Antidiabetic Activity of *Sida Rhombifolia (L)* leaf extract

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Abstract— Diabetes mellitus is a metabolic disease that is a major health problem today, the prevalence of which in latest years has been rapidly increasing worldwide. It was also considered as an untreatable metabolic illness that affects about 2.8 % of the inhabitants of the world. This study was aim to analyses the quantitative and qualitative phytochemistry and antidiabetic activity of Sida rhombifolia leaf extract. Maximum scavenging activity of alpha amylase inhibition assay of different extract was observed that the ethanol exhibited comparable to the another concentration activity in the maximum activity to compare with another value in the inhibition. Glucose uptake by yeast activity of different extract was observed the ethyl acetate exhibited comparable are responsible to maximum percentage protection. Result indicated that the sida rhombifolia(L.)plant ethyl acetate extract shows 82.30% of activity was recorded the Sida rhombifolia(L.) leaf extract cum. The overall results have clearly indicated that the extract could be used as a therapeutic agent. Though further analysis is needed as clinical trials and other advanced approaches to confirm this as a safer therapeutic medicine.

Keywords : Antidiabetic, Alpha amylase, inhibition assay, Glucose, Phytochemical analysis

I. INTRODUCTION

Diabetes mellitus (DM) is a chronic disorder associated by insulin production defect and or both of its intervention. In many other metabolism within the human body, this contributes to extended hyperglycemia with alterations [1]. Diabetes has an implicit relationship with the most prevalent endocrine disorder in many other illnesses. Approximately 200 million individuals globally were projected to have benefited from DM in 2010, and by 2025 it is anticipated to achieve 300 million [2]. Depending on the disease etiology, diabetes mellitus is categorized into distinct categories, but the two main kinds are commonly adopted: type 1 (IDDM) and type 2 (NIDDM) [3]. In addition, there is also another short-term diabetes-related disease recognized as mellitus gestational diabetes (GD). It relates to the incidence of glucose intolerance or initial identification throughout the pregnancy era. Other diseases involve genetic pancreatic ß-cell disorders or mutations of insulin receptors or deformities of post-receptors [3].

Fasting plasma glucose (FPG) concentration assessment, which takes place early in the morning, is the most common diabetes diagnostic methods. It is regarded normal for patients with FPG below 100 mg / dl ; those between 100 and 125 mg / dl are pre-diabetes while those above 125 mg / dl are diabetic[4]. The current oral antidiabetics involve sulfhonyl ureas that reduce blood sugar, primarily by increasing insulin production from Langerhans islets. They are coupled with receptors of sulfonyl urea on ß cells leading to the closure of adenosine triphosphate-dependent potassium channels. As a result, depolarization of the cell membrane and the subsequent influx of calcium accompanied by secretion of stored insulin from secretory granules occurs within the cells. This process only performs when insulin is present [4]. The "starch blockers" alpha-glucosidase inhibitors block certain enzymes that cause carbohydrate decomposition in the small intestine. They function primarily by reducing the absorption level of carbohydrate in the body [5, 6]. In addition, by adding to the carbohydrate-binding region and interacting with their hydrolysis in mono-saccharides, acarbose reversibly inhibits both pancreatic alpha-amylase and ß-glucosidase enzymes[7]. This findings in slower absorption and reduced levels of postprandial blood sugar.In order to eliminate these side effects, it is better to in troduce hypoglycemic herbs so that they can be used in conj unction with standard drugs.

II. MATERIALS AND METHODS

Collection of sample:

The flowering plant of *Sida rhombifolia* leaves were collected from road sides and around the Vellore district, Tamil nadu during **December, 2018** .The sample was authenfied from botanical survey of India. The leaves were gently washed with tap water, then rinsed with distilled water. Then air dried and powered to mesh and preparation.

Sequential extraction:

Sida rhombifolia leaves about (100gm) of dried powder was placed in 300ml of hexane for added 24 hours .After 24 hours mixed and then filtered by using whatmann's filter paper. The filtered thus obtained was cool and concentrated to dryness. Then dry leaf powder (100gm) was placed in 300ml of ethyl acetate for added 24 hours in maceration. After 24 hours mixed and then filtered thus obtained was

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cooled and concentrated to dryness. Then dry powder of leaves in (100gm) was placed 300ml of ethanol for 24hours maceration. After 24hours mixed and filtered thus obtained to cool. Then the extraction mixture was filtered and red concentrated of distilling unit. Then its residues were weighed and kept in bottle to use phytochemical and other biological screening methods. Then collected filtrate**.**

Alpha-Amylase Inhibition Assay:

Various compound concentrations (200-1000µg) and incubate 500µl of 0.02mol / l sodium phosphate buffer (pH 6.9with 0.006m mol / l Nacl) with porcine pancreatic alpha amylase enzyme (0.5mg / ml) at 25^{o}c for 10minThe reaction mixture was introduced after the 500µl of 1 % starch solution in 0.02mol / l of sodium phosphate buffer (pH 6.9 with 0.006mol / l Nacl). The reaction mixture was then incubated for 10min at 25^{o}cThe procedure was lastly halted by incubation in the boiling water bath (5min) and cooled to room temperature, followed by the addition of 1.0ml of Dinitrosalicylic acid (DNS). With 10ml of distilled water, the reaction mixture was diluted. The colorimetric absorption measure at 540 nm. The reaction mixture of all reagents and enzymes with the exception of the test sample was used as control. The inhibition percentage was shown as an outcome of alpha amylase inhibitor activity. The inhibition percentage was calculated using the given formula,

% Inhibition = (Abs Control – Abs Sample)/ Abs Control *100

Yeast Cell Glucose Uptake:

The yeast cell was formed by Cirillo et al (1962) technique. *Saccharomyces cerevisiae* was suspended distilled water and was washed by repeated centrifugation (4200rpm, 5min) in distilled water until pellet appeared clear. A 5 %(w/w) suspension of washed yeast was prepared in distilled water.

The yeast cell's glucose uptake was evaluated. Four concentrations of this compound (25, 50, 75,100 µg / ml) were prepared. A standard glucose concentration of 100mM/L was prepared. Different extract concentrations were introduced to the 1ml of glucose solution and incubated at 37oc for 10minitues. The reaction was initiated with 100 µl of vortexed yeast suspension and further incubated at 37oc for 60 minutes. After incubation the tubes were centrifuged (3800rpm, 5min) and Glucose was measured using anthrone technique in the supernatant.

Increase in glucose uptake (%) = (Abs control-Abs sample) / Abs control *100

Where, Abs control is the control reaction absorbance (which contains all reagents except the sample) and Abs sample is the test sample absorbance

III. RESULTS AND DISCUSSION

Alpha-Amylase Inhibition Assay Sida Rhombifolia (L.)

α - Amylase inhibition assay scavenging activity in hexane, ethyl acetate and ethanol extract *Sidarhombifolia(L)*

Table 1: The inhibition percentage of Hexane

Concentration in µg	%Inhibition(C-S) /Cx100	%Inhibition(C-S) /Cx100
200	68.63%	89.28%
400	59.06%	89.92%
600	46.05%	93.36%
800	31.65%	94.38%
1000	18.26%	95.66%

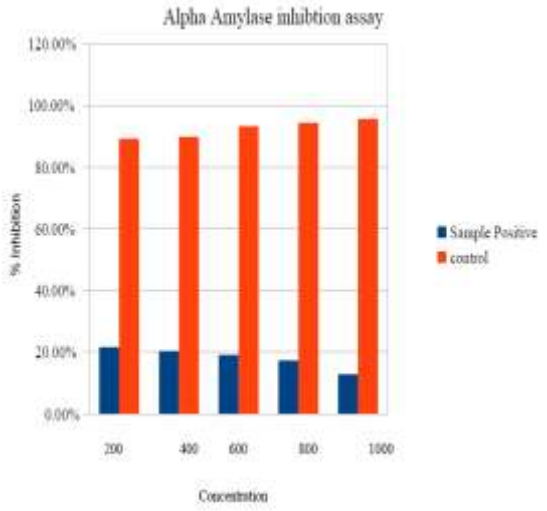

Figure 1: Alpha amylase inhibition scavenging activity was observed in hexane extract of *Sida rhombifolia(L.)*

Table.2:The inhibition percentage of Ethyl Acetate

Concentration in µg	%Inhibition(C-S) /Cx100	%Inhibition(C-P. C)/Cx100
200	20.35%	89.28%
400	19.26%	89.92%
600	16.78%	93.36%
800	14.43%	94.38%
1000	13.01%	95.66%

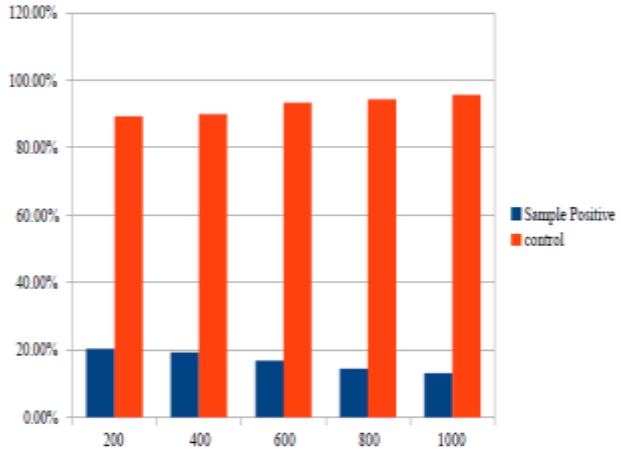

Figure 2: Alpha amylase inhibition scavenging activity was observed in ethyl acetate extract of *Sida rhombifolia(L.)*

Table.3:The Inhibition Percentage of Ethanol

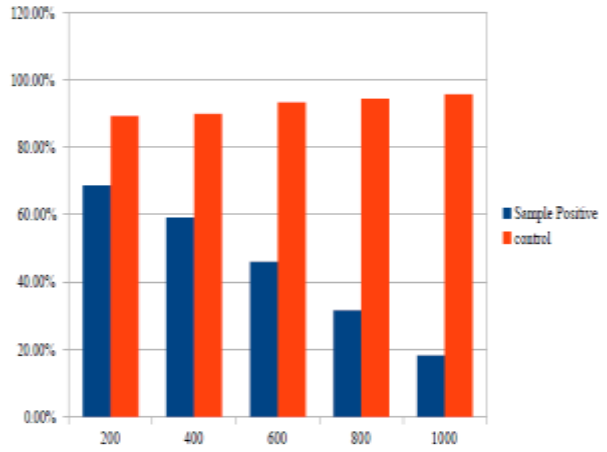

Figure 3: Alpha amylase inhibition scavenging activity was observed in ethanolic extract of *Sida rhombifolia(L.)*

The results are summarized Table1, Table2, Table3. It was observed that the water and ethanol soluble of content as selected leaf extract (10mg/ml) were found be potent α-amylase inhibition assay agents. The maximum scavenging activity was observed in ethanolic extract of *Sida rhombifolia(L.)retusa* (68.63%) while maximum activity of compare with another value of inhibition.

Yeast glucose uptake:

The antidiabetic activity of the drug should be evaluated with glucose uptake by yeast. Then the percentage will be increased in glucose concentration.

α – The glucose uptake by the yeast in hexane, ethyl acetate and ethanol extract of *Sidarhombifolia(L)*

Table.4: The Increase In Glucose Uptake (%) of Hexane

Mean % Inhibition		
Concentration in µg	Compound%	Acarbose%
25	73.32%	43.45%
50	62.94%	31.03%
75	54.89%	25.36%
100	45.73%	14.93

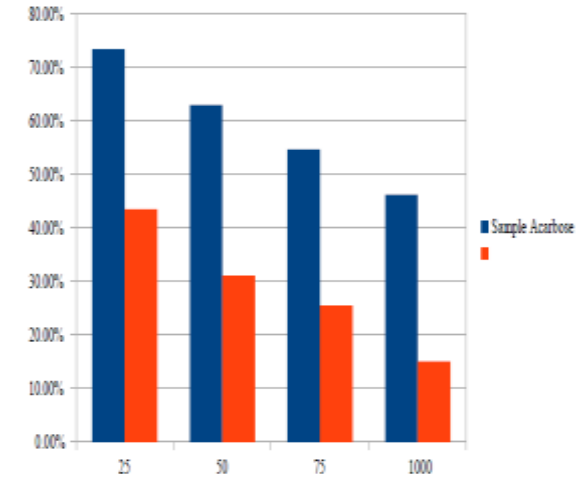

Figure 4: The Glucose uptake by yeast in hexane extract of *Sidarhombifolia(L)*

Table.5: The Increase In Glucose Uptake (%) Ethyl Acetate:

Mean % Inhibition		
Concentration in µg	%Compound	%Acarbose
25	82.30%	43.45%
50	73.70%	31.03%
75	64.42%	25.36%
100	55.83%	14.93%

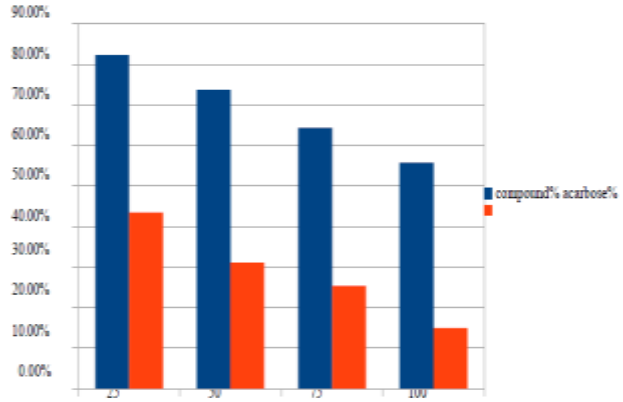

Figure 5: The Glucose uptake by yeast in ethyl acetate extract of *Sidarhombifolia(L)*

Table.6: The Increase In Glucose Uptake (%) Ethanol:

%Mean inihibition		
Concentration in µg	%Compound	%Acarbose
25	72.18%	43.45%
50	65.35%	31.03%
75	52.24%	25.36%
100	46.63%	14.93%

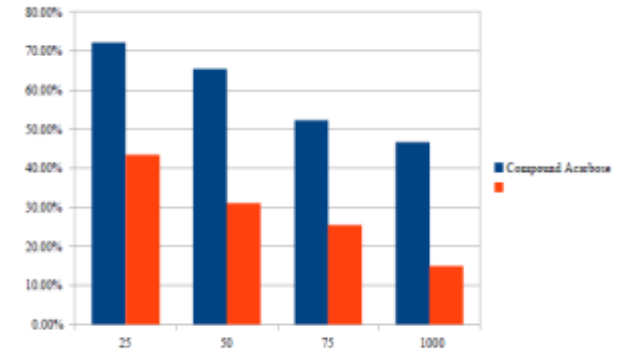

Figure 6: The Glucose uptake by yeast in ethanolic extract of *Sidarhombifolia(L)*

The glucose uptake by yeast activity of different extract was observed the ethyl acetate exhibited to comparable are responsible to maximum % protection. Result indicated that the Sida rhombifolia(L.) leaves from ethyl acetate extract shows 82.30% of activity was recorded to comparable to another extract of cum through inhibition.

IV. CONCLUSION

Diabetes mellitus is a terrible disease triggered by increas ed manufacturing of hepatic glucose and impaired action of i nsulin. Over the past three centuries, the use of herbal medic ine has grown dramatically in both developing and develope d countries. The alpha amylase inhibition assay in showed that the extract was highly effective and the result of induced cum treated with comparable to the control activity inhibition. The effect of *Sida rhombifolia* extract on the other hand is comparable to treated with antidiabetic drug. This was confirmed by the glucose uptake by yeast analysis. The effect is attributed to the rich chemical composition of *Sida rhombifolia* extract such saponins, flavonoids, alkaloids, phenols etc. The overall results have clearly indicated that the extract could be used as therapeutic agent. Through further analysis is needed as clinical trials and other high tech approaches to confirm the same in order to confirm as safer therapeutic medicine in antidiabetic activity significant.

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