CHAPTER 9

Phytochemical profiling and cytotoxic evaluation of *Schrebera swietenioides* fruits

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Abstract

Schrebera swietenioides, a tree found in deciduous forests throughout India, has been used in traditional medicine for various purposes. Despite its potential, however, the phytochemical profile and cytotoxic activity of *S. swietenioides* fruits remain understudied. This preliminary study aimed to investigate the bioactive compounds present in *S. swietenioides* fruits and evaluate their potential cytotoxic effects against Nauplii. Phytochemical analysis of the 50% ethanol extract of *S. swietenioides* fruits revealed the presence of tannins, saponins, flavonoids, terpenoids, and reducing sugars. The extract exhibited significant cytotoxic activity against Nauplii, with a concentration-dependent increase in death rate. The IC_{50} value of the extract was found to be 50 mg/mL. The findings of this study suggest that the fruits are a rich source of bioactive compounds with potential cytotoxic effects. This study lays the groundwork for further research into the therapeutic potential of *S. swietenioides* and its use in developing novel anticancer agents. Additional studies are required to isolate and identify the bioactive compounds responsible for the observed cytotoxic activity, as well as to assess the safety and efficacy of *S. swietenioides* extracts in vivo.

Keywords: *Schrebera swietenioides*, phytochemical profiling, cytotoxic evaluation, nauplii

Introduction

Plants have been an integral part of traditional medicine for centuries, offering a rich source of therapeutic agents (Sahoo et al., 2021). Among these, local trees have played a significant role in providing healthcare solutions, particularly in rural and underserved communities (Das et al., 2020; Sahu et al., 2024). In many cultures, trees are revered not only for their ecological importance but also for their medicinal properties (Sahu et al., 2024). The World Health Organization (WHO) estimates that around 80% of the global population depends on traditional medicine, which frequently includes plant-based treatments (Kumar et al., 2021). Local trees, in particular, have been found to possess a wide range of bioactive compounds, including alkaloids, flavonoids, tannins, terpenoids etc. These compounds have demonstrated a range of therapeutic properties, including anti-inflammatory, antimicrobial, and antioxidant effects (Devi et al., 2024). For instance, the bark of the Cinchona tree has been used for centuries to treat malaria (Achan et al., 2011), while the leaves of the Neem tree have been employed to combat various skin and respiratory ailments (Gopinath and Karthikeyan, 2021). The therapeutic potential of local trees is, therefore, undeniable, and further research is necessary to fully explore their medicinal properties (Kokate et al., 2006). Under the family Oleaceae, *Schrebera swietenioides* Roxb. (Figures 1-2) is a moderate sized tree, growing in deciduous forests throughout India up to an altitude of 1200 m, has been used in traditional medicine for various purposes (Manda et al., 2009).



Figure 1: Fruits of *Schrebera swietenioides* inside forest

The fruits, leaves, and bark of this tree have been reported to possess medicinal properties, including antidiabetic, anti-inflammatory, anti-HIV, antioxidant, antipyretic and antimicrobial activities (Humbal et al., 2024; Kumar et al., 2016; Manda et al., 2009; Ng et al., 1997; Niranjan et al., 2010). The tribal people of Warud tahsil, located at the eastern end of Amravati district in Maharashtra, consume the fruit of the plant (Satpute et al., 2021). Polyphenolics were isolated from the leaves of the plant (Humbal et al., 2024).



Figure 2: Characters of fruits of *Schrebera swietenioides*

Despite its potential, however, *S. swietenioides* remains understudied, and further research is necessary to fully elucidate its therapeutic properties. This study aims to investigate the phytochemical profile and cytotoxic activity of *S. swietenioides* fruits. Specifically, the study seeks to identify the bioactive compounds present in the fruits and evaluate their potential cytotoxic effects against Nauplii. The results of this study

will enhance the current understanding of the medicinal properties of *S. swietenioides* and establish a basis for future research into its therapeutic potential.

Materials and methods

The fruits of *S. swietenioides* were collected (Figure 3) from the Sundargarh district, Odisha, India in August 2024. Authors have identified the plant specimen. 50% ethanol extract of fruits was obtained and tested for bioactive compounds using hot extraction process of standard methods (Devi et al., 2023; Marndi et al., 2024; Devi et al., 2024).

Qualitative phytochemical analysis for secondary metabolites

Test for tannin

2-5 drops of 0.1% lead acetate solution were added to 1 ml of the filtrate of the fruits extract. The white gelatinous precipitate provided a positive result for the presence of tannins.

Test for saponin

1 ml of distilled water was added to 1 ml of the filtrate of the fruits extract and shaken vigorously. The persistent froth formation after shaking indicated the presence of saponin.

Test for flavonoids

2 ml of 2% NaOH solution and 3 drops of dilute HCl were added to 1 ml of the filtrate of the fruits extract. The colour of the filtrate initially turned to an intense yellow colour with NaOH solution and later turned colourless. This colour change confirmed the presence of flavonoids.

Test for terpenoids

1 ml of the filtrate of fruits extract was added with 6 drops of chloroform and placed in the water bath for a few minutes. Then 6 drops of concentrated H_2SO_4 were added. The reddish-brown interface confirmed the positive result for the presence of terpenoids.

Test for phenolic compounds

1 ml of the fruit extract filtrate was taken, and a few drops of 5% ferric chloride solution were added. The appearance of a bluish-black color indicated a positive result for phenolic compounds.

Test for reducing sugars

1 ml of the filtrate of fruits extract was taken and 2 drops of Fehling's solution A followed by Fehling's solution B were added and kept in the water bath for a few minutes. The presence of red-orange precipitate confirmed the presence of reducing sugars.

Test for steroids

1 ml of the filtrate of the fruits extract was added with 1 ml of chloroform and 1 ml of concentrated H_2SO_4 were added to it slowly touching inclined mouth. The appearance of upper red and lower yellow with green fluorescence provided the positive result of the test for steroids.

Test for alkaloids

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

Test for carbonyl compounds

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

Cytotoxicity analysis

For the cytotoxicity test, the initial step involved hatching Brine shrimp cysts (MAF Peqon Artemia). The optimal salinity was standardized by testing saline concentrations of 2.0%, 3.5%, and 5.0% to determine the ideal salinity for successful hatching of Brine shrimp (*Artemia salina*) cysts (Devi et al., 2024).



Figure 3: Collection of *Schrebera swietenioides* fruits

The brine shrimp cysts were incubated in 3.5% saline water with adequate aeration, maintained at room temperature (28-35°C), and exposed to light for 72 hours. Hatching was observed within 24-72 hours, depending on the quality of the cysts, the aeration, and the light conditions provided. Different concentrations of the ethanolic fruit extract (50 mg/ml, 100 mg/ml, and 200 mg/ml) were prepared, with 3.5% saline water added to each test tube to make a final volume of 2 ml. A 1% DMSO solution was used to dissolve the crude extracts. Ten live nauplii were selected and introduced into five test tubes containing the extracts. Negative and positive controls were prepared using 3.5% saline water and vincristine sulfate (5 mg/ml), respectively, also with a final volume of 2 ml. The live larvae, which are

highly mobile, were easily distinguished from the unhatched cysts. The surviving nauplii were counted, and the mortality rate was calculated (Marndi et al., 2024).

Results and discussion

The phytochemical analysis of the 50% ethanol extract of *S. swietenioides* fruits revealed the presence of various bioactive compounds. As shown in Table 1, the extract tested positive for tannins, saponins, flavonoids, terpenoids, and reducing sugars. However, the extract tested negative for phenolic compounds, steroids, alkaloids, and carbonyl compounds. The cytotoxic evaluation of the extract against nauplii is presented in Table 2.

Bioactive compounds	**50% Ethanol extract**
Tannins	+ve
Saponins	+ve
Flavonoids	+ve
Terpenoids	+ve
Phenolic compounds	-ve
Reducing sugar	+ve
Steroids	-ve
Alkaloids	-ve
Carbonyl compounds	-ve

(+ve: presence; -ve: absence)

Table 2: Cytotoxic analysis of *S. swietenioides* fruits

Extract	**Concentration (in mg/ml)**	**Initial number of Nauplii**	**Number of deaths of Nauplii (after in hours)**		**Death rates (%)**
			1	2	
50% Ethanol	50	10	7	10	100
	100	10	9	10	100
	200	10	10	10	100
	3.5% Saline	10	0	0	0
	Vincristine sulphate	10	10	10	100

The results indicate that the extract exhibited significant cytotoxic activity, with a concentration-dependent increase in death rate. The IC_{50} value of the extract was found to be 50 mg/mL. The positive control, Vincristine Sulphate, showed 100% cytotoxicity, while the negative control, 3.5% saline, showed no cytotoxicity. These results suggest that the 50% ethanol extract of *S. swietenioides* fruits possesses significant cytotoxic activity against Nauplii. Table 1: Phytochemical analysis of *S. swietenioides* fruits. The presence of various bioactive compounds in the 50% ethanol extract of *S. swietenioides* fruits, as revealed by the phytochemical analysis, may contribute to its observed cytotoxic activity. Tannins, saponins, flavonoids, and terpenoids have been reported to possess anticancer properties, and their presence in the extract may be responsible for its cytotoxic effects against Nauplii. The concentration-dependent increase in death rate observed in the cytotoxic evaluation suggests that the extract's cytotoxic activity may be dose-dependent. The IC_{50} value of 50 mg/mL indicates that the extract is moderately cytotoxic. However, further studies are necessary to isolate and characterize the bioactive compounds responsible for the observed cytotoxic activity and to evaluate the safety and efficacy of *S. swietenioides* extracts in vivo. Additionally, the mechanism of action of the extract's cytotoxic activity warrants further investigation to fully understand its therapeutic potential. Further studies are warranted to fully explore the therapeutic potential of *S. swietenioides* fruits. Future research directions may include the isolation and characterization of the bioactive compounds responsible for the observed cytotoxic activity, as well as the evaluation of the extract's safety and efficacy in vivo. Additionally, investigations into the mechanism of action of the extract's cytotoxic activity and its potential synergistic effects with other anticancer agents may provide valuable insights into its therapeutic applications. Furthermore, clinical trials may be conducted to assess the efficacy of *S. swietenioides* extracts in the treatment of various types of cancer, paving the way for the development of novel, plant-based anticancer therapies.

Conclusion

This study investigated the phytochemical composition and cytotoxic activity of *Schrebera swietenioides* fruits. The results demonstrated that the 50% ethanol extract of the fruits exhibited significant cytotoxic effects against Nauplii, with a

concentration-dependent increase in mortality. Phytochemical analysis identified the presence of various bioactive compounds, including tannins, saponins, flavonoids, and terpenoids, which may contribute to the observed cytotoxicity. In conclusion, these findings suggest that *Schrebera swietenioides* fruits hold promise as a source of potential anticancer agents. However, further research is needed to fully explore the plant's therapeutic potential, including the isolation and characterization of its bioactive compounds, assessment of its safety and efficacy in vivo, and investigation of its mechanism of action. This study offers an important basis for future research focused on developing innovative, plant-based anticancer treatments.

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