

NOTE

Anthelmintic Activity of Stems of *Opuntia vulgaris* Mill.

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The anthelmintic activity of aqueous extract of dried stems and the isolated saponin from fresh stems of *Opuntia vulgaris* were evaluated separately on adult Indian earthworm (*Pheretima posthuma*) and the activities were compared with standard drug piperazine citrate. Both aqueous extract and saponin exhibited dose dependent action on inhibition of spontaneous motility (paralysis) and death of earthworms. Saponin showed remarkably higher anthelmintic activity than the standard drug piperazine citrate and the aqueous extract.

Key Words: Anthelmintic activity, *Opuntia vulgaris* Mill, Piperazine citrate, Saponin.

Helminthic infections are now being recognized as a cause of much chronic ill health and sluggishness amongst the tropical people. More than half of the population in the world suffers from worm infection of one or the other. Helminths also affect the domestic animals and livestock causing considerable economic losses. Traditional system of medicine reports the efficacy of several natural products eliminating helminths. Keeping this in view, the present communication deals with the evaluation of anthelmintic activity of stems of *Opuntia vulgaris* Mill.

Opuntia vulgaris Mill. (*Sappattukkalli* in Tamil, *Nagophenia* in Oriya, Family: Cactaceae) is a large succulent shrub distributed throughout India, but is more common in Northern India¹. It is indigenous to Brazil and Argentina. The plant is bitter, laxative, stomachic, carminative, antipyretic; cures biliousness, tumours, anemia, ulcers and enlargement of the spleen. The stems are made into emollient cataplasms. The juice of the plant is used in the treatment of syphilis in Ayurveda^{2,3}. The aqueous extract of *O. vulgaris* stem on preliminary chemical analysis is found to contain saponin and alkaloid^{4,5}. In the present study, we have evaluated the anthelmintic activity of aqueous extract of dried stems and isolated saponin from fresh stems of *O. vulgaris* Mill.

The fresh stems of *O. vulgaris* were collected during the month of September locally (Mayurbhanj district of Orissa) and were authenticated by Dr. H.J. Chowdhury, Joint Director, Central National Herbarium, Botanical Survey of India,

Howrah, West Bengal. A voucher specimen has been preserved in our laboratory for further references (DM1). After collection, the stems were washed properly and fungal infected parts were cut out.

Preparation of aqueous extract: Shade-dried, powdered, sieved (40 mesh size) stems were exhaustively soxhlet extracted with distilled water. The aqueous extract was then evaporated to complete dryness. The yield of aqueous extract was 10.2% w/w.

The aqueous extract gave positive qualitative tests for the presence of saponin and alkaloid.

Isolation of saponin: Fresh stems were ground in a mixture grinder and stored overnight in a refrigerator for the deposition of some pigments, dispersed and other materials. The supernatant was taken and sufficient amount of activated charcoal was added and mixed well. It was, then, filtered repeatedly to get a colourless solution, which was taken in a separating flask. To it sufficient amount of petroleum ether was added, shaken thoroughly and a jelly like mass was fractionated. This crude saponin SAP M was collected, dried in air (yield 5.1% w/w with respect to fresh stems) and gave positive tests for the presence of saponin (foam test, Libermann-Burchard reaction, high hemolytic activity)⁶.

This crude saponin on preparative TLC using silica gel G as stationary phase and chloroform : ethanol (1 : 1) as solvent system gave a fraction SAP 1 (m.p. 203–205°C, λ_{\max} 222 nm in water, R_f value 0.89) which showed characteristic IR (Jasco 530 FTIR, in KBr) peaks (cm^{-1}): 3547 $\nu(\text{OH})$, 1690 $\nu(\text{COOH})$, 1610 ν (for aromatic and unsaturation), 1140 $\nu(\text{C—O—C})$, 690 $\nu(\text{—CH})$; 599 cm^{-1} suggests the structural similarities with triterpenoid saponin^{7–9}.

Evaluation of anthelmintic activity: Anthelmintic activity was evaluated for both aqueous extract of dried stems and isolated saponin from fresh stems separately. The activity was tested according to method discussed in detail by Kailashraj and Kurupa¹⁰. *Pheretima posthuma* (earthworm obtained from Horticulture Department) of nearly equal size (8 ± 1 cm) was selected for the present study due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings^{11,12}.

Piperazine citrate, diluted with normal saline to obtain 0.06, 0.20 and 0.03 g %, served as standard and poured into petridishes. The tested compounds were prepared in minimum quantity of distilled water and diluted to 15 mL with normal saline to obtain the final concentration of 0.06, 0.20, 0.03 g % and taken into petridishes. Normal saline served as control for standard. Six earthworms of nearly equal size were placed in each petridish at room temperature. The times taken to complete paralysis and death were recorded. The mean paralysis time and mean lethal time for each sample was recorded (each reading was taken six times). The time taken by the worms to become motionless was noted as paralysis time and to ascertain the death, earthworms were frequently applied with external stimuli, which stimulate and induce movement if alive^{13,14}.

The anthelmintic activity^{15,16} of the title compounds on *P. posthuma* is exhibited in Table-1. The data indicate that saponin isolated from fresh stems of *O. vulgaris* Mill. was found to possess markedly higher anthelmintic activity than the standard (piperazine citrate). The aqueous extract of dried stems was also found to possess comparable activity in a dose dependant manner.

TABLE-1
ANTHELMINTIC ACTIVITY OF STEMS OF *O. VULGARIS* MILL.

S.No.	Compound	Concentration (g %)	Time (min.)	
			For paralysis	Death
1.	Control (normal saline)	0.06	–	–
		0.20	–	–
		0.30	–	–
2.	Piperazine citrate	0.06	96 ± 1.20	130 ± 1.10
		0.20	50 ± 0.95	79 ± 1.00
		0.30	42 ± 0.87	53 ± 0.85
3.	SAP M	0.06	86 ± 1.23	116 ± 1.99
		0.20	41 ± 0.78	69 ± 0.94
		0.30	18 ± 0.82	28 ± 0.65
4.	AE	0.06	–	–
		0.20	68 ± 1.03	171 ± 2.03
		0.30	55 ± 0.93	122 ± 1.88

Results are expressed as mean ± SEM from six observations.

AE = aqueous extract of dried stem. SAP M = Crude saponin

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