of which are thought to be pathogenic for the development of diabetic complications (Kangralkar, *et al.*, 2010). Since diabetes is a multifactorial chronic disease, many therapeutic goals and variety of drugs can be

used in diabetotherapy. However, the currently available diabetes medicaments, with the exception of metformin, remain largely unavailable for most patients in Africa and other developing countries. Therefore, these patients often consult traditional healers who rely heavily on medicinal plants to treatment of diabetes (Ekor, 2014).

Hyperlipidemia is a lipid disorder hence alters lipid profile. It is characterized by elevated serum total cholesterol, low density lipoprotein, very low density lipoprotein and decreased high density lipoprotein levels (Rerkasen, *et al.*, 2008) though may be asymptomatic. Hyperlipidemia causes atherosclerotic cardiovascular disease which eventually affects organs such as the kidney, leading to glomerular injury, interstitial fibrosis and tubular atrophy, ischemic nephropathy and End Stage Renal Disease (Mühlfeld, *et al.*, 2004) Increase in body weight and certain organs such as liver and spleen are also associated with hyperlipidemia due to increase in cholesterol and triglycerides and infiltration of these lipids to the organs respectively

*Buchholzia coriacea*, is a forest tree with large, sleek leaves and conspicuous cream white flower in racemes at the end of the branches (Adisa, *et al.*, 2011). The plant is effortlessly diagnosed with the aid of using the compound pinnate leaves and the lengthy slim angular end result containing large, generally aligned seeds. In Nigeria, *B. coriacea* is a perennial plant which grows as a tree. It belongs to the family of capparaceae and its neighborhood include “Uwuro” (Yoruba-Nigeria), “esson bossi” (Central Africa), “Uke” (Igbo-Nigeria), the plant has diverse not unusual place names including “Ovu (Bini), and Aponmu (Akure) (Keay, *et al.*, 1964). *Buchholzia coriacea* is being utilized in different part of Africa for treatment of diverse illnesses and infection. Its utility vary from country to country in Africa. In the Ivory coast the twig bark decoction of the plant *B. coriacea* is used for the treatment of rheumatism and kidney ache, it's also used for the treatment of infections of the eye (bark gruel poured into the flat of the hand and inhaled) and for the treatment of pain in the back (fruit pulp massaged in)

In Nigeria, the fruit of the plant *B. coriacea* is used as an anthelmintic. In Liberia, the seeds of the plant *B. coriacea* are used internally towards worms and pain, externally towards pores and skin diseases. In Cameroon, *B. coriacea* seed is used for the treatment of coughs. Young leaves of the plant *B. coriacea* are utilized in a gruel poultice for ulcers and boils. In Gabon, pounded bark of the plant *B. coriacea* is used as a lotion towards scabies. In former times younger warriors had been given fresh roots of *B. coriacea* to stimulate them earlier for the battle (Nwachukwu, *et al.*, 2014). This study was therefore carried out to explore the possible efficacy of preventive and therapeutic administration of *B. coriacea* flavonoid-rich seed extract on streptozotocin (STZ)-induced diabetic rats.

## **METHODS**

### **PLANT MATERIAL**

Fresh seeds of *Buchholzia coriacea* (5kg) Cameroun were purchased at a local market (Relief Market) in Imo State, Nigeria. The seeds were identified and authenticated by a botanist, Dr. C.N. Duru of Biotechnology Department, Federal University of Technology, Owerri, Imo State. The

seeds were immediately washed to remove earthly materials, peeled, chopped and shade-dried for 1 week on laboratory trays. The dried seeds were pulverized to powder and weighed.

## **EXTRACTION**

The powdered seeds (800 g) were soaked in 95% methanol for 48 hrs with intermittent shaking, and filtered. The filtrate was concentrated by evaporation to dryness using a rotary evaporator, under reduced pressure at a temperature of 40 °C. The crude extract of *Bulchhozia coriacea* (BCE), was stored in airtight container in a refrigerator until used.

## **EXTRACTION OF FLAVONOID (Chu *et al.*, 2002)**

Two hundred grams (200g) was dissolved in 20 ml of 10% H<sub>2</sub>SO<sub>4</sub> and hydrolyzed by heating in water bath for 30 min at 100°C. The mixture was placed on ice for 15 min for precipitation of the flavonoid aglycones. The flavonoid aglycones were then dissolved in 50 ml of warm 95% ethanol, filtered and concentrated by rotary evaporation at 55°C.

## **EXPERIMENTAL ANIMALS**

Albino rats were purchased from the college of veterinary, Michael Okpara University of Agriculture, Nigeria. The animals were housed in animal cages under standard laboratory conditions, allowed free access to standard rat pellet and water. The animals were acclimatized for 10 days prior to commencement of the experiment. All experimental animals were in accordance with the guidelines of both the University's ethical committee and the International Guidelines for Handling of Laboratory Animals (Derrell, 1996).

## **INDUCTION OF TYPE 2 DIABETES**

Type 2 diabetes was induced according to the method as described by Rachel and Shahidul (2012). After giving water containing 10% fructose to rats for 14 days, streptozotocin (40 mg/kg body weight) in ice-cold 0.1 M citrate buffer (pH 4.5) was administered to the animals after an overnight fast. After 72 hours of streptozotocin administration, blood glucose level was checked using glucometer. Animals with blood glucose levels  $\geq$  250 mg/dl were considered diabetic.

## **3.8 EXPERIMENTAL DESIGN**

Twenty-five (25) albino rats were used for this study. The animals were randomly placed groups with five animals each. Group 1 served as the normal control (feed and water only), Group 2 were the diabetic control (STZ plus water and feed only), Group 3 served as the positive control (glibenclamide plus feed and water only), while group 4 and 5 served as the test groups with the concentrations: 100mg/kg and 500mg/kg body weight of the plant extract.

All animals were administered with the extract for 6 weeks, and sacrificed 24 hours after the last treatment.

**Table 3.1 Table of Experimental Design**

groups	Number of animals	Treatment (all the administration measurement are in mg/kg body weight).
<b>1 (Normal control)</b>	5	Receives feed and water only
<b>2 (Diabetic control)</b>	5	Streptozotocin 40mg/kg + feed and water only
<b>3 (positive drug control)</b>	5	5 mg/kg glibenclamide + feed and water only
<b>4 (Test)</b>	5	100 mg/kg of <i>B. coriacea</i> flavonoid extract
<b>5 (Test)</b>	5	500 mg/kg of <i>B. coriacea</i> flavonoid extract

### FASTING BLOOD SUGAR LEVEL ASSESSEMENT

This was carried out on weekly bases. The animals were fasted overnight for the fasting blood sugar level checks. They were weighed before blood samples were collected through ocular puncture. A glucometer was used to check the sugar level. Rats with FBS higher than 126 mg/dL were used in the experiments.

### DETERMINATION OF LIPID PROFILE

Total serum cholesterol was determined by the method of Allain *et al.*, (1974).

The method of Friedwald *et al.*, (1972) was used to calculate LDL-cholesterol

HDL- cholesterol was determined by the method of Grove (1979) and Burstein *et al.*, (1980).

The method of Fossati and Principle. (1982) used in determining plasma triacylglycerol.

### RESULTS

**Table 1: Result showing effect of *Buchholzia coriacea* seed flavonoid extract on fasting blood sugar concentration (mg/dl) of fructose-streptozotocin induced type II diabetes.**

Groups	Week 1	Week 2	Week 3	Week4	Week 5	Week 6
<b>Normal control</b>	120.00±3.74 <sup>a</sup>	124.00±5.05 <sup>a</sup>	130.25±4.65 <sup>a</sup>	132.25±4.86 <sup>ab</sup>	119.75±5.74 <sup>a</sup>	111.75±8.50 <sup>a</sup>
<b>(Diabetic)</b>	135.75±4.35 <sup>b</sup>	166.88±4.63 <sup>b</sup>	170.50±1.92 <sup>b</sup>	173.00±2.94 <sup>c</sup>	163.50±16.46 <sup>b</sup>	158.75±11.79 <sup>b</sup>
<b>Negative control</b>						
<b>Positive Control</b>	134.25±5.91 <sup>ab</sup>	132.25±6.70 <sup>a</sup>	130.75±4.65 <sup>a</sup>	126.25±3.86 <sup>ab</sup>	118.00±4.69 <sup>a</sup>	113.75±5.32 <sup>a</sup>
<b>100mg/kg b.w.</b>	118.75±18.55 <sup>a</sup>	126.38±15.20 <sup>a</sup>	126.50±19.16 <sup>a</sup>	117.25±18.39 <sup>a</sup>	107.25±17.65 <sup>a</sup>	106.50±17.75 <sup>a</sup>
<b>500mg/kg b.w.</b>	146.50±9.04 <sup>a</sup>	164.63±4.61 <sup>b</sup>	162.75±7.80 <sup>b</sup>	142.75±25.93 <sup>b</sup>	119.75±7.89 <sup>a</sup>	116.25±9.64 <sup>a</sup>

Treatments with superscripts a,b,c,d showed significant difference ( $p < 0.05$ ) compared with the diabetic control rat group. Values are expressed as mean  $\pm$  standard deviation (n=4). Values with the same superscript are not significantly different. ( $p > 0.05$ )

From the result table, there was an increase in the blood sugar level in negative (diabetic) group. This could be as a result of increase in the feed consumption and infection from the environment. The most reduction effect was seen in the group 100mg/kg b.w. of the seed extract as compared to the positive control group which was treated with glibenclamide. This shows that, it is dose-dependent in action as there is increment in sugar level in group 500mg/kg b.w. as compared to the normal control group. The reduction of sugar level seen in 100mg/kg b.w. group is an indication that the plant seed extract was able to restore to normal level as that of normal control group as the week progresses.

**Table 2: Result showing the result for lipid profile level in streptozotocin induced type II diabetes in albino rat.**

GROUPS	HDL (mmol/L)	LDL (mmol/L)	VLDL (mmol/L)	CHOL. (mmol/L)	TAGS (mmol/L)
<b>Normal Control</b>	59.71±2.93 <sup>a</sup>	23.28±2.68 <sup>a</sup>	20.24±4.53 <sup>a</sup>	103.23±7.42 <sup>a</sup>	124.18±5.50 <sup>a</sup>
<b>Negative Control</b>	51.29±2.19 <sup>b</sup>	55.19±3.23 <sup>d</sup>	38.80±6.21 <sup>c</sup>	145.28±6.35 <sup>d</sup>	158.96±5.57 <sup>d</sup>
<b>Positive control</b>	56.03±3.08 <sup>a</sup>	33.57±4.51 <sup>b</sup>	28.59±1.57 <sup>b</sup>	118.19±8.23 <sup>b</sup>	141.99±7.23 <sup>bc</sup>
<b>100mg/kg</b>	48.59±2.94 <sup>b</sup>	47.94±1.61 <sup>c</sup>	39.23±4.81 <sup>c</sup>	135.76±5.01 <sup>cd</sup>	148.91±1.78 <sup>c</sup>
<b>500mg/kg</b>	58.01±2.04 <sup>a</sup>	36.41±3.68 <sup>b</sup>	31.65±1.91 <sup>b</sup>	126.07±5.75 <sup>bc</sup>	135.39±5.68 <sup>b</sup>

Result showing the result for lipid profile level. High Density Lipoprotein (HDL), Low Density Lipoprotein (LDL), Very low Density Lipoprotein (VLDL), Cholesterol (CHOL.) and Triglycerides or Triacylglycerols (TAGS). Treatments with superscripts a,b,c,d showed significant difference ( $p < 0.05$ ) compared with the diabetic control rat group. Values are expressed as mean  $\pm$  standard deviation (n=4). Values with the same superscript are not significantly different. ( $p > 0.05$ )

From the table, the result shows that, the negative (Diabetic) group recorded increase in LDL VLDL CHOL. and TAG'S while reducing HDL which significantly different ( $P < 0.05$ ) as compared to the normal control group. However, the 500mg/kg b.w. of the seed extract shows high level of activities seen in all the parameters; there is reduction recorded in LDL VLDL CHOL. and TAG'S while increasing HDL which is not significant ( $P > 0.05$ ) when compared to the positive (glibenclamide) control group

## Discussion

Traditional medicines all over the world encompass a wide variety of natural drugs for the treatment of symptomatology associated with chronic disorders such as diabetes mellitus. Scientists are discovering within nations' traditional medicine to find future antidiabetic agents. This study, set out to investigate the effect of *Buchholzia coriacea* flavonoid seed extract on the fasting blood sugar levels of fructose-supplemented streptozotocin-induced type II diabetes in albino rats. According to the data reported on Table 1, on the fasting blood sugar levels of the studied rats, there was a significant elevation in the sugar levels of the rats in the early days of the study in the negative control (diabetic) group. This can be attributed to insulin resistance as a result of the induction of the streptozotocin (STZ) to the animals, as well as stress and inflammatory responses triggered by diabetes as a result of the destruction of the  $\beta$ -cells of the pancreas that disrupted the normal functioning of insulin. Type 2 diabetes is caused by impaired  $\beta$ -cells function and capacity to secrete sufficient insulin, coupled with a decline in target tissue sensitivity to insulin. The 100mg/kg b.w. of the plant seed extract group showed a significant reduction ( $p < 0.05$ ) in the mean sugar level when compared with the 500mg/kg b.w. of the plant seed extract group and also as compared with the positive (glibenclamide) control group. That was strongly indicative of the dose-dependent action of the extract. According to Parthsarti *et al.*, (2013) as in this study, mean fasting blood sugar (FBS) levels were decreased significantly ( $p < 0.05$ ) when *Dolichos biflorus* was given to diabetic rats from mean  $\pm$ SD values of  $362 \pm 63.36$  to  $118 \pm 38.55$ . The reduction of sugar level seen in 100mg/kg b.w. group is an indication that the plant seed extract was able to restore its level to the baseline or that of the normal control group as the weeks progressed. That strongly indicated that the plant seed extract could be used in the treatment of diabetes based on its hypoglycemic effects, probably by enhancing insulin secretion, which in turn enhances glucose uptake by the liver, adipose tissue and muscles inhibits glucose absorption by the intestine and gluconeogenesis by the liver. This result is in agreement with that of Ezeigbo (2011), who reported that, treatment of alloxan-induced diabetic mice with the crude extracts of *B. coriacea* seed brought down the raised blood glucose levels significantly ( $P_s = 0.043$ ) in a dose-dependent manner. Also, studies by Hossam, *et al* (2019) strongly suggested that methanol extract of *Adansonia digitata* leaf could have strong antidiabetic and hypolipidemic effects in a dose-dependent manner and also improved essential hematological properties and redox parameters in the experimental diabetic rats studied.

As shown on Table 4.5., there was significant increase in LDL, VLDL, total cholesterol and triacylglycerol while decreasing HDL, the good cholesterol, in the negative diabetic control rats. Diabetes is characterized by increased serum triacylglycerols (TG), a drop in HDL, and an elevated LDL (LDL). Abnormal lipid metabolism in diabetes accelerates atherosclerosis (Balikai *et al.*, 2020; Pinakesty and Azizah, 2020). The 100 and 500mg/kg b.w. of the plant seed extract could cause significant ( $p < 0.05$ ) decreases in LDL, VLDL, total cholesterol and triacylglycerol while HDL level increased. This suggested that the extract was effective in reversing the abnormalities associated with lipid metabolism in the diabetic rats. That was in agreement with that of Ukpabi *et al.*, (2019), who reported in that rats treated with aqueous seed extracts of *Garcinia kola* and insulin lowered fasting blood glucose levels while rats treated with aqueous seed extracts of *Garcinia Kola* and atorvastatin significantly lowered TG, TC and LDL concentrations in a dose-dependent manner while significantly elevating HDL concentrations when compared to untreated

control rats. Lipid abnormalities in patients with diabetes, often termed “Dyslipidemia”, a typically characterized by high total cholesterol (TC), high triglycerides (Tgs,) low high-density lipoprotein cholesterol (HDL-C) and increased level of low-density lipoprotein particles. It is a well-known fact that dyslipidemia is an independent risk factor for cardiovascular disease. Elevated blood glucose level combined with dyslipidemia increases atherosclerosis-related inflammation and make it more extensive (Yusuf, *et. al.*, 2004).

The results from this study indicated that administration of flavonoid from seed extract of *Buchholzia coriacea* to streptozotocine-induced diabetic rats at the experimental doses and the duration of administrations showed that the extract had hypoglycemic effect as it reduced the serum fasting blood sugar level to normal as compared with positive control. Also it has hypolipidemic effects as it reduced TG, TC, LDL, VLDL and subsequently increased HDL.

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