

Investigation of the Anti-Inflammatory Activity of *Barringtonia asiatica* (Lecythidaceae) Ethanolic Leaf Extract in Albino Mice

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Abstract: The leaves of *Barringtonia asiatica* (Lecythidaceae) have long been employed in traditional medicine for their therapeutic benefits, including anti-inflammatory properties. This study evaluated the anti-inflammatory effects of ethanolic leaf extracts of *B. asiatica* in vivo using carrageenan-induced paw edema in Swiss albino mice. Mice were administered extract concentrations of 25%, 50%, and 75%, while diclofenac sodium served as a positive control. Phytochemical analysis revealed the presence of flavonoids, saponins, and other bioactive constituents. The 75% extract demonstrated significant anti-inflammatory effects, comparable to diclofenac. Acute oral toxicity testing confirmed the extract's safety. These findings support the traditional medicinal use of *B. asiatica* and its potential as a safe, plant-based anti-inflammatory agent.

Keywords: *Barringtonia asiatica*, Anti-Inflammatory, Ethanolic Extract, Carrageenan, Paw Edema, Phytochemicals.

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I. INTRODUCTION

Inflammation is a fundamental biological defense mechanism triggered by infection, tissue damage, or irritants. While acute inflammation is beneficial, persistent or chronic inflammation can contribute to the development of numerous diseases, including rheumatoid arthritis, cardiovascular disorders, and metabolic syndromes. Nonsteroidal anti-inflammatory drugs (NSAIDs) such as diclofenac sodium are commonly prescribed to alleviate inflammation, yet their use is frequently limited due to adverse effects such as gastrointestinal disturbances and hepatotoxicity (Chippada et al. 2011).

Barringtonia asiatica, also called the Fish Poison Tree and known locally as "Botong" in the Philippines, is a tropical coastal tree from the Lecythidaceae family. It has been utilized in folk medicine for its anti-inflammatory, antimicrobial, and wound-healing properties. The leaves of *B. asiatica* contain bioactive compounds such as saponins,

flavonoids, and polyphenols, which are believed to contribute to its therapeutic potential (Lee et al. 2019).

However, the scientific validation of its anti-inflammatory potential, particularly using ethanolic leaf extracts, remains insufficient.

The literature survey revealed that there are no scientific studies carried out regarding the anti-inflammatory activity of the leaves of *B. asiatica*. Hence, this study is focused to investigate the anti-inflammatory potentials of *B. asiatica* ethanolic using the carrageenan-induced paw edema model in Swiss albino mice. The findings aimed to provide empirical evidence supporting the plant's ethnopharmacological claims and contribute to the discovery of safer plant-derived anti-inflammatory agents.

II. MATERIALS AND METHODS

➤ Plant Material Collection and Identification

Fresh leaves of *B. asiatica* were harvested from Sta. Filomena, Iligan City, Philippines, in December

2024. Specimens were authenticated by botanists from the Mindanao State University - Iligan Institute of Technology.

➤ Preparation of Ethanolic Leaf Extract

Collected leaves were washed, air-dried at ambient temperature ($22 \pm 3^\circ\text{C}$) for 3-4 weeks, pulverized, and sieved. The powdered leaves were macerated in 90% ethanol (1:5 ratio) for 72 hours using an orbital shaker, followed by filtration and concentration with a rotary evaporator at 40°C . The resulting crude extract was stored at 4°C .

➤ Phytochemical Screening

Qualitative tests were performed to detect the presence of alkaloids, flavonoids, tannins, saponins, phenolics, terpenoids, steroids, and glycosides, following standard procedures.

Table 1 Phytochemical Constituent Concentration Result Present in *B. asiatica*

Phytochemical Constituent	Result
Alkaloids	++
Flavanoids	+++
Phenols	+++
Saponins	+++
Tannins	++
Steroids	+++
Terpenoids	++

The table below showed the result of the Phytochemical screening of the crude extract of *B. asiatica* the result of the following is interpreted as (++) means moderate or good concentration of the Phytochemical constituent, while (+++) meant abundant or strong indicating high concentration of a phytochemical constituent.

➤ Experimental Animals

Healthy male Swiss albino mice (20–25 g) were housed in controlled conditions and acclimatized for seven days. The study protocol adhered to OECD Guideline 423 and was approved by the institutional animal ethics committee.

➤ Acute Oral Toxicity Test

Initially, a single dose of 2000 mg/kg body weight was administered orally, and the animals were observed for clinical signs of toxicity and mortality for a period of 14 days. No mortality or signs of adverse effects were observed.

Subsequently, a higher dose of 5000 mg/kg was administered to a separate group of mice. Similarly, no signs of toxicity, behavioral changes, or mortality were noted throughout the observation period. These results suggest that the extract has a relatively high safety margin and is non-toxic at doses up to 5000 mg/kg.

➤ Induction of Inflammation and Treatment Groups

Paw inflammation was induced by subplantar injection of 0.1 mL of 1% carrageenan into the right hind paw. Mice were assigned to the following groups:

- Negative control (PNSS),
- Positive control (Diclofenac sodium, 10 mg/kg),
- *B. asiatica* extract at 25%, 50%, and 75% concentrations (administered orally).

➤ Measurement of Paw Edema

Paw thickness was measured using a digital vernier caliper at 0, 1, 2, 3, 4, and 5 hours post- carrageenan injection. Percentage inhibition of inflammation was calculated using the formula:

$$\% \text{ Inhibition} = [(V_c - V_t) / V_c] \times 100$$

Where V_c = mean paw thickness of control;

V_t = mean paw thickness of treated group.

➤ Statistical Analysis

Results were expressed as mean \pm SEM. One- way ANOVA followed by Dunnett's test was used to assess statistical significance ($p < 0.05$).

III. RESULTS

➤ Anti-Inflammatory Activity

Table 2 and Figure 1 show the mean paw thickness and percentage inhibition of inflammation among the treatment groups. The 75% *B. asiatica* extract exhibited a significant reduction in paw edema ($p < 0.01$), indicating strong anti-inflammatory activity. Its effect closely approached that of the standard drug, diclofenac sodium. Lower concentrations (25% and 50%) also demonstrated a reduction in inflammation, although the effects were less marked compared to the 75% concentration, supporting a dose-dependent response.

Table 2 Effect of *B. asiatica* Ethanolic Extract on Carrageenan-Induced Paw Edema in Mice

Treatment Group	Paw Thickness at 3 rd Hour (mm)	% Inhibition
Negative Control (PNSS)	0.76 ± 0.03	—
Positive Control (Diclofenac Sodium)	0.33 ± 0.02 **	56.58%
25% <i>B. asiatica</i>	0.61 ± 0.02 *	19.74%
50% <i>B. asiatica</i>	0.52 ± 0.03 **	31.58%
75% <i>B. asiatica</i>	0.37 ± 0.02 **	51.32%

*Significant at $p < 0.05$; **Significant at $p < 0.01$

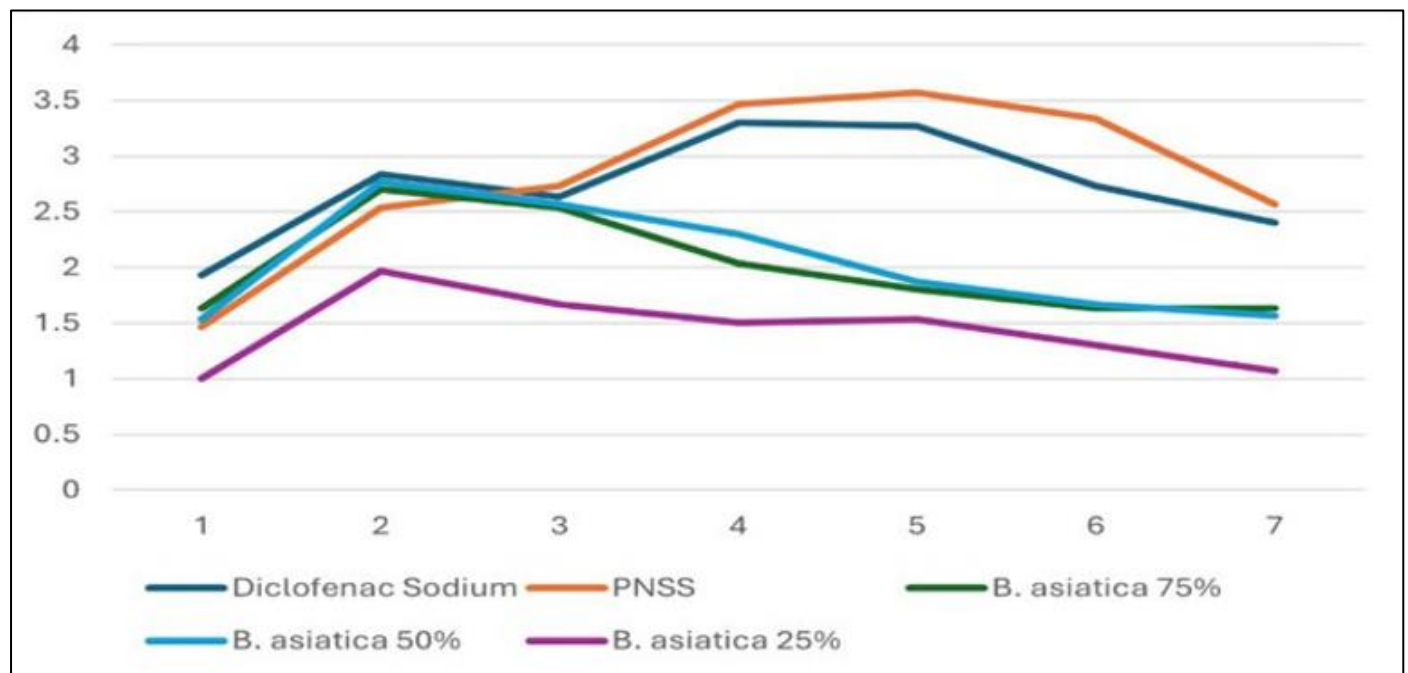


Fig 1 Line Graph Result of *B. asiatica* Ethanolic Leaf Extract (25%, 50%, 75%) and the Control Groups (Positive and Negative)

Figure 1 illustrates the progression and suppression of carrageenan-induced paw edema in Swiss Albino mice treated with different concentrations of *Barringtonia asiatica* ethanolic leaf extract, categorized as low, middle, and high doses, alongside diclofenac sodium (positive control) and PNSS (negative control). Paw volumes were recorded at specific intervals: before carrageenan injection (initial), immediately after (0 hour), and then hourly up to the 5th hour.

The PNSS group showed a sharp increase in paw volume, peaking at the 3rd hour, with minimal reduction afterward, indicating sustained inflammation. The diclofenac group also peaked at the 3rd hour but showed a noticeable decline thereafter. The high dose extract group demonstrated the most significant reduction in inflammation, with paw volume steadily decreasing from the 2nd hour and nearing baseline by the 5th hour. The middle dose followed a similar trend but with a slower decrease, while the low dose group showed only a mild and gradual reduction.

This trend highlights a dose-dependent anti-inflammatory effect, with the high dose performing comparably to the standard drug. The clearest treatment effects were observed between the 2nd and 5th hour, the typical peak of inflammation in this model. The consistent response in the high and middle dose groups also suggests low variability among subjects and supports the potential of *B. asiatica* as a reliable anti-inflammatory agent.

IV. DISCUSSION

The findings of this study support the traditional claims regarding the anti-inflammatory potential of *B. asiatica*. The ethanolic extract exhibited dose-dependent inhibition of inflammation in the carrageenan-induced paw edema model. The 75% concentration was particularly effective, showing nearly equal efficacy to the NSAID diclofenac.

The mechanism is likely multifactorial. Flavonoids, saponins, and phenolic compounds are known to suppress

inflammatory mediators such as prostaglandins, histamine, and serotonin. The extract's capacity to reduce paw volume in both the early (0–2 hours) and late (3–5 hours) phases of inflammation suggests interference with multiple inflammatory pathways.

Comparable findings were observed in related species such as *Barringtonia racemosa*, where bartogenic acid-rich fractions significantly reduced inflammatory markers in both acute and chronic models. The phytochemical profile of *B. asiatica* may thus offer similar bioactivities.

V. CONCLUSION

The study demonstrates that *Barringtonia asiatica* ethanolic leaf extract possesses significant anti-inflammatory properties, with the 75% concentration being the most effective. Its favorable safety profile and bioactive content make it a promising candidate for further drug development and phytopharmaceutical research. Isolation of active constituents and mechanistic studies are recommended to further validate these findings.

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