

## Research Article

# Anti-Diabetic Potential of Methanol Extract of Cooked Corn Silk (Stigma Maydis) on Alloxan-Induced Diabetes in Albino Mice

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## Abstract

The use of herbal remedies, including corn silk, classified as *Stigma* or *Zea mays* are popular as an alternative to standard Western allopathic medicine for a variety of problems, including prostate disorders, a diuretic as well as for bedwetting and obesity. In the present study the ant diabetic potential of methanol extract of cooked, dried and pulverized *Stigma* was assessed in alloxan-induced diabetic albino mice. Hyperglycemia induced by the injection of 150 mg/kg (i.p.) of alloxan monohydrate freshly dissolved in physiological saline. Doses (250, 500 and 750 mg/kg) per os, of the extract were separately administered to three groups of five diabetic mice each in the study. The activity compared with reference standard glibenclamide (2 mg/kg, p.o.) and negative control of physiological saline. Treatment of the alloxan-induced diabetic mice with the methanol extract of *Stigma* decreased the raised blood glucose levels in a dose-dependent manner. The methanol extract exhibited relatively significant ( $P<0.05$ ) anti-diabetic effect after 72 h and 96 h of treatment with the doses of 500 and 750 mg/kg body weight, respectively. Results of photochemical screening of the methanol extract of cooked *Stigma* have indicated the presence of cardiac glycoside, steroids, terpenoids, alkaloids, flavonoids, carbohydrates and anthraquinones.

## Objective of the Study

All studies done on the anti-diabetic activity of corn silks did not include cooked corn silk. Of course, therefore, some photochemical may be lost into the boiling water while cooking corn silks. This may or may not include corn silk polysaccharides which were reported to be ant diabetic [1] therefore interested in finding out the effect of boiling the corn silk. I don't know whether this is not a drive to carrying out a research.

**Keywords:** Alloxan Monohydrate; Cooked Corn Silk; Diabetes Mellitus; Physiological Saline; Glibenclamide; Albino Mice

## Introduction

Corn silks are scientifically referred to as *Maydis* stigma or *Zea mays*. There are five species of *zea* genus, which are *Z. diploperennis*, *Z. luxurians*, *Z. nicaraguensis*, *Z. parviflora* and *Z. mays*

L. [2]. However, *Z. mays* subspecies is the only cultivated species while the other species and subspecies are wild grasses. *Zea Mays* is simply called maize or corn. As a monoecious plant, corn produces both male and female flowers. The corn silk that grows out of the ear is the female part of a corn plant while the male part of the plant is the tassel growing out the top of the corn stalk. The corn silk is the stigma and style of the female part of the corn. The stigma is the sticky end of the silk where pollen attaches and the style is the tube from the stigma to the ovary, where the kernel forms on the cob. Corn silk elongates beyond the cob covering the edible part of the plant. The silks are fine, soft, relatively long (10-20 cm) with a mild sweetish taste and look like a tuft of hairs [3]. The color of the corn silks, at first are usually light green and later turn into red, yellow or light brown *Zea mays* are used as either a food, as animal feed or as industrial raw material. In developing countries like Nigeria maize is mainly used as a staple food whereas more than 60 percent of it is used as animal feed in the de-

veloped countries. Corn silk as an herb has been reported to have properties of antioxidant, anti-prostitutes and antispasmodic [4]. In addition, corn silk is well known in treating infection and cystitis [5], kidney stone and other renal illness [6]. One study, however, has shown that corn silk has no antibacterial activity when investigated against many bacterial species [7]. Based on folk remedies, corn silk has been used as an oral ant diabetic agent in China for decades. A part from the ant diabetic use, corn silk is also used as a diuretic and a decoction of the silk is taken for the treatment of urinary troubles and gallstones [8]. In fact, although not scientifically proven, corn silk tea has been claimed to have so many health benefits to human such as lowering blood pressure, decrease prostate inflammation, diabetic and urinary tract infection, edema, obesity and promote relaxation [9]. However, in spite of its widespread use, the mechanisms underlying hypoglycemic activity of corn silk not yet understood [10]. With regards to toxicity, a study using male and female Westar rats has confirmed that corn silk is non-toxic in nature [11]. The intake of corn silk as such has also been reported to have no adverse effects [12] and this supports the safety of corn silk for human consumption and its use as an herbal drug for healthcare applications.

## Mechanism of action

Glibenclamide simply works mainly by stimulating  $\beta$ -cells of the pancreas to produce more insulin. With chronic administration of glibenclamide the blood glucose lowering effect persists despite a gradual decline in the insulin secretary response to the drug. This may explain also why the decrease in blood glucose level in my research prolonged.

## Metabolism of glibenclamide in the liver

Glibenclamide has a short plasma half-life (2-10 hrs) but prolonged biological effect due to the formation of active metabolites! Generally, sulphonylureas (glibenclamide inclusive) cause hypoglycemia (as a side effect). Glibenclamide metabolites (3-cis-hydroxyglibenclamide and 4-trans-hydroxyglibenclamide - where the cyclohexyl ring is hydroxylated at positions 3 and 4, respectively) exert some hypoglycemic effect that contributes to the prolonged decrease in blood glucose levels in test animals. Retention of 4-trans-hydroxyglibenclamide prolongs the hypoglycemic effect of the agent in those with severe renal impairment. Clearance may therefore be substantially decreased in those with severe renal impairment. However, I did not carry out renal impairment test on the test animals.

## Materials and methods

Plant material Maize or corn purchased in Wudil local market, Wudil Local Government area, Kano state, Nigeria. Locally

maize is roasted or cooked and the seeds removed from the cobs and eaten using teeth. Before roasting, both the husks and silks have to be removed. The whole maize containing the husks and silks are usually cooked in pure water. In this study the cooked maize used and after the cooking done the silks, mostly attached in between the rows of the seeds, were removed, air dried for four weeks and pulverized to coarse powder using pestle and mortar. The powdered material (dark brown) kept in an airtight and dried white polyethene bag.

### Sample extraction

1kg of the corn silk powder divided into 2 portions of 500g each and macerated with distilled methanol (2.0 L) at room temperature in maceration bottles. The maceration bottles were occasionally shaken for three days to obtain a reasonable amount of extract and then decanted and filtered using filter paper. Fresh solvent (1.5 L) added to the remains in the two bottles and the bottles occasionally shaken for two days. The extracts were decanted and again filtered into the previous filtered extracts. The filtrates of the two extracts of each portion were combined and concentrated using rotary evaporator to obtain a methanol extract (brownish-black paste) and the solvent recovered.

### Photochemical screening

Photochemical screening carried out on the methanol extract of corn silk using standard procedures and tests to determine the presence of alkaloids, flavonoids, anthraquinones, steroids, terpenoids, tannins, cardiac glycosides, reducing sugars and saponins.

### Pharmacological study

#### Animals

All experimental animals used were male and female albino mice (weighing between 24-33g and were obtained from Nigerian Institute for Tripanosomiasis Research (NITR), Kaduna State, Nigeria. The mice were kept in clean and dry aluminium cages under standard environmental conditions and were fed using growers marsh feed and given water ad libitum. The mice were fasted, however, for 12 hours before experimentation. The experimental methods involving animals have been approved by Animal Research Ethics Committee of Kaduna State University, Kaduna-Nigeria.

#### Experimental design

Diabetes induced by a single intraperitoneal injection of 150mg/kg body weight alloxan monohydrate freshly dissolved in regular saline 0.9 % physiological saline immediately before used to overnight fasted albino mice (free access to water allowed). Seven days after, animals with fasting blood glucose level  $\geq 7.0$  mmol/dL (or  $\geq 126$  mg/dL) or more were considered diabetic and

employed in the study. The diabetic mice were then grouped into 5 groups of five mice each as follows

Group I: Served as positive control and received glibenclamide (2 ml/kg bodyweight)

Group II: Received Methanol extract at 250 mg/kg bodyweight

Group III: Received Methanol extract at 500 mg/kg body weight

Group IV: Received Methanol extract at 750 mg/kg body weight

Group V: Served as negative control receiving physiological saline (10 ml/kg body weight)

The animals were treated once and blood glucose concentrations were measured at 0, 24, 48, 72 and 96 hours. Blood samples were taken by a snip-cut at the tip of the tail and blood glucose levels were measured with a glucometer (a ONE TOUCH Ultra easy blood glucose monitoring system, Life Scan Europe Division

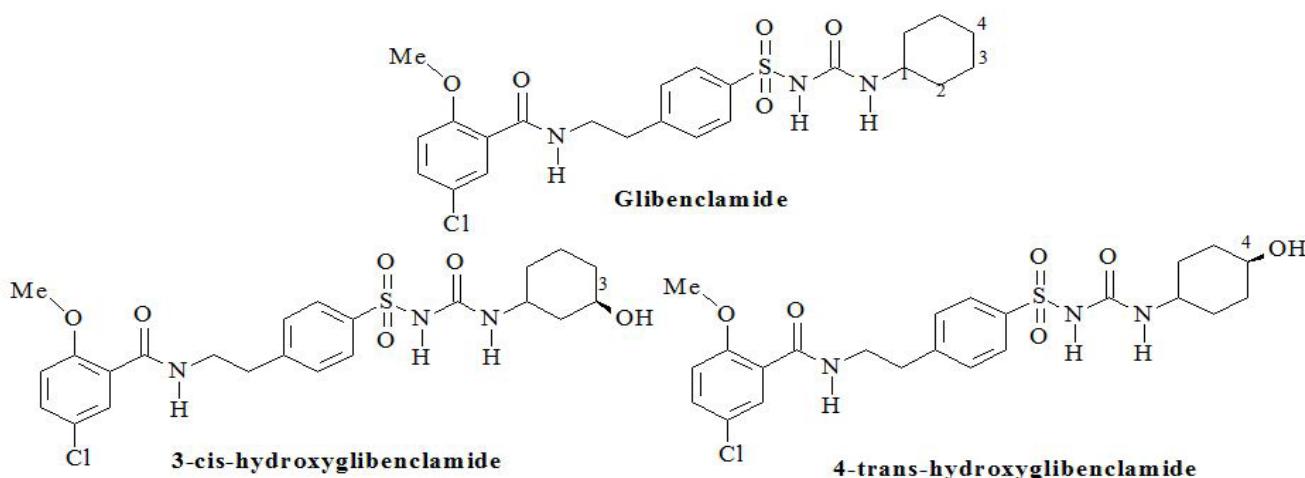
of Cilag GmbH international 6300 Zug Switzerland).

#### Statistical analysis and data evaluation

All the values of blood glucose concentrations were expressed as mean  $\pm$  Standard Error Of Mean (SEM.) and analysis of variance, ANOVA, followed by Student's t-test.

#### Protein binding

Glibenclamide is extensively (~99%) bound by serum proteins (and also the metabolite 4-trans-hydroxyglibenclamide is greater than 97% bound to serum proteins) and is recycled hepatically. Both of these factors contribute to prolonged duration of action of glibenclamide. Protein binding is primarily non-ionic making glibenclamide less likely to displace or be displaced by drugs that are ionically bound to proteins. This makes glibenclamide to rather spend more time in the system.



## Results and Discussion

### Photochemical screening

The results of photochemical studies on the methanol extract of cooked corn silk have been provided in (Table 1)

Photochemical	Observation
Alkaloids	+
Saponins	+
Steroids	+
Flavonoids	+

Cardiac Glycoside	+
Tannins	+
Anthraquinones	+
Carbohydrates	+
Key : + =	positive

**Table 1:** Photochemical analysis of the extract of cooked *Allium cepa*.

### Pharmacological study

The effects of the methanol extracts of the cooked corn silk on fasting blood glucose levels in alloxan monohydrate-induced diabetic albino mice have been given in (Table 2)

Test material	Groups	Blood glucose levels in (mmol/L) sampling time in hours				
		0	24	48	72	96
Glibenclamide (positive control)	Group I	11.5±1.9	8.3±2.0	6.5±2.4	5.4±3.7	4.1±2.1
250 mg/kg	Group II	11.1±3.5	10.1±2.5	9.8±3.0	9.3±2.7	8.7±3.2
500 mg/kg	Group III	9.8±2.1	9.2±1.3	8.8±1.4	7.4±2.2	6.5±1.1
750 mg/kg	Group IV	10.4±1.7	9.3±2.3	8.6±0.8	6.5±2.1	5.4±2.5
Physiological sane (negative control)	Group V	10.5±1.3	10.7±3.0	9.2±2.1	10.6±1.5	11.2±1.5

Values of blood glucose levels are given as mean ± SEM (n = 5), and the values indicated significant (P<0.05) anti-diabetic effect with respect to both positive and negative controls. Differences between groups were also considered significant at 5% level of significance i.e. P ≤ 0.05

**Table 2:** Antidiabetic effect of methanol extracts of cooked Allium cepa in alloxan- induced diabetic mice.

The results of photochemical screening of the methanol extract of cooked corn silk have shown the presence of alkaloids, saponins, steroids, flavonoids, terpenoids, cardiac glycosides, tannins, anthraquinones and carbohydrates. Corn silk reported to be rich in phenolic compounds, particularly flavonoids [6,13]. Another photochemical screening of various extracts of sweet corn silk has shown positive results for flavonoids, alkaloids, phenols, steroids, glycosides, carbohydrates, terpenoids, anthraquinones and tannins [14]. The results of these studies also correlate well with the report given by [15]. The potential medicinal uses of corn silk are related to its pharmacologically active photochemical. The ant diabetic activities of the graded extracts tested in this study could therefore be attributed to the photochemical found in the extract, some of which have been reported to be hyperglycemic by many researchers. For example, flavonoids and tannins have been known to possess ant diabetic activity [16] and in other study flavonoids, sterols, terpenoids, alkaloids, saponins and phenolics have been reported to be ant diabetic principles [17]. A study conducted on *Moringa Oleifera* Leaves reported that the presence of flavonoids, terpenoids, tannins and saponins explains why the plant is used for treating diabetes [16,18]. These secondary metabolites are used ethno-pharmacologically to treat diabetes and hyperglycemia [19]. Two anthraquinones have been isolated from the ethanol extract of rhubarb rhizome and indicated to have anti-diabetic properties [21]. Alkaloids have also been implicated to have hypoglycemic activity and it found that the neem seed kernel powder contained hypoglycemic alkaloids that lowered blood sugar levels in alloxan-induced diabetic rabbits [22]. Results of a study on the polysaccharides isolated from corn silk have shown that 100-500 mg/kg body wt. of the polysaccharides decreased the blood glucose levels and improved the glucose tolerance of streptozotocin (STZ) - induced diabetic rats [1]. This indicated that the corn silk polysaccharides may be useful as an anti-diabetic agent.

The presence of flavonoids in corn silk has already been reported and the pharmacological activities of corn silk also much

related to the flavonoids. Flavonoids are a widely distributed group of plant phenolic compounds which are effective as antioxidants [23]. A recent study showed that the total flavonoids content of the butanol extract of corn silk is in good correlation with the total phenolic content [13]. More recently five different flavonoids as well as some falconoid derivatives have been identified in corn silk [11]. The isolated flavonoids from corn silk were found to act as anti- fatigue and ant diabetic agents [23] and the phenolics and flavonoids were thought to give corn silk its antioxidant properties [24].

The aqueous extract of corn silk has been found to reduce hyperglycemia and use can be made of it as a hypoglycemic food for diabetic patients [25]. In another study the effect of corn silk aqueous extract on glycemic metabolism using an alloxan-induced hyperglycemia in mice, with Xiaoke pills (a Chinese diabetic medicine) as positive control and saline as negative control, revealed that the blood glucose levels of the Xiaoke pill and extract treated groups decreased appreciably [9]. In the present studies comparison of the values of blood glucose levels in the treated and the control groups of alloxan-induced diabetic mice suggested some favourable ant diabetic effect of the cooked corn silks. Statistical analysis using student's t-test revealed that there is a statistically valid difference between the treated and the control groups [26]. The methanol extract exhibited relatively significant (P<0.05) anti-diabetic effect at 72 h and 96 h at the doses of 500 and 750 mg/kg body weight, respectively. From the results it can be suggested that the methanol extract exhibited dose dependent action in a similar mechanism as glibenclamide i.e., by stimulation of surviving  $\beta$ -cells to release more insulin [27].

## Conclusion

The aqueous extract of corn silk has already been investigated and found to reduce hyperglycemia. In the present studies comparison of the values of blood glucose levels in the treated

and the control groups of alloxan-induced diabetic mice suggested some favourable ant diabetic effect of the cooked *corn silks even though, while cooking, some photochemical may be lost into the boiling water*. The experimental results have illustrated that the methanol extract of the cooked corn silk exhibited dose dependent action in a similar mechanism as glibenclamide where surviving beta cells are stimulated to release more insulin.

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