

**PHYTOCHEMICAL ANALYSIS AND IN VITRO ANTIDIABETIC ACTIVITY OF
ETHANOL EXTRACT OF IPOMOEA STAPHYLINA LEAVES**

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ABSTRACT

Diabetes mellitus is a metabolic disorder which is characterized clinically as hyperglycaemia due to the deficiency of insulin secretion. The intestinal digestive enzymes β -glucosidase and α amylase are playing vital role in digestion process. Anti-diabetic therapeutic approach reduces the post prandial glucose level in blood by the inhibition of these enzymes. In recent years there is a fast increase in the occurrence of diabetes mellitus. In the present study *Ipomoea Staphylina* was screened for in vitro anti-diabetic activity and phytochemical bioactive compounds analyses were studied. The assay result suggests that the presence of bioactive compounds, could be responsible for the versatile medicinal properties of this plant including diabetes, the extract exhibit the dose-dependent increase in inhibitory effect on α -glucosidase (up to 84.14%), and α amylase enzyme up to (98.11%). The current study proves that the anti diabetic activity of ethanol extract of *Ipomoea Staphylina* leaves by *in vitro* studies

Keywords: *Ipomoea Staphylina*, α -glucosidase, α –amylase, phytochemical and antidiabetes.

Introduction

Herbal medicine and alternative medicines used for through the world. The herb product derived as medications and it is used for various treatment on high blood pressure, diabetes and other illness (Monica Loizzo *et al.*, 2008). It's is a word borrowed from the Greek word meaning a siphon because the affected individuals experience polyuria and pass water like a siphon.

There are more no of diabetes mellitus cases has been increasing through over the world for the recent years. The year of 2000, the world health organization estimated a total of 171 million of people with diabetes mellitus from the global population, and this report projected to increase to 366 million by 2030 (Wild *et al.*, 2004). Diabetes affects mainly the developing countries like India. Indeed, India presently has the largest number of diabetic patients in the world and has been infamously dubbed as the 'diabetic capital of the world' (Adate N 2007). Not now no treatment has pursued for permanent cure of diabetes. In spite of many drawbacks like anorexia, brain atrophy and fatty lever etc., still insulin therapy treatment is the only accepted treatment for diabetes.

Diabetes mellitus is a one of the most common disease and affecting the citizens of the countries. All over the world, its estimated 25% of the population is affected by this disease. Diabetes mellitus is caused by the abnormality of carbohydrate metabolism it is linked to low blood insulin level of target organs (Arumugam *et al.*, 2013). It is a group of chronic and metabolic disorders its poses a major challenge through over the world. (Ayesha Noor *et al.*, 2013). It's a diverse group of metabolism disorders it's mainly affecting the following metabolism carbohydrate, fat, and protein (Manikandan *et al.*, 2013). It is a complex, multifactorial disease which affects the quality, quantity, and style of an individual's life (Anil Kamboj *et al.*, 2013).

The present study was undertaken to evaluate the phytochemical analysis and *in vitro* antidiabetic activity of ethanol extract of *Ipomoea Staphylinaleaves*.

Materials and Methods

Plant material and extraction

The fresh leaves of *Ipomoea staphylina* were collected locally and authenticated by the Department of Botany, St. Joseph College, Trichy. The shade dried *Ipomoea*

staphylina leaves were powdered mechanically and stored in an air tight container. The extraction was carried out by hot percolation method using Soxhlet apparatus. The solvent used was ethanol. About 250 gm of powder was extracted with 500 ml of ethanol. The extract was concentrated to dryness under controlled temperature 40- 50°C. The percentage yield was found to be 15.65%. The extract was preserved in refrigerator till further use.

Phytochemical Screening

For preliminary phytochemical analysis, the freshly prepared ethanol extracts of *Ipomoea Staphylina* leaves were tested for the phytoconstituents by using standard phytochemical procedures (Evans, W.C and Evans, T. 2003).

In vitro methods employed in antidiabetic studies

Inhibition of alpha-amylase enzyme

A starch solution (0.1% w/v) was obtained by stirring 0.1g of potato starch in 100 ml of 16 mM of sodium acetate buffer. The enzyme solution was prepared by mixing 27.5 mg of alpha-amylase in 100 ml of distilled water. The colorimetric reagent is prepared by mixing sodium potassium tartarate solution and 3, 5 di-

nitrosalicylic acid solution 96 mM. Both control and plant extracts were added with starch solution and left to react with alpha-amylase solution under alkaline conditions at 25°C. The reaction was measured over 3 minutes. The generation of maltose was quantified by the reduction of 3, 5 di-nitrosalicylic acid to 3-amino-5-nitro salicylic acid. This reaction is detectable at 540 nm (Malik and Singh, 1980).

Inhibition of alpha-glucosidase enzyme

The inhibitory activity was determined by incubating a solution of starch substrate (2 % w/v maltose or sucrose) 1 ml with 0.2 M Tris buffer pH 8.0 and various concentration of plant extract for 5 min at 37°C. The reaction was initiated by adding 1 ml of alpha-glucosidase enzyme (1U/ml) to it followed by incubation for 40 min at 35°C. Then the reaction was terminated by the addition of 2 ml of 6N HCl. Then the intensity of the colour was measured at 540nm (Krishnaveniet *al.*, 1984).

Calculation of 50% Inhibitory Concentration (IC₅₀)

The concentration of the plant extracts required to scavenge 50% of the radicals (IC₅₀) was calculated by using the percentage scavenging activities at five different concentrations of the extract.

Percentage inhibition (I %) was calculated by

$$I \% = (Ac-As)/Ac \times 100, \text{ (Shaiet al., 2010)}$$

where Ac is the absorbance of the control and As is the absorbance of the sample.

Result:

Phytochemical Analysis:

Table 1 shows the phytochemical analysis of ethanol extract of *Ipomoea Staphylinaleaves*. The ethanol extract of *Ipomoea Staphylinarevealed* the presence of Carbohydrate, protein, aminoacids, vitamins, alkaloids, Flavonoids, steroids, terpenoids, phenols, Phytosterols, Phlobatannins, coumarins and tannins.

Table 1: Phytochemical analysis of ethanol extract of *Ipomoea Staphylinaleaves*

S.No	Phytochemicals	Ethanol extract
1	Carbohydrate	+
2	Protein	+
3	Test for Vitamins	+
4	Amino Acids	-
5	Inorganic elements	+
6	Flavonoids	-
7	Glycosides	-
8	Test for Tannins	+
9	Saponins	-
10	Phlobatannins	+
11	Phytosterols	+

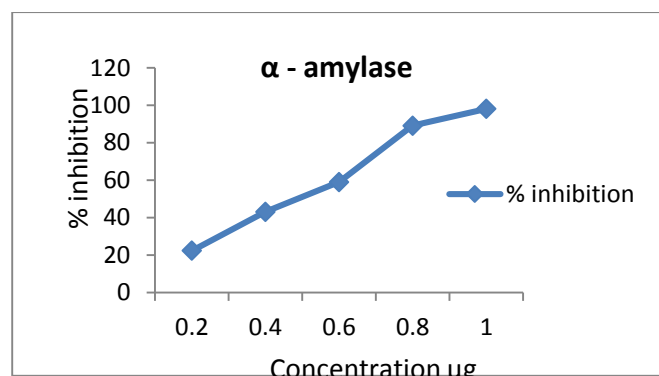
12	Alkaloids	+
13	Phenolic compounds	+
14	Steroids	+
15	Test for coumarins	+
16	Test for quinones	+

In Vitro*antidiabetic activity of *Ipomoea Staphylinaleaves

Inhibition of α -amylase enzyme:

*In vitro*antidiabetic activity of alpha-amylase enzyme of *Ipomoea Staphylinaleaves* was showed in figure 1. There was a dose dependent increase in percentage inhibitory activity alpha-amylase enzyme. At a concentration of 0.2 ml of plant extract showed a percentage inhibition of 22.31% and 1.0 ml of plant extract showed inhibition of 98.11%.

Figure 1: *In vitro*antidiabetic activity of alpha-amylase enzyme

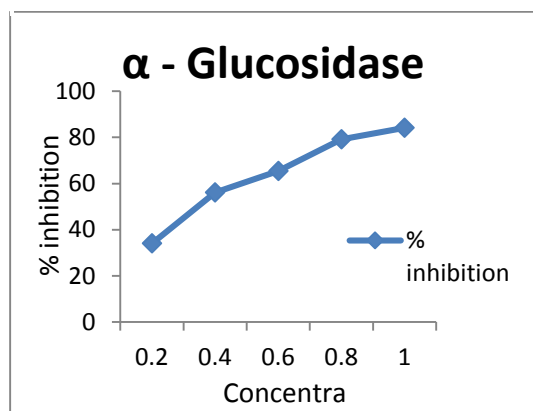


Inhibition of α -glucosidase enzyme:

*In vitro*antidiabetic activity of alpha-glucosidase enzyme of *Ipomoea Staphylinaleaves* was showed in Figure 2. There was a dose dependent increase in

percentage inhibitory activity alpha-glucosidase enzyme. At a concentration of 0.2 ml of plant extract showed a percentage inhibition of 34.12% and 1.0 ml of plant extract showed inhibition of 84.14%.

Figure 2: In vitro antidiabetic activity of alpha glucosidase enzyme



Discussion:

The major qualities of using herbal medicines seem to be their supposed efficacy, low incidence of serious adverse effects and low cost (PrasannaShama and Shastry, 2012). More than 25% of world population is affected from this disease. It's caused by the abnormality of carbohydrate metabolism deficiency of insulin secretion. Herbal drugs with anti diabetic activity it's commercially formulated as a medicine. These plants provide a potential source of hypoglycaemic agents because plant compound is used for prepared for drugs. Most of the Indian plant is used for various

types of diabetes and reported (Kavishankaret *al.*, 2011). At present extracts from more than 150 plants are used to treat diabetes. This leads to an active gain in popularity of using herbal drugs to control this disease. Many natural sources have been investigated with respect to suppression of glucose production from the carbohydrates in the gut or glucose absorption from the intestine (Matsui, T 2001).

Alpha -amylase catalyses the hydrolysis of alpha-1,4-glycosidic linkages of starch, glycogen and various oligosaccharides. Alpha-glucosidase further breaks down the disaccharides to simple sugars, readily available for intestinal absorption. The inhibition of their activity in the digestive tract of humans is considered to be effective tool to control diabetes (Hara and Honda 1990).

The present phytochemical screening of methanol extract of *Ipomoea Staphylina leaves* confirmed the presence of bioactive compounds like Carbohydrate, protein, aminoacids, vitamins, alkaloids, Flavonoids, steroids, terpenoids, phenols, Phytosterols, Phlobatannins, coumarins and tannins. Any of these secondary metabolites, singly or in combination with others could be responsible for the *in*

vitro antidiabetic activity. The major outcome of this study reveals that ethanol extract of *Ipomoea Staphylina* leaves have exhibited potent inhibition of alpha-amylase and alpha-glucosidase enzyme activity

Conclusion:

The present study was formulated to understand the phytochemical analysis and *in vitro* antidiabetic properties of *Ipomoea Staphylina* leaves. In this study the phytochemical analysis revealed the presence of Carbohydrate, protein, aminoacids, vitamins, alkaloids, Flavonoids, steroids, terpenoids, phenols, Phytosterols, Phlobatannins, coumarins and tannins. Further studies are required to elucidate whether *Ipomoea Staphylina* possess *in vivo* antidiabetic activity. The plant showed significant inhibition activity, so further compound isolation responsible for the activity has to be done for the usage of antidiabetic agent.

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