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Investigation on Anti-Microbial Activity of *Pterospermum reticulatum* White and Arn

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ABSTRACT

The present study was aimed to investigate the activities of different solvent extracts of *Pterospermum reticulatum* by performing its antimicrobial studies and comparing it with standard drugs. Successive soxhlation was performed on powdered leaf and stem parts of *Pterospermum reticulatum* using solvents in increasing order of polarity. Percentage yield and extract weight are calculated from which ethanol extract exhibited higher yield. The results of antimicrobial activity of petroleum ether, chloroform, ethanol and water extract of *Pterospermum reticulatum* against *Proteus vulgaris* was carried out. It was found that almost all extracts showed similar pattern with trace inhibition according to concentration administered. Antimicrobial potential of the petroleum ether, chloroform, ethanol and water on leaf and stem extracts for the said concentration could not be established to a satisfactory level.

INTRODUCTION

Natural products are the active components not only of most traditional medicines but also many modern medicines. They are often used as starting point of drug discovery. They are often divided into two classes: primary and secondary metabolites. As the secondary metabolites have the ability to modulate biochemical and signal transduction pathways, they are used as medicines.

A natural product is a chemical compound or substance produced by a living organism. They will include a large and diverse group of substances from a variety of sources. They may include plant, animal, mineral or marine sources. The isolated compound from the extracted source can choose for the evaluation and prove their medicinal activity [1,2].

Pterospermum species is an herb distributed throughout the world. The leaves of the plant are widely used for diabetes and as a hemostatic in the medicines. The plant is used as antioxidant, antiulcer, anti-inflammatory, analgesic, hypoglycemic and anthelmintic.

Pterospermum reticulatum Wight and Arn is considered as rare and threatened species belonging to the family Sterculiaceae. Pterospermum in greek word referred to winged seed. According to IUCN Red Data Book and commonly called Malavuram in Malayalam and Mulipolavu in Tamil. It is a tree growing up to 25 meters tall, bark is grayish brown, leaves are simple, alternate, oblong, obovate, white tomentose and stellate hair beneath.

Pterospermum reticulatum is a medicinally important plant. The stem bark of the plant was used by tribes to treat ulcers, wounds and inflammations. The crude extracts obtained from the leaf and stem of *Pterospermum reticulatum* used to treat bacterial and fungal diseases by local tribes. It is also used in diarrhea and dysentery [3].

Antimicrobial study conducted on *Pterospermum reticulatum* was shown to have valuable activities against the following strains:

1) Ethanol and Water extract:

- Bacterial strain: *S. aureus, B. subtilis, & P. aureginosa*
- Fungal strain: *Aspergillus niger, Candida albicans & Trichoderma viridae*

2) Water extract:

- *Candida albicans*:

The population from natural habitat is rapidly declining due to various factors such as habitat destruction and reproductive constrains. This study is to investigate unknown activities of *Pterospermum reticulatum* and to apply it into modern medicine and to conserve this plant species from the threat of extinction [4].

TAXONOMY [5]

Kingdom	:	Plantae
Phylum	:	Tracheophyta
Class	:	Magnoliopsida
Order	:	Malvales
Family	:	Sterculiaceae
Genus	:	Pterospermum
Species	:	*Pterospermum reticulatum*

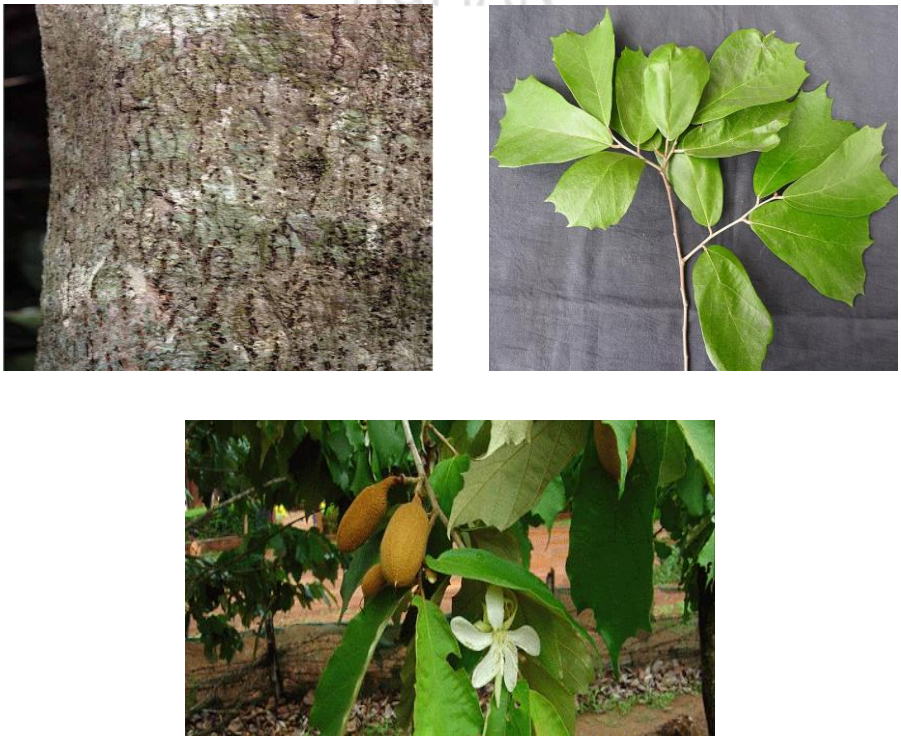

Figure No. 1: *Pterospermum reticulatum*

OBJECTIVE

A large number of phytochemicals belonging to several chemical groups having inhibitory effects on several types of pathogenic microbes are drawing attention of scientists to search for potential drug. Thus the potential of the plants and their ability to compact infectious disease justifies the continued identification and characterization of medicinal plant to quantify their antimicrobial potentials.

The main aim was to find out the activities of different extracts of *Pterospermum reticulatum*, by performing its antimicrobial studies and comparing it with standard drugs.

METHODOLOGY[6,7]

- **Collection**

Pterospermum reticulatum was collected from Kerala Forest Research Institute (KFRI) Peechi and Nilambur teak museum Malapuram. Soxhlet apparatus and all other chemicals and reagents purified by standard techniques were used.

- **Authentication:**

The collected plant were identified and authenticated by Dr. Vivek P J, Asst. Professor, Department of Botany, Sree Neelakanda Government Sanskrit College, Pattambi.

- **Drying:**

The leafs from whole plant were separated, washed in running water to remove dirt, dried under shade at low temperature in oven at a temperature of 30-40°C. The leaves of individual plants were suitably size reduced and then stored separately in labeled airtight containers.

- **Size reduction:**

The collected leaves was subjected to size reduction manually.

- **Extraction:**

Extraction of dried leaves of *Pterospermum reticulatum* were carried out using successive extraction.

- **Preparation of extract:**

About 16 g of dried leaf and bark powder were taken and packed into the column of Soxhlet apparatus, extracted exhaustively with 250 ml of various solvents, Extraction was carried till the colour of siphon changes. The extracts were filtered, cooled and concentrated to a syrupy consistency. The yield of each extract was noted.

ANTIMICROBIAL ACTIVITY [8]

Antimicrobial testing can be used for drug discovery and for the prediction of therapeutic outcome. A variety of laboratory methods are available for the screening or evaluation of antimicrobial activity of plant extract. The most known techniques are disk diffusion and broth or agar dilution method.

1) Preparation of nutrient agar:

- Measure 1.62 g of agar and 60 ml of distilled water in a conical flask.
- Stir well with a sterilized glass rod.
- Cover the mouth of conical flask with aluminum foil.
- Autoclaved for 30 minutes.
- Allow to cool and pour the mixture in to a sterilized Petri dish.
- Allow the mixture for settle down.

2) Addition of bacterial strain

Proteus vulgaris is selected as a bacterial strain for the evaluation of antimicrobial activity. *Proteus vulgaris* is a rod shaped gram negative bacterium.

Addition of strain to the nutrient agar medium is done by cotton swab method. After addition of bacterial strain, the medium is allow to settle properly.

3) Preparation of standard solution:

Accurately weighed 1g of ciprofloxacin and dissolved in DMSO. It was made up to 100 ml in standard flask to make a concentration of solution.

4) Selection of blank solution

DMSO is considered blank solution due to their easy solubility.

5) Well, diffusion technique:

Agar well diffusion is widely used to evaluate the antimicrobial activity of plants or microbial extracts. The wells are punched aseptically with the help of sterile cork borer or a tip with 6 to 8mm.

6) Addition of Standard, Test and Blank:

Mark the well respectively as standard, test and blank. The samples are added into the well with the help of micropipette having a capacity of 25µl.

7) Incubation

The Petri dish are kept in the BOD incubator for 24hrs at a temperature 30-40°C.

Table No. 1: Calculation of percentage yield of leaf extract

Raw material	Extract	Weight of raw material before extraction (g)	Weight of raw material after extraction (g)	Weight of extract (g)	Percentage yield (%)
Pterospermum reticulatum	Petroleum ether	16.077	15.8	0.3	1.851
	Chloroform	16.2	16.0	0.2	1.226
	ethanol	16.3	15.0	0.8	5.333
	water	15.0	13.0	0.7	5.001

Table No. 2: Calculation of percentage yield of bark extract

Raw material	Extract	Weight of raw material before extraction (g)	Weight of raw material after extraction (g)	Weight of extract (g)	Percentage yield (%)
Pterospermum reticulatum	Ethanol	21.0	19.915	1.532	7.692

Table No. 3: Zone of inhibition of leaf extract

Microorganism	Zone of inhibition(mm)									
	Chloroform extract (µg/ml)			Petroleum ether (µg/ml)			Ethanol extract (µg/ml)	water extract (µg/ml)	Standard-Ciprofloxacin (µg/ml)	Blank-DMSO
	25	50	100	25	50	100	100	100	10	100
Proteus vulgaris	6	9	18	5	8	21	18	11	32	4

Table No. 4: Zone of inhibition of bark extract

Microorganism	Zone of inhibition(mm)		
	Ethanol extract (100mg/ml)	Standard-Ciprofloxacin(10µg/ml)	Blank-DMSO
Proteus vulgaris	9	35	3

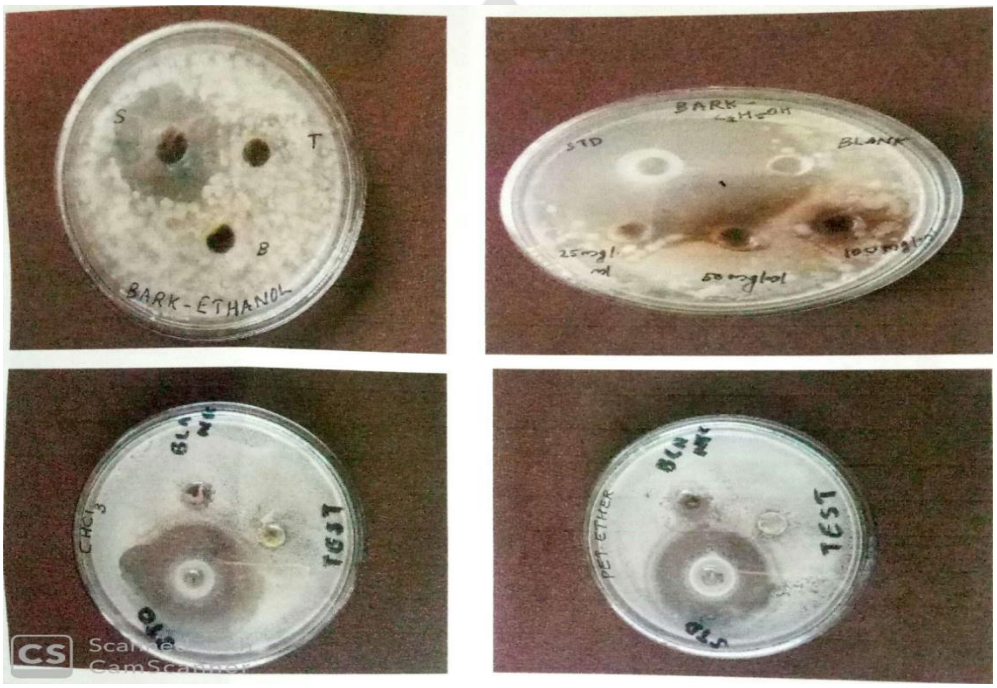

Figure No. 2: Zone of inhibition in bark

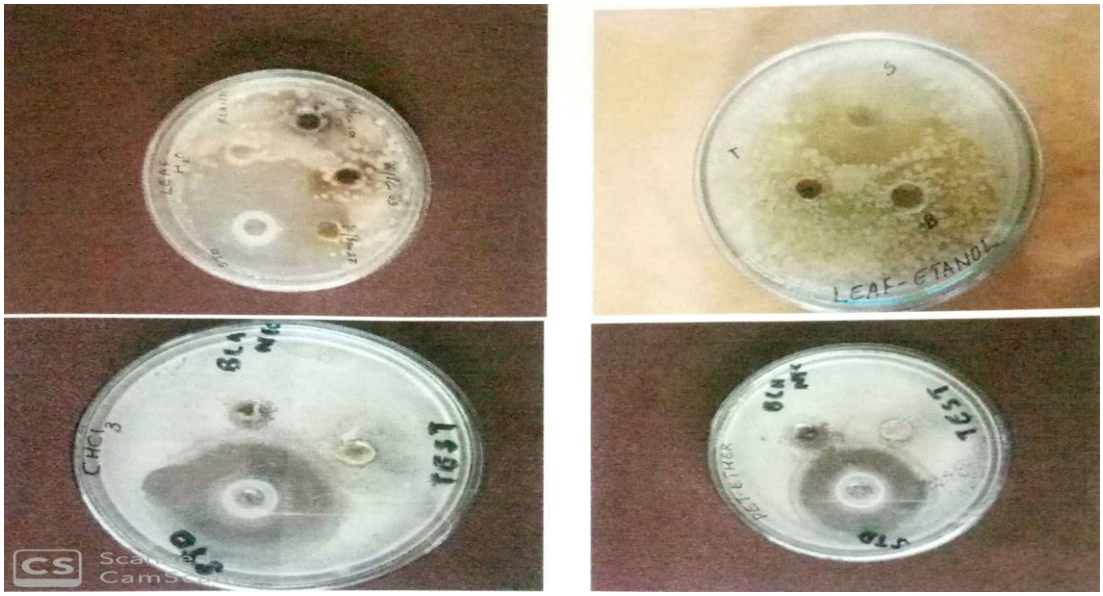

Figure No. 3: Zone of inhibition in leaf

RESULTS

Extract weight and yield:

Successive soxhlation was performed on powdered leaf and stem parts of *Pterospermum reticulatum* using solvents in increasing order of polarity. Percentage yield and extract weight are given in Table: 1. Ethanol extract exhibited higher yield.

Antimicrobial activity:

The results of antimicrobial activity of petroleum ether, chloroform, ethanol and water extract of *Pterospermum reticulatum* against Proteus vulgaris was tabulated. It was found that almost all extracts showed similar pattern with trace inhibition according to concentration administered.

Antimicrobial potential of the petroleum ether, chloroform, ethanol and water on leaf and stem extracts for the said concentration could not be established to a satisfactory level.

DISCUSSION

Secondary metabolites in plant products are responsible for several biological activity in man and animals. Antimicrobial studies are one of the initial steps towards the study of such

activity. The extracts of plants can be a very good source of antibiotics against various bacterial pathogens.

The present study carried out on the plant revealed percentage yield of leaf using various solvents of increasing polarity and for bark extract using ethanol. Ethanol and water gave good yield with highest yield reported in ethanol extract and least in chloroform extract of leaf while ethanol extract of bark alone showed on overall high value. However, these high yield extracts failed to produce significant anti-bacterial effect in the prepared concentrations.

The crude alcohol and water extracts could be further studied for their antimicrobial potential by increasing their concentration to a high value as the extracts exhibited satisfactory yield.

CONCLUSION

Pterospermum reticulatum is a species of flowering plant in the family Sterculiaceae. It is found only in India. It is threatened by habitat loss. The current study is conducted for reveal the importance of *Pterospermum reticulatum*. The other subspecies from Pterospermum showing a number of medicinal activities.

The study clearly demonstrated that the plant extracts exhibited potent antimicrobial activity. Ethanol and water extract of bar and leaf gave a good activity.And the highest yield is reported in ethanol extract and least in chloroform extract of leaf while ethanol extract of bark alone showed on overall high value.

From the current study revealed that the *Pterospermum reticulatum* shows antimicrobial potential only in high concentration to exhibit a satisfactory action.

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