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Treatment of Diabetic Condition in Streptozotocin Induced Diabetic Wister Rats Using Food Blends Such as Unripe Plantain, Soybean and Ginger

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Authors' contributions

This work was carried out in collaboration between all authors. Author II designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Authors SM and AJ managed the analyses of the study. Author AJ managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aims: To investigate the effect of food blends (plantain, soybean and ginger) on the blood glucose, lipid profile and haematological indices on *streptozotocin* induced diabetic rats.
Methodology: A total of 35 rats of mean body weight 219.07 g separated into7 groups (5 per group) where induced by a single intraperitoneal (I.P) injection of *streptozotocin* (0.1 g dissolved in 5 ml of freshly prepared sodium citrate buffer 0.1 M, pH 4.5) at a dose of 40 mg/kg body weight after fasting for 12 hours and fed with flours/blends. The flours were produced from plant materials for different treatments/blends (blend A=100% unripe plantain, B=80% unripe plantain, 14% soybean, 6% ginger, C=70% unripe plantain, 26% soybean, 4% ginger, D= 60% unripe plantain, 38% soybean, 2% ginger, E= 50% unripe plantain, 50% soybean) and the phytochemicals and minerals content were determined. Blood glucose was determined at 5 days interval for 25 days. Diabetes was confirmed in rats with blood glucose concentrations ≥200 mg/dl. After 25 days rats were

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anaesthetized with chloroform vapour and blood samples collected by cardiac puncture for haematology and lipid profile determination.
Results: The results showed that unripe plantain, soya beans and ginger in adequate proportion(C=70% unripe plantain, 26% soybean, 4% ginger or D= 60% unripe plantain, 38% soybean, 2% ginger) could help to reduce blood glucose, improve haematological parameters and lipid profile. Significant reduction was observed in the blood glucose level of rats fed blends C and D from 286 to 85 mg/dl and 307 to 90 mg/dl respectively at the end of experiment. These results also demonstrated that the inclusion of ginger at 6% causes rise in blood glucose level. Total cholesterol (TC) increased in all the blends. However, the lowest concentration of TC was observed in blends C and D. The highest packed cell volume (60%) and Haemoglobin (20 g/dl) level observed in rats fed blend C was significantly higher than the normal control fed conventional feeds. The increase in packed cell volume (PCV) (50%) and Hb (17 g/dl) in diabetic rats demonstrated that the formulated blend C was able to raise PCV and Hb above 50% and 17 g/dl (Normal control NC) respectively. Significant increase ($P<0.05$) in low density lipoprotein cholesterol (LDLc) was also observed in all the blends with blend C having the least (4.0 mg/dl) close to NC (2.0 mg/dl).
Conclusion: From the results it is evident that blend C will manage and improve the health status of diabetic patients.

Keywords: Diabetes mellitus; streptozotocin (STZ); haematology; lipid profile; plant materials.

1. INTRODUCTION

Diabetes mellitus has become a major global problem in our world today. It is a common disorder associated with increased morbidity and mortality and can be defined as a group of metabolic diseases characterized by chronic hyperglycemia due to defective insulin secretion, insulin action, or both, resulting in impaired carbohydrate, lipid, and protein metabolism [1].

The combat against diabetes mellitus must be made a matter of top priority by all due to the continual increase in the global prevalence of this social ill. Globally the prevalence was estimated to increase in year 2000 to 2010 from 14.2 million to 17.5 million in North America, 15.6 million to 22.5 million in South America, 26.5 million to 32.9 million in Europe, 9.4 million to 14.1 million in Africa, 84.5 million to 132.2 million in Asia and 1.0 million to 1.3 million in Australia giving a total global increase in prevalence from 151 million people in 2000 to 221 million people in 2010 [2]. This was projected to 324 million by 2025 by Zimmet et al. [3] and 366 million 2030 [4] and this is expected to rise to 592 million by 2035 [5].

Currently, approximately 425 million adults (20-79 years) were living with diabetes; by 2045 this will rise to 629 million. The proportion of people with type 2 diabetes is increasing in most countries, 79% of adults with diabetes were living in low- and middle-income countries. The greatest number of people with diabetes was between 40 and 59 years of age. 1 in 2 (212 million) people with diabetes were undiagnosed. Diabetes caused 4 million deaths. Diabetes caused at least USD 727 billion dollars in health expenditure in 2017 – 12% of total spending on adults. More than 1,106,500 children were living with type 1 diabetes. More than 21 million live births (1 in 7 births) were affected by diabetes during pregnancy. 352 million people were at risk of developing type 2 diabetes [6] (Fig. 1).

The increasing interest in herbal medicine for the treatment of diabetes and many prevailing diseases is not surprising. This may be attributed to the upsurge in cases of drug resistance, cost and several side effects associated with most orthodox medicines. The use of plant materials as spices, condiments and for medicinal purposes has therefore become more popular and as such more plants materials such as plantain and soybean that have low carbohydrate content with high mineral values are being exploited.

There is therefore no doubt that orthodox medicine itself appears to be strongly anchored on traditional medicine [7]. The fact that the tropics into which majority of Africa lies is host to about 2/3 of the world's flora and fauna means that a lot of medicinal plants can be found here for both curative and management of diseases [8].

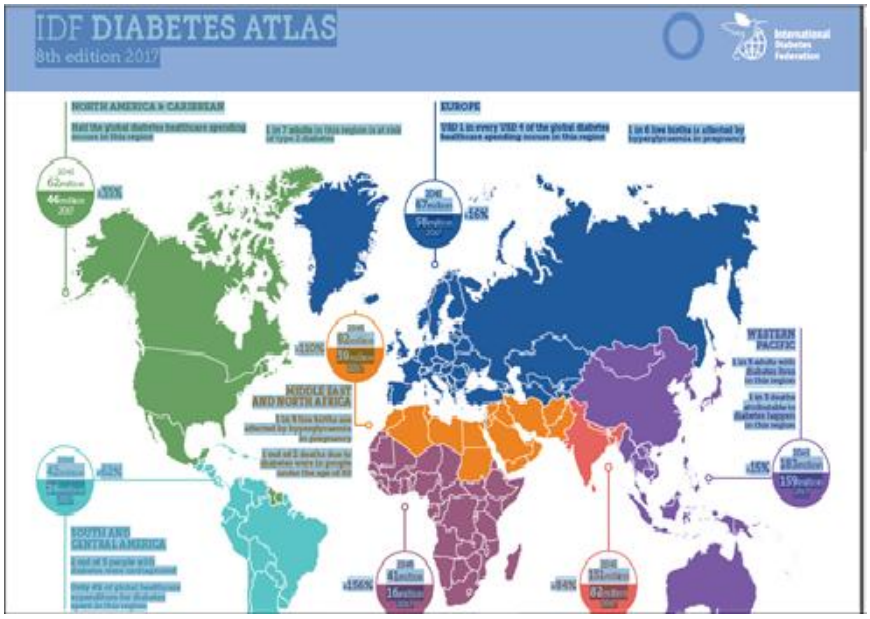

Fig. 1. International Diabetic Federation [6]

Plantain (*M. paradisiaca*) is a staple food crop in West Africa where its starchy fruits are generally cooked or fried before consumption. During unripe plantain ripening, the starch is changed to reducing sugars and sucrose. The medicinal value of plants have assumed a more important dimension in the past decades owing largely to the discovery that their extracts contain not only minerals but also a diverse array of secondary metabolites with antioxidant potentials [9,10]. These antioxidants have been implicated in the therapeutic effects of several plants and vegetables that are used in traditional medicine [11,12].Plantain contains a high fiber content, and thus is capable of lowering cholesterol and helps to relieve constipation and hence prevention of colon cancer. Besides, its high potassium content is found to be useful in the prevention of rising blood pressure and muscle cramp [13]. Various parts of the plant such as the leaves, root, fruit stalk, bract and fruit have been used for medicinal and domestic purposes.

Soybean is known as the “Golden bean” or the super legume of the twentieth century, because it contains a good proportion of oil more than 20 percent. Soybean is also categorized as oilseed, represents an excellent source of unsaturated fatty acids, high quality proteins and fiber. Soybean contains very small amount of saturated fatty acid but do not contain any Trans fatty acid. Both omega-6 and omega-3 fatty acids such as linoleic acid (56 % of total fat) and alpha lenolenic acid (7-8% of total fat) are present in soybean. Cooked Soybeans are rich in iron, phosphorous, magnesium, vitamin B2 (riboflavin) and folate. Kadam et al. [14] stated that legumes have been known as “a poor man’s meat”. They supply protein, complex carbohydrates, fiber and essential vitamins and minerals to the diet, which are low in fat and sodium and contain no cholesterol.

Spices are food adjunct commonly added to food to improve the sensory properties but many spices have been observed to exert medicinal effects. Some spices which have been reported to exert hypoglycemic effect both in laboratory animals and human subjects are: Fenugreek seeds *(Trigonella foenum),* garlic *(Allium sativum),* Onion *(Allium cepa),* turmeric *(Curcuma longa),* cumin seeds *(Curminum cyminum),* ginger *(Zingiber officinale),* mustard *(Brassica nigra),* curry leaves *(Murraya koenigi)* and coriander *(Coriandum sativum)* [15].

Ginger is a perennial plant with narrow, bright green, grass-like leaves. It is cultivated in the tropics for its edible rhizomes and has been found to be useful for both culinary and medicinal purposes [16,17]. Fresh ginger contains 80.9% moisture, 2.3% protein, 0.9% fat, 1.2% minerals, 2.4% fiber and 12.3% carbohydrates. The minerals presented in ginger are iron, calcium and phosphorous. It also contains vitamins such as thiamine, riboflavin, niacin and vitamin C. The

composition varies with the type, variety, agronomic conditions, curing methods, drying and storage conditions [18].

Several studies have reported the hypoglycemic effect of different forms of ginger in both animals and human subjects. Among the fairly recent reports are: Arablou et al. [19]; Mozaffari-Khosravi et al. [20] and Mahluji et al. [21] used ginger powder in type 2 diabetic patients; Son et al. [22] used 6-gingerol isolated from ginger in obese diabetic mice; Sukalingam et al. [23] used 6-gingerol in STZ-induced diabetic rats; Abdulrazaq et al. [24] used aqueous ginger extract STZ-induced diabetic rats; while Jafri et al. [25] used aqueous extract in alloxan-induced diabetic rats. Very limited studies have reported the hypoglycemic effect of ginger juice while there is abject scarcity of scientific findings on hypoglycemic effect of cooked ginger extract, which is highly needed since the spice is mostly consumed in cooked forms in various cuisines. Hence, the objective of this study is to determine the effect of food blends (plantain, soybean and ginger) on the blood glucose, lipid profile and haematological indices on streptozotocin induced diabetic rats.

2. MATERIALS AND METHODS

2.1 Materials

Unripe plantain and ginger roots were bought from Jattu market in Auchi, Edo State, Nigeria; defatted soy bean flour (Variety TGX 1448-2E) was purchased from Benin City in Edo State Nigeria. Streptozotocin (STZ) Sigma NO SO130 was a product of Sigma-Aldrich chemical company, UK. Every other chemical used were bought from Promise laboratory in Ekpoma, Edo State, Nigeria.

2.1.1 Processing of plantain flour

Fresh unripe plantain was peeled, sliced using slicer and dried in an oven at 60°C for 48 hours. Dried sample was ground into powder (plantain flour).

2.1.2 Processing of soybeans to defatted flour

Soybean seeds were cleaned and sorted manually to remove dirt leaves and stones. The clean soybean seeds were coarsely milled to separate the coat from the cotyledon. The dehulled seeds were milled to fine soybean flour using an attrition mill. The fine soybean flour was then defatted using cold extraction with n-hexane. The defatted flour was then air-dried and the clumps broken into fine flour, then sieved through a mesh screen.

2.1.3 Processing of ginger powder

Fresh ginger roots were sorted and washed to remove soil and other foreign materials then sliced to thin layers and dried in an oven at 60^{o}C for 24 hours before milling to powder.

2.1.4 Formulation of unripe plantain, soybeans and ginger flour blends

Five samples were prepared from the combinations of unripe plantain, defatted soybean and ginger as blends:

A=100% unripe plantain
B=80% unripe plantain, 14% soybean, 6% ginger
C=70% unripe plantain, 26% soybean, 4%ginger
D= 60% unripe plantain, 38% soybean, 2% ginger
E= 50% unripe plantain, 50% soybean

2.1.5 Mineral content Determination

Analyses of mineral content were carried out using an Atomic Absorption Spectrophotometer (Perkin Elmer Analyst 200) according to the method of AOAC [26].

2.1.6 Phytochemical determination

Flavonoid determined by the method of [27]:- 10 g of the sample was extracted repeated with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through Whatman filter paper No 42. The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

Alkaloid was determined by the alkalin precipitation gravimetric method described by [28]. Five grams (5 g) of the sample was weighed into 50 ml of 10% acetic acid solution in ethanol in a 250 ml beaker. The mixture was shaken and allowed to stand for 4 hours. The mixture was then filtered with Whaman No. 42 filter paper. The filtrate was concentrated to one quarter of its original volume by evaporation using a steam bath. Alkaloid in the extract was precipitated by

drop-wise addition of ammonium hydroxide (NH4OH) until full turbidity was obtained. The alkaloid precipitate was recovered by filtration using a weighed filtered paper and washed with 1% ammonia solution (NH4OH), dried in the oven at 80^{o}C for 1 hour. It was later cooled in a desiccator and reweighed. By weight difference, the weight of alkaloid was determined and expressed as percentage of the sample analyzed, using the formula.

$$\% \text{ Alkaloid} = \frac{W_2 - W_1}{W} \times \frac{100}{1}$$

Where:

W = weight of sample
W1= weight of empty filter paper
W2= weight of paper + alkaloid precipitate

Saponin was determined by [29]. 20 g of sample was dispersed in 200 ml of 20% ethanol. The suspension was heated over a hot water bath for 4 h with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90°C. The concentration was transferred into a 250 ml separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ethyl layer was discarded. The purification process was repeated. 60 ml of n-butane extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporations the sample were dried in the oven into a constant weight. The saponin content was calculated in percentage.

Tannin was determined using Follins Dennis spectrophotometric method according to [30]. Five gram (5 g) of the sample was dispersed in 50 ml of distil water and shaken. The mixture was allowed to stand for 30 min at room temperature and shaken every 10 min. at the end of the 30 min, the mixture was filtered through Whatman filter paper and the filtrate was used for the experiment. Two milliliters (2 ml) of the extract was measured into 50 ml volumetric flask. Similarly, 5 ml of standard tannic acid solution and 5 ml of distilled water were measured into separate flask to serve as standard and blank respectively. They were further diluted with 35 ml distilled water separately and 1 ml of Follin-Dennis reagent was added to each of the flask, followed by 2.5 mls of saturated sodium carbonate solution (NA_2CO_3). The content of each flask was then made up to 50 ml at room temperature. The absorbance of the developed colour was measured at 620 nm wavelength in spectrophotometer. Readings were taken with the reagent blank at zero.

2.1.7 Induction of diabetes in Wister rats

A total of 35 adult male albino rats with mean body weight of 219.07 g were obtained from the disease free stock of the animal house, attached to Ambrose Alli University. The rats were separated into seven groups with five rats per group including NC and DC as shown in Table 1.

Prior to experimentation, the rats were acclimatized to laboratory condition and fed with rat pellet and water adlibitum for a week. Diabetes was induced in rats by a single intraperitoneal (I.P) injection of freshly prepared solution of streptozotocin (0.1 g dissolved in 5 ml of freshly prepared sodium citrate buffer 0.1 M, pH 4.5) at a dose of 40 mg/kg body weight after fasting for 12 hours. Good hygiene was maintained by constantly cleaning and removal of faeces and spilled feeds from cages daily. Fasting blood glucose (FBG) was determined using Accuchek Active glucometer, Roche Germany, with blood obtained from the tail vein of the rats. This test was repeated on day 5, 10, 15, 20 and 25. Diabetes was confirmed in STZ treated rats with blood glucose concentrations $\geq$200 mg/dl.

2.1.8 Collection and analysis of blood

The rats were anaesthetized with chloroform vapour, twelve hours (12 h) after last day of feed administration, and blood samples were collected by cardiac puncture into a set of plain and fluoride oxalate sample bottles.

2.1.9 Hematological parameters

The packed cell volume (PCV) was measured by the micro hematocrit centrifuge. Hemoglobin (Hb) concentration was determined by the cyanomethemoglobin technique [31]. The white blood cell components were also determined.

2.1.10 Lipid Profile Studies

Blood sample was centrifuged to collect plasma which was used to estimate total cholesterol, high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C), and triglycerides (TG) using commercial kits obtained from Randox Laboratories, UK.

Table 1. Rat groups and treatments

Groups	Number of rats	Treatments
A	5	STZ-induced diabetic rats fed with100% unripe plantain),
B	5	STZ-induced diabetic rats fed with 80% unripe plantain, 14% soybean, 6% ginger),
C	5	STZ-induced diabetic rats fed with 70% unripe plantain, 26% soybean,4% ginger) and
D	5	STZ-induced diabetic rats fed with 60% unripe plantain, 38% soybean,2% ginger)
E	5	STZ-induced diabetic rats fed with 50% unripe plantain and 50% soybean),
NC	5	Not induced and fed with rat pellet
DC	5	Induced and fed with rat pellet).

2.2 Data Analysis

Data generated were subjected to analysis of Variance (ANOVA) to test significant variations ($P<0.05$) among mean values obtained. Duncan's multiple range test was applied to indicate where significant differences ($P<0.05$) occurred using Genstat statistical package 2005, 8th edition (Genstat Procedure Library Release PL16).

3. RESULTS AND DISCUSSION

3.1 Mineral Composition of Formulated Food

Table 2 depicts the composition of the studied minerals. Food blend E had the highest potassium content (1099.42 ppm), this was followed by D (944.79 ppm) while the lowest potassium content was observed in A (704.80 ppm). The highest potassium observed in food blend E could be attributed to its high inclusion of soybean (50%) which is known to be a rich source of potassium. Potassium is an important mineral in the body that regulates fluid balance, muscle contraction and nerve signals. High potassium may reduce blood pressure and water retention, protect against stroke and prevent osteoporosis and kidney stones.

Food blend A had the highest sodium content (75.65 ppm), this was followed by B (67.19 ppm) while the lowest content (47.80 ppm) was observed in E. sodium is essential for life. It helps to control the body's fluid balance. It send nerve impulses and affects muscle function.

Food blend E had the highest calcium content (804.02 ppm), this was followed by D (626.91 ppm) and C (435.71 ppm) while the lowest calcium content (236.16 ppm) was observed in food blends A. calcium plays an important role in muscle contraction, transmitting messages through the nerves and the release of hormones. Calcium is also important mineral in the formation of teeth and bones.

Food blend E had the highest iron content (141.49 ppm), this was followed by D (121.42 ppm) and C (114.64 ppm) while the lowest content (28.60 ppm) was observed in food blends A. Iron is an important component of haemoglobin, the substance in red blood cell, responsible for carrying oxygen and transports it throughout the body.

The mineral content (potassium, calcium and iron except sodium) of the blends, increased with increasing soybean inclusion level (Table 2), depicting that soybean is rich in these minerals.

3.2 Phytochemical Properties

Table 3 shows the phytochemical compositions of the blends. The lowest tannin content (tannin 0.27 mg/100 g) was observed in food blend A and was followed by blend B (0.55 mg/100 g). Blends C, D and E had the same tannin content (0.61 mg/100 g).

The highest alkaloid content (6.43%) was observed in blend A and was followed by B (6.23%), C (5.99%), D (5.75%), and E (4.84%) in that decreasing order.

Blends B and C had the same flavonoid content (0.42 mg/100 g) which was higher than the other blends. The lowest flavonoid content (0.11 mg/100 g) was observed in blend A; this was followed by E (0.31 mg/100 g) and D (0.35 mg/100 g).

Blend A (0.16 mg/100g) had the lowest saponin content and was followed by B (2.39 mg/100 g),

C (3.99 mg/100 g), D (4.22 mg/100 g), and E (6.33 mg/100 g) in that increasing order.

Saponins are known to possess both beneficial (cholesterol lowering) and deleterious (cytotoxic permeabilization of the intestine and paralysis of the sensory system) properties [32]. Flavonoids, alkaloids and tannins are polyphenolic compounds with antioxidant properties. In addition, phenolic compounds existing in plants are also responsible for their contribution to colour, sensory and antioxidant properties of food [33].

The low phytochemical values (Table 3) recorded in this study are significantly lower than ($P<0.05$) the results of Eleazu et al. [34] who recorded significant values (saponin 1.827, flavonoid 0.981 and tannin 1.577) in unripe plantain flour. However, he further reported that the levels of saponin in the flour are quite too low to cause any deleterious effects.

Food and nutrients play vital role in the normal functioning of the body. In this study, plant materials such as unripe plantain, soybean and ginger were used to formulate food blends with the aim of studying its effect on the haematological parameters, lipid profile and blood glucose level of diabetic rats.

The analysis of variance showed significant difference ($P<0.05$) in the packed cell volume (PCV) and haemoglobin (Hb) level of the diabetic rats (Fig. 2). The highest PCV and Hb level (60%, 20 g/dl respectively) were observed in rat fed blend C that contains 70% unripe plantain, 26% soybean and 4% ginger. This was significantly higher ($P<0.05$) than the normal control (not induced) (50%, 17 g/dl) fed conventional feeds. The increase in PCV and Hb in diabetic rats showed that the formulated blends were able to raise the PCV and Hb above 50% and 17 g/dl.

The degree of anemia in diabetic patients can be associated with a number of factors such as glomerular filtration rate and glycated h (HbAlc) level. Thomas et al. [35] reported that anemia is due to diminished erythropoietin production by failing kidneys and increased non enzymatic glycosylation of red blood cell (RBC) membrane protein. In this study, increase in PCV and Hb level of some of the diabetic rats does not depict occurrence of anemia rather shows its potency in the management of the ailment (diabetes). This could be attributed to the phytochemicals and mineral present in the blends. The antioxidant properties of these phytochemicals especially flavonoids have been reported in several studies. Onat et al. [36] reported the anti-sickling properties. This according to Palacious et al. [37] it prevents oxidation of RBC and Hb that often lead to haemolysis. According to Egunyomi et al. [38] it may also stimulate formation or secretion of erythroprotein in the stem cells of the animals as evidenced by the increased level of PCV and Hb. There is no significant difference ($P < 0.05$) in the lymphocytes of the formulated blends (A and D) from the normal control. The diabetic control rat had lymphocytes (72%) significantly higher ($P<0.05$) than every other rat. The high lymphocytes level could be attributed to unknown infection. The values of Neutrophiles, Ecsinophiles, Basophiles and Monocytes obtained in rats fed with blends C, D and E were significantly lower ($P<0.05$) than the normal control rats.

3.3 Changes in Blood Glucose and Body Weight of Streptozotocin induced Wister Rats

Blood glucose (Table 4) and body weight (Fig. 3) were monitored for total duration of 25 days. At 5 days interval blood glucose level and body weight were determined. The initial measurements were taken before induction at day 0 for glucose level and body weight. The various rat groups had blood glucose level between 93-120 mg/dl and body weight between 205-270 g (day 0). They were induced and fed formulated food blends and water *adlibitum*.

On the 5^{th} day, all the induced groups had significant increase ($P<0.05$) in glucose level>200 mg/dl (Table 4) with corresponding decrease in body weight (Fig. 2). Thus the rats were considered diabetic at $\geq$200 mg/dl. The rat fed blend D had the highest blood glucose level 307 mg/dl. NC rats had the lowest blood glucose level (110.0 mg/dl) and showed no significant ($P>0.05$) change throughout the period of experiment.

There was steady significant increase ($P<0.05$) in the blood glucose of group A, B, E and DC throughout the period of this experiment. The results showed that at 0% and 6% inclusion of ginger in blends A and B respectively, the rat were hyperglycemic. This demonstrated that the inclusion of ginger at 6% causes rise in BGL. Significant reduction ($P<0.05$) was observed in the blood glucose level

of rats fed with blends C and D from 286 to 73 mg/dl and 307 to 99 mg/dl from day 5 to 10 respectively. This same trend was observed in blood glucose level for 15, 20 and 25 days with rat fed blends C (99, 101 and 85 mg/dl) and D (114, 103 and 90 mg/dl) respectively, having normal blood glucose <200 mg/dl. This shows the potency of the blends C (70% unripe plantain, 26% Soybean and 4% ginger) and D (60% unripe plantain, 38% soybean and 2% ginger) in the management of the ailment (diabetes). This could be attributed to the combination levels of the plant materials particularly the inclusion of ginger at 4% and 2% in blend C and D respectively. Ginger provides an amount of potassium that could help stroke and diabetes and adult requires 2000 mg of potassium each day. Potassium is important for diabetic patients and those at the risk of it. The findings of recent study published by researchers from university of Sydney in 2012 revealed that ginger extract helps to increase cell absorption of glucose even independent of insulin (www.naturlnews.com). The predominant pungent compound in ginger is responsible for its benefit to humans [39]. According to Andallu et al. [40] ginger has a therapeutic benefit of lowering fasting serum blood glucose level in Type 2 diabetes. According to Singh et al. [41]. many of the putative activities of ginger (antioxidant, anti-inflammatory, hepatoprotective, anti-obesity) are often associated with the etiology and pathophysiology of type 2 diabetes, which suggest the possibility that ginger may not have a direct effect on diabetes but acts indirectly by suppressing factors that lead to impaired glucose control. Thus, was supported by a study showing that ginger root powder (200 mg/kg body weight) in type 2 diabetic rat model reversed symptoms of metabolic syndrome, blood glucose, blood lipid and decreased oxidative stress [42]. Although blend B had ginger inclusion at 6%, the glucose level was >200 mg/dl throughout the period of this experiment. This shows that ginger inclusion at 6% could result in hyperglycemic condition. However, at day 10, a rise was observed in the glucose level of rats fed blend A (272-334 mg/dl), B (245-301 mg/dl) and E (247-370 mg/dl) with corresponding decrease in body weight (Figure 3). Thus this indicates that the formulation for A, B and E could not control the diabetic condition. The DC rats fed with conventional rat feed increased in blood glucose level and body weight steadily throughout the period of experiment, while the body weight decreased from 228 to 220g and increased from 235 to 241 g in rats fed blends C and D respectively.

Table 2. Mineral composition of formulated food blends

	Minerals (ppm)			
Blends	**Potassium**	**Sodium**	**Calcium**	**Iron**
A	704.80^{e}	75.65^{a}	236.16^{e}	28.60^{e}
B	931.82^{d}	67.19^{b}	430.77^{d}	92.89^{d}
C	942.17^{c}	66.00^{b}	435.71^{c}	114.64^{c}
D	944.79^{b}	62.08^{c}	626.91^{b}	121.42^{b}
E	1099.42^{a}	47.80^{d}	804.02^{a}	141.49^{a}
SEM	0.05	0.54	0.06	0.05

Means with the same letters down the column are not significantly different (P>0.05)
A=100% unripe plantain; B=80% unripe plantain, 14% soybean, 6% ginger; C=70% unripe plantain, 26% soybean, 4%ginger; D= 60% unripe plantain, 38% soybean, 2% ginger ; E= 50% unripe plantain, 50% soybean; SEM= Standard error of mean

Table 3. Phytochemical properties of formulated food blends

	Phytochemicals			
Blends	**Tannin (mg/100g)**	**Alkaloid (%)**	**Flavonoids (mg/100g)**	**Saponin (mg/100g)**
A	0.27^{c}	6.43^{a}	0.11^{d}	0.61^{d}
B	0.55^{b}	6.23^{b}	0,42^{a}	2.39^{c}
C	0.61^{a}	5.99^{c}	0.42^{a}	3.99^{b}
D	0.61^{a}	5.75^{d}	0.35^{b}	4.22^{b}
E	0.61^{a}	4.84^{e}	0.31^{c}	6.33^{a}
SEM	0.008	0.014	0.005	0.008

Means with the same letters down the column are not significantly different (P>0.05)
A=100% unripe plantain; B=80% unripe plantain, 14% soybean, 6% ginger; C=70% unripe plantain, 26% soybean, 4%ginger; D= 60% unrip e plantain, 38% soybean, 2% ginger; E= 50% unripe plantain, 50% soybean; SEM= Standard error of mean

Table 4. Blood glucose of streptozotocin rats

	Days of induction						
Blends	**0**	**5**	**10**	**15**	**20**	**25**	**SEM**
A	104.40^{qrs}	272.00^{ij}	334.00^{f}	504.00^{b}	409.00^{c}	413.20^{c}	
B	93.00^{rst}	245.00^{k}	301.00^{h}	562.00^{a}	559.00^{a}	506.00^{b}	
C	107.00^{pqr}	286.00^{h}	73.00^{u}	99.00^{rst}	101.00^{qrst}	85.00^{tu}	4.15
D	103.00^{qrs}	307.00^{g}	99.00^{rst}	114.00^{p}	103.00^{qrst}	90.00^{stu}	
E	109.00^{pqr}	247.00^{j}	370.00^{e}	392.00^{d}	375.00^{e}	402.00^{cd}	
NC	108.00^{pqr}	110.00^{pq}	133.00^{n}	109.00^{pqr}	106.00^{pqr}	103.00^{qrst}	
DC	120.00^{i}	229.0^{j}	184.00^{m}	214.00^{lm}	225.00^{l}	283.00^{i}	

Means with same superscript down the column and along the row are not significantly different (P>0.05)
A=100% unripe plantain; B=80% unripe plantain, 14% soybean, 6% ginger; C=70% unripe plantain, 26% soybean, 4%ginger; D= 60% unripe plantain, 38% soybean, 2% ginger; E= 50% unripe plantain, 50% soybean; NC= Normal control, fed conventional feed (not induced); DC= Diabetic control, fed conventional fed (induced)

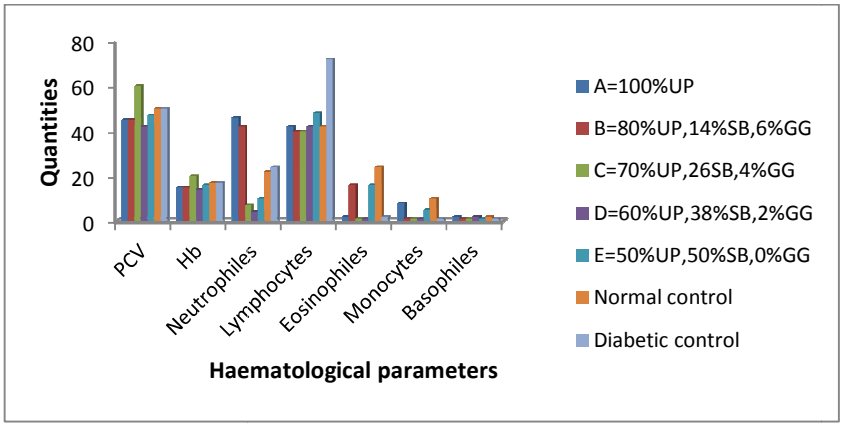

Fig. 2. Haematological quantities

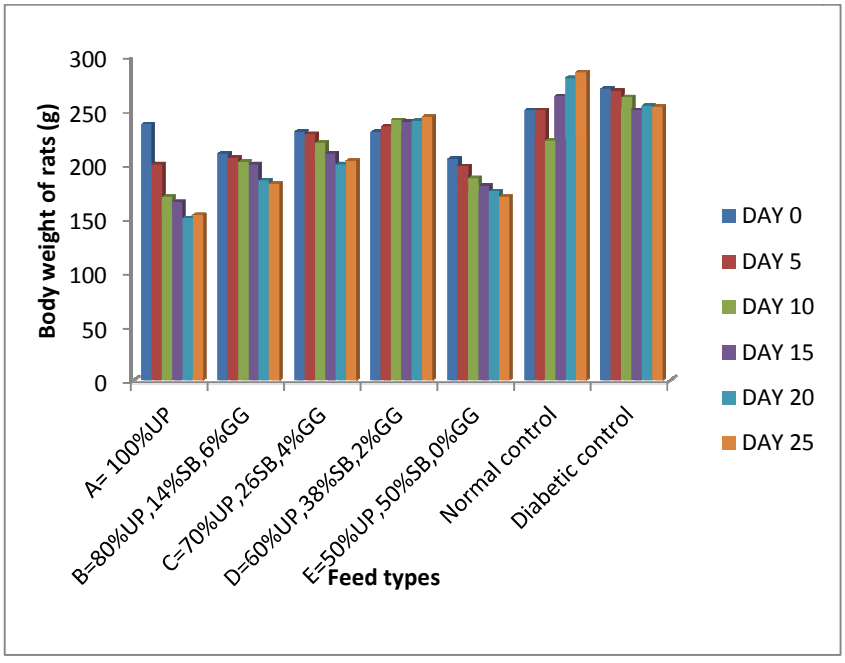

Fig. 3. Body weight of rats fed formulated and commercial feeds (g)

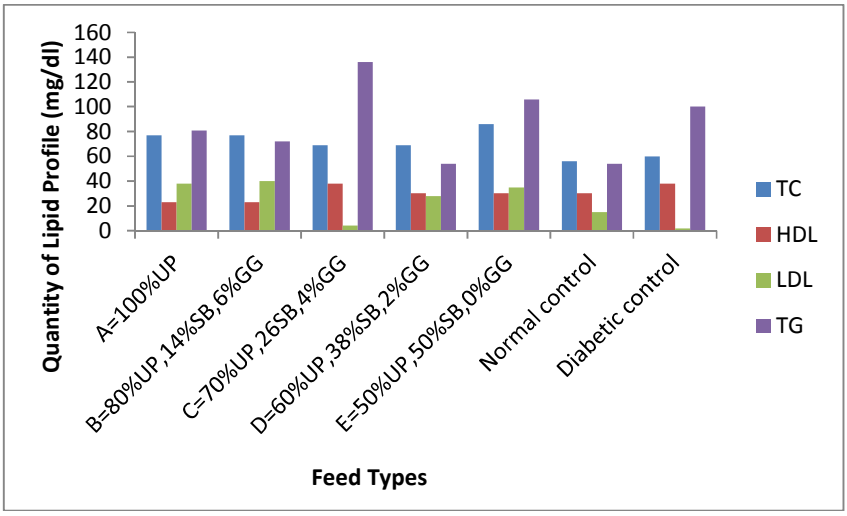

Fig. 4. Lipid profile of streptozotocin induced wister rats

3.4 Effect of Food Blends (Diet) on Serum Lipid Profile

Serum lipid concentration of streptozotocin induced rats fed with food blends and conventional feed in this study is shown in Fig. 3. From the results, serum high density lipoprotein cholesterol (HDLc) concentration in rats fed blends C (38 mg/dl) and NC (38mg/dl)were same but higher and significantly different ($P<0.05$) from HDLc of rats fed with other blends.

Total cholesterol (TC) increased in all the blends. However, the lowest concentration in TC was observed in blends C and D (Fig. 3). Thus this depicts that blends C (69 mg/dl) and D (69 mg/dl) are better having lower cholesterol concentration.

For low density lipoprotein cholesterol (LDLc) significant increase ($P<0.05$) was observed in all the blends. However, blend C (4.0 mg/dl) was next to NC (2.0 mg/dl) while the highest was observed in blend B (40.0 mg/dl).

The increased in LDLc, TC, and decreased in HDLc agrees with the findings of Adaramoye et al. [43] for diabetic rats. Besides, the formulated diets are plant materials containing phytochemicals. Ezekwe and Obidoa, [44] reported that action of plant extract in reducing plasma cholesterol concentration could be due to the ability of one or more of the phytochemicals in the plant to activate the functioning enzymes of the rats responsible for cholesterol absorption.

4. CONCLUSION

In this research work, it was observed that the blends of unripe plantain, soya beans and ginger in adequate proportion(C=70% unripe plantain, 26% soybean, 4% ginger or D= 60% unripe plantain, 38% soybean, 2% ginger) could help to reduce blood glucose, improve haematological parameters and lipid profile. The mineral content (potassium, calcium and iron except sodium) of the blends increased with increasing soybean inclusion level, depicting that soybean is rich in these minerals. Significant reduction ($P<0.05$) was observed in the blood glucose level of rats fed blends C and D from 286 to 85mg/dl and 307 to 90 mg/dl respectively. The lowest concentration of TC was observed in blends C and D. This depicts that blends C (69 mg/dl) and D (69 mg/dl) are better and preferred to the other blends. In addition, blend C also had the least value (4.0 mg/dl) of low density lipoprotein cholesterol (LDLc).Hence, blend C is most preferred to prevent and control diabetes as well as improve the health status of diabetic patients.

ETHICAL APPROVAL

As per international standard or university standard ethical approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Akah JA, Lemji JA, Salawa OA, Okoye TC, Offiah NV. Effects of *Vernonia amygdalina* on biochemical and haematological parameters in diabetic rats. Asian Journal of Medicinal Science. 2009;1(3):108-113.

2. Amos A, McCarty D, Zimmet P. The rising global burden of diabetes and its complications: Estimates and projections to the year 2010. Diabetic Med. 1997;14: 1-85.
3. Zimmet P, Shaw J, Albert KG. Preventing type 2 diabetes and the dysmetabolic syndrome in the real world: A realistic review. Diabet. Med. 2003;20: 693-702.
4. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: Estimates for the year 2000 and projections for 2030. Diabetes Care. 2004; 27(5):1047-1053.
5. Guariguata L, Whiting DR, Humbleton I, Beagley J, Linnenkamp U, Shaw JE. Global estimates of diabetes prevalence for 2013 and projections for 2035. Diabetes Research and Clinical Practice. 2014;103(2):137-149.
6. International Diabetic Federation (IDF). The IDF Diabetes Atlas. 8th Edition; 2017.
7. Nweze EI. Justification for the use of *Ocimum gratissimum* in herbal medicine and itsalteration with disc antibiotics; 2009. Available:www.biomedicine.com
8. Sofowora A. Medicinal plants and traditional medicine in Africa. John Wiley and Sons Ltd. 1993;33-34.
9. Akinmoladun AC, Ibukun EO, Afor E, Akirinlola BL, Onibon TR, Akinboboye AO, Obuotor EM, Farombi EO. Chemical constituents and antioxidant activity of Alstoniaboonei. Afr. J. Biotechnol. 2007; 6(10):1197-1201.
10. Ahenkora K. Kyei MA. Marfo EK. Banful B. Nutritional composition of false horn Apantuba plantain during ripening and processing. J. Food Chem.1998;455- 458.
11. Kumar RS, Sivakuma T, Sunderem RS, Gupta M, Murujesh K, Rajeshwa Y, Kumar MS, Kumar KA. Antioxidant and anti-microbial activities of *Bauhima recemosa* L Stem Bark. J. Med. Biol. Res. 2005;38: 1015 -1024.
12. Marthur NK, Marthur V. Antioxidants: Natural ingredients and additives for food. Beverage Food World. 2001;5:13-15.
13. Ng SP, Fong CS. Banana enhances your anti-cancer power. In: Health discovery. Petaling Jaya Malalysia: Life Publisher Berhad. of cocoyam (*Colocassia esculenta*) leafwith some green leafy vegetables. Nig J. Nutr Sci. 2000;2(1):22-26.
14. Kadam ML, Salve RV, Mehrajfatema ZM, More SG. Development and evaluation of composite flour for messi roti/ chapatti. Journal of Food Process Technology. 2012;3(1):01-07.
15. Srinivasan K. Plant foods in the management of diabetes mellitus: Spices as beneficialanti diabetic food adjunct. International Journal of Food Science and Nutrition. 2005;56(6):399-414.
16. Grant KI, Ginger Am. J. Health Sys. Pharm. 2000;57:945-957.
17. Ursell A. The complete guide to healing foods. Dorling Kindersley Ltd, London. 2000;112-114.
18. Suekawa M, Ishige A, Yuansa K, Sudo K, Aburada M, Hosoya E. Pharmacological studies on ginger- Pharmacological actions of pungent constituents of 6-gingerol and 6-shogaol. J. Pharmacoblodyn. 1984;7: 836-848.
19. Arablou T, Aryaeian N, Valizadeh M, Shariffi F, Hosseini A, Djalali M. The effect of ginger consumption on glycemic status, lipid profile and some inflammatory markersin patients with type 2 diabetes mellitus. International Journal of Food Sciences and Nutrition. 2014;65(4):515-520.
20. Mozaffari-Khosravi H, Talaei B, Jalali BA, Najarzadeh A, Mozayan MR. The effect of ginger powder supplementation on insulin resistance and glycemic indices inpatients with type 2 diabetes: A randomized double-blind placebo-controlled trial. Complementary Therapies in Medicine. 2014;22(1):9-16.
21. Mahluji S, Attari VE, Mabassori M, Payahoo L, Ostadrahimi A, Golzari SEJ. Effect of ginger (*Zingiber officinale*) on plasma glucose level, HbA1c and Insulin sensitivity in type 2 diabetic patients. International Journal of Food Sciences and Nutrition. 2013;64(6):682-686.
22. Son MJ, Miura Y, Kazum Y. Mechanism of anti diabetic effect of gingerol in cultured cells and obese diabetic model mice. Cytotechnology; 2014.
23. Sukalingam K, Ganesan K, Gani SB. Hypoglycemic effect of 6-gingerol, an active principle of ginger in streptozotocin-induced diabetic rats. Journal of Pharmacology and Toxicological Studies. 2013;1(2):23-30.
24. Abdulrazaq NB, Cho MM, Win NN, Zaman R, Rahman MT. Beneficial effects of ginger (*Zingiber officinale*) on carbohydrate

metabolism in streptozotocin-induced diabetic rats. British Journal of Nutrition. 2012;108(7):1194-1201.
25. Jafri SA, Abass S, Qasim M. Hypoglycemic effect of Ginger (*Zingiber officinale*) inalloxan-induced diabetic rats (*Rattus norvagicus*). Pakistan Veterinary Journal. 2011; 31(2):160-162.
26. Association of Official Analytical Chemist-(AOAC). Official methods of analysis of the association of official analytical chemist. 16th Edition, Washington; 2000.
27. Boham BA, Kocipal-Abyazan R. Flavonoids and condensed tannin from leaves of Hawaiian *Vaccinium vaticulatum* and *V. calycinium*. Pacific Science. 1994; 48:458-463.
28. Harbone JB. Phytochemical methods; a guide to modern techniques of plant analysis. Chaman and Hall Ltd., London, UK. 1973;20-28.
29. Nahapetian A, Bassiri A. Changes in concentration and interrelationship of phytate, P, Mg. Cu, Zn in wheat during maturation. Journal of Agricultural and Food Chemistry. 1974;32: 1179-1182.
30. Pearson D. Chemical analysis of food. 7th Edition, Church-hill Livingston, Edinburgh, UK. 1976;10-15.
31. Dacie JV, Lewis SM. Practical Haematology. 8th Ed., Longman Group Ltd., Hong Kong. 1994;49.
32. Price KR Johnson IT, Fenwic CR. The chemical and biological significance of saponins in food and feeding stuff. Unpublished Crit. Rev. Food Sci. Nutr. 1987;26:27-135.
33. Robbins R J. Phenolic acids in foods. An overview of analytical methodology. J. Agric. Food Chem. 2003;51:2886-2887.
34. Eleazu CO, Okafor PN, Amajor J, Awa E, Ikpeama AI, Eleazu KC. Chemical composition, antioxidant activity, functional properties and inhibitory action of unripe plantain (*M. Paradisiacae*) flour. African Journal of Biotechnology. 2011;10(74): 16948-16952.
35. Thomas MC, Maclsaac RJ, Tsalamandris C. Unrecognized anemia in patients with diabetes: A cross sectional survey. Diabetes care. 2003;26:1164-1169.
36. Onat A, Can G, Kaya H, Hergene G. Atherogenic index of plasma (log10 triglycerides/high density lipoprotein cholesterol) predict high blood pressure, diabetes and vascular event. Journal Clin. Lipid. 2010;4:89-98.
37. Palacios I, Lozano M, Moro C. Antioxidant properties of phenolic compounds occurring in edible mushrooms. Food chem. 2011;128:674-678
38. Egunyomi A, Moody JO, Eletu OM. Antisickling activities of two ethnomedicinal plant recipes used for the management of sickle cell anaemia in Ibadan, Nigeria. Afri. J. Biotechnol. 2009;8:20-25
39. Choudhari SS, Kareppa BM. Identification of bioactive compounds of *Zingiber officinale* roscoe rhizomes through gas chromatography and mass spectrometry. Int J Pharm Res Dev. 2013;5:16-20.
40. Andallu B, Radhika B, Suryakantham V. Effect of aswagandha, ginger andmulberry on hyperglycemia and hyperlipidemia. Plant Foods Hum Nutr. 2013;58:1-7
41. Singh AB, Akanksha SN, Maurya R, Srivastava AK. Anti-hyperglycemic, lipidlowering, and anti-oxidant properties of [6]-gingerol in db/db mice. Int J Med Med. Sci. 2009;1:536-44.
42. Madkor HR, Mansour SW, Ramdan G. Modulatory effect of garlic, ginger, turmeric and their mixture on hyperglycemia, dyslipidemia and oxidative stress in streptozotocin-nicotinamide diabetic rats. Br. J. Nutr. 2010;1:105-107.
43. Adaramoye OA, Nanneri VO, Anyaanwu KC. Possible anti atherogenetic effect of Kolaviron (a *Garcinia kola* seed extract) in hypercholesterolemia rats. Clin. Exp. Pharmacol. Physiol. 2005;32(1-2):40-46.
44. Ezekwe CI, Obidoa O. Biochemical effect of *Vernonia amygdalina* on rat liver microsomes. Niger. J. Biochem. Mol. Biol. 2001;16:1745-1798.

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