

Chapter 1

Phytochemical and toxicity evaluation of *Vincetoxicum fasciculatum* fruits: an unexplored plant of Apocynaceae

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Abstract: *Vincetoxicum fasciculatum*, a plant species of Apocynaceae family with potential medicinal properties, was subjected to phytochemical analysis and cytotoxicity evaluation. Phytochemical screening of the hydroethanolic extract of the fruits revealed the presence of tannins, saponins, flavonoids, phenolic compounds, and reducing sugars. Cytotoxicity analysis using brine shrimp nauplii showed that the extract exhibited significant cytotoxic activity, with a death rate of 100% at concentrations of 50-200 mg/mL after 3 hours of exposure. These findings suggest that *V. fasciculatum* possesses bioactive compounds with potential therapeutic applications and draw attention towards further investigation in pharmacological aspects.

Keywords: Bioactive compounds; phytochemicals; wild fruits

INTRODUCTION

The Apocynaceae family, comprising about 5300 species (Straub et al., 2020) and a rich source of medicinal plants with a long history of traditional use (Lv et al., 2020). Many members of this family have been employed in various folk medicine practices (Bhadane et al., 2018) to treat a range of ailments, including cancer

(Wong et al., 2011), diabetes (Bindu and Narendhirakannan, 2019) and cardiovascular diseases (Bachheti et al., 2022). The Apocynaceae family is particularly notable for its high diversity of bioactive compounds, including alkaloids, glycosides, and terpenoids, which have been shown to possess potent medicinal properties (Almagro et al., 2015; Fanta-Yadang et al., 2019; Kanwal et al., 2020; Naidoo et al., 2021; Mollica et al., 2022). Genus *Vincetoxicum* Wolf, an unexplored member of the Apocynaceae family, with potential medicinal applications (Prameela et al., 2024). Therefore, *V. fasciculatum* (Buch. -Ham. ex Wight) Kuntze is selected for present study. It is an unexplored plant and the secondary metabolites and biological activities remain undiscovered.



Figure 1: Fruit of *Vincetoxicum fasciculatum*

Present study aimed to investigate the phytochemical profile and cytotoxicity of the hydroethanolic extract of *V. fasciculatum* fruits, with a view to identifying potential therapeutic applications and contributing to the discovery of new medicinal agents from the Apocynaceae family.

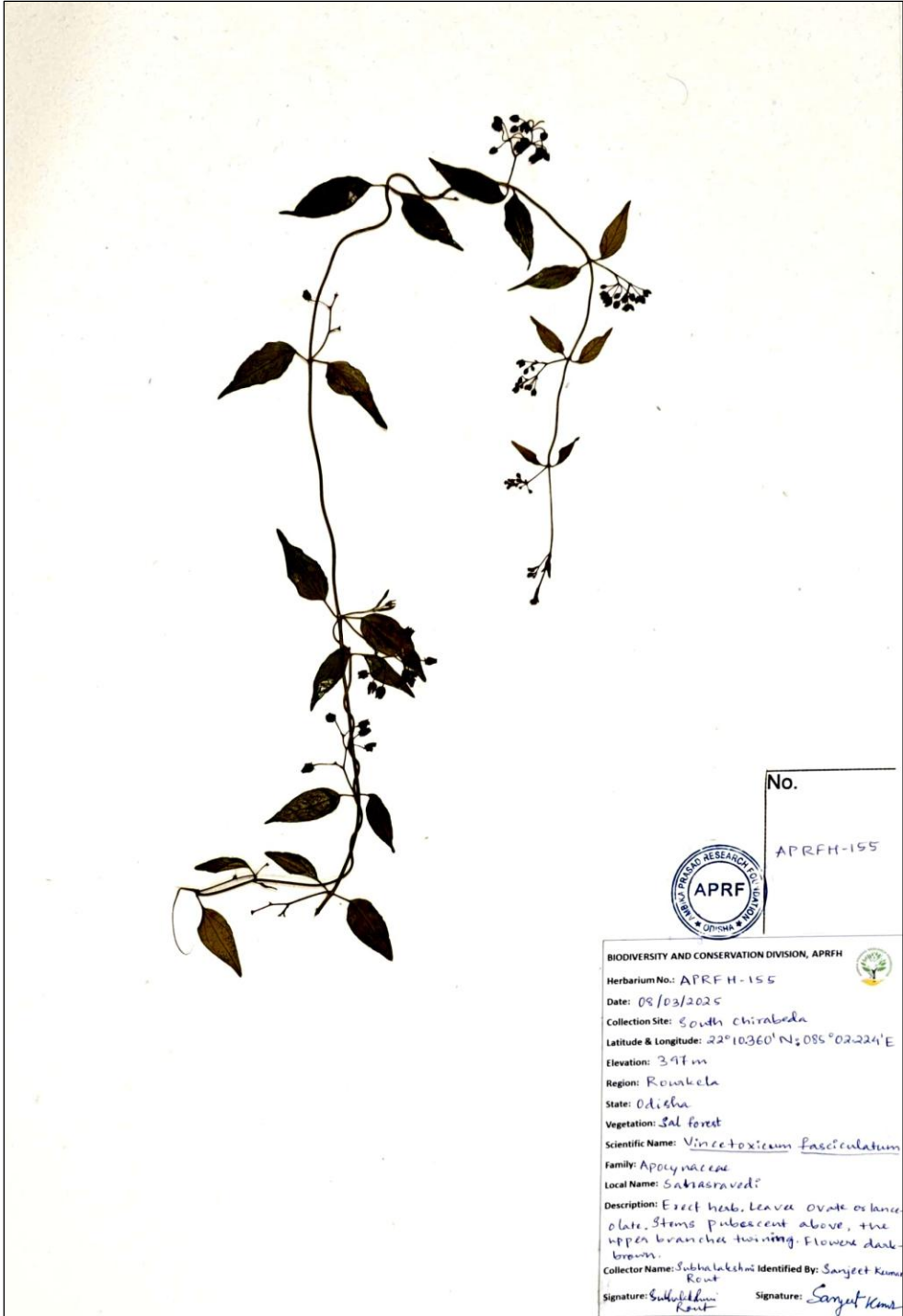


Figure 2: Herbarium specimen of *Vincetoxicum fasciculatum*

MATERIALS AND METHODS

The fruits of *V. fasciculatum* were collected from the Rourkela Forest Division of Sundargarh district of Odisha, India (Figure 1). The plant species was identified by the authors and a specimen was submitted at APRFH (Figure 2). The Soxhlet extraction method was adopted using different solvents for phytochemical analysis (Devi et al., 2023; Marndi et al., 2024; Jena et al., 2024). For toxicity analysis, hatching of Brine cysts was the initial process. Standardization of the optimum was done at 2.0, 3.5 and 5.0 % saline concentration to check the optimum salinity for proper hatching of Brine shrimp (*Artemia salina*) cysts (Kumar et al., 2012). The brine shrimp cysts are then incubated in 3.5% saline water with proper aeration, room temperature between 28-35°C and light for 48 hours. Proper hatching could be observed within 24-48 hours depending upon the quality of cysts, proper aeration and light provided. Different concentrations (13.4 mg/ml, 26.87 mg/ml, 40.31 mg/ml, 53.75 mg/ml and 67.18 mg/ml) of the fruit extracts were taken and 3.5% saline water was used to make the volume 2 ml in each test tube. 1% DMSO was used to dissolve the crude extracts. Ten nauplii were selected and introduced to five test tubes of the extracts. Positive and negative controls were prepared using vincristine sulphate (5 mg/ml) or potassium dichromate and 3.5% saline water respectively with a volume of 2 ml. The live larvae are highly mobile and thus differentiate from the unhatched cysts. The survivor nauplii were counted and analysed the death percentage (Kumar et al., 2012).

RESULTS & DISCUSSION

The phytochemical analysis of the hydroethanolic extract of *V. fasciculatum* fruits revealed the presence of several bioactive compounds, including tannins, saponins, flavonoids, phenolic compounds, and reducing sugars (Table 1).

Table 1: Phytochemical analysis of fruits of *Vincetoxicum fasciculatum* using hydroethanolic extract

Bioactive compounds	Hydroethanolic extract
Tannin	+ve
Saponin	+ve
Flavonoids	+ve
Terpenoids	-ve
Phenolic compounds	+ve
Reducing sugar	+ve
Steroids	-ve
Alkaloids	-ve
Carbonyl compounds	-ve

(+ve: Present; -ve: Absent)

Table 2: Cytotoxicity analysis of fruits of *Vincetoxicum fasciculatum* using hydroethanolic extract

Extract	Concentration (in mg/ml)	Initial number of Nauplii	Number of deaths of Nauplii			Death rates (%)
			After 1 hour	After 2 hours	After 3 hours	
Hydro- ethanolic extract	25	10	4	7	10	100
	50	10	7	10	10	100
	100	10	8	10	10	100
	200	10	10	10	10	100
	3.5% Saline	10	0	0	0	0
	Vincristine sulphate	10	10	10	10	100

The presence of these compounds is consistent with previous reports on the phytochemical composition of other Apocynaceae species. The cytotoxicity analysis of the hydroethanolic extract against brine shrimp nauplii showed significant cytotoxic activity, with a death rate of 100 % at concentrations of 50-200 mg/ml after 3 hours of exposure (Table 2). The LC₅₀ value of the extract was

found to be 25 mg/ml, indicating potent cytotoxic activity. The cytotoxic activity of the extract may be attributed to the presence of bioactive compounds (Kumar et al., 2013; Hossain et al., 2025). The results of present study suggest that *V. fasciculatum* may be a potential source of anticancer agents (Kumar et al., 2012). The results are also consistent with previous reports on the medicinal properties of other Apocynaceae species. The presence of bioactive compounds such as tannins, saponins, and flavonoids in the extract may contribute to its medicinal properties (Aradhana et al., 2022). Further studies are needed to isolate and characterize the bioactive compounds responsible for the cytotoxic activity and to evaluate the extract's anticancer potential in vitro and in vivo. Other researchers have also reported the medicinal values. Saheb (2014) reported that leaves of *Tylophora fasciculata* are used to treat wounds and ulcers during the survey of medicinal climbers from Nallamalais, Andhra Pradesh, India. Bhakshu et al., (2016) observed the anticandidal activity in *T. fasciculata*, and it showed inhibitory activity against *Candida albicans*. He also reported that the leaf pastes of *T. fasciculata* were applied externally to the wounds for treatment. Lakshmi et al., (2023) reported the presence of alkaloids, flavonoids, glycosides, saponins, and volatile oils in *T. fasciculata*. Mostafa et al., (2023) documented the ethno-medicinal uses of *T. fasciculata*. The above reports and present study indicate that there is a need for more explorer and bioactivity analysis works.

CONCLUSION

The present study demonstrates the phytochemical and cytotoxic properties of *Vincetoxicum fasciculatum* and highlights its potential as a source of medicinal agents. Further research is needed to fully explore the medicinal properties of this plant species.

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