An *in vitro* evaluation of the anthelmintic activity of *Hedychium spichatum* rhizomes and *Zingiber zerumbet* rhizomes on the Pheritima Posthuma model: A comparative study

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ABSTRACT

Ethanolic extracts of *Hedychium spichatum* rhizomes and *Zingiber zerumbet* rhizomes were taken for *in vitro* comparative studies on the anthelmintic activity against *Pheritima posthuma*. Different concentrations (25, 50, 100 mg/ml) of both the extracts were used for the activity. Varying albendazole concentrations (25, 50, 100 mg/ml) were used as a reference standard and normal saline (0.9% NaCl) was used for the control treatment. The results were expressed in terms of time in minutes to report the paralysis and time of death of the earthworms. The results obtained from the study indicate toward the anthelmintic activity, supporting folk use of both the plants when compared with the standard. The results also established that *Z. zerumbet* was a more potent candidature of as compared with *H. spichatum*.

Key words: Albendazole, anthelmintic activity, *Hedychium spichatum*, *Zingiber zerumbetw*

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INTRODUCTION

From the ancient times, indigenous drugs have been used in the Indian medicinal system to treat different ailments and to provide therapeutic benefits. Our traditional system of medicine has made use of the different parts of plants in different types of diseases, including anthelmintic, anti-inflammatory and antimicrobial activities. Kavirajes and Hakims are still using several medicinal plants to treat helminthiasis. During the recent years, medicinal chemistry has made great strides, especially in synthetic chemistry but, for the sake of therapeutic effect up to the level and nontoxic treatment for helminthiasis, the research of plant-derived drug therapy is mostly needed.[1]

Rhizomes of *Hedychium spichatum*, commonly known as Gandhapalashi or Kapur-kachari, belong to the Zingiberaceae family, and have been reported for their folklore use in treating inflammatory and hyperglycemic conditions.[1] The plant has also been evaluated for its cytotoxic activity by Reddy *et al.*[2] The survey published by Akhtar *et al.* reported the use of *H. spichatum* as an anthelmintic in the Indian–Pakistan region.[3-4]

Zingiber zerumbet, commonly known as Narkachur and Banadrak, also belongs to the Zingiberaceae family. Rhizomes of *Z. zerumbet* have been reported for antipyretic and analgesic activities.[5] Somchit *et al.* demonstrated the anti-inflammatory property of the ethanolic and aqueous extracts of *Z. zerumbet*.[6] Nadia *et al.* also reported that Zerumbone isolated from *Z. zerumbet* inhibited cancer cell growth of human ovarian and cervical origin.[7] However, Iqbal *et al*, performed a study on other plants of the Zingiberaceae and Cucurbutaceae families (*Zingiber officinale, Curcurbita mexicana*) in different anthelmintic models,[8] but *H. spichatum* and *Z. zerumbet* have not been evaluated scientifically for anthelmintic activity. In light of the above facts, this study has been designed to evaluate *H. spichatum* and *Z. zerumbet* for their anthelmintic activity against a Pheritima posthuma model.

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MATERIALS AND METHODS

Collection of plants

The plant *H. spichatum* (specimen no. 97377) and *Z. zerumbet* rhizome (specimen no. 97769) were collected from the fields of Kusumha village (Kushinagar, Uttar Pradesh). The plant was authenticated by the National Botanical Research Institute (NBRI), Lucknow.

Collection of earthworms

Earthworms were collected from Tendua, Gorakhpur, and were identified and deposited in the Department of Pharmacy, ITM, GIDA, GKP, India.

Preparation of the extracts

Shade-dried small pieces of *H. spichatum* and shade-dried powder of the *Z. zerumbet* rhizomes were subjected to hot percolation by the Soxhlet apparatus using ethanol as a solvent.

Procedure

The anthelmintic activity was performed according to the method of Ghosh *et al.* (2005) on the adult Indian earthworm *Pheritima posthuma*.[9,10] Albendazole, the standard drug, was diluted with normal saline to obtain 25, 50 and 100 mg/ml concentrations and was poured into Petri dishes. Ethanolic extracts of both plants were diluted with normal saline to obtain 25, 50 and 100 mg/ml concentrations. Normal saline (0.9% NaCl) alone served as the negative control.

All these dilutions were poured into the Petri dishes accordingly. Six groups of earthworms ($n = 6$) were taken for the study. Earthworms, nearly equal sizes (about 8 cm), were placed in each Petri dish at room temperature. Time for paralysis was noted down when no movement of any sort could be observed, except when the worms were shaken vigorously. Time of death for worms was recorded after ascertaining that the worms neither moved when shaken vigorously nor when dipped in warm water (50°C). The paralysis time and lethal time were recorded in terms of minutes.

RESULTS AND DISCUSSION

In vitro anthelmintic activity was performed and the paralysis time and lethal time were recorded. Statistical evaluation of the data was performed by one-way ANOVA. The results were expressed as mean ± SD using Graph Pad Instat 3 ($n = 6$).

The results show that for the 25 mg/ml concentration, albendazole showed the best activity for death time (124.83 ± 6.99 min) and the ethanolic extract of *H. spichatum* and *Z. zerumbet* showed a death time of 146 ± 2.828 min and 125.83 ± 5.23 min, respectively. Also, for the 50 mg/ml concentration, albendazole showed the highest activity against the worms (95.5 ± 4.84 min) and the ethanolic extract of *H. spichatum* and *Z. zerumbet* showed a death time of 137.5 ± 9.75 min and 112.33 ± 5.87 min, respectively. For the 100 mg/ml concentration, albendazole showed the least death time of 73.83 ± 4.167 min, and the ethanolic extract of *H. spichatum* and *Z. zerumbet* showed a death time of 96.66 ± 3.266 min and 76.2 ± 1.75 min, respectively. The paralysis and death times of both the plants along with the standard is given in Table 1. The study revealed that both the plants' ethanolic extracts had significant activity at the higher concentration (100 mg/ml). *Z. zerumbet* has shown better activity than *H. spichatum* at a higher concentration (100 mg/ml) compared with the standard, albendazole (100 mg/ml). The comparison of the death time of both the plants in different concentrations with respect to the standard is given in Figure 1.

CONCLUSION

The present study enabled us to conclude the potential use of ethanolic extracts of both *H. spichatum* and *Z. zerumbet* as anthelmintic agents against Pheritima posthuma. Extensive research is needed to determine the individual component responsible for the anthelmintic activity and molecular mechanism responsible for the same.

Table 1: *In vitro* anthelmintic effect of *Hedychium spichatum* and *Zingiber zerumbet* against *Pheritima posthuma*

Treatment	Concentration	Paralysis time (min)	Death time (min)
Albendazole (Std)	25 mg/ml	55.66 ± 4.59	124.83 ± 6.99
	50 mg/ml	43.33 ± 4.32	95.5 ± 4.84
	100 mg/ml	34.66 ± 3.327	73.83 ± 4.167
Hedychium spichatum (ethanolic extract)	25 mg/ml	78.16 ± 3.656	146 ± 2.828
	50 mg/ml	62.33 ± 4.131	137.5 ± 9.752
	100 mg/ml	44 ± 4.382	96.66 ± 3.266
Zingiber zerumbet (ethanolic extract)	25 mg/ml	74.35 ± 2.805	125.83 ± 5.23
	50 mg/ml	51.34 ± 1.24	112.33 ± 5.87
	100 mg/ml	37.86 ± 3.031	76.2 ± 1.75

± SD value, n = 6, $P < 0.01$

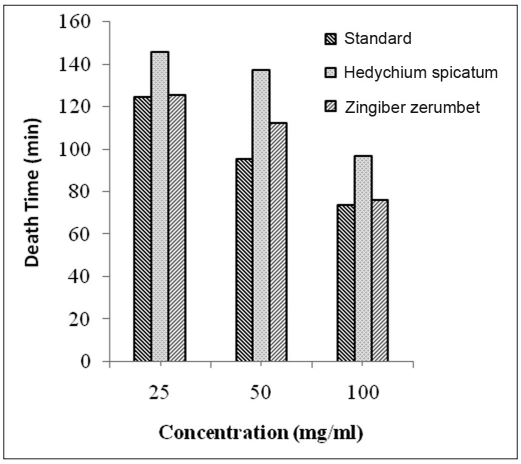

Figure 1: Comparative studies of death time of *Hedychium spichatum* and *Zingiber zerumbet* and the standard albendazole

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