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Original Article

Anti-diabetic activity of Sesbania grandiflora - alpha amylase inhibitory effect

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ABSTRACT

Introduction: Diabetes mellitus is characterised by high blood sugar levels caused due to ineffectiveness of the insulin produced at the peripheral tissues. Due to the side effects of insulin and oral hypoglycaemic agent, demand to use natural products with anti-diabetic activity has been increased recently. Materials and Methodology: Sesbania grandiflora extract used in the study is obtained from Green Chem Herbal Extracts and Formulations, Bengaluru, India. Acarbose, Dinitrosalicylic acid, DMSO is obtained from Sigma Aldrich Co. India. α-Amylase enzyme were procured from HiMedia, Mumbai. In vitro amylase inhibition was studied by the method of Bernfeld and the percentage inhibition of α-Amylase enzyme was calculated. Results: A maximum inhibition of 81% was observed at 1000µg/ml which was compared with that of standard Acarbose that showed 93% inhibition. The IC_{50} of the extract was found to be 50.95µg/ml and for Acarbose 34.83 uM. Conclusion: The study showed that this plant species exhibited anti-diabetic property and can be used to explore possibilities of developing a natural drug for the management of diabetes mellitus rather than using synthetic drugs which produce side effects.

Keywords: Sesbania grandiflora, anti-diabetic activity, glycation, diabetes mellitus, alpha amylase.

Introduction

Sesbania grandiflora commonly known as vegetable hummingbird is a small tree in the genus Sesbania. The tree thrives under full exposure to sunshine and is extremely frost sensitive [1]. This fast growing tree is cultivated in many parts of India and Sri Lanka and has many traditional uses [2]. The leaf extract inhibits the formation of advanced glycation end-products. It contains linolenic acid and aspartic acid, which were found to be the major compounds responsible for the anti-glycation potential of the leaf extract [3,4-8].

The flowers have an astringent action on the body used in liver and spleen disorders, rhinitis, night blindness, abdominal pain etc. and helps in the detoxification of the body. The leaves possess tonic properties which is helpful in worm infestations and bleeding disorders. The leaf paste is also used to treat oral and throat problems. The root bark is externally applied to reduce pain and inflammation in arthritis and gout. This plant also possesses anti-anxiolytic and anti-proliferative properties against cancer cells in humans [5,9,10].

Diabetes mellitus is a complex chronic condition that is a major source of ill health worldwide. It is characterised by hyperglycaemia and imbalance of carbohydrate, protein, and fat metabolisms [6,11-15]. This may be attributed to insulin inactivity or resistance, as a direct result of destruction or dysfunction of the beta-cells of the pancreas [7,16,17].

India is emerging as the diabetic capital of the world. Facing the burden of the consequences that the disease brings; it is currently estimated that every fifth diabetic in the world is an Indian [8,18,19]. Moreover, statistical projection about India suggests that the number of diabetics will rise from 15 million in 1995 to 57 million in the year 2025, the highest number of diabetics in the world. Reasons for this rise include increase in sedentary lifestyle, consumption of energy-rich diet, obesity, higher life span, etc. [9,20]. Inhibition of α-amylase enzymes involved in the digestion of carbohydrates, can significantly decrease the postprandial increase of blood glucose after a mixed carbohydrate diet and therefore can be an important strategy in

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the management of postprandial blood glucose level in type 2 diabetic patients and borderline patients [10,21,22].

Control of postprandial hyperglycaemia is the key to the management of type 2 diabetes mellitus and its associated complications. Conventional approach to this growing epidemic is associated with many side effects, and moreover, it is expensive and inaccessible to certain communities [11, 12,23]. Due to this, evaluation of plant products is of growing interest since they contain many bioactive substances. Currently there is renewed interest in plant-based medicines modulating physiological effects in the prevention and cure of diabetes and obesity [13,24,25].

Hence the aim of this research is to scientifically investigate the anti-diabetic activity of Sesbania grandiflora in order to develop a natural drug instead of a synthetic one.

Materials and Methodology

Plant materials:

Sesbania grandiflora extract used in the study obtained from Green Chem Herbal Extracts and Formulations, Bengaluru, India.

Chemicals used:

Acarbose, Dinitrosalicylic acid, DMSO is obtained from Sigma Aldrich Co. India. α-Amylase enzyme was procured from HiMedia, Mumbai.

In-vitro α-Amylase inhibitory assay (Bernfeld 1955):

In vitro amylase inhibition was studied by the method of Bernfeld. In brief, 100µl of the different concentrations of Sesbania (1,3,10,30,100,300 and 1000 µg/ml) was allowed to react with 200 µl of α-Amylase (Hi media RM 638) and 100µl of 2mM of phosphate buffer (pH- 6.9). After 20-minute incubation, 100 µl of 1% starch solution was added. For the controls, the same process was done that 200 µl of the enzyme was replaced by buffer. Acarbose had the role of being used as the reference standard. 500 µl of Dinitrosalicylic acid reagent was added to both control and test after incubation for 5 minutes. 5 minutes was the period for keeping them in boiling water. Using spectrophotometer, the absorbance was recorded at 540 nm and using the following formula, the percentage inhibition of α-Amylase enzyme was calculated:

$$\%\text{inhibition} = [(\text{Control} - \text{Test})/\text{Control}] * 100$$

Suitable reagent blank and inhibitor controls were carried out simultaneously.

Results

α– amylase inhibitory activity:

The results of the present study exhibited strong α– amylase inhibitory activity of the Sesbania extract which is compared with standard Acarbose. As shown in Figure 1, maximum inhibition of 81% was observed at a concentration of 1000µg/ml by Sesbania extract, which was compared with that of standard Acarbose that showed inhibition of about 93%. The IC_{50} of the extract was found to be 50.95µg/ml and for Acarbose 34.83µM.

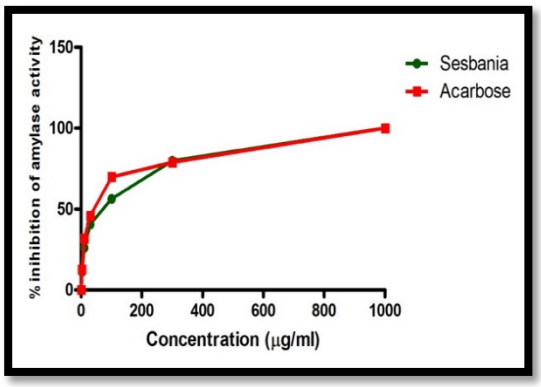

Figure 1. alpha amylase inhibitory assay

Discussion

Many herbal extracts are being used directly or indirectly for the preparation of many modern medicines. In this study, an in vitro alpha-amylase inhibitory effect of Sesbania grandiflora is performed to evaluate the anti-diabetic activity. It was seen in figure 1 that this plant had lowered the blood glucose level by showing a maximum inhibition of 81%. This is due to the presence of lignin, terpenes, tripenes and other constituents [26]. However, acarbose being the standard showed 93% inhibition. When the IC_{50} values of Sesbania and acarbose is compared, it was found that it's more than acarbose. Alpha-amylase catalyses the hydrolysis of alpha-1,4-glycosidic linkages of starch, glycogen and various oligosaccharides. The disaccharides will be broken down to simple sugars by alpha- glucose daze making it available for intestinal absorption. The inhibition of their activity in the digestive tract of humans is considered to be effective tool to control diabetes [27].

Other studies regarding the α – amylase inhibitory activity were also performed. It showed that D. Villosa showed an increase ranging from 10.71 to 71.88% with a concentration range of 1.5µg/ml - 1000µg/ml and the IC_{50} value was calculated as 72.44 µg / ml when compared with acarbose, IC_{50} value 83.23 µg/mL [28]. IC_{50} values of α-amylase inhibitory effects of ethanol and hexane extracts of P. amarus was also found. The hexane extract (at a concentration 100 µg/mL) showed 75.32% of α-amylase inhibitory activity with IC_{50} value 48.92 µg/mL. Both ethanol and hexane extracts showed appreciable α-amylase inhibitory effects when compared with acarbose [29]. In another study, a weak α-amylase inhibitory activity of 20% (v/v) extract of A. paniculata was seen. It showed similar α-glucosidase and amylase inhibitory effect. The IC_{50} values show that 20% (v/v) eth- anolic extract has a lower potency and has a preference for α-glucosidase over α-amylase [30].

Therefore, Sesbania grandiflora showed considerable α-amylase inhibitory effects when compared with acarbose. Several studies of anti-anxiolytic and anti-inflammatory properties have been performed too. From the above mentioned results, this extract can be used as an anti-diabetic agent and incorporated into the pharmacological preparations.

C

onclusion

A good understanding of the anti-diabetic activity of the Sesbania grandiflora has been created and now is being utilised effectively for other research studies in the pharmacological arena. These newer discoveries will be very helpful in the treatment of one of the most common metabolic disorder- Diabetes. Thus, establishing a promising future in this field.

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