

***In vitro* antidiabetic activity of *Cyperus tegetum* Roxb. root extract**

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ABSTRACT

Cyperus tegetum Roxb is herb growing naturally in marshy land in the humid tropics in India. *Cyperus tegetum* belongs to the family Cyperaceae. The stem of the plant is used for making mats for floor covering as well as decoration. The plant is commonly known as mat stick, madur kathi (Bengali). Methanol extract of *Cyperus tegetum* Roxb was carried out by extraction procedure and phytochemical test was evaluated to confirm the presence of alkaloid, glycosides, proteins, amino acids, phenolic compounds, flavonoid and tannins in extract. Diabetes is a clinical syndrome characterized by hyperglycemia due to absolute or relative deficiency of insulin. Recent decades have experienced a sharp increase in the incidence and prevalence of diabetes mellitus. The objective of present work was to evaluate the antidiabetic activity of petroleum ether and aqueous extract of root of *Cyperus tegetum* Roxb. The samples were studied for their effect on inhibition of alpha amylase. α -Amylase inhibition was in a dose dependent manner.

Keywords: *Cyperus tegetum* Roxb, Methanolic extract, Phytochemicals, Antidiabetic activity.

INTRODUCTION

Natural products, such as plants extract, either as pure compounds or as standardized extracts, provide unlimited opportunities for new drug discoveries because of the unmatched availability of chemical diversity [1]. The plant *Cyperus tegetum* Roxb. belonging to the family Cyperaceae, a glabrous and robust perineal sedge found throughout India up to an altitude 1800m (Fig.1). The plant is commonly known as mat stick, madur kathi (Bengali) and cultivated as an economic crops in west Medinipur district of West Bengal and traditionally used by the tribal people for the treatment of cachexia, atrophy and snake bite [2].



Figure.1 Root of CT

In recent years, substantial efforts have been made to identify efficient antidiabetic agents, as synthetic hypoglycemic agents are associated with many disorders and their effectiveness is limited and prone to a variety of side effects. Decreased physical activity, increasing obesity, stress and changes in food consumption have been implicated in this increasing prevalence in the past two decades [3,4].

Diabetes is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion. Food habits and genetic factors are responsible for diabetes [5]. A study revealed that urbanization of rural India has doubled the rate of diabetes. In India, between 1995 and 2025 the number of people with diabetes is projected to rise from 19 to 57 million. As per national urban diabetic survey, the incidence of diabetes was found to be high. This paper provides details in extraction, common phytochemical screening assay and the in-vitro anti-diabetic activity. Despite considerable progress in the management of diabetes mellitus by conventional synthetic drugs, research work on natural agents has greatly increased all over the world. Plants used in folk medicine to treat diabetes mellitus represent a viable alternative for the control of this disease. The present experiment was carried out to evaluate in-vitro antidiabetic activity of methanol extract and aqueous extract of rhizomes of this plant [6].

MATERIALS AND METHODS

Plant materials

The plant *Cyperus tegetum* Roxb (Family: Cyperaceae) was collected from the cultivated land of Paschim Medinipur, West Bengal. The rhizomes were washed, dried at room temperature and then grounded in a mixer grinder to make powder form.

Extraction of plant materials

The extraction process was carried out to get the clear solution so that the photochemical test to be carried out and in vitro antidiabetic activity to be carried out. The powdered rhizomes were extracted by extraction method using a methanol. The procedure was carried out by taking 5g of matstick powder and it was dissolved in 500ml of methanol in the Soxhlet apparatus the experiment was carried out for 6hrs till the clear solution is obtained. After the extraction vacuum filtration was carried out and filtrate was collected [2].

Preliminary phytochemical analysis

The phytochemical tests were performed using various reagents. The MeOH extract was tested for the presence or absence of alkaloid, glycosides, tannins, steroids, reducing sugars, proteins and amino acids, phenolic compounds and flavonoids [2].

In-vitro methods employed in Anti-diabetic studies

Inhibition of alpha amylase enzyme

The assay was carried out following the standard protocol with slight modifications. Starch (2 mg) was suspended in 0.2 mL of 0.5M Tris-HCl buffer (pH 6.9) containing 0.01 M CaCl₂ (substrate solution). The test tubes containing substrate solution were boiled for 5 min and then pre incubated at 37°C for 5 min. Methanol extract *Cyperus tegetum* roxb was dissolved in DMSO in order to obtain concentrations of 250, 500, 750 and 1000 µg/mL. Then, 0.2 mL of plant extract of particular concentration was added to the tube containing the substrate solution. In addition, 0.1 mL of alpha-amylase in Tris-HCl buffer (2 units/mL) was added to the tube containing the plant extract and substrate solution. The reaction was carried out at 37°C for 10 min. The reaction was stopped by adding 0.5 mL of 3,5-dinitrosalicylic acid. The reaction mixture was centrifuged at 3000 rpm for 5 min at 4°C. The absorbance of resulting supernatant was measured at 595 nm using UV-VIS spectrophotometer. Metamorphin, a known α-amylase inhibitor was used as a standard drug. The experiments were repeated twice [7].

The α-amylase inhibitory activity was calculated by using following formula:

$$\text{Inhibition of } \alpha\text{-Amylase (\%)} = \frac{\text{Abs sample} - \text{Abs control}}{\text{Abs sample}} \times 100$$

RESULTS AND DISCUSSION

The *Cyperus tegetum* Roxb was determined by prescribed phytochemical tests, which indicated the absence of steroids, glycosides and alkaloids, flavonoids, tannins, saponin, glycosides, proteins and reducing sugar are present (Table 1).

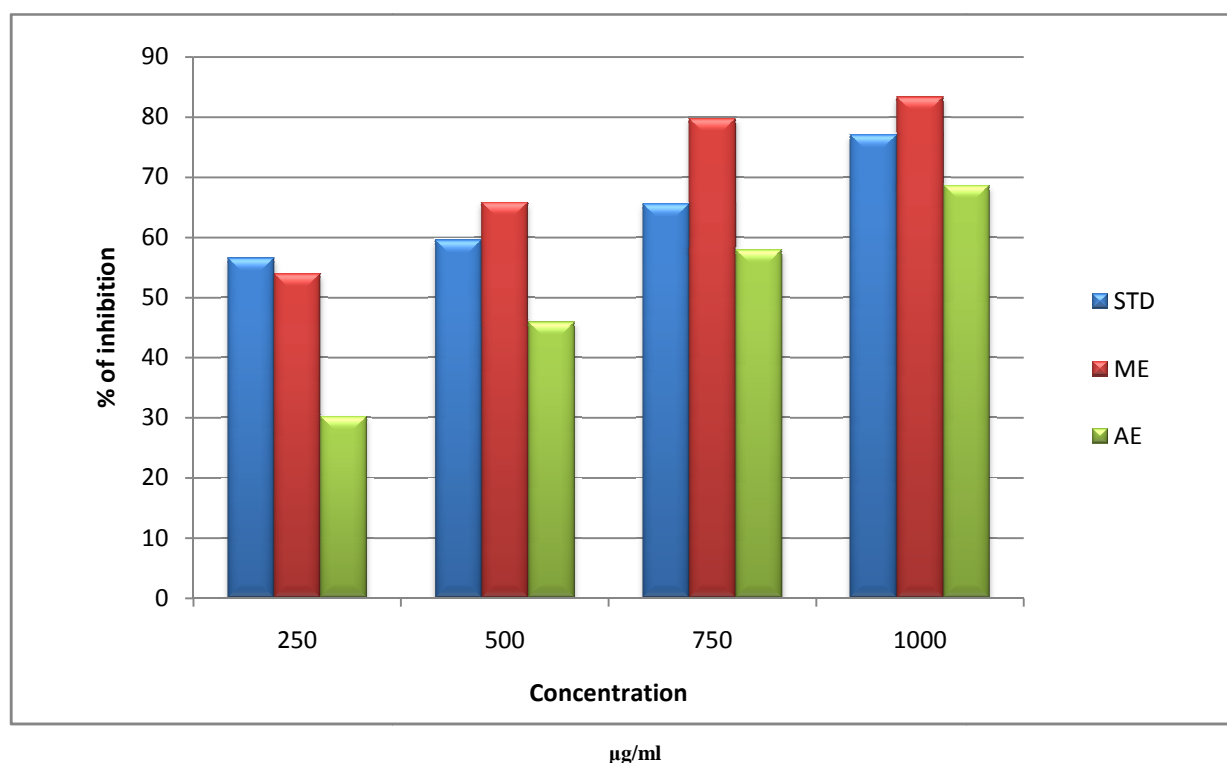
Table 1. Qualitative phytochemical evaluation of the *Cyperus tegetum* Roxb. Extracts

Phytoconstituents	Test performed/reagents used	Results
Alkaloids	Meyers test	+
	Dragendorff's test	+
	Hangers test	+
Steroids	Liebermann-Burchard test	-
Flavonoids	Shinoda test	+
Tannins	Ferric chloride	+
	Lead acetate	+
Saponin	Test for stable foam	+
Glycosides	Borntager test	-
Proteins and amino acids	Ninhydrin test	+
Reducing sugar	Benedict and Fehling's test	+

Presence (+) or absence(-)

Alpha- amylase inhibition method

Alpha amylase is an enzyme that hydrolyses alpha-bonds of large alpha linked polysaccharide such as glycogen and starch to yield glucose and maltose. Alpha amylase inhibitors bind to alpha- bond of polysaccharide and prevent break down of polysaccharide in mono and disaccharide. As the result shows methanol and aqueous extract of *Cyperus tegetum* Roxb shows significant activity as compared to metamorphin standard drug, and 500 and 1000 µg/ml concentration of methanol extract shows greater activity than Metamorphin (Fig 2).

Figure 2. Percentage of α-amylase inhibitory effects of acarbose (standard drug), methanol extract and aqueous extract *Cyperus tegetum* Roxb.

Many herbal extracts have been reported to have antidiabetic activities and are used in Ayurveda for the treatment of diabetes. Herbal extracts have been used directly or indirectly for the preparation of many modern medicines. In this study, an *in vitro* inhibitory effect of different extracts of *Cyperus tegetum* Roxb on alpha- amylase activities was evaluated. Antidiabetic activities of *Cyperus tegetum* Roxb have been reported with its roots.

We compared %inhibition of α-amylase inhibitory effects of methanol extracts of *Cyperus tegetum* Roxb and aqueous extract with Metamorphin. At the same time, methanol extracts showed appreciable α-amylase inhibitory effects when compared with metamorphin. It may be due to the presence of more chemical constituents such as, terpenes, tripenes, flavonoids (quercetin, quercetrin, rutin), and alkaloids in the ethanol extracts. The plant-based α-amylase inhibitor offers a prospective therapeutic approach for the management of diabetes. In this study, whole plants of *Cyperus tegetum* Roxb showed considerable α-amylase inhibitory effects when compared with metamorphin.

CONCLUSION

First of all the methanol extraction was carried out to get the clear solution so that all the test should be carried out in proper manner after doing the methanol extraction photochemical test was performed it was found that the absence of steroids, glycosides and alkaloids, flavonoids, tannins, saponin, glycosides, proteins and reducing sugar are present. The in-vitro anti-diabetic application of *Cyperus tegetum Roxb* was carried. It showed an appreciable α -amylase inhibitory effects. This study supports the ayurvedic concept that *Cyperus tegetum Roxb* could be useful in management of diabetes.

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