Anthelmintic activity of *Platycladus orientalis* leaves extract

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Abstract- Ethanol extract from the leaves of *Platycladus orientalis* were investigated for their anthelmintic activity against *Pheretima posthuma*. Three concentrations (1%, 2.5% and 5%) of extract were studied in activity, which involved the determination of time of paralysis and death of the worm. The extract exhibited significant dose dependent anthelmintic activity. Piperazine citrate in same concentration as that of extract was included as standard reference and distilled water as control. The anthelmintic activity of ethanol extract of *Platycladus orientalis* has therefore been demonstrated for the first time.

Keywords: Anthelmintic Activity, *Platycladus Orientalis*, *Pheretima posthuma*.

Introduction

Helminth infections are among the most widespread infections in humans, distressing a huge population of the world. Although the majority of infections due to helminths are generally restricted to tropical regions and cause enormous hazard to health and contribute to the prevalence of undernourishment, anaemia, eosinophilia and pneumonia. Parasitic diseases cause ruthless morbidity affecting principally population in endemic areas. The gastro-intestinal helminthes becomes resistant to currently available anthelmintic drugs therefore there is a foremost problem in treatment of helminthes diseases. Hence there is an increasing demand towards natural anthelmintics [1, 2, 3]. *Platycladus* is a distinct genus of evergreen coniferous tree in the cypress family Cupressaceae, containing only one species, *Platycladus orientalis*, also known as Chinese Arborvitae or Biota. It is native to northwestern China and widely naturalized elsewhere in Asia east to Korea and Japan, south to northern India, and west to northern Iran. It is a small, slow-growing tree, to 15-20 m tall and 0.5 m trunk diameter (exceptionally to 30 m tall and 2 m diameter in very old trees). The foliage forms in flat sprays with scale-like leaves 2-4 mm long. The cones are 15-25 mm long, green ripening brown in about 8 months from pollination, and have 6-12 thick scales arranged in opposite pairs. The seeds are 4-6 mm long, with no wing. The different parts of the plant are traditionally used as a diuretic, anticancer, anticonvulsant, stomachic, antipyretic, analgesic and anthelmintic [4, 5]. The plant has not been explored for its anthelmintic activity so far. The present study was therefore aimed at investigating the anthelmintic activity of the leaves extracts with a view to justifying the use of the plant in the treatment of helminthes.

Materials and Methods

Collection and preparation of Plant Extract

The leaves of *Platycladus orientalis* were collected in the month of February from the local field of Etawah, Uttar Pradesh state, India, and authenticated by Dr. Harish K. Sharma, Ayurvedic Medical College, Davangere, Karnataka, India. A voucher specimen was submitted at Institute's herbarium department for future reference (AN 102). Dried leaves were collected from local moist place, washed and approximately equal size (15 cm). They were kept in water.

**Phytochemical screening**

Qualitative assay, for the presence of plant phytocconstituents such as carbohydrates, alkaloids, glycosides, flavonoids, tannins and saponins were carried out on the powdered leaves following standard procedure [6, 7].

**Animal**

Healthy adult Indian earthworms, *Pheretima posthuma*, due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings [8, 9, 10] were used in the present study. All earthworms were of approximately equal size (15 cm). They were collected from local moist place, washed and kept in water.

**Drugs**

Piperazine citrate was purchased from GSK Pvt.Ltd. The solvents and other chemicals of analytical grade were used during experimental protocol.

**Anthelmintic Activity**

Alcohol extract from the Leaves of *Platycladus orientalis* were investigated for their anthelmintic activity against *Pheretima posthuma*. Various concentrations (1%, 2.5%, and 5%) of extract were tested in the bioassay, which involved determination of time of paralysis and time of death of the worms. Piperazine citrate was included as standard reference and distilled water as control. The anthelmintic assay was carried as per the method of Ajaiyeoba et al. with minor modifications. Deore S. L.et al 2009[11]. The assay was performed on adult Indian earthworm, *Pheretima posthuma*, collected from...
moist soil and washed with normal saline to remove all faecal matter were used for the anthelmintic study. The earthworms of 3-5 cm in length and 0.1-0.2 cm in width were used for all the experimental protocol. In the first set of experiment, six groups of six earthworms were released in to 10 ml of solutions of Piperazine citrate, ethanol extracts of Leaf of Platycladus orientalis (1%, 2.5% and 5% each) in distilled water. Observations were made for the time taken to paralysis and death of individual 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.

Results and Discussion
The leaves extracts of the Platycladus orientalis displayed a significant anthelmintic activity (p<0.05) in dose dependent manner as shown in Table 1. The predominant effect of Piperazine citrate on the worm is to cause a flaccid paralysis that result in expulsion of the worm by peristalsis. Piperazine citrate by increasing chloride ion conductance of worm muscle membrane produces hyperpolarisation and reduced excitability that leads to muscle relaxation and flaccid paralysis. The extracts demonstrated paralysis as well as death of worms at a time comparable to Piperazine citrate especially at higher concentration of 5%. Phytochemical screening of the crude extracts revealed the presence of flavonoids and polyphenolic compound as one of the major chemical constituents. Polyphenolic compounds shown anthelmintic activity; chemically tannins are polyphenolic compounds [12]. Some synthetic phenolic anthelmintics e.g., niclosamide, oxyclozanide and bithionol are shown to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation[13] . It is possible that tannins contained in the extracts of Platycladus orientalis produced similar effects. Another possible anthelmintic effect of tannins is that they can bind to free protein in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and cause death [14].

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References
[1] Bundy D. A. (1994) Trans Royal Soc Trop Med Hyg, 8: 259-261.
[2] Tagbota S., Townson S. (2001) Adv Parasitol, 50:199-205.
[3] Sondhi S.M., Shahu R., Magan Archana. (1994) Indian Drugs, 31(7): 317-320.
[4] Kirtikar K.R. and Basu B.D. (1999) Indian Medicinal Plants, Allahabad, Vol.-II, 856.
[5] Asima C., Satyesh C.P. (1995) the Treatise on Indian Medicinal Plant. Editors, Publication and Information Directorate, New Delhi, Vol.6, 149.
[6] Kokate C.K. (1999) Practical Pharmacognosy, 4th edn, Vallabha Prakashan, New Delhi, 149-156.
[7] Khandewal K.R. (2000) Practical Pharmacognosy Technique and Experiments, 2nd edn, Nirali Prakashan, Pune, 149-56.
[8] Vidyarthi R.D. (1977) A Textbook of Zoology, 14th edn, Chand and Co. Press,New Delhi,329-31.
[9] Thorn G.W. (1977) Harrison’s Principles of Internal Medicine, Mc Grew Hill, New York, 1088-90.
[10] Vigar Z. (1984) Atlas of Medical Parasitology, 2nd edn.,Publishing House,Singapore,216-18.
[11] Ajaiyeoba E.O., Onocha P.A., Olarenwaju O.T. (2001) Pharm Biol, 39: 217-20.
[12] Bate-Smith E.C. (1962) J Soc Bot, 58: 95-103.
[13] Martin R.J. (1997) Vet. J., 154:11-34.
[14] Mali R.G., Wadkar R.R. (2008) Indian J Pharm.Sci, 70: 131-133.
### Table 1 - Anthelmintic effect of the leaves extract of Platycladus orientalis

| Group                  | Concentration of extract in % | Time taken in Minutes ± SEM | Paralysis | Death       |
|------------------------|-------------------------------|----------------------------|-----------|-------------|
|                        |                               |                            |           |             |
|                        | 1.0                           | 13.00±0.33                 | 35.00±1.45|             |
|                        | 2.5                           | 11.00±1.52                 | 33.00±1.41|             |
|                        | 5.0                           | 10.00±0.88                 | 31.00±1.20|             |
|                        | 1.0                           | 19.00±1.73                 | 38.00±2.73|             |
|                        | 2.5                           | 14.00±1.24                 | 34.00±1.71|             |
|                        | 5.0                           | 8.00±1.20                  | 32.00±1.26|             |
|                        | --                            | --                         | --        | --          |

**Piperazine citrate**

**Ethanol extract**

**Control**