



PRELIMINARY PHYTOCHEMICAL SCREENING AND ANTIOXIDANT PROPERTY OF VARIOUS EXTRACTS OF *ALBIZIASAMAN* LEAVES



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Abstract

Albizia saman is very important medicinal plant, it is used in many ayurvedic preparations. The medicinal plant having anti-asthma, anti-diarrheal, anti-fungal activity etc. In the current research work carried out to find the phytochemical constituents and antioxidant property of various extracts of *Albizia saman* leaves were performed. The leaves were shade dried and powdered. The powdered leaves were defatted with Petroleum Ether in Soxhlet apparatus and then extracted with absolute methanol for 24 hours separately in Soxhlet apparatus. The results of preliminary phytochemical screening of the methanolic extract of *Albizia saman* revealed the presence of alkaloids, glycosides, steroids, flavonoids, saponins, tannins, carbohydrates and reducing sugar. The antioxidant activity of the methanolic extract was measured in terms of reducing power and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging capacity and the total antioxidant capacity of the methanolic extract of *Albizia saman* were determined by phosphomolybdenum method, the result shows the highest peak of free radical scavenging and antioxidant activity. The result supports the use of *Albizia saman* in traditional medicine for anti-diabetic activity.

INTRODUCTION

Albizia saman formerly called *Samanea samam* (family Leguminosae), is a fast growing tree commonly used as a pasture, shade or for ornamental purposes. The leaves, bark, root, seeds and pods of the tree are used in traditional system of medicine. The alcoholic extract of the leaves inhibited *Mycobacterium tuberculosis*, the alkaloid fraction of leaves is effective on the CNS and its infusion is used as laxative. Seeds are chewed for sore throat. A decoction of the inner bark and fresh leaves are used for the treatment of diarrhoea, colds and intestinal ailments¹. The chemical literature search indicated that *Albizia saman* contains alkaloids, glycosides, terpenoids etc². The mature pods of *Albizia saman* are black brown, oblong, lumpy, 10 to 20 cm long, 15 to 19 mm wide, 6 mm thick, straight or slightly curved, not dehiscent but eventually cracking irregularly, and filled with a sticky, brownish pulp that is sweet and edible³. The bottom line is that the pods of the *Albizia saman* tree are rarely used as a plant part source for herbs. Therefore, information on the phytochemical components and biological activity of the

Pods of *Albizia saman* tree is scanty. Knowledge of the phytochemical constituents of plant parts is required in understanding the basis for any therapeutic effect. For instance, the flavonoids, which are ubiquitous in higher plants and common part of human diet⁴ could significantly inhibit microbes which are resistant to conventional antibiotics⁵. Recently, isolated flavonoids were reported to exhibit antimicrobial activity⁶. In addition, flavonoids through their free-radical scavenging activity have evoked multiple biological functions, including vasodilatory, anti-carcinogenic, anti-inflammatory, anti-bactericidal, immune stimulatory, anti-allergic and anti-viral functions⁷. Phytochemicals are naturally occurring biochemicals in plants that help to give plants their characteristic colour, flavour, smell and texture. Apart from that, phytochemicals could prevent diseases (including cancer and cardiovascular diseases) and inhibit pathogenic microorganisms⁸. Indeed, the proper composition of phytochemicals appears to confer plants and plant parts with peculiar medicinal properties. This might explain the heightened routine interest to screen

higher plants for active agents with antifungal and antimicrobial/antibacterial activities against plant and human pathogens.

Tannins (polyphenols with widely varying chemistry) are one of the major phytochemicals found in many higher plants. Tannins have a characteristic strange smell and astringent taste and could bind to proteins (and consequently precipitate proteins) through the effective formation of strong complexes with proteins and other macromolecules. Thus, they could have a major impact on animal nutrition, including inhibition of growth rate digestive enzymes⁹. Tannins have been implicated with various pharmaco-therapeutic effects¹⁰. Considering the immense medicinal property of Leguminosae, the present study was carried out the phytochemical constituents of various extracts of *Albizia samanto* compare the free radical scavenging activity and total activity among the various extracts *Albizia saman*.

MATERIALS AND METHODS

Plant material collection

The fresh *Albizia saman* leaves were collected from Ramanathapuram. Selected

samples were authenticated by Botanical Survey of India, Coimbatore.

Preparation of extract

The leaves were shade dried at room temperature for 10 days. The dried leaves were ground to powder with a help of amechanical grinder. The powder was sieved through a 1 mm mesh and was stored in an air-tight container for future use. To 20 g of the powdered leaves the different solvents were separately added and then extracted using Soxhlet extractor for 2 h. The Benzene, Chloroform, Ethanol, Ethylacetate, Methanol, Petroleum ether were separately concentrated using rotary evaporator and then preserved individually at 5°C in an air tight bottle until used for the phytochemical and antioxidant activity.

Phytochemical screening

The phytochemical analysis of various extracts of *Albizia saman* leaves were carried out by standard Procedure^{11&12}.

Determination of Free Radical Scavenging Activity (DPPH Radical Scavenging Activity)

Free radical scavenging is one of the mechanisms involved in antioxidant action, a good antioxidant (AH) able to scavenge

the DPPH radical and retain its own stability due to its reduction ability as shown in the equation below¹³.



The scavenging activity of the extract was measured according to the method¹⁴. Briefly, 6×10^{-5} M solution of DPPH in methanol was prepared. A 100 μl of DPPH solution was added to 100 μl of samples or standard prepared in methanol in different concentrations. Only samples from the ethanol extract were analyzed in this study. The assay was carried out in a 96 well microplate with each concentration was performed in triplicate. After 30 minutes, the absorbance of mixture was measured at 517 nm using microplate reader. A control was 100 μl of 6×10^{-5} M DPPH in methanol + 100 μl methanol. The DPPH radical scavenging activity was calculated according to the following equation

$$\% \text{ inhibition} = \left[\frac{(\text{Ac}-\text{Ae})}{\text{Ac}} \right] 100$$

Where Ac is the absorbance of the control and Ae is the absorbance of the extract/standard.

A graph of % inhibition against the plant extract concentration were plotted to

determine the IC_{50} , which is the concentration of the extracts necessary to inhibit 50% of the initial DPPH concentration. The IC_{50} of each extractswere compared with standards; BHT and Quercetin.

Determination of Antioxidant Capacity by Phosphomolybdenum Method:

The antioxidant activity of the methanolic extract was determined by the phosphomolybdenum method as described by Prieto¹⁵. Briefly, 0.3 ml of extract was mixed with 3 ml of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). The reaction mixture was incubated at 95 °C for 90 min and cooled to room temperature. Finally, absorbance was measured at 695 nm using a spectrophotometer¹² against blank. Methanol (0.3 ml) in place of extract was used as the blank. The total antioxidant capacity was expressed as the number of equivalents of ascorbic acid (AAE) in micrograms per milligram of extract¹⁶.

Total antioxidant activity

$$= 100 \left[1 - \frac{(\text{Ao}-\text{At})}{(\text{Aoo}-\text{Ato})} \right]$$

Where A_0 is the OD of the sample at time to minutes and A_t is the time of the sample at time $t = 90$ minutes. A_0 and A_t represent the OD of the control at time $t = 0$ minutes and $t = 90$ minutes respectively.

RESULTS AND DISCUSSION

Qualitative analysis of the presence of phytochemicals

The phytochemical screening of the various extract of *Albizia saman* indicates the presence of saponins, steroids, alkaloids, flavonoids, tannins, terpenoids and glycosides.

Methanolic extract of *Albizia saman* gave maximum extraction of phytochemicals than any other extract. Methanolic extract was followed by ethanolic extract for the presence of phytochemical constituents. Carbohydrates, flavonoids, tannins, terpenoids and phenols were present in all the extracts. Phytochemical constituents of various extracts of *Albizia saman* shown in table 1.

Free Radical Scavenging Activity of *Albizia saman*

The DPPH scavenging activities of various extracts such as benzene, chloroform,

ethanol, ethyl acetate, methanol and petroleum ether of the selected samples were analyzed to estimate their antioxidant properties. Figure 1 indicates the DPPH radical scavenging activity of various extracts of *Albizia saman*.

Methanolic extract of *Albizia saman* showed the highest DPPH scavenging activity which was followed by ethanol and chloroform extracts. This may be due to the extraction of maximum phytochemicals in methanolic extract. The least activity was noted in petroleum ether extract.

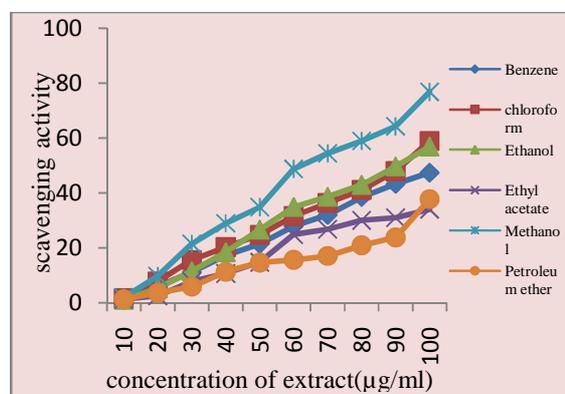


Figure 1 (DPPH scavenging activity)

Total Antioxidant Activity of *Albizia saman*

Plants have good antioxidant ability and are safer than the synthetic antioxidants¹⁷. Secondary metabolites from medicinal

plants function as small molecular weight antioxidants, but their particular mechanism of action are variable and depend both on the structure and environment¹⁸. Antioxidants can significantly delay or prevent the oxidation of easily oxidizable substances¹⁹. The major action of antioxidants in cells is to prevent damage due to the action of reactive oxygen species.

The present investigation has shown the total antioxidant potential of various extracts of *Albizia saman* (Figure 2)

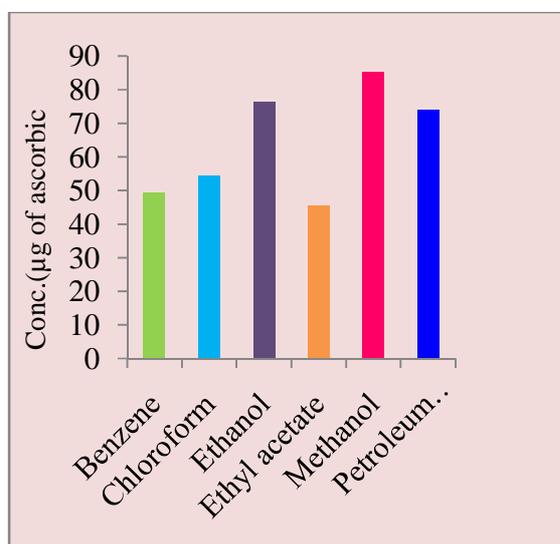


Figure 2(Total antioxidant activity)

Methanolic extract of *Albizia saman* exhibited strongest antioxidant activity (85.24 µg of ascorbic acid/mg of extract) among all the other extracts, while ethyl acetate extract yielded the lowest (45.5 µg of ascorbic acid/mg of extract).

CONCLUSION

The preliminary qualitative phytochemical screening of *Albizia samanis* found to contain phytochemicals namely, tannins, flavonoids, steriods, saponins, cardiac glycosides and terpenoids. It may be concluded that *Albizia saman* is a rich source of phytochemical and has antioxidant property. Phytochemical were extracted best in methanol among the solvents and methanolic extract showed the maximum DPPH and total antioxidant activities.

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Table 1

Phytochemical constituents of various extracts of *Albizia saman*

Phytochemicals	Inference					
	B	C	E	EA	M	PE
Amino acids	-	-	+	-	+	-
Anthraquinones	+	+	+	+	+	-
Alkaloids	-	-	+	-	+	-
Carbohydrates	-	+	+	+	+	-
Flavonoids	+	+	+	+	+	-
Glycosides	+	+	+	+	+	-
Saponins	-	-	-	-	-	-
Steroids	+	-	+	+	+	-
Tannins	+	+	+	+	+	-
Terpenoids	+	+	+	+	+	-
Phenols	+	+	+	+	+	-
Fixed oils and fats	-	-	-	-	-	+

+ Presence; -absence

B - Benzene; C - Chloroform; E - Ethanol; EA - Ethyl acetate; M - Methanol; PE-Petroleum ether

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