

In-Vitro Anti-Diabetic Activity of *Ficus krishnae* Stem Bark Extracts

Amarvani P Kanjekar, Aruna L H, Ramesh L Londonkar*

Department of Biotechnology, Biopharmaceutical and Nanobiotechnology Laboratory, Gulbarga University, Kalburagi, India

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ABSTRACT

Ficus krishnae (Moraceae) is one of the folklore medically important native plant to India. It has been used extensively by ayurvedic practitioner in India to treat various ailments such as ulcers, vomiting, fever, inflammations, leprosy, syphilis, biliousness, dysentery and inflammation of liver. The present study aims the screening of *Ficus krishnae* stem bark extracts for in-vitro anti-diabetic study. The in vitro alpha-amylase inhibitory study was performed using different concentration of extract and compared with a standard drug. The results reveal that, there was a dose dependent increase in the percentage inhibitory activity against intestinal enzymes by petroleum ether, chloroform and aqueous extracts. Our findings suggests that the petroleum ether extract and acarbose have an efficient anti-diabetic activity.

Keyword: *Ficus krishnae*, soxhlet extract, anti-diabetic, alpha amylase.

INTRODUCTION

Medicinal plants are the good source in health related therapeutic aid for alleviating diseases of humankind. Over the 2500 years, there has been a strong traditional system of medicines such as Chinese, ayurvedic and unani born and practiced more in eastern continent. These traditional system of medicine is being practiced to treat about 80% of the human population in the developing countries for their primary health remedy¹. International diabetes federation has estimated that in 2010 year, it has risen to 285 million, contributing 6.4% of the world adult population and it is predicted that in 2030 the number of people with diabetes mellitus (DM) will rise to 438 million. India has declared as the Diabetes capital of the world. Currently 40.9 million people in India suffering from DM and in 2030 there may be 79.44 million diabetes patients may found in India². Diabetes mellitus is one of the syndrome characterized by hyperglycemia, caused due to decrease in insulin production by pancreatic islet cells which leads to increase in blood glucose level³. Diabetes insipidus is another diabetic condition caused due to deficiency of antidiuretic hormone (ADH) or vasopressin hormon secreted by the posterior pituitary gland. Symptoms of diabetes include weight loss, polyuria (excess urine), polydipsia (thirst) and polyphagia (excessive food)⁴. The genus *Ficus* has high importance; many species of this genus are used in traditional medicine. Various tribes, sub-tribes and races of the great assamese society have valued several species of *Ficus* in their own way. As the genus is rich in diversity, this region possesses tremendous scope of exploitation of its members, as many species belonging to this genus have carried good properties which will benefit the mankind⁵. *Ficus krishnae* belongs to the family Moraceae is known by different names Krishna fig, Krishna's butter cup (in English) &

Makkhan Katori (in Hindi) and native to India. It is large, fast growing, evergreen tree grow up to 30 m tall, with spreading branches and aerial roots^{6,7,8}. The unique structure of the tree is that the leaves have a pocket-like fold at the base. All parts of this plant are useful in treatment of ulcers, vomiting, vaginal complaints, fever, inflammations and leprosy. It has also proved as an anti-diabetic and antihyperlipidemic agent^{9,10}. However; no in-vitro anti-diabetic works of stem bark of this plant has been reported. Hence, this study will be useful in documentation and forming the guidelines for further study on *Ficus krishnae*.

MATERIALS AND METHODS

Collection of plant materials

Ficus krishnae stem barks were collected from Dev Dev Vana botanical garden, Bidar, Karnataka. India, allowed to dry in shade for 2-4 weeks, after drying the bark was powdered and stored in airtight container for further use.

Soxhlet extraction

The bark powder (100 g) was successively extracted by hot soxhlet extraction with Petroleum ether, chloroform and aqueous solvent (600 mL) for 6, 8 and 6 hours respectively. After extraction the extracts were stored for future use.

Evaluation of in vitro anti-diabetic activity

Inhibition of alpha amylase enzyme activity method

A total of 500 µl of test samples (Petroleum ether, chloroform and aqueous extracts) and standard drug (50 - 1000 mg/ml) were added to 500 µl of 0.20 mM phosphate buffer (pH 6.9) containing α-amylase (0.5 mg/ml) solution and incubated at 25 °C for 10 mins. After this, 500µl of a 1 % starch solution in 0.02 M sodium phosphate buffer (pH 6.9) was added to each tube. The reaction mixtures were then incubated at 25 °C for 10 mins. The reaction was stopped with 1.0 ml of 3, 5 dinitrosalicylic (DNS) acid

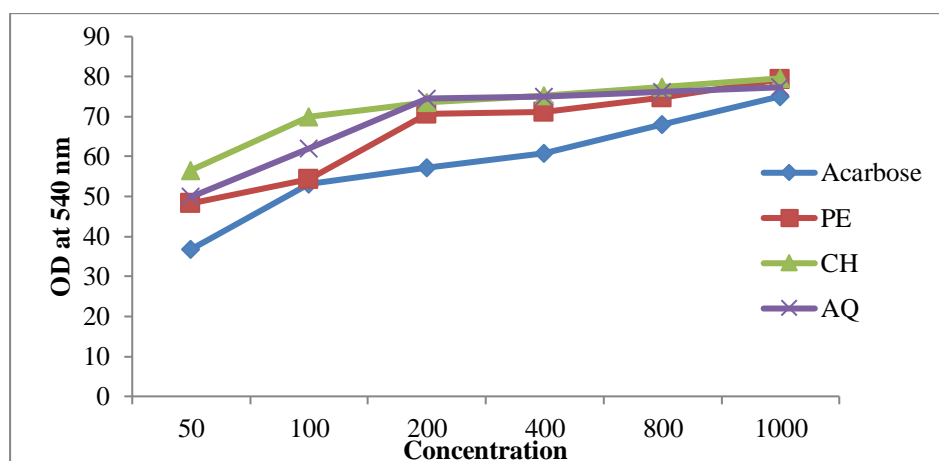


Figure 1: In Vitro alpha amylase inhibition by acarbose, PE, CH and AQ extracts of *Ficus krishnae* stem bark extracts.

colour reagent. The test tubes were then incubated in a boiling water bath for 5 min, cooled to room temperature. The reaction mixture was then diluted after adding 10 ml distilled water and absorbance was measured at 540 nm¹¹. Acarbose was used as a standard drug for assay. The control samples represent 100 % enzyme activity and were prepared without any plant extract. Each test was performed for six times and the mean absorption was calculated the percentage of α - amylase inhibition.

% Inhibition was calculated according to the formula:

$$\% \text{ Inhibition} = \frac{A_{540} \text{ control} - A_{540} \text{ Sample}}{A_{540} \text{ control}} \times 100$$

RESULT AND DISCUSSION

Fig.1 reveal that the PE, CH and AQ extracts of *F.krishnae* have showed a significant inhibition in the activity of α -amylase enzyme, when compared to standard acarbose. The inhibitory activity increases with increase in concentration of all different extracts and acarbose. At the minimum concentration of 50µg/ml of *F.krishnae* three extracts and acarbose have showed 48.28%, 56.59%, 49.90% and 36.80% respectively. At maximum concentration of 1000 µg/ml extracts and acarbose have showed 79.28%, 79.55%, 77.30% and 75.06% respectively. Whereas *F. glomerata* aqueous extracts of gum indicated 82.76 % at concentration of 1000µg/ml¹². Alpha amylase is a hydrolysis enzyme that acts on alpha-bonds of large linked polysaccharide such as glycogen and starch to yield glucose and maltose. Alpha amylase inhibitors bind to alpha bonds of polysaccharides that prevent the degradation of mono and disaccharides¹³.

CONCLUSION

In present investigation the presence of various bioactive compounds in the petroleum ether extract of *Ficus krishnae* plant are responsible for anti- diabetic property compared to other extracts that have showed a good inhibition of alpha-amylase enzyme activity. Further studies are required for the isolation of pure compound which is responsible for anti-diabetic activity.

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AUTHORS' CONTRIBUTORS

The authors of this study claim sole responsibility for the concepts included herein. All authors read and approved the final manuscript.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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