

Analgesic, CNS Depressant and Anthelmintic Activity of *Sarcostemma viminalis*

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ABSTRACT

Various extracts of *Sarcostemma viminalis* were screened for central analgesic activity by hot plate test, peripheral analgesic activity by acetic acid-induced writhings, CNS depressant activity by pentobarbitone-induced sleep and locomotor activity testing methods and anthelmintic activity on the earthworms *Pheretima posthuma*. Results showed that petroleum ether and ethyl acetate extracts had good central analgesic activity; ethyl acetate and methanol extracts had good peripheral analgesic activity; chloroform and ethyl acetate extracts had good CNS depressant activity and methanol extract had good anthelmintic activity.

Keywords: *Sarcostemma viminalis*, Analgesic, CNS depressant, Anthelmintic, Photoactometer, *Pheretima posthuma*

Sarcostemma viminalis syn *S. brevistigma* is a leafless trailing or twining shrub with branches, commonly known as 'Ransher or Somyel'. Flowers are small, in sessile with umbels and seeds are comose [1]. Chemical investigation of the plant showed presence of pregnane derivatives like bregenin [2] and pregnane glycosides like brevine and brevinine [3]. Genin G and H [4], triterpenes like β -amyrin and friedelin [5] and some toxic constituents like sarcovimisine [6] were reported from the plant. Medicinally plant is useful as antiallergic [7], anti-inflammatory [8] and tocolytic [9].

Objective of the study was to evaluate analgesic, CNS depressant and anthelmintic potential of the plant.

MATERIALS AND METHODS

Plant Material

Twigs of *S. viminalis* were collected from Ahmednagar district in October 2005 and authenticated at Botanical Survey of India, Pune (Voucher specimen No. GAS1).

Preparation of Extract

Dried and coarsely powdered twigs of *S. viminalis* were subjected to successive solvent extraction in Soxhlet extractor using petroleum ether, chloroform, ethyl acetate and methanol as solvent. All the extracts

were vacuum dried to produce PEE, CLE, EAE and ME respectively.

Animals

Female Swiss Albino mice, weighing between 25-30 g were used for all experimental protocols. Animals were housed at least one week in the laboratory animal room prior to testing. Food and water were given *ad libitum*. All procedures described were reviewed and approved by Institutional Animal Ethical Committee.

Indian adult earthworms (*Pheretima posthuma*) collected from moist soil and washed with normal saline to remove all the 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 due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings [10, 11].

Analgesic Activity

i. Hot Plate Test

Central analgesic activity was evaluated using hot plate method [12]. Mice were divided into six groups of six animals each. The first group served as control and received only vehicle, second group was administered standard drug pentazocine (5 mg/kg, i.p.). The animals of third to sixth group were treated with PEE, CLE, EAE and ME (50 mg/kg, i.p., each) of twigs of *S.*

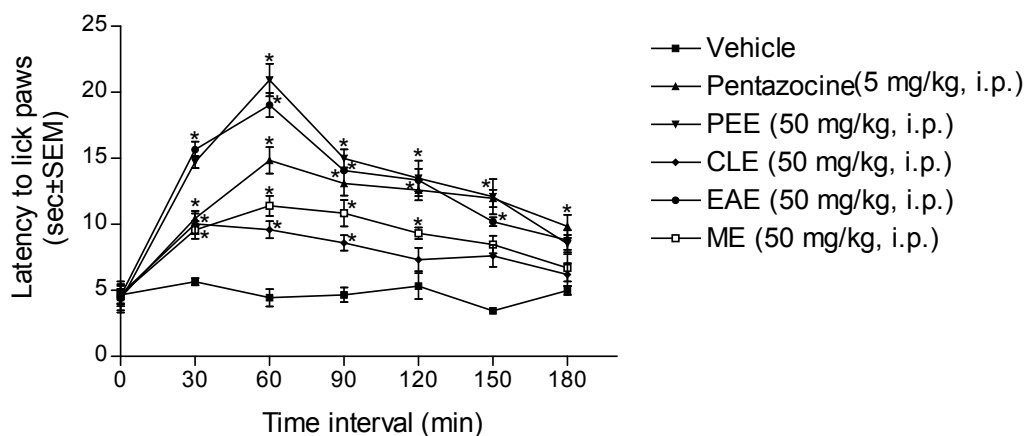


Fig 1. Effect of various extracts of *S. viminale* on thermic stimulus-induced pain in mice (Hot plate test). All the values are expressed as mean \pm SEM; $n=6$, $*p<0.05$ significant compared to control.

viminale, respectively. Mice were placed individually on the hot plate maintained at $55\pm 1^{\circ}\text{C}$ and latency of nociceptive response such as licking, flicking of a hind limb or jumping was noted. The readings were taken at 30, 60, 90, 120, 150 and 180 min after administration of extracts. The experiment was terminated 20 sec after their placement on the hot plate to avoid damage to the paws.

ii. Writhing Test

Peripheral analgesic activity was evaluated using acetic acid-induced writhing test [13]. Mice were divided into six groups of six animals each. The animals received PEE or CLE or EAE or ME (50 mg/kg, i.p., each) or standard drug paracetamol (50 mg/kg, i.p.) or vehicle, 30 min before intraperitoneal injection of 0.1 ml of 0.6% solution of acetic acid. Mice were placed individually into glass beakers after administration of acetic acid and five minutes were allowed to elapse. The mice were then observed for the period of 30 minutes and then number of writhes recorded for each animal.

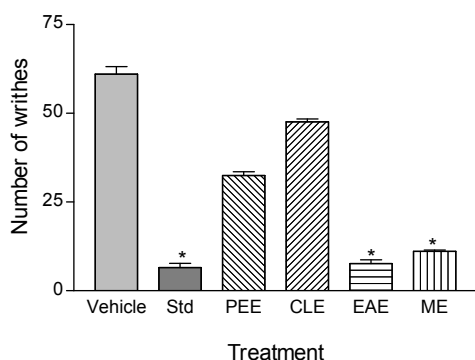


Fig 2. Effect of various extracts of *S. viminale* on acetic acid-induced writhings in mice. All the values are expressed as mean \pm SEM; $n=6$, $*p<0.05$ significant compared to control.

CNS Depressant Activity

i. Locomotor activity testing [14]

Male mice were divided into six groups ($n=6$). First group received vehicle only, second group received diazepam (2 mg/kg, i.p.). Third to sixth groups received PEE, CLE, EAE and ME (50 mg/kg, i.p., each). Mice were placed individually in photoactometer. Basal reaction time was noted before and 30 min after the administration of treatment. A count is recorded when the beam of light falling on the photocell of photoactometer is cut off by mice.

ii Pentobarbitone-induced sleeping time [14]

Male mice were divided into five groups ($n=6$). First group received vehicle only, second to fifth groups received PEE, CLE, EAE and ME (50 mg/kg, i.p., each) 30 min before administration of pentobarbitone sodium (40 mg/kg, i.p.) and duration of sleep was measured. The sleeping time was measured as the duration for which the righting reflex was lost.

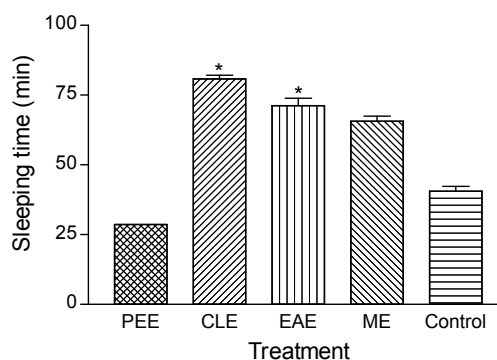


Fig 3. Effect of various extracts of *S. viminale* twigs on pentobarbitone-induced sleep in mice. All the values are expressed as mean \pm SEM; $n=6$, $*p<0.05$ significant compared to control.

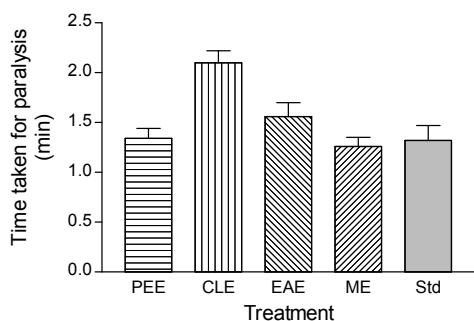


Fig 4. Anthelmintic activity of various extracts of *S. viminale* twigs. All the values are expressed as mean \pm SEM; n=6, control worms were alive up to 24 hrs.

Anthelmintic Activity [15]

Six groups, of six earthworms each were released into 10 ml of desired solution as follows; vehicles (5% DMF in normal saline), albendazole (20 mg/ml) or PEE or CLE or EAE or ME (20 mg/ml, each) of twigs of *S. viminale* in normal saline containing 5% DMF. Observations were made for the time taken to paralysis of individual worm. Paralysis was said to occur when the worms did not revive even in normal saline.

Statistical significance

The results were analyzed for statistical significance using students 't' test. $p < 0.05$ was considered as significant.

RESULTS AND DISCUSSION

All the extracts of *S. viminale* showed significant central analgesic activity in hot plate test at the dose of 50 mg/kg, i.p. (Fig.1). Analgesic activity was comparable with standard drug pentazocine. Among all the extracts, petroleum ether and ethyl acetate extracts showed highest increase in reaction time.

Thermic painful stimuli are known to be selective to centrally active drugs [16]. Among all the extracts petroleum ether and ethyl acetate extracts showed highest increase in reaction time. Prostaglandins and bradykinins were suggested to play an important role in

analgesia [17, 18]. Flavonoids and sterols are reported to inhibit prostaglandin synthesis [19]. A number of flavonoids have been reported to produce analgesic activity [20]. As phytochemical tests showed presence of flavonoids in ethyl acetate extract and sterols in petroleum ether extract, it might suppress the formation of prostaglandins and bradykinins and exert its activity.

Peripheral analgesic activity was assessed by acetic acid-induced writhing test. Ethyl acetate and methanolic extracts showed significant suppression of writhings induced by acetic acid (Fig. 2). It was observed that onset of writhing was delayed and duration of writhing was shortened. Acetic acid is known to trigger the production of noxious substances within the peritoneum, which induces the writhing response [21]. The effect of the extracts against the noxious stimulus may be an indication that it depressed the production of irritants and thereby reduction in number of writhes in the animals.

Results in fig. 3 indicate that the sleeping time induced by pentobarbitone sodium was more prolonged significantly after administration of CLE followed by EAE.

Results in Table 1 revealed that the locomotor activity counts in CLE and EAE treated groups were significantly reduced compared to vehicle group.

Prolongation of sleeping time in pentobarbitone-induced sleeping time test is may be because of enhancement in brain GABA as it is known to have depression action in brain [22,23]. In locomotor activity testing, decrease in rearing along with locomotor activity is observed, that reveals depressive effect on CNS [24].

Methanolic extract showed most potent anthelmintic activity followed by petroleum ether extract and ethyl acetate extract of *S. viminale* twig. So it can be concluded that active principle responsible for anthelmintic activity is a polar compound present in *S. viminale* twig.

In conclusion we can say that petroleum ether and ethyl acetate extracts were having good central analgesic activity and ethyl acetate and methanolic extracts were having good peripheral analgesic activity. Chloroform and ethyl acetate extracts were having good CNS depressant activity and methanolic extract is having good anthelmintic potential.

Table 1. Effect of various extracts of *S. viminale* twigs on locomotor activity of mice.

Treatment (Dose: mg/kg, i.p.)	Number of movements (for 2 min)	
	Before administration of drug	After 30 min of administration of treatment
Vehicle	40.22 \pm 0.23	42.95 \pm 0.36*
Diazepam (2)	47.32 \pm 1.01	20.75 \pm 0.67*
PEE (50)	45.25 \pm 1.65	37.5 \pm 0.87*
CLE (50)	46.75 \pm 0.88	23.5 \pm 2.34*
EAE (50)	46.25 \pm 1.67	23 \pm 2.76*
ME (50)	43.75 \pm 0.76	28.75 \pm 0.99*

All values are expressed as mean \pm SEM; n=6, * $p < 0.01$ significant compared to vehicle.

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