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Evaluation of anthelmintic activity of seeds of *Sesamum indicum* L. and fruits of *Capsicum frutescens* L.

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Abstract
The present study was aimed to evaluate anthelmintic activity of methanolic extract of seeds of sesame (*Sesamum indicum* L.) and fruits of *Capsicum frutescens* L. on aquarium worm *Tubifex tubifex* by using three concentrations *viz.*, 2.5, 5 and 10 mg/ml of each extracts were studied which was mainly concerned with the determination of time of paralysis and time of death of the worms. The gradual increased in a dose exhibited a gradual increase in the activity. The results showed that both the alcoholic extracts exhibited significant anthelmintic activity at highest concentration of 10 mg/ml as compared with levamisole (1 mg/ml) was evaluated as standard reference and distilled water as control.

Keywords: *Sesamum indicum* L., *Capsicum frutescens* L., methanolic extract, anthelmintic activity, *Tubifex tubifex*, levamisole.

1. Introduction
Plant-based foods contain significant amounts of bioactive compounds, which provide desirable health benefits beyond basic nutrition. Epidemiological evidence suggests that consumption of a diet rich in vegetables and fruits has positive implications for human health. The World Health Organization reported that 80% of the world populations rely chiefly on indigenous medicine and that the majority of traditional therapies involve the use of plant extracts or of their active constituents [1] and over 25% of modern medicines that are commonly used worldwide contains compounds extracted from medicinal plants [2]. In Bangladesh there is abundant of medicinal plants and ninety percent of the medicinal plants are wild sourced [3, 4].
Helminthic infestations are now being recognized as a cause of chronic ill health and sluggishness amongst the children. World Health Organization estimated 2 billion people infected with helminthes and it was also estimated that 100% of all age group of school children are at risk of morbidity [5]. The major phyla of helminthes are nematodes (round worms) which are soil transmitted helminths that mostly cause the intestinal infection, filarial worms cause the onchocerciasis and lymphatic filariasis, while platihelminths (flatworms) also known as trematodes like schistosomes and cestodes causes cyticerosis [6]. Current estimates suggest that over half of the world population is infected with intestinal helminths, such as *Ascaris*, hookworms, *Trichuris*, *Enterobius*, *Strongyloides*, and tapeworms, and that most of these infected people live in remote rural areas in the developing countries [6, 7]. In case of other animals also gastrointestinal parasites causes infections that diminish the animal survival, growth rates and reproductive performance [8]. Morbidity from nematodes is common with diabetes and lung cancer. The helminths parasites mainly subsist in human body in intestinal tract, but they are also found in tissue, as their larvae migrate towards them [9]. Chemical control of helminthes coupled with improved management has been the important worm control strategy throughout the world. Side effects of anthelmintic commonly include intestinal gastro-intestinal disturbances nausea and giddiness, while various studies and reviews have showed the resistance to anthelmintic is increasing day to day [10]. Henceforth it is important to look for alternative strategies against gastrointestinal nematodes, which have led to the proposal of screening medicinal plants for their anthelmintic activity.
Sesame (*Sesamum indicum* L.) is an oleaginous seed of the family Pedaliaceae, widely used as a seasoning and in bread products. It is a pharmaceutically important plant specially its seeds

which accumulates a variety secondary metabolites including phenolic compounds, terpenes and steroids for which its used traditionally as herbal medication for many years both for the benefits of whole body and also cosmetic preparation as a free radical scavenger. Its seed is composed of about 55% lipids and 20% protein and also contains vitamins and minerals [11]. Sesame oil is rich in unsaturated fatty acids, to which is attributed its effectiveness in reducing blood cholesterol levels. It is a very rich in lecithin, a phospholipid that acts as a powerful emulsifier, facilitating the dissolution of fat in an aqueous medium [12]. Sesame oil is also widely consumed as a nutritious food, very beneficial to health, as a cooking oil, in pharmaceuticals, in shortening and margarine, as a soap fat and as a synergist for insecticides [13]. Beside seeds the other parts of plant are also useful like wound healing activity [14], analgesic activity [15], flowers (cancer, alopecia, and constipation), roots (antifungal activity) and leaves (infant cholera, diarrhoea, dysentery, and for urinary infections). They serve as a good source of copper, manganese and calcium which are effective in reducing pain, in osteoporosis [16].

Capsicum frutescens (Family: Solanaceae), commonly known as chilli pepper, hot pepper or red pepper, is a perennial plant with shrubs as high as eight (8) feet. It is commonly cultivated and when grown in gardens, it is treated as annul, raising seeds every year. The red colour of the fruit is partly due to its high vitamin A content. Woody plants can accumulate in their cells a great variety of phytochemicals including alkaloids, flavonoids, tannins, saponins, cyanogenic glycosides, phenolic compounds, lignin and lignans. It is widely used as a food flavouring agent, a colouring agent, and an additive in livestock feed and in the food and pharmaceutical industries [17]. It has been noted to stimulate gastric activities, increase circulation and enhances blood flow [18], antioxidative, hypocholesterolemic [19], hypolipidemic, immune-modulatory [20] and anti-mutagenic [21]. Internally, it has been used to treat asthma, pneumonia, toothache, pharyngitis and laryngitis [22].

The present study was undertaken to investigate the anthelmintic activity of methanolic extract of seeds of sesame (*Sesamum indicum* L.) and fruits of *Capsicum frutescens* L.

2. Materials and Methods

2.1 Chemicals

All chemicals used were of analytical reagent grade. Methanol was purchased from Merck, Germany. Normal saline solution was purchased from Beximco Infusion Ltd. Levamisole was purchased from ACI Limited, Bangladesh.

2.2 Plant Materials

Fresh fruits of *Capsicum frutescens* and seeds of *Sesamum indicum* L. for this study were purchased from the local market of Chittagong, Bangladesh and were authenticated by Dr. Sheikh Bokhtear Uddin, Associate Professor, Department of Botany, University of Chittagong, Chittagong-4331, Bangladesh.

2.3 Preparation of Crude Extract

The collected fruits of *Capsicum frutescens* and seeds of *Sesamum indicum* L were dried for a period of 2 weeks under shade and ground. The ground fruits and seeds were soaked in sufficient amount of methanol for one week at room temperature with occasional shaking and stirring. The sediments were filtered and the filtrates were dried at 40 °C in a water bath. The solvent was completely removed by filtering with Whatman number-1 filter paper. The solvent was evaporated under reduced pressure at room temperature to yield semisolid. The extract was then preserved in a refrigerator till further use [23].

2.4 *In-vitro* Anthelmintic Assay

The anthelmintic activity of methanolic extract of seeds of sesame (*Sesamum indicum* L.) and fruits of *Capsicum frutescens* L. were carried out as per the procedure of Ajaiyeoba *et al.* [24] with some minor modifications. The aquarium worm *Tubifex tubifex* were used in the present study because it has anatomical similarity and belongs to the same group of intestinal worm i.e. annelida [25, 26, 27]. The worm were collected from the local market of Chittagong, average size of worms 2-2.5 cm. were taking study. The standard drug levamisole and three different concentrations of methanol extracts (2.5, 5 and 10 mg/ml) in double distilled water [28, 29] were prepared freshly and used for the study of anthelmintic activity. One group was composed of water and it was considered as controlled group. The anthelmintic activity was determine at two different stage ‘time of paralysis’ and ‘time of death’ of the worms. Time for paralysis was noted when no movement of any sort could be observed except when the worms were shaken vigorously. Death was concluded when the worms lost their motility followed with fading away of their body colors [30]. Death was also confirmed by dipping the worms in slightly warm water. The mortality of parasite was assumed to have occurred when all signs of movement had ceased [31].

3. Results

Results of study were recorded as shown in table-1 as in the form of time required to get consecutive attacks of paralysis and at the end time required for complete death of parasite. From the observations made, higher concentration of extract produced paralytic effect much earlier and the time to death was shorter for all worms. From the above study it was seen that the methanolic extract showed dose dependent anthelmintic activity as compared to a standard drug levamisole. In case of *C. frutescens* the mean paralyzing time of *Tubifex tubifex* with the dose of 2.5, 5 and 10 mg/ml were found to be 18.28 ± 0.245, 5.08 ± 0.165 and 3.43 ± 0.281 minutes respectively and in case of *S. indicum* the mean paralyzing time were found 35.38 ± 0.263, 19.56 ± 0.836 and 12.28 ± 0.352 minutes respectively for the same dose. In the meantime levamisole at a dose of 0.5, 0.8 and 1 mg/ml causes paralysis in the above helminth in 14.41 ± 0.643, 6.26 ± 0.261 and 3.30 ± 0.645 minutes respectively. The mean death time of *Tubifex tubifex* with the dose of 2.5, 5 and 10 mg/ml were found to be 37.92 ± 0.213, 21.04 ± 0.743 and 5.89 ± 0.273 minutes respectively for *C. frutescens; and* 57.25 ± 0.211, 36.47 ± 0.712 and 21.68 ± 0.301 for *S. indicum* in the same dose . In the meantime levamisole at a dose of 0.5, 0.8 and 1 mg/ml causes death in the above helminth in 51.32 ± 0.825, 12.21±0.512 and 6.50 ± 0.314 minutes respectively.

Table 1: Anthelmintic activity of methanolic extract of seeds of sesame (*Sesamum indicum* L.) and fruits of *Capsicum frutescens* L.

Test Sample	Concentration Mg/ml	Time taken for paralysis (min)	Time taken for Death (min)
C. frutescens	10	3.43 ± 0.281	5.89 ± 0.273
	5	5.08 ± 0.165	21.04 ± 0.743
	2.5	18.28 ± 0.245	37.92 ± 0.213
S. indicum	10	12.28 ± 0.352	21.68 ± 0.301
	5	19.56 ± 0.836	36.47 ± 0.712
	2.5	35.38 ± 0.263	57.25 ± 0.211
Standard (Levamisole)	1	3.30 ± 0.645	6.50 ± 0.314
	0.8	6.26 ± 0.261	12.21 ± 0.512
	0.5	14.41 ± 0.643	51.32 ± 0.825
Control (water)		--	--

All results are mean ± SEM of three consecutive experiments.

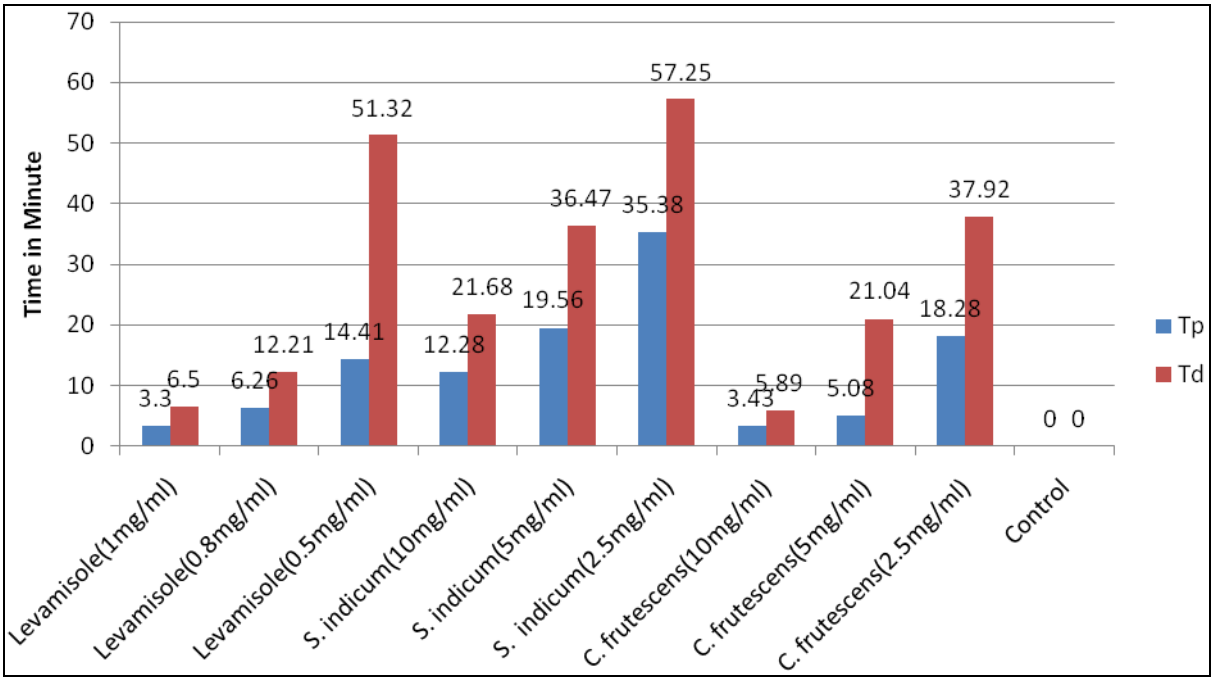

Fig 1: Anthelmintic activity methanolic extract of seeds of sesame (*Sesamum indicum* L.) and fruits of *Capsicum frutescens* L. Where **Tp**- Time for paralysis, **Td**- Time for death.

4. Discussion

Anthelmintics are the drugs that expel out parasitic worms (helminthes) from the body by either causing paralysis or by directly killing them by damaging its cuticle, leading to partial digestion or rejection by immune mechanisms [32]. Levamisole works as a nicotinic acetylcholine receptor agonist that causes continued stimulation of the parasitic worm muscles, leading to paralysis. The literature have been reported that the presence of flavonoids, tannins and polyphenolic compounds show anthelmintic activity, [33] as they can bind to free protein in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and thereby causes death [34]. Some synthetic phenol anthelmintics e.g. niclosamide, oxyclozanide and bithionol are shown effects to interfere with energy generation in antihelminth parasites by uncoupling oxidative phosphorylation and phosphorylation [35].

Finally study concludes that the plant under study has found to possess significant anthelmintic activity in dose dependent manner. The plant might have potential to be developed as useful economic and safe anthelmintic alternative, but it demands more thorough study to find out the exact chemical responsible for anthelmintic activity of plant so as to isolate and extract it separately so as to improve the potency.

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