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Evaluation of Anti-Inflammatory Properties of *Saurauia Roxburghii* Leaf Extract in Rats: Insights from an *In-Vivo* Study

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ABSTRACT:

Inflammation is a natural defense mechanism involving physiological changes that help minimize tissue damage and eliminate pathogenic infections. The ethanolic extract from the leaves of Saurauia roxburghii was evaluated for its anti-inflammatory effects in vivo. The adverse side effects associated with conventional synthetic drugs used to treat acute inflammatory conditions highlight the need for developing new therapeutic agents with reduced toxicity and improved safety profiles. Preliminary phytochemical analysis revealed the presence of various bioactive compounds including alkaloids, saponins, tannins, flavonoids, glycosides, phenolics, terpenoids, and volatile oils. The extract's anti-inflammatory activity was assessed using a carrageenan-induced paw edema model in rats at oral doses of 200 mg/kg and 400 mg/kg, with indomethacin (10 mg/kg) used as a standard reference. Paw volume variations were measured with the help of a mercury column plethysmometer. The findings revealed a statistically significant decrease in inflammation ($p<0.05$), with the 400 mg/kg dose producing a 37.94% reduction in paw edema by the 5th hour, closely comparable to the 39.48% inhibition observed in the indomethacin-treated group. The results of the study validate the potential of this plant for managing conditions related to inflammatory disorders.

Keywords: Inflammatory activity, Plethysmometer, Indomethacin, leaves of S. roxburghii

1. Introduction

The world possesses a vast wealth of medicinal plants, which have historically formed the cornerstone of traditional healthcare practices in India. Inflammation represents a fundamental immune reaction triggered by infection or injury, essential for defending the body by removing harmful substances and promoting the repair of damaged tissues to restore normal function. It is astronomically distributed into two forms: acute and chronic.

Inflammation is driven by a complex cascade of biological events, with arachidonic acid metabolism playing a central role. This fatty acid can be processed via the cyclooxygenase (COX) pathway to produce prostaglandins and thromboxane A2, or through the 5-lipoxygenase (5-LOX) pathway to generate hydroperoxyl-eicosatetraenoic acids (HPETEs) and leukotrienes—both of which act as crucial intercessors in seditious responses. When neutrophils are activated, arachidonic acid is released from membrane phospholipids and subsequently transformed into leukotrienes and prostaglandins via the 5-LOX and COX pathways, respectively. Blocking these enzymes reduces the formation of these inflammatory mediators, suggesting that such inhibition could offer anti-inflammatory and pain-relieving benefits, potentially with fewer gastrointestinal side effects. Additionally, reactive oxygen species (ROS), generated during leukocyte activation, play a significant role in the inflammation process.

Inflammation is a protective response of the body aimed at minimizing tissue injury and eliminating harmful pathogens. It involves a series of physiological adaptations designed to control damage and initiate healing. Inflammatory diseases significantly contribute to human illness and death. This process represents a core pathological response, encompassing a complex interplay of cellular and chemical reactions within affected blood vessels and surrounding connective tissues. These reactions are triggered by injury or abnormal stimuli—whether physical, chemical, or biological in nature—and result in:

1. Structural changes at the site of inflammation due to localized responses.
2. Elimination or neutralization of the harmful agents.
3. Activation of mechanisms that promote tissue repair and recovery. These processes are marked by the classic signs of inflammation: redness (rubor), heat (calor), swelling (tumor), and pain (dolor).

Saurauia roxburghii, commonly known as Eastern Gogan, is a large shrub or small evergreen tree that typically grows between 4 to 10 meters in height. It belongs to the Actinidiaceae family, which also includes the kiwifruit. Native to regions extending from Peninsular Malaysia to eastern Nepal, this

species is distinguished by its broad, elliptical leaves that are densely covered with rusty hairs on the underside, and its clusters of pink blossoms that bloom loosely. Eastern Gogan is widely found across several regions of India, especially in states such as Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland, Sikkim, Tripura, and West Bengal. Beyond India, its natural range extends to multiple countries including Bangladesh, Bhutan, Laos, Malaysia, Myanmar, Nepal, Sri Lanka, and Thailand.

2. Material and Methods

Plant selection

The selection of this plant was influenced by its established use in traditional medicine. Furthermore, the presence of phytochemicals in the ethanolic extract of *Saurauia roxburghii* suggested possible analgesic and anti-inflammatory effects, reinforcing the decision to utilize its leaves for the proposed study.

Collection, identification and authentication of Plant material-

The plant sample was obtained from a local plant vendor in Assam and was taxonomically identified as *Saurauia roxburghii* (family: Actinidiaceae) by Dr. S.N. Dwivedi, Professor of Botany at Janata PG College, under the academic affiliation of APS University, Rewa (Madhya Pradesh). The authenticated specimen was submitted and recorded under the voucher number**.**

Preparation of extracts

The extraction was performed using a Soxhlet apparatus. Finely powdered, air-dried leaves of *Saurauia roxburghii* underwent a 24-hour hot extraction process (soxhlation) using ethanol as the solvent, maintained at approximately 60°C. The resulting extract was then concentrated by distillation in a porcelain evaporating dish and further dried over a boiling water bath, yielding a brown, semi-solid residue.

Phytochemical screening

The ethanolic leaf extract of *Saurauia roxburghii* was subjected to qualitative analysis to identify the presence of key phytochemical groups, including alkaloids, saponins, tannins, flavonoids, glycosides, phenolic compounds, terpenoids, and volatile oils.

Experimental Animals

Wistar rats, weighing between 150–200 grams, were procured from the animal facility of Swami Vivekanand College of Pharmacy, Indore, India. The animals were housed individually in clean, spacious cages maintained at a controlled temperature of 22ºC ± 3ºC, under a 12-hour light and 12-hour dark cycle. They were provided unrestricted access to standard food and water. Before beginning the experimental procedures, the rats were given a one-week acclimatization period. All experimental procedures were conducted following approval from the Institutional Animal Ethics Committee (Approval No: IAEC/SVCP/2024/02), adhering strictly to the guidelines established by the CCSEA.

Acute oral toxicity

According to literature ethanolic extract of leaves of Saurauia roxburghii was found safe at 300 mg/kg and 500 mg/kg body weight, no mortality was observed at both doses and therefore, LD_{50} of ethanolic extract of plant was reported 2g/kg body weight.

Evaluation of Anti-inflammatory activity and analgesic

The anti-inflammatory activity of the extract was evaluated through the carrageenan-induced paw edema method in rats, a well- established experimental model for assessing acute inflammation.

3. Experimental design

A total of 20 healthy adult albino rats of the Wistar strain were randomly assigned into 5 separate groups.

Table 1: Grouping of experimental animals

S.No.	Groups	No. of Animals
1.	Normal Control	2
2.	Negative Control	3
3.	Low dose of test extract (200 mg/kg)	5
4.	High dose of test extract (400 mg/kg)	5
5.	Standard Drug Indomethacin (10 mg/kg)	5
	Total	20

Procedure:

Figure 1: Carrageenan Administration in Rat

Carrageenan induced-paw edema method for anti-inflammatory activity

Healthy Wistar rats were individually weighed and marked with ink just above the tibio-tarsal joint on both hind limbs for consistent reference. Baseline paw volumes were measured using the mercury displacement method. Except for the normal control group, all other rats were given oral doses of either the test extract or the standard drug before inflammation was induced. The 1st group, designated as the normal control, received only normal saline orally and was not subjected to any inflammatory agent. The 2nd group, serving as the negative control, had inflammation induced by a sub-plantar injection of 1% carrageenan solution into the right hind paw. For the 3rd group, rats were pre-treated orally with a lower dose (200 mg/kg) of *Saurauia roxbughii* extract, followed an hour later by a carrageenan injection in the same manner. The 4th group received a higher dose (400 mg/kg) of the extract and was also injected with carrageenan after an hour. The 5th group acted as the standard treatment group and was pre-treated with indomethacin at a dose of 10 mg/kg. An hour later, carrageenan was administered into sub-plantar region to induce inflammation in the right hind paw.

Paw edema volume was recorded using a mercury plethysmometer at specific time intervals—immediately after carrageenan injection and then at 1, 2, 3, 4, and5 hours post-injection. The mean paw volume was calculated for each group and compared against both the control and standard treatment groups. A noticeable decrease in paw swelling in the groups pre-treated with *Saurauia roxburghii* extract, relative to the untreated control group, was interpreted as evidence of anti-inflammatory activity.

Statistical analysis

The results were expressed as mean ± standard error of the mean (S.E.M.). Statistical evaluation was performed using one-way analysis of variance (ANOVA) followed by Tukey's HSD post-hoc test. Comparisons were made considered to indicate statistical significance across all analyses.

4. Results

Extraction

The yield of the ethanolic extract of S. roxburghii leaves was 27.32% w/w.

Preliminary phytochemical screening of the leaf extract of *Saurauia roxburghii* was conducted to identify the presence of various bioactive compounds:

The ethanolic leaf extract of *Saurauia roxburghii* underwent qualitative phytochemical evaluation, which indicated the presence of numerous biologically active compounds such as alkaloids, flavonoids, glycosides, essential oils, saponins, tannins, and diverse phenolic substances.

Anti-inflammatory response

Reduction of paw edema in rats following carrageenan-induced inflammation.

The in-vivo anti-inflammatory effects of the ethanolic extract of *Saurauia roxburghii leaves* were assessed and compared to the standard drug, Indomethacin, and the control group over a five-hour period following the induction of inflammation. The results were presented as mean ± standard deviation, showing the inflammation index and the percentage of edema inhibition in the rat paw, as outlined in below tables. It was observed that the high dose (400 mg/kg) of S. roxburghii extract exhibited a more pronounced anti-inflammatory effect over time compared to the lower dose (200 mg/kg). Rats were pre-treated with varying doses of the ethanolic extract or Indomethacin, and after one hour, edema was induced by administering a carrageenan solution. The anti-inflammatory efficacy revealed that at the 5th hour, the percentage inhibition of paw edema in Group-III (low dose 200 mg/kg) was 31.79%, while Group-IV (high dose 400 mg/kg) showed a 37.94% reduction. This was closely compared to Group-V (Indomethacin 10 mg/kg), which exhibited a 39.48% reduction in paw edema at the same time point. The inhibition of paw edema with the 400 mg/kg dose was statistically significant

($p<0.05$) when compared to the Indomethacin-treated group at all time intervals (1, 2, 3, 4, and 5 hours). The maximum reduction in paw volume observed with the 400 mg/kg dose was 37.94%, which was nearly equivalent to the effect of Indomethacin (39.48%) at the 5th hour.

Calculation of Paw Edema Inhibition Percentage Using Ethanolic Extract and Standard Drug.

The extent to which paw edema was inhibited by both the ethanolic extract and the reference drug was determined using the following formula.

Percentage Inhibition of Paw Edema was calculated using the formula:

Inhibition (%) = (Oc − Ot) / Oc × 100.

where **Oc** represents the paw edema volume in the control group, and **Ot** denotes the paw edema volume in the treated groups.

Table 2- Rat Paw Edema Inhibition (%) Induced by Ethanolic Extract of S. roxburghii and Standard Anti-inflammatory Agent (Indomethacin).

			% of inhibition of paw edema				
Groups	**Treatment**	**Dose**	**At 1st Hour**	**At 2nd Hour**	**At 3rd Hour**	**At 4th Hour**	**At 5th Hour**
Group-II	Carrageenan	1% Carrageenan	0	0	0	0	0
Group-III	Ethanolic extract	200 mg/kg	6.74	13.25	17.20	23.56	31.79
Group-IV	Ethanolic extract	400 mg/kg	89.55	15.46	24.19	30.89	37.94
Group-V	Indomethacin	10 mg/kg	9.55	16.57	25.26	33.50	39.48

Note: Data indicate % inhibition of paw edema in rats, compared to the standard group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post hoc test, with significance set at $p < 0.05$ versus the control.

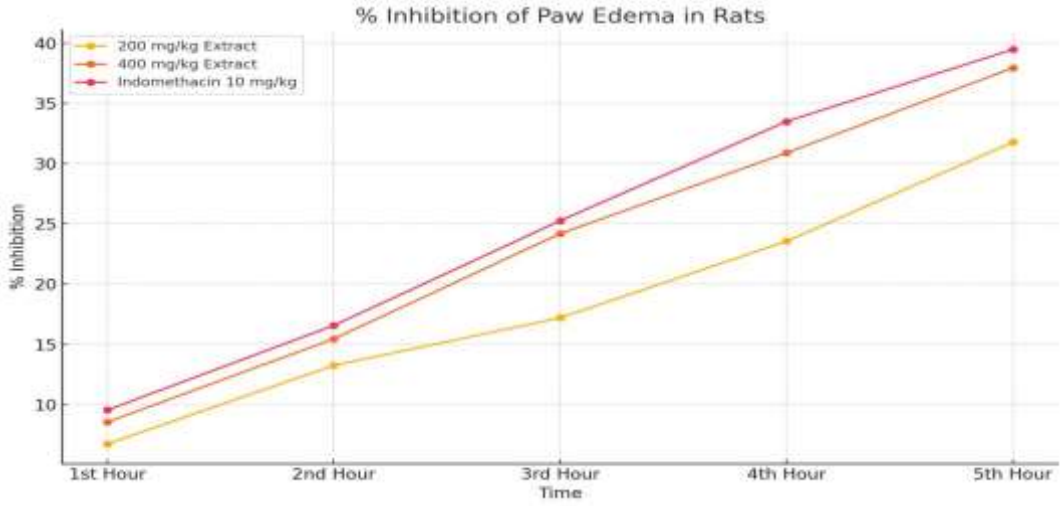

Figure 2: Comparative % inhibition of paw edema in rats at distinct time points after treatment with *S. roxburghii* ethanolic extract, relative to control.

5. Discussion

Phytochemicals are naturally occurring bioactive compounds found in a wide range of plant parts, including medicinal herbs, fruits, vegetables, flowers, leaves, roots, and fibers. These compounds serve as the plant's natural defense system, offering protection against diseases. In the current investigation, the ethanolic extract of *Saurauia roxburghii leaves* was evaluated for its anti-inflammatory effects through in vivo models, specifically using carrageenan-induced paw edema in rats. This investigation was rooted in traditional medicinal claims suggesting the therapeutic potential of the plant. Extraction was performed using ethanol as the solvent in a Soxhlet apparatus, yielding 27.32% of crude extract from the powdered leaves. Preliminary phytochemical screening revealed the presence of several bioactive compounds, including alkaloids, were identified using standard laboratory reagents. To assess anti-inflammatory activity, the acute paw edema model induced by carrageenan was employed. Carrageenan triggers inflammation by promoting the accumulation of protein-rich exudate and infiltration of neutrophils. The paw volume was measured using a mercury plethysmometer at specified intervals. The extract, at both 200 mg/kg and 400 mg/kg doses, demonstrated a dose-dependent reduction in paw edema, with results being comparable to those observed in the Indomethacin-treated group (10 mg/kg). These findings confirm that the ethanolic extract of *S. roxburghii leaves* possesses significant anti-inflammatory properties. The ethanolic extract administered at a dose of 400 mg/kg produced a significant reduction ($p<0.05$) in paw edema in rats. At the 4th and 5th hour, this dose resulted in 30.89% and 37.94% inhibition of inflammation, respectively. In comparison, rats treated with Indomethacin at 10 mg/kg exhibited 33.50% and 39.48% inhibition during the same time points in the carrageenan-induced paw edema model.

6. Conclusion

The current investigation demonstrated that the ethanolic extract of *S. roxburghii leaves* possesses notable anti-inflammatory properties. These effects are likely associated with the presence of various bioactive phytochemicals including alkaloids, saponins, tannins, flavonoids, glycosides, and phenolic compounds identified within the extract. The carrageenan-induced paw edema model in rats remains one of the most extensively employed and reliable methods for assessing anti-inflammatory activity.

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