

Short Communication

Evaluation of Hypoglycaemic Efficacy of Aqueous Seed Extract of *Aframomum melegueta* in Alloxan-induced Diabetic Rats

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ABSTRACT

The hypoglycaemic efficacy of aqueous seed extract of *Aframomum melegueta* was investigated in alloxan-induced diabetic rats. Twenty five albino rats (*Rattus norvegicus*, average weight: 150g) were randomly divided into five groups of five (5) rats each. For the diabetic group, diabetes mellitus was induced by intraperitoneal injection of 5% solution of alloxan at a dose of 150 mg/kg body weight. Diabetes was confirmed 72 hours after alloxan injection if fasting blood glucose (FBG) was ≥ 10 mmol/l. Two control groups: non-diabetic (positive) and diabetic (negative) were administered tap water as vehicle solvent, throughout the duration of the experiment. The remaining three groups received 100 mg/kg of metformin, 200 and 400 mg/kg orally and aqueous seed extract of *Aframomum melegueta* respectively and simultaneously. Fasting blood glucose was evaluated daily. The results showed that oral administration of aqueous extract of *A. melegueta* to diabetic rats lowered blood glucose to normal level within 6 days of administration, while metformin took 14 days. There was no significant difference in the duration of lowering the blood glucose by the two doses of extract administered. In conclusion, oral administration of aqueous seed extract of *Aframomum melegueta* has potent hypoglycaemic activity in alloxanised diabetic rats.

Keywords: *Aframomum melegueta*, Alloxan, Aqueous seed extract, Diabetic rats, Hypoglycaemia

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INTRODUCTION

Diabetes mellitus is a metabolic disorder characterised by hyperglycaemia and alteration in carbohydrate, fat and protein metabolism and it is associated with absolute or relative deficiencies in insulin secretion or insulin action (Bhupesh *et al.*, 2009). The incidence of diabetes has reached an epidemic proportion worldwide. It is estimated that about 2.8% of the world's population suffer from diabetes, according to World Health Organisation (Wild *et al.*, 2004). The incidence is increasing rapidly and it is anticipated that by the year 2030, it will double the current figure (Ghost *et al.*, 2004). This rapid increase in the incidence of the disease is associated with rapidly changing lifestyle and environmental factors (Lang *et al.*, 2008; Mozaffarian *et al.*, 2009).

The use of herbal medicine is widespread throughout the world. Many medicinal plants have

been confirmed to have hypoglycaemic properties. Some of them include *Pterocarpus massupum* (Indian Kino), *Allium sativum* (Garlic), *Azadirachta indica* (Neem), *Carica papaya* (unripe fruit), *Trigonella foerum* (Fenugreek), *Ocimum santum* (Tulsi), *Aloe barbadensis* miller (Adesokan *et al.*, 2006; Akinola, 2010). *Aframomum melegueta* is an herbaceous perennial plant species of the ginger family that is native to the swampy habitats along the West African coast. The *Aframomum* plant is trumpet-shaped with purple flowers that develop into 5-7 cm long with numerous reddish-brown seeds. Depending on the country, it is called several names including grains of paradise, melegueta pepper, alligator pepper and Guinea pepper.

In Nigeria, it is called *ata ire* (Yoruba), and *chitta* (Hausa). It has been established to be a potent antimicrobial agent (Stroev, 1989); effective as an

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antifungal (Okigbo and Ogonnaya, 2006) and used to treat dysentery and diarrhoea (Stroev, 1989). The seed is also used in treating inflammatory conditions of the throat, fever and exanthemata (Apata, 1979); snake bites (Nebojsa *et al.*, 2009) and has antioxidant and antitumor effects (Chan *et al.*, 2004). Although, the anti-diabetic effectiveness of *Aframomum melegueta* is acclaimed traditionally (Gbolade, 2009), there is no proven scientific evidence to ascertain this. This study reports on the anti-diabetic potential of aqueous seed extract of *Aframomum melegueta* in alloxan-induced diabetic rats.

MATERIALS AND METHODS

Plant Material

The seeds of *Aframomum melegueta* were obtained from Oja-Oba market, Ilorin and were authenticated in the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria.

Preparation of Seed Extract

Seeds of *A. melegueta* were washed with water and air-dried under the shade to constant weight. The seeds were ground into powdery form with an electric blender (model MS- 223). The 20g of the powdered seed was soaked in 100ml of distilled water with constant stirring by a magnetic stirrer for 48 hours. It was filtered with muslin cloth and the filtrate evaporated in a water bath at 40°C. 0.4g of the extract yielded 2% of 20g and was stored in a refrigerator at 8°C, from where stock solution was prepared.

Experimental Animals

Twenty five albino rats (*Rattus norvegicus*) of both sexes and with an average weight of 150±2g, were obtained from the Small Animal Holding Unit of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria. They were exposed to 12 hour daylight and 12 hour darkness, fed rat chew, water *ad libitum*, and were allowed to acclimatise for two weeks. They were randomly separated into five groups of 5 rats each according to their sexes. Group A which served as positive control, was given distilled and diabetes was not induced. Group B which served as the negative control, had diabetic induced and was left untreated. Group C also developed diabetes and was treated with 100 mg/kg body weight of anti-diabetic drug, metformin, (Hovid, Bhd, Ipoh, Malaysia) procured from a registered pharmacy shop within Ilorin metropolis. Groups D and E were induced diabetes and treated with 200 and 400 mg/kg body weight of aqueous seed extract of *Aframomum melegueta* respectively, based on

preliminary studies. All the albino rats were kept under laboratory conditions (25±2°C and relative humidity of 50±15%) and their cages were cleaned of metabolic waste twice daily.

Induction of Diabetes Mellitus

Diabetes was induced by intraperitoneal injection of 5% aqueous solution of alloxan at a dose of 150 mg/kg body weight. Alloxan (2,4,5,6-tetraoxypyrimidine; 2,4,5,6-pyrimidine tetrone), is an oxygenated pyrimidine derivative and a toxic glucose analogue, which specifically destroys insulin producing β -cells of the pancreas. It therefore causes insulin dependent diabetes mellitus (IDDM). Fasting blood glucose was determined in each rat prior to the injection of alloxan to exclude those that could be naturally diabetic. Diabetes was confirmed if fasting blood glucose was ≥ 10 mmol/L.

Determination of Blood Glucose

Fasting blood glucose was determined by glucose oxidase method using Accu chek glucometer (Roche diagnostics, Germany). The tail of the rat was cut swiftly with sterile scalpel and a drop of blood was squeezed onto the test area of strip inserted into the glucometer. The animals were fasted for 12 hours before each glucose determination, which was repeated every 48 hrs till the end of the experiment.

Statistical Analysis

All values were expressed as mean \pm SD and the means were subjected to two way analysis of variance using the software – SPSS. *P* values of <0.05 were considered statistically significant.

RESULTS

The normal control (positive) group remained normoglycaemic throughout the duration of the experiment, while the negative control group B became hyperglycaemic 72 hrs after alloxan injection and remained so for 10 days, before all the animals died. Group C, that received metformin, showed gradual reduction of blood glucose and became normoglycaemic by the 14th day. The test groups D and E that received 200 and 400 mg/kg body weight of aqueous seed extract of *A. melegueta* respectively, showed rapid reduction of blood glucose that returned to normal within 6 days. The two extract doses did not show any difference in the rate and duration of reduction of blood glucose, meaning that the effect was not dose dependent. These results are summarised in Table 1.

Table 1: Fasting Blood Glucose Concentrations (mmol/L) of Control Groups and those Administered Metformin and Aqueous Seed Extract of *Aframomum melegueta* in Alloxan Induced-diabetic Rats

Animal Groups	Day 0	Day 2	Day 4	Day 6	Day 8	Day 10	Day 12	Day 14
Normal: Control Group A	5.27±0.25	5.27±0.25	5.27±0.25	5.27±0.25	5.27±0.25	5.27±0.25	5.27±0.25	5.27±0.25
Diabetic: Control Group B	27.62±0.01	26.62±0.01	26.54±0.01	26.67±0.02	26.73±0.18	26.00±0.01	25.00±0.01	25.00±0.01
Metformin: Group C	23.81±0.01	18.67±0.01	17.12±0.01	11.70±0.00	9.46±0.01	7.12±0.01	5.74±0.00	4.52±0.01
<i>A.melegueta</i> 200 mg/kg: Group D	25.89±0.02	17.19±0.01	11.10±0.00	4.74±0.01	4.74±0.01	4.74±0.01	4.74±0.01	4.74±0.01
<i>A.melegueta</i> 400 mg/kg: Group E	23.58±0.00	14.45±0.00	10.03±0.01	4.95±0.01	4.95±0.01	4.95±0.01	4.95±0.01	4.95±0.01

DISCUSSION

This study, scientifically confirmed the locally acclaimed glucose lowering effect of aqueous seed extract of *Aframomum melegueta* by traditional medicine practitioners. The extract possesses a more potent hypoglycaemic property than metformin. It took 14 days to attain normoglycaemia in the group that received metformin and only 6 days in the two groups that received extract of *A. melegueta*. The hypoglycaemic effect of the plant extract could be insulinotropic, since alloxan induced hyperglycaemia, by causing insulin dependent diabetes mellitus (IDDM), or may be due to a combination of inhibition of hepatic glycogenolysis and gluconeogenesis (Chattopadhyay, 1999). Metformin causes hypoglycaemia by delaying glucose uptake from gastrointestinal tract, increase peripheral glucose utilisation mediated by increased insulin sensitivity; inhibits hepatic and renal gluconeogenesis (Clark and Pierce, 2000).

Metformin has also been shown to reduce low density lipoprotein cholesterol and triglyceride levels, and prevent cardiovascular complications associated with diabetes (Clark and Pierce, 2000). Conversely, the observed hypoglycaemic effect of the plant extract could be insulinotropic (Dunstan *et al.*, 2002), since alloxan induced hyperglycaemia, by causing insulin dependent diabetes mellitus (IDDM), or may be due to a combination of inhibition of hepatic glycogenolysis and gluconeogenesis (Chattopadhyay, 1996; 1999). The fact that insulin is completely absent in alloxan-induced diabetes, perhaps explain the reason the extract of *A. melegueta* is more efficient than metformin. Earlier workers have however identified gingerols and related compounds in *A. melegueta*, which may be responsible for its hypoglycaemic property

(Bhupesh *et al.*, 2009). Essential oil, paradol, among others, is the active flavour constituent of *A. melegueta*, that has been found to possess antioxidant and antitumor properties (Chan *et al.*, 2004).

In conclusion, the plant *Aframomum melegueta* can be of immense use in phytomedicine especially for the management of diabetes mellitus. This is a preliminary study and further studies on precise mechanism of action of the plants in reducing hyperglycaemia would be necessary. It would also be worthwhile to isolate the acclaimed active ingredients responsible for the pharmacologic activity in subsequent research.

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