

**COMPARATIVE STUDY OF ANTHELMINTIC ACTIVITY BETWEEN ACETONE AND ETHANOLIC STEM BARK EXTRACTS OF *SPONDIAS PINNATA* (LINN.F) KURZ**Panda B.K<sup>1\*</sup>, Patro V.J<sup>2</sup>, Mishra U.S<sup>3</sup>, Panigrahi B.K<sup>1</sup><sup>1</sup>Seemanta institute of pharmaceutical sciences, Jharpokharia, Orissa, India<sup>2</sup>Roland institute of pharmaceutical science, Berhampur, Orissa, India<sup>3</sup>Royal college of pharmacy and health sciences, Berhampur, Orissa, India

Received on: 03/06/2011 Revised on: 12/07/2011 Accepted on: 03/08/2011

**ABSTRACT**

*Spondias Pinnata* (Linn.F) Kurz is found in tribal area of Mayurbhanj district and extensively used traditionally by the tribal people as Anthelmintic, Anti-inflammatory, Regulate menstruation, Anti-pyretic, Anti-tumor and Anti-bacterial activity<sup>1-6</sup>. The present study is attempted to explore the anthelmintic activity of acetone and ethanol extract of bark of plant *Spondias Pinnata* in a comparative study. The various doses of acetone and ethanol extracts were evaluated for their anthelmintic activities on adult Indian earthworms, *Pheretima postuma*. All extracts of both the solvents were able to show anthelmintic activity at (10, 25, 50 and 100 mg/ml) concentration. The activities are well comparable with standard drugs, Piperazine citrate and Albendazole (10 mg/ml). All the doses of acetone and ethanol extract of *Spondias Pinnata* showed better anthelmintic activities than the standard drugs. When the dose of extract is increased, a gradual increase in anthelmintic activity was observed. Ethanol extract showed better anthelmintic activity in comparison to the acetone extract of *Spondias Pinnata*. The data was verified as statistically significance by using ANOVA at 5% level of significance ( $P < 0.05$ ).

**KEY WORDS:** *Spondias Pinnata*, Anthelmintic activity, Ethanol extract, Acetone extract, *Pheretima postuma*.

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**INTRODUCTION**

Helminthes infections are among the most common infections in man, affecting a large proportion of the world's population. In developing countries they pose a large threat to public health and contribute to prevalence of mal nutrition, anemia, eosinophilia and pneumonia. Although the majority of infections due to worms are generally limited to tropical regions, they can occur to travelers who have visited those areas and some of them can develop in temperate climate<sup>7</sup>. Parasitic diseases cause severe morbidity, including lymphatic filariasis (a cause of elephantiasis) onchocerciasis (river blind ness), and schistosomiasis. These infections can affect most population in endemic areas with major economic and social consequence<sup>8</sup>.

*Spondias Pinnata* (Linn.F) Kurz belongs to family *Anacardiaceae* and also called Indian hog-plum (English), amara (hindi), amra (Bengali), amrataka (Sanskrit), ambalam (Tamil), avimamadi (Telgu), ambula (Oriya). It is a glabrous tree 9-10.5 m. high;

trunk straight; bark smooth, ash-colored; branches nearly horizontal. Leaves 30-45 cm long, the common petioles slender, terete, smooth, striate; leaflets 3-5 pairs and a terminal one 7.5-18 by 3.8-7.5 cm. oblong or elliptic-oblong, acuminate, quite entire, more or less oblique; main nerves numerous, horizontal, straight, joined by strong intra-marginal one; petiolules 5-6 mm long. Flowers 1 or 2 sexual, sessile, numerous, pinkish-green, in sparingly branched glabrous terminal panicles 25-38 cm long. Calyx-teeth minute, triangular petals 2.5-3 mm long, ovateoblong, acute. Disk 10-crenate. Stamen-10, about half as long as the petals. Drupes ovoid, yellow, about 3.8 cm long; stone woody, hard, rough with irregular furrows and cavities, fibrous outside. Seed usually 1, more rarely 2 or 3<sup>9</sup>.

The plant is reported to contain  $\beta$ -Amyrin and oleanolic acid, glycine, cystine, serine, alanine and leucine in fruits. Lignoceric acid,  $\beta$ -sitosterol and its glucoside in aerial parts<sup>10</sup>.

The literature survey reveals that various parts of *Spondias Pinnata* (Linn.F) Kurz have been used as folklore medicine for curing various ailments like dysentery and diarrhea, rheumatism, vomiting (bark); regulating menstruation (roots); anti-tubercular (plant); flavoring agent, dysentery (leaves); aphrodisiac (unripe fruits); constipation and antiscorbutic (ripe fruits)<sup>11</sup>.

There are no reports on systematic and scientific study of anthelmintic activity of bark extracts. In the present study, we report the anthelmintic activity of acetone and ethanolic extracts of the barks of *Spondias Pinnata* and their comparative study.

## MATERIALS AND METHODS

### Plant Materials

The bark of *Spondias Pinnata* (Linn.F) Kurz (*Anacardiaceae*) was collected from young matured plants at the rural belt of Mayurbhanj district in the month of sept-2006, and was authenticated by taxonomist of botanical survey of India, Shibpur, Howrah, West Bengal (Letter No. CNH/I-I/(177)/2007/Tech. II/113, dt. 12-07-2007), Kolkata and Voucher Specimen was deposited there. The bark was shade-dried, pulverized in a mechanical grinder and stored in a room temperature in a closed container for further use.

### Preparation of Extract

The powdered plant materials (350g) was repeatedly extracted in a 2000ml round bottomed flask with 1500ml solvents of increasing polarity starting with petroleum ether, Acetone and ethanol. The reflux time for each solvent was 40 cycles. The extracts were cooled at room temperature, filtered and evaporated to dryness under reduced pressure in a rotary evaporator.

### Anthelmintic Bioassay

The earth worm *Pheretima posthuma* (Annelida, *megascolecidae*) was used for evaluating the anthelmintic activity of crude extract using the reference substance for comparison. All earth worms were of approximately equal size (8 cm). They were collected from Horticulture Department, Baripada and are identified at the department of Zoology, M.P.C Autonomous College, Orissa.

### Drugs

The both extract of *Spondias Pinnata* (Linn.F) Kurz was tested in various doses in each group. Normal saline water was used as control. Piperazine citrate and Albendazole were used as standard drugs for comparative study with Acetone and Ethanol extract.

### Experimental Method

The method of Nargund<sup>12</sup> was followed for the screening of anthelmintic activity. Anthelmintic activity was evaluated on adult Indian earth worm, *Pheretima*

*posthuma*. Earth worms were divided in to eleven group (5 each). The first group (I) served as normal control, which received saline water only. The second (II) and third (III) groups received the standard drugs is Piperazine citrate and Albendazole at a dose level of 10mg/ml. Group (IV) to (VII) received doses of acetone extract of 10mg/ml, 25mg/ml, 50mg/ml, 100mg/ml respectively. Group (VIII) to (XI) received doses of ethanolic extracts of 10mg/ml, 25mg/ml, 50mg/ml and 100mg/ml respectively.

Observations were made for the time taken to cause paralysis and death of individual worms for 2 hours. Paralysis was said to occur when the worms do not revive even in normal saline water. Death was concluded when the worms lost their motility followed with fading away of their body colors.

### Statistical Analysis

The data on biological studies were reported as mean  $\pm$  standard deviation ( $n = 5$ ). For determining the statistical significance, standard error mean and analysis of variance (ANOVA) at 5% level significance was employed.  $P < 0.05$  were considered significant<sup>13</sup>.

## RESULTS AND DISCUSSION

Both the extracts of *Spondias pinnata* exhibited anthelmintic activity in dose dependant manner, as shown in the table. The anthelmintic activity of both acetone and ethanolic extracts was comparable with that of standard drugs. The normal saline was used as control. The mortality was noted with 50mg/ml of acetone extract within 82 minutes and as the concentration of this extract is increase to 100mg/ml, the death time has significantly come down to 54 minutes.

Ethanol extract also produced dose dependant paralysis and death at various concentration ranging from 10mg/ml to 100mg/ml. At concentration of 50mg/ml it produced death within 44 minutes and a dose of 100mg/ml the death come down to 31 minutes.

The higher concentration of each crude extract produced paralytic effect much earlier and time to death was shorter.

In this study, we have evaluated the effect of *Spondias pinnata* bark extract on earth worms. Ethanol extract showed significant wormicidal activity. Acetone extract has comparatively less potency than that of ethanolic extract.

Further studies are required to identify the actual chemical constituents that are present in the crude extracts of this plant, which are responsible for anthelmintic activity.

## CONCLUSION

From the above result and discussion it is clear that the extracts of *Spondias pinnata* have good anthelmintic

activity and comparable to the standards. This study provides a scientific support to the traditional uses of the plant in the treatment of worm infections by the traditional healers of this region.

**ACKNOWLEDGEMENT**

The authors are grateful to college authorities for constant support throughout this work.

**REFERENCES**

- Bhandary MJ, Chandrasekhar KR, Kaveriappa KM. Medical ethnobotany of the siddis of Uttara kannada district, Karnataka. J. Ethnopharmacol 1995; 47 Suppl 3: 149- 158.
- Wasawat S. A list of Thai Medicinal Plants. Research Report A.S.R.C.T., No.1 on research project 1967; 17.
- Burkill IH. A Dictionary of the Economic Products of the Malay Peninsula. Ministry of agriculture and co-operatives, Malaysia, 1996.
- Mokkhasmit M, Ngarmwathana W, Sawasdimongkol K, Permpiphat U. Pharmacological evaluation of Thai Medicinal Plants. J. Med Ass 1971; 54 Suppl 7: 490-504.
- Maurakami A, Nakamura Y, Ohigashi H, Koshimizu K. Cancer Chemo preventive Potentials of edible Thai Plants and some of their active Constituents. Mem. Sch. Biol. Oriented Sci Technol Kinki University 1997; 1: 1-23.
- Valsaraj R, Pushpangadan P, Smitt UW, Adersen A, Nyman U. Anti-microbial screening of selected Medicinal Plants from India. J. ethnopharmacol 1997; 58 Suppl 2: 75-83.
- Bundy DA. Immunoepidemiology of intestinal helminthic infection. I: The global burden of Intestinal Nematode Disease. Trans Royal Soc Trop Med Hyg 1994; 8: 259-61.
- Togbato S, Townson S. Anti-parasitic Properties of Medicinal and Other Naturally Occurring Products. Adv Parasitol 2001; 50: 199-295.
- Kirtikar KR, Basu BD. Indian Medicinal Plants. Sri Satguru Publication, Delhi: India; 2000; 03: p.944-948.
- Tandon S, Rastogi RP. Studies on the Chemical Constituents of Spondias Pinnata. Planta- medica. 1976; 29: 190-192
- Warrier PK, Nambier VPK, Ramankutty C. Indian Medicinal Plants, Aryavaidya Sala, Kottakkal. Hyderabad: Orient Longman Limited; 2004. p.186-189.
- Nargund VLG. Anthelmintic activity of 8-Fluoro-9-substitued (1, 3)-Benzothiazolo(5,1-b)-1, 3, 5-triazoles on *Phertima postuma*. Indian Drugs 1999; 36(2): 137-39.
- Bolton S. In Pharmaceutical Statistics-Practical and Clinical Application. New Work: Marcel Dekker; 1997. p. 69-78.
- Khandelwal KR, Pawar AP, Kokate CK and Gokhale SB. Practical Pharmacognosy. Pune: Nirali Prakashan; 2001. p.153.

**Table 1: CHEMICAL TEST FOR EXTRACTS OF SPONDIAS PINNATA<sup>14</sup>**

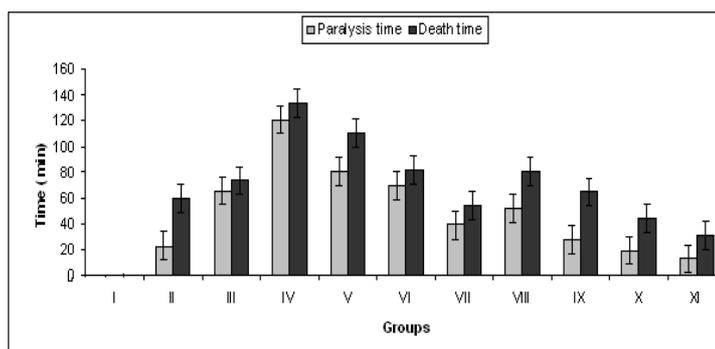
Sr.No.	Chemical test	Extract	
		Acetone	Ethanol
01	Steroids	+	+
02	Glycosides	+	+
03	Saponin	-	+
04	Carbohydrate	-	-
05	Alkaloids	-	-
06	Flavonoids	+	+
07	Tannins	+	-
08	Protein	-	-
09	Gum and Mucilage	-	-
10	Tri-terpenoids	-	+

(+) - Present, (-) - Absent

**Table 2: ANTHELMINTIC ACTIVITY OF ACETONE AND ETHANOL EXTRACTS OF SPONDIAS PINNATA.**

Groups	Treatments	Dose (mg/ml)	Time taken for paralysis (min) (X ± S.D)	Time taken for death (min) (X ± S.D)
I	Control (Normal saline water)	-	-	-
II	Standrd-1 (Piperazine citrate)	10	23.0± 0.2	60.0± 0.5
III	Standrd-2 (Albendazole)	10	65.6± 0.31	73.4± 0.82
IV	Acetone extracts	10	120.05 ±0.52	133.0±0.23
V	Acetone extracts	25	80.23 ±0.74	110.0±0.66
VI	Acetone extracts	50	69.58 ±0.67	82.0± 0.51
VII	Acetone extracts	100	39.28± 0.77	54.0± 0.81
VIII	Ethanol extracts	10	50.2± 0.63	80.36± 0.35
IX	Ethanol extracts	25	28.0± 0.2	65.0± 0.3
X	Ethanol extracts	50	19.0 ±0.2	44.0± 0.5
XI	Ethanol extracts	100	13.0 ±0.2	31.0± 0.2

Each value is represented as mean ± standard deviation (n = 5). Data are found to be significant by testing through one way ANOVA at 5% level of significance (p<0.05).



**Figure 1: Anthelmintic activities of acetone and ethanol stem bark extracts of Spondias pinnata (Linn.f) Kurz on Indian earth worm Phertima postuma. Each bar is represented as mean ± standard deviation (n=5)**

Group I-Control(Normal saline water), group II-standrd-1(Piperazine citrate), group III-standrd-2 (Albendazole), group IV to VII-Acetone extract of dose 10,25,50 and 100 mg/ml respectively and group VIII to XI-Ethanol extract of dose 10,25,50 and 100 mg/ml respectively.

Source of support: Nil, Conflict of interest: None Declared