



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 Maydis* 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 antidiabetic potential of methanol extract of cooked, dried and pulverized *Stigma Maydis* was assessed in alloxan-induced diabetic albino mice. Hyperglycemia was 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 was 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 Maydis* 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 phytochemical screening of the methanol extract of cooked *Stigma Maydis* have indicated the presence of cardiac glycoside, steroids, terpenoids, alkaloids, flavonoids, carbohydrates and anthraquinones.

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. parennis* and *Z. mays* L. [1]. 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. [2] The color of the corn silks, at first are usually light green and later turn into red, yellow or light brown. [3]

Zea mays is 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 developed countries. Corn silk as a herb has been reported to have properties of antioxidant [4], anti-prostatitis and antispasmodic. [5] In addition, corn silk is well known in treating infection and cystitis [6], kidney stone and other renal illness. [7] One study, however, has shown that corn silk has no antibacterial activity when investigated against



many bacterial species. [8] Based on folk remedies, corn silk has been used as an oral antidiabetic agent in China for decades. A part from the antidiabetic 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. [9] 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. [10] However, in spite of its widespread use, the mechanisms underlying hypoglycemic activity of corn silk was not yet understood. [11] With regards to toxicity, a study using male and female Wistar rats has confirmed that corn silk is non-toxic in nature. [12] The intake of corn silk as such has also been reported to have no adverse effects [13] and this supports the safety of corn silk for human consumption and its use as an herbal drug for healthcare applications.

Materials and Methods

Plant material

Maize or corn was 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 was used and after the cooking was 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) was kept in an airtight and dried white polyethene bag.

Sample extraction

1000g of the corn silk powder was 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) was 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.

Phytochemical Screening

Phytochemical screening was carried out on the methanol extract of *corn silk* using standard procedures and tests to determine the presence of alkaloids, flavonoids, anthraquinones, steroids, 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 was 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 was allowed). Seven days after, animals with fasting blood glucose level ≥ 7.0 mmol/dL (or ≥ 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 body weight)

Group II: Received Methanol extract at 250 mg/kg body weight

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.

Results and Discussion

Phytochemical Screening

The results of phytochemical studies on the methanol extract of cooked *corn silk* have been provided in Table 1.0.

Table 1.0: Phytochemical Analysis of the Extract of Cooked Corn Silk

| Phytochemicals | Observation |
|-------------------|-------------|
| Alkaloids | + |
| Saponins | + |
| Steroids | + |
| Flavonoids | + |
| Cardiac Glycoside | + |
| Tannins | + |
| Anthraquinones | + |
| Carbohydrates | + |

Key: + = positive

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 2.0.

Table 2.0: Antidiabetic Effect of Methanol Extracts of Cooked Corn Silk in Alloxan-induced Diabetic Mice

| Test Material | Group | Blood Glucose Levels in mmol/L Sampling Time in Hours | | | | |
|--|-------|---|----------------|---------------|----------------|----------------|
| | | 0 | 24 | 48 | 72 | 96 |
| Glibenclamide (positive control) | I | 11.5 \pm 1.9 | 8.3 \pm 2.0 | 6.5 \pm 2.4 | 5.4 \pm 3.7 | 4.1 \pm 2.1 |
| 250 mg/kg | II | 11.1 \pm 3.5 | 10.1 \pm 2.5 | 9.8 \pm 3.0 | 9.3 \pm 2.2 | 8.7 \pm 3.2 |
| 500 mg/kg | III | 9.8 \pm 2.1 | 9.2 \pm 1.3 | 8.8 \pm 1.4 | 7.4 \pm 2.2 | 6.5 \pm 1.1 |
| 750 mg/kg | IV | 10.4 \pm 1.7 | 9.3 \pm 2.3 | 8.6 \pm 0.8 | 6.5 \pm 2.1 | 5.4 \pm 2.5 |
| Physiological Saline (negative control) | V | 10.5 \pm 1.3 | 10.7 \pm 3.0 | 9.2 \pm 2.1 | 10.6 \pm 1.5 | 11.2 \pm 1.5 |

Values of blood glucose levels are given as mean \pm 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 \leq 0.05

The results of phytochemical 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 was reported to be rich in phenolic compounds, particularly flavonoids. [7, 14] Another phytochemical screening of various extracts of sweet corn silk has shown positive results for flavonoids, alkaloids, phenols, steroids, glycosides, carbohydrates, terpenoids, anthraquinones and tannins. [15] The results of these studies also correlate well with that reported. [16] The potential medicinal uses of corn silk are related to its pharmacologically active phytochemicals. The antidiabetic activities of the graded extracts tested in this study could therefore be



attributed to the phytochemicals found in the extract, some of which have been reported to be hyperglycaemic by many researchers. For example, flavonoids, sterols, terpenoids, alkaloids, saponins and phenolics have been reported to be antidiabetic 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. [18, 19] These secondary metabolites are used ethno-pharmacologically to treat diabetes and hyperglycemia. [20] 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 hypoglycaemic activity and it was found that the neem seed kernel powder contained hypoglycaemic 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. [23] 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 was also much related to the flavonoids. Flavonoids are a widely distributed group of plant phenolic compounds which are effective as antioxidants. [24] 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. [4] More recently five different flavonoids as well as some flavonoid derivatives have been identified in corn silk. [12] The isolated flavonoids from corn silk were found to act as anti-fatigue and antidiabetic agents [25] and the phenolics and flavonoids were thought to give corn silk its antioxidant properties. [3] 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. [26] 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 [10]. 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 antidiabetic 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. 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 β -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 antidiabetic effect of the cooked *corn silks even though, while cooking, some phytochemicals 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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