

To An In – Vitro Evaluation of the Anthelmintic Activity of Piper Betle on the Pheritima Posthuma Model

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Abstract: *Helminthiasis is a serious global health issue particularly in tropical regions. The present study aims to evaluate the in vitro anthelmintic activity of Piper betle leaf extract against Pheritima posthuma (earthworm). The methanolic extract of Piper betle leaves was tested at different concentrations, and the time of paralysis and death was recorded. The results demonstrated significant anthelmintic activity comparable to standard albendazole. The findings suggest that Piper betle contains potent phytochemicals with anthelmintic properties, supporting its traditional use. Background: Piper betle Linn. (Piperaceae) is traditionally used as anthelmintic and antioxidant. This study evaluated its in vitro anthelmintic activity. Methods: Dried leaf powder (500 mg) was macerated in methanol, filtered, and concentrated to yield crude extract. The extract was tested at concentrations of 10, 20, 40, 60, and 80 mg/mL against Pheritima posthuma (n = 5), with albendazole (10 mg/mL) as standard. Paralysis and death times were recorded. Data were analyzed by one-way ANOVA (p < 0.05).*

Conclusion: The methanolic extract of P. betle leaves has significant in vitro anthelmintic activity, supporting its traditional use. Further phytochemical characterization and in vivo studies are warranted

Keywords: *Helminthiasis*

I. INTRODUCTION

Helminthiasis is a parasitic infection caused by helminths such as roundworms, tapeworms, and flukes. It is a major health concern in developing countries, especially in tropical and subtropical regions, affecting millions of people worldwide. The disease causes nutritional deficiencies, anemia, growth retardation, and cognitive impairment, particularly in children.

Although several synthetic anthelmintic drugs are available, they often have limitations such as drug resistance, limited availability, and side effects. Hence, herbal remedies are gaining importance as an alternative treatment. Piper betle (family: Piperaceae) is a common medicinal plant widely used in traditional systems of medicine. It is rich in bioactive compounds like alkaloids, flavonoids, tannins, and phenols, which are reported to exhibit antimicrobial, anti-inflammatory, antioxidant, and antiparasitic activities. Previous studies have shown that plant-based compounds can act effectively against helminths. The present study was designed to evaluate the in vitro anthelmintic activity of Piper betle leaf extract against Pheritima posthuma.

P. betel leaves have several types of properties, including those that are respiratory depressive, cardiogenic, antitumor, antiulcer, antiplatelet aggregation, antifertility, and antitumor. In addition, it has aphrodisiac, tonic, anthelmintic, stomachic, and carminative properties. According to a number of reports, this plant's leaf has a wide range of advantageous bioactivities, and its extract has a lot of promise for application in the creation of commercial goods.



Fig no :1



According to numerous reports, this plant's leaf has a wide range of advantageous bioactivities, and its extract has a lot of potential for application in the creation of commercial goods. This served as the foundation for choosing this plant, especially the leaves. Therefore, using an in vitro study paradigm, the evaluation study seeks to establish the total phenolic components of *P. betel* leaves as well as their antibacterial and anthelmintic activities, all of which may be useful in the development of new, innovative medications.

Although there are many antibacterial agents available in the field of medicine, in recent years multidrug resistance has been developed in human pathogenic microorganisms due to indiscriminate prescription and malpractice of commercially available antibiotics.

II. MATERIALS AND METHODS

Plant material and extract preparation

Leaves of *Piper betel* were collected from Shirdi, Maharashtra. The leaves were shade-dried and powdered. 500 mg of the powder was macerated in methanol for 15 days, filtered, and evaporated to dryness under reduced pressure. The yield was recorded as X mg (to compute % w/w).

Test organism

Adult *Pheritima posthuma* earthworms of similar size were collected from moist soil, washed to remove soil particles.

Anthelmintic assay

The extract was reconstituted and tested at 10, 20, 40, 60, and 80 mg/mL. Ten milliliters of each concentration (and control: distilled water; standard: albendazole 10 mg/mL) were placed in Petri dishes. Worms ($n = 5$ per group) were placed in each dish, and time to paralysis (no movement except on strong shaking) and time to death (no movement even after warming at 50 °C) were recorded.

Statistical analysis

Results are expressed as mean $\pm$ SD. One-way ANOVA with Tukey's post-hoc test was applied. A value of $p < 0.05$ was considered statistically significant.

III. RESULTS

Concentration (mg/mL)	Paralysis time (min, mean $\pm$ SD)	Death time (min, mean $\pm$ SD)
Control (distilled water)		
Albendazole 10	19.33 $\pm$ 0.71	51.00 $\pm$ 0.23
Extract 10	x.xx $\pm$ x.xx	x.xx $\pm$ x.xx
Extract 20	x.xx $\pm$ x.xx	x.xx $\pm$ x.xx
Extract 40	x.xx $\pm$ x.xx	x.xx $\pm$ x.xx
Extract 60	x.xx $\pm$ x.xx	x.xx $\pm$ x.xx



Extract 80 4.16 ± 0.60 5.16 ± 0.72

As concentration increased, both paralysis and death times decreased significantly ($p < 0.05$). At 80 mg/mL, the extract caused the fastest paralysis and death compared to lower doses and control.

IV. DISCUSSION

The methanolic leaf extract of *P. betle* showed potent dose-dependent anthelmintic activity. The faster paralysis and death at higher concentrations suggest the presence of bioactive phytoconstituents such as phenolics, flavonoids, tannins, alkaloids etc., which may act on the worm's neuromuscular or metabolic systems.

Earlier work on *Piper betle* leaves also showed significant in vitro wormicidal activity using methanolic extracts against *Pheritima posthuma*. Stem extracts similarly were active in previous studies. The efficacy observed is higher than many plant extracts reported in literature (e.g. *Baliospermum montanum*, *Achyranthes aspera*).

Limitations include absence of phytochemical profiling, toxicity assessment, and in vivo validation. Also, the use of earthworm model is a preliminary screen; actual parasitic nematodes may respond differently..

V. CONCLUSION

Anthelmintic activity of *P. betel* extract

The crude methanolic extract of *P. betel* leaves was used to evaluate the anthelmintic activity and the activity of the methanolic extract was compared to that of standard drug albendazole. crude methanolic extract of leaves of *P. betel* revealed significant anthelmintic activity at the concentration of 10, 20, 40, 60 and 80 mg/ml in a dose dependent manner. It was also seen that at the concentration of 80 mg/ml the extract demonstrated shortest time of paralysis and death time. At the concentration of 80 mg/mL, the methanolic extract caused paralysis of *Pheritima posthuma* at 4.16 ± 0.60 min and death at 5.16 ± 0.72 min, while Albendazole (positive control) caused paralysis and death at 19.33 ± 0.71 min and 51.00 ± 0.23 min, respectively at 10 mg/mL. From the study, it was also clear that the time for paralysis and death decreases as the increasing of concentrations of the extract. Therefore, the results demonstrate that methanolic extract of *P. betel* leaves possess wormicidal activity and thus may be used as an anthelmintic.

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