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EXTRACTION AND PHYTOCHEMICAL SCREENING OF *MIKANIA SCANDENS* LINN. AND EVALUATION OF ITS METHANOLIC EXTRACT FOR ANALGESIC ACTIVITY

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ABSTRACT

Keywords:

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Mikania scandens is a rapidly growing herbaceous to semi-woody, perennial vine. It grows quickly over other plants such as young trees, smothering them; it can climb trees up to 25 m tall. Although not as serious a weed as *M. chordata*, in Southeast Asia it has become a hard to eradicate weed of tea, rubber, and other plantation crops. It also reduces the carrying capacity of pasture. The achenes are spread by wind, water and animals. The plant also reproduces by old rootstocks, runners and suckers. It is still under investigation. Although it has some analgesic and anti-inflammatory properties, but these uses are not widely accepted till now. Folkloric uses include utilisation of the herb as an anticoagulant or other medicinal purpose. The aim of the undertaken work is to identify and isolate the chemical constituents of the plant *Mikania scandens* and to evaluate its reported analgesic activity.

INTRODUCTION: Herbal medicine is the oldest form of healthcare known to mankind. Herbs had been used by all cultures throughout history. It was an integral part of the development of modern civilization. Pain is a sensorial modality and primarily protective in nature, but often causes discomfort. It is the most important symptom that brings the patient to physician.

Analgesics relieve pain as a symptom, without affecting its cause. The name *Mikania scandens* was once used to refer to most of the slender twiners in tropical and temperate America. The naming is done after the Czech botanist Johann Christian Mikan. *Mikania* originates from South America. A few species, such as *Mikania scandens*, are found in temperate areas of North and South America, tropics.

Distribution: Native to Central and South America, Mexico and the West Indies. Naturalized in India, Pakistan, Sri Lanka, Southeast Asia, United States.

Plant Profile:

Scientific Classification

Kingdom	:	Plantae – Plants
Sub kingdom	:	Tracheobionta – Vascular plants
Superdivision	:	Spermatophyta – Seed plants
Division	:	Magnoliophyta – Flowering plants
Asterales		
Family	:	Asteraceae – Aster family
Genus	:	<i>Mikania</i> Willd – hempvine
Species	:	<i>Mikania scandens</i> (L.) wild.

The aim of the undertaken work is to identify and isolate the chemical constituents of the plant *Mikania scandens* and to evaluate its reported analgesic activity¹.

The experimental work is categorized under two broad heading namely phytochemistry part and pharmacological evaluated for analgesic activity^{1,2}.

MATERIAL & METHODS:

The collection of Plant Material: The plant material of *Mikania scandens* was collected from Kolkata and suburban regions in the month of September 2011. Here we use the whole plant material including stems and leaves for the investigation purpose.

Extraction by Soxhlet Process: The solid dry material of the plant *Mikania scandens* was placed inside a thimble made from thick filter paper, which is loaded into the main chamber of the Soxhlet extractor¹². The Soxhlet extractor is placed onto a flask containing the extraction solvent. Here we use three types of solvent to get maximum extracted compound from this plant. We used Methanol, Petroleum Ether and Chloroform respectively as solvent in the extraction process. At the end of the extraction process, we got three type of extract of *Mikania scandens*^{3,12}.

- a) Petroleum Ether extract
- b) Chloroform extract

The solvent is completely removed by extraction and obtained deep green gummy exudates. This crude extract was used for further investigation.

Phytochemical Screening for Constituents: Phytochemical analyses of the crude extract revealed the presence of flavonoids, steroid, alkaloid, tannin and saponin and other constituents such as sesquiterpenes³.

Pharmacological Evaluation: From the review of literature, it became evident that *Mikania scandens* was reported to have some analgesic and anti-inflammatory properties. Here, our aim is to prove and reevaluate the pharmacological properties of *Mikania scandens*².

It can be done by two methods namely, Oral Toxicology test and Analgesic screening by tail flick method².

Acute Toxicity Studies:

Experimental Animals: Swiss albino mice weighing 20-25 gm of either sex were used for the study⁴. The animals were procured and housed in the animal

house maintained under standard hygienic conditions at 20±2°C, humidity (60±10%) complying with the norms of the Institutional Animal Ethical Committee (IAEC) as per CPCSEA guidelines under optimum conditions and facilities for experimentation.

Oral Toxicology Test: The acute oral toxicity studies were performed to study the acute toxic effects and to determine minimum lethal dose of the drug extracts. Swiss albino mice of either sex weighing 20-25gm were used for the study. The aqueous petroleum ether, methanol and chloroform extracts were administered orally to different groups of overnight fasted mice at the doses of 1000, 2000 and 3000 mg/kg body weight. After administration of the extracts, animals were observed continuously for the first three hours for any toxic manifestation. Thereafter, observations were made at regular intervals for 24 hrs².

Analgesic Screening by Tail Flick Method: Analgesic activity of methanol extract of *Mikania scandens* was studied by using the tail flick method in mice.

MATERIALS AND METHODS:

Animal used: For the experiment, Swiss Albino mice of either sex, 3-4 weeks of age, weighing between 20-25 g, were collected. Animals were maintained under standard environmental conditions (temperature: (24.0±1.0°C), relative humidity: 55-65% and 12h light/ 12 h dark cycle) and had free access to feed and water. The animals were acclimatized to laboratory condition for one week prior to experiments^{5,6}.

Apparatus : A warm water bath.

Tail Immersion Test: Male or female albino mice weighing between 20-25gm were fasted for 24hours with water given *ad libitum* maintained at room temperature and was divided into 3 groups of 3 mice. Group I served as control. Group II served as standard and were given nimesulide (50 mg/kg body weight) orally. Group III (test) were treated orally with aqueous extract of 250 mg/kg body weight. Analgesic effect of the test substances was determined by the hot tail-flick method described by Sewell and Spencer (1976). One to two cm of the tail of mice was immersed in warm water kept constant at 50°C.

The reaction time was the time taken by the mice to deflect their tails. The first reading is discarded and the reaction time was taken as a mean of the next two readings. The latent period of the tail-flick response was taken as the index of analgesia and was determined before and at 0, 10, 20, 30, 40, 50 and 60

min after the administration of drugs. The maximum reaction time was fixed at 15 seconds.

Results of the Test: The result of the tail immersion test yielded results given subsequently and indicates a positive analgesic activity of the crude extract of the herb. (**Table. 1**)

TABLE 1: THE RESULTS OF BASAL REACTION TIME OF ANIMALS IN THE TAIL IMMERSION TEST PERFORMED ON SWISS ALBINO MICE IN THE EXPERIMENT TO EVALUATE THE ANALGESIC ACTIVITY OF THE METHANOLIC EXTRACT OF CRUDE DRUG

Group Name	Body Weight (gm)	Basal Reaction Time (sec)	0 (sec)	10 (sec)	20 (sec)	30 (sec)	40 (sec)	50 (sec)	60 (sec)
Control (normal saline)	20	2	2	3	2	2	1	2	1
Coloured									
Coloured	22	2	2	2	1	2	1	2	1
Colourless	20	1	1	2	1	1	2	1	2
Standard (Nimesulide)									
Coloured	22	2	2	3	4	5	6	5	5
Coloured	23	3	3	4	4	5	6	5	4
Colourless	23	2	2	4	5	5	6	3	2
Test (Methanolic extract of <i>Mikania scandens</i>) Coloured	25	3	2	3	4	2	2	5	6
Colourless	24	2	2	4	5	4	5	5	6
Colourless	25	2	3	3	4	3	4	6	6

RESULT AND DISCUSSION:

Phytochemical Screening: Phytochemical analyses of the crude extract revealed the presence of flavonoid steroid, alkaloid, tannin and saponins etc.

Acute Toxicity Studies: The acute oral toxicity studies were performed to study the acute toxic effects and to determine minimum lethal dose of the drug extracts. Swiss albino mice were used for the study. The aqueous, pet ether, methanol and chloroform extracts were administered orally at the doses of 1000,2000 and 3000mg/kg body weight. And observed for 24 hours. But no mice behavioral change is observed neither anyone was killed by the dose. Hence it is proved that the drug has no toxic effect in this level.

Tail Immersion Test: The tail withdrawal reflex time following administration of the extract of *M scandens* was found to increase with increasing dose of the sample. The result was statistically significant ($p < 0.05$ - 0.001) and was comparable to the reference drug Nalbuphine.

CONCLUSION: The undertaken work is a study of the phytochemistry of *M. scandens* and investigates its pharmacological properties in vivo in animal models. And from the above experiments, it can be safely concluded that the polyherbal extract of *M. scandens* indeed have a distinct analgesic property and it may act by elevating the pain threshold in the body.

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