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Preliminary Phytochemical Screening of Leaf and Stem Extracts of *Homalanthus Populifolius* Graham

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ABSTRACT

This study explores the phytochemical composition and screening of leaf and stem extracts from *Homalanthus populifolius*, a member of the Euphorbiaceae family, collected from the shola forests of Coonoor in the Western Ghats. Our goal is to evaluate the effectiveness of extraction methods, such as petroleum ether, hydroalcoholic, and aqueous, on yield and bioactivity. Using a modified maceration technique, we analysed the phytochemical properties of the extracts. Key findings include the presence of alkaloids and flavonoids in both leaf and stem extracts, tannins predominantly in hydroalcoholic extracts, proteins in leaf extracts, steroids in aqueous and some petroleum ether extracts, and consistent phenols across all solvents. Saponins showed inconsistent presence in leaf extracts. These results underscore the phytochemical diversity, indicating potential for pharmacological applications and traditional medicine.

Key words: Homalanthus populifolius, phytochemical screening.

Introduction

The exploration of plants for their therapeutic properties has long been a cornerstone of traditional medicine, bridging ancient wisdom and modern science. Among the vast array of plant species, *Homalanthus populifolius* emerges as a distinguished representative of the Euphorbiaceae family, known not only for its aesthetic appeal but also for its rich biochemical profile. This plant, native to the biodiverse Western Ghats in India, particularly thrives in the serene, mist-laden shola forests of Coonoor, an area celebrated for its unique ecosystems and endemic species.

Research on bioactive compounds produced from plants, both quantitative and qualitative, depends on the selection of the most effective extraction method (Sasidharan *et al.*, 2011). Extraction, the first stage in researching a medicinal plant, significantly affects the results. The term "sample preparation techniques" is another term for extraction methods. The bulk of the time, this field of study is neglected and conducted by inexperienced researchers (Azmir *et al.*, 2013), despite sample preparation methods occupying almost two-thirds of an analytical chemist's time resources. A survey found that most scholars believe sample preparation is essential for any analytical investigation (Huie, 2002). Moreover, the study aims to intricately analyse the relationship between the identified secondary metabolites and their potential therapeutic benefits, underscoring the crucial need to conserve our rich botanical heritage. Through this research, we aspire to contribute valuable insights into the medicinal uses of plants while championing the preservation of biodiversity in the remarkable landscapes of the Western Ghats.

Aim

This study aims to perform a preliminary phytochemical screening of extracts from *Homalanthus populifolius* in the Euphorbiaceae family by evaluating different extraction methods.

Objectives

- > To compare the efficacy of different extraction solvents (petroleum ether, hydroalcoholic, and aqueous)
- > To analyse the phytochemical constituents in the plant extracts that contribute to their medicinal properties.



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Homalanthus populifolius

Assessment by: Botanic Gardens Conservation International (BGCI) & IUCN SSC Global Tree Specialist Group



The newly identified *Homalanthus populifolius*, first documented in the enchanting landscapes of Coonoor, India, and the majestic Western Ghats, stands as a testament to nature's resilience. It is categorised as Least Concern (LC) in the IUCN Red List. *H. populifolius* inspires us to cherish and protect the wonders of our biodiversity.

Materials and methods

Plant collection and authentication

The plant was collected from the shola forests of Coonoor in the Western Ghats, which are home to the most significant wild medicinal plants. The Botanical Survey of India, Southern Regional Centre, Coimbatore, Tamil Nadu, has confidently identified and authenticated.

We expertly collected the leaves and stem barks of *H. populifolius*, which were thoroughly washed under running water before being shade-dried and finely powdered.

Extraction of medicinal plants using a modified maceration technique:

This study extracted bioactive components from medicinal plants using a modified maceration process. To make extraction easier, 500 millilitres of different solvents were used to suspend around 100 gms of dried leaf and stem powder, which had been precisely weighed. Separate solvent systems, such as petroleum ether for non-polar molecules, hydroalcoholic combinations to target a wide range of polar and non-polar components, and distilled water for polar phytochemicals, were used to treat the plant materials (Abubakar AR and Haque M. 2020).

To improve solvent action and encourage component solubility, the mixtures were macerated for 48 hours at a constant temperature for each extraction, which was carried out under carefully monitored circumstances (Ghenabzia, I et al., 2023). The extracts were concentrated using a rotary evaporator set at 40°C under reduced pressure after the maceration time. To eliminate the solvent without destroying the delicate bioactive substances included in the extracts, this step is essential.

Following the extraction and concentration procedures, each extract's percentage yield was computed as weight/weight (% w/w), enabling a comparison of how well each solvent extracted the targeted phytochemicals. This improved method guarantees the preservation of the medicinal qualities of the plant components that are extracted while also optimising production.

Preliminary phytochemical screening

Standard techniques were used to detect nine secondary metabolites.

Phytochemical screening of the extracts and fractions, standard procedures were employed (Kokate *et al.*, 2006; Evans, 2009; Kumar *et al.*, 2013). Researchers looked into the presence of alkaloids (Dragendorrf's test), flavonoids (alkaline reagent test), tannins (Ferric chloride test), protein (Ninhydrin test), steroids (Salkowski test), phenolics (Ferric chloride test), saponins (foam test), cardiac glycosides (Kellerkilliani test), and fixed oil and fat (spot sample) Brain and Turner 1975 and Evans 1996.

Test for tannin:

The leaf and bark extracts were taken in approximately 1 millilitre. It was mixed with three to five drops of a 10% lead acetate solution. The presence of tannin was verified by the production of a gelatinous precipitate.

Test for saponin:

One millilitre of the leaf and bark extracts was mixed with one millilitre of distilled water. After adding, give it a good shake and look for the lingering foam to check that saponin is present.

Test for flavonoids:

A volume of one millilitre was extracted from the leaves and bark. 3–4 drops of diluted HCL and 2 millilitres of 2% NaOH solution were added. When exposed to NaOH solution, the hue first became intensely yellow before becoming colourless. This colour shift verified that flavonoids were present.

Test for phenolic compounds:

We took around 1 millilitre of the filtrate. We added a few drops of a 5% ferric chloride solution. The phenolic group test yields a positive result when the colour is dark, bluish-black.

Test for steroids:

The sample was mixed with 1 mL of concentrated sulfuric acid and 1 mL of chloroform. Steroids are present when the upper layer is red and the lower layer is yellow, with green fluorescence.

Test for alkaloids:

About 1 ml of the bark extract was taken, and 3 to 4 drops of Dragendroff's reagent. The formation of a reddish-brown precipitate confirmed the presence of alkaloids.

Test for cardiac glycoside:

The Keller-Killiani test was used to evaluate the extracts test was used. After mixing 2 mL of extract with 1 mL of acetic acid and two drops of ferric chloride, 2 mL of concentrated sulphuric acid was added, and the colour shift was noted. A positive test result for cardiac glycosides was determined by the appearance of a reddish-brown colour.

Test for protein

A few drops of the 0.2% ninhydrin reagent were added to 2 millilitres of the samples, and they were heated for five minutes. The presence of amino acids is shown by the creation of the blue hue.

Test for fats and oils

Heat each of the six extracts in a water bath for one to two hours after treating each one separately with 0.5N alcoholic potassium hydroxide and a drop of phenolphthalein. The presence of fixed oils and fats in these extracts is indicated by the soap production or partial neutralisation.

Result

The percentage yields of the extracts:

The hydroalcoholic stem extract of *H. populifolius*, prepared using the maceration technique, had the lowest yield at 2%. In contrast, the Petroleum ether leaf extract of *H. populifolius*, also prepared by the maceration method, had the highest yield at 8%.

Preliminary phytochemical screening of the leaves and stems of *H. populifolius*.

The phytochemical analysis of various extracts of H. populifolius is summarised in Table 1. The preliminary phytochemical findings indicate that H. populifolius contains a range of compounds in its different extracts, including alkaloids, flavonoids, fixed oils and fats, phenols, proteins, saponins, tannins, terpenoids, and cardiac glycosides. Notably, cardiac glycosides were absent in the leaf and fixed oils and fats, as well as in the stem extracts. However, alkaloids, flavonoids, fixed oils and fats, phenols, and terpenoids were present across all three types of extracts: petroleum ether, hydroalcoholic, and aqueous. In contrast, the overall extracts from the leaf's petroleum ether, hydroalcoholic, and aqueous contained alkaloids, flavonoids, fixed oils and fats, phenols, proteins, saponins, tannins, terpenoids, and glycosides. Phenols were found in all three extracts.

Table 1: Preliminary phytochemical screening of leaf and stem.

Compounds	Tests	Leaf			Stem		
		Petroleum ether	Hydroalcoholic	Aqueous	Petroleum ether	Hydroalcoholic	Aqueous
Alkaloids	Dragendroff's test	+	+	+	+	+	-
Flavonoids	Ammonia test	+	+	+	+	+	-
Tannins	Lead Acetate test	+	+	+	-	+	-
Proteins	Ninhydrin test	+	+	+	-	-	+
Steroids	Salkowski's test	+	-	+	-	+	-
Phenols	Ferric chloride test	+	+	+	+	+	+
Saponins	Foam test	+	+	+	-	-	+
Cardiac Glycosides	Foam test	-	-	-	+	+	-
Fixed oils and fats	Spot test	+	+	+	-	-	-

(+) = Present; (-) = Absent

Discussion

The study of *Homalanthus populifolius* shows it contains various beneficial compounds, including alkaloids, flavonoids, phenols, and saponins. Notably, it lacks cardiac glycosides, indicating a unique chemical profile Table 1. The consistent presence of alkaloids and flavonoids suggests potential health benefits, including the reduction of inflammation and oxidative stress. The presence of phenols supports its strong antioxidant properties. Some compounds, such as cardiac glycosides and certain tannins, are not found in all extracts, which indicates selective biochemical processes. Overall, *H. populifolius* shows promise for further research into its health applications.

For *H. macradenius*, the study highlights its antioxidant, antibacterial, and anti-inflammatory properties. The presence of phenolics and flavonoids likely contributes to its health benefits (Mabolo *et al.*, 2025). Safety tests indicate it can be safely consumed at the tested doses, but more research is needed to confirm long-term safety and proper dosing. *H. populneus* (Giesel.) Pax. indicates its potential as an alternative HIV treatment, as it reduces CD4 T cells while increasing cytotoxic T cells, thus enhancing immune responses. Further research is needed to determine effective concentrations and explore their mechanisms (Sintya *et al.*, 2019). Collaboration across fields is vital to uncover *H. populneus* potential in treating HIV/AIDS, especially in areas with limited access to standard treatments. Overall, this research opens new avenues for HIV therapy.

CONCLUSION

According to the results of Preliminary qualitative phytochemical screening indicates that the plant contains various secondary bioactive compounds, such as alkaloids, flavonoids, phenols, tannins, steroids, and cardiac glycosides. These compounds in the leaves and stems can be used for traditional treatments and pharmaceutical drug preparation.

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