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Research Article

Preliminary Assessment of Phytochemical Constituents and Antibacterial Activity of Crude Leaves Extracts of *Simarouba glauca*

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Abstract

Simarouba glauca is well known as paradise tree in India and other parts of world. Natural products and their derivatives have historically been exploited as a valuable source of novel therapeutic agents. Medicinal plants play a significant role in the production of novel and valuable drugs used in modern medicine. In present study the primary detection of important phytochemical properties of *Simarouba glauca* was attempted. In phytochemical analysis of crude leaves extracts of *Simarouba glauca* showed presence of carbohydrates, amino acids, proteins, glycosides, tannins, terpenoids, phenols and saponins. This study was also conducted to determine the antibacterial potential of crude leaves extracts of plant against Gram positive and Gram negative clinical isolates. The acetone extract showed remarkable antibacterial potential against test isolates. It exhibited higher zones of inhibition against *E.coli* and *Salmonella paratyphi B* i.e. 13 and 12 mm respectively. The Gram negative test organisms were found more susceptible as compared to Gram positive to acetone and methanol extracts.

INTRODUCTION:

The World Health Organization (WHO) reveals that most percentage of the world's population still relies on traditional remedies including plants as their primary health care aid (HerbDay, 2007). Nature is rich source of various important medicines but due to inadequacy of information about them, some of these sources are still remained unrevealed. More than 2000 species of plants detected therapeutic significance and used as medicinal purposes (Palanisamy *et al.*, 2015). *Simarouba glauca* is an evergreen tree and has a long history of herbal medicine (Sharanya *et al.*, 2016). *Simarouba* is indigenous to the rainforest and other tropical areas in Mexico, Cuba, Haita, and Central America (Patil and Gaikwad, 2011;Lakshmi *et al.*, 2014; Osagie-Eweka*et al.*, 2016). *Simarouba glauca* is found to cultivated in several parts of India. It was brought in India in 1960s by Plant Genetic Resources in the research station at Amravati in Maharashtra and University of Agricultural Sciences, Bangalore in 1986 (Patil and Gaikwad, 2011; Lakshmi *et al.*, 2014). *Simarouba glauca* belongs to family Simaroubaceae. It produces bright green leaves 20 to 50 cm in length, small white flowers, and small red fruits (Lakshmi *et al.*, 2014).Leaves of *Simarouba glauca* are pinnately compound with 3-21 leaflets oblong and often notched or smooth at apex; alternate, even, bluish oily green (Osagie-Eweka *et al.*, 2016).

Medicinal plants synthesizes large variety of chemical substances known as secondary metabolites which include alkaloids, steroids, flavonoids, terpenoids, glycoside, saponia, tannins, phenolic compounds and so on (Ghahi A, 1990; Bargah, 2015). Therefore the basic phytochemical investigation is vital. *Simarouba glauca* was reported with pharmacological properties such as haemostatic, antihelmenthic, antiparasitic, antidysentric, antipyretic and anticancerous (Patiland Gaikwad, 2011). The present study deals with the preliminary assessment of phytochemical constituents of crude leaves extracts of plant *Simarouba glauca* and further determination of antibacterial properties of them.

MATERIALS AND METHODS

The leaves of the *Simarouba glauca* were collected from local region of Baglan of Nshik district, M.S., India and brought to laboratory. Leaves were cleaned by washing with distilled water to remove dust, and other particulate contaminants. Leaves were shade dried into laboratory at room temperature for 10 days. The dried leaves were grounded to make fine powder. The fine powder leaves was soaked in sterile water, ethanol and acetone to extract the bioactive materials.

Figure1: Leaves of *Simarouba glauca*

EXTRACTION OF PLANT MATERIAL:

1. Aqueous extract:

40 gm of fine powder of leaves was dissolved in to 200 ml of sterile distilled water in Erlenmeyer flask to make aqueous extract. The flask was placed on orbital shaker for 24hrs for extraction process. The extract was then evaporated in a rotary evaporator at 60°C (Kandil *et al.*, 1994). The color of extract was green. The final dried samples were stored in labeled sterile bottles and kept at 4°C.

2. Methanol extract:

Each dried plant sample was ground and extracted with methanol. 20 gm of powder was soaked into 200 ml of methanol. The extraction process was carried out for 24 hours at room temperature on orbital shaker. The ethanol extract was dried under a reduced pressure at 40°C. The dried extract was stored in sterile bottles until further use.

3. Acetone extract:

Powdered sample (20 g) from plant leaves were extracted with acetone by continuous agitation for 24 hrs. The solvent was removed using a rotary vacuum evaporator at 40 °C to give a concentrated extract, which was then stored at 4°C till use.

Preliminary Phytochemical Analysis of crude extracts:

The crude extracts of *Simarouba glauca* were subjected to preliminary phytochemical screening for the detection of major chemical groups. The different qualitative chemical tests were carried out on the aqueous extract using standard procedures to identify the constituents (Trease and Evans, 1989; Harborne, 1973;Veerachari and Bopaiah, 2011;Sahira and Cathrine, 2015;Sengottuvel*et al.*, 2016; Tiwari*et al.*, 2011; Njoku and Obi, 2009). The phytochemical analysis of all three crude leaves extracts of *Simarouba glauca* were conducted for detection of carbohydrates, amino acids, proteins, glycosides, tannins, terpenoids, phenols and saponins.

Determination of antibacterial activity:

Agar diffusion assay was performed by using sample impregnated /control discs of size 5 mm. Muller-Hinton Agar plates were used for antibacterial assay. The plates with basal medium were poured with seed agar containing each test organism. After solidification of seed agar, the sample impregnated discs were placed gently on surface aseptically.

The plates were incubated at 37°C for 24 hrs. After incubation diameter of zone of inhibition was measured for each crude extract.

Test organisms for antibacterial activity:

Antibacterial activity against clinical isolates, Gram positive bacteria such as *Staphylococcus aureus, Streptococcus species* and Gram negative bacteria such as *E. coli, Salmonella paratyphi B, Pseudomonas aeruginosa and Proteus mirabilis* studied by diffusion assay using Muller-Hinton media.

RESULTS AND DISCUSSION

Preliminary phytochemical analysis of crude leaves extracts:

The phytochemical analysis of plants is important to determine the presence of various secondary metabolites. Knowing of presence of phytochemical constituents of plant may be significant to prepare pharmacological formulation (Nandagopalan *et al.,* 2016). Most of phytoconstituents plays important role as antimicrobials and reported to use in medicines against various acute and chronic infections and diseases. In present study the analysis of phytochemicals in crude leaves extracts of *Simarouba glauca* showed presence of carbohydrates, amino acids, proteins, glycosides, tannins, terpenoids, phenols and saponins **(Table 1)**. These secondary metabolites of plants make them of enormous importance in medicines. Saponins of plants have often considered as antibacterial, anti-inflammatory and antitumor activities (Lakshmi *et al.*, 2014; Sparg *et al.*, 2004). Flavonoids, alkaloids and terpenoids are considered to be antimicrobial and antidiarrheal (Tiwari *et al.*, 2011). The secondary metabolites of plants have been proved their therapeutic values which plays vide range of activities in herbal remedies.

Table 1: The Phytochemical analysis of crude extracts of leaves of *Simarouba glauca*

Phytochemical	Aqueous Extract	Methanol Extract	Acetone Extract
Carbohydrates	+	+	+
Aminoacids and peptides	+	+	+
Glycosides	+	+	+
Tannins	+	+	-
Terpenoids	-	+	+
Phenols and	+	+	+
Saponins	+	-	+

(Present: +, Absent: -)

Antibacterial activity of crude leaves extracts *of Simarouba glauca*

The results obtained were compared with standard antibiotic drugs. In this study, extracts of *Simarouba glauca* were found to be active against Gram-positive, Gram-negative clinical isolates. Secondary metabolites synthesized by plant such as saponins, flavonoids and tannins are believed to exert detrimental effect on microorganisms (Koche *et al.,* 2012) Aqueous extract showed less antimicrobial activity as compared to methanol and acetone extracts (**Table 2**). The clinical isolates, *E. coli, Salmonella paratyphi B and Proteus mirabilis* were found more susceptible to both methanol and acetone extracts as compared to aqueous extract. Gram negative test organisms *E. coli*, *Salmonella paratyphi Band Proteus mirabilis* were found susceptible whereas *Pseudomonas aeruginosa* found less sensitive. From this study it was found that plant extracts exhibited remarkable bactericidal activity against both Gram positive and Gram negative test organisms. This study showed the antibacterial activity crude leaves extracts of *Simarouba glauca* which suggests that plant leaves extracts contains various antibacterial components and further analysis of bioactive compounds of *Simarouba glauca* may dictate its potential in herbal medicines.

Table 2: The antibacterial effect of aqueous, methanol and acetone extracts of *Simarouba glauca* against test organisms

Test Organism	Extract	Diameter of Zone of Inhibition (mm)		
		Extract	Penicillin (50µg/ml)	Streptomycin(50µg/ml)
Staphylococcus aureus	Aqueous	5	16	-
	Methanol	8		-
	Acetone	10		-
Streptococcus species	Aqueous	4	14	-
	Methanol	8		-
	Acetone	8		-
E. coli	Aqueous	4	-	18
	Methanol	12	-	
	Acetone	13	-	
Salmonella paratyphi B	Aqueous	3	-	14
	Methanol	12	-	
	Acetone	14	-	
Pseudomonas aeruginosa	Aqueous	3	-	16
	Methanol	7	-	
	Acetone	9	-	
Proteus mirabilis	Aqueous	2.5	-	19
	Methanol	10	-	
	Acetone	11	-	

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