

# Comparative Study on Anti-Diabetic Effect Between *Cissus Quadrangularis* and *Trigonella Foenum-Graecum*

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## ABSTRACT

**Background:** The increase in the prevalence of diabetes is parallel with an increase in associated risk factors such as being overweight or obese. The revolution of traditional medicine is growing nowadays. Natural products are found to be the better alternative in the current scenario for replacing routine drugs.

**Aim:** The aim of this study is to compare the antidiabetic activity of *Cissus Quadrangularis* and *Trigonella Foenum-graecum*.

**Methods:** The  $\alpha$ -amylase inhibition assay was performed using the 3,5-dinitrosalicylic acid (DNSA) method. A sample of *Cissus Quadrangularis* and *Trigonella Foenum-graecum* extract at five different concentrations were made and a positive control sample was prepared using metformin (500mg) (1  $\mu$ g/ml) and the reaction was performed similarly to the reaction with plant extract as mentioned above.

**Results:** As the concentration of the sample increased from 20% to 100% microgram/milliliter, there was a noticeable trend of increased inhibition percentage, indicating a dose-dependent response. It was found that fenugreek has better inhibition of  $\alpha$ -amylase when compared to pirandai even at the lowest concentration.

**Conclusion:** Although *Cissus quadrangularis* and *Trigonella foenum-graecum* both have antidiabetic characteristics; the evidence indicates that *Trigonella foenum-graecum* is more effective.

**Keywords:** *Cissus quadrangularis*, *Trigonella foenum-graecum*, Anti-diabetic,  $\alpha$ -amylase inhibition, Diabetes mellitus, Herbal medicine.

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## INTRODUCTION

Diabetes mellitus (DM) is a metabolic disorder characterized by chronic hyperglycemia either because the pancreas does not produce enough insulin or the peripheral target tissues are unable to respond to the normal concentration of insulin (1,2). It is a major cause of morbidity and mortality with an increasing prevalence and the fastest growing disease worldwide (3,4).

The universal prevalence of diabetes has nearly doubled since 1980, rising from 4.7% to 8.5% in the adult population. Moreover, the prevalence of diabetes has also been found to steadily increase for the past 3 decades and has risen faster in low- and middle-

income countries compared to high-income countries. The increase in the prevalence of diabetes is parallel with an increase in associated risk factors such as being overweight or obese. If not properly treated or controlled, diabetes may cause blindness, kidney failure, lower limb amputation, and other long-term consequences that impact significantly on the quality of life (5). Interestingly, the WHO also projects that diabetes will be the seventh leading cause of death in 2030 (6).

Natural products, particularly of plant origin, are the main quarry for discovering promising lead candidates and play an imperative role in the upcoming drug development programs (7). Ease of availability,

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low cost, and least side effects make plant-based preparations the main key player of all available therapies, especially in rural areas (8). Moreover, many plants provide a rich source of bioactive chemicals, which are free from undesirable side effects and possess powerful pharmacological actions.(9) Plants also have always been an exemplary source of drugs with many of the currently available drugs being obtained directly or indirectly from them (10). The recent review of Durazzo et al. gives a current snapshot of the strict interaction between the main biologically active compounds in plants and botanicals by giving a mini overview of botanicals features, a definition of the study, and examples of innovative (i.e., an assessment of the interaction of bioactive compounds, chemometrics, and the new goal of biorefineries) and a description of existing databases (i.e., plant metabolic pathways, food composition, bioactive compounds, dietary supplements, and dietary markers); in this regard, the authors marked the need for categorization of botanicals as useful tools for health research.(11)

Currently, the only available treatment for T1DM is insulin therapy and has many side effects (12). Therefore, testing the efficacy of natural antidiabetic agents for the treatment of diabetes offers least/no side effects. Two medicinal plants easily available in Indian subcontinent are *Cissus quadrangularis* L. and *Trigonella foenum-graecum* L.. *C. quadrangularis* L. commonly known as Veldt Grape from the family Vitaceae is a perennial herb native to India. Phytoconstituent analysis of *C. quadrangularis* by GC-MS revealed presence of alkaloids, terpenes and terpenoids, ascorbic acid (vitamin C), tocopherols (Vitamin E), tannins, phenols, saponins, flavonoids and phytosterols (13). The plant extract is also rich in calcium ions and studies reported the formation of calcite crystal (14). *Trigonella foenum-graecum* L. (fenugreek), from the family Fabaceae, is common to many European, Middle Eastern, and Asian countries including India. Dried ripe seeds of *T. foenum-graecum* are used as spices by Indians and have medicinal values too. Extract of dried ripe seed of *T. foenum-graecum* are well known to possess antidiabetic properties (15). *T. foenum-graecum* seed extract also possess antihyperlipidemic, antihypercholesterolemic, antioxidant and estrogenic activities and is reported to improve hemorheological properties of blood(16,17). The rationale of selecting combination of *C. quadrangularis* and *T. foenum-graecum* is their importance as antidiabetic agent, respectively in traditional medicine.

## MATERIALS AND METHODS

### ANTIDIABETIC ACTIVITY

#### Inhibition of Alpha Glucosidase

The  $\alpha$ -amylase inhibition assay was performed using the 3,5-dinitrosalicylic acid (DNSA) method suggested by Wickramaratne et al., 2016. The plant extract was dissolved in minimum amount of 10% DMSO and was further dissolved in buffer ((Na<sub>2</sub>HPO<sub>4</sub>/NaH<sub>2</sub>PO<sub>4</sub> (0.02 M), NaCl (0.006 M) at pH 6.9) to give concentrations ranging from 10 to 1000  $\mu$ g/ml. A volume of 200  $\mu$ l of  $\alpha$ -amylase solution (2 units/ml) was mixed with 200  $\mu$ l of the extract and was incubated for 10 min at 30 °C. Thereafter 200  $\mu$ l of the starch solution (1% in water (w/v)) was added to each tube and incubated for 3 min. The reaction was terminated by the addition of 200  $\mu$ l DNSA reagent (12 g of sodium potassium tartrate tetrahydrate in 8.0 mL of 2 M NaOH and 20 mL of 96 mM of 3,5-dinitrosalicylic acid solution) and was boiled for 10 min in a water bath at 85–90 °C. The mixture was cooled to ambient temperature and was diluted with 5 ml of distilled water, and the absorbance was measured at 540 nm using a UV-Visible spectrophotometer. The blank with 100% enzyme activity was prepared by replacing the plant extract with 200  $\mu$ l of buffer. A blank reaction was similarly prepared using the plant extract at each concentration in the absence of the enzyme solution. A positive control sample was prepared using Metformin(500mg) (1  $\mu$ g/ml) and the reaction was performed similarly to the reaction with plant extract as mentioned above.

#### Procedure

100  $\mu$ l of 0.1 U glucosidase was taken in different tubes. To this 50  $\mu$ l of sample and standard of different concentrations were added (should not mix) and incubated at 25°C for 10 min. Then 50  $\mu$ l of p-nitrophenyl alpha- D-glucosidase was added, vortexed and incubated at 25°C for 5 min. Add 800  $\mu$ l of stop solution (0.1 M sodium carbonate) was added. Absorbance was measured at 405 nm.

$$\% \text{ of alpha glucosidase inhibition} = \frac{\text{OD control} - \text{OD sample}}{\text{OD control}} \times 100$$

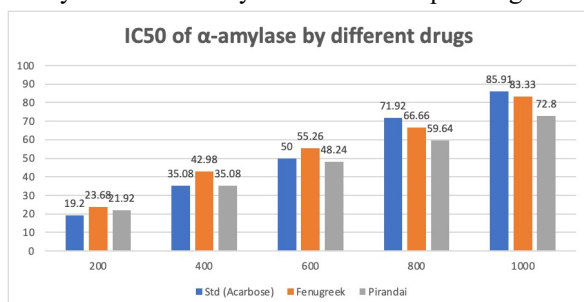
## RESULTS

### Table 1: IC<sub>50</sub> of $\alpha$ -amylase by different drugs

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Concentration	Std (Acarbose)		Fenugreek		Pirandai	
	OD	% of Inhibition	OD	% of Inhibition	OD	% of Inhibition
200	0.92	19.29	0.87	23.68	0.89	21.92
400	0.74	35.08	0.65	42.98	0.74	35.08
600	0.57	50	0.51	55.26	0.59	48.24
800	0.32	71.92	0.38	66.66	0.46	59.64
1000	0.16	85.96	0.19	83.33	0.31	72.80
<b>IC50 Value</b>	571.051 $\mu\text{g/ml}$		538.587 $\mu\text{g/ml}$		638.463 $\mu\text{g/ml}$	

The table 1 show that Trigonella Foenum-Graecum has a lower IC50 value (538.587 $\mu\text{g/mL}$ ) and a higher percentage inhibition at similar concentrations (compared to 638.463  $\mu\text{g/mL}$  for cissus quadrangularis) further indicates that it has a stronger ability to inhibit  $\alpha$ -amylase than cissus quadrangularis.



**Figure 1: Bar graph showing IC50 of  $\alpha$ -amylase by different drugs**

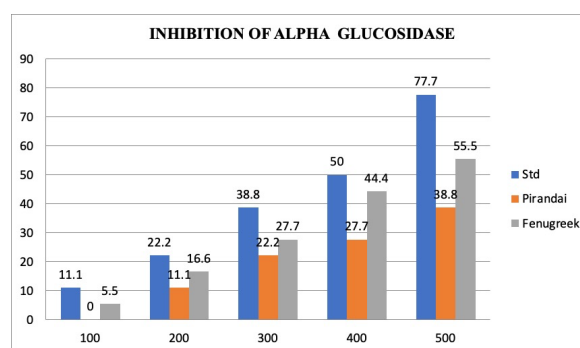
**Table 2:  $\alpha$ -amylase inhibition assay conducted at various concentrations ( $\mu\text{g/mL}$ )**

Sample Concentration ( $\mu\text{g}$ )	20	40	60	80	100
<b>Std. Acarbose</b>	0.16	0.14	0.11	0.09	0.04
<b>% of inhibition</b>	11.1	22.2	38.8	50	77.7
<b>Pirandai</b>	0.18	0.16	0.14	0.13	0.11

% of inhibition	0	11.1	22.2	27.7	38.8
<b>Fenugreek</b>	0.17	0.15	0.13	0.10	0.08
<b>% of inhibition</b>	5.5	16.6	27.7	44.4	55.5

The table 2 presents the results of an  $\alpha$ -amylase inhibition assay conducted at various concentrations ( $\mu\text{g/mL}$ ) of a sample substance. Each concentration was evaluated for its percentage of inhibition compared to the control. Cissus Quadrangularis And Trigonella Foenum-Graecum

As the concentration of the sample increased from 20% to 100% microgram/milliliter, there was a noticeable trend of increased inhibition percentage, indicating a dose-dependent response. It was found that fenugreek have better inhibition of  $\alpha$ -amylase when compared to pirandai even at the lowest concentration.



**Figure 2: Bar graph showing % inhibition of  $\alpha$ -amylase by different groups**

### SUMMARY AND CONCLUSION:

The study evaluated the antidiabetic activity of two samples, Cissus quadrangularis and Trigonella foenum-graecum, by measuring their " $\alpha$ -amylase" enzyme inhibition. There was a steady rise in inhibition amount of Trigonella foenum-graecum with increasing concentration, reaching 55.5% at 100 $\mu\text{g/mL}$ . whereas, Cissus quadrangularis showed the inhibition only at 40  $\mu\text{g/mL}$  and at maximum concentration the inhibition was 38.8%.

This study indicates that Trigonella foenum-graecum has a stronger ability to inhibit  $\alpha$ -amylase than Cissus quadrangularis. Based on this information, Trigonella foenum-graecum may be able to work better as an antidiabetic agent by blocking " $\alpha$ -amylase". Trigonella foenum-graecum Powder may have a greater potential for antidiabetic activity than Cissus quadrangularis due to its ability to inhibit  $\alpha$ -amylase.

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### CONCLUSION

Although *Cissus quadrangularis* and *Trigonella foenum-graecum* both have antidiabetic characteristics, the evidence indicates that *Trigonella foenum-graecum* is more effective. As a diabetes treatment, *Trigonella foenum-graecum* works well.

Further studies and clinical trials would be beneficial to confirm these findings and explore the therapeutic potential of these natural products.

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