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ANTHELMINTIC ACTIVITY OF ETHANOLIC BUD EXTRACT OF SYZYGIUM AROMATICUM AGAINST PHERETIMA POSTHUMA

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Abstract:-The aim of the present study was to investigate anthelmintic potential of ethanolic bud extract of *Syzygium aromaticum* by using *Pheritma posthuma* as test worms. Three concentrations (2.5, 5 and 10 mg/ml) of etahnolic bud extracts of *S.aromaticum* were tested in the assay. Albendazole (20 mg/ml) was used as a standard reference and distilled water as a control. The time of paralysis and the the time of death of worms were considered as the parameters to asses the anthelmintic action. It was noticed in the investigation that the time of paralysis and the death of worms was dose dependent and was much earlier in all three test concentrations of ethanolic bud extract of *S. aromaticum* as compared to Albendazole (Standard). From the result it is concluded that the ethanolic bud extract of *S. aromaticum* is much potent anthelmintic natural source which might be helpful against the helminthes infection.

Keywords: Syzygium aromaticum, anthelmintic activity, Pheritima posthuma.

INTRODUCTION:

Medicinal plants and derived medicine are widely used in traditional cultures all over the world and they are becoming increasingly popular in modern society as natural alternatives to syntheitic chemicals (Ben et al., 2009). In the last few decades there has been an exponential growth in the field of herbal medicine. It is getting popularized in developing and developed countries owing to its natural origin and lesser side effects (Patel et al., 2001). Anthelmintics are the drugs that either kill (Vermicide) or expel (Vermifuge) infesting helminthiasis. Helminthiasis is prevalent globally and almost 1/3rd of worlds population harbours them but it is more common in developing countries with poorer personal and environmental hygiene. They harm the host by depriving him of food causing blood loss, injury to organ, intestinal or lymphatic obstruction and by secreting toxins (Tripathi, 2003 and Mahadik, 1998). The palnts are known to provide a rich source of botanical anthelmintics (Satyavati, 1926, Lewis, 1977). A number of medicinal plants have been used to treat parasitic infections in man and animals (Nadkarni, 1954; Chopra, 1956; Said, 1969; Akthar, 2000 and Iqbal, 2004). The *S. aromaticum* traditionally used for antibacterial (Pandey and Singh, 2011)), anti-inflammatory (Kadam, et al., 2009) , antioxidant (Shyamala et al., 2003), antiplatelet (Shrivastava et al., 1993), antifungal (Pinto et al., 2009), antiviral (Banerjee et al., 2005) purpose.

MATERIALS AND METHODS

Experimental Animals

The earthworms were collected from their natural habitat. They were brought in the Department of Zoology and identified based on the available literature as *Pheretima posthuma*. Then they were cultured in laboratory. The adult earthworms of 5-7 cm length were used for the present experimentation.

Plant Material

Syzygium aromaticum bud extract is used as an anthelmintic drug. It is commonly called as the clove tree that belongs to the plant family Myrtaceae. This aromatic tree grows to height of 25 to 40 feet and has opposite oval leaves. Its flowers grow in bunches at the end of the branches. The flowers are red and white when developed, but are usually harvested before they

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open. They are dried in sun and the yellowish coloured buds are changed into dark brown. The dried buds, which are about $\frac{1}{2}$ inch long, are used in kitchen as spice. The dried flower buds of the tree which are pungent and aromatic are used for therapeutic purpose. Cloves are harvested when 1.5-2 cm long and consist of long calyx, terminating in four spreading sepals and four unopened petals which form a small ball in the center.

The dried buds of S. aromaticum were marketed and used for the preparation of ethanolic extract.

Preparation of Ethanolic Extract

Plant extract was prepared by the method of Alade and Irobi (1993) with minor modifications suggested by Ahmad and Beg (2001). The buds were grinded to fine powder with the help of mixer grinder and obtained the fine powder by using the muslin cloth as sieve. Then these powdered materials were used for preparation of ethanolic extracts by using 25g powder mashed in 100 ml absolute ethanol for 72 hours. The mixture was stirred every 24 hour using a sterile glass rod. At the end of extraction, the extract was filtered and the filtrate was evaporated at 500C and paste form extract was stored at 40C until further use.

Sample and standard preparation

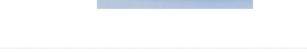
1.Sample preparation: Three samples (2.5, 5 and 10 mg/ml) of ethanolic bud extract of S.aromaticum were prepared and used as test concentrations.

2.Standard used for the activity: Albendazole solution having strength of 20 mg/ml was prepared and used as standared.

EVALUATION OF ANTHELMINTIC ACTIVITY

The assay was performed invitro using adult earthworm (*P. posthuma*) owing to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings for preliminary evaluation of anthelmintic activity of ethanolic bud extract of *S.aromaticum*. Test samples were prepared at concentrations 2.5 mg/ml, 5 mg/ml and 10 mg/ml in distilled water. Three worms of approximately 5-7 cm length were selected for the experimentation. They were first washed with distilled water and placed in petridish containing 20 ml each of the above three test solutions. Albendazole (20 mg/ml) was used as reference standard and dsitilled water as control. All the test solutions and standard drug solution were prepared freshly before starting the experiments. Observations were made for the time taken for paralysis and the death of worms. The time for paralysis of worms was noted at different time interval when less or no movement was noticed. The time for death of worms was noted when no movement of worms was noticed even when they were shaken vigoursly or showed no response to the any stimuli (Physical or chemical). The results are shown in the observation table No1





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(Plate- I: Fig.1. Dried *Syzygium aromaticum* bud, Fig.2. Earthworm (*P. posthuma*) Fig.3. Ethanolic bud extract of *S. aromaticum*)

RESULTS AND DISCUSSION

| Sr. | Extracts | Concentration | Pheretima posthuma | |
|-----|---------------------------|---------------|--------------------|----------------------------|
| No. | | (mg/ml) | Paralysis (P) | Death (D) |
| 1. | Albendazole (Standard) | 20 | 1185.33 ± 79.02 | 1608.33 ± 78.57 |
| 2. | Distilled water (Control) | | | |
| | Ethanolic buds extract of | 2.5 | 4.267 ± 0.252 | 45 ± 2.00 |
| 3. | S.aromaticum | 5 | 1.433 ± 0.306 | 35 ± 4.35 |
| | | 10 | 1.033 ± 0.551 | $\textbf{2.333} \pm 0.351$ |

Observation Table No.1 Anthelmintic Activity of Ethanolic Bud Extract of S. Aromaticum

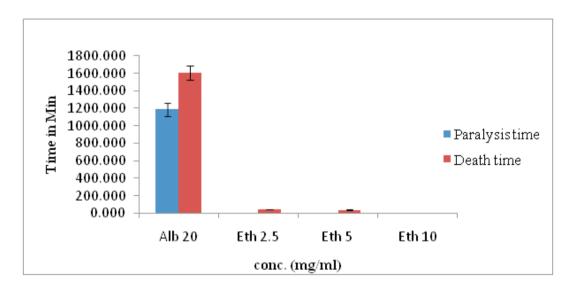


Fig : Histogram showing anthelmintic activity of standard and test extract activity of ethanolic bud extract of *S. Aromaticum*

The results obtained in the present investigation indicated that the ethanolic crude extract of *S.aromaticum* buds produced dose dependent paralysis ranging from loss of motility to loss of response to external stimuli which eventually progressed to death. In case of three crude test samples (2.5, 5 and 10 mg/ml conc.) paralysis was evident at 4.267 ± 0.252 , 2.433 ± 0.306 and 1.033 ± 0.551 minutes while death occurred within 45 ± 2.00 , 35 ± 4.35 and 2.333 ± 0.351 minutes respectively as compared to Albendazole (20 mg/ml conc.) where paralysis was evident at 1185.33 ± 79.02 while death was noticed at 1608.33 ± 78.57 minutes respectively. These finding clearly indicated that though crude samples of ethanolic extract of *S.aromaticum* buds showed significant anthelmintic activity in a dose dependent manner it was more significant at higher concentration which can be evident from haemorrhagic and necrotic spots observed externally on the body of worms exposed

to higher concentration (10 mg/ml) and much earlier paralysis and death as compared to other test samples and the standard.Lokesh et al.(2012) stated that the predominant effect of albendazole on the worm is to cause a flaccid paralysis that result in the worm by peristalsis.Piperazine citrate by increasing chloride ion conductance of worm muscle membrane

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produces hyperpolarisation and relaxation and flaccid paralysis (Martin R.J.(1985). Lakshmi et al.(2012) investigated anthelmintic activity of Carica papaya latex. According to them papaya latex contains many biological active compounds having anthelmintic activity. Though the phytochemical analysis of S.aromaticum buds have not been done in the present investigation it is proposed that it may also contained such an active ingradient having anthelmintic action.

CONCLUSION

In the light of the results obtained in the present investigation and points of discussion it is concluded that the crude ethanolic extract of S.aromaticum buds may have such an active ingradients that may paralyse and kill the earthworms. Hence, it can also be used against helminthes infection in human beings and other animals. Future scope however, involves phytochemical analysis and isolation of phytoconstituents of S. aromaticum responsible for anthelmintic activity.

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