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# **Research Article**

# SCREENING OF ANALGESIC ACTIVITY OF ETHANOLIC EXTRACT of STYLOSANTHES FRUTICOSA BY HOT PLATE METHOD

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# ABSTRACT

In the present study, whole plant of Stylosanthes fruticosa was screened for its analgesic activity in mice by hot plate method. From the results obtained by phytochemical screening, ethanolic extract of Stylosanthes fruticosa (EESF) was selected for its analgesic activity screening. After toxicity studies, three doses i.e., 200, 400 and 800mg/kg were selected. Aspirin was used as a standard drug. EESF showed a significant results and the results were compared with that of standard drug.

KEYWORDS: Stylosanthes Fruticosa, Analgesic, EESF, Toxicity, Aspirin.

### INTRODUCTION

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**P**ain can be defined as a somatic sensation of acute discomfort, a symptom of some physical hurt or disorder, or even emotional distress. Pain is a crucial aspect of the body's defense mechanisms & it is a part of a rapid warning relay instruction the motor neurons of the central nervous system to minimize physical harm. Pain may be contained to a discrete area, as in an injury or it can be more diffuse <sup>[1]</sup>.

Analgesia simply means the absence of pain without losing consciousness. When employed as analgesics, these drugs usually are effective only against pain of low-to-moderate intensity, such as dental pain. Although their maximal efficacy is generally much less than the opioids, NSAIDs lack the unwanted adverse effects of opiates in the CNS, including respiratory depression and the development of physical dependence. NSAIDs do not change the perception of sensory modalities other than pain. Chronic postoperative pain or pain arising from inflammation (eg, somatic pain) is controlled particularly well by NSAIDs <sup>[2]</sup>.

*Stylosanthes fruticosa* belongs to the family fabaceae which is commonly known as shrubby pencil - Flower, wild Lucerne and Arfrican stylo. In Telugu it is called as Saile kampa,

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Saali kampa. *Stylosanthes fruticosa* native range that extends from South Africa to Ethiopia, India, and Srilanka. It is a woody herb, Annual and Perennial legume. It is used as an analgesic and as anti-bacterial agent. It is helpful in erosion control as it can contribute to the stabilization and sustainable use of degraded lands. Prostate forms can be promoted to protect against soil erosion [3, 4].

An extended literature review reveals that, leaves of the plant was already explored for its antibacterial activity <sup>[5]</sup>, dried whole plant extracts was studied for phytochemicals, proximate analysis <sup>[6]</sup>, antimicrobial and antioxidant patience and leaf extracts were studied to characterize the bioactive constituents using UV-Visible, FTIR and GC-MS <sup>[7]</sup>. Whole plant of *Stylosanthes fruticosa* was not scientifically explored for its analgesic activity, hence an effort has been taken in this study to explore analgesic activity of *Stylosanthes fruticosa*.

### MATERIALS AND METHODS

#### **Collection and authentication of the plant:**

Whole plant of *Stylosanthes fruticosa* were collected from Sri Venkateswara University Campus, tirumala gardens of Chittoor District of Andhra Pradesh, India and the same was authentified by Assistant Professor, Dr.K.Madhava Chetty, Department of Botany, S.V.University, Tirupathi, A.P. Voucher specimen was deposited (Voucher No:98) at Department of Pharmacognosy for further reference.

### Preparation of extracts:

The shade dried plant material was reduced to moderately course powder and extracted successively with various solvents of increasing polarity using Soxhlet apparatus. The extraction was carried out until the extractive become colorless. The solvent was completed removed from the marc in each case before the next extraction was carried out. The solvents were recovered from their extracts by distillation

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under reduced pressure. The dried extracts thus obtained were kept in a desiccator and was used for further experiments.

### Preliminary phytochemical screening: [8, 9]

Cyclohexane, diethyl ether, acetone, ethanol and aqueous extracts of whole plant powder of *stylosanthes fruticosa* was subjected to preliminary phytochemical screening for the identification of the presence or absence of various constituent such as phytosterols, triterpenoids, carbohydrates, glycosides ,alkaloids, saponins, tannins/phenols and flavonoids.

### Acute toxicity studies:

Acute toxicity studies were performed for ethanolic extract of whole plant of *Stylosanthes Fruticosa* [EESF] using different doses according to OECD guidelines. For the pharmacological studies, the amount of dose administered was adjusted on the basis of observation during toxicity studies <sup>[10]</sup>.

### **Experimental design:**

#### Hot plate method:

The paws of mice are very sensitive to heat even at temperatures which do not damage the skin. They respond by jumping, withdrawal of paws and licking of paws. The time until these responses occur can be prolonged after administration of centrally acting analgesics.

Five groups of mice each consisting of six were divided. The basal reaction time was observed previously in animals based on hind paw licking or jump response whichever appears first when placed on hot plate maintained at  $55^{\circ}$ C.

**Group I** : Control animals received 2ml/kg of 1% NaCMC **Group II** : Standard group animals received Aspirin 100mg/kg **Group III**: Test group animals received EESF 200mg/kg **Group IV**: Test group animals received EESF 400mg/kg

### **Group V** : Test group animals received EESF 800mg/kg

The reaction time was measured in seconds at 0 min (before drug challenge), 15 min, 30 min, 60 min and 120 min after administration of the above scheduled drugs <sup>[11-14]</sup>.

### **RESULTS AND DISCUSSION**

The results of the preliminary phytochemical screening test indicated the presence of carbohydrates, alkaloids, glycosides, flavonoids, saponins, phytosterols/ triterpenoids and phenolic compounds/tannins in ethanolic extract. The results also indicated the presence of carbohydrates, glycosides, flavonoids and saponins in aqueous extract. From the identified phytoconstituents, ethanolic extract of whole plant of *Stylosanthes fruticosa* was selected for the further study.

The results of the acute toxicity study showed that EESF was safe to use to animals up to the dose of 2000mg/kg orally. From the results obtained, 200mg/kg, 400mg/kg and 800mg/kg of extracts of *Stylosanthes fruitcosa* were selected for the experimental studies.

The tabulated form of the results of hot plate method is given in table-1. In this study a cut off period of 15 sec was observed to avoid damage to the paws of animals. The reaction time of standard group animals increased from  $6.87 \pm 0.48$  to  $14.04 \pm 0.33$  at 120 min and in EESF 200mg/kg treated animals, the reaction time increased from  $7.32 \pm 0.43$  to  $9.84 \pm 0.46$ , in 400mg/kg treated animals from  $6.98 \pm 0.46$  to  $11.50 \pm 0.56$  and in 800mg/kg treated animals reaction time increased from  $7.04 \pm 0.38$  to  $13.76 \pm 0.40$ .

Groups		Reaction time (Seconds)				
		0 min	15 min	30 min	60 min	120 min
Ι	Control	7.26 ± 0.49	$7.02 \pm 0.58$	6.97 ± 0.38	$7.18 \pm 0.36$	$7.00 \pm 0.44$
Π	Standard	6.87 ± 0.48	$8.79 \pm 0.44^*$	10.63 ± 0.65**	12.47 ± 0.52**	14.04 ± 0.33**
III	EESF 200mg	$7.32 \pm 0.43$	7.36 ± 0.38	7.48 ± 0.52	8.17 ± 0.54*	9.84 ± 0.46**
IV	EESF 400mg	6.98 ± 0.46	7.63 ± 0.67	8.24 ± 0.53*	9.96 ± 0.42**	11.50 ± 0.56**
V	EESF 800mg	$7.04 \pm 0.38$	8.56 ± 0.56*	$10.28 \pm 0.61^{**}$	12.34 ± 0.56**	13.76 ± 0.40**
						-

### Table No. 1: Analgesic effect of EESF on mice by hot plate method

Each value represents the mean  $\pm$  SEM, n=6. \* p < 0.05, \*\* p < 0.01, Groups II to V are compared with group I.

### SUMMARY AND CONCLUSION

 $S_{tylosanthes}$  fruticosa belongs to the family fabaceae which is commonly known as shrubby pencil - Flower, wild Lucerne and Arfrican stylo. In Telugu it is called as Saile kampa, Saali kampa. *Stylosanthes fruticosa* native range that extends from South Africa to Ethiopia, India, and Srilanka. It is a woody herb, Annual and Perennial legume. It is used as an analgesic and as anti-bacterial agent. It is helpful in erosion control as it can contribute to the stabilization and sustainable use of degraded lands. Prostate forms can be promoted to protect against soil erosion.

Preliminary phytochemical screening study confirmed the presence of carbohydrates,alkaloids, glycosides, flavonoids, saponins, phytosterols / triterpenoids and phenolic compounds/tannins in ethanolic extract. Acute toxicity study of ethanolic extract of *Stylosanthes fruticosa* was tested in mice's. It has been found that these extracts were safe to use to animals at a dose of 2000mg/kg per orally.

Analgesics relieve pain without loss of consciousness. Various literatures recommend the use of natural products in various traditional medical systems to treat pain. Nociceptive reaction towards thermal stimuli is well-validated model for detecting centrally acting analgesic activity. The centrally acting analgesics generally elevate pain threshold of mice towards heat.

In the present study hot plate method was used to study the analgesic property of EESF. In this model, EESF 800mg/kg was found more active than other test doses and results were comparable to the standard analgesics Aspirin.

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