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## Anti-Inflammatory Effects of Roots of *Terminalia paniculata* and *Boswellia ogadensis*

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### Abstract:

*Terminalia paniculata* and *Boswellia ogadensis* most widely found plants in India. Methanolic and nHexane extract of *Terminalia paniculata* roots and *Boswellia ogadensis* roots were analysed for anti-inflammatory activity. The effect of methanolic and n-Hexane extracts of roots of *Terminalia paniculata* Brandis and *Boswellia ogadensis* significantly in 200,400 and 600 mg/kg p.o dosage, showed anti-inflammatory activity (acute and chronic models).

**KEYWORDS:** Methanol, n-Hexane, acute and chronic inflammatory models.

### INTRODUCTION

Animals' existence on this earth has been made possible only because of the vital role played by plants. Plants and animals coexist together and their dependency on each other gives importance to living life. Medicinal plants existing even before human being made their appearance on the earth.<sup>[1,2]</sup>

*Terminalia paniculata* is herb belongs to the family Combretaceae. *Terminalia paniculata* is semi-evergreen tree species.<sup>[3]</sup> *Boswellia ogadensis* (Burseraceae) is a narrow endemic and endangered deciduous tree species.<sup>[4]</sup> Present study focus mainly on acute and chronic anti-inflammatory effect of n-Hexane and methanolic extracts of *Terminalia paniculata* Brandis roots and *Boswellia ogadensis* roots.

### MATERIALS AND METHODS

#### Collection and Authentication

The roots of *Terminalia paniculata* Brandis and *Boswellia ogadensis* most widely found in the India. The plant was collected from Alwar Rajasthan, India. The plant species were authenticated by Taxonomist, Department of Botany, Sunrise University

#### Extraction

The fresh roots were collected, cleaned, shade dried at room temperature for a week and powdered using mixer grinder. 25 gms of the coarse powder was continuously extracted with 100 ml of methanol (SD Fine, India) for 18 hrs at 60°C using Soxhlet apparatus. The powder was successively extracted with n-Hexane (SD Fine, India). All extracts were filtered through a Whatman no.1 filter paper. Thereafter, the extracts was concentrated using rotary flash evaporator (50°C) to 30ml volume. And

finally 30 ml extracts were concentrated to constant weight in vacuum oven at 30 to 50°C. The evaporated residues with constant weight were stored prior to analysis in dark at 4°C.

Methanolic residue of *Terminalia paniculata* was given name as METP and n-Hexane residue of *Terminalia paniculata* was given name as HETP. Methanolic residue of *Boswellia ogadensis* was given name as MEBO and n-Hexane residue of *Boswellia ogadensis* was given name as HEBO [5].

### ***In vivo* Anti-Inflammatory Activity<sup>[6]</sup>**

#### **Acute Anti-inflammatory Activity (Formalin-induced Paw Oedema in Rats)**

Female Albino rats weighing 180-230g were used for the study. Acute inflammation was induced by injecting formalin (NICE Chemicals, Edappally) (0.1 ml of 1% suspension in 0.9% saline) in sub-plantar region and paw volume was measured 0, 1, 2, 3, 4 and 5 hour, with the help of Plethysmometer

All the treatment compounds were administered 30 minutes, prior to formalin. Acute inflammation was induced in right hind paw. A mark was put on the leg second at the leg at the malleolus region to facilitate the dipping of the leg to the same level at the second and subsequent times.

The initial reading was taken at 0 hour, i.e., immediately after injecting formalin and the procedure was repeated at 1, 2, 3, 4 and 5 hour after formalin injection. The difference between 0 hour reading and one of the subsequent readings provides the actual edema volume at the time. The mean paw volume at different times was calculated and compared with the control and the percentage inhibition was then calculated by using the formula;

$$\text{Percent inhibition} = 100 * (1 - T_t/T_c)$$

Where,  $T_t$  and  $T_c$  are the average increase in paw volume of drug treated and control group respectively.

Group-I: Distilled water was supplied and served as control.

Group-II: Animals received a dose of 10 mg/kg of Diclofenac sodium (NICE Chemicals, Manimala Road,,Edappally) i.p. and served as standard

Group-III: Animals received a dose of 200 mg/kg of METP / MEBO p.o.

Group-IV: Animals received a dose of 400 mg/kg of METP / MEBO p.o.

Group-V: Animals received a dose of 600 mg/kg of METP / MEBO p.o.

Group-VI: Animals received a dose of 200 mg/kg of, HETP / HEBO p.o.

Group-VII: Animals received a dose of 400 mg/kg of, HETP / HEBO p.o.

Group-VIII: Animals received a dose of 600 mg/kg of HETP / HEBO p.o.

#### **Chronic Anti-inflammatory Activity (Formalin Induced Paw Oedema)**

Albino wistar rats weighing 170-230 mg/kg were divided into six groups of six in each group. All these animals were fasted for 18 hours before the beginning of the experiment and water was given ad libitum. In animals of all the groups chronic inflammation was produced by sub plantar injection of 20µl of freshly prepared 2% suspension of formalin in normal saline in right hind paw of rat was used as the oedematogenic agent. Animals were treated with drugs for 6 consecutive days.

The paw volume was measured using a plethysmometer before and 6 days after formalin challenge in each group. The increase in paw volume and percent of inhibition was calculated.

Group-I: Distilled water was supplied and served as control.

Group-II: Animals received a dose of 100 mg/kg of Diclofenac sodium i.p. and served as standard

Group-III: Animals received a dose of 200 mg/kg of METP and MEBO p.o.

Group-IV: Animals received a dose of 400 mg/kg of METP and MEBO p.o. Group-V: Animals received a dose of 600 mg/kg of METP and MEBO p.o. Group-VI: Animals received a dose of 200 mg/kg of, HETP and HEBO p.o. Group-VII: Animals received a dose of 400 mg/kg of, HETP and HEBO p.o. Group-VIII: Animals received a dose of 600 mg/kg of, HETP and HEBO p.o.

## RESULTS

### Acute Anti-inflammatory Activity (Formalin-induced paw edema in Rats)

All the test compounds (METP, HETP, MEBO and HEBO) were tested with the diclofenac sodium (10 mg/kg) as a standard for the anti-inflammatory activity. Presently diclofenac showed significant 84.38 % inhibition of inflammation at 5<sup>th</sup> hour ( $0.20 \pm 0.018$ ) when compared with control ( $1.28 \pm 0.042$ ). The test compounds showed maximum percentage of inhibition edema at 5<sup>th</sup> hour significantly in respective dose levels of 200, 400 and 600mg/kg for the test compounds of METP and HETP as 69.53%, 77.34 %, 82.03% and 62.50%, 71.09%,

76.56% respectively. The values are tabulated in the Table I. The test compounds showed maximum percentage of inhibition of oedema at 5<sup>th</sup> hour significantly in respective dose levels of 200, 400 and 600mg/kg for the test compounds MEBO and HEBO as 66.41%, 73.44%, 83.59% and 50.78%, 64.84%, 78.91% resp. The values are tabulated in the Table II.

### Chronic Anti-inflammatory Activity for *Terminalia paniculata* Formalin-induced paw Oedema in Rats

Formalin induced paw oedema is one of the most suitable test procedure to screen chronic anti-inflammatory agents. The results obtained as mean increase in paw volume (ml) and % inhibition are represented in Table III and IV. The mean response of standard was 89.73% inhibition of increase in paw thickness after 6 days. In this model at 200, 400 and 600 mg/kg dose levels of METP and HETP extracts showed 37.05%, 54.02%, 75.45%, and 25.45%, 38.84%, 68.30% inhibition of increase in paw thickness after 6 days. The values are tabulated in table no.3. 200, 400 and 600 mg/kg dose levels of MEBO and HEBO extracts showed 43.30%, 67.41%, 81.25%, and 29.46%, 46.88%, 72.32% inhibition of increase in paw thickness after 6 days. The values are tabulated in table no.4. All the results were compared with solvent control and diclofenac sodium reference drug control.

**Table I: Effect of METP and HETP on Formalin-induced paw Oedema in Rats (Acute Model)**

Groups	Treatment	Paw Oedema Volume (hr)						
		0 hr	1hr	2hr	3hr	4 hr	5 hr	% Inhibition
Group-I	Saline	0.18 ± 0.021	0.78 ± 0.064	1.09 ± 0.037	1.14 ± 0.063	1.19 ± 0.038	1.28 ± 0.042	-
Group-II	Diclofenac sodium (10mg/kg i.p.)	0.19 ± 0.035	0.34 ± 0.028***	0.57 ± 0.046***	0.32 ± 0.024***	0.23 ± 0.047***	0.20 ± 0.018***	84.38%
Group-III	METP (200mg/kg p.o.)	0.16 ± 0.025	0.62 ± 0.043**	0.89 ± 0.037**	1.02 ± 0.041***	0.79 ± 0.056***	0.39 ± 0.023***	69.53%
Group-IV	METP (400mg/kg p.o.)	0.17 ± 0.027	0.56 ± 0.040**	0.78 ± 0.054***	0.53 ± 0.028***	0.40 ± 0.037***	0.29 ± 0.021***	77.34%
Group-V	METP (600mg/kg p.o.)	0.18 ± 0.039	0.45 ± 0.033***	0.69 ± 0.047***	0.43 ± 0.026***	0.31 ± 0.034***	0.23 ± 0.028***	82.03%
Group-VI	HETP (200mg/kg p.o.)	0.16 ± 0.022	0.70 ± 0.045 <sup>ns</sup>	0.99 ± 0.052 <sup>ns</sup>	1.12± 0.053*	0.94 ± 0.038***	0.48 ± 0.041***	62.50%
Group-VII	HETP (400mg/kg p.o.)	0.17 ± 0.020	0.61 ± 0.038**	0.81 ± 0.043***	0.67 ± 0.029***	0.54 ± 0.032***	0.37 ± 0.022***	71.09%
Group-VIII	HETP (600mg/kg p.o.)	0.18 ± 0.034	0.52 ± 0.043***	0.76 ± 0.038***	0.53 ± 0.036***	0.39 ± 0.025***	0.30 ± 0.019***	76.56%

Values are Mean ± SEM (n=6) one way ANOVA followed by Tukey-Karmer's test. Where, \*\*\* P<0.001, \*\* P<0.01, \* P<0.05 and ns: Not significant. METP- Methanolic extract of *Terminalia paniculata*, HETP: n-Hexane extract of *Terminalia paniculata*

**Table II: Effect of MEBO and HEBO on Formalin-induced paw oedema in Rats (Acute Model)**

Groups	Treatment	Paw Oedema Volume (hr)						
		0 hr	1hr	2hr	3hr	4 hr	5 hr	% Inhibition
Group-I	Saline	0.18 ± 0.021	0.78 ± 0.064	1.09 ± 0.037	1.14 ± 0.063	1.19 ± 0.038	1.28 ± 0.042	-
Group-II	Diclofenac sodium (10mg/kg i.p.)	0.19 ± 0.035	0.34 ± 0.028***	0.57 ± 0.046***	0.32 ± 0.024***	0.23 ± 0.047***	0.20 ± 0.018***	84.38%
Group-III	MEBO (200mg/kg p.o.)	0.20 ± 0.032	0.67 ± 0.032*	0.92 ± 0.026*	1.110 ± 0.054**	0.83 ± 0.049***	0.43 ± 0.031***	66.41%
Group-IV	MEBO (400mg/kg p.o.)	0.19 ± 0.047	0.61 ± 0.037**	0.85 ± 0.043**	0.68 ± 0.035***	0.47 ± 0.020***	0.34 ± 0.028 ***	73.44%
Group-V	MEBO (600mg/kg p.o.)	0.18 ± 0.028	0.53 ± 0.030***	0.72 ± 0.032***	0.47 ± 0.022***	0.29 ± 0.014***	0.21 ± 0.017***	83.59%
Group-VI	HEBO (200mg/kg p.o.)	0.18 ± 0.019	0.73 ± 0.036 <sup>ns</sup>	1.02 ± 0.043 <sup>ns</sup>	1.13 ± 0.046 <sup>ns</sup>	0.98 ± 0.034***	0.63 ± 0.025***	50.78%
Group-VII	HEBO (400mg/kg p.o.)	0.19 ± 0.029	0.68 ± 0.031*	0.84 ± 0.036**	0.73 ± 0.021***	0.59 ± 0.027***	0.45 ± 0.018***	64.84%
Group-VIII	HEBO (600mg/kg p.o.)	0.16 ± 0.036	0.59 ± 0.034**	0.78 ± 0.027***	0.60 ± 0.032***	0.41 ± 0.019***	0.27 ± 0.012***	78.91%

Values are Mean  $\pm$  SEM (n=6) one way ANOVA followed by Tukey-Karmer's test. Where, \*\*\* P<0.001, \*\* P<0.01, \* P<0.05 and ns: Not significant. MEBO- Methanolic extract of *Boswellia ogadensis*, HEBO: n-Hexane extract of *Boswellia ogadensis*.

**Table III: Effect of METP and HETP on Formalin-induced paw Oedema in Rats (Chronic Model)**

Groups	Treatment	Initial Paw Volume	Paw Volume After 6 Days	Increase in Paw Volume	% of Inhibition
Group-I	Saline	1.32 $\pm$ 0.045	3.56 $\pm$ 0.212	2.24 $\pm$ 0.143	-
Group-II	Diclofenac sodium (100 mg/kg i.p.)	1.29 $\pm$ 0.082	1.52 $\pm$ 0.092	0.23 $\pm$ 0.065	89.73%
Group-III	METP (200mg/kg p.o.)	1.30 $\pm$ 0.063	2.71 $\pm$ 0.241	1.41 $\pm$ 0.132	37.05%
Group-IV	METP (400mg/kg p.o.)	1.25 $\pm$ 0.068	2.28 $\pm$ 0.195	1.03 $\pm$ 0.178	54.02%
Group-V	METP (600mg/kg p.o.)	1.28 $\pm$ 0.047	1.83 $\pm$ 0.180	0.55 $\pm$ 0.113	75.45%
Group-VI	HETP (200mg/kg p.o.)	1.31 $\pm$ 0.050	2.98 $\pm$ 0.291	1.67 $\pm$ 0.186	25.45%
Group-VII	HETP (400mg/kg p.o.)	1.27 $\pm$ 0.039	2.64 $\pm$ 0.214	1.37 $\pm$ 0.109	38.84%
Group-VIII	HETP (600mg/kg p.o.)	1.26 $\pm$ 0.043	1.97 $\pm$ 0.173	0.71 $\pm$ 0.102	68.30%

Results are expressed on mean + SEM from four observations Paw Volume was measured after 6 days. METP- Methanolic extract of *Terminalia paniculata*, HETP: n-Hexane extract of *Terminalia paniculata*

**Table IV: Effect of MEBO and HEBO on Formalin-induced paw oedema in Rats (Chronic Model)**

Groups	Treatment	Initial Paw Volume	Paw Volume After 6 Days	Increase in Paw Volume	% of Inhibition
Group-I	Saline	1.32 $\pm$ 0.045	3.56 $\pm$ 0.212	2.24 $\pm$ 0.143	-
Group-II	Diclofenac sodium (100 mg/kg i.p.)	1.29 $\pm$ 0.082	1.52 $\pm$ 0.092	0.23 $\pm$ 0.065	89.73%
Group-III	MEBO (200mg/kg p.o.)	1.32 $\pm$ 0.043	2.59 $\pm$ 0.305	1.27 $\pm$ 0.262	43.30%
Group-IV	MEBO (400mg/kg p.o.)	1.28 $\pm$ 0.077	2.01 $\pm$ 0.215	0.73 $\pm$ 0.138	67.41%
Group-V	MEBO (600mg/kg p.o.)	1.26 $\pm$ 0.093	1.68 $\pm$ 0.146	0.42 $\pm$ 0.053	81.25%
Group-VI	HEBO (200mg/kg p.o.)	1.29 $\pm$ 0.062	2.87 $\pm$ 0.297	1.58 $\pm$ 0.235	29.46%
Group-VII	HEBO (400mg/kg p.o.)	1.30 $\pm$ 0.048	2.49 $\pm$ 0.234	1.19 $\pm$ 0.186	46.88%
Group-VIII	HEBO (600mg/kg p.o.)	1.31 $\pm$ 0.051	1.93 $\pm$ 0.172	0.62 $\pm$ 0.121	72.32%

Results are expressed on mean + SEM from four observations Paw Volume was measured after 6 days. MEBO- Methanolic extract of *Boswellia ogadensis*, HEBO: n-Hexane extract of *Boswellia ogadensis*.

## Discussion

It is well known that inhibition of formalin-induced pedal oedema in rats is one of the most suitable test procedures to

screen anti-arthritic and anti-inflammatory agents as it closely resembles human arthritis. Injection of formalin subcutaneously into hind paw of rats produces localized inflammation and pain. The nociceptive effect of formalin is biphasic, an early neurogenic component followed by a later tissue mediated response. Thus formalin-induced arthritis is a model used for the evaluation of an agent

with probable anti-proliferative activity.<sup>[7]</sup> This experiment is associated with the proliferative phase of inflammation. Results with *Terminalia paniculata* and *Boswellia ogadensis* of METP, HETP, MEBO and HEBO (600mg/kg, p.o.) are showed quite compatible with those of the standard drug diclofenac sodium. Therefore, the drug appears to be effective against formalin-induced arthritis. Formalin induced paw oedema is one of the most suitable test procedure to screen chronic anti-inflammatory agents. The mean response of standard 85.02% was inhibition of increase in paw thickness after 6 days. In this model at 200, 400 and 600 mg/kg dose level of METP, HETP, MEBO and HEBO extracts showed significantly inhibition of increase in paw thickness after 6 days

### Conclusion

The effect of methanolic and n-Hexane extracts of roots of *Terminalia paniculata* Brandis and *Boswellia ogadensis* significantly in 200, 400 and 600 mg/kg p.o dosage, showed anti-inflammatory activity (acute and chronic models).

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