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Evaluation of anthelmintic activity of some members of family Moraceae

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The present investigation was developed to evaluate the anthelmintic activity of some members of family Moraceae. The selected plants were *Ficus bengalensis, Ficus religiosa, Ficus glomerata, Morus indica* and *Morus laevigata*. Anthelmintic activity was carried out by the preparation of methanolic extracts of different parts of these selected plants. For anthelmintic activity, the dried leaves of *F. bengalensis, F. religiosa and F. glomerata,* dried fruits and leaves of *M. indica* and *M. laevigata* were extracted with water and methanol for anthelmintic activity. Anthelmintic studies indicated that the methanolic extracts of *F. bengalensis, F. religiosa, F. glomerata, M. indica* and *M. laevigata* significantly caused paralysis (12.50 ± 0.50, 25 ± 1.00, 19.50 ± 0.50, 15.50 ± 0.50 and 9.50 ± 0.50, respectively), and death (70.50 ± 0.50, 68.5 ± 0.50, 69.00 ± 1.00, 43.50 ± 0.50 and 40.00 ± 1.32, respectively) of worms especially at higher concentration (100 mg/ml) compared with reference drug sample of Albendazole (Paralysis time: 10.60 ± 0.53 and Death time: 36.27 ± 1.42).

Key words: Anthelmintic activity, Moraceae, methanolic extracts, aqueous extracts.

INTRODUCTION

Treatment by herbal medicines has become a part of general health care by the people of different tribes. The use of recent medicines either it is synthetic or semi-synthetic in origin healing field participate in quick result in less time. These traditional medicines, mainly obtained from plants have played a significant role in supporting diseases free human subsistence on this earth (Cotton, 1996).

In ethnomedicine, up to 79% the global inhabitants in growing countries use plant resources as the basis of major health care (Farnsworth et al., 1985). Until now, there is no certification, even though few articles, of conventional veterinary cure in Pakistan on the contrary to other countries where particular concentration has been payed (Anonymous, 1996). The taxonomic healing of the family Moraceae constitutes great taxa of up to 50 genera and almost 1,400 species, counting a number of significant groups (Ventakamaran, 1972). Pakistan has a diverse atmosphere and is affluent in herbal medicines, scattered over a huge area. There are almost 600 plant species known as having medicinal purposes (Shinwari and Khan, 1996).

In Pakistan, plants of medicinal values are chiefly used by Tibbia Dawakhana (herbal medical center of native healers known as hakims). In Ayurvedic scheme of herbal dealing, pharmaceutical industries are being commercially oppressed for the withdrawal of a mixture of ingredients (Mahmood et al., 2003). An elevated percentage of medicinal drugs which are derivative of ordinary substances in western medicine and in traditional medicine of striking society can be a rich basis for progress of new drugs (Hassan et al., 2008).

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Table 1. Data of selected medicinal plants.

Botanical name of plants	Local name	Family	Selected part of plant
F. bengalensis	Banyan	Moraceae	Leaves
F. religiosa	Peeple	Moraceae	Leaves
F. glomerata	Gullar	Moraceae	Leaves
M. indica	Toot	Moraceae	Leaves and fruits
M. laevigata	Shah toot	Moraceae	Leaves and fruits

Table 2. Percentage of extracts/Kg.

Name of plant	Part used	Fresh weight (g)	Dry weight (g)	Moisture content (%)	Extract/kg
F. bengalensis	Leaves	1000	580	42	236
F. religiosa	Leaves	1000	440	46	247
F. glomerata	Leaves	1000	410	59	97
M. indica	Leaves and fruits	1000	390	61	139
M. laevigata	Leaves and fruits	1000	400	60	120

According to the World Health Organization (WHO) “a plant with medicinal value is a plant in which one or more of its organs consists on any material which can be used for healing purposes, and these are semi-synthetically chemo-pharmaceutical” (WHO, 2010). Anthelmintics are drugs or agents that destroy the ejection of parasitic worms of intestine. Helminthes infections are well-known as basis of frequent persistent as well as perceptive ill health among humans and cattles. Almost half of the population of the world suffers from illness, and majority of cattles suffers from infections of worms. Worms are killed with anthelmintic drug. (Singh et al., 2002).

Helminths are the most significant infectious agent that causes serious diseases. In third world countries, these diseases are extremely widespread due to poor management practices chiefly (Dhar et al., 1982). The main objectives of the current studies are:

1. To develop the knowledge of the herbal medicines within natives.
2. To encourage the use of plant as to evade the side effects of synthetically healing medicine.
3. To find out the potential of Family Moraceae for Anthelmintic activity.

METHODOLOGY

Selection and collection of plants

Five plants (*Ficus bengalensis, Ficus religiosa, Ficus glomerata, Morus indica, Morus laevigata*) of the family Moraceae were selected for anthelmintic activities. These plants were selected on the basis of easy growth, easy approach and easy availability and chiefly on the basis of ethnomedicinal importance (Table 1). These plants were collected from different areas (Lahore College for Women University, Lahore and Lawrence Garden Lahore). Plant specimens were deposited as voucher specimen in Prem Madam Herbarium of Lahore College for Women University, Lahore, for record and further reference.

Preparation of plant material

The plant material was washed with water and dried in shade, then cut into small pieces and grind in an electrical grinder to be obtained in a powder form. Powdered plant material was placed in a thimble of Soxhlet apparatus and extracted with methanol at 50°C. (Table 2).

Preparation of stock solution and dilutions

Stock solution was prepared from crude methanolic extracts of plants and stock solution was further diluted into different dilutions according to the need of the activity either in milligram or microgram. Four different concentrations (25, 50, 75, 100 mg/ml) from crude methanolic plant extract were prepared in distilled water. 10 ml of these dilutions were taken in different Petri dishes and earthworms were placed in these Petri dishes.

Anthelmintic activity

Selection of animals

The assay was carried out on earthworms *Pheretima posthuma*. Because of easy availability and its resemblance with intestinal roundworm of human beings, earthworms have been widely used for anthelmintic activity (Ajaiyeoba et al., 2001).

Collection and identification/authentication of worms

Earthworms were collected from the moist soil of Wapda Town, Lahore and from Lawrence Garden Lahore. Earthworms were

Table 3. Anthelmintic activity of *F. bengalensis, F. religiosa* and *F. glomerata*.

Conc. of plant extract (mg/ml)	Mean ± SD	
	Paralysis time (min)	Death time (min)
	F. bengalensis	
25	52.50±0.50	1130.33±0.57
50	39.50±0.50	100.50±0.50
75	24.50±0.50	89.66±0.76
100	12.50±0.50	70.50±0.50
	F. religiosa	
25	72.66±0.76	117.5±0.50
50	59.50±0.50	93.16±1.25
75	47.16±1.25	79.5±0.50
100	25±1.00	68.5±0.50
	F. glomerata	
25	62.83±0.76	121.16±1.60
50	49.50±0.50	99.66±0.76
75	31.50±0.50	82.83±0.76
100	19.50±0.50	69.00±1.00

washed with water to remove soil and other dirt particles. Earthworms were 9 to 10 cm in length and 0.2 to 0.3 cm in width. The collected worms were identified for its authentication from the Department of Zoology, Lahore College for Women University, Lahore.

Experimental plan

All earthworms were divided into 7 groups (A, B, C, D, E, F and G) and groups (C, D, E, F and G) further divided into subgroups.

Group A: Earthworms of group A were placed in distilled water and it served as controlGroup B: Earthworms of group B were placed in Zental (Albendazole) suspension at the dose of 400 mg/10 ml and it served as standard reference drug.
Group C: Earthworms of group C were placed in different concentrations (C_1 = 25, C_2 = 50, C_3 = 75 and C_4 = 100 mg/ml) of methanolic extract of *F. bengalensis* according dosage. This group was further divided into subgroups.
Group D: Earthworms of group D were placed in different concentrations (D_1 = 25, D_2 = 50, D_3 = 75 and D_4 = 100 mg/ml) of methanolic extract of *F. religiosa*. This group was further divided into subgroups.
Group E: Earthworms of group E were placed in different concentrations (E_1 = 25, E_2=50, E_3=75 and E_4=100 mg/ml) of methanolic extract of *F. glomerata*. This group was further divided into subgroups.
Group F: Earthworms of group F were placed in different concentrations (F_1 = 25, F_2 = 50, F_3 = 75 and F_4 = 100 mg/ml) of methanolic extract of *M. indica*. This group was further divided into subgroups.
Group G: Earthworms of group G placed in different concentrations (G_1 = 25, G_2 = 50, G_3 = 75 and G_4 = 100 mg/ml) of methanolic extract of *M. laevigata*. This group was further divided into subgroups.

Observations were made by taking the time of paralysis and time of death. Time for paralysis was noted when no movement of any type can be observed. Time of death was noted when the earthworms lost their movement by changing their body color.

Statistical analysis

Mean and standard deviation (SD) of the data was determined with the help of statistical package for social sciences (SPSS) software.

RESULTS AND DISCUSSION

The mean paralysis time of *P. posthuma* with the extract of *F. bengalensis* at the dose of 25, 50, 75 and 100 mg/ml were found to be 52.50 ± 0.50, 39.50 ± 0.50, 24.50 ± 0.50 and 12.50 ± 0.50 min, respectively. The mean death time of *P. posthuma* at the dose of 25, 50, 75 and 100 mg/ml were found to be 1130.33 ± 0.57, 100.50 ± 0.50, 89.66 ± 0.76 and 70.50 ± 0.50 min, respectively (Table 3). The mean paralysis time of *P. posthuma* with the extract of *F. religiosa* at the dose 25, 50, 75, and 100 mg/ml were found to be 72.66 ± 0.76, 59.50 ± 0.50, 47.16 ± 1.25 and 25 ± 1.00 min, respectively. The mean death time of *P. posthuma* at the does 25, 50, 75, and 100 mg/ml were found to be 117.5 ± 0.50, 93.16 ± 1.25, 79.5 ± 0.50 and 68.5 ± 0.50, respectively (Table 3). *F. glomerata* showed paralysis time with the dose of 100 mg/ml, 19.50 ± 0.50 min while death time on this dose concentration was observed within 69.00 ± 1.00 min (Table 3). *M. indica* had the paralysis time in dose concentrations of 25, 50, 75 and 100 mg/ml 63.33 ± 1.25, 46.33 ± 1.04, 31.16 ± 1.25 and 15.50 ± 0.50, and death time at dose concentrations of 25, 50, 75 and 100 mg/ml were observed to be 91.66 ± 0.76, 72.33 ± 1.52, 60.16 ± 1.25 and 43.50 ± 0.50, respectively (Table 4). *M. laevigata* has shorter time for paralysis (9 to 29 min) of worms and death time for worms was (40 to 50 min), and mean of paralysis time at the dose of 100 mg/ml was

Table 4. Anthelmintic activity of *Morus indica* and *Morus laevigata.*

Conc. of plant extract (mg/ml)	Mean ± SD	
	Paralysis time (min)	Death time (min)
M. indica		
25	63.33±1.25	91.66±0.76
50	46.33±1.04	72.33±1.52
75	31.16±1.25	60.16±1.25
100	15.50±0.50	43.50±0.50
M. laevigata		
25	50.33±1.25	81.83±1.04
50	37.00±1.00	69.33±1.04
75	21.66±0.76	52.50±0.50
100	9.50±0.50	40.00±1.32

Table 5. Anthelmintic activity of control group.

Conc. of dose	Mean ± SD	
	Paralysis time (min)	Death time (min)
Albendazole 100 mg/ml	10.60±0.53	36.27±0.42
Distilled Water 100 mg/ml	-	-

9.50 ± 0.50, and mean death time at this dose was 40.00 ± 1.32 (Table 4).

It was observed that both death time and paralysis time decreased as the concentration was increased. For *F. bengalensis,* lowest paralysis time (70.5 min) and lowest death time (12.5 min) were observed at 100 mg/ml. For *F. religiosa,* minimum paralysis and death times were 68.5 and 25 min, respectively at the 100 mg/ml while for the third specie of *Ficus* (*F. glomerata*), the paralysis time was slightly higher than the *F. religiosa* (69 min) but the death time was lowest than other two species (19.5 min). Among Morus species, *M. laevigata* showed minimum times for paralysis (40 min) and death (9.5 min).

Polyphenolic compounds showed anthelmintic activity. Chemically, tannins and phlobatannins are polyphenolic compounds. It is possible that tannins and phlobatannins in the extracts of plants produce similar effects. Tannins have the ability of binding to free proteins of the gastrointestinal tract of host organism or bind to glycoprotein in the cuticle of the worms which may cause death of the worms (Smith, 1962) (Table 5).

Conclusion

It was an important study to check the paralysis and death times of some members of family Moraceae. Results showed that *M. laevigata* required minimum time to paralyze and kill the worms at 100 mg/ml concentration.

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