academicJournals

Vol. 7(11), pp. 263-267, December 2015
DOI: 10.5897/JPP2015.0359
Article Number: 7437AEC56729
ISSN 2141-2502

http://www.academicjournals.org/JPP

Journal of Pharmacognosy and Phytotherapy

Full Length Research Paper

Pharmacological studies of methanolic extracts of *Sonchus arvensis* from Kathmandu

Bhupendra Kumar Poudel*, Jagnnath Prasad Sah, Shyam Raj Subedi, Mohan Prasad Amatya, Sadhana Amatya and Tirtha Maiya Shrestha

Institute of Medicine, Tribhuvan University, Nepal.

Received 5 August, 2015; Accepted 17 November, 2015

***Sonchus arvensis*, a perennial sowthistle, is a common but underutilized species of Kathmandu, Nepal. Several uses like sedative, antioxidant and kidney stone eradicating properties have been identified till date, but study on other pharmacological activities is not yet explored. Therefore, this plant was collected from Kathmandu; aerial parts of the plant were dried, crushed, and extracted using a Soxhlet apparatus. The methanolic extract was then concentrated for screening pharmacological effects. While comparing with the standards, the plant was found to possess strong anti-inflammatory activity and inhibitory effect in Gastro Intestinal (GI) motility and locomotor activity in a dose-dependent fashion. The plant, however, did not show skeletal muscle relaxant activity as observed in the traction test and inclined plane test. Thus, it is concluded that the plant possess strong phyto-chemicals having anti-inflammatory activity and inhibitory effect in locomotion and GI motility.**

Key words: Sonchus, anti-inflammatory, motility, locomotion, extraction.

INTRODUCTION

In Nepal, the concept of ethno-medicine has developed since the late 19th century (1885-1901 A.D). The first book "Chandra-Nighantu regarding medical plants was published by the Royal Nepal Academy in 1969. Majority of the population is still dependent on botanical medicines which indicates the importance of herbal medicines in the primary health care (Rajbhandari et al., 2001; Amatya et al., 2009). *Sonchus arvensis*, a vigorous herbaceous perennial plant with milky sap and creeping roots is abundantly available in 1000 to 4100 m in range and grows in sandy, loamy or clayey soils (Holm et al. 1997). In many areas, this sowthistle is considered a noxious weed, as it grows quickly in a wide range of conditions and its wind-borne seeds allow them to spread rapidly (APNI, 2012). Since, this plant is least explored and there are only few pharmacological studies, this study is done to lay a strong foundation for the future development of herbal medicines from this plant's methanolic extract.

MATERIALS AND METHODS

Sample preparation

Samples of the plant were collected from Nagarjun Hill, Kathmandu, Nepal; the aerial parts were dried, crushed, and extracted using a

*Corresponding author. E-mail: poudel.bhupen@gmail.com. Tel: 977-9841380139.

Soxhlet apparatus. Twenty grams of the powdered material was extracted with 200 ml volumes of petroleum ether, diethyl ether, methanol, and water, each in a stepwise manner. The extracts were concentrated and stored in refrigerator. Only methanolic extract was used for pharmacological screening. Male albino mice (30±2.12 g) were used for gastrointestinal motility, spontaneous locomotor activity, and skeletal motility activity tests, whereas male wistar rats (280±3.43 g) were used for anti-inflammatory test.

Gastrointestinal motility: Charcoal meal test

Mice were divided into 4 groups of 3 mice each; the first and second group were administered 500 and 100 mg/kg of methanol extract made in normal saline solution intraperitoneally (i.p), respectively; third group were administered atropine sulphate (5 mg/kg, i.p) and fourth group were administered control vehicle i.p. After 30 min, all animals were fed with 1 ml of charcoal meal (animal charcoal 12 g, tragacanth 2 g, and water 130 ml) intragastrically with the aid of feeding needle. All the mice were killed after 30 min by inhalation of chloroform. The abdomen was opened, the intestine quickly isolated and the small intestine from pylorus to ceacum was cut by scissor and its length was measured by ruler. The intestinal distance moved by the charcoal meal from pylorus was measured to calculate percent of charcoal movement from pylorus to caecum as (Rouf et al., 2003):

$$\%\ \text{of Charcoal movement} = \frac{\text{Distance travelled by the charcoal meal}}{\text{Total length of the small intestine}} \text{X } 100$$

Spontaneous locomotor activity

Mice were divided into 5 groups, each consisting of 3 mice. First group, serving as control, received i.p, the vehicle only and second group received i.p 5 mg/kg of diazepam serving as standard, while the remaining groups were injected i.p with 125, 250 and 500 mg/kg of the methanolic extract i.p. After 30 min of administration of extract, each mouse was placed in an open square field 50×50 cm surrounding and subdivided into 25 squares of 10×10 cm. The percentage inhibition in locomotion was calculated as (Kulkarni 2000):

$$\%\ \text{Inhibition} = 100 - \frac{\text{No. of squares crossed in test}}{\text{No. of squares crossed in control}} \text{ X } 100$$

Skeletal muscle activity: Traction technique

Mice were divided into five groups of five mice in each. Each mouse of first three groups were given methanolic extract i.p at the dose of 125, 250, and 500 mg/kg respectively. One group received only distilled water as a control and another group received diazepam at the dose of 5 mg/kg as a standard. After 30 min, the capability of the mice to touch the wire with at least one of the hind paws within 5 s was observed and percentage failure was calculated by the formula (Chattopadhyay et al., 2003):

$$\%\ \text{Failure} = \frac{\text{No. of mice failed to grasp the wire}}{\text{Total no. of mice}} \text{ X } 100$$

Skeletal muscle activity: Inclined plane test

Mice were divided in 5 groups of 4 mice each. First group was given 125mg/kg, methanol extract, second group 250mg/kg methanol extract, third group500 mg/kg, fourth group diazepam 5 mg/kg and the fourth group i.e. control group was given distilled water only i.p. After 30 min they were placed in the inclined plane 45° for 2 min and their ability to remain at such inclination was noted.

Percentage failure was calculated by the following formula (Chattopadhyay et al., 2003):

$$\%\ \text{failure} = \frac{\text{no of mice failed to remain in the inclined plane}}{\text{total no of mice}} \text{ x } 100$$

Anti-inflammatory activity: Carrageenan test

It was studied in male wistar rats (280±3.43 g) as per the method described by Winter et al. (1962). Rats were divided into 5 groups, each consisting of 3 rats. One group serving as control received i.p the vehicle (normal saline) only and the other group received i.p 100 mg/kg of aspirin and 10 mg/kg diclofenac serving as standard, while the remaining groups were injected i.p with 200 and 400 mg/kg of the methanol extract. One hour later each animal was injected with 0.1 ml of 1% carrageenan suspension in normal saline into the sub-plantar region of right hind paw. The diameter of the paw was measured in each rat by vernier caliper before and 3 h after carrageenan injection. The diameter of oedema was recorded as the difference between the two readings.

The percentage inhibition of oedema was calculated as (Arumozhi et al., 2005):

$$\%\ \text{Inhibition} = 100 - \frac{\text{Change in diameter of test group}}{\text{Change in diameter of control group}} \text{ X } 100$$

RESULTS AND DISCUSSION

The extractive value for methanol extract was higher than for other extracts; and hence was used for pharmacological screening. Similar higher percentage yield was found for methanolic extract of *S. arvensis* than with other fractions (Ali 2012).

In this study, the extract decreased propulsion of the charcoal meal through the gastrointestinal tract of mice dose dependently when compared with the control group. The 500 mg/kg; intraperitoneal (ip) methanol and 5 mg/kg; ip atropine had comparable inhibitory activity in intestinal motility (Figure 1). This reduction in gastrointestinal motility by methanol extract of *S. arvensis* may be due to antisecretory effects. The numerous phytochemicals like tannins, polyphenolic compounds, flavonoids, quercetrin and other chemical compounds may be speculated for antimotility effect (Ezekwesili et al., 2010). Hence, this activity of the plant may be useful in treatment of diarrhoea as an antimotility agent.

Similarly, methanol extract significantly inhibited locomotor activity on mice in dose dependent fashion indicating antidepressant property (Figure 2). Therefore, a standardized *S. arvensis* extract or its purified constituents could be of potential interest for the GI motility disorders.

The traction test and inclined plane test revealed that the methanol extract didn't possess skeletal muscle relaxant activity (Table 1 and 2).

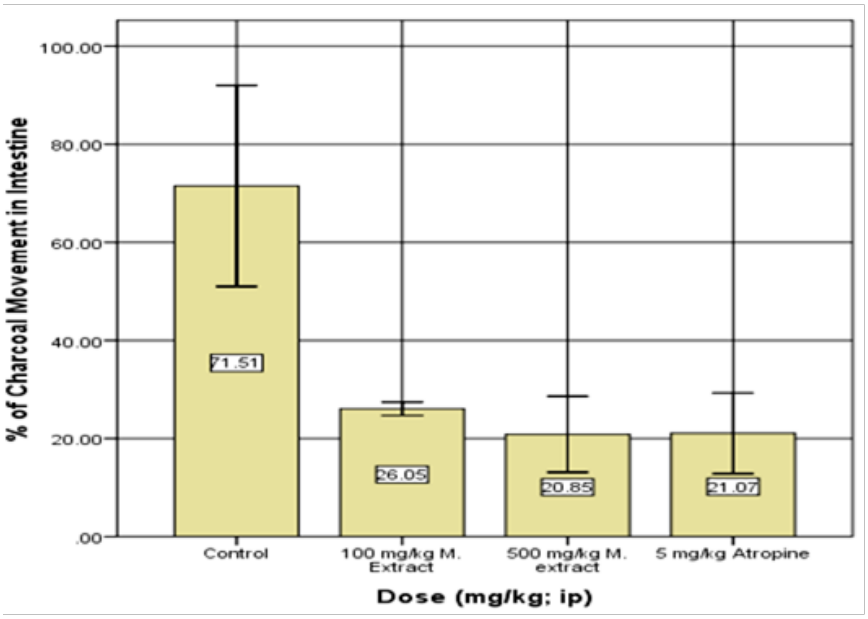

Figure 1. Effect of methanol extract on intestinal motility.

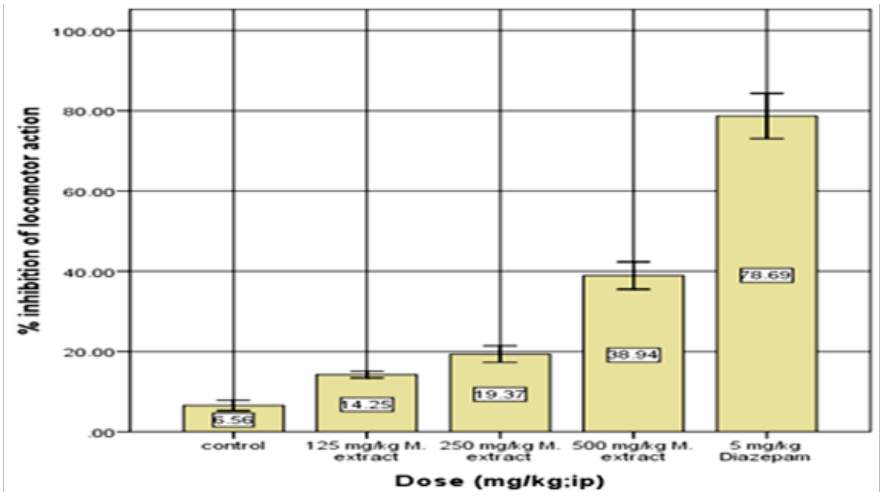

Figure 2. Effect of methanol extract on locomotor activity.

Further, methanol extract inhibited carrageenan induced acute paw oedema in dose dependent manner indicating anti-inflammatory activity. 400 mg/kg; ip methanol extract had higher anti-inflammatory activity than 10 mg/kg; ip diclofenac (Figure 3). Carrageenan releases prostaglandins (Winter et al., 1962) and inflammation occurs because of a proteolytic process with formation of kinin-like mediator(s) (Rosa and Sorrentino 1968).

S. arvensis contains various compounds palmitic acid, β-sitosterol, daucosterol, quercetin, apigenin-7-*O*-β-glucopyranoside, luteolin-7-*O*-β-D-glucopyranoside, quercetin-3-*O*-β-D-glucopyranoside and rutin (Jiang et al. 2009) which might have produced above pharmacological effects. However, further studies are required to isolate the major bioactive constituents and to verify the findings.

Conclusions

This study revealed significant anti-inflammatory, inhibitory effect on both gastrointestinal and locomotor activity. The study, however, revealed no skeletal muscle activity. It may, therefore, be concluded from this study that the plant possessed constituents that revealed the

Table 1. Effect of methanol extract of on skeletal muscle of mice by traction test.

Treatment	Dose (mg/kg; ip)	No. of mice failed to grasp the wire	Failure (%)
Control	-	0	0
Methanol extract	125	0	0
Methanol extract	250	0	0
Methanol extract	500	0	0
Diazepam	5	4	100%

Table 2. Effect of methanol extract on skeletal muscle of mice by inclined plane test.

Treatment	Dose (mg/kg; ip)	Animal falling after treatment	Failure (%)
Control	-	0	0
Methanol extract	125	0	0
Methanol extract	250	0	0
M Methanol extract	500	0	0
Standard (Diazepam)	5	4	100

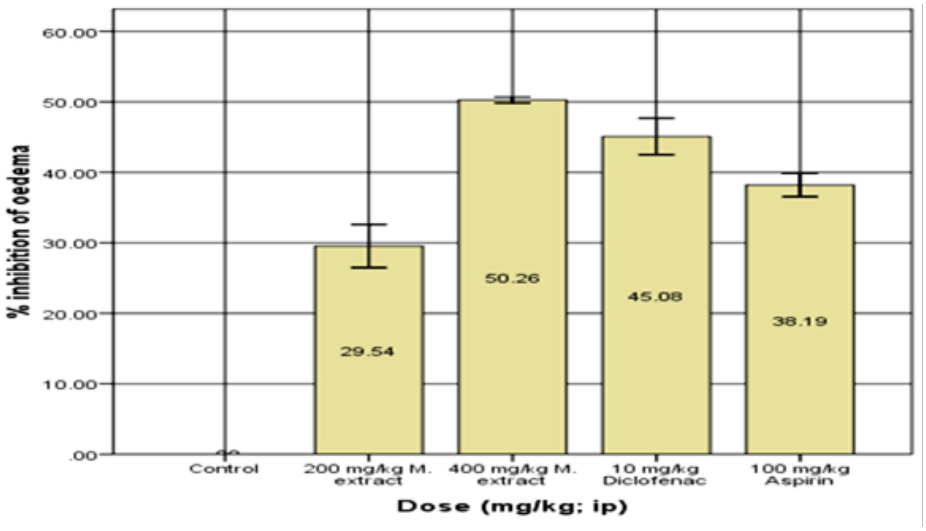

Figure 3. Effect of methanol extract on anti-inflammatory activity.

above pharmacological properties which may generate lead molecules for development of newer drugs. However, to reach any conclusive decision, a detailed phyto-chemical study for isolation, purification, identification, and characterization of the compound and biological studies with exact mechanism of action responsible for the particular activity, is necessary. Hence, further scientific investigation and specific studies are highly recommended for better evaluation of the potential effectiveness of the plant.

ACKNOWLEDGEMENT

The authors are thankful to Department of Pharmacy, Institute of Medicine, Tribhuvan University for providing laboratory and laboratory materials to conduct experiments.

Conflict of Interests

The authors have not declared any conflict of interest.

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