PHARMACOLOGICAL STUDIES OF METHANOLIC EXTRACTS OF *SONCHUS ARVENSIS* FROM KATHMANDU

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**ABSTRACT:** *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, the 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, didn’t showed skeletal muscle relaxant activity as observed in the traction test and inclined plane test. Thus, it is concluded that the plant possess strong phytochemicals having anti-inflammatory activity and inhibitory effect in locomotion and GI motility.

**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 population is still dependent on botanical medicines which indicates the importance of herbal medicines in the primary health care 1,2. *Sonchus arvensis*, a vigorous herbaceous perennial plant with milky sap and creeping roots is abundantly available in 1000m to 4100m in range and grows in sandy, loamy or clayey soils 3. In many areas, this sow thistle is considered noxious weed, as it grows quickly in a wide range of conditions and it’s wind-borne seeds allow them to spread rapidly 4. 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 then dried, crushed, and extracted using a 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, skeletal motility activity tests whereas male wistar rats (280±3.43 g) 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 mg/kg and 100mg/kg of methanol extract made in normal saline solution intraperitoneally (i.p) respectively; third group with atropine sulphate (5mg/kg) i.p and fourth group with control vehicle i.p. After 30 minutes all animals were fed with 1ml of charcoal meal (animal charcoal 12g, tragacanth 2g, and water 130ml) intra-gastrically with the aid of feeding needle. All the mice were killed after 30 minutes by inhalation of chloroform. The abdomen was opened, the intestine quickly isolated and the small intestine from pylorus to caecum was cut by scissors 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:

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

**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 5mg/kg of Diazepam serving as standard, while remaining groups were injected i.p with 125, 250 and 500mg/kg of the methanolic extract i.p. After 30 minutes of administration of extract, each mouse was placed in an open square field 50X50 cm surrounding and subdivided into 25 squares of 10X10 cm. The percentage inhibition in locomotion was calculated as:

\[
\text{% Inhibition} = 100 - \frac{\text{No. of squares crossed in test}}{\text{No. of squares crossed in control}} \times 100
\]

**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 125mg/kg, 250mg/kg and 500mg/kg respectively. One group received only distilled water as a control and another group received Diazepam at the dose of 5mg/kg as a standard. After 30 minutes, the capability of mice to touch the wire with at least one of the hind paws within 5 seconds was observed and percentage failure was calculated by the following formula:

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

**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 group 500 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:

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

**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 other group received i.p 100mg/kg of Aspirin and 10mg/kg Diclofenac serving as standard, while remaining groups were injected i.p with 200mg/kg and 400mg/kg of the Methanol extract. One hour later each animal was injected with 0.1ml 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 hours after carrageenan injection. The diameter of oedema was recorded as the difference between the two readings.

The percentage inhibition of oedema was calculated as:

\[
\text{% Inhibition} = 100 - \frac{\text{Change in diameter of test group}}{\text{Change in diameter of control group}} \times 100
\]

**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\(^9\).

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 (Fig. 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, quercetin and other chemical compounds may be speculated for antimotility effect\(^{10}\). Hence, this activity of the plant may be useful in treatment of diarrhoea as an antimitotility agent.

Similarly, methanol extract significantly inhibited locomotor activity on mice in dose dependent fashion indicating antidepressant property (Fig. 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).

### TABLE 1: EFFECT OF METHANOL EXTRACT OF ON SKELETAL MUSCLE OF MICE BY TRACTION TEST

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg; ip)</th>
<th>No of mice failed to grasp the wire</th>
<th>% failure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>-</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>M.Extract</td>
<td>125</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>M.Extract</td>
<td>250</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>M.Extract</td>
<td>500</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Diazepam</td>
<td>5</td>
<td>4</td>
<td>100%</td>
</tr>
</tbody>
</table>

### TABLE 2: EFFECT OF METHANOL EXTRACT ON SKELETAL MUSCLE OF MICE BY INCLINED PLANE TEST

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg; ip)</th>
<th>Animal falling after treatment</th>
<th>% failure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>-</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>M.Extract</td>
<td>125</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>M.Extract</td>
<td>250</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>M.Extract</td>
<td>500</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>Standard (Diazepam)</td>
<td>5</td>
<td>4</td>
<td>100%</td>
</tr>
</tbody>
</table>

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 (Fig. 3). Carrageenan releases prostaglandins \(^{11}\) and inflammation occurs because of a proteolytic process with formation of kinin-like mediator(s) \(^{12}\).

*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 \(^{13}\) 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 activity 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 above pharmacological properties which may generate lead molecules for development of newer drugs. However, to reach any conclusive decision a detailed phytochemical 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.
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REFERENCES:


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