

**Evaluation of Anthelmintic Activity of Aqueous Leaf  
Extract of *Clitoria ternatea* Linn.**Kamrun Nahar<sup>1</sup>, Muhammad Ashikur Rahman<sup>2</sup>, Most. Nazma Parvin<sup>1</sup> and  
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**ABSTRACT**

Development of anthelmintic resistance and high cost of conventional anthelmintic drugs led to the evaluation of medicinal plants as an alternative source of anthelmintics. The current study was aimed to evaluate the possible anthelmintic effects of crude fresh juice of leaves of *Clitoria ternatea* Linn. using of adult earth worm *Pheretima Posthuma*. Three concentrations (25, 50, 100mg/ml) of juice were studied for the determination of time of paralysis and death of the earth worms. Albendazole in same concentration as that of juice was considered as standard reference and normal saline as control. The result of the present study reveals that fresh juice significantly showed paralysis and also caused death of worms especially at higher concentration of 100mg/ml, as compared to standard reference Albendazole.

**Key word:** *Clitoria ternatea*, anthelmintic, earthworm, paralysis, Albendazole.

**INTRODUCTION**

Anthelmintics or antihelminthics are drugs that expel parasitic worms (helminths) from the body, by either stunning or killing them (The Mark Index, 1996). The WHO estimates that a staggering two billion people harbor parasitic worm infections. Parasitic worm also infect livestock and crops affecting food production with a resultant economic impact. Despite this prevalence of parasitic infections, the research on anthelmintic drug is poor. As per WHO, only few drugs are frequently used in the treatment of these parasite infections (Manoj et al., 2008). Traditional system of medicine reports the efficacy of several natural plants in eliminating worms (Dwivedi et al., 2009).

The plant *Clitoria ternatea* Linn. is a strangling and climbing herb with a strong base, ovate-oblong leaves and beautiful blue flowers, grown as an ornamental plant in gardens. This plant belongs to family Papilionaceae and is commonly called "Butterfly pea plant" in English and locally known as Aparajita and Nila in Bengali. The useful parts are root, bark, seeds and flowers. The plant used as colic gonorrhoea and skin disease. Root is used as, laxative and demulcent, aperients. Seeds are used as cathartic. Roasted & powdered seeds are used in the treatment of ascites, enlargement of abdominal viscera, weakness of sight, sore throat, tumors, dropsy & skin diseases (Ghani, 1998).

The plant has not been explored for its anthelmintic activity so far. The present study was therefore aimed to investigate the anthelmintic activity of the leaves of fresh juice with a view to justifying the use of the plant in the treatment of helminths.

**MATERIALS AND METHODS****Collection and Identification of plants**

The leaves of *Clitoria ternatea* Linn. were collected from Boldha Garden, Dhaka, Bangladesh and were identified at the Bangladesh National Herbarium, Mirpur, Dhaka where the Voucher specimen no: 35202 has been deposited. The collected plant parts were separated from undesirable materials or plants or plant parts. They were dried for one week. The plant parts were ground into a coarse powder with the help of a suitable grinder. The powder was stored in an airtight container and kept in a cool, dark and dry place until analysis commenced.

### Preparation of Fresh juice extract

Collected leaves were weighed (75g) and blended into liquification in 150ml of water. The mixture was then centrifuged at 150 rpm. The supernatant was filtered through sterile filter paper in to conical flask as the study extract 1ml of filtrate is expected to contain 0.5g i.e.500mg/ml.

### Chemicals and drugs

All chemicals and drugs were obtained commercially and were of analytical grade. Albendazole was collected from Square Pharmaceuticals Ltd., Bangladesh. DMF were purchased from Merck, Germany.

### Selection of worm

The assay was performed on adult earthworm, *Pheretima posthuma* due to its anatomical and physiological resemblance with the intestinal round worm parasites of human beings. Because of easy availability, earthworms have been widely used for the initial evaluation of anthelmintic compounds. Adult earthworms (*Pheretima posthuma*) were collected from moist soil of Savar area, of Dhaka in Bangladesh and washed with normal saline to remove all faecal matters. Then the worms were used for anthelmintic study. The earthworms of 3-5cm in lengths and 0.1-0.2cm in width were used for all the experimental protocol.

### Phytochemical Screening

The freshly prepared crude extract was qualitatively tested for the presence of various phytochemical constituents (Ghani, 2003).

### Anthelmintic Activity

Aqueous extracts from the *Clitoria ternatea* Linn. Leaves were investigated for their anthelmintic activity against *Pheretima posthuma*. The anthelmintic assay was carried as per the method found in the literature (Ajaiyeoba et al., 2001) with minor modifications. Fresh juice extracts of leaves of *Clitoria ternatea* Linn. were dissolved in minimum amount of DMF and the volume was adjusted to 10ml with saline water. All drugs and extract solutions were freshly prepared before starting the experiment. In each case, 3 earthworms were released into 10ml of desired formulation as follows: vehicle (5% DMF in normal saline), Albendazole (25mg/mg, 50mg/ml, 100mg/ml), or fresh juice extract (25mg/mg, 50mg/ml,100mg/ml) of leaves of *Clitoria ternatea* Linn..Observation was made for the time taken to paralysis and death of individual worm. Paralysis was said to occur when the worms were not able to move even in saline solution.

## RESULTS AND DISCUSSION

The results of phytochemical screening and *in vitro* anthelmintic activity of *Clitoria ternatea* Linn. of the aqueous extract are shown in Table1 and Table 2 respectively.

**Table 1: Results of Phytochemical screening of *Clitoria ternatea* Linn.**

Extract	Carbohydrate	Alkaloid	Glycoside	Tannin	Gum	Flavonoid	Saponin
AECTL	+	++	+++	+++	+	++	-

AECTL: Aqueous extract *Clitoria ternatea* Linn. leaves; (+): Present; (-): Absent

**Table 2: Results of *in vitro* evaluation of anthelmintic activity of *Clitoria ternatea* Linn.**

Test sample	Conc. mg/ml	Time taken for paralysis (minutes)	Time taken for death (minutes)
Vehicle (5% DMF in normal saline)	-----	-----	-----
Fresh aqueous juice extract of <i>Clitoria ternatea</i> Linn. leaves (n=3)	25	12.4±0.25	13.3±0.24
	50	10.4±0.24	11.3±0.26
	100	10.3±0.14	12.2±0.17
Albendazole	25	25.4±0.14	55.3±0.23
	50	18.3±0.28	38.2±0.17
	100	11.1±0.09	20.3±0.10

All Values represent Mean ± SEM; Values are significantly different from reference standard (Albendazole) where SEM: Standard error mean, n= Number of earth worms in each groups.

Preliminary phytochemical screening of fresh leaf juice extract of *Clitoria ternatea* Linn. showed the presence of glycoside, tannin, carbohydrate, flavonoid. The anthelmintic activity Fresh juice extract was observed in a dose dependent manner. The results of anthelmintic activity were comparable with standard drug, Albendazole. Literature review reveals that tannins which are chemically polyphenolic compounds (Bate-Smith et al., 1962) are responsible to produce anthelmintic activity (Niezen et al., 1995; Khadatkhar et al., 2008; Ghosha et al., 2007; Mali, 2007). As phytochemical analysis of the crude leaf extract of *Clitoria ternatea* Linn revealed the presence of the tannin among, other chemical constituents, it is possible that tannins contained in the extracts produced similar effects. Reported anthelmintic effect of tannins is that they can bind to free proteins in the gastrointestinal tract of host animal (Athnasiadou et al., 2001) or glycoprotein on the cuticle of the parasite (Thompson et al., 1995) and may cause death.

## CONCLUSION

The fresh juice extract of the leaves of *Clitoria ternatea* Linn. displayed profound anthelmintic activity in the study. But this study is preliminary type and it would be interesting to carry out further study for isolating the possible phytoconstituents and characterization of the active constituents which may be responsible for the mechanism of action.

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