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Exploring the Antioxidant and *In Vitro* Anti-Diabetic Properties of *Stachytarpheta urtifolia* Leaves

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ABSTRACT

Diabetes mellitus is a chronic metabolic disorder caused by carbohydrate, fat, and protein metabolism problems. Genetic predispositions and environmental influences contribute to the disease, presenting significant challenges to patients' health. There are two types of diabetes, Type I and Type II. Both lead to elevated blood glucose levels (hyperglycemia). Several complications are associated with persistent hyperglycemia, including microvascular problems affecting the eye, kidneys, and nerves, as well as macrovascular problems like accelerated artery hardening, which increases heart disease, stroke, and peripheral artery disease risks. It is essential to focus on the prevention and management of diabetes-related complications in order to mitigate their impact on micro and macrovascular health. It is important to consider alternative treatment options as these medications can pose undesirable side effects. Medication, dietary adjustments, and regular exercise are all part of comprehensive diabetes management. Our study was aimed of the present study is to explore the anti-oxidant and anti-diabetic activity in the leaves of *Stachytarpheta urticifolia.* A comprehensive study was conducted to examine *Stachytarphata urticifolia*, a traditional botanical remedy, for its phytochemical composition, antioxidant potential, and effectiveness against diabetes. In this study, *Stachytarphata urticifolia* was found to exhibit significant diabetic-fighting and anti-oxidant properties. In addition to phenolic compounds, flavonoids, alkaloids, and terpenoids, *Stachytarphata urticifolia* contains hundreds of bioactive compounds. Phytochemicals such as these, which possess anti-oxidant and anti-diabetic properties, are considered to be promising therapeutic choices for humans who are suffering from diabetes or metabolic disorders. *Stachytarphata urticifolia* has shown remarkable antioxidant activity over the past few decades, which has been demonstrated to be helpful in preventing oxidative damage and preventing the onset of chronic diseases like diabetes by battling free radicals and combating oxidative stress. The in vitro studies of *S. urticifolia* were conducted in order to evaluate its anti-diabetic potential. There have been studies conducted *in vitro* that examined key enzymes associated with carbohydrate metabolism in an attempt to determine whether *Stachytarphata urticifolia* inhibits carbohydrate digestion and absorption by inducing moderate inhibitory effects on amylase, glucose oxidase, and glucose uptake. There was no doubt that *S. urticifolia* is capable of reducing complications associated with diabetes and these findings suggest that it may be useful for patients with diabetes who need to mitigate these complications. As part of this study, we examined *S. urticifolia*, a well-known traditional plant, for its phytochemical composition and potential anti-oxidant and anti-diabetic effects. The anti-

oxidant potential of *S. urticifolia* and its moderate anti-diabetic activity were also found in our study. Based on the results of this study, *S. urticifolia* might be of benefit to diabetic patients as a therapeutic agent.

KEYWORDS: Antioxidant, *Stachytarpheta urtifolia,* Anti-Diabetic, Plant Extract.

INTRODUCTION:

Diabetes mellitus:

Diabetes mellitus is a chronic metabolic disorder caused by carbohydrate, fat, and protein metabolism problems. Genetic predispositions and environmental influences contribute to the disease, presenting significant challenges to patients' health. There are two types of diabetes, Type I and Type II [1]. Both lead to elevated blood glucose levels (hyperglycemia). Several complications are associated with persistent hyperglycemia, including microvascular problems affecting the eye, kidneys, and nerves, as well as macrovascular problems like accelerated artery hardening, which increases heart disease, stroke, and peripheral artery disease risks. It is essential to focus on the prevention and management of diabetes-related complications in order to mitigate their impact on micro and macrovascular health [2].

Prevalence of diabetics in India:

Diabetes has become a widespread health condition in India that crosses all social strata, transcending its traditional association with affluence. Among individuals of Indian descent, diabetes is proliferating rapidly due to social shifts and lifestyle changes, exacerbated by geographic and ethnic predispositions [3].

With diabetes becoming the leading cause of death worldwide, India stands on the verge of a crisis. In 2030, an estimated 80 million Indians will suffer from diabetes, a significant increase from 32 million in 2000 [4]. According to projections, this number will grow to a staggering 100 million by the year 2050.

Risk factors of Diabetes Mellitus [5]:

- Diabetes can stem from insufficient insulin production or resistance to insulin within the body.
- Hereditary factors play a significant role in the transmission of diabetes across generations.
- Excessive weight or obesity is a prominent risk factor, particularly for individuals with a family history of diabetes mellitus.
- Certain viral infections, such as influenza, mumps, and Coxsackie virus, have been implicated in the destruction of beta cells in the pancreas, leading to diabetes development.
- Various medications, including diuretics, corticosteroids, diazoxide, phenytoin, and others, can contribute to the onset of diabetes.
- Inflammation of the pancreas is also recognized as a potential trigger for diabetes onset.

Symptoms:

- The sudden onset of insulin-dependent diabetes mellitus (IDDM) is characterized by the abrupt appearance of symptoms such as frequent urination, extreme thirst and/or hunger, rapid weight loss, irritability, weakness and exhaustion, as well as nausea and vomiting.
- Conversely, non-insulin-dependent diabetes mellitus (NIDDM) typically develops more gradually, with symptoms that may be subtle. These can include any of the aforementioned symptoms, alongside itching, blurry vision, obesity, tingling or numbness in the feet, slow healing of the skin or gums, and recurrent bladder infections.

Diabetic's treatments:

Diabetes mellitus is treated with multiple strategies aimed at reducing symptoms, controlling blood glucose levels, and preventing complications. The use of insulin therapy remains an effective way to control blood sugar despite the fact that some of its drawbacks include insulin resistance, brain atrophy, anorexia, and fatty liver. Various hypoglycemic agents, including biguanides and sulfonylureas, can reduce hyperglycemia along with insulin.

It is important to consider alternative treatment options as these medications can pose undesirable side effects. Medication, dietary adjustments, and regular exercise are all part of comprehensive diabetes management.

A medication may be adjusted based on individual responses and tolerances for controlling blood sugar levels. It is essential to eat balanced meals, monitor carbohydrate intake, and avoid sugary foods to maintain diabetes. Blood sugar control is enhanced by regular exercise, which improves insulin sensitivity and promotes weight management.

Furthermore, prevention of complications is incorporated into treatment beyond glycemic control. Diabetes cardiovascular risk factors can be reduced with lifestyle modifications, including smoking cessation and moderate alcohol consumption. Early detection and management of complications depends on monitoring blood pressure, cholesterol, and kidney function.

Herbal medicines for DM:

As synthetic drugs are associated with adverse reactions and undesirable side effects, herbal medicines are becoming increasingly popular for treating diabetes. Alternatives to conventional medications include natural products and dietary modulators with anti-diabetic properties. In comparison to synthetic drugs, herbal medicines are perceived to be less toxic, have fewer side effects, and are relatively less expensive.

Numerous pharmaceuticals are derived from plants directly or indirectly, as plants are a primary source of medicinal compounds. Glucophage, a drug derived from Galega officinalis, is an example of an antihyperglycemic drug.

A plant-based remedy for diabetes management has this therapeutic potential. There have been scientific studies that show the potential benefits of several herbs and botanical extracts as antidiabetic remedies. Some traditional medicines use bitter melon (Momordica charantia), fenugreek (Trigonella foenumgraecum), cinnamon (Cinnamomum verum), and ginseng (Panax ginseng) for their purported anti-diabetic effects.

Compounds in bitter melon can help regulate blood sugar levels and improve insulin sensitivity. Fasting blood glucose levels are reduced and glucose tolerance is improved by fenugreek seeds. It is also possible to enhance insulin sensitivity and lower blood glucose levels with cinnamon extract [6, 7, 8, 9]. The use of herbal medicines holds promise as complementary or alternative therapies for diabetes management, but patients should work with healthcare professionals before starting treatment. Herbal remedies for diabetes may interact with other medications and differ in potency and purity [10, 11]. It is important to make informed decisions and monitor closely. There is still much to learn about herbal medicines and their efficacy, safety, and optimal dose.

Plant review:

Figure 1: *Stachytarpheta urtifolia* leaves

Morphological characters:

This perennial herb thrives in the wild across Bangladesh's Sylhet and Chittagong districts, commonly called blue snakeweed or nettle leaf velvet berry. Moreover, it is cultivated as an ornamental weed in these areas [12]. There are also populations of this species found on various continents, including Africa, the Indian Ocean islands (such as Seychelles, Comoros, Madagascar, Reunion, and Mauritius), Southeast Asia, eastern Australia, and New Caledonia. Morphology of plant is presented in Figure 1.

Medicinal uses:

Traditionally, S. Urticifolia has been used to treat fever, rheumatism, venereal disease, dropsy, and ulcers [13]. Diarrhea and dysentery are treated with the bark infusion. Moreover, tribal communities in Bangladesh use it as an abortifacient.

Pharmacological uses:

Cytotoxicity:

The leaf and root bark extracts of S. urticifolia, along with the isolated compounds ipolamiide and α-spinasterol, showed moderate cytotoxic effects against brine shrimp nauplii (Artemia salina). Vincristine sulfate was used as a reference standard in this evaluation.

Anti-oxidant property:

Significant antioxidant activity was observed in the methanolic extracts obtained from the leaves and inflorescences of *S. urticifolia*.

Anti-bacterial property:

The root aqueous extract of *S. indica* and the leaf methanolic extract of S. urticifolia exhibited notable activity against different bacterial species [14], comparable to the standard antibiotics streptomycin and ampicillin, respectively. Various research studies have also highlighted the antimicrobial effectiveness of *S. jamaicensis* extracts against pathogenic microorganisms, including both bacteria and fungi.

Aim:

The aim of the present study is to explore the anti-oxidant and anti-diabetic activity in the leaves of *Stachytarpheta urticifolia.*

Objectives:

- To extract the phytoconstituents from the leaves of *Stachytarpheta urticifolia* by Soxhlet extraction with hydro alcohol (50:50).
- To perform phytochemical screening on extract.
- To perform the Anti-oxidant estimation by DPPH method.
- To perform the Inhibition assay of α- amylase and α-glucosidase activity and glucose uptake study.

Methodology:

Plant collection and authentication:

Fresh leaves of *Stachytarphata urticifolia* was collected local area in Chennai, Tamil Nadu from during December.

Extraction of plant materials:

Leaves of *Stachytarphata urticifolia* (1kg) were dry and pulverized. Then it undergoes extraction by means of Continuous hot percolation method (Soxhlet apparatus). A mechanical grinder was used to finely powder the dried leaves of *S.urticifolia* until a particle size of 40-100 mm was achieved. Afterwards, the powder was stored at room temperature in polythene bags. To extract the powdered leaves material, 100 grams were subjected to a continuous percolation process, utilizing the Soxhlet extraction method. Solvent extraction was facilitated by heating the solid matrix with reflux in the SOX thimble to facilitate Soxhlet operation. Solutes were transferred to the reservoir through condensation and extraction with fresh solvent. In most cases, this extraction process took 24 hours to complete once exhaustive extraction had been achieved. A constant 70°C temperature was maintained throughout the extraction process. Hydro-alcohol served as the solvent for the extraction process. Following this, the extract was filtered through a Whatman No. 1 filter paper and evaporated to dryness using a rotary evaporator under reduced pressure. In order to further process the extract, dark glass bottles were used to store the crude extract.

Extraction yield:

The powdered sample of *S. urticifolia* leaves was effectively extracted with hydro-alcohol employing a Soxhlet extractor. A yield of approximately 1.12% was obtained from the extraction process.

Phyto-chemical screening:

A phytochemical analysis of *S. urticifolia* hydroalcoholic extract revealed the presence of some phytoconstituents, despite the absence of some compounds. In Table 1, phytoconstituents identified in the extract are summarized, along with their presence or absence.

Table: 1 Phytochemical screening of hydroalcoholic extract of *S. urticifolia*

Constituents	(Presence) or (Absence) of Phytochemicals
Flavonoids	Presence
Tannins	Absence
Alkaloids	Presence
Protein	Absence
Saponins	Presence
Steroids	Absence

Glycosides	Presence

Anti-oxidant assay (DPPH method):

Antioxidant activity of *S. urticifolia* hydroalcoholic extract was tested against DPPH free radicals. At all concentrations tested, the plant extract was found to significantly scavenge DPPH free radicals. In particular, the highest concentration tested (0.5 mg of crude extract) inhibited DPPH free radicals 94.23%, as shown in Table 2, 3.

Table 2: Scavenging ability of standard drug (ascorbic acid)

S.no	Concentration (mg)	Absorbance @520 nm Control	Absorbance @520 nm Standard (ascorbic acid)	%inhibition
1	0	0.82	0.03	0
2	0.1	0.84	0.07	90.4
3	0.2	0.87	0.04	93.8
4	0.3	0.84	0.05	92.8
5	0.4	0.84	0.05	92.8
6	0.5	0.84	0.03	94.2

Table 3: Scavenging ability of Sample (*S. urticifolia* hydro alcoholic extract)

S.no	Concentration (mg)	Absorbance @520 nm CONTROL	Absorbance @520 nm EEVN	%INHIBITION
1	0	0.82	0.02	0
2	0.1	0.84	0.06	80.2
3	0.2	0.87	0.03	86.8
4	0.3	0.84	0.04	90.8
5	0.4	0.84	0.03	90.8
6	0.5	0.84	0.02	92.4

Thin Layer Chromatography of *S. urticifolia* hydro alcoholic extract:

The hydro alcoholic extract of *S. urticifolia* was subjected to TLC profiling using 100% methane. The analysis revealed the presence of seven major compounds in the extract, each characterized by distinct retention factor (R_f) values: 0.27, 0.36, 0.41, 0.56, 0.65, 0.71, and 0.86.

Anti-diabetic assay:

In vitro α-amylase inhibition assay:

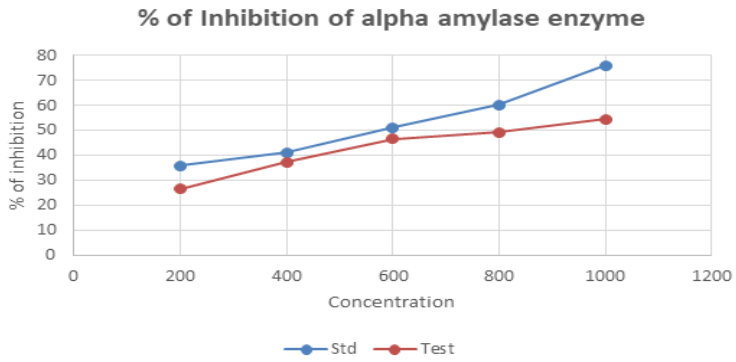

Figure 2: In vitro α-amylase inhibition activity

Based on the results presented in the table, *S.urticifolia* (HAESU) exhibited moderate activity against amylase. There was a discernible impact of the extract on the activity of the amylase enzyme, even if its impact was not particularly strong or weak, but rather fell in the middle. HAESU has a moderate activity that suggests it may interact with amylase in a noticeable way and affect its function, which is interesting from a biological and pharmacological perspective. Inhibition activity of standard and test are presented in Figure 2.

In vitro α-glycosidase inhibition assay:

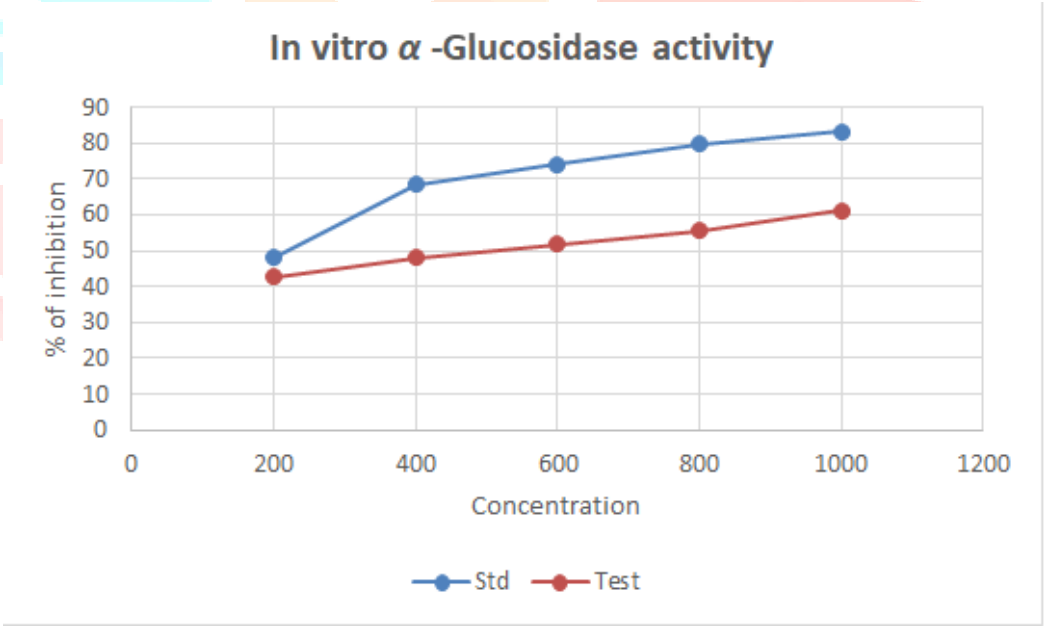

Figure 3: *In vitro* α-glycosidase inhibition activity

Based on the results presented in the table, *S.urticifolia* (HAESU) exhibited moderate activity against glycosidase. There was a discernible impact of the extract on the activity of the glycosidase enzyme, even if its impact was not particularly strong or weak, but rather fell in the middle. HAESU has a moderate activity that suggests it may interact with amylase in a noticeable way and affect its function, which is interesting from a biological and pharmacological perspective. Inhibition activity of standard and test is presented in Figure 3.

Table 4: Glucose uptake assay

Groups	Concentration (µl)	Glucose uptake by Yeast cells	
		Absorbance at565nm	% Glucose uptake by Yeast cells
Control		0.74	
Standard Metformin(1mg/ml)	25	0.35	54.41
HAESU	25	0.54	46.21

The mechanism by which yeast cells take up glucose may be different from those of other eukaryotic or human cells. The exact mechanism by which yeast cells uptake glucose remains unclear, unlike humans, where glucose transport involves enzymes and processes. The concentration of glucose within yeast cells and its subsequent metabolism can influence glucose uptake in yeast cells. The internal glucose concentration decreases when it is rapidly metabolized into other compounds, allowing the cell to absorb glucose more readily. Compared with the standard drug, HAESU significantly enhanced glucose uptake by yeast cells following treatment with the samples, as shown in Table 4.

DISCUSSION:

A comprehensive study was conducted to examine *Stachytarphata urticifolia*, a traditional botanical remedy, for its phytochemical composition, antioxidant potential, and effectiveness against diabetes. In this study, *Stachytarphata urticifolia* was found to exhibit significant diabetic-fighting and anti-oxidant properties. In addition to phenolic compounds, flavonoids, alkaloids, and terpenoids, *Stachytarphata urticifolia* contains hundreds of bioactive compounds. Phytochemicals such as these, which possess anti-oxidant and anti-diabetic properties, are considered to be promising therapeutic choices for humans who are suffering from diabetes or metabolic disorders. *Stachytarphata urticifolia* has shown remarkable antioxidant activity over the past few decades, which has been demonstrated to be helpful in preventing oxidative damage and preventing the onset of chronic diseases like diabetes by battling free radicals and combating oxidative stress.

The in vitro studies of *S. urticifolia* were conducted in order to evaluate its anti-diabetic potential. There have been studies conducted *in vitro* that examined key enzymes associated with carbohydrate metabolism in an attempt to determine whether *Stachytarphata urticifolia* inhibits carbohydrate digestion and absorption by inducing moderate inhibitory effects on amylase, glucose oxidase, and glucose uptake. There was no doubt that *S. urticifolia* is capable of reducing complications associated with diabetes and these findings suggest that it may be useful for patients with diabetes who need to mitigate these complications.

Phytochemically, *S. urticifolia* was thought to be rich in phytochemicals, particularly flavonoids and phenolics, which have been shown to have anti-hyperglycemic, insulin-sensitizing, as well as beta-cell protective effects, contributing to its anti-diabetic properties. Moreover, *S. urticifolia* has shown that its antioxidant activity may contribute to reducing inflammation as well as oxidative stress, which have both been implicated in the development of diabetes. There is no doubt that from a phytochemical standpoint, *S. urticifolia* is a promising candidate when it comes to anti-diabetic interventions because of its ability to regulate blood sugar levels and enhance glucose tolerance, as well as its capability to enhance blood glucose tolerance.

CONCLUSION:

As part of this study, we examined *S. urticifolia*, a well-known traditional plant, for its phytochemical composition and potential anti-oxidant and anti-diabetic effects. The anti-oxidant potential of *S. urticifolia* and its moderate anti-diabetic activity were also found in our study. Based on the results of this study, *S. urticifolia* might be of benefit to diabetic patients as a therapeutic agent.

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