



**EVALUATION OF ANALGESIC, ANTI-INFLAMMATORY AND ANTIPYRETIC  
ACTIVITY OF FLOWER BUDS METHANOLIC EXTRACT OF *MICHELIA CHAMPACA*  
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**ABSTRACT**

Present study was intended to evaluate the analgesic anti-inflammatory and antipyretic activity of methanolic extract of *Michelia champaca* in experimental standard modals i.e. albino rats following oral administration. The results showed that the methanolic extract significantly reduce the edema induced by carrageenan within 1 to 5 hrs. Post dosing at all the dose levels used. On the analgesic property acetic acid induce writhing was significantly reduce in the formalin test, the extract also significantly decreases the painful stimulus in both phases of test which confirms central and peripheral effects of the drugs. Its effects on antipyretic activity were also appreciable it significantly reduces fever at higher doses within 2 hrs. On yeast induce hyperthermia in rats.

**KEYWORDS:** Analgesic, Anti-inflammatory, Antipyretic, *Michelia champaca*, albino rats.

**INTRODUCTION**

*Michelia champaca* Linn. known as *champaca* is belonging to the family of Magnoliaceae.<sup>[1]</sup> It consists of 12 genera and 220 species of evergreen trees and shrubs, native to tropical and subtropical South and Southeast Asia (Indomalaya), including southern China. It's commonly referred as yellow *champaca*. There are three species of *Michelia* available in Malaysia. They are *Michelia Alba* (white chempaka), *Michelia champaca* (orange chempaka) and *Michelia figo* (dwarf chempaka) with *Michelia champaca* and *Michelia Alba* being the most popular species within the family.<sup>[2]</sup> In recent times there are several reports of medical specialty roles and activities of *Michelia champaca* and its active principals on the circulatory system, *Michelia*, known by the scientific name *Michelia champaca*, is a very tall tree that grows up to 30m tall. The young branches are covered with grey hairs. The leaves are ovate in shape and are up to 30.5cm long and 10.2cm wide narrowing to a fine point at the apex. Flower buds of *Michelia champaca* Linn. belonging to the family Magnoliaceae is commonly used by many traditional healers in most of the herbal preparations for diabetes<sup>[3]</sup> and kidney diseases.<sup>[4]</sup> Traditionally, it is being used in fever, colic, leprosy, post partum protection<sup>[5]</sup> and in eye disorders.<sup>[6]</sup> It has been reported to possess antipyretic, anti-inflammatory,<sup>[7]</sup> insecticidal,<sup>[8]</sup> antimicrobial<sup>[5]</sup>, and

leishmanicidal activities.<sup>[9]</sup> The active constituents reported in this plant are alkaloids, saponins, tannins, sterols, flavonoids and triterpenoids.<sup>[5]</sup> From the above reference this study was conducted to evaluate the extract of *Michelia champaca* flower buds for central analgesic activity along with marked antipyretic and anti-inflammatory activity in rats.

**MATERIALS AND METHODS**

**Plant material**

Flower buds of *M. champaca* were collected in March 2014 from tirupati, India. The taxonomical identification of the plant was done by Dr. V.S Raju, Department of Botany Kakatiya University. The herbarium was prepared and a voucher specimen (Sample No .04, Ref no.Gen/05-04/2014) was deposited.

**Preparation of extracts**

Dried and powdered Flower buds material of *Michelia champaca* (500 g) was successively Soxhlet extracted with petroleum ether (60-80°), chloroform, acetone, Methanol and water for 72 h each. Crude aqueous extract of this plant was prepared separately by boiling the plant material (25 g) with 200 ml of water for 15 min. The obtained extracts were evaporated in vacuum to give residues and their percentage yields were determined.

### Animals and treatment

Healthy Wistar rats of either sex (150-180 g), with no prior drug treatment, were used for the present studies. The animals were fed with commercial pellet diet and water *ad libitum*. The animals were acclimatized to laboratory hygienic conditions for 10 days before starting the experiment. Animal study was performed in the Division of Pharmacology,

### Acute toxicity studies

The acute toxicity test of the extracts was determined according to the OECD (Organization for Economic Co-operation and development) guidelines No. 420. Female Wistar rats (150-180 g) were used for this study. After the sighting study, starting dose of 2000 mg/kg (p.o.) of the test samples was given to various groups containing five animals in each group. The treated animals were monitored for 14 days, for mortality and general behaviour. No death was observed till the end of the study. The test samples were found to be safe up to the dose of 2000 mg/kg, and, from the results, 400 mg/kg dose was chosen as the maximum dose for further experimentation.

### ANALGESIC STUDY

Analgesic effects was evaluated using three different models: the writhing test, tail flick test and formalin test.

### WRITHING TEST

Male Swiss rats (180-200 g) were used according to the method described previously by.<sup>[10]</sup> The total number of writhings, following intraperitoneal (i.p.) administration of 0.6% acetic acid, was recorded for 20 min. starting 10 min. after injection. The animals were pretreated with hydroalcoholic extracts (HAEs) from Flower buds of *M. champaca* 100, 150, 200 mg/kg b.wt respectively.

### Analgesic activity

Table 1: writhing test

Group	Treatment	n	Dose mg/kg	Route of administration	No. Of writhes	Inhibition Of Writhing Response (%)
I	Control	6	-	i.p	49.06±4.08	-
II	Aspirin	6	300	i.p	12.05±2.14	90
III	Flower buds of <i>M. champaca</i>	6	50	i.p	20.65±2.10	40
IV	Flower buds of <i>M. champaca</i>	6	150	i.p	18.85±1.78	65
V	Flower buds of <i>M. champaca</i>	6	200	i.p	8.68±1.14	85

Mean = S.E.M. of 6 animals. \*\* = P≤0.001= highly significant. Group II ,III , IV, and V compared with Group I.

### TAIL-FICK TEST

The basal reaction time of each mouse was determined using tail-withdrawal response when one-third of the tail was immersed in water bath at 51°C.<sup>[11]</sup> The cutoff time for immersion was 180 s. The reaction time was evaluated 30, 60, 90, 120 and 240 min after oral administration of extracts, distilled water or acetylsalicylic acid.

### FORMALIN TEST

The method used in our study was similar to that described previously.<sup>[12]</sup> Twenty microliter of 5% formalin was injected subcutaneously into the right hind paw of mice. The time (in seconds) spent in licking and biting responses of the injected paw was taken as an indicator of pain response. Responses were measured for 5 min after formalin injection (early phase) and 20–30 min after formalin injection (late phase). *Michelia champaca* flower buds M extracts (0.5 and 1.0 g/kg, i.p.) were administered 60 min before formalin injection. Indomethacin (10 mg/kg, i.p.) was administered 30 min before formalin injection. Control group received the same volume of saline by oral administration.

### Anti-inflammatory

#### *Carrageenan induced hind paw edema in rats*

Paw edema was produced in rats by carrageenan following the methods of Winter et al. (1962) respectively.<sup>[13]</sup> Male rats weighing 100–120 g were divided into groups of six animals. A volume of 0.05 ml of 1% carrageenan in normal saline solution (NSS) in 0.2M carbonate buffer was injected intradermally into the plantar side of the right hind paw of the rat. Test drugs and vehicle were given 1 h prior to carrageenan injection. Paw volumes were measured using a plethysmometer (model 7150, Ugo Basile, Italy) before as well as 1, 3 and 5 h after carrageenan, injection. Results obtained were compared with those obtained from their.

**Anti-inflammatory activity****Table2 Carrageenan –induced paw edema method.**

Group	n	Dose (mg/kg)	Pawvolume increase(ml)			Inhibition (%)		
			1hr	3hr	5hr	1hr	3hr	5hr
Control	6		0.36±0.07	0.69±0.05	0.82±0.03			
Aspirin	6	300	0.10±0.02**	0.21±0.02**	0.27±0.03**	72	70	67
Flower buds of <i>M. Champaca</i> ME	6	100	0.22±0.04*	0.47±0.01*	0.51±0.02*	39	32	38
Flower buds of <i>M. Champaca</i> ME	6	150	0.18±0.03*	0.39±0.01*	0.42±0.04*	50	43	49
Flower buds of <i>M. Champaca</i> ME	6	200	0.07±0.01**	0.24±0.02**	0.35±0.02**	81	65	57

n = 6 animals in each group. \* = p≤0.01 (significant). \*\* = p≤0.01 (highly significant) control groups, which received vehicle only

**Antipyretic activity**

*Yeast induced hyperthermia in rats* In order to determine the antipyretic activity, rats treated with FCA (adjuvant-induced arthritis model) were employed.<sup>[14]</sup> The heat on the surface of left and right hind paws of each rat

was measured every other day with a clinical contact digital thermometer (Prima long) and the difference between these two values was compared with that of control animals and results were evaluated statistically.

**Antipyretic activity****Table3: Brewer's yeast induced pyrexia in rats.**

Group	n	Dose (mg/kg)	Rectal temperature in °C at time (hr.)					
			18 <sup>a</sup> hr	0 <sup>b</sup> hr	1hr	3hr	5hr	6hr
Control	6		38.65±0.27	39.20±0.04	39.91±0.11	39.76±0.14	39.64±0.07	39.83±0.06
Aspirin	6	300	38.47±0.25	39.32±0.13**	37.28±0.13**	36.11±0.08**	35.03±0.11**	36.03±0.21**
Flower buds of <i>M. champaca</i> ME	6	100	38.13±0.12	39.41±0.12ns	39.31±0.12*	39.30±0.26 ns	38.65±0.20 ns	38.40±0.1 ns
Flower buds of <i>M. champaca</i> ME	6	150	38.25±0.14	39.63±0.10 ns	38.08±0.15**	38.54±0.21 ns	38.62±0.11 ns	38.22±0.11 ns
Flower buds of <i>M. Champaca</i> ME	6	200	38.68±0.09	39.48±0.14 ns	38.24±0.09**	38.03±0.02**	38.03±0.08**	37.42±0.08**

n = 6 animals in each group, ns = non-significant, \* = p≤0.01 (significant) \*\* = p≤0.01 (highly significant), a = temperature just before yeast injection b = temperature just before drug administration

**RESULT**

*Analgesic activity of administration of Michelia champaca* flower buds M extract at the dose level of 100, 150 and 200 mg/kg b. wt. to the rats produced weak effect on the writhing induced by the injection of 0.6% acetic acid when compared with the aspirin (300mg/kg) by 79% while the treated group with *Michelia champaca* flower buds M extracts inhibited the writhing by 40%, 65%, 85% respectively (table 1). The methanolic extract of *Michelia champaca* flower buds (100- 200 mg/kg) produced inhibition of formalin induce biphasic pain response (neurogenic and inflammatory pain) in rats. The analgesic effect of this fraction occurred predominately during the II phase; 200 mg dose level was more efficient in the late phase. *Anti-inflammatory activity of Michelia champaca* flower buds, The inhibitory activity on carrageenan induced rat hind paw edema, caused by the subplanar administration of *Michelia champaca* flower buds M extract, at various assessment times after

carrageenan injection are shown in table 2. aspirin, a cyclooxygenase inhibitor, at the dose of 300mg/kg body weight exhibited significant (p≤0.01) edema inhibition. *Michelia champaca* flower buds M extract at doses of 50, 100, 150 mg/kg body weight also possessed significant (p≤0.001) inhibitory effect on carrageenan induced paw edema at all recorded times. This increase was observed at 1 hr. and was maximum at 5hr. after administration of carrageenan in the vehicle group. *Antipyretic activity of Michelia champaca* flower buds M extract produced a reduction (p≤0.01) in hyperpyrexia induced by dried yeast injection in rats, with activity being pronounced within 18 hrs. After administration of the extracts (table-3).

**DISCUSSION**

The inflammatory effect, analgesic, antipyretic properties of *Michelia champaca* flower buds M extract methanolic extracts were investigated in the present

study The writhing test allows us to identify central and peripheral analgesic compound.<sup>[15]</sup> The tail formalin test is recent algesiometric assay in which only behaviour suggestive of pain is the licking of tail. The lack of 2 distinct phases after the administration of formalin in the tail may be due to a different pattern of the release of the chemical pain mediators at both the spinal and peripheral levels and this method mainly identifies peripheral analgesic.<sup>[16]</sup> The thermal model of the tail flick test is considered to be spinal reflex, but could also involved higher neural structures at this method identify mainly central analgesic.<sup>[17]</sup> The extracts derived from *Michelia champaca* flower buds M extract exhibited analgesic activity in albino rats by inhibiting acetic acid induce writhing, which is a model of visceral pain.<sup>[18]</sup> Acetic acid induced writhing is a highly sensitive and useful test for analgesic drug development, but not a selective pain test as it gives false positives with sedatives, muscles relaxants and other pharmacological activities.<sup>[19]</sup> The analgesic property of *Michelia champaca* flower buds M extract can also probably be to the blockade of the effects or the synthesis and /or release of PGs and /or other endogenous substances that excite pain nerve endings.<sup>[20]</sup> Although the writhing response test is very sensitive it has poor specificity as in analgesic screening test, therefore the formalin test and tail flick test were conducted to confirm and study the possible analgesic mechanism of the tested plants. The formalin test consist of 2 distinct phases that possibly reflecting different types of pain mechanisms. The I phase immediately after injection of formalin and last about 5 minutes this is due to result chemical, peripheral stimulation of nociceptors.<sup>[21]</sup> The II phase starts approximately 15-20 min after formalin injection and lasts for 20- 40 min. the formalin test is sensitive to nonsteroidal anti-inflammatory drugs and other mild analgesics. In the study aspirin inhibited analgesic behaviour during both the early and late phases. The carrageenan test was selected because of its sensitivity in detecting orally active inflammatory agents particularly in the acute phase of inflammation.<sup>[22]</sup> The intraplantar injection the carrageenan in rats leads to paw edema. Its first phase that is 1 hour after infection results from the concomitant release of mediators, histamine, serotine and kinins on the vascular permeability. The II phase is correlated with elevated production of prostaglandins, oxygen derive free radicals & production of inducible cyclooxygenase.<sup>[23]</sup> Oral administration of the *Michelia champaca* flower buds M extract extracts suppressed that edematous response 1 hr after carrageenan injection & this effect continues up to 5 hr It is well known that most of the anti-inflammatory analgesic drugs posses antipyretic activity *Michelia champaca* flower buds M extract revealed weak antipyretic effect at low dose i.e. 100mg/kg b.wt. but at higher dose i.e. 150 and 200mg/kg bwt. It produce marked antipyretic effect in brewel yeast induce fibril rats. In general, anti-inflammatory drugs produce their antipyretic action through inhibition of prostaglandin synthesis within the hypothalamus.<sup>[24]</sup> Although there is no direct evidence of

*Michelia champaca* flower buds M extract to interfere with prostaglandin synthesis in hypothalamus but it can be supported by a related study in which *D. Odorifera* extract was found to inhibit prostaglandin by synthesis<sup>[25]</sup> From these results is concluded that the extract from *Michelia champaca* flower buds M extract possess both peripheral and central analgesic activity along with marked antipyretic and anti-inflammatory activity in rats and also the present study provoke the traditional use of *Michelia champaca* flower buds M extract for the purpose various ailments like analgesic, anti-inflammatory and antipyretic.

## CONCLUSION

The methanol extract of *Michelia champaca* flower buds has moderate and safe oral both peripheral and central analgesic activity along with marked antipyretic and anti-inflammatory activity in rats.

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