

**IN VITRO ANTHELMINTIC ACTIVITY ETHANOLIC LEAVES EXTRACT OF GYNURA PROCUMBENS,
A PROSPECTIVE MEDICINAL PLANT**

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Abstract

Anthelmintic activity of crude *Gynura procumbens* leaves extract was evaluated using adult earthworms. *Gynura procumbens* (Lour.) Merr. (Family Asteraceae) is a medicinal plants often found in tropical Asian countries such as China, Thailand, Indonesia, Malaysia, Vietnam. Traditionally, it is widely used in a wide variety of countries to treat a wide variety of health disorders such as kidney discomfort, rheumatism, diabetes mellitus, constipation and hypertension. On the basis of traditional uses of *G. procumbens plant's leaves*, it seems to possess high therapeutic effect for treatment of various diseases making it a target for pharmacological studies aiming to validate and provide scientific evidence for the traditional claims of its efficacy. For anthelmintic activity standard albandazole preparation 15mg/ml showed paralysis time 11.8 and death time 20.6. The extract of *G. Procumbens* 25mg/ml (paralysis time 36.4 and death time 52.4), 50mg/ml(paralysis time 22 and death time 41) and 100mg/ml (paralysis time 16 and death time 26). The results of present study specify that the ethanolic extract significantly indicate paralysis and also caused death of worms in dose dependent manner.

Keywords: Anthelmintic activity, *Gynura procumbens*, Albandazole, Earthworms.

Introduction

For biological potent drugs natural sources are the major and important site for drug discovery in the new era of civilization [1]. In the different parts of the world there has been an expanding interest in the study of medicinal plants as natural products [2]. In third world countries helminthiasis is the common disease of all ages. The parasitic diseases cause severe morbidity by affecting population in autochthonous areas with major economic and social consequences [3]. Helminthic infections are among the most common infections in man, affecting a large proportion of the world's population. In developing countries, they pose a major threat to public health and contribute to the spread of malnutrition, pneumonia, anemia, eosinophilia and anemia. Parasitic diseases causing severe morbidity include lymphatic filariasis (a cause of elephantiasis), onchocerciasis (river blindness), and schistosomiasis. [4]. Anthelmintic or anti-helminthic are drugs that expel parasitic worms (helminths) from the body, by either stunning or killing them. They may also be called vermifuges (stunning) or vermicides (killing). Helminth infections are among the most widespread infections in humans, distressing a huge population of the world.

The development of anthelmintic drug-resistance in helminthes against synthetic drugs has been reported in number of countries [5], which gives a clear indication that control programs based exclusively on their use are not sustainable. Most of the worm infections are limited to tropical areas, but these may occur in individuals who have visited such infected areas and then infected in temperate areas. Several anthelmintics, such as praziquantel and albendazole, are contraindicated for certain groups of patients, such as pregnant or lactating women. This has led to the increase in interest of ethno medical practices across the world for the use of medicinal plants in treatment of helminthic diseases [6]. Day by day the progression of anthelmintic drug-resistance in helminthes against synthetic drugs has been reported in number of countries, which gives a clear or logical evidence that control programs based exclusively on their use are not sustainable. Most of the worm infections are limited to tropical areas, but these may occur in

individuals who have visited such infected areas and then infected in temperate areas. Praziquantel and albendazole are common anthelmintics, but these drugs are contraindicated for certain groups of patients, such as pregnant or lactating women [8]. This has encouraged the patients to increase in interest of ethno medical practices all over the world for the use of medicinal plants in treatment of helminthic diseases [9].

The objective of the present study is to evaluate anthelmintic activity of ethanolic extract of *Gynura Procumbens* as well as to rationalize the use in helminthiasis in folk medicine.

Methods

Experimental animal and preparation of Phosphate buffer saline

Live parasites *Paramphistomum cervi* (Trematoda) were collected from freshly slaughtered cattle at local abattoirs and identified by experts. After cleaning, parasites were stored in 0.9% phosphate-buffered saline (PBS) of pH 7.54 prepared with 8.01 g NaCl, 0.20 g KCl, 1.78 g Na₂HPO₄ and 0.27 g KH₂PO₄ in 1 liter of distilled water at 37±1°C.

Plant material

The plant leaves of *Gynura procumbens* were collected from Mirpur-12 (DOHS) and leaves were separated from the undesirable materials and dried at room temperature for two weeks. The plant leaves were ground into a coarse powder with the help of suitable grinder. About 350 gm of powder material was taken in a clean and dried glass beaker and soaked in 600 ml of ethanol. The container containing the contents was sealed and stored for 10 days.

Preparation of sample

To prepare the suspension of ethanolic extract of *G. Procumbens* the concentrations of 25, 50 and 100 mg/ml; .25, .5 and 1g of extract were taken and triturated with Tween 80 as a suspending agent and final volume was made to 50 ml for respective concentration with PBS. For the preparation of standard albendazole at concentrations of 15 mg/ml; 150 mg of albendazole powder were taken and triturated with Tween 80 as a suspending agent and final volume was made to 10 ml for respective concentration with PBS. [10][11]

The anthelmintic activity of *Gynura procumbens* was evaluated according to the method of Kratika et al 2010 [12 – 15].

The animals were divided into five groups containing six earthworms. 10ml of control (Distilled water), standard (Albendazole) and extract (*Gynura procumbens*) of each concentration were taken in different petri dishes. Experimental six animals parasites of both types were taken in each different petri dishes. The concentration of standard and extract were, Standard Albendazole 15mg/ml, Plant's extract 25mg/ml, plant's extract 50mg/ml and plant's extract 100mg/ml. I recorded the time of paralysis when motion was not observed unless shaken violently. The death time was recorded after evaluating that the parasites did not move when shaken vigorously, dipped in warm water (50°C) or subjected to external stimuli. Anthelmintic activity is expressed as the time required for paralysis and death of parasites as compared to control [16][17].

Results and Discussion

Gastrointestinal tract infections by helminthic of humans and animals have been recognized to unfavorably affect the healthy lifestyle of large number community with a resultant lowering of resistance to other diseases. In the finding for active compounds with anthelmintic activity, a number of substances have been separated using different species of worms, for example, earthworms, *Ascaris*, *Nippostrongylus* and *Heterakis*. For the evaluation of anthelmintic activity of compounds by in vitro study from above mention all of these species specially earthworms have been used widely because of their physiological resemble compare with intestinal "worms" that are present in our GI tract. It has been demonstrated that all anthelmintics are toxic to earthworms and a substance toxic to earthworms is worthy for investigation as an anthelmintic [18]

Ethanol extract of *Gynura procumbens* leaves was tested for anthelmintic activity on live parasites *P. cervi*. Standard Albendazole drugs were used for comparative study. The above discussion showed that ethanolic extract of the leaves of *Gynura procumbens* has an active compound that shows anthelmintic activity against helminthes. Death time of standard Albendazole is 20.6min (15mg/ml) and

on the other hand, if we increase the concentration of the plant's extract that decrease the death time at 100mg/ml concentration death time is 26 min.

From the above result, it is culminated that the extracts of the plant have potent anthelmintic activity when compared with the conventionally used drugs and is equipotent to standard drug. Further results, using in vivo models are required to carry out and establish the effectiveness and pharmacological rationale for the use of the plant as anthelmintic drug.

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Table1: Anthelmintic activity of *Gynura procumbens* the basis of paralysis time on helminthic.

| Treatments Concentration (mg/ml) | Worm No. | Time taken for paralysis in min. | Mean time taken for paralysis in min. | SD |
|----------------------------------|----------|----------------------------------|---------------------------------------|-------|
| Control in water | C1 | ----- | ----- | ----- |
| | C2 | ----- | | |
| | C3 | ----- | | |
| | C4 | ----- | | |
| | C5 | ----- | | |
| Standard Albendazole 15 | S1 | 12 | 11.8 | 1.48 |
| | S2 | 11 | | |
| | S3 | 12 | | |
| | S4 | 10 | | |
| | S5 | 14 | | |
| Plant's extract 25 | E1 | 39 | 36.4 | 1.94 |
| | E2 | 35 | | |
| | E3 | 37 | | |
| | E4 | 34 | | |
| | E5 | 37 | | |
| Plant's extract 50 | E1 | 25 | 22.2 | 1.92 |
| | E2 | 22 | | |
| | E3 | 23 | | |
| | E4 | 20 | | |
| | E5 | 21 | | |
| Plant's extract 100 | E1 | 16 | 16 | .70 |
| | E2 | 17 | | |
| | E3 | 16 | | |
| | E4 | 15 | | |
| | E5 | 16 | | |

Table2: Anthelmintic activity of *Gynura procumbens* the basis of death time on helminthic.

| Treatments Concentration (mg/ml) | Worm No. | Time taken for death in min. | Mean time taken for death in min. | SD |
|--|-------------|------------------------------------|---|-------|
| Control in water | C1 | ----- | ----- | ----- |
| | C2 | ----- | | |
| | C3 | ----- | | |
| | C4 | | | |
| | C5 | | | |
| Standard Albendazole 15 | S1 | 22 | 20.6 | .89 |
| | S2 | 20 | | |
| | S3 | 21 | | |
| | S4 | 20 | | |
| | S5 | 20 | | |
| Plant's extract 25 | E1 | 53 | 52.4 | 1.94 |
| | E2 | 55 | | |
| | E3 | 50 | | |
| | E4 | 53 | | |
| | E5 | 51 | | |
| Plant's extract 50 | E1 | 39 | 41 | 2.30 |
| | E2 | 43 | | |
| | E3 | 44 | | |
| | E4 | 40 | | |
| | E5 | 39 | | |
| Plant's extract 100 | E1 | 27 | 26 | 2.23 |
| | E2 | 26 | | |
| | E3 | 29 | | |
| | E4 | 25 | | |
| | E5 | 23 | | |

Figure: Graphically representation of different doses of extracts and their effect.

