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Research Article

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## Antinociceptive Activity of *Symphytum Officinale* Linn. Root on Swiss Albino Mice

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### ABSTRACT

Methanolic extract of the root of *Symphytum officinale* Linn was investigated for the analgesic activity. Experiments were carried out with this extract for their central antinociceptive potential on eddy's hot plate method in mice. In this model, methanolic extract showed significant increase in reaction time at doses of extract 200mg and 400mg/kg b.wt when compared to control.

**Keywords:** *Symphytum officinale* Linn, Anti nociceptive potential, Eddy's hot plate method

### ARTICLE INFO

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### 1. Introduction

*Symphytum officinale* Linn (common name: Comfrey) is a herb belongs to the family boraginaceae. Comfrey is native to Europe and temperate Asia and is common throughout England. It is generally found in wet areas and along road sides and in ditches. Traditionally, roots or leaves were taken internally against lung disorders, gastritis, and stomach ulcer and bleeding. The root being more active and

they can be taken internally and used externally. It is used as analgesic, astringent, demulcent, diuretic, expectorant and hemoptysis (bronchial or pulmonary hemorrhage). As a part of our research studies on the medicinal plants we investigated the analgesic activity of methanolic extract of root of *Symphytum officinale* Linn and herein, report the results of our examinations. [ 1,2]

## 2. Materials and method

### Materials

#### Collection of plant material

The roots of *Symphytum officinale* Linn was collected in the month of March 2013 from Tirumala hills, Tirumala, Chittoor Dt, A.P, India. It was authenticated by Dr. K. Madhava chetty, Department of botany, Sri Venkateswara University, Tirupati, A.P, India.

#### Chemicals

All the chemicals used for the study are of analytical grade.

#### Selection of experimental animals

Swiss albino mice of either sex (20-30 gm) were used in the study. Animals were housed individually in polypropylene cages in a ventilated room under ambient temperature of  $22 \pm 2$  C and 45-65 % relative humidity, with a 12 hour light followed by 12 hour dark. All the animals were acclimatized for at least 7days to the laboratory conditions prior to experimentation .Tap water and food pellets were provided ad libitum. Food pellets was with held overnight prior to dosing. Mice were handled and maintained strictly as per guidelines of "Guide for the care and Use of Laboratory animals".(Institute of Laboratory Animals Resources, National Academic Press 1996: NIH Publication number # 85-23, revised 1996).

#### Methods

#### Extraction of *Symphytum officinale* Linn roots

*Symphytum officinale* Linn. roots was shade dried and made into coarse powdered which was passed through a# 40 mesh sieve to get uniform particle size and was extracted using methanol by continuous hot percolation process using soxhlet apparatus<sup>3</sup>.

#### Preliminary phytochemical analysis

The methanolic extract of roots of *Symphytum officinale* Linn (MERSO) is subjected to preliminary phytochemical analysis to test for presence or absence of various phytochemical constituents used standard procedures<sup>4</sup>.

**Acute toxicity study of *Symphytum officinale* Linn root extract:** Acute toxicity study was performed for methanolic extract of roots of *Symphytum officinale* Linn according to Organization for Economic Co-operation and Development (OECD)-423 guidelines. Female mice selected by random sampling technique were employed in this study. The animals were fasted prior to dosing. MERSO was administered orally to different groups at the dose levels of 5, 50, 300, 2,000 and 5,000 mg/kg body weight. The animals were observed 24hrs for mortality with special attention during first 2hr and intermittently for 14 days<sup>5</sup>.

#### Analgesic activity

**Eddy's hot plate method:** The method originally described by Woolfe and Mac Donald (1944) has been modified by several investigators. Mice were divided into four groups of 6 mice each. Group I: Control group received normal saline 10ml/kg body weight. p. o. route, Group II :Standard group received pentazocine 5mg/kg body weight. i.p. route, Group III &IV: Test group 1&2 received MERSO 200 &400mg/kg body weights. p. o. route. Mice were pre-selected on the hot plate at  $55 \pm 0.5^{\circ}$  C. Licks on the rear paws were the parameters of observation. Animals showing a reaction time (defined as the latency for licking the hind feet or jumping.) greater than 20s were discarded. The

animals were then treated with vehicle, pentazocine, and MERSO of both doses 200 & 400 mg. Latency time (in seconds) for each mouse was determined on the hot plate during a maximum period of 20 sec, at the intervals of 30, 60, 90, 120 and 150min before and after the administration of the vehicle, pentazocine and extract<sup>6</sup>.

#### Statistical analysis

Statistical analysis was carried out using Instat 3 software. All results were expressed as Mean  $\pm$  S.E.M. The statistical analysis of all the results was done using one way analysis of variance (ANOVA) followed by Dunnett's test.

## 3. Results and Discussion

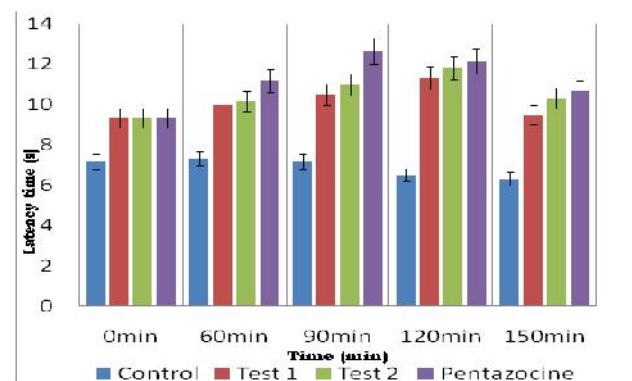
### 3.1. Preliminary phytochemical analysis of methanolic extract of *Symphytum officinale* Linn root.

The revealed results of the preliminary phyto chemical analysis of methanolic extract of *Symphytum officinale* Linn roots were shown below were tabulated in table 1

**Acute toxicity:** In acute toxicity study, no mortality was observed in animals treated with methanolic extract of roots of *Symphytum officinale* Linn up to a high dose of 5,000mg/kg body weight.

#### Analgesic Activity:

Analgesics are drugs that act on peripheral or central nervous system to selectively relieve pain without significantly altering consciousness<sup>7</sup>It has been reported that a number of flavonoids possess anti-inflammatory<sup>8</sup> and analgesic<sup>9</sup> activities. Hot plate method produces two measureable behavioural components in response to thermal pain, with regard to their reaction times. Responses such as paw licking and jumping in rats are considered to be supraspinally integrated<sup>10</sup>. As illustrated in Table 2 & figure1, oral administration of methanolic extract of roots of *Symphytum officinale* Linn significantly ( $P < 0.05$ ,  $P < 0.01$ ) increased the reaction time as compared to control group. The maximum analgesic effect was exhibited by methanolic extract of roots of *Symphytum officinale* Linn at 120 minutes. The maximum analgesic effect shown by Pentazocine at 90 minutes.



**Figure 2:** Effect of methanolic extract of roots of *Symphytum officinale* Linn and Pentazocine on latency to hot plate.

n=6. The observations are mean  $\pm$  SEM; \* $P < 0.05$ , \*\*  $P < 0.01$ , as compare to control (ANOVA followed by Dunnett's test); Test = Methanolic extract of roots of *Symphytum officinale* Linn.

**Table 1:** Preliminary phytochemical analysis

S.No	Compound	Test	MERCO
1	Reducing agents	Fehling's	+
		Benedicts	+
2	Test for Gums	Hydrolyze with HCl & performed Fehling's test	+
3	Test for proteins	Biuret	+
4	Test for Amino acids	Million's	+
5	Test for Cardiac glycosides	Legal	-
6	Test for flavanoids	Lead acetate	+
7	Test for Tannins	5% FeCl <sub>3</sub>	+
		Lead acetate	+
8	Test for Starch	Iodine	-
9	Test for Alkaloids	Dragendorff's	-
		Hager's	-
		Wagner's	-
10	Test for Saponin Glycosides	Foam	-
11	Test for Steroids	Salkowski	-

+ = Present, - = absent

**Table 2:** Effect of methanolic extract of roots of *Symphytum Officinale* Linn on latency to hot plate test in mice.

Groups	Dose (mg/kg)	0 min.	60 min.	90 min.	120min.	150min
Control (Normal saline)	10ml	7.16±0.47	7.33±0.55	7.16±0.4	6.5±0.56	6.33±0.66
Test -1	200	9.33±0.7*	10±0.44*	10.5±0.88**	11.33±0.49**	9.5±0.42**
Test-2	400	9.33±0.55*	10.16±0.7*	11±0.51**	11.83±0.30**	10.33±0.49**
Standard (Pentazocine)	5	9.33±0.62*	11.16±0.7**	12.66±0.84**	12.16±0.29**	10.66±0.49**

n=6. The observations are mean ± SEM. \* P<0.05, \*\* P<0.01, as compare to control. (ANOVA followed by Dunnett's test). Test = Methanolic extract of roots of *Symphytum officinale* Linn.

#### 4. Conclusion

The methanolic extract of *Symphytum officinale* Linn. root was subjected for phytochemical investigation and LD<sub>50</sub> studies. It was found that methanolic extract contained reducing sugars, gums, proteins, aminoacids, flavonoids and tannins. Flavonoids and tannins are responsible for analgesic activity. The extract was tested for their lethal effect up to the dose level of 5000mg/kg. No mortality was observed in mice. The administration of extract at doses of 200mg/kg, 400mg/kg, by oral administration, produced a significant (P<0.05-0.01) anti nociceptive effect in the hot plate induced pain in mice.

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