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Research Article

POTENTIAL USE OF PLANT EXTRACTS OF *ALBIZIA SAMAN* AS AN ANTI-DIABETIC AGENT

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ABSTRACT

Diabetes mellitus is the capital metabolic syndrome of the world. Currently India represents 49% of the world diabetes burden. Diabetic patients encounter considerable morbidity and mortality from various dreadful complications. To have a check over the rising diabetic status, good metabolic control, long lasting changes in lifestyle and pharmacological treatments are required. Chronic use of allopathic drugs results in side effects and an alternate medicine is sought through herbal drugs. Based on the framed objective, the present study focuses on identifying a hypoglycemic agent using plant extract. The aim of the present investigation is to evaluate and identify the active ingredients present and explore the functional groups in 50% hydroethanolic extracts of leaf and bark of *A. saman* through techniques like GC-MS and FTIR. To substantiate, in vitro inhibitory assay of the plant extracts using alpha- Amylase, alpha-Glucosidase and Glucose diffusion inhibition assays are carried out to confirm the presence of any significant anti-diabetic effect. The results provided a scientific validation of the folklore use of *Albizia saman* and suggested that this plant (leaf and bark) has promising therapeutic activity for the maintenance of diabetes mellitus.

Keywords: Morbidity, mortality, chronic, allopathic drug, hypoglycemic agent, folklore.

INTRODUCTION

Diabetes mellitus is the most common endocrine disorder affects more than 400 million people around the world and this count may increase to more than 640 million in 2040 worldwide. It is a group of metabolic disorders characterized by hyperglycemia and abnormalities in carbohydrate, protein and fat metabolism and is one of the five leading cause of death in the world and typically there are two categories namely, Type I and Type II Diabetes.[1]

Prevention of diabetes and its complications is not only a major challenge for the future, but is essential if health is to be an attainable target and strongly emphasize the optimal, rational use of traditional and natural indigenous medicines. The morbidity from type 2 diabetes predominantly relates to its micro vascular and macro vascular complications. Patients with type 2 diabetes are at higher risk of stroke and cardiovascular disease as well as renal impairment, retinopathy and peripheral nerve damage.

Oxidative stress is currently suggested as the mechanism underlying diabetes and its complications. Consequently, antioxidant therapy has been thought to be effectual for the prevention and treatment of various diseases including diabetes in recent times[2].

Herbal medications are the most widely and commonly preferred method to control blood sugar. Various Indian medicinal herbs have been scientifically validated for their efficacy and safety. Plant- based medicine is gaining momentum and takes an upper hand over hypoglycemic drugs. In recent years, medicinal plants have been extensively investigated for the treatment of various diseases and fewer side effects on its use are catching a trend all over the world[3]. Herbal remedies for treating diabetes possess hypoglycemic effect, decreases the elevated plasma glycemic level by restoring the activity of pancreas and reduce the intestinal absorption of glucose. They have abundant levels of glycosides, alkaloids, flavonoids, terpenoids, carotenoids, etc and altogether show anti-diabetic effects[4]. In addition, some herbal alternatives provide symptomatic relief, prevent secondary complications, regenerate beta cells, overcome resistance, possess high antioxidant activity, lower cholesterol levels and maintain a steady blood sugar levels. Chronic use of allopathic drugs results in side effects and an alternate medicine is sought through herbal drugs[5]. Based on the framed objective, the present study focuses on identifying a hypoglycemic agent using plant extract.

The choice of plant targeted in this study *is Samanea saman (Jacq) Merr* also called *Albizia saman* belongs to the family Leguminosae and is popularly called as Rain tree or Monkey pod. *Albizia* species is a socially significant large tropical tree which usually reaches a height of 50-80 feet with rough wrinkled bark.

Literature reveals that it has been widely used in folklore medicine as a remedy for various disease treatments[6]. The present investigation focuses on the exploitation of the different parts of the plant species as a hypoglycemic agent and validating through in vitro experiments.

The phytoconstituents present in the leaf and bark extracts were quantitatively analyzed and found to contain a good concentration of cardiac glycosides, phenols and saponin as major phytochemical constituents making the plant of high medicinal importance. The future perspective demands the isolation and identification of the active principle and encourages further research of its potential bioactive compounds for the evolution of

preventive health care without any harmful side effects. Therefore, the study has been initiated to characterize the active ingredients and analyze the biological activities of the compounds identified through GC-MS analysis and to explore the functional groups present through FTIR analysis in the 50% hydroethanolic leaf and bark extracts. In vitro inhibitory activity of the 50% hydroethanolic extracts of leaf and bark of *A. saman* on alpha-Amylase, alpha-Glucosidase and Glucose diffusion inhibition assays are also carried out to find out the presence of any significant anti-diabetic effect in the plant extracts. Thus, the present study becomes conclusive for validating the parts of the plant material *Albizia saman* as an anti-diabetic source.

MATERIALS AND METHODS

Based on the objective set, the methodology adopted for the present work on "Potential use of plant Extracts of *Albizia saman* as an Anti-Diabetic Agent" was discussed.

Collection of Plant Material

Fresh leaves and bark of *Albizia saman* were collected from Aravind Nagar, Puduchatram of Dindigul district and the specimen were deposited and authenticated by Botanical Survey of India, Southern Regional Centre, T.N.A.U. Campus, Coimbatore. **BSI/SRC/5/23/2016/Tech/1948**- *Albizia saman (Jacq.) Merr*. (Basionym: Mimosa saman Jacq.)-Mimosaceae.

Extraction and Storage

100 g of the coarse powder of leaf and bark was then subjected to extraction in 600 ml of 50% hydroethanol solvent by using Soxhlet apparatus[7]. Soxhlet method of extraction was the preferred method and was employed wherein instead of many portions of warm solvent being passed through the sample; just one batch of solvent is recycled to obtain the two hydroethanolic extracts of leaf and bark. The collected extracts were evaporated to dryness in a rotary evaporator and were stored in an air-tight container and used for further analysis.

Identification of the Active Ingredient of the Plant Materials

GC–MS Analysis

GC–MS analysis was carried out using a high resolution, double focusing instrument JEOL GCMATE II GC-MS (Agilent Technologies 6890 N) with data system whose maximum resolution is 6000 and maximum calibrated mass is 1500 Daltons. 2µl of the 50% hydroethanolic extract of the leaf and bark of *Albizia saman* was subjected to GC and MS analysis. The data were obtained on a fused silica column (HP5) of 50 m x 0.25 mm I.D. Analysis conditions were 20 min at 100°C, 3 min at 235°C for column temperature, 240°C for injector temperature. Helium was the carrier gas used with a flow rate of 1ml/min and split ratio of 5:4. 1µl of the sample was evaporated in a split less injector at 300°C and the run time was 25 min. The active ingredients were identified by gas chromatography coupled with mass spectrometry[8].

Identification of Phytocompounds

The molecular weight and structure of the components of test materials were ascertained by interpretation of mass spectrum of GC-MS using the database of National Institute Standard and Technology (NIST) having more than 62,000 spectral patterns. The mass spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library database. The identity of the spectra above 95% was needed for the identification of components. The name, molecular weight and structure of the components of the leaf and bark extract were ascertained. The relative percentage amount of each component was calculated by comparing its average peak area with the total area[9].

FTIR Analysis

Fourier Transform Infrared Spectrophotometry (FTIR) is the most useful technique for identifying the functional groups in the extracts. It utilizes the principle that different substances will absorb Infrared light with a unique pattern[10].

The 50% hydroethanolic extracts of the leaf and bark were milled with potassium bromide (KBr) to form a very fine powder. This powder was then compressed into a thin pellet which can be analyzed. The technique was performed in the spectral range of 4000-400 cm^{-1} with resolution on 4 cm^{-1}. (Shimadzu IR prestige-21V FTIR).A small drop of powder was placed on the KBr plate and a second plate was placed over it to obtain a nice thin film. Then the plate was placed in a sample holder and the spectral analysis was carried out. The results were recorded on a chart and this chart can be compared to drug standards or a library database.

In vitro Evaluation Of The Anti-Diabetic Activity

The anti-diabetic activity of the 50% hydroethanolic extract of leaf and bark was evaluated by various in vitro methods.

Alpha-Amylase Inhibitory Assay

In α-amylase inhibition method, the enzyme solution was prepared by dissolving α-amylase in 20mM phosphate buffer (6.9) at the concentration of 0.5mg/ml. 1ml of the leaf and bark extract of various concentrations (20, 40, 60, 80 & 100 µg/ml) and 1ml of enzyme solution were mixed together and incubated at 25°C for 10min. After incubation, 1ml of starch (0.5%) solution was added to the mixture and further incubated at 25°C for 10min. The same was performed for the control where 1ml of enzyme was replaced by the buffer. The reaction was then stopped by adding 2ml of dinitrosalicylic acid (DNS, color reagent), heating the reaction mixture in a boiling water bath (5min). After cooling, the absorbance was measured colorimetrically at 565 nm[11].
The inhibition percentage was calculated using the given formula,

Inhibition (%) = Abs_{565} (control) – Abs_{565} (extract) / Abs_{565}(control) * 100

The IC 50 values were determined from plots of percent inhibition versus log inhibitor concentration. Acarbose was used as the reference alpha amylase inhibitor. All tests were performed in triplicate.

Alpha-Glucosidase Inhibitory Activity

In this method, different concentrations of leaf and bark extracts were incubated with 10ml of enzyme solution for 1o mins at 37°C. Maleate buffer solution of pH 6.0 is used to make the volume. The enzyme reaction is started by adding solution of p-nitrophenyl alpha D-glucopyranoside and further incubated at 37°C for 30 min. The reaction is terminated by treating the mixture in boiling water bath for 5 min. After the addition of 0.1M disodium hydrogen phosphate solution, the absorbance of liberated p-nitrophenol is read at 400nm with acarbose as the standard[12].

Glucose Diffusion Inhibition Assay

This assay was performed by taking 2 ml of 0.15 M NaCl containing 0.22mM D-glucose loaded into a dialysis tube (6 cm×15 mm), containing 50% hydroethanolic extract of leaf and bark and the dialysis tube was sealed. The sealed tube was then placed in a centrifuge tube containing 45 ml of 0.15 M NaCl and kept in an orbital shaker at room temperature at 100 rpm. The diffusion of glucose into the external solution was monitored by

measuring the glucose concentration in the external solution every 60min by Glucose Oxidase kit method[13].

RESULTS

Based on the objective in assessing the phytopotential of the leaf and bark extract of *Albizia saman* (Rain Tree), the study has been executed and the findings of the study is discussed extensively.

Studies are conducted using the leaf and bark extracts of *Albizia saman* (Rain tree). Comparison on using leaf and bark extract as an anti-hyperglycemic agent was validated. The results of the study shall emphasize the commercial use of the best material (Leaf and bark) of the tree for pharmaceutical companies in the production of new hypoglycemic agents for the treatment of diabetes.

GC-MS Profile of Hydroethanolic Extract of Leaf and Bark of *Albizia Saman*

Mass spectrometry has become a vital tool in the hands of organic chemists and biochemists because of its potential to supply definitive, qualitative and quantitative information on molecules based on their structural compositions. Gas chromatography attached to a Mass spectrometer (GC-MS) enables mixture of small molecules to be separated and analyzed.

The hydroethanolic extract of leaf and bark were subjected to gas chromatography - mass spectrum analysis. Figure no. 1 and 2 shows the GC-MS spectral of the hydroethanolic extracts of leaf and bark of *Albizia saman* and Table no. 1 and 2 indicates the active ingredients present in the extracts. The biological activities of compounds identified by GC-MS are represented in Table 3.

FTIR Analysis

FTIR spectroscopy is a rapid, non-invasive, high resolution diagnostic tool for identifying the types of bonds in a molecule by creating an infrared absorption spectrum that resembles a molecular fingerprint. The spectrum obtained was used to identify the functional groups of the active components present in the leaf and bark extracts, based on the peak values in the region of IR radiation. The hydroethanolic extract subjected to FTIR analysis was separated based on its peak ratio. FTIR analysis of 50% hydroethanolic extracts confirmed the presence of alcohol, alkenes, alkanes, aldehydes, amines, aromatic and nitro compounds. FTIR analysis of the extracts was carried out with spectral frequency range of 4000-750 cm^{-1}. The FTIR absorption spectrum was represented in Figure 3 and 4 and the Functional groups present in the leaf and bark extract were shown in Table 4 and 5.

In vitro Evaluation of the Anti-Diabetic Activity

This phase of study was carried out to evaluate the in vitro inhibitory effect of the hydroethanolic extracts of leaf and bark of *A. saman* on alpha- Amylase, alpha-Glucosidase and Glucose diffusion inhibition assays.

Alpha-Amylase Inhibitory Assay

Inhibition of alpha-amylase varied from 19.33 to 55.34% in the concentration range of 20 to 100µg/ml in the bark extract. IC_{50} value of the bark extract was found to be 87.41 µg /ml. The hydroethanolic leaf extract of *A. saman* exhibited α-amylase inhibitory activity ranging from 13.34 to 48.53 % with an IC_{50} value 99.21µg/ml. Acarbose (at a concentrations 100 µg/ml) showed 60.51% inhibitory effects on the α-amylase activity with an IC_{50} value 79.8 µg/ml indicated in Table 6.

Alpha-Glucosidase Inhibitory Activity

Hydroethanolic leaf extract (at a concentration 100 µg/ml) showed 55.7% inhibitory effects on the α-amylase activity with an IC_{50} value 88.6 µg/ml. Whereas the hydroethanolic bark extract (at a concentrations 100 µg/ml) showed 68.7% inhibitory effects on the α-amylase activity with an IC_{50} value 69.7 µg/ml. The standard Acarbose (at a concentrations 100 µg/ml) showed 79.8% inhibitory effects on the α-amylase activity with an IC_{50} value of 59.7µg/ml represented in Table 7.

Glucose Diffusion Inhibition Assay

The assay revealed that the hydroethanolic bark extract of *A. saman* significantly decreased the glucose movement across the membrane and showed a glucose concentration of 989.5mg/ml when compared to the control which indicated a glucose concentration of 1253.6mg/ml in the external solution at the end of 5 hr. Hydroethanolic bark extract significantly decreased the glucose movement across the membrane when compared to the leaf extract which showed a glucose concentration of 1150.5mg/ml. The results of the glucose diffusion assay were depicted in Table 8.

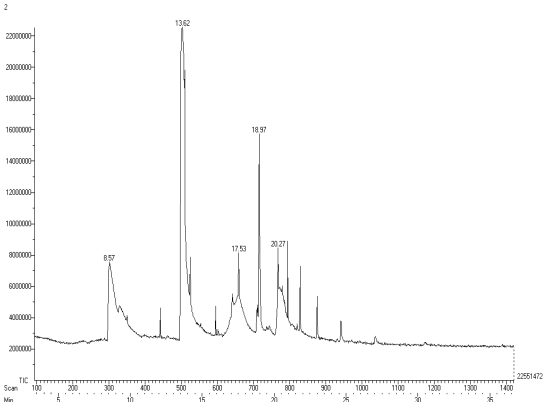

Figure 1: GC MS analysis of hydroethanolic extract of leaf of *A.saman*

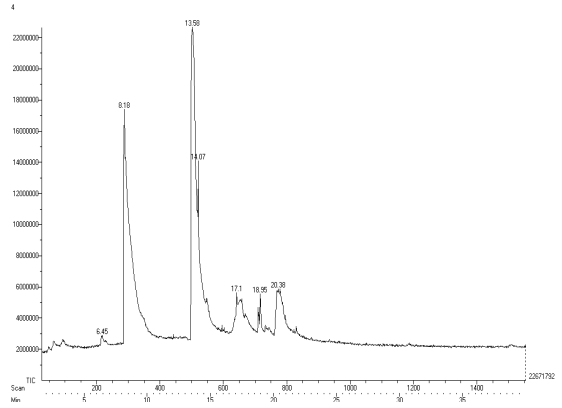

Figure 2: GC MS analysis of hydroethanolic extract of bark of *A.saman*

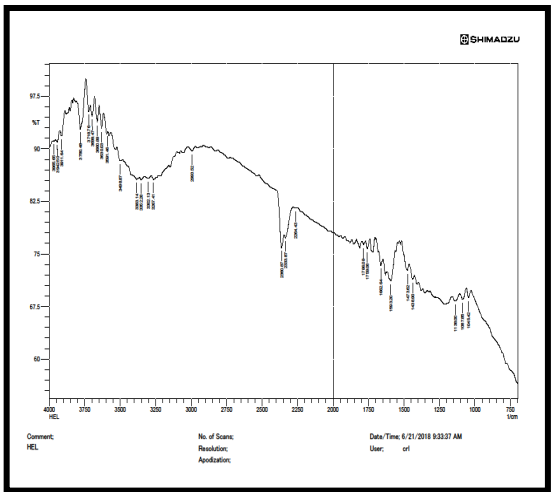

Figure 3: FTIR absorption spectrum of the hydroethanolic extract of leaf

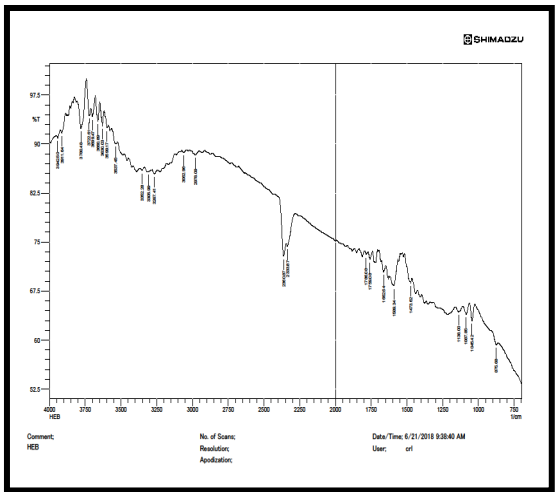

Figure 4: FTIR absorption spectrum of the hydroethanolic extract of bark

Table 1: GC- MS Analysis of the Hydroethanolic Extract of Leaf of *Albizia saman*

RT	Name of the compound	Molecular formula	Molecular weight	Peak area %	Type of compound
8.57	2-Amino benzoyl hydrazide	$C_7H_9N_3O$	151.17	12.16	Heterocyclic compound
13.62	Diethyl phthalate	$C_{12}H_{14}O_4$	222.24	64.14	Ester
17.53	Z,E-2-methyl-3,13-octadecadien-1-ol	$C_{19}H_{36}O$	280.49	4.05	Alcohol
18.97	Phytol	$C_{20}H_{40}O$	296.54	13.43	Diterpene alcohol
20.27	Oxiraneundecanoic acid-3-pentyl-methyl ester trans	$C_{19}H_{36}O_3$	312.49	6.223	Ester

Table 2: GC- MS Analysis of the Hydroethanolic Extract of bark of *Albizia saman*

RT	Name of the compound	Molecular formula	Molecular weight	Peak area %	Type of compound
06.45	L-Glucose	$C_6H_{12}O_6$	180.16	1.82	Aldohexose Monosaccharide
08.18	Benzoic acid, 2-amino methyl ester	$C_8H_9NO_2$	151.25	35.90	Ester
13.60	Phthalic acid diethyl ester	$C_{12}H_{14}O_4$	222.24	6.23	Ester
14.07	Benzoxazole-3-acetic acid,2,3-dihydro-2-oxo-ethyl ester	$C_9H_9NO_3$	221.19	31.27	Ester
17.10	6-O-methyl 2,4-methylene-a-sedoheptitol	$C_9H_{18}O_7$	238.24	11.37	Alcohol
18.95	Phytol	$C_{20}H_{40}O$	296.68	13.04	Diterpene alcohol
20.42	9-oximino-2,7-diethoxyfluorene	$C_{17}H_{17}NO_3$	283.32	0.37	Alkene

Table 3: Biological activities of compounds identified through GC-MS

Name	Activity	References
Diethyl phthalate	Antimicrobial Antifungal	Shashikala *et al.,*2016 Wyson e*t al*., 2016
Phytol	Anti oxidant Antimicrobial Anticancer Anti-inflammatory Anti-diuretic Immunostimulatory Anti-diabetic	Camila *et al.,* 2013 Venkataraman *et al.,* 2012 Vijayalakshmi *et al.,* 2011 Elmar *et al.,* 2013
9-oximino-2,7-diethoxyfluorene	Bacteriostatic Anti-pathogenic	Olawale *et al*., 2017 Hussein, 2016

Table 4: FT-IR Analysis of hydroethanolic extract of Leaf

Frequency (cm^{-1})	Bond	Functional group
1138	C-O stretch	Alcohol, carboxylic acid, ester, ether
1438	C-C stretch	Aromatics
1593	N-H bend	Primary amine
1662	-C=C- stretch	Alkene
1759	C=O Stretch	Carboxylic acid
2264	-CΞC- stretch	Alkyne
2993	C-H stretch	Alkane
3267	O-H stretch	Carboxylic acid, Alcohols, Phenols
3302	-CΞC-H:C-H stretch	Alkyne (terminal)
3352	N-H stretch	Primary & secondary amine, amides
3498	O-H stretch, H- bonded	Alcohols, Phenols
3630	O-H stretch, free hydroxyl	Alcohols, Phenols

Table 5: FT-IR Analysis of hydroethanolic extract of Bark

Frequency (cm^{-1})	Bond	Functional group
875	C-H	Aromatics
1087	C-O stretch	Alcohol, carboxylic acid, ester, ether
1138	C-N stretch	Aliphatic amine
1473	C-C stretch	Aromatics
1589	N-H bend	Primary amine
1662	-C=C- stretch	Alkene
1759	C=O Stretch	Carboxylic acid
2978	C-H stretch	Alkane
3062	=C-H stretch	Alkene
3267	N-H stretch	Primary & secondary amine, amides
3305	-CΞC-H:C-H stretch	Alkyne (terminal)
3352	N-H stretch	Primary & secondary amine, amides
3630	O-H stretch, free hydroxyl	Alcohols, Phenols

Table 6: Alpha-Amylase inhibitory assay

Alpha-Amylase inhibitory assay			
Concentration of the extract (µg /ml)	50% Hydroethanolic extract of leaf	50% Hydroethanolic extract of bark	Acarbose
20	13.34±0.21	19.33±0.45	21.41±0.35
40	22.98±0.53	26.81±0.61	29.05±0.39
60	30.45±0.25	35.72±0.33	39.74±0.19
80	40.37±0.70	43.33±0.45	47.02±0.27
100	48.53±0.39	55.34±0.48	60.51±0.56
IC_{50}	99.21	87.41	79.8

Values are in mean ± SD (n=3)

Table 7: Alpha-Glucosidase inhibitory activity

Alpha-Glucosidase inhibitory activity			
Concentration of the extract (µg /ml)	50% Hydroethanolic extract of leaf	50% Hydroethanolic extract of bark	Acarbose
20	12.2±0.14	16.78±0.25	19.35±0.33
40	21.8±0.38	29.5±0.19	34.87±0.53
60	36.2±0.17	45.6±0.41	52.78±0.58
80	44.5±0.48	58.2±0.55	69.4±0.42
100	55.7±0.34	68.7±0.21	79.8±0.57
IC_{50}	88.6	69.7	59.7

Values are in mean ± SD (n=3)

Table 8: Glucose diffusion inhibition assay

	Glucose diffusion inhibition assay		
	Glucose concentration in external solution (mg/ml)		
Time (hr)	Control	50% Hydroethanolic extract of leaf	50% Hydroethanolic extract of bark
0	192.2±0.85	97.4±0.9	62.35±1.3
1	304.3±1.1	254.7±1.15	203.2±1.55
2	555.2±1.21	470.7±1.3	380.6±0.99
3	826.4±0.89	710.7±0.79	605.7±0.63
4	1109.1±1.23	1008.2±1.56	807.2±1.45
5	1253.6±1.23	1150.5±0.87	989.5±0.55

Values are in mean ± SD (n=3)

DISCUSSION

The present research work evaluated and identified the active ingredients present and explored the functional groups in the 50% hydroethanolic extracts of leaf and bark through techniques like GC-MS and FTIR.

In GC-MS analysis, identification of the compounds is based on the peak area, retention time, molecular formula and molecular weight. The major compounds identified by GC- MS of leaf extract includes five compounds namely 2-Amino benzoyl hydrazide, Diethyl phthalate, Z,E-2-methyl-3,13-octadecadien-1-ol, Oxiraneundecanoic acid-3-pentyl-methyl ester trans and phytol; and that of bark extract includes seven compounds, namely L-Glucose, Benzoic acid- 2-amino methyl ester, Phthalic acid diethyl ester, Benzoxazole-3-acetic acid, 2, 3-dihydro-2-oxo-ethyl ester, 6-O-methyl 2,4-methylene-a-sedoheptitol, 9-oximino-2,7-diethoxyfluorene and phytol.

The biological activities of compounds identified by GC-MS include:

The GC-MS analysis of the identified compounds from the hydroethanolic extract of the leaf and bark of *Albizia saman* possess specific activities such as anti-fungal, anti-oxidant, antimicrobial, anticancer, anti-inflammatory, anti-diuretic, immunostimulatory, anti-diabetic, bacteriostatic and anti-pathogenic activities. The presence of phytol in both leaf and bark extract revealed that they possess anti-diabetic activity and could be exploited for the development of hypoglycemic agent to treat diabetes.

In FTIR analysis, spectral reflectance of the 50% hydroethanolic leaf extract showed the presence of alcohols and phenols group with bands at 3267cm^{-1}, 3498cm^{-1} and 3630cm^{-1} (O-H stretch, H-bonded) . A peak at 1138 cm^{-1} and 1759cm^{-1} indicates the presence of carboxylic acid, ester and ether (C-O stretch). The prominent peak at 1438 cm-1 reveals the presence of functional group like aromatics (C-C stretch). The peak at 1593cm^{-1} and 3352cm^{-1} is assigned for primary & secondary amines and amides (N-H bend, N-H stretch). A peak at 1662 cm^{-1}, 2264 cm^{-1} & 3302 cm^{-1} and 2993 cm^{-1} indicates the presence of Alkene(-C=C- stretch), Alkyne (-CΞC- stretch, -CΞC-H:C-H stretch) and Alkane (C-H stretch) respectively.

Spectral reflectance of the 50% hydroethanolic leaf extract showed the presence of alcohols and phenols group with bands at 3630cm^{-1} (O-H stretch). A peak at 1087 cm^{-1} and 1759cm^{-1} indicates the presence of carboxylic acid, ester and ether (C-O stretch). The prominent peak at 1473 cm^{-1} and 875 cm^{-1} reveals the presence of functional group like aromatics (C-C stretch, C-H "oop"). The peak at 1138cm^{-1} and 1589cm^{-1}, 3267cm^{-1} & 3352cm^{-1} is assigned for aliphatic amine (C-N stretch) and primary amine (N-H bend) & secondary amines and amides (N-H stretch) respectively. A peak at 1662 cm^{-1} (-C=C- stretch) & 3062 cm^{-1} (=C-H stretch) indicates the presence of Alkene , 3305 cm^{-1} and 2978 cm^{-1} reveals the presence of Alkyne (-CΞC-H:C-H stretch) and Alkane (C-H stretch) respectively. These functional groups in the plant extracts of *Albizia saman* represent the presence of active constituents responsible for their pharmacological activities.

In vitro inhibitory activity of the 50% hydroethanolic extracts of leaf and bark of *A. saman* on alpha- Amylase, alpha-Glucosidase and Glucose diffusion inhibition assays carried out confirmed the presence of significant anti-diabetic effect in both the plant extracts.

Alpha amylase is an enzyme that hydrolyses alpha-bonds of polysaccharide to yield high levels of glucose and maltose. Alpha amylase inhibitors bind to **α**-bond of polysaccharide and prevent break down of polysaccharide into mono and disaccharide. In vitro inhibitory assay of α-amylase was performed with different concentrations of the leaf and bark extract ranging from 20 to 100 µg /ml. From the data obtained, it was found that hydroethanolic extract of the bark efficiently inhibited alpha amylase enzyme in vitro. There was a dose dependent increase in percentage inhibitory activity against alpha amylase and the inhibition. The present study indicated that the bark extract of *Albizia saman* could be useful in the management of postprandial hyperglycemia by effectively inhibiting the alpha- amylase activity. The results followed the same pattern of findings observed in the methanol extract of *S. brevistigma* in significantly inhibiting the activity of α-amylase with IC_{50} value of 250µg/ml and might be considered as a potential source of natural anti-diabetic agents.

The inhibitory activities of methanolic extracts of *Artocarpus altilis*, *Cinnamomum zeylanicum*, *Piper betel* and *Artocarpus heterophyllus* on wheat alpha amylase and Baker's yeast alpha glucosidase at varying concentrations were shown and clearly indicated the potential of these extracts to manage hyperglycemia. Our findings also revealed that the hydroethanolic extracts of the bark and leaf efficiently inhibited alpha-glucosidase enzyme in vitro. There was a dose dependent increase in percentage inhibitory activity against alpha-glucosidase by both the plant extracts. Thus the results clearly depicted that the hydroethanolic bark extract of *A. saman* showed the greater % inhibition of the alpha glucosidase enzyme compared to the other plant extract.

At the beginning of the glucose diffusion inhibitory assay, glucose concentration in the dialysis bag was 0.22mM D. The level of inhibition of glucose diffusion into the external solution from the dialysis bag by the plant extracts at various intervals of time ranging from 0 to 5 hr were assayed and compared with the control in the absence of the plant extract. The data clearly suggested that hydroethanolic bark extract was significant in inhibiting glucose diffusion which in turn states that the plant is capable of regulating glucose movement out of the cells into the blood stream thereby controlling post prandial glucose levels. The leaves of *G. hirsuta* were subjected to various in vitro assays like glucose diffusion and results of the study suggested that *Grewia hirsuta* possesses significant hypoglycemic potential. The expected bioactive components could be flavonols or phenolic acids as literature shows a clear link between polyphenols and anti-diabetic activity of herbal extracts.

CONCLUSION

From the findings, results can be concluded that the 50% hydroethanolic extract of bark had better control on glycogenolysis than the leaf extract. This could be validated from the above results that the bark extract inhibited alpha amylase and alpha glucosidase activity and this is further substantiated through the glucose diffusion inhibition assay. From the above in vitro assays, it could be suggestive of bark extract possessing better anti-diabetic and hypoglycemic activity than the leaf extract. Thus in the future, pre-clinical studies using animal models can be carried out to validate the anti-diabetic property of the plant extract of *Albizia saman*. It may be concluded that *Albizia saman* shall be considered as a promising plant with various therapeutic properties and can be further explored in curing various diseases.

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