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PHYTOCHEMICAL SCREENING AND ANTHELMINTIC ACTIVITY OF ALCOHOLIC EXTRACT OF FRUITS OF *ELEOCHARIS DULCIS*

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Abstract

Eleocharis dulcis is the scientific name of a plant which are locally known as Chinese water chestnut or water chestnut or pani faul, belongs to family of Cyperaceae . The plant is indigenous to eastern Asia and China, Indonesia and Australia are grown. Usually, it is grown in the tropical, subtropical, and warming areas. This plant is grown in natively to eastern Asia and China, Indonesia and Australia . The eatable part of this plant is the underground corm, which is typically harvested. It was especially used for treating jaundice, abdominal mass, conjunctival congestion, throat swelling and pain, excrescence, bloody diarrhoea, hypertension, chronic nephritis, constipation. For anthelmintic activity, methanol fruit extract of *Eleocharis dulcis* was investigated for their action against *Paramphistomum cervi*. Different doses of each extract were examined in the bioassay, which included assurance of time of paralysis and time of death of the worms. The reference standard albendazole showed paralysis time 14.6 minutes and death time 16.8 minutes. On the other hand, methanol extract showed at dose 150 mg/ml significant effect which has paralysis time 23.0 minutes and death time 29.8 minutes. The potential of these activities may be due to the presence of most of the phytochemicals which supports previous claims and validate its uses as an expected folk medicine.

Keywords: *Eleocharis dulcis*, fruit extract, albendazole, anthelmintic activity.

Introduction

Helminthes infections, over and again entitled helminthiasis are among the most inescapable infection and a preeminent degenerative disorder upsetting an enormous extent of world's population [1]. The helminths parasites mainly subsist in human body in intestinal tract, but they are also found in tissue, as their larvae migrate towards them [2]. Intestinal parasitic infections are one of the fundamental wellbeing issues, particularly in developing countries, and especially in children. Deficient sanitation and poor cleanliness might be the primary purpose for it [3]. The parasitic gastroenteritis is caused by mixed infection with many species of stomach and intestinal worms, which results weaknesses, loss of appetite, decreased feed efficiency, reduced weight gain as well as decreased productivity [4]. Vaccinations may also play a role, as in the case of lungworms. Be that as it may, issues have developed with the utilization of anthelmintics, eminently the improvement of obstruction in helminths to different anthelmintic mixes and classes, also as substance buildup and lethality issues . Furthermore, acknowledgment of the antigenic multifaceted nature of parasites has eased back antibody improvement [5,6]. Several anthelminthics, inclusive of praziquantel and albendazole, are contraindicated for certain agencies of patients, including pregnant or lactating wo. This has led to the boom in hobby of ethno clinical practices across the world for the use of medicinal plant life in remedy of helminthic sicknesses [7]. *Eleocharis dulcis* is the botanical name of a medicinal plant which is locally known as Pani Faul, Singara and Water Chestnut, belongs to the family of Cyperaceae and it's especially eaten as a fruit in their ripen condition [8]. This plant is grown in natively to eastern Asia and China, Indonesia and Australia [9, 10]. The eatable part of this plant is the underground corm, which is typically harvested. The leaves of this plant are the hollow shape and it would be on an average 1.5 feet in height, and appearance like grassy [11]. The fruit is drop-shaped to broadly dropshaped as well as it is a perennial herb with short rhizomes [12]. It was especially used for treating jaundice, abdominal mass, conjunctival congestion, throat swelling and pain, excrescence, bloody diarrhoea, hypertension, chronic nephritis, constipation[13,14].

Therefore, the aim of the present study is to investigate the anthelmintic activity of *Eleocharis dulcis* fruit extract of that might explore the remedial potential of this plant to a great extent.

Methods

Plant Materials

The edible (fruit) part of the plant of *Eleocharis dulcis* was collected from near Jahangirnagar University fields, Dhaka, Bangladesh. The plant was identified and authenticated by Bangladesh National Herbarium.

Drying and Grinding

The collected fruits were separated from undesirable materials. Then these were dried in for one week in the sunlight and these were cut into small pieces. The fruit parts were converted into coarse powder by using a suitable grinder. The powder was kept in a cool, dark and dry place and stored in an airtight container until analysis initiated.

Preparation of Fruit Extract

At first, a clean flat flat-bottomed glass container was taken and added about 400 gm of a powdered sample into the container. Then 1500 ml of 90% methanol added into the container and soaked the powder into the methanol. Afterward, the container was sealed with its contents and kept for a period of 10 days accompanying occasional shaking and stirring. After that, the coarse part of the fruits was separated from the mixture by using white cotton. Then the liquid portion was also filtered three times with the help of white cotton. Then again, it was filtered through Whatman filter paper. Then the filtrate was kept in a Rotary evaporator machine that separates the solvent and the desirable crude extract was obtained.

Experimental worms

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 8.01g NaCl, 0.20 g KCl, 1.78 g

Na_2HPO_4 and 0.27 g KH_2PO_4 in 1 liter of distilled water at 37±1°C.

Phytochemical screening

Different phytochemical groups such as alkaloids, glycosides, Phenols, flavonoids, tannins, gums, saponins, steroids were identified by characteristic colour change using standard chemical tests [15]. Molisch Test and Fehling's test were used for carbohydrate existence. Biurets's Test was used for Proteins detection. Flavonoid Test was used for the detection of flavonoids. Alkaloids were detected using the Dragendroff's, Mayer's and Hager's test. For identification of tannin potassium dichromate test, ferric chloride, and lead acetate tests were followed. Keller- Kiliani tests were performed to identify glycosides. Frothing Test for saponins existence, Sulphuric acid test was performed for the detection of steroid. Molisch test was performed for detecting the amount of existence of gum in the samples.

Anthelmintic Activity

Preparation of sample

To prepare the suspension of methanol fruit extract of *Eleocharis dulcis* the concentrations of 50, 100 and 150mg/ml; respectively. 0.5, 1, 1.5 gm 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 20 mg/ml; 200 mg of albendazole powder were taken and triturated with of Tween 80 as a suspending agent and final volume was made to 10 ml for respective concentration with PBS [16,17]. The anthelmintic activity of gynura procumbens was evaluated according to the method of Kratika et al 2010 [18]. The animals were divided into five groups containing five earthworms. 10ml of control (Distilled water), standard (Albendazole) and extract of each concentration were taken in different petri dishes. Experimental five animal parasites of both types were taken in each different petri dish. The concentration of standard and extract were Standard Albendazole 20 mg/ml, both extracts 50, 100 and 150mg/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 [19].

Results

In the phytochemical screening, the fruit extract revealed the presence of some of the pharmacologically active phytochemicals. After completing a wide range of chemical tests for the identification of major classes of therapeutically important compounds, alkaloid, carbohydrate, Phenols, Glycoside, tannins, flavonoids, Saponin, and terpenoids (Table 1).

The reference standard albendazole showed paralysis time 14.6 minutes and death time 16.8 minutes. On the other hand, methanol extract showed at doses 100 and 150 mg/ml significant effect which has paralysis time 25.6 and 23.0 minutes and death time 44.4 and 29.8 minutes; respectively.

Discussion

Phytochemical analysis of the crude extracts revealed the presence of tannis which were shown to produce anthelmintic activities as well as chemically tannins are polyphenolic compounds [20,21]. Some synthetic phenolic anthelmintics such as niclosamide, oxyclozanide, bithionol etc., are reported to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation [22].

The bio-activities of terpenes on several disorder are still unclear. The mechanisms of action seems to be related to the interaction with the parasite redox cycling system leading to enzyme inhibition as well as parasite killing [23]. Tannin containing plants increase the supply and absorption of digestible protein by animals. This is achieved by formation of protein complexes in the rumen by tannins, which later dissociate at low pH in the abomasum to release more protein for metabolism in the small intestines of ruminant animals. Moreover, tannins or their metabolites have a direct effect on the viability of the preparasitic stages of helminths [24]. Other phytochemicals reported to have an anthelmintic effect include flavonoids and terpenoids [25].

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Table 1. Chemical group tests of *Eleocharis dulcis*

Tested groups	Methanol extract
Glycoside	+
Alkaloid	+
Saponin	+
Tannin	+
Gum	-
Carbohydrate	+
Terpenoids	+
Phenols	+
Flavonoids	+

(+) Indicates presence, (-) Indicates absence.

Table 2: Anthelmintic activity of methanol fruit extract of *Eleocharis dulcis*.

Treatment	Concentration(mg/ml)	*Paramphistomum cervi*	
		Paralysis	Death
Control (Distilled water)	-	-	-
Standard (Albendazole)	20 mg/ml	14.6±1.8	16.8±1.9
Fruit (methanol) extract	50 mg/ml	29.4±2.7	53.2±5.3
	100 mg/ml	25.6±2.7	44.4±5.2
	150mg/ml	23.0±2.7	29.8±3.4

All Values represent Mean± SD; n=5 in each group.

Figure 1. Anthelmintic activity of *Eleocharis dulcis* fruit extract on *Paramphistomum cervi*.