

Research Article

Preliminary Phytochemical Investigation and Antimicrobial Activity of *Sinapis Alba*

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Abstract: The plant leaves contain a number of medicinally important compounds. The present study was carried out to identify the phytochemicals and evaluate antimicrobial activity of hexane extract of *Sinapis alba* seeds. The antimicrobial activity was determined by well diffusion method and tested against different pathogens of three bacteria and two fungus. Antimicrobial activity of 50, 100 and 150% Hexane extract of *Sinapis alba* (white mustard), has been evaluated against streptococcus pneumoniae, Aspergillus niger, Salmonella typhi, klebsiella pneumoniae, Trichophyton Violaceum. The seeds of hexane extract contain terpenoids and fattyacids. In 150% concentration *Sinapis alba* showed the highest 20mm antibacterial zone against streptococcus pneumoniae. In 100% and 150% concentration *Sinapis alba* showed the highest 15mm, 20mm, antimicrobial zone respectively against Aspergillus niger. The extract showed inhibitory effects due to the presence of terpenoids and terpenoids.

Keywords: Aspergillus niger, fatty acids, terpenoids, streptococcus pneumonia, salmonella typhi.

INTRODUCTION

Most traditional medicines are developed from nature. They have not yet fulfilled the scientific requirements, so as to be classified as modern medicines [1]. For purposes of scientific backup, a study is needed to examine their bioactive components, their efficacy and safety [2]. Usually, most components that are useful for medicinal purposes are secondary metabolites. White mustard is native to the Mediterranean region. It grows on fields and waste areas, on calcareous soils. [3] White mustard is an annual plant, with an erect stem and numerous branches spring from the main stem. Leaves are alternately arranged, etiolate and serrated, with short, white bristles along the veins. Flowers are pale yellow, forming a shape of a cross. Flowering occurs from June to August. Seeds are globular, dark reddish-brown [4-7].

White mustard seeds are considered to be analgesic, carminative, expectorant and stimulant. Traditionally, they have been applied in cases of digestive complaints. An infusion of the seeds is useful in treatment of chronic bronchitis, rheumatism, muscular and skeletal pains (it stimulates circulation in the pain area) [8]. Externally it is usually made into mustard plasters, poultices or added to the bath water. It can also be used for skin eruptions. Mustard tea is used for fevers and colds. The seed contains substances that can irritate the skin and mucous membranes. It is advisable to consult your doctor or herbalist before consumption of white mustard [9].

Moreover, mustard oil works as an appetizer as it stimulates digestive juices. Besides, regular consumption of mustard seeds reduces the frequency of migraines. Mustard benefits in the treatment of cough, cold, flu and so on. In terms of nutritional benefits of mustard, it is rich in vitamins, minerals, antioxidants, and phytonutrients. This spice contains magnesium, selenium, iron, calcium, manganese, potassium, phosphorus, vitamin B complex etc [10]. The aim of the present study was to determine the possible phytochemical and antimicrobial activity of hexane extract of *Sinapis alba* seeds.

MATERIALS AND METHODS

Plant Material

Plants were collected from Thanjavur District of Tamilnadu. The botanical identity of the plant was confirmed by Dr. John Brito, Rapinet Herbarium, St. Joseph's College, Thiruchirappalli.

Preparation of Extract

The powder (1kg) was extracted with Hexane at room temperature for 48h. The extracts were filtered and concentrated under reduced pressure in a rotary evaporator. The extracts were subjected to screening of phytochemical and antimicrobial activity.

Phyto Chemical Analysis

Phytochemical analysis involves the qualitative analysis of herbal plants. The preliminary qualitative tests have been attempted in *Sinapis alba* seeds to find out the presence or absence of certain bio active Compounds. Chemical tests were carried out on the

hexane extract using standard procedures to identify the constituents as described by Harbone[11].

Test for Tannins

About 0.5g of the dried powdered samples was boiled in 20ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride were added. Absence of brownish color indicating the absence of tannins.

Test for Phlobatannins

Extract boiled with 1% aqueous hydrochloric acid. No red precipitate is formed. absence of phlobatannins.

Test for Saponin

About 2g of the powdered sample was boiled in 20ml of distilled water in a water bath and filtered. 10ml of the filtrate was mixed with 5ml of distilled water and shaken vigorously for a stable persistent froth. No change in solution, absence of Saponin.

Test for Flavonoids

Few drops of 1% aluminum solution were added to a portion of extract. No change in color. Absence of flavonoids.

Test for Oil and Fat

Stain test:

Small Quantity of extract was pressed between two filter paper. An oily stain on filter paper indicates the presence of oil.

Test for Steroids

2ml of acetic anhydride was added to 0.5g ethanolic extract with 2ml sulphuric acid. No change in color, absence of steroids.

Test for Terpenoids (Salkowski Test)

5ml of extract was mixed in 2ml of chloroform, and 3ml of conc. Sulphuric acid was carefully added to form a layer. A reddish brown colouration of the interface was formed to show positive results for the presence of terpenoids.

Test for Alkaloid

Mayer Test

0.5 ml of extract is added with 1 ml of mayer reagent. Yellow colour is absent. Absence of alkaloid.

Dragandroff's test

0.5ml of extract is added with 1ml of dragandroff's reagent orange red colour is absent. Absence of Alkaloid.

Wagner Test

0.5ml Sample is treated with 1 ml of wagner reagent. No change in colour. Absence of alkaloids.

Test for Phenol

0.5ml of extract is treated with phenol reagent. No change in colour. Absence of phenol.

Micro Organism

Trichophyton violaceum, Aspergillus niger, Salmonella Typhi, Streptococcus pneumoniae, Klebsiella were the pathogenic micro organisms included in the study. All the cultures were obtained in pure form from the culture collection of Institute of Microbial Technology (IMTECH), Chandigarh, India.

Media Preparation

Bacteria Media

36gm of Muller Hindon Media (Hi-Media) was mixed with distilled water and then sterilized in autoclave at 15lb pressure for 15 minutes. The sterilized media's were poured into petridishes. The solidified plates were bored with 5mm dia cork borer. The plates with wells were used for the antibacterial studies.

Fungal Media

200mg of potato slices were boiled with distilled water. The potato infusion was used as water source of media preparation. 20gm of dextrose mixed with potato infusion. 20gm of agar was added as solidifying agent. These constituents were mixed and autoclaved. The solidified plates were bored with 6mm dia cork borer.

Well Diffusion Method

Antibacterial and anti-fungal activity of the plant extract was tested using well diffusion method [12]. The prepared culture plates were inoculated with different selected strains of bacteria and fungi using streak plate method. Wells were made on the agar surface with 6mm cork borer. The extracts were poured into the well using sterile syringe. The plates were incubated at 37°C±2°C for 24 hours for bacterial and 25°C ± 2°C for 48 hours for fungal activity. The plates were observed for the zone formation around the wells was measured in mm (millimeter). For each treatment three replicates were maintained.

The diameter of inhibition zones was measured in mm and the results were recorded. Inhibition zones with diameter less than 12mm were considered as having no antimicrobial activity. Diameters between 12 and 16mm were considered moderately active and these with 16 mm were considered highly active.

RESULTS AND DISCUSSION

The result of the phytochemical screening and antimicrobial activity against the tested pathogens are given in the Table 1, 2, 3. The inhibition effects of all the pathogens are presented in fig: 1, 2. Terpenoids, fatty acids were present in *Sinapis alba* and alkaloids, Tannins, Steroids, flavonoids, saponins were absent in *Sinapis alba*.

Table 1: Qualitative Analysis of phytochemicals of *Sinapis alba*

S.NO	PHYTOCHEMICAL	RESULT
1	Alkaloids	Absent
2	Phenol	Absent
3	Tannin	Absent
4	Terpenoid	Present
5	Saponin	Absent
6	Flavonoids	Absent
7	Steroids	Absent
8	Fatty acid	present

Table 2: Antibacterial activity of different concentrations of hexane extract of *Sinapis alba*.

Name of the pathogens	Inhibition in mm				
	Control	Antibiotic	50%	100%	150%
Salmonella Typhi	0	20	Nil	Nil	Nil
Streptococcus Pneumoniae	0	22	10	11	20
Klebsiella Pneumoniae	0	44	Nil	Nil	Nil

Table 3: Anti fungal activity of hexane extract of *Sinapis alba* seeds

Name of the species	Inhibition in mm				
	Control	Antibiotic	50%	100%	150%
Trichophyton Violaceum	0	31	Nil	Nil	Nil
Aspergillus Niger	0	32	13	15	20

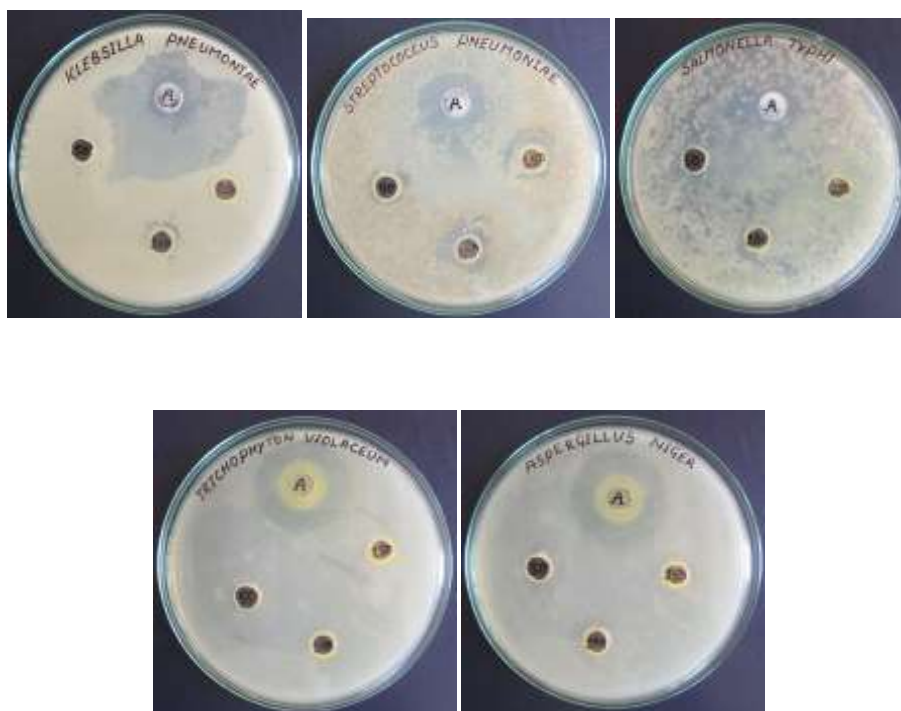


Figure 2: Inhibition effect of selected species

As a result, the comparing of hexane extract of *Sinapis alba* against bacteria and Fungai, In 150% concentration, antimicrobial activity of hexane extract of *Sinapis alba* showed the highest 20mm zone against streptococcus pneumoniae. In 100% and 150% concentration, *Sinapis alba* showed the highest 15mm, 20mm antimicrobial zone respectively against *Aspergillus niger*. In 50%, 100% and 150% concentration, antimicrobial activity of hexane extract of *Sinapis alba* in *Salmonella yyphi*, *klebsiella pneumoniae*, *trichophyton violaceum* not showed.

Ethnobotanical approach is one of the common methods that are employed in choosing the plants for pharmacological study. India is one of the twelve mega biodiversity centers having more than 45,000 plant species. Its diversity is unmatched due to the presence of sixteen different agroclimatic zones, 10 vegetative zone and 10 vegetative zone and 15 biotic provinces [13].

Use of plants as a source of medicine has been inherited and is an important component of the health care system. Approximately 20% of the plants found in the world have been submitted to pharmacological or biological tests [14]. The systematic screening of plant extracts for antibacterial activity is a continuous effort to find new antibacterial compounds. Considering the rich diversity of plants in Karnataka, it is necessary to screen plants for their antibacterial activity.

Conclusion

The present investigation shows the *Sinapis alba* plant seeds had high antimicrobial activity of comparing other medicinal plants. So, it was consider the *Sinapis alba* seeds had medicinal values and some biological activities of hexane extract. The different concentration of hexane extract *Sinapis alba* has been demonstrated for the first time further investigation is in progress to isolate and characterized the active principle.

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