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Indigenous Knowledge of Ethnic Community on Usage of Kripa (Lumnitzera racemosa) and its preliminary screening

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Abstract

Kripa (*Lumnitzera racemosa*) is an evergreen branched tree of medicinal value found in the mangrove areas of the Indian subcontinent and traditionally used by local rural communities to treat various ailments and their symptoms. Kripa was identified as one of the many mangrove species that occur in the Sunderbans delta that are being used for its therapeutic properties. The traditional usage of Kripa leaves and bark were learnt through interaction with the locals. The plant parts were collected from the Medicinal Plant Conservation Area (MPCA) in Bonnie Camp and a preliminary phytochemical analysis was conducted in methanolic extraction by following standard methodology. The locals reported that the most common use of the plant were to treat itches, bites (inflammation) and occasionally even symptoms of diabetes. The preliminary phytochemical screening reveals that the leaves of Kripa contains glycosides, alkaloids, phenols, tannins, flavonoids, etc.

Keywords: Ethnic community, Kripa, medicinal value, phytochemical.

Introduction

Due to the important role and possibilities for treatment of human diseases, research on medicinal plants has been the focus of grate fascination during the last few decades (Pan et al., 2010 and Boots et al., 2008). Medicinal plants or herbs are the essence of India's traditional medicine practice (Adhikari et al., 2018). Using plants to create formulations to treat various ailments and their symptoms play a vital part in the history of traditional methods of Indian medicine like Ayurveda. Ethnomedicine is a study of the traditional medicine practiced by ethnic communities, specially by indigenous people. The combination of ethnomedicine and ethnobotany is currently of great interest, due to their important role in finding out or succeeding new medicine.

Sunderban is one the most diverse areas of the nation in terms of flora and fauna and the entire vegetation is mostly of mangrove type. Since, mangrove species of plants are indigenous and unique to this region (Pattanaik et al., 2008). Mangrove plants having medicinal values are traditionally used

by several ethnic groups for the treatment of various ailments (Revathi et al., 2013). Medicinal plants are not only a major resource base for the traditional medicine and herbal industry but also provide livelihood and health security to a large segment of Indian population (Anon, 2010).

The major objectives of the study includes

• To identify the community of people in and around Bonnie Camp area those who still uses and practices medicinal plants for the treatment of various diseases.

• To explore the ethnobotanical usage of *Lumnitzera racemosa* from indigenous knowledge of local tribal people.

• To screen the phytochemicals present in the roots of *Lumnitzera racemosa* collected from Bonni Camp MPCA.

Materials and Methods

A literature review has been done before visiting the field. The literature review primarily focused to identify the community of people in areas surrounding Bonnie Camp area that are populated, who still inherit traditional knowledge regarding hodopathy and practice regularly. During field work, interviews were conducted with local knowledgeable villagers, the herbal healers called 'Vaidyas or Ojhas' (local physicians in Indian System of Medicine), old woman and medicinal plant vendor (Shaw et al., 2015).

Study area

The study area selected was the Bonnie Camp area of the Sundarbans. Sundarban National Park is located between 21°432' – 21°55' N latitude and between 88°42' – 89°04' E longitude. The average altitude of the park is 7.5m above sea level. 54 small islands compose the park and several distributaries of the Ganges River intersect it. Sundarban was named after the Sundari Tree. Like this tree, many other trees indigenous to such mangrove environments have specialized roots called pneumatophores which emerge above ground and help in gaseous exchange i.e. respiration. During the rainy season when the entire forest is waterlogged, the spikes rising from the ground has their peak in the air and aid in the process of respiration of the trees.

The area was already opted for establishment of Medicinal Plants Conservation Area (MPCA) by the West Bengal State Forest Department under the 'National Programme on promoting conservation of Medicinal Plants and traditional knowledge for enhancing the Health and Livelihood Security'. MPCA have been suggested to be managed as 'hands off' area with certain interventions to encourage identification, preservation, natural regeneration, monitoring etc. by involving Forest Protection Committee (FPC) members through awareness and confidence building exercises. National programmes on promoting conservation of medicinal plants and ancient traditional practices to improve health and lifestyle are gaining more and more importance with time.

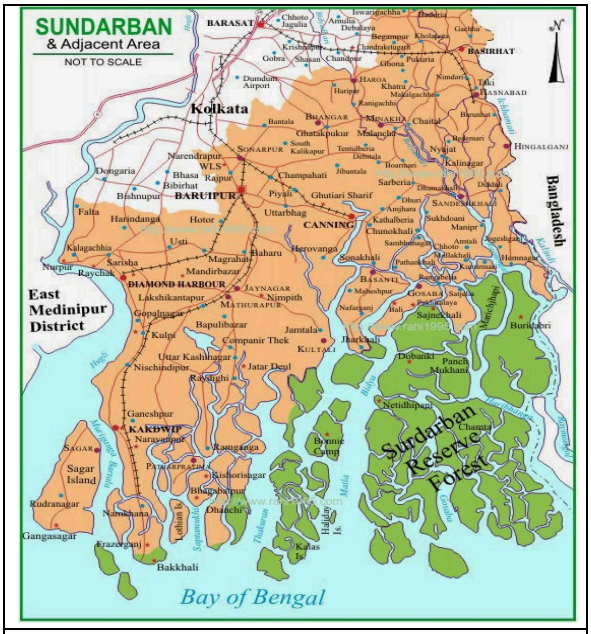

Fig 1. Map of Sundarban National Park.

There are few parameters that are kept in mind while choosing an MPCA. They are -

1. The area should have a varied diversity of vegetation including species with proven medicinal value.
2. Area should be relatively undisturbed and should have easy accessibility.
3. The vegetation cover of the area should represent the general type of flora of the region.
4. The area should be traditionally known for its medicinal plants.
5. The area should be under the legal protection of the government, as in, it should be part of a protected area such as a Biosphere Reserve or the like.

Bonnie Camp is the major MPCA of the South 24 Parganas district of West Bengal, as a part of the "Silviculture South Division". The initial stage in studying medicinal plants is the preparation of plant samples to preserve the biomolecules in the plants prior to extraction. Leaves of each plant were dried in shade for approximately a couple of weeks. Direct heat or even sunlight was avoided so as to prevent possible decomposition of the active compounds of the chemical composition. Once sufficiently dry, the leaves were cut into smaller bits for ease of processing. A clean, dry and sterilized mixer-grinder was used to blitz the leaves into a rough powder form. The resulting product was physically ground to make it finer for better potential extraction. Lowering particle size increases surface contact between samples and extraction solvents. Grinding resulted in coarse smaller samples; meanwhile, powdered samples have a more homogenized and smaller particle, leading to better surface contact with extraction solvents (Azwanida, 2015). After reviewing some research articles, methanol was chosen as the preferred reagent/solvent for extraction. Methanol is a good solvent for extraction and it is frequently used in biology because of its polarity. It is capable of extracting both lipophilic and hydrophilic molecules or substances. The other advantage is that it be removed easily at room temperature because it is highly volatile.

The study of medicinal plants starts with the pre-extraction and the extraction procedures, which is an important step in the processing of the bioactive constituents from plant materials. Extraction is the separation of medicinally active portions of plant using selective solvents through standard procedures. The purpose of all extraction is to separate the soluble plant metabolites, leaving behind the insoluble cellular marc (residue). The initial crude extracts using these methods contain complex mixture of many plant metabolites, such as alkaloids, glycosides, phenolics, terpenoids and flavonoids.

Phytochemical Analysis

This study is carried out for the development of phytochemical parameters and to observe the active chemical components that have medicinal properties, present in methanolic extract obtained from the collected specimens (processed leaves).

Test for Triterpenoids

Libermann-Buchard test

10mg of test sample was dissolved in 1.0 ml of chloroform; 1.0 ml of acetic anhydride was then added to the mixture followed by 2.0 ml of concentrated sulphuric acid. Appearance of reddish violet ring at the junction of the two layers confirms the presence of triterpenoids and steroids.

Salkowski Test

1.0 ml of concentrated sulphuric acid was added to 10 mg of test sample dissolved in 1.0 ml of chloroform. Appearance of reddish blue colour exhibited by chloroform layer and green fluorescence by the acid layer suggests the presence of steroids.

Tests for Alakaloids

Mayer test

A small quantity of test sample was treated with few drops of dilute hydrochloric acid and filtered. The filtrate was treated with Mayer's reagent. Appearance of yellowish buff colored precipitate indicates the presence of alkaloids.

Dragendorff test

A small quantity of test sample was treated with few drop of dilute hydrochloric acid and filtered. The filtrate was then treated with Dragendorff's reagent. Appearance of orange brown precipitate indicates the presence of alkaloids.

Wagner test

A small quantity of test sample was treated with few drop of dilute hydrochloric acid and filtered. The filtrate was treated with Wagner's reagent. Failure to produce reddish brown precipitate suggested the absence of alkaloids.

Hager test

A small quantity of test sample was treated with few drop of dilute hydrochloric acid and filtered. The filtrate was treated with Hager's reagent. Appearance of yellowish precipitate demonstrates the positive test for alkaloids.

Test for Tannins

A small quantity of test sample was dissolved in minimum amount of distilled water and filtered. The filtrate was treated with 10% aqueous potassium dichromate solution. Development of yellowish brown precipitate demonstrates the presence of tannins. A small quantity of test sample was dissolved in minimum amount of distilled water and filtered. The filtrate was allowed to react with 10% lead acetate solution. Formation of yellow colour precipitate indicates the positive test for tannins. A small quantity of test sample was dissolved in minimum amount of distilled water and filtered. The filtrates were then allowed to react with 1.0 ml of 5% ferric chloride solution. Formation of greenish black coloration demonstrates the presence of tannins. A small quantity of test sample was dissolved in minimum amount of distilled water and filtered. The filtrates are then allowed to react with 1.0 ml of 1% gelatin and 1.0 ml of 10% sodium chloride solution. Formation of white buff colored precipitate demonstrates the presence of tannins.

Test for Saponins

A small quantity of test sample was dissolved in minimum amount of distilled water and shaken in a graduated cylinder for 15 minutes. Formation of stable foam suggests the presence of saponins. A small quantity of test sample was dissolved in methanol. 1.0 ml of extract solution was treated with 1% lead acetate solution. Formation of white precipitate indicates the presence of saponins.

Test for Carbohydrate

Molisch test

The test solution is combined with a small amount of Molisch's reagent (α-naphthol dissolved in ethanol) in a test tube. After mixing, a small amount of concentrated sulfuric acid is slowly added down the sides of the sloping test-tube, without mixing, to form a layer. A positive reaction is indicated by appearance of a purple ring at the interface between the acid and test layers.

Benedict test

A small quantity of test sample was treated in minimum amount of distilled water and filtered. To the filtrate equal volume of Benedict's reagent was added and heated for few minutes. Formation of brick red precipitate confirms the presence of reducing sugars.

Fehling test

A small quantity of test sample was treated in minimum amount of distilled water and filtered. To the filtrate equal volume of Fehling's A & B solution were added and heated for few minutes. Development of brick red colour demonstrates the presence of reducing sugars.

Fig 2. Whole Kripa plant.

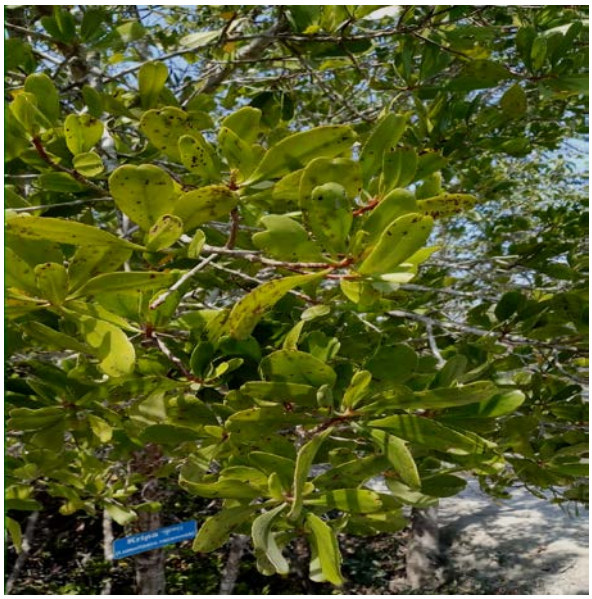

Fig 3. Kripa leaves.

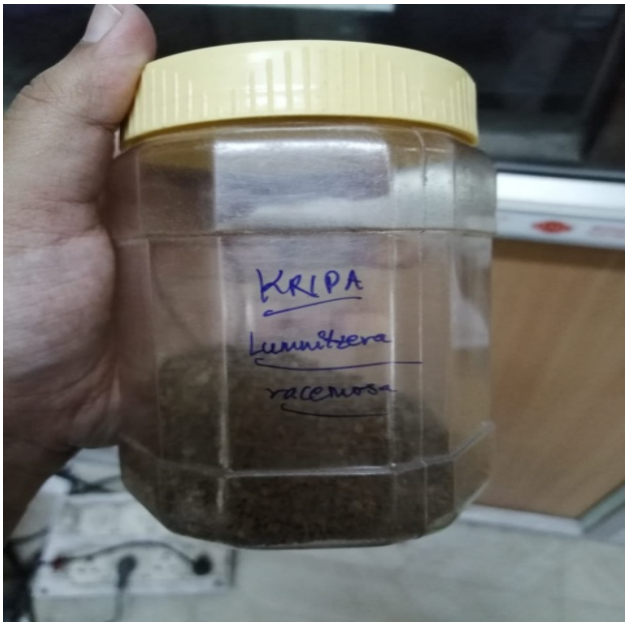

Fig 4. Dried and crushed Kripa leaves.

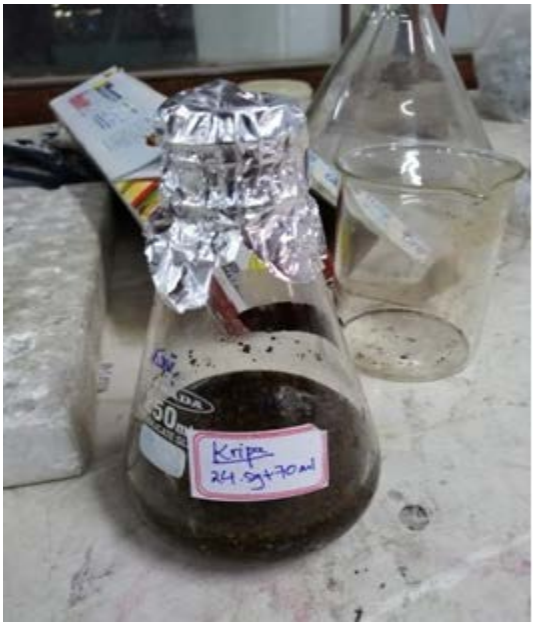

Fig 5. Making of methanolic extract from Kripa leaves.

Test for Flavonoids

Shinoda test

To dry powder or extract, add 5 ml of 95% ethanol, few drops of conc. HCl and 0.5 g magnesium turnings. Pink colour is observed. To small quantity of residue, add lead acetate solution. Yellow coloured precipitate is formed. Addition of increasing amount of sodium hydroxide to the residue show yellow coloration, which decolouration after addition of acid.

Results and Discussions

The ethnic community living in the Bonnie Camp area of the Sundarbans includes mainly Oraon, Ho, Munda and Santhal tribes who still dependent of

the local medicinal plants available in the surrounding forest. Focus group discussion reveals that *Lumnitzera racemosa* is the most frequently used medicinal plants by the local community and also traded commercially. It was identified as flagship species for conservation of medicinal plants in the newly created Medicinal Plant Conservation Area (MPCA).

Morphological study has shown that the plant is an evergreen shrub or small tree growing up to 8 metres tall with white flower, flat and spoon shaped lives with emarginated tips are mainly used in indigenous medicine, though the other parts of the plants are also have importance from different point of views.

Table 1. Preliminary Phytochemical Screening.

Compound/ Group	Test	Result
Tannins	Ferric chloride	+
	Lead acetate	+
Alkaloids	Wagner	+
	Dragendorff	+
	Hager	+
Phenols	Neutral $FeCl_3$	+
	$FeSO_4$ and Sodium Potassium Tartarate	+
Glycosides	Modified Borntragor	+
Carbohydrates	Molisch	+
	Fehling	+
Flavonoids	Shinoda	+
Steroids	Salkowski	_
Saponins	Lead acetate	_
Note: Presence (+) and absence (-).		

Local people are using this plant for the preparation of different herbal medicines, either singly or in combinations like with water, honey, or other plant species etc. The local tribes also reported that the most common use of leaves for the treatment of various ailments like antihypertensivity, antiplasmodial, antiangiogenic, anti-inflammation. The preliminary phytochemical screening reveals that the leaves of Kripa contains-tannins, alkaloids, phenols, glycosides, carbohydrates and flavonoids as responded in methanolic extract. However, no metabolites are found in the methanolic extract. The response details of preliminary phytochemical screening are represented in the table 1.

Lumnitzera racemosa is being used for the treatment of various ailments like itches, herpes, and even diabetes in some cases. It is also used in animal bites. From the preliminary tests results it may be assume that the phytochemical present in the eaves of Kripa supports the above uses, however further confirmation is need to be established through further detailed investigation

Conclusion

It is extensively used by the rural population for the purposes described above. Preliminary screening confirm the presence of many metabolites as shown in the table. More detailed tests and investigations are required for the plant, including actual quantitative analysis of the metabolites. This is to see how much of each of these active compounds are really present in the plant, so as to use that information to develop medicines for proper use. Since this plant is proven to be very important and in demand for its high medicinal properties by the local community, scientific harvesting and sustainable conservation needs to be promoted.

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