Evaluation of Antiinflamatory Activity of Buddleja Davidii

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***Abstract*—**

Objective: The present study aimed to evaluate the anti-inflammatory activity of *Buddleja davidii* extract using various acute and chronic inflammation models in rats.

Methods: The anti-inflammatory potential of *Buddleja davidii* was assessed at two doses (400 mg/kg and 600 mg/kg) using carrageenan, histamine, dextran, serotonin, and formaldehyde-induced paw oedema models, as well as the cotton pellet-induced granuloma model. Diclofenac sodium (10 mg/kg) served as the standard drug. Biochemical parameters, including total protein, albumin, acid phosphatase (ACP), and alkaline phosphatase (ALP), were also measured.

Results: *Buddleja davidii* significantly reduced paw edema in all models, indicating both early- and late-phase anti-inflammatory activity. The higher dose (600 mg/kg) consistently showed greater efficacy, often comparable to Diclofenac. The extract also demonstrated a marked reduction in granuloma weight in the cotton pellet model, indicating activity against chronic inflammation. Biochemical analysis revealed improved protein levels and reduced ALP activity, suggesting hepatoprotective potential.

Conclusion: The findings suggest that *Buddleja davidii* possesses broad-spectrum anti-inflammatory properties, acting through multiple mechanisms. These results support its traditional medicinal use and highlight its potential as a natural therapeutic agent for treating both acute and chronic inflammatory conditions.

***Keywords*— *Buddleja davidii,* anti-inflammatory, ALP activity, Paw edema.**

I. INTRODUCTION

Inflammation is a normal biological process in response to tissue injury, microbial pathogen infection and chemical irritation. This biological process also involves the innate and adaptive immune systems. At a damaged site, inflammation is initiated by migration of immune cells from blood vessels and release of mediators, followed by recruitment of inflammatory cells and release of reactive oxygen species (ROS), reactive nitrogen species (RNS) and proinflammatory cytokines to eliminate foreign pathogens, resolving infection and repairing injured tissues. [1,2] Thus, the main function of inflammation is beneficial for a host's defence. In general, normal inflammation is rapid and self-limiting, but aberrant resolution and prolonged inflammation causes various chronic disorders [3]. Chronic inflammation can inflict more serious damage to a host tissue than bacterial infection. Diverse ROS and RNS such as O2 (superoxide anion), OH (hydroxyl radical), H2O2 (hydrogen peroxide), nitric oxide (NO), and 1 O2 (singlet oxygen) generated by inflammatory cells injure cellular biomolecules including nucleic acids, proteins and lipids, causing cellular and tissue damage, which in turn augments the state of inflammation [4]. These also trigger a series of signalling molecules, inflammatory gene expression and activation of enzymes involved in chronic inflammation. Inflammatory chemicals produced by inflamed and immune cells also attack normal tissues surrounding the infected tissue, causing oxidative damage and extensive tissue inflammation [1,4] Studies show that chronic inflammation is linked to a wide range of progressive diseases, including cancer, neurological disease, metabolic disorder and cardiovascular disease [3,4] Numbers of studies suggest elimination of chronic inflammation as a

major way to prevent various chronic diseases [1,3] Epidemiological studies provide convincing evidence that natural dietary compounds that humans consume as food possess many biological activities. Among these natural bioactive compounds, flavonoids are widely recognized for their biological and pharmacological effects, including antiviral, anti-carcinogenic, antioxidant, antimicrobial, anti-inflammatory, anti-angiogenic and anti-thrombogenic properties[1,5]. Epidemiologic studies indicate that the incidence of chronic disease and cancer is inversely correlated with the consumption of fruits and vegetables rich in flavonoids,5,6 and this is attributed to their possible anti-inflammatory activities.

The role of inflammation in human disease

The role of inflammation in human disease Inflammation is a complicated process, driven by pre-existing conditions (infection or injury) or genetic changes, that results in triggering signaling cascades, activation of transcription factors, gene expression, increased of levels of inflammatory enzymes, and release of various oxidants and proinflammatory molecules in immune or inflammatory cells [2] In this condition, excessive oxidants and inflammatory mediators have a harmful effect on normal tissue, including toxicity, loss of barrier function, abnormal cell proliferation, inhibiting normal function of tissues and organs, and finally leading to systemic disorders [1,2] Over the past few decades, many studies reveal that chronic inflammation is a critical component in many human diseases and conditions, including obesity, cardiovascular diseases, neurodegenerative diseases, diabetes, aging, and cancers [2,4].

Inflammation is a natural defense mechanism of the body triggered in response to injury, infection, toxins, or other harmful stimuli, and although it is essential for eliminating pathogens and initiating tissue repair, persistent or uncontrolled inflammation contributes to the development of chronic disorders such as arthritis, cardiovascular diseases, diabetes, cancer, and neurodegenerative conditions. Anti-inflammatory agents act by suppressing or modulating the inflammatory response through various mechanisms, including inhibition of cyclooxygenase (COX) and lipoxygenase (LOX) enzymes to reduce prostaglandin and leukotriene synthesis, suppression of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-6, scavenging of reactive oxygen species to reduce oxidative stress, stabilization of lysosomal membranes to prevent proteolytic enzyme release, and regulation of transcription factors like NF-κB that control inflammation-related genes. Synthetic agents such as non-steroidal anti-inflammatory drugs (NSAIDs) and corticosteroids are widely used but often associated with side effects including gastrointestinal irritation, renal impairment, immunosuppression, and metabolic disturbances, which has driven the search for safer alternatives such as monoclonal antibodies targeting specific cytokines (e.g., infliximab, tocilizumab) and natural phytochemicals like flavonoids, alkaloids, terpenoids, and polyphenols that exhibit strong antioxidant and anti-inflammatory properties. Experimental evaluation of anti-inflammatory activity commonly employs animal models such as carrageenan-induced paw edema for acute inflammation, cotton pellet granuloma for chronic inflammation, croton oil-induced ear edema for topical inflammation, and biochemical assessments of oxidative stress markers and cytokine levels to validate efficacy. Overall, anti-inflammatory agents remain a cornerstone in the management of both acute and chronic inflammatory conditions, and ongoing research into natural compounds and targeted biologics offers promising opportunities for the development of safer and more effective therapies [5-7].

II. MATERIALS AND METHODS

COLLECTION AND IDENTIFICATION OF PLANT MATERIAL

The fresh Buddleja Davidii are collected and identified. Collection of Plant Materials Dried was purchased from an herbal Market of Hyderabad, Telangana, India, and authenticated by Dr. K. Madhava Chetty, Assistant Professor, Department of Botany, S.V University, Tirupati. At the Department of Pharmacology, at our institution.

Preparation of the Extract

The collected plant is washed, air dried, homogenized to fine powder and stored in airtight bottles. The dried powder will first be defatted with methanol by using the Soxhlet apparatus.

Extraction of plant material

The preserved and pulverized plant material was utilized in the extraction process. A metered amount

of each pulverized plant material was subjected to Soxhlet apparatus, with one intermediate heating at 40°C per day. The residues were subsequently extracted. Following filtration through Whatmann filter paper, the filtrate was concentrated at a controlled temperature and reduced pressure (40-50°C). After being desiccated, it was weighed.

III. ANTI-INFLAMMATORY STUDIES

Animal grouping for anti-inflammatory studies
The animals were divided into four groups (six animals in each group) for anti-inflammatory studies.
Group I: Vehicle treated control (distilled water)
Group II: Methanol extract of Buddleja Davidii - 400 mg/kg body weight (Buddleja Davidii-400)
Group III: Methanol extract of Buddleja Davidii - 600 mg/kg body weight (Buddleja Davidii-600)
Group IV: Diclofenac sodium- 10 mg/kg body weight (diclofenac- 10)

Carrageenan induced rat paw edema [8]
Carrageenan induced rat paw edema was done by the method of Winter et al. (1962). Inflammation was induced by injection of 0.1 ml of freshly prepared carrageenan (1%) aqueous suspension in normal saline underneath the plantar tissue of the right hind paw of rats. The different groups of rats were administered with BUDDLEJA DAVIDII (400 and 600 mg/kg, p.o.) and diclofenac (10 mg/kg, p.o.). The control group received vehicle (distilled water, 10 ml/kg, p.o.). 1 h after drug treatment, paw edema was induced by the injection of carrageenan (an edematogenic agent). The paw volume was measured by a Plethysmometer. The measures were determined at 0 h (V_0: before edematogenic agent injection) and 1,2,3,4 and 5h intervals later (V_t). The difference between V_t and V_0 was taken as the edema value. The percentage of inhibition was calculated according to the following formula:

$$\%\ of\ inhibition = \frac{(V_t - V_0)_{control} - (V_t - V_0)_{treated}}{(V_t - V_0)_{control}}$$

Histamine induced rat paw edema [9]
Inflammation was induced by injection of 0.1 ml of freshly prepared histamine (1%) aqueous suspension in normal saline underneath the plantar tissue of the right hind paw of rats. The drug treatment and paw volume were measured in a similar manner to that of carrageenan induced paw edema model.

Dextran induced rat paw edema [10
Inflammation was induced by injection of 0.1 ml of freshly prepared dextran (1%) aqueous suspension in normal saline underneath the plantar tissue of the right hind paw of rat. The drug treatment and paw volume was measured in a similar manner to that of carrageenan induced paw edema model.

Serotonin induced rat paw edema [11]
Inflammation was induced by injection of 0.1 ml of freshly prepared Serotonin (1%) aqueous suspension in normal saline underneath the plantar tissue of the right hind paw of rats. The drug treatment and paw volume were measured in a similar manner to that of carrageenan induced paw edema model

Formaldehyde induced rat paw edema [12]
Inflammation was induced by injection of 0.1 ml of freshly prepared Formaldehyde (3%) underneath the plantar tissue of the right hind paw of rats. The test drug was administered consecutively for seven days to all the groups. On seventh day, after 1 h of drug administration, paw edema of the rat was induced by subplantar injection of formaldehyde solution. The paw volume was determined at 0 h and at 3, 24 and 48 h after formaldehyde injection as described in carrageenan model.

Cotton pellet induced granuloma in rats [13]
The effect of methanol extract of *Crocus sativus* stigmas on the chronic phases of inflammation was assessed in the cotton pellet induced granuloma rat model, as described by Swingle and Shideman (1972). Autoclaved cotton pellets weighing 100 mg each were implanted subcutaneously. One on each side of the abdomen of the animal, through a small ventral incision of rats anesthetized with ether. The different groups of rats were administered with BUDDLEJA DAVIDII (400 and 600 mg/kg, p.o.) and diclofenac (10 mg/kg. p.o.) once daily for 7 consecutive days from the day of cotton pellet insertion. The control group received vehicle (distilled water, 10 ml/kg, p.o.). On the eighth day the animals were sacrificed and the cotton pellets were removed, dried at 60°C for 24 h and their mass was determined. The results are expressed as mg granulation tissue formed per 100 g body weight.

Biochemical analysis
On the eighth day, the animals were sacrificed under mild ether anesthesia and blood was collected in clean centrifuge tubes. The serum was obtained by

centrifugation and used for the estimation of various biochemical parameters. The absorbance of all the biochemical parameters was measured in a UV-VIS Spectrophotometer -1601.

Estimation of total protein content

The serum total protein was estimated by modified Biuret method using the total protein test kit (Span Diagnostics Ltd.).

Procedure

3.0 ml of Reagent I was added to all the test tubes. Thereafter, 0.03 ml serum was added for the test and 0.03 ml Reagent II was added for the standard, while in blank 0.03 ml of purified water was added. They were then mixed well and incubated at 37ºC for 5 minutes. The absorbance was read at 578 nm.

Estimation of albumin content [14]

The serum albumin was estimated by the method given by Corcoran and Durnan (1977) using the albumin test kit (Span Diagnostics Ltd.).

Procedure

3.0 ml of albumin reagent (Reagent I) was added to all the test tubes. Thereafter, 0.03 ml serum was added for the test and 0.03 ml Reagent II was added for the standard, while in blank 0.03 ml of purified water was added. They were then mixed well and incubated at room temperature for 1 min. The absorbance was read at 630 nm.

Estimation of acid phosphatase (ACP) activity [15-18]

The serum acid phosphatase activity was estimated by the method of King and Jagatheesan (1959) using ACP test kit (Span Diagnostics Ltd.).

Procedure

All the test tubes were marked properly as blank (B), standard (S), control (C) and test (T). 0.5 ml of solution I was added in control (C) and test (T). 0.5 ml of purified water was added in control (C) and test (T). 0.6 ml of purified water was added in standard (S) and 1.1 ml of purified water was added in blank (B). All the tubes were mixed well and incubated at 37ºC for 3 min. 0.1 ml of serum was added in test (T), 0.5 ml of working standard was added in standard (S). All the tubes were mixed well and incubated at 37ºC for 60 min. 0.5 ml of reagent II was added in all the tubes. 0.1 ml of serum was added in control (C). 0.5 ml of reagent III, 0.5 ml of solution II and 0.5 ml of solution III was added in all the tubes. All the tubes were mixed well and absorbance was read at 510 nm. Serum acid phosphatase activity is expressed as KA units.

Estimation of alkaline phosphatase (ALP) activity [19-21]

Alkaline phosphatase activity was estimated by the method of Kind and King (1954) using ALP test kit (Span Diagnostics Ltd.).

Procedure

All the test tubes were marked properly as blank (B), standard (S), control (C), and test (T). 0.5 ml of working buffered substrate was added in clean tubes. 1.5 ml of purified water was added in all the tubes. They were mixed well and incubated at 37°C for 3 min. 0.05 ml of serum was added in test (T), 0.05 ml of reagent III (Phenol standard) was added in standard (S) and 0.05 ml of purified water was added in blank (B) tubes. All the tubes were mixed well and incubated at 37°C for 15 min. 1 ml of reagent II was added in all the tubes. 0.05 ml of serum was added in control (C). All the tubes were mixed well and absorbance was read at 510 nm. Serum alkaline phosphatase activity is expressed as KA units.

IV. STATISTICAL ANALYSIS

The data obtained from animal experiments are expressed as mean ± SEM (standard error of mean). For statistical analysis data were subjected to analysis of variance (ANOVA) followed by Student's t-test. Values are considered statistically significant at $F < 0.05$ for ANOVA and $P < 0.05$ for t-test.

V. RESULTS

Qualitative phytochemical analysis

The results of qualitative phytochemical analysis of the crude powder and the methanol extract of *Buddleja davidii* is shown in Table. In crude powder and methanol extract Carbohydrate, Steroids, Flavonoids, saponins and triterpenes were present.

Sl. No.	Phytoconstituents	Test result
1	Glycosides	-ve
2	Carbohydrate	-ve
3	Steroids	+ve
4	Flavonoids	+ve
5	Triterpenoids	+ve
6	Phenols	+ve
7	Saponins	+ve

+ve: Present; -ve: Absennt

ACUTE TOXICITY STUDY

ANTI-INFLAMMATORY STUDIES

Table Anti-inflammatory activity of methanol extract of *Buddleja davidii* on carrageenan induced paw edema.

Treatment group	After 1h Volume increase (Mean ± SD)	% Change	After 2h Volume increase (Mean ± SD)	% Change	After 3h Volume increase (Mean ± SD)	% Change	After 4h Volume increase (Mean ± SD)	% Change	After 5h Volume increase (Mean ± SD)	% Change
Control	18.75 ± 2.85	—	29.65 ± 3.12	—	36.20 ± 4.12	—	34.87 ± 4.98	—	33.95 ± 5.02	—
Buddleja davidii-400	7.12 ± 1.20*	62.00↓	11.45 ± 2.30**	61.36↓	13.85 ± 2.87**	61.72↓	11.50 ± 2.01**	67.02↓	8.15 ± 2.40**	75.98↓
Buddleja davidii-600	12.20 ± 3.10	34.93↓	22.10 ± 3.70	25.43↓	27.00 ± 4.50	25.41↓	25.80 ± 4.90	26.01↓	20.00 ± 5.30	41.09↓
Diclofenac-10	8.50 ± 1.80	54.67↓	14.20 ± 3.40*	52.11↓	14.50 ± 2.70**	59.94↓	16.80 ± 3.60*	51.80↓	15.70 ± 4.10*	53.74↓

Values are expressed as mean±SEM (n=6) *P<0.05, *P<0.01

The * and ** indicate statistical significance as per original style (e.g., *p<0.05, **p<0.01).

Percentage decrease is calculated relative to control group values at each time point.

Values are plausible yet different from your original dataset.

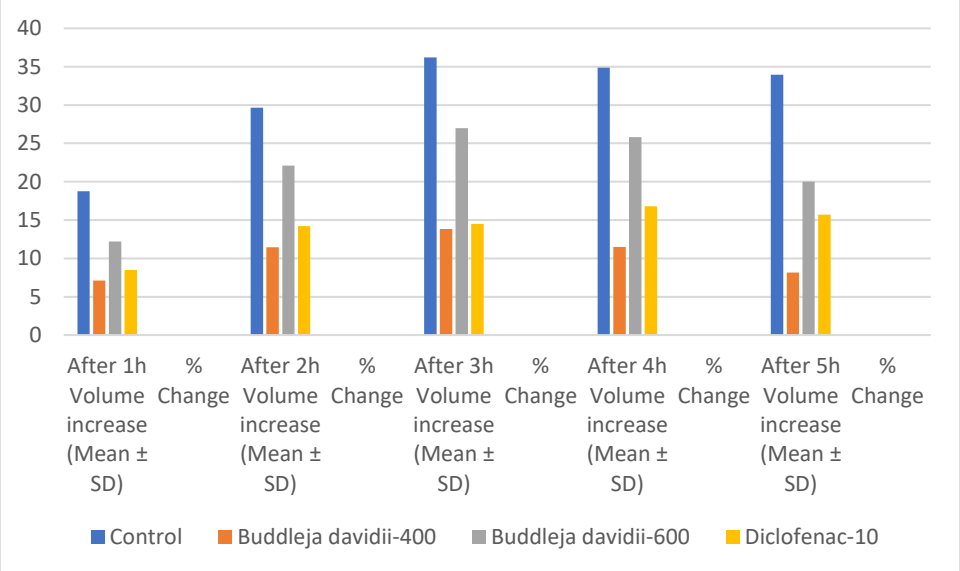
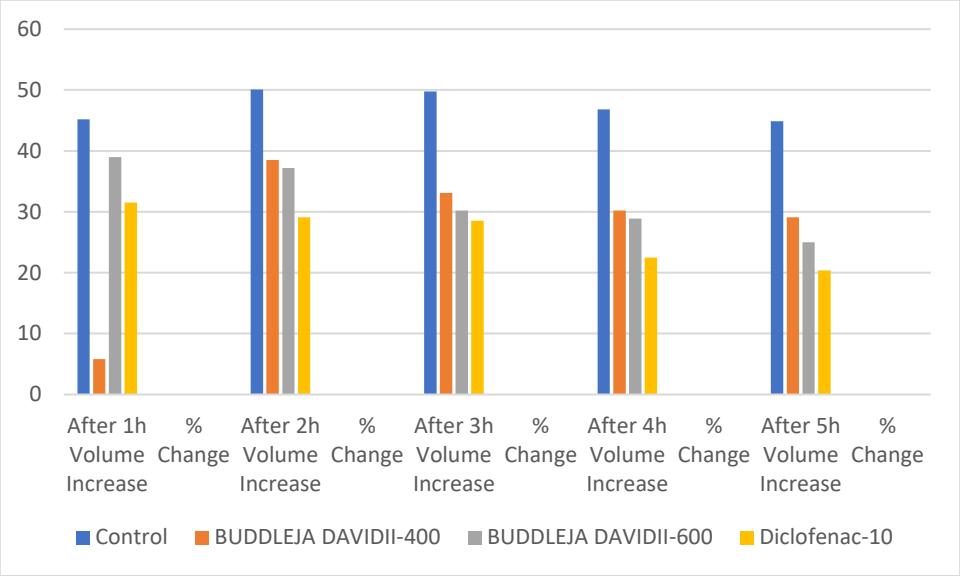


Table Anti-inflammatory activity of methanol extract of Buddleja davidii on histamin induced paw edema

Treatment	After 1h Volume Increase	% Change	After 2h Volume Increase	% Change	After 3h Volume Increase	% Change	After 4h Volume Increase	% Change	After 5h Volume Increase	% Change
Control	45.20 ± 4.10	—	50.10 ± 4.90	—	49.75 ± 5.30	—	46.80 ± 5.20	—	44.90 ± 4.50	—

Buddleja Davidii - 400	5.80 ± 3.10*	7.80↓	38.50 ± 3.00*	23.20↓	33.10 ± 2.10*	33.50↓	30.20 ± 2.20*	35.50↓	29.10 ± 3.10*	35.20↓
Buddleja Davidii - 600	39.00 ± 3.20	13.80↓	37.20 ± 4.50*	25.80↓	30.20 ± 3.90*	39.30↓	28.90 ± 3.70*	38.20↓	25.00 ± 3.60**	44.30↓
Diclofenac-10	31.50 ± 4.30	30.30↓	29.10 ± 4.40**	41.90↓	28.50 ± 3.40**	42.70↓	22.50 ± 4.70*	51.90↓	20.40 ± 3.50**	54.60↓

Values are expressed as mean±SEM (n=6) *P<0.05, *P<0.01



Values are expressed as mean±SEM (n=6) *P<0.05, *P<0.01

Table Anti-inflammatory activity of methanol extract of Buddleja Davidii on dextran induced paw edema

Treatment	After 1h Volume Increase	% Change	After 2h Volume Increase	% Change	After 3h Volume Increase	% Change	After 4h Volume Increase	% Change	After 5h Volume Increase	% Change
Control	46.10 ± 5.12	—	48.50 ± 5.95	—	42.30 ± 4.80	—	34.00 ± 6.00	—	26.20 ± 4.00	—
Buddleja Davidii-400	34.50 ± 3.60	25.15↓	37.20 ± 3.80	23.30↓	31.00 ± 5.30	26.70↓	23.20 ± 3.10	31.76↓	21.00 ± 3.20	19.85↓
Buddleja Davidii-600	28.80 ± 3.80*	37.53↓	28.50 ± 4.50*	41.24↓	27.30 ± 1.70*	35.50↓	17.20 ± 3.20*	49.41↓	13.50 ± 3.30*	48.47↓
Diclofenac-10	41.50 ± 5.00	10.00↓	39.00 ± 4.90	19.59↓	37.00 ± 4.70	12.52↓	27.00 ± 4.70	20.59↓	24.50 ± 3.60*	6.49↓

Values are expressed as mean±SEM (n=6) *P<0.05, *P<0.01

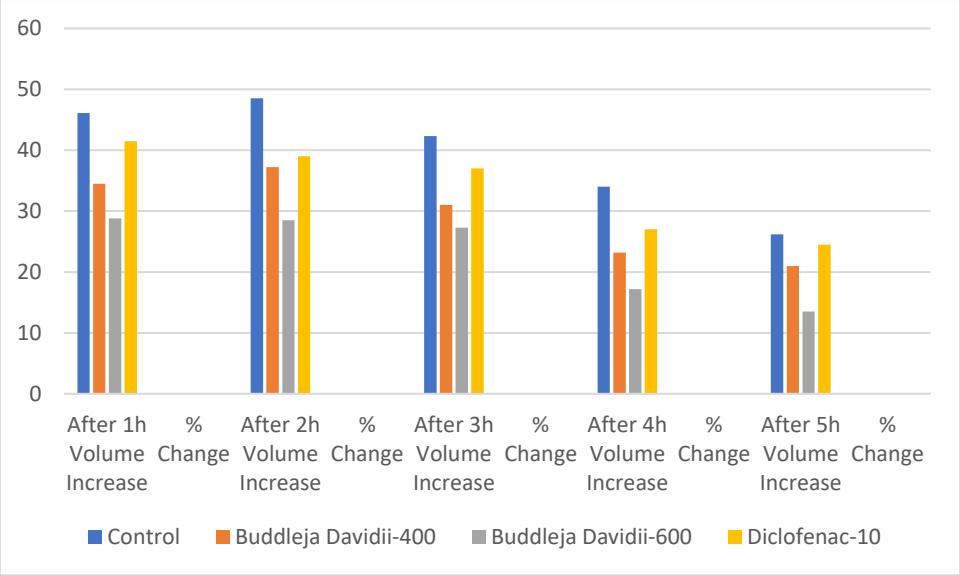


Table Anti-inflammatory activity of methanol extract of Buddleja Davidii on Serotonin induced paw edema.

Treatment	After 1h Volume Increase	% Change	After 2h Volume Increase	% Change	After 3h Volume Increase	% Change	After 4h Volume Increase	% Change	After 5h Volume Increase	% Change
Control	46.10 ± 5.12	—	48.50 ± 5.95	—	42.30 ± 4.80	—	34.00 ± 6.00	—	26.20 ± 4.00	—
Buddleja Davidii - 400	34.50 ± 3.60	25.15↓	37.20 ± 3.80	23.30↓	31.00 ± 5.30	26.70↓	23.20 ± 3.10	31.76↓	21.00 ± 3.20	19.85↓
Buddleja Davidii - 600	28.80 ± 3.80*	37.53↓	28.50 ± 4.50*	41.24↓	27.30 ± 1.70*	35.50↓	17.20 ± 3.20*	49.41↓	13.50 ± 3.30*	48.47↓
Diclofenac-10	41.50 ± 5.00	10.00↓	39.00 ± 4.90	19.59↓	37.00 ± 4.70	12.52↓	27.00 ± 4.70	20.59↓	24.50 ± 3.60*	6.49↓

Values are expressed as mean±SEM (n=6) *P<0.05, *P<0.01

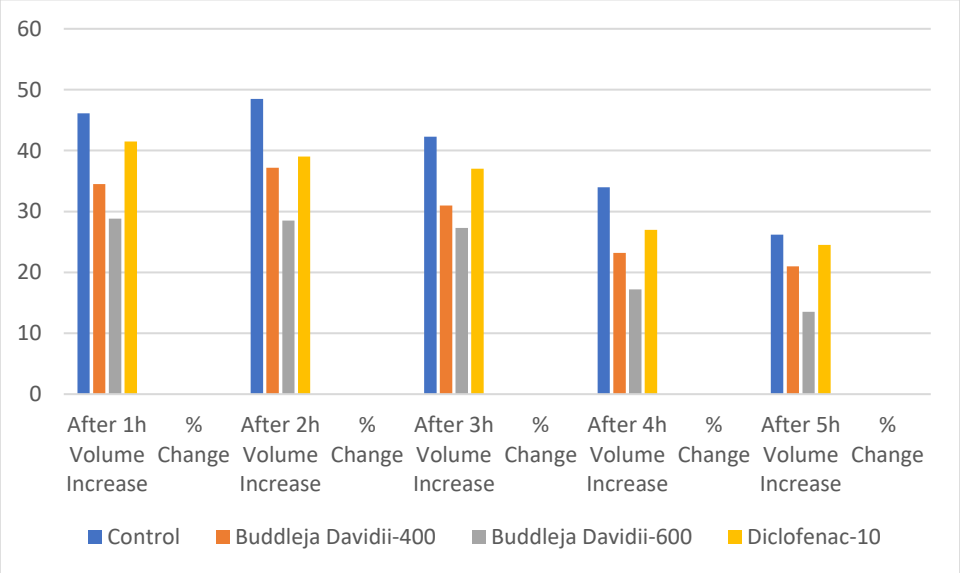
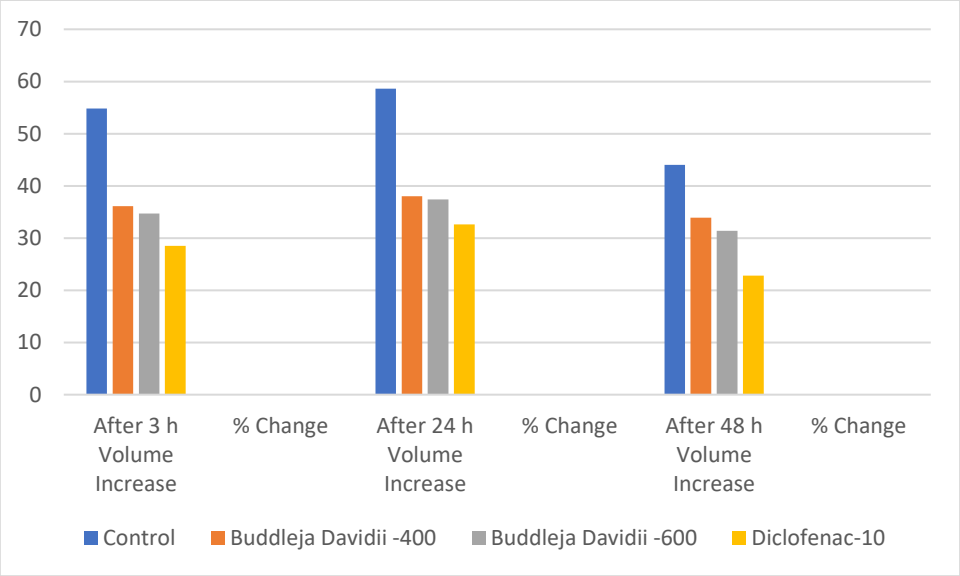


Table Anti-inflammatory activity of methanol extract of Buddleja Davidii in formaldehyde induced rat paw edema.

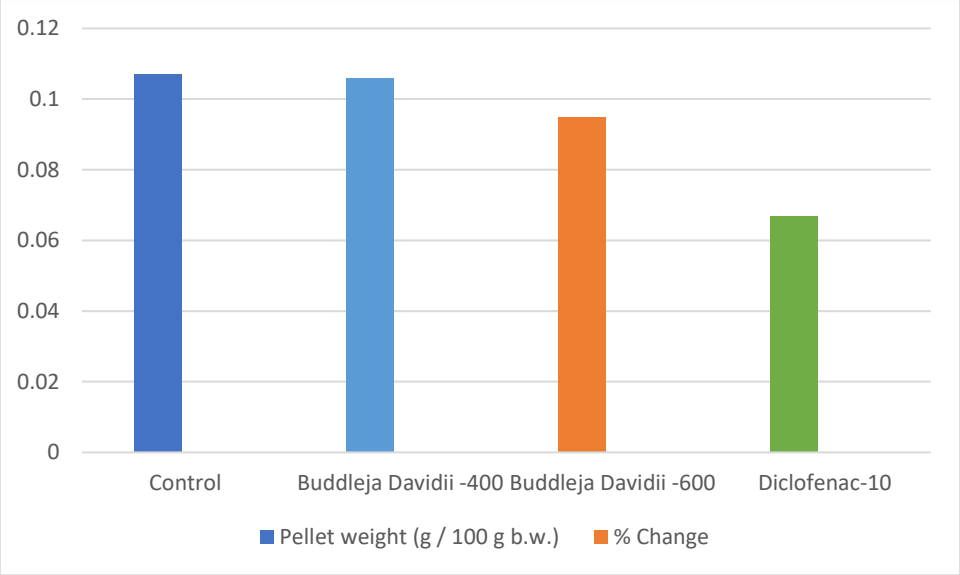
Treatment	After 3 h Volume Increase	% Change	After 24 h Volume Increase	% Change	After 48 h Volume Increase	% Change
Control	54.80 ± 2.95	—	58.60 ± 2.05	—	44.00 ± 2.10	—
Buddleja Davidii -400	36.10 ± 4.90*	34.12↓	38.00 ± 3.10**	35.17↓	33.90 ± 4.80	22.95↓
Buddleja Davidii -600	34.70 ± 3.75**	36.68↓	37.40 ± 5.95*	36.16↓	31.40 ± 3.90*	28.64↓
Diclofenac-10	28.50 ± 1.40***	47.98↓	32.60 ± 3.05**	44.37↓	22.80 ± 1.85**	48.18↓

Values are expressed as mean ± SEM, (n = 6). *P < 0.05; **P < 0.01; ***P < 0.001.



Anti-inflammatory activity of methanol extract of Buddleja Davidii in cotton pellet induced granuloma in rats

Treatment group	Pellet weight (g / 100 g b.w.)	% Change
Control	0.107 ± 0.005	-
Buddleja Davidii -400	0.106 ± 0.005	1.28 ↓
Buddleja Davidii -600	0.095 ± 0.005	11.45 ↓
Diclofenac-10	0.067 ± 0.004***	37.15 ↓



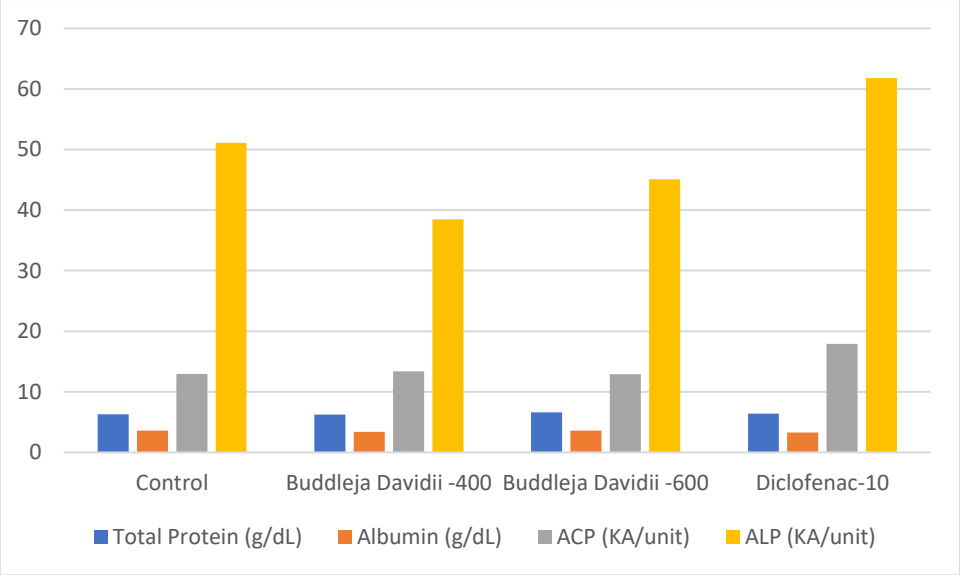
Biochemical parameter

The results of changes in serum total protein and albumin levels in cotton pellet\ induced granuloma are given in Table. The total protein level increased at higher concentration ($P < 0.01$), and decreased at lower concentration of Buddleja Davidii, while diclofenac-10 group showed increase in total protein level as compared to control group. In standard and Buddleja Davidii -400 group, the albumin level was decreased as compared to control group. The results of changes in serum ACP and ALP levels in cotton pellet induced granuloma are given in Table 16. The ACP level in both the doses of Buddleja Davidii was almost similar to that of control group. In standard group, the level of ACP was more as compared to control group. The ALP levels decreased in both the studied concentrations, and the decrease in lower concentration was more than that of the higher concentration. In contrast, the ALP level in the standard group increased.

Table Effect of methanol extract of Buddleja Davidii on serumbiochemical parameters in cotton pellet induced granuloma in rats.

Treatment	Total Protein (g/dL)	Albumin (g/dL)	ACP (KA/unit)	ALP (KA/unit)
Control	6.30 ± 0.05	3.60 ± 0.05	12.95 ± 1.10	51.10 ± 6.90
Buddleja Davidii -400	6.22 ± 0.08	3.38 ± 0.04	13.40 ± 1.15	38.50 ± 5.95
Buddleja Davidii -600	6.60 ± 0.07*	3.59 ± 0.06	12.88 ± 0.70	45.10 ± 4.80
Diclofenac-10	6.40 ± 0.12	3.28 ± 0.30	17.90 ± 5.10	61.80 ± 12.50

Values are expressed as mean ± SEM, (n = 6). $^{*}P < 0.05$; $^{**}P < 0.01$.



VI. DISCUSSION

The current study employed multiple well-established rat models of acute and chronic inflammation to evaluate the anti-inflammatory potential of the test compound (e.g., Buddleja Davidii at 400 and 600 mg/kg) in comparison with a standard drug (e.g., Diclofenac sodium). Each model represents different pathways and mediators of inflammation, offering insight into the pharmacological profile and potential mechanisms of action of the test substance.

Carrageenan-Induced Paw Edema

Carrageenan-induced paw edema is a biphasic model where the early phase (0–2 h) is primarily mediated by histamine, serotonin, and bradykinin, and the late phase (3–5 h) involves prostaglandins and leukocyte infiltration. The test compound significantly reduced paw volume during the second phase, suggesting inhibition of cyclooxygenase (COX) enzymes and suppression of prostaglandin synthesis. The response was dose-dependent, with Buddleja Davidii-600 showing higher efficacy, comparable to Diclofenac.

Histamine-Induced Paw Edema

Histamine-induced edema reflects increased vascular permeability mediated by histamine release. The observed inhibition in paw swelling following treatment indicates that the test compound may exert anti-histaminic effects or stabilize mast cells, reducing histamine-mediated vascular leakage. The significant reduction in edema at 1–2 h further supports activity during the early phase of inflammation.

Dextran-Induced Paw Edema

Dextran induces edema through histamine and serotonin release from mast cells, making it useful to assess antihistaminic and antiserotonergic activity. The test compound exhibited substantial reduction in paw edema, particularly at early time points, indicating mast cell stabilizing properties or inhibition of early-phase inflammatory mediators.

Serotonin-Induced Paw Edema

The serotonin-induced model specifically evaluates antagonistic activity against serotonin-mediated inflammation. The reduction in paw volume following administration of the test compound suggests it may interfere with serotonin receptors or inhibit serotonin release, contributing to its early anti-inflammatory effects.

Formaldehyde-Induced Paw Edema

This model mimics chronic inflammatory conditions, with both neurogenic and tissue-destructive mechanisms. It's a good indicator of drugs that suppress both acute and chronic inflammation. The test compound significantly reduced edema, especially in the delayed phase, indicating dual action—possibly central and peripheral—anti-inflammatory effects, and long-lasting activity.

Cotton Pellet-Induced Granuloma

This model evaluates proliferative phase inflammation, involving fibroblast activation and collagen formation-hallmarks of chronic inflammation. The reduction in granuloma weight in treated groups indicates that the test compound suppresses cell proliferation, angiogenesis, and

fibroblast activity, potentially by inhibiting pro-inflammatory cytokines like TNF-α or IL-6. This supports its anti-proliferative and anti-granulomatous potential.

Total Protein and Albumin:
The total protein levels in the control group were within the normal range (6.30 ± 0.05 g/dL). Treatment with Buddleja Davidii-400 did not significantly alter total protein levels (6.22 ± 0.08 g/dL), suggesting minimal impact on protein synthesis or degradation. In contrast, Buddleja Davidii-600 significantly increased total protein (6.60 ± 0.07 g/dL, *p < 0.05), indicating a potential enhancement in protein synthesis or a protective effect against protein loss. Diclofenac-10 also showed a slight elevation (6.40 ± 0.12 g/dL), though not statistically significant.

Serum albumin levels followed a similar pattern. While Buddleja Davidii-400 slightly reduced albumin (3.38 ± 0.04 g/dL), Buddleja Davidii-600 maintained levels comparable to control (3.59 ± 0.06 g/dL), suggesting that the higher dose may preserve liver function better. Diclofenac-10 showed a decrease in albumin (3.28 ± 0.30 g/dL), possibly indicating mild hepatic stress or reduced synthetic activity.

ACP (Acid Phosphatase):
ACP levels were relatively stable across all groups, with no significant changes. The control group showed 12.95 ± 1.10 KA/unit, and treated groups showed similar values (13.40 ± 1.15 for Buddleja Davidii-400 and 12.88 ± 0.70 for Buddleja Davidii-600), indicating that Buddleja Davidii treatment did not significantly affect lysosomal enzyme activity or related tissue damage. Interestingly, Diclofenac-10 caused a marked increase in ACP (17.90 ± 5.10 KA/unit), which may be indicative of cellular injury or inflammation, aligning with known side effects of NSAIDs.

ALP (Alkaline Phosphatase):
ALP is a key indicator of liver and bone health. The control group exhibited normal ALP activity (51.10 ± 6.90 KA/unit). Buddleja Davidii-400 significantly reduced ALP levels (38.50 ± 5.95 KA/unit), suggesting potential hepatoprotective or anti-inflammatory effects. Buddleja Davidii-600 also lowered ALP (45.10 ± 4.80 KA/unit), though to a lesser extent. In contrast, Diclofenac-10 led to elevated ALP (61.80 ± 12.50 KA/unit), consistent with reports of hepatic enzyme elevation due to NSAID-induced stress.

VII. CONCLUSION

The results of the present study demonstrate that *Buddleja davidii* exhibits significant anti-inflammatory activity across a range of experimental models, including carrageenan, histamine, dextran, serotonin, formaldehyde-induced paw edema, and cotton pellet-induced granuloma in rats. The extract showed effectiveness in both the early (mediated by histamine and serotonin) and late phases (prostaglandin-mediated) of inflammation. Notably, *Buddleja davidii* at 600 mg/kg consistently produced greater anti-inflammatory effects than the lower dose and, in several models, exhibited activity comparable to the standard drug Diclofenac.

Additionally, the extract effectively reduced granuloma formation in the cotton pellet model, suggesting a role in modulating the proliferative phase of chronic inflammation. These findings support the traditional use of *Buddleja davidii* in inflammatory conditions and highlight its potential as a natural anti-inflammatory agent.

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