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Biochemical basis of the use of cocoyam (Colocassia esculenta L.) in the dietary management of diabetes and its complications in streptozotocin induced diabetes in rats

Chinedum Ogbonnaya Eleazu^{1*}, Polycarp Nnaecheta Okafor², Ijeh Ifeoma²

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ABSTRACT

Objective: To investigate the biochemical basis of the anti-diabetic action of cocoyam [Colocassia esculenta (C. esculenta)] in streptozotocin diabetic rats.

Methods: Blood glucose of the rats was measured with a glucometer. The relative heart weight, relative pancreatic weight, serum proteins, urea, creatinine, albumin, amylase, lipase, serum and hepatic aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP) and chemical analysis of the test feed were determined using standard techniques.

Results: The diabetic rats fed with cocoyam had significant elevation (P<0.05) of relative pancreatic weight, hepatic AST, ALT, ALP and serum proteins and albumin, but had significant reduction (P<0.05) of blood glucose, serum urea, creatinine, amylase, lipase, AST, ALT and ALP compared with the diabetic control rats while these parameters were significantly altered (P<0.05) in the diabetic control rats compared with the non-diabetic rats. There were no significant differences (P>0.05) in the relative heart weights of all the rats in the three groups. The test feed contained considerable amounts of phenolics, crude fiber, Ca, Fe, K, Na but low amounts of oxalate and phytate.

Conclusions: *C. esculenta* may exert its anti-diabetic action by delaying/regulating the rate at which dietary starch is hydrolyzed to glucose or possibly through inhibition of acute pancreatitis. Finally, the study also shows the potentials of *C. esculenta* in the dietary management of obesity.

1. Introduction

Diabetes mellitus is a global health problem that is ravishing humanity with its prevalence being at an alarming increase^[1]. It was reported that approximately 4% of the global population suffers from this disease, a percentage which is expected to reach 5.4% in 2025, thereby making this disease endemic globally^[1].

Several therapeutic strategies that are currently being used to treat this metabolic disorder have been met with several limitations thereby leading to increased search for alternative therapies. This trend has been further intensified by increasing doubts surrounding current dietary and other lifestyle behaviors together with growing interests in functional foods and nutraceuticals[1].

In addition, herbal remedies have proven to be more accessible and affordable than conventional drugs as the former represents the first line of treatment available for the world's teeming population^[2].

Cocoyam [Colocasia esculenta L. (C. esculenta)] is an herbaceous perennial plant belonging to the Araceae family. In most African countries, it is mainly cultivated by small—scale farmers[3]. Like many plants of the Araceae family, cocoyam grows from the fleshy corm (tuber) that can be boiled, baked or mashed into a meal and used as staple food or snack.

The corms supply easily digestible starch and are known to contain substantial amounts of protein, vitamin C, thiamine, riboflavin, niacin and significant amounts of dietary fiber^[4]. In ethnomedicine, cocoyam is used in the management of diabetes mellitus, treatment of ringworm^[5], cough, sore throat and wounds^[6,7].

¹National Root Crops Research Institute, Umudike, Nigeria

²Michael Okpara University of Agriculture, Umudike, Nigeria

 $[\]hbox{``Corresponding author: Chinedum Ogbonnaya Eleazu, National Root Crops Research Institute, Umudike, Nigeria.}$

Tel: +2348034164686

E-mail: eleazon@yahoo.com

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Previous studies have shown the hypoglycemic activity of the roots and leaves in diabetic rat models[8–10].

Although cocoyam has been scientifically documented as an anti-diabetic plant, the biochemical basis of its anti-diabetic action has not been fully investigated. The need for this investigation was further buttressed by the conflicting report given by Alegbejo *et al.* on the potential high glycemic index of this crop[11].

Considering the increasing concern over the alarming rate of diabetes mellitus and in light of the promising potentials that cocoyam might have with regards to amelioration of diabetes and its complications, this study was commenced upon to gain insights into the biological activities of cocoyam towards the pancreatic amylase, pancreatic lipase, liver, kidney function parameters, relative pancreas and relative heart weights of streptozotocin (STZ) diabetic rats which was hoped to serve as a vehicle towards understanding the biochemical basis of the anti-diabetic action of cocoyam.

2. Materials and methods

2.1. Plant materials

Fresh samples of *C. esculenta* variety locally known in the eastern parts of Nigeria as Edeofe were obtained at harvest from the National Root Crops Research Institute, Umudike, Nigeria. They were identified by Dr. Chukwu, Coordinator of Cocoyam Programme, National Root Crops Research Institute, Umudike as well as by a Taxonomist in Michael Okpara University of Agriculture, Umudike, Nigeria and deposited in their herbarium for authentication.

2.2. Chemicals

The STZ and potassium chloride were products of Sigma and Aldrich Chemical Company, United Kingdom. The creatinine, bilurubin, total protein, albumin and amylase assay kits used were purchased from Biosystems, Barcelona, Spain.

2.3. Processing of the plant material

The samples were soaked in water for 10 min, washed, peeled, rewashed and oven dried at 70 °C for 48 h until constant weight was obtained and processed to flour. The processed flour was pelletized, oven dried for 24 h at 90 °C before feeding to the rats.

2.4. Animal experiments

2.4.1. Selection of animals

Forty eight male albino rats of the Wistar strain (150–218 g) obtained from the animal house of the Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria, were used for the study. The animals were kept in metabolic cages in the animal house of the Department of

Biochemistry, Michael Okpara University of Agriculture, Umudike, Nigeria. They were acclimatized for 2 weeks to their diets prior to the commencement of the experiment and were maintained under a constant 12–h light and dark cycle and at room temperature. All animal experiments were approved by the Ethical Committee of Michael Okpara University of Agriculture, Umudike which was in line with the Guide for the Care of Use of Laboratory Animals as reported by the National Academy Press^[12].

2.4.2. Induction of diabetes

After 2 weeks of acclimatization, freshly prepared solution of STZ (0.1 g dissolved in 5 mL of freshly prepared sodium citrate buffer 0.1 mol/L, pH 4.5) was injected intraperitoneally to 42 of the rats (65 mg/kg body weight) at fasting state while six of the remaining rats served as the non-diabetic group and received standard rat pellets. Blood was collected from the tail vein and blood glucose concentration was analyzed in the STZ treated rats prior to the commencement of the dietary feeding using a blood glucose meter (Double G glucometer, USA) and subsequently, twice in a week, throughout the duration of the experiment. The STZ-treated rats with fasting blood glucose levels >200 mg/dL after 12 days of induction of STZ were considered to be diabetic and were used for the study.

2.4.3. Experimental procedure

The STZ induced rats with stable diabetic condition were then divided into two subgroups (Groups 2 and 3) comprising of six animals per group while the non-diabetic group formed the first group as follows:

Group 1. Normal rats given standard rat pellets (non-diabetic control)

Group 2. Diabetic rats fed standard pellets (diabetic control)

Group 3. Diabetic rats fed cocoyam pellets (810 g/kg)

Their diets and water were both given *ad libitum* for 28 d, after which the rats were stunned by blow, sacrificed and blood was collected intravenously from their heart using 10 mL syringes. The heart and pancreas were collected and weighed. The body weights of the rats were recorded on a daily basis, using an electronic weighing balance (Model Scout Pro, Ohaus Corporation, USA) and the percentage change in weight was calculated as:

Percentage change in weight =
$$\frac{\text{Final weight-Initial weight}}{\text{Final weight}} \times 100$$

Similarly, the percentage change in fasting blood glucose was calculated as:

Percentage change in fasting blood glucose (FBG) =
$$\frac{\text{Final FBG - Initial FBG}}{\text{Final FBG}} \times 100$$

The relative heart weight was expressed in g/100g and was calculated as:

Relative heart weight
$$(g/100 \text{ g}) = \frac{\text{Total heart weight}}{\text{Final body weight}} \times 100$$

The relative pancreatic weight was expressed in g/100g and

was calculated as:

Relative pancreatic weight (g/100 g) = $\frac{\text{Total pancreatic weight}}{\text{Final body weight}} \times 100$

2.5. Determination of marker enzyme activities in the liver

The liver was quickly washed with ice—cold physiological saline and stored at -20 °C until analyzed. Ten percent homogenate (w/v) of the liver was prepared in 150 mmol/L KCl using a homogenizer at 4 °C and centrifuged for 15 min at 4 °C[13], and the supernatant was analyzed for aspartate transaminase (AST), alanine transaminase (ALT) and alkaline phosphatase (ALP) using the Randox kit method.

2.6. Determination of liver and kidney function parameters in the serum

The serum AST and ALT levels of the rats were determined spectrophotometrically at 540 nm using the Biosystems diagnostic kits as described by Gella *et al*[14]. The total and direct (conjugated) bilurubin levels of the rats were determined using the Biosysytems diagnostic kits and the principle was based on the reaction of the serum bilurubin with diazo reagent to form a coloured complex which was measured at 540 nm with a spectrophotometer[15]. The serum creatinines of the rats were determined using the Biosystems kit and the principle was based on the reaction of the creatinine in the sample with picrate in alkaline medium to form a colored complex which was measured spectrophotometrically at 500 nm. The serum amylase, lipase, total proteins, albumin and urea levels of the rats were also determined using Biosystems diagnostic kits using the methods described by previous researchers[15–17].

2.7. Preparation of plant extract for total phenolic assays

About 6 g of the flour of the cocoyam sample was soaked in 60 mL of water and left overnight. The mixture was filtered (Whatman No. 1 filter paper) and centrifuged at $3000 \times g$ for 10 min for the assay of the phenolic contents of the flour.

2.8. Chemical analysis of the flour

The total phenolic content of the flour was determined using the Folin–Ciocalteu method as described by Singleton *et al*^[18]. Gallic acid was used as the standard and results were expressed in mg GAE/100 g. The crude fiber, oxalate and phytate contents of the test plant were determined using the methods of Association of Official Analytical Chemists^[19]. The atomic absorption spectrophotometer (Analyst 200, Perkin Elmer, Waltham, MA, USA) was used to analyze the iron (Fe) and calcium (Ca) contents of the flour while a flame photometer was used to analyzed the sodium (Na) and potassium (K) contents of the flour.

2.9. Statistical analysis

Data was subjected to analysis using the SPSS version

15.0. Results were presented as the mean \pm SD of triplicate experiments. One—way analysis of variance (ANOVA) was used for comparison of the means. Differences between means were considered to be significant at P<0.05 using the Duncan's new multiple range test.

3. Results

Administration of STZ at a dosage of 65 mg/kg body weight to the rats of Groups 2 and 3 produced stable diabetic condition within 12 d in most of the experimental rats. Intake of cocoyam by the diabetic rats of Group 3 resulted in 233.42% decreases in hyperglycemia compared with the diabetic control rats (Table 1).

Table 1
Fasting blood glucose of rats (mg/dL).

Groups	Week 1	Week 3	Week 4	Change (%)
Group 1	83.75±4.98 ^a	72.67±5.57 ^a	85.50±12.39 ^a	2.05 (i/c)
Group 2	241.00±69.24°	293.67±41.73°	312.75±62.56°	22.94 (i/c)
Group 3	311.75±73.68 ^a	74.50±13.96 ^a	93.50±26.73 ^a	-233.42 (d/c)

Values are means±SD. ^{abc}: Different superscripts (row) are significantly different (*P*<0.05). i/c: Increase; d/c: Decrease.

The relative heart weights (%) of the non-diabetic, diabetic control and diabetic rats fed cocoyam pellets were (0.42 ± 0.07)%, (0.45 ± 0.03)% and (0.38 ± 0.04)% respectively. There were no significant differences (P>0.05) in the relative heart weights of all the rats in the three groups.

The relative pancreatic weights (%) of the non–diabetic, diabetic control and diabetic rats fed cocoyam pellets were $(0.54\pm0.12)\%$, $(0.36\pm0.02)\%$ and $(0.43\pm0.08)\%$ respectively. The relative pancreatic weights of the diabetic rats were significantly lower (P<0.05) than those of the non–diabetic rats while the relative pancreatic weights of the diabetic rats fed cocoyam pellets were significantly higher than those of the diabetic control rats.

The diabetic rats fed cocoyam pellets recorded 23.29% loss of weight, compared with diabetic control rats that recorded 45.36% loss of weight unlike the non-diabetic rats that recorded 13.42% gain in weight (Table 2).

Table 2
Body weight of rats (g).

Groups	Initial weight	Final weight	Percentage change (%)
Group 1	175.75±24.58 ^a	203.00±13.04°	13.42 (i/c)
Group 2	169.20±38.52 ^a	116.40±15.63 ^a	-45.36 (d/c)
Group 3	221.00±21.79 ^b	179.25±10.50 ^b	-23.29 (d/c)

Values are means±SD. ^{abc}: Different superscripts (row) are significantly different (*P*<0.05). i/c: Increase; d/c: Decrease.

As shown in Table 3, there were significant increases (P<0.05) in the serum AST, ALT, ALP, total and conjugated bilirubin contents of the diabetic control rats compared with the non-diabetic rats while there were significant decreases (P<0.05) in the serum AST, ALT, ALP, total and conjugated bilirubin contents of the diabetic rats fed cocoyam pellets compared with the diabetic control rats.

Table 3
Liver function parameters in the serum of rats

Groups	AST (IU/L)	ALT (IU/L)	ALP (IU/L)	Total bilirubin	Conjugated
Groups				(mg/dL)	bilirubi
Group 1	15.56±13.08 ^a	12.96±11.21 ^a	35.65±6.14 ^a	0.55±0.09 ^a	0.37±0.12 ^a
Group 2	41.90±12.33 ^b	25.11±17.56 ^b	55.30±2.41°	1.36±0.20 ^b	0.78 ± 0.16^{c}
Group 3	18.46±12.27 ^a	15.48±11.07 ^a	47.49±3.46 ^b	0.62±0.11 ^a	0.47 ± 0.10^{b}

Values are means \pm SD. ^{abc}: Different superscripts (row) are significantly different (P<0.05).

There were significant decreases (P<0.05) in the hepatic AST, ALT and ALP activities of the diabetic control rats compared with the non-diabetic rats while there were significant increases (P<0.05) in the hepatic AST, ALT and ALP activities of the diabetic rats fed cocoyam pellets compared with the diabetic control rats (Table 4).

Table 4
Liver function parameters in the liver of rats.

Groups	AST (IU/L)	ALT (IU/L)	ALP (IU/L)
Group 1	24.68±12.54 ^b	21.68±2.78 ^b	47.38±5.14 ^b
Group 2	12.12±2.27 ^a	12.03±0.36 ^a	18.77±2.46 ^a
Group 3	45.33±19.73°	20.92±4.93 ^b	42.38±8.10 ^b

Values are means \pm SD. ^{abc}: Different superscripts (row) are significantly different (P<0.05).

There were significant (P<0.05) decreases in the serum total protein and albumin levels of the diabetic control rats compared with the non-diabetic rats while there were significant increases in the serum total protein and albumin levels of the diabetic rats fed cocoyam pellets compared with the diabetic control rats (P<0.05) (Table 4).

There were significant increases (P<0.05) in the serum urea and creatinine levels of the diabetic control rats compared with the non-diabetic rats while there were significant decreases (P<0.05) in the serum urea and creatinine levels of the diabetic rats fed cocoyam pellets compared with the diabetic control rats (P<0.05) (Table 5).

Table 5
Renal function parameters of rats.

Groups	Protein (g/dL)	Albumin (g/dL)	Urea (mg/dL)	Creatinine (mg/dL)
Group 1	7.64±0.44°	3.53±0.03 ^d	37.33±1.58 ^a	0.68±0.21 ^a
Group 2	5.37±0.61 ^a	2.34±0.31 ^a	66.50±14.99 ^b	1.47±0.03°
Group 3	6.71±0.71 ^b	2.93±0.07 ^b	40.30±15.83 ^a	0.69 ± 0.04^{a}

Values are means \pm SD. ^{abc}: Different superscripts (row) are significantly different (P<0.05).

The serum amylase levels of the non-diabetic, diabetic control and diabetic rats fed cocoyam were (109.28±20.93) IU/L, (81.44±2.24) IU/L and (72.24±5.02) IU/L respectively. There were significant decreases (*P*<0.05) in the serum amylase activities of the diabetic control rats compared with the non-diabetic rats. In addition, there were significant decreases (*P*<0.05) in the serum amylase activities of the diabetic rats fed cocoyam pellets compared with the diabetic control rats.

The serum lipase levels of the non-diabetic, diabetic control and diabetic rats fed cocoyam were (11.60 \pm 6.10) IU/L, (45.85 \pm 8.46) IU/L and (19.75 \pm 6.08) IU/L respectively. There were significant increases (P<0.05) in the serum lipase activities of the diabetic control rats compared with the

non-diabetic rats but significant decreases (P<0.05) in the serum lipase activities of the diabetic rats fed cocoyam pellets compared with the diabetic control rats (Table 6). Furthermore, there were no statistical significant differences (P>0.05) in the serum lipase levels of the diabetic rats fed cocoyam compared with the non-diabetic rats.

The composition of the cocoyam pellet consisted of 81% cocoyam flour, 9% soya bean flour, 4% vitamin mixture, 2% salt and 4% groundnut oil.

Chemical analysis of the cocoyam pellet showed that it contained (6.77±0.05)% crude fiber, (7.50±1.73) mg GAE/g of sample, (0.64±0.02)% Ca, (0.84±0.03)% K, (0.23±0.00)% Fe, (0.41±0.03)% Na, (7.50±1.73) mg GAE/g of sample, (0.35±0.14) mg/100 g oxalate and (0.13±0.04) mg/100 g phytate (Figure 1).

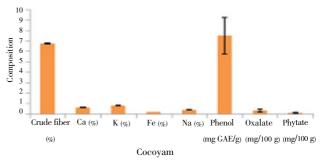


Figure 1. Chemical compostion of cocoyam pellets.

4. Discussion

The normalization of the blood glucose levels of the diabetic rats fed cocoyam pellets to the extent that was observed in this study, suggests the hypoglycemic action of cocoyam.

The non-significant changes in the relative heart weights of the rats in the three groups can be attributed to the non-susceptibility of the heart to STZ attack as the heart expresses GLUT 4 transporter^[20].

The decrease in the relative pancreatic weights of the diabetic untreated rats suggests destruction of the pancreatic beta cells while the increase in the relative pancreatic weights of the diabetic rats fed cocoyam, indicates regeneration of the pancreatic β -cells.

The percentage loss of weight by the diabetic untreated rats can be explained on the basis of increased muscle wasting and loss of structural proteins that contribute to body weight[21,22], while the improvement in the body weights of the diabetic rats fed cocoyam is indicative of the protective action of cocoyam against muscle wasting[22].

Bilirubin is excreted by the liver and any interference with the normal liver function affects its rate of conjugation and excretion^[23], thereby making bilirubin a good marker of liver function and bile excretion status.

The increase in the levels of total and conjugated bilirubin in the diabetic control rats indicates defect in the normal liver function of the rats of this group while the reduction in the total and conjugated bilurubin levels of the diabetic rats fed cocoyam pellets, suggests the ability of cocoyam to enhance liver function. ALP is a membrane bound enzyme and it is ubiquitous in nature. The primary role of extracellular phosphatases is to provide inorganic phosphate for cell growth by hydrolysis of external phosphates which do not enter the cell membrane[24].

AST and ALT are cytosolic enzymes localized within the cells of the liver, heart, gill, kidney, muscles and other organs^[24]. These enzymes are directly associated with the conversion of amino acids to keto acids which are routed for complete metabolism through the TCA cycle and the electron transport chain^[25].

The increased gluconeogenesis and ketogenesis observed in diabetes may be due to high levels of activity of these transaminases^[26]. Measurement of enzymic activities of aminotransferases and phosphatases is of clinical and toxicological importance as changes in their activities is indicative of tissue damage by toxicants or in disease conditions^[27,28].

The present study indicates an increase in the level of the liver enzymes in the serum of STZ diabetic rat models. This increase is attributable to toxicity of STZ to tissues that express GLUT 2 transporter such as hepatocytes and renal tubular cells[29].

The effect of STZ on the levels of diagnostic enzymes in the liver has remained unraveled. While some authors reported increased activities of AST, ALT[30,31] and ALP[24,32] in the liver of alloxan and STZ diabetic rat models, another group reported no alteration in the levels of these enzymes in the liver of diabetic rats[29]. In a study carried out by Sing *et al.*[13], they reported non–significant decreases in the activities of AST and ALT in the liver of STZ diabetic rat models but a significant increase in the activity of ALP in the liver of streptozotocin diabetic rat models.

The decreased hepatic levels of AST, ALT and ALP in the diabetic control rats as observed in this study lend credence to earlier works carried out by El-Demerdash *et al.* who reported decreased hepatic levels of AST, ALT and ALP in diabetic rats^[33]. This decrease may have resulted from leakage of these enzymes from the liver cytosol into the blood as a result of STZ toxicity. However, the increase in the levels of the diagnostic enzymes in the liver of the diabetic rats fed cocoyam indicates the ameliorative actions of cocoyam on liver damage.

The cocoyam variety demonstrated protection against nephrotoxicity which was evident from the increase in the serum protein and albumin as well as the decrease in the serum creatinine and urea of the diabetic rats fed the test plant.

Amylase acts on the α -1-4-glycosidic linkages of starch in the small intestine, converting them to maltose^[34] which is further hydrolyzed to glucose moieties that are absorbed in the small intestine and transported to the blood stream. Previous reports are found that amylase activity is decreased in STZ diabetic rat models^[35,36]. The decrease in the serum amylase levels of the diabetic untreated rats is attributed to the inhibition of STZ on Ca and Mg homeostasis and amylase gene expression^[37].

The increased serum amylase levels of the non-diabetic rats as observed in this study may be related to fluctuations in the rates of amylase synthesis and its secretion by secretory glands. Such fluctuations have been attributed to ageing of animals^[38,39].

The decreased serum amylase levels of the diabetic rats fed the test feed suggests that the anti-diabetic function of *C. esculenta* may be connected with a decrease in the activity of pancreatic amylase which is indicative of lesser products of carbohydrate digestion formation and subsequently absorption, leading to a decreased percentage spike in random blood glucose or the decrease may be due to endocrine, exocrine and paracrine changes in the parenchyma of the pancreas^[34,40,41].

Pancreatic lipase is the primary enzyme that hydrolyzes dietary fat, converting triglycerides to monoglyceride and fatty acid. It is a more specific marker of pancreatic dysfunction than pancreatic amylase[36] and it has also been employed in human and animal model studies involving the evaluation of natural products for their usefulness as anti-obesity and anti-diabetic agents[1]. Elevation of the serum lipase levels of the diabetic control rats as observed in this study establishes a case of acute pancreatitis for the rats of this group. The progression to diabetic state for the rats of this group suggests that their exocrine tissues may have been extensively damaged by the STZ administered. However, the reduced serum pancreatic lipase levels of the diabetic rats fed cocoyam indicates the ameliorative action of cocoyam on acute pancreatitis as playing a key role in its management of diabetes and it also indicates the potentials of cocoyam in the management of obesity.

Calcium is an important component of intracellular processes that occur within insulin responsive tissues like skeletal muscle and adipose tissue^[40]. Alteration in calcium flux can have adverse effects on insulin secretion which is a calcium–dependent process^[41].

Calcium deficiency has been suspected as having a major role in the development of type I and type II diabetes. In people without diabetes, hypocalcemia was associated with impairment of insulin release^[41]. The considerable amount of Ca in the cocoyam variety investigated could be useful in a dietary formulation for diabetics.

The higher amount of K than Na in the cocoyam pellets is considered of comparative advantage. This is because intake of diets with higher Na to K ratio has been related to the incidence of hypertension^[42].

Iron is an essential component of hemoglobin and it is critical to the proper functioning of the immune system and the production of energy^[42].

Diets rich in dietary fiber decrease the reabsorption of bile acids, thus reducing cholesterol level and increasing glucose tolerance^[43]. Dietary fiber also inhibits the activity of pancreatic amylase, thereby regulating the rate at which glucose is released into the blood. The considerable amount of crude fibre in the cocoyam variety investigated, suggests that these compounds could have contributed to the anti-diabetic properties of this cocoyam variety.

Phenolic acids refer to a class of polyphenolic compounds in the diet. Phenolics have received considerable attention because of their potential antioxidant activity and effects on carbohydrate metabolism involving the inhibition of α -glucosidase and α -amylase, the key enzymes responsible for digestion of dietary carbohydrates to glucose^[44].

Phytates and oxalates are anti-nutrients that adversely affect mineral bioavailability by forming insoluble salts with mineral elements such as zinc, calcium and iron, thereby preventing their utilization. The presence of these toxic principles in cocoyam tends to limit its utilization.

Eugene *et al.* reported that cooking, boiling, baking and oven drying at 105 °C removes acridity that results from high concentration of phytate and oxalate in root crops^[45]. In a study carried out by Famurewa *et al.*^[46], they reported a non–significant decrease in the phytate and oxalate contents of cocoyam after oven drying at 60 °C, 70 °C and 80 °C respectively although the duration of oven drying was short (50 to 250 min). Oxalates in food were classified as toxic (2 to 5 g/100g) and lethal (above 5 g/100 g)^[47].

In this study, it was observed that soaking in water for 10 min, oven drying at 70 °C for 48 h at 70 °C and subsequently for 24 h at 90 °C significantly decreased the concentration of oxalate and phytate in the test feeds to non-toxic levels, indicating the bioavailability of the minerals these active principles form complexes with. This decrease could be attributed to the water solubility as well as heat susceptibility of these compounds.

Apart from affecting mineral bioavailability, phytates in the diet also have some beneficial roles, one of which is reduction of blood glucose response (glycemic index) in humans^[48]. This they carry out by binding starch either directly through hydrogen bonds or indirectly through the proteins associated with starch or by complexing with Ca²⁺ to inhibit pancreatic amylase activity. In addition, their inhibitory action of pancreatic lipase activity makes them useful anti-obesity agents.

It is therefore possible that the crude fibre, phenolic compounds and phytate contents of this cocoyam variety may be the explanation for its pancreatic amylase and pancreatic lipase inhibitory activity or a combination of these active principles and the mineral contents of this cocoyam variety could work in synergy to confer anti-diabetic actions to it.

The study shows that *C. esculenta* may exert its antidiabetic action by delaying/regulating the rate at which dietary starch is hydrolyzed to glucose or possibly through inhibition of acute pancreatitis. Finally, the study also shows the potentials of *C. esculenta* in the dietary management of obesity.

Conflict of interest statement

We declare that we have no conflict of interest.

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References

- [1] Aloulou A, Hamden K, Elloumi D, Ali MB, Hargafi K, Jaouadi B, et al. Hypoglycemic and anti-lipidemic properties of kombucha tea in alloxan-induced diabetic rats. *BMC Complement Altern Med* 2012; **12**: 63.
- [2] Okpe O, Ibrahim S, Njoku GC, Ndidi US, Atabo S. Pancreatic islet regeneration and some liver biochemical parameters of leaf extracts of *Vitex doniana* in normal and streptozotocin–induced diabetic albino rats. *Asian Pac J Trop Biomed* 2014; 4(2): 124–130.
- [3] Tattiyakul J, Asavasaksakul S, Pradipasena P. Chemical and physical properties of flour extracted from Taro *Colocasia* esculenta (L.) Schott grown in different regions of Thailand. Sci Asia 2006; 32: 279–284.
- [4] Niba LL. Processing effects on susceptibility of starch to digestion in some dietary starch sources. Int J Food Sci Nutr 2003; 54(1): 97-109.
- [5] Mwenye OL, Labschagne MT, Herselman L, Benesi IRM, Chipungu FP. Ethno-botanical and morphological characterisation of cocoyams (Colocasia esculenta L. Schott and Xanthosoma sagittifolum L. Schott) germplasm in Malawi. Second RUFORUM Biennial Meeting; 2010 Sep 20–24, Entebbe, Uganda. 2010, p. 193–199.
- [6] Chatterjee A, Pakrashi SC. The treatise on Indian medicinal plant. New Delhi: National Institute of Science Communication; 2001, p. 32.
- [7] Kirtikar KR, Basu BD. Indian medicinal plant with illustrations. Dehradun: Oriental Enterprises; 2001, p. 3598–3602.
- [8] Patel DK, Kumar R, Laloo D, Hemalatha S. Diabetes mellitus: an overview on its pharmacological aspects and reported medicinal plants having anti-diabetic activity. *Asian Pac J Trop Biomed* 2012; 2(5): 411-420.
- [9] Eleazu CO, Iroaganachi M, Eleazu KC. Ameliorative potentials of cocoyam (Colocasia esculenta L.) and unripe plantain (Musa paradisiaca L.) on the relative tissue weights of streptozotocin-induced diabetic rats. J Diabetes Res 2013; doi: 10.1155/2013/160964.
- [10] Kumawat NS, Chaudhari SP, Wani NS, Deshmukh TA, Patil VR. Anti-diabetic activity of ethanol extract of *Colocasia esculenta* leaves in alloxan induced diabetic rats. *Int J PharmTech Res* 2010; 2(2):1246–1249.
- [11] Alegbejo JA, Ameh DA, Ogala WN, Ibrahim S. Glycemic index of boiled cocoyam and stew. Sahel Med J 2008; 11(3): 80–83.
- [12] National Academy Press. *Guide for the care and use of laboratory animals*. Washington D.C.: National Academy Press; 1996.
- [13] Singh SN, Vats P, Suri S, Shyam R, Kumria MM, Ranganathan S, et al. Effect of an anti-diabetic extract of *Catharanthus roseus* on enzymic activities in streptozotocin induced diabetic rats. *J Ethnopharmacol* 2001; 76: 269–277.
- [14] Gella FJ, Gubern G, Vidal R, Canalias F. Determination of total

- and pancreatic alpha-amylase in human serum with 2-chloro-4-nitrophenyl-a-D-maltotrioside as substrate. *Clin Chim Acta* 1997; **259**: 147–160.
- [15] Tietz NW. Clinical guide to laboratory tests. 3rd ed. Philadelphia: WB Saunders Company; 1995, p. 518–519.
- [16] Young DL, Friedman L. Effects of disease on clinical laboratory tests. 4th ed. Washington D.C.: AACC Press; 2001, p. 133.
- [17] Young DS. Effect of drugs on clinical laboratory tests. 3rd ed. Washington D.C.: AACC Press; 1997, p. 147.
- [18] Singleton VL, Orthofer R, Lamnela-Ravenlos RM. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-ciocalteu reagent. *Methods Enzymol* 1999; 299: 152-178.
- [19] Association of Official Analytical Chemists. Official methods of analysis. 15th ed. Arlington: Association of Official Analytical Chemists; 1990, p. 121.
- [20] Medina RA, Owen GI. Glucose transporters: expression, regulation and cancer. *Biol Res* 2002; **35**(1): 9–26.
- [21] Bellamkonda R, Rasineni K, Singareddy SR, Kasetti RB, Pasurla R, Chippada AR, et al. Antihyperglycemic and antioxidant activities of alcoholic extract of *Commiphora mukul* gum resin in streptozotocin induced diabetic rats. *Pathophysiology* 2011; 18: 255–261.
- [22] Rajiv GG, Sasikumar P. Anti-diabetic effect of Merremia emarginata Burm. F. in streotozotocin induced diabetic rats. Asian Pac J Trop Biomed 2012; 2(1): 281-286.
- [23] Usha K, Kasturi GM, Hemalatha P. Hepatoprotective effect of Hygrophila spinosa and Cassia occidentalis on carbon tetrachloride induced liver damage in experimental rats. Indian J Clin Biochem 2008; 22(2): 132–135.
- [24] Radhika R, Ragavan B, Sharad PD, Sudarsanam D. Action of marker enzymes of *Rheum emodi* in alloxan induced diabetic rats. *Asian J Exp Biol Sci* 2012; 3(2): 420–423.
- [25] Onaolapo AY, Onaolapo OJ. Ocium gratissimum Linn. causes dose dependent hepatotoxity in streptozotocin induced diabetic wistar rats. Maced J Med Sci 2011; 5(1): 17–25.
- [26] Maiti R, Jana D, Das U, Ghosh D. Anti-diabetic effect of aqueous extract of seed of *Tamarindus indica* in streptozotocin diabetic rats. *J Ethnopharmacol* 2004; 92: 85–91.
- [27] Singh SN, Praveen V, Shoba S, Radhey S, Kumria MML, Ranganathan S, et al. Effect of an anti-diabetic extract of *Catharanthus roseus* on enzymic activities in streptozotocin induced diabetic rats. *J Ethnopharmacol* 2001; 76: 269-277.
- [28] Adesokan AA, Oyewole OI, Turay BM. Kidney and liver function parameters in alloxan-induced diabetic rats treated with aloe barbadensis juice extract. Sierra Leone J Biomed Res 2009; 1(1): 33-37.
- [29] Eleazu CO, Eleazu KC, Chkwuma SC, Udeme N. Review of the mechanism of cell death resulting from streptozotocin challenge in experimental animals, its practical use and potential risk to humans. J Diabetes Metab Disord 2013; 12: 60.
- [30] Jordá A, Gomez M, Cabo J, Grisolía S. Effect of streptozotocin diabetes on some urea cycle enzymes. Biochem Biophys Res Commun 1982; 106: 37–43.
- [31] Rajesh M. Protective effects of *Carthamus tinctorius* on streptozotocin induced diabetic complications in rats and the possible morphological changes in the liver and kidney. *Int J Sci*

- Innovat Discov 2012; 2(5): 502-510.
- [32] Yadav UC, Moorthy K, Baquer NZ. Effects of sodium—orthovanadate and *Trigonella foenum*—graecum seeds on hepatic and renal lipogenic enzymes and lipid profile during alloxan diabetes. *J Biosci* 2004; **29**(1): 81–91.
- [33] El-Demerdash FM, Yousef MI, El-Naga NI. Biochemical study on the hypoglycemic effects of onion and garlic in alloxan-induced diabetic rats. Food Chem Toxicol 2005; 43: 57-63.
- [34] Mori DM, Baviera AM, de Oliveira Ramalho LT, Vendramini RC, Brunetti IL, Pepato MT. Temporal response pattern of biochemical analytes in experimental diabetes. *Biotechnol Appl Biochem* 2003; 38: 183–191.
- [35] Nayak S, Shivnanda NB. Maniple manual of clinical biochemistry. New Delhi: Jaypee Brothers Medical Publishers; 2007, p. 161–162.
- [36] Patel R, Yago MD, Victoria EM, Shervington A, Singh J. Mechanism of exocrine pancreatic insufficiency in streptozotocininduced diabetes mellitus in rat: effect of cholecystokininoctapeptide. *Mol Cell Biochem* 2004; 261(1): 83-89.
- [37] Omoruyi FO, Budiama A, Eng Y, Olumese FE, Hoesel JL, Ejilemele A, et al. The potential benefits and adverse effects of phytic acid supplement in streptozotocin-induced diabetic rats. Adv Pharmacol Sci 2013; doi: 10.1155/2013/172494.
- [38] Pepato MT, Baviera AM, Vendramini RC, Brunetti IL. Evaluation of toxicity after one-month treatment with *Bauhinia forficate* decoction in streptozotocin-induced diabetic rats. *BMC* Complement Altern Med 2004; 4: 7.
- [39] Kim JD, Kang SM, Seo BI, Choi HY, Choi HS, Ku SK. Anti-diabetic activity of SMK001, a poly herbal formula in streptozotocininduced diabetic rats: therapeutic study. *Biol Pharm Bull* 2006; 29(3): 477–482.
- [40] Pittas AG, Lau J, Hu FB, Dawson-Hughes B. Review: the role of vitamin D and calcium in type 2 diabetes. A schematic review and metal analysis. J Clin Endocrinol Metab 2007; 92(6): 2017– 2029.
- [41] O'Connell B. Select vitamins and minerals in the management of diabetes. *Diabet Spect* 2001; 14: 133–148.
- [42] Chen XH, Xia LX, Zhou HB, Qiu GZ. Chemical composition and antioxidant activities of *Russula griseocarnosa* sp. nov. *J Agric Food Chem* 2010; **58**: 6966–6971.
- [43] Vasundev V. Fundamentals of biochemistry, textbook of Biochemistry. 2nd ed. New York: John Wiley & Sons Inc.; 2006; p. 110–214.
- [44] Iwai K. Anti-diabetic and antioxidant effects of polyphenols in brown alga *Ecklonia stolonifera* in genetically diabetic KK-A(y) mice. *Plant Foods Hum Nutr* 2008; **63**: 163–169.
- [45] Eugene NO, Joyce OA, Jonathan CI. Effect of heat processing on the proximate composition and energy values of selected Nigerian staple foods from oil-producing areas of the Niger Delta. *Biochemistry* 2008; 20(1): 1-9.
- [46] Famurewa JAV, Olatujoye JB, Ajibode A. Drying phenomenon and influence on the anti–nutritional and pasting properties of cocoyam (Taro). *J Sci Res Rep* 2014; **3**(2): 275–283.
- [47] Hui YH. Encyclopedia of food science and technology. New York: John Wiley & Sons Inc; 1992, p. 58.
- [48] Kumar V, Sinha AK, Makkar HPS, Becker K. Dietary roles of phytate and phytase in human nutrition: a review. Food Chem 2010; 120: 945–959.