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Kigellia Africana dried leaf Methanolic extract's Antidiabetic potential in diabetic rats caused by Alloxan

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Abstract
The objective of the current investigation is to ascertain the antidiabetic potential of a methanolic extract of dried *Kigellia Africana* leaves. Historically, this nasty medication was used to reduce blood glucose levels. The continuous heat extraction method was used to obtain the methanolic extract. Using albino rats, the extract's antihyperglycemic efficacy was examined. Animals received an oral dose of 200mg/kg of extract as opposed to the normal dose of glibenclamide and the control. Male and female rats were used in the experiment, which lasted 21 days. Blood drawn from animals utilising the retro orbital approach was tested using a glucometer for blood sugar levels. The extract significantly outperformed the norm in terms of anti-diabetic efficacy. It was concluded that the herbal formulation for antidiabetic application can employ the current crude medication.

Keywords: *Kigellia Africana*, alloxan, glibenclamide, antidiabetic activity

Introduction
Nearly 10% of the population is affected by diabetes (Madhumeha), a serious illness whose prevalence is rising quickly. Diabetes can cause major side effects like blindness, kidney, heart, and impotence issues, as well as neuropathy and impotence. Diabetes and its consequences remain a serious medical issue despite the development of hypoglycemic drugs. Diabetes management without any side effects continues to be a problem for the medical system. The fact that herbal medicines are compatible with human systems, easily metabolized, and have fewer side effects makes them a prime candidate for this venture.
More over 180 million people worldwide are estimated [2] to have diabetes, according to the World Health Organization (WHO). An estimated 1.1 million persons lost their lives to diabetes in 2005. Consequently, despite the availability of numerous anti-diabetic medicines on the market, diabetes and related complications continues to be a major medical problem. Urgent breakthroughs are needed in combating this disease. This leads to increasing demand for natural products with anti-diabetic activity and less side effects.
The sausage tree, or *Kigelia africana*, is a spreading tree with long, pendulous racemes of black, mottled blooms that resemble candelabra. It is a member of the Bignoniaceae family. Its fruits have long, woody stalks that resemble cords and have a sausage-like look [3]. The flowers have two-lipped, irregular bell-shaped corollas that are 9 to 13 cm long, yellowish on the surface, and purple on the inside. They are found in spring or summer and dangle on auxiliary panicles that can be up to 2 metres long. Oblong, hard, 30–50 cm long, and hanging on a stalk for several months, the fruits are difficult to divide. The fruit has purgative effects and is used in Africa as a treatment for rheumatism, syphilis, and ulcers. Fruits that have been pickled in vinegar are used as an appetiser and to treat constipation and to remove kidney stone. Most traditional healers use it to treat a wide range of skin ailments like, fungal infection, boils, psoriasis and eczema. It also has internal application including the treatments in dysentery, ringworm, tapeworm, post- partum haemorrhage, malaria, diabetes, pneumonia and toothache [4].
The 12–20 cm long, 3–7 leaflet leaves are grouped in threes at the ends of the branches. The inflorescence is a 35–75 cm long panicle. The tubular flowers have a terrible odour and are dark red with yellow veins. The fruits are sausage-shaped, measuring between 6.5 and 80 cm in length and diameter. Fruits can be dried and fermented, but both ripe and unripe fruits are harmful to humans. Sometimes leaves are used to make a general tonic for better development and health. Aqueous fruit solutions are used as a wash or a massage to help babies gain weight and have analgesic effects. Digestive diseases are treated with the roots, bark, leaves, stems,

twigs, and fruits of this plant. The roots bark and ripe or unripe fruits are taken as a laxative or emetic, to treat chronic and acute digestive disorders and against gastric infections. Remedies containing the fruits of *Kigelia africana* are taken internally to relieve constipation or haemorrhoids, anti-inflammatory, antimalerial, antidiarrhoeal and syphilis. Stem bark showing antibacterial activity. Traditionally this plant has been used for several important medicinal purposes like wide range of skin ailments like, fungal infection, boils, psoriasis and eczema, dysentery, ringworm, tapeworm, post-partum haemorrhage, malaria, diabetes, pneumonia. Despite a long tradition of use, no work has been carried out to justify its traditional claims, specially, analgesic, antidiabetic properties [5]. Traditionally this plant has been used for several important medicinal purposes like wide range of skin ailments like, fungal infection, boils, psoriasis and eczema, dysentery, ringworm, tapeworm, post- partum haemorrhage, malaria, diabetes, pneumonia.

Exploiting India's herbal integrity can be done to treat each of these diseases. In addition to treating chronic diseases like diabetes mellitus, plants are also capable of treating linked conditions including polyuria, polydipsia, glucosuria, etc. The Charak Samhita [6], one of our Vedic texts, mentions the use of plants, herbs, and their derivatives for the treatment of diabetes mellitus. About 700 recipes that are used to treat diabetes mellitus in about two thirds of the world's population contain ingredients from more than 400 plants. The hypoglycaemic property of numerous plants, which has already been documented in many literatures, has been demonstrated in a vast number of in vivo investigations on animals to test the stated activity. Present study was therefore aimed to investigate possible antidiabetic effect of methanolic leaves extract of *Kigellia Africana* against alloxan induced diabetic rats.

Materials and Methods

Plant material

The leaves of *Kigellia aricana* were collected during the month of October from district- Sangli region (M.S) and were authentified by botanist Dr. G. G. Potdar, Y. C. College, Karad. The voucher specimen SGG1 was maintained and kept for future reference.

Chemicals

Alloxan obtained as gift sample from Explicit Chemicals Pvt. Ltd, Mumbai. India and Glibenclamide from Shri Krishna Drugs Ltd. Medak (AP), India. All other chemicals and reagents used were of analytical grade purchased from Research lab fine chem., Mumbai.

Preparation of extract

The leaves were air dried under shady conditions. By utilising a dry grinder and sieve number 40, the dried leaves were reduced in size to a coarse powder. A soxhlet device was used to pack the 40gm of *Kigellia Africana* powder before it was extracted [7] with petroleum ether for deffating [8]. The extracted substance was dried using a heating mantle until a solid or semi-solid mass was produced, and it was then kept in desiccators.

Before giving each extract to the animals orally, a weighed portion of each extract was dissolved in 1% gum acacia solution in filtered water. The oral feeding needle was used to deliver the extracts orally. The dose chosen and the animal's weight were used to calculate the volume of extract delivered [9].

Animals used

Wistar albino rats (150-200 gm) of both sexes were employed in this study. The rats were maintained under standard laboratory conditions at 25 ± 2 °C, relative humidity 50 ± 15% and normal photo period [12 h dark/12 h light] were used for the experiment. Commercial pellet diet [Chakan Mill, Sangli] and water were provided ad libitum. The experimental protocol has been approved by the Institutional Animal Ethics committee

Induction of Diabetes [10]

Prior to solubilization with distilled water, alloxan monohydrate was first weighed individually for each animal in accordance with their weight. Alloxan was injected at a dose of 200 mg/kg body weight to cause diabetes. The animals were given meal and 5% dextrose solution an hour after receiving alloxan to help them recover from the initial hypoglycemic period. Accucheck Glucometer was used to measure blood glucose after 72 hours. The diabetic rats (glucose levels of 200–300 mg/dl) were placed into four groups, each with six animals.

Experimental design

Different groups of rats were used to study the effects of *Kigellia Africana* methanolic leaves extract (KAME). The rats were divided into four groups each consisting of six rats.

Group I: Normal Control. Received the vehicle (1% gum acacia).

Group II: Diabetic Control. Received the vehicle (Distilled water).

Group III: Diabetic Reference treated with glibenclamide in a dose of 5 mg/kg, orally.

Group IV: Diabetic animals treated with KAME at a dose of 200 mg/kg, orally.

Design-

Group -I: normal control group contain six animals and all animals were first acclimatised and provided water and pellets. Prepared 1% gum acacia solution in purified water and treated to all animals (1ml) through oral route by oral feeding needle. After 0,7th, 14th and 21th day blood was withdrawn by retro orbital technique. About one drop of blood was put on glucometer strip and measured the blood glucose level.

Group II- Diabetic control group contain six animals and all animals were kept in suitable environmental condition. Acclimatised before started the activity. All animals were given alloxan dose 200mg/kg body weight of each animal. Dose of alloxan was given through oral route by oral feeding needle. After 0,7th, 14th and 21th day blood was windrowed by retro orbital technique. The treatment was carried out upto 21 days.

Group III- standard control group contain six animals in group and all animals were kept in suitable environmental condition provided water and feed. All animals were first made diabetic by alloxan then prepared 5mg/kg solution of glibenclamide and given to each rat through oral route by oral feeding needle. Each rat then provided water and feed. Blood was windrowed by retro orbital technique after 0,7th, 14th and 21th day and blood glucose level was measured by glucometer.

Group IV- this group contains six animals. All animals were provided with water and feed. All animas were made diabetic by alloxan. Then prepared the solution of KAME in 1% gum acacia of dose 200mg/kg and given to all animals through oral route by oral feeding needle. Then after 0,7th, 14th and 21th

day blood was windrowed by retro orbital technique and blood glucose level was measured by Accucheck Glucometer
At the time of study following parameters were studied-
1. Blood glucose level
2. Body weight

Body weight measurement [9-10]
Using an animal weighing scale, body weight was measured a total of five times during the course of the study period (i.e., prior to alloxan induction (starting value), and then on days 7, 14, and 21 of the treatment period after alloxan induction).

Statistical analysis [11]
All of the data were reported as Mean SEM, statistical analysis was performed using ANOVA, followed by a comparison to the appropriate control group using Dunnett's test. Statistics was considered significant at a value of P 0.05.

Results
Effect on blood glucose level
Alloxan treatment caused a considerable rise in blood glucose levels in diabetic rats compared to normal controls. In the study, a dose-dependent decrease in blood glucose levels was caused by daily injection of the KAME extract for three weeks. At the 200 mg/kg KAME dose, the blood glucose level at the end of the experiment (the 21 day) was 121 mg/dl. Table 1 also displays the anti-diabetic effects of KAME on the blood glucose levels in diabetic rats.

Effect on body weight
The body weight of the diabetic controls (group II) significantly decreased compared with the normal controls (group I). During the weekly of observation of the KAME treated diabetic rats at dose of 200 mg/kg there were significant ($p< 0.05$) weight gains on day 21 relative to day 0 as shown in Table 2.

Discussion
Alloxan can kill the pancreatic beta cells that make insulin, which is how it causes diabetes [12-13]. In vitro research has demonstrated that alloxan selectively damages pancreatic beta cells, inducing cell necrosis. In the current work, the hypoglycemic impact of a methanolic extract of *Kigellia Africana* leaves was assessed in diabetic rats caused by alloxan.
When compared to normal rats, the injection of alloxan significantly raised blood glucose levels. From day 7 on, diabetic rats' blood glucose levels were significantly and consistently reduced after receiving 200 mg/kg of the methanolic leaves extract. It was discovered that a dose of 200 mg/kg was equivalent to a dose of glibenclamide at 0.5 mg/kg. According to the experimental findings on the effects of *Kigellia Africana*'s methanolic extract in rats without diabetes (Table 1), blood sugar levels considerably decline starting on day 7 (p=0.05) and continuing through day 21 (p=0.001). The methanolic extract of *Kigellia Africana* demonstrated statistically significant hypoglycemic activities when compared to conventional glibenclamide, demonstrating effective antidiabetic activity. The hypoglycemic effect comparable to glibenclamide suggested that the extract may act by regenerating the β-cells in alloxan-induced diabetes.
A 21-day period of *Kigellia Africana* administration had an impact on body weight. Normal, healthy animals were discovered to have steady body weights, whereas diabetic animals displayed weight loss. Diabetes caused an increase in muscle wasting and a loss of tissue proteins, which led to a fall in weight [16]. In the trial, extracts therapy reduced body weight loss after 21 days of treatment in a dose-dependent manner. Additionally, the extract improved body weight.

Conclusion
Thus, *Kigellia Africana* leaf methanolic extract demonstrated hypoglycemic efficacy in diabetic rats that was comparable to the common medication glibenclamide. Additionally, it supports the conventional wisdom that *Kigellia Africana* has anti-diabetic properties. The extract of this unprocessed medication may lower blood sugar levels by regenerating pancreatic beta cells, causing glucose to accumulate in muscle, or breaking down glucose quickly. The findings of this study could help develop a comprehensive strategy for treating diabetes.

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Conflict of interest
The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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