

Chapter 3

Qualitative phytochemical analysis of *Phyllanthus virgatus* G. Forst.: a riparian herb

Nibedita Jena¹, Sanjeet Kumar¹ and Bhagwati Prashad Sharma^{2*}

¹Ambika Prasad Research Foundation, Odisha, India

²Sidharth Government College, Nadaun, Himachal Pradesh, India

*Correspondence author's email-Id: bp76sharma@gmail.com

Abstract

The whole plant of *Phyllanthus virgatus* was selected for qualitative phytochemical analysis as per its traditional uses. Whole plant paste is used to cure skin itching. The Soxhlet method was adopted for extraction using three solvents (aqueous, ethanol, and methanol). Results revealed that whole plant extracts showed the presence of tannin, which might be responsible for treating skin itches. Further, more experimental works are needed to validate ethnomedicinal claims on *P. virgatus* and the development of therapeutic drugs against skin itches.

Keywords: Herb, river areas, skin infections

Introduction

Rivers give life in the earth. They are also the place where early civilizations proliferated and share a good quality of life in every aspect (Devi *et al.*, 2024). They give water and good quality of soil. People globally use to stay near the river for getting food, water, transport, and fertile agricultural land (Kumar *et al.*, 2018). River basin and sand bar are also home of diverse flora and fauna including edible stuffs for humans as well as livelihood. River banks and sand bar is also home of different types of medicinal plants (Lal *et al.*, 2024). During the floral diversity survey of Mahanadi River areas in June 2024, authors observed a plant, *Phyllanthus virgatus*. Authors collected the plant as it had very good population and discussed with local fishing community and found that the whole plant paste is used to cure skin itches.

Keeping this in view, an attempt has been made to document the available bioactive compounds in *P. virgatus* and an effort is made to validate the ethnomedicinal claims through phytochemical screening. *P. virgatus* is a small herb belonging to Phyllanthaceae family. Important morphological characters is its capsules are depressed spheres, about 2.8-3.2 mm wide. It usually grows near moist areas, river banks, agricultural land and near marshy areas. The present study highlights the importance of riparian herb as medicinal agent.

Methodology

Collection of plant sample and preparation of extracts

The whole plants of *Phyllanthus virgatus* were collected from the bank of Mahanadi River area of Cuttack district of Odisha state in May 2024 (Figure 1). The plant was identified by the senior author. The Soxhlet method of extraction was adopted using different solvents for phytochemical analysis (Devi *et al.*, 2023). Detection of nine secondary metabolites was carried out using standard methods (Tallur *et al.*, 2024; Valigatla *et al.*, 2024). An herbarium specimen is deposited to the Herbarium Unit, Biodiversity and conservation Lab, APRF, Cuttack, Odisha (Figure 2).

Qualitative phytochemical analysis

Test for tannin

About 1 ml of the filtrate of whole plant extract was taken. Added about 3-5 drops of 0.1% lead acetate solution into it. The gelatinous precipitate formation confirmed the presence of tannin (Kumar *et al.*, 2013).

Test for saponin

About 1 ml of the filtrate of whole plant extract was taken and mixed with 1 ml of distilled water and allowed to shake it vigorously. The persistent froth formation after shaking confirmed the presence of saponin (Kumar *et al.*, 2017).

Test for flavonoids

About 1 ml of the filtrate of whole plant extract was taken and added 2 ml of 2 % NaOH solution followed by H₂SO₄ into it. The colour initially turned to an intense yellow colour with the addition of NaOH solution and later become colourless with sulphuric acid. This change in colour confirmed the presence of flavonoids.

Test for terpenoids

About 2 ml of the filtrate of whole plant extract was added with 6 drops of chloroform and placed it in the water bath for few minutes. Then 6 drops of concentrated H₂SO₄ were added. The appearance of reddish-brown interface confirmed the presence of terpenoids (Kumar and Jena, 2014).

Test for phenolic compounds

About 1 ml of the filtrate of whole plant extract was taken. Few drops of 5% Ferric chloride solution were added into the filtrate extract. The appearance of bluish black colour provides the positive result of the phenolic compounds (Chandra *et al.*, 2024).



Figure 1: Collection of Whole plant of *P. virgatus* for experimental work

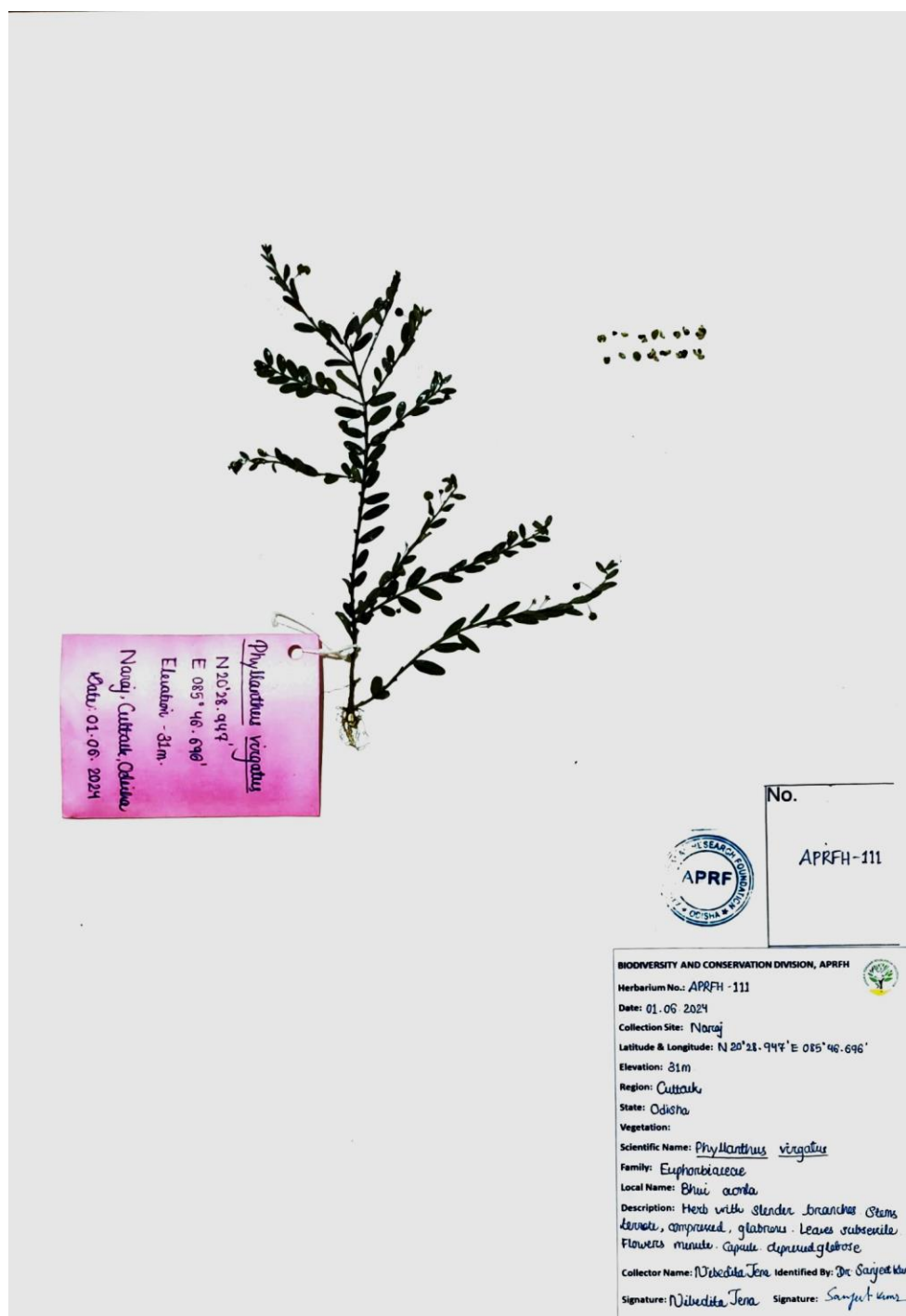


Figure 2: Herbarium specimen of *P. virgatus*



Figure 3: Habitat of *P. virgatus*

Test for reducing sugar

About 1 ml of the filtrate of whole plant extract was taken and 2 drops of Fehling's solution A followed by Fehling's solution B were added and kept it in water bath for few minutes. The presence of red orange precipitate confirmed the positive result of the test for reducing sugar (Vajha *et al.*, 2024).

Test for steroids

About 1 ml of the filtrate of whole plant extract was taken. 1 ml of chloroform and 1 ml of concentrated sulphuric acid was added into it. The appearance of upper red and lower yellow with green fluorescence provide the positive result for the presence of steroids.

Test for alkaloids

About 1 ml of the filtrate of whole plant extract was taken and added 3 to 4 drops of Dragendroff's reagent. The formation of reddish-brown precipitate confirmed the presence of alkaloids.

Test for carbonyl compounds

About 1 ml of the filtrate of whole plant extract was taken and added 3 to 4 drops of 2,4- Di-nitrophenylhydrazine reagent. The formation of yellow crystal confirmed the presence of carbonyl compounds.

Results and discussion

During field survey near Mahanadi River areas, authors found that whole plant paste of *P. virgatus* is used to treat skin itches and keeping this in view, phytochemical screening was carried out. The results of phytochemical screening revealed that tannin, saponin, phenolic compounds and reducing sugars are present in aqueous and methanol extracts (Table 1; Figures 4-6) but terpenoids are also present in ethanol extract. The presence of tannin might be responsible to treat skin itches (Nakamura *et al.*, 2018).

Table 1: Phytochemical screening using different extracts of *P. virgatus* whole plant

Bioactive compounds	Aqueous	Methanol	Ethanol
Tannin	+ve	+ve	+ve
Saponin	+ve	+ve	+ve
Flavonoids	-ve	-ve	-ve
Terpenoids	-ve	-ve	+ve
Phenolic compounds	+ve	+ve	+ve
Reducing sugar	+ve	+ve	+ve
Steroids	-ve	-ve	-ve
Alkaloids	-ve	-ve	-ve
Carbonyl compounds	-ve	-ve	-ve

(+ve: positive; -ve: negative)

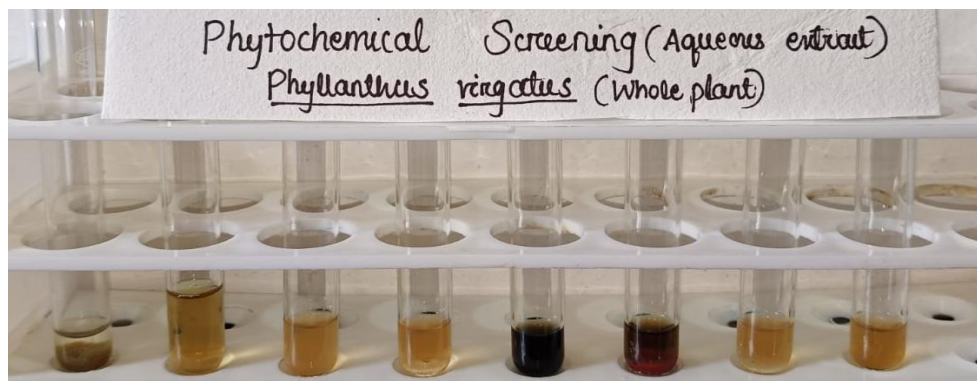


Figure 4: Screening of secondary metabolites using aqueous extract of *P. virgatus* whole plant

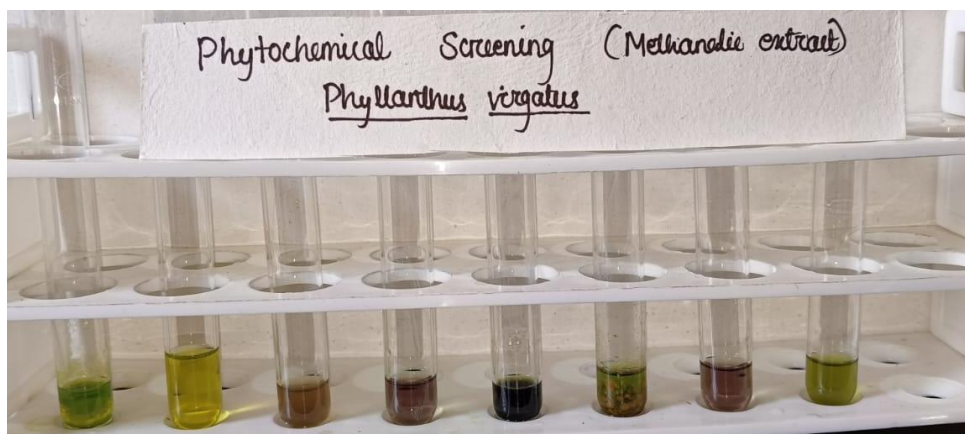


Figure 5: Screening of secondary metabolites using methanolic extract of *P. virgatus* whole plant



Figure 6: Screening of secondary metabolites using ethanolic extract of *P. virgatus* whole plant

Conclusion

The study concludes that riverine plants have a wide range of applications in the field of ethnopharmacology. To design novel drugs, it is necessary to investigate the ethnomedicinal applications of plants found in Indian river basins.

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