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PHARMACOLOGICAL EVALUATION OF ANTI-INFLAMMATORY ACTIVITY OF ETHANOLIC EXTRACT OF BY USING IPOMEA HEDERIFOLIA *IN VIVO* STUDIES

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Abstract:

Inflammation is a complex biological response to tissue injury, infection, or irritation and is associated with the release of various inflammatory mediators and oxidative stress. The present study was designed to evaluate the anti-inflammatory activity of the ethanolic extract of Ipomoea hederifolia using in vivo experimental models. The anti-inflammatory activity was assessed using carrageenan-induced paw edema for acute inflammation and cotton pellet-induced chronic inflammation in Wistar albino rats. Animals were divided into five groups: normal control, negative control, standard diclofenac-treated group (10 mg/kg), and two extract-treated groups receiving Ipomoea hederifolia extract at doses of 200 mg/kg and 500 mg/kg, respectively. In the carrageenan-induced paw edema model, the extract produced a significant and dose-dependent reduction in paw swelling compared with the negative control group. In the chronic inflammation model, the extract significantly reduced inflammatory tissue formation, indicating inhibition of the proliferative phase of inflammation. Biochemical analysis demonstrated a significant decrease in pro-inflammatory cytokines, including TNF-α, IL-1β, IL-6, and C-reactive protein (CRP), in extract-treated animals. Furthermore, the extract restored antioxidant enzyme levels such as superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH), while significantly reducing malondialdehyde (MDA) levels. The higher dose (500 mg/kg) exhibited greater anti-inflammatory and antioxidant effects, comparable to those of diclofenac sodium. The findings suggest that the ethanolic extract of Ipomoea hederifolia possesses significant anti-inflammatory activity, possibly mediated through the suppression of inflammatory mediators and enhancement of antioxidant defense mechanisms. Therefore, Ipomoea hederifolia may represent a promising natural source for the development of anti-inflammatory therapeutic agents.

Keywords: *Ipomoea hederifolia, Anti-inflammatory activity, Carrageenan-induced paw edema, Chronic inflammation, Diclofenac sodium, Pro-inflammatory cytokines.*

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INTRODUCTION:

Medicinal plants have served as an important source of therapeutic agents since prehistoric times. The use of plants for the treatment of diseases is deeply rooted in human civilization and has been documented in several ancient texts, including the Rigveda, the Bible, and the Quran. More than 6,000 years ago, ancient Chinese civilizations were among the first to systematically utilize natural vegetation for medicinal purposes. In India, the Ayurvedic system of medicine has been practiced for over 3,000 years and remains one of the oldest traditional healthcare systems in the world. Hippocrates, widely regarded as the "Father of Medicine," was the first to provide a scientific basis for understanding diseases and their treatment. Traditional Indian systems of medicine encompass Ayurveda, Siddha, Unani, Tibetan medicine, and Naturopathy. Herbal therapy offers a rational and holistic approach to healthcare, providing effective treatment for numerous chronic and difficult-to-manage diseases that are often challenging to cure through other medical systems. In addition to treating illnesses, herbal medicine also emphasizes disease prevention, health promotion, and the maintenance of overall well-being [1-2].

History of Herbal Drugs

The use of medicinal plants for the treatment of human diseases dates back thousands of years. Early humans discovered the healing properties of plants through observation, intuition, and trial-and-error methods. The earliest references to medicinal plants are found in the *Rig Veda*, which was composed between 4000 and 1600 B.C. A more extensive description of the medicinal use of plants is available in the *Atharva Veda*. The science of *Ayurveda*, regarded as an *Upa Veda*, provides detailed information on the properties and therapeutic applications of various medicinal plants.

One of the earliest and most important Ayurvedic texts, the *Charaka Samhita* (around 600 B.C.), describes approximately 341 medicinal plants used in healthcare. Another classical text, the *Sushruta Samhita*, also contains valuable information on medicinal plants and their applications. Renowned scholars such as Dhanvantari and Nagarjuna possessed extensive knowledge of medicinal herbs and their therapeutic properties. *Rauwolfia serpentina*, a medicinal plant that later gained worldwide recognition, is mentioned in ancient Hindu scriptures and in the *Charaka Samhita* as an effective remedy for snake bites and insect stings.

Herbal medicines gained popularity because of their effectiveness, easy availability, affordability, and relatively low incidence of adverse effects. The medicinal value of plants is attributed to the presence of bioactive chemical constituents known as secondary metabolites. These include alkaloids, glycosides, tannins, essential oils, fixed oils, resins, gums, and mucilages. Such compounds are distributed in different parts of plants, including roots, stems, leaves, seeds, bark, flowers, and fruits, and are responsible for their therapeutic activities.

The widespread acceptance of herbal medicine over conventional allopathic medicine is due to several advantages, including lower cost, fewer side effects, easy accessibility, and their ability to provide holistic treatment by addressing the underlying causes of diseases. Herbal remedies are also considered beneficial in the management of various chronic and difficult-to-treat disorders [3-5].

SCOPE OF HERBAL MEDICINES

Herbal medicines play a significant role in healthcare systems worldwide and continue to gain importance due to their therapeutic efficacy, safety, affordability, and cultural acceptance. The growing interest in natural products has expanded the scope of herbal medicines in the prevention, diagnosis, and treatment of various diseases. According to the World Health Organization (WHO), a large proportion of the global population relies on traditional herbal remedies for primary healthcare needs. The scope of herbal medicines extends beyond traditional healing practices and includes modern pharmaceutical research, nutraceuticals, cosmetics, and preventive healthcare. Medicinal plants serve as valuable sources of bioactive compounds that have led to the discovery and development of numerous modern drugs. Herbal medicines are widely used in the management of chronic diseases such as diabetes, hypertension, arthritis, obesity, cardiovascular disorders, liver diseases, neurological disorders, and various infectious diseases.

Advances in phytochemistry, pharmacology, biotechnology, and analytical techniques have enhanced the scientific validation of herbal medicines. Research on medicinal plants has facilitated the identification of active constituents, mechanisms of action, and therapeutic potential of herbal formulations. The integration of traditional knowledge with modern scientific approaches has opened new avenues for drug discovery and development. Herbal medicines also have significant applications in preventive healthcare, where they are used to improve immunity, maintain overall well-being, and reduce the risk of disease. The increasing demand for natural and plant-based products has contributed to the growth of the herbal pharmaceutical industry, creating opportunities for cultivation, processing, standardization, quality control, and commercialization of medicinal plants.

Furthermore, herbal medicines have an important role in complementary and alternative medicine systems such as Ayurveda, Siddha, Unani,

Traditional Chinese Medicine, and Homeopathy. Their widespread acceptance, coupled with ongoing scientific research, suggests that herbal medicines will continue to be a valuable resource for global healthcare and future therapeutic innovations [6-7].

INFLAMMATION

Inflammation is a complex biological response of the body's vascularized tissues to harmful stimuli such as pathogens, damaged cells, toxins, irritants, or physical injury. It serves as a protective mechanism aimed at eliminating the initial cause of cell injury, removing damaged tissues, and initiating the healing process. Although inflammation is an essential component of the body's defense system, excessive or prolonged inflammatory responses can contribute to the development of various acute and chronic diseases.

The inflammatory response involves a coordinated interaction among immune cells, blood vessels, and molecular mediators. When tissues are injured or infected, immune cells release chemical mediators such as histamine, prostaglandins, leukotrienes, cytokines, and chemokines. These mediators increase blood flow to the affected area, enhance vascular permeability, and recruit leukocytes to the site of injury. As a result, the classical signs of inflammation become evident, including redness (*rubor*), heat (*calor*), swelling (*tumor*), pain (*dolor*), and loss of function (*functio laesa*). Inflammation can be classified into acute and chronic forms. Acute inflammation is a rapid and short-term response that develops within minutes or hours following tissue injury or infection. It is characterized by the exudation of plasma proteins and the migration of neutrophils to the affected site. The primary objective of acute inflammation is to eliminate the causative agent and restore normal tissue function. If the injurious stimulus persists, the inflammatory process may progress to chronic inflammation.

Chronic inflammation is a prolonged inflammatory response that may last for weeks, months, or even years. It is characterized by the infiltration of macrophages, lymphocytes, and plasma cells, along with tissue destruction and repair processes such as fibrosis and angiogenesis. Chronic inflammation has been implicated in the pathogenesis of numerous disorders, including rheumatoid arthritis, inflammatory bowel disease, asthma, diabetes mellitus, cardiovascular diseases, neurodegenerative disorders, and cancer.

Several molecular pathways regulate the inflammatory process. Among these, the nuclear factor-kappa B (NF-κB) signaling pathway plays a crucial role in the expression of pro-inflammatory genes. Other important mediators include tumor necrosis factor-alpha (TNF-α), interleukins (IL-1β, IL-6), cyclooxygenase (COX) enzymes, and inducible nitric oxide synthase (iNOS). These mediators contribute to the initiation and maintenance of inflammatory responses and are common targets for anti-inflammatory therapies. The management of inflammation involves the use of steroidal and non-steroidal anti-inflammatory drugs (NSAIDs), immunosuppressive agents, and biological therapies. However, the long-term use of these medications may be associated with adverse effects such as gastrointestinal irritation, renal dysfunction, cardiovascular complications, and immunosuppression. Consequently, there is growing interest in medicinal plants and herbal products as potential sources of safer and more effective anti-inflammatory agents. Numerous plant-derived compounds, including flavonoids, alkaloids, terpenoids, tannins, and phenolic compounds, have demonstrated significant anti-inflammatory activity by modulating various inflammatory pathways. Therefore, understanding the mechanisms of inflammation and identifying novel anti-inflammatory agents remain important areas of biomedical and pharmaceutical research for the prevention and treatment of inflammatory diseases [8-10].

Acute Inflammation

Acute inflammation is the immediate and short-term protective response of the body to tissue injury, infection, trauma, or exposure to harmful agents. It develops rapidly, usually within minutes or hours, and is characterized by the classic signs of redness, heat, swelling, pain, and loss of function. The process involves vasodilation, increased vascular permeability, and the migration of neutrophils to the site of injury to eliminate pathogens and remove damaged tissue. Acute inflammation generally lasts for a few hours to a few days and usually resolves completely once the causative factor is removed, leading to tissue repair and restoration of normal function.

Chronic Inflammation

Chronic inflammation is a prolonged inflammatory response that persists for weeks, months, or even years when the causative agent cannot be eliminated or when there is repeated tissue injury. It is characterized by the infiltration of macrophages, lymphocytes, and plasma cells, along with ongoing tissue destruction and attempts at healing through fibrosis and angiogenesis. Chronic inflammation may result from persistent infections, autoimmune disorders, prolonged exposure to toxic substances, or unresolved acute inflammation. Unlike acute inflammation, chronic inflammation often causes progressive tissue damage, scarring, and organ dysfunction, contributing to diseases such as Tuberculosis, Rheumatoid Arthritis, and Inflammatory Bowel Disease.

Problems Associated with Chronic Inflammation

Chronic inflammation is a prolonged inflammatory response that can cause significant tissue damage and contribute to the development of various diseases. Continuous activation of inflammatory cells and release of inflammatory mediators lead to the destruction of normal tissues, fibrosis, and loss of organ function. Chronic inflammation is associated with several disorders, including rheumatoid arthritis, inflammatory bowel disease, asthma, chronic obstructive pulmonary disease (COPD), and autoimmune diseases. It also plays a major role in the development of cardiovascular diseases such as atherosclerosis, where inflammation contributes to plaque formation and blood vessel damage. Persistent inflammation can promote cancer development by causing DNA damage, stimulating abnormal cell growth, and creating a favorable environment for tumor progression. Additionally, chronic inflammation is linked to metabolic disorders such as obesity, insulin resistance, type 2 diabetes mellitus, and non-alcoholic fatty liver disease. Thus, chronic inflammation is a major health concern due to its role in tissue destruction, organ dysfunction, and the progression of numerous chronic diseases [11-12].

Symptoms of Inflammation

Inflammation is characterized by five classical signs: redness (rubor), heat (calor), swelling (tumor), pain (dolor), and loss of function (functio laesa). Redness and heat occur due to increased blood flow to the affected area, while swelling results from the accumulation of fluid and immune cells in the tissues. Pain is caused by the release of inflammatory mediators, such as prostaglandins and bradykinin, which stimulate nerve endings. Loss of function may occur because of pain, swelling, or tissue damage, limiting the normal activity of the affected organ or tissue. In severe or systemic inflammation, additional symptoms such as fever, fatigue, loss of appetite, muscle aches, and increased white blood cell count may also be observed. These symptoms represent the body's protective response aimed at eliminating harmful stimuli and promoting tissue repair.

Causes of Inflammation

Inflammation is a protective response of the body's immune system to harmful stimuli that cause tissue injury or damage. The major causes of inflammation include infectious agents such as bacteria, viruses, fungi, and parasites, which trigger immune responses to eliminate the invading microorganisms. Physical agents like trauma, burns, radiation, extreme temperatures, and mechanical injuries can also induce inflammation by damaging tissues. Chemical agents, including acids, alkalis, toxins, and environmental pollutants, may cause cellular injury and initiate inflammatory reactions. Inflammation can also result from immune reactions, such as allergies, hypersensitivity reactions, and autoimmune diseases, where the immune system mistakenly attacks the body's own tissues. Additionally, tissue necrosis caused by lack of blood supply, ischemia, or infarction releases cellular contents that stimulate inflammation. The presence of foreign bodies such as splinters, dust particles, surgical sutures, or other non-degradable materials can also provoke chronic inflammatory responses. Thus, inflammation serves as a defense mechanism against a wide variety of harmful stimuli to protect the body and promote healing.

Mediators of Inflammation

Inflammatory mediators are chemical substances that initiate, regulate, and amplify the inflammatory response. They are produced by various cells such as mast cells, macrophages, neutrophils, platelets, and endothelial cells, as well as by plasma proteins. The major mediators of inflammation include histamine, which causes vasodilation and increased vascular permeability; serotonin, which contributes to vascular changes; and prostaglandins, which are responsible for pain, fever, and vasodilation. Leukotrienes increase vascular permeability and attract leukocytes to the site of injury. Cytokines, such as tumor necrosis factor-alpha (TNF-α), interleukin-1 (IL-1), and interleukin-6 (IL-6), regulate immune and inflammatory responses and promote the recruitment of inflammatory cells. Chemokines direct the migration of leukocytes to sites of inflammation. Bradykinin causes pain, vasodilation, and increased vascular permeability. The complement system generates components such as C3a and C5a, which enhance inflammation and attract immune cells. Nitric oxide (NO) contributes to vasodilation and antimicrobial activity, while reactive oxygen species (ROS) help destroy pathogens but may also damage host tissues. Together, these mediators coordinate the complex processes involved in inflammation and tissue repair [13-15].

Prostaglandins

Prostaglandins are biologically active lipid compounds derived from arachidonic acid through the action of the cyclooxygenase (COX) enzymes. They are produced by many types of cells in response to tissue injury, infection, or inflammation and act as important mediators of the inflammatory process. Prostaglandins cause vasodilation and increase vascular permeability, thereby contributing to redness and swelling at the site of inflammation. They also sensitize nerve endings, leading to pain, and play a key role in the development of fever by acting on the hypothalamus. Different

prostaglandins perform specific functions; for example, PGE_2 is involved in pain, fever, and inflammation, while PGI_2 (prostacyclin) causes vasodilation and inhibits platelet aggregation. Because of their role in inflammation, prostaglandin synthesis is inhibited by nonsteroidal anti-inflammatory drugs (NSAIDs) such as aspirin and ibuprofen, which block cyclooxygenase enzymes and reduce pain, fever, and inflammation. Thus, prostaglandins are essential mediators that regulate numerous physiological and pathological processes, particularly inflammatory responses.

Role of Cyclooxygenase (COX) in Inflammation

Cyclooxygenase (COX) is a key enzyme involved in the synthesis of prostaglandins from arachidonic acid, which is released from cell membrane phospholipids during tissue injury or inflammation. Prostaglandins produced through the COX pathway play an important role in mediating the signs and symptoms of inflammation, including pain, fever, redness, and swelling. There are two main isoforms of cyclooxygenase: COX-1 and COX-2. COX-1 is constitutively expressed in most tissues and is responsible for normal physiological functions such as protection of the gastric mucosa, regulation of renal blood flow, and platelet aggregation. In contrast, COX-2 is induced at sites of inflammation by cytokines, growth factors, and other inflammatory stimuli, leading to increased production of inflammatory prostaglandins. These prostaglandins promote vasodilation, increase vascular permeability, and sensitize pain receptors, thereby contributing to the inflammatory response. Nonsteroidal anti-inflammatory drugs (NSAIDs) such as aspirin, ibuprofen, and diclofenac exert their anti-inflammatory effects by inhibiting cyclooxygenase activity and reducing prostaglandin synthesis. Therefore, cyclooxygenase, particularly COX-2, plays a central role in the development and maintenance of inflammation [16].

Inflammatory Diseases

Inflammatory diseases are disorders characterized by persistent or excessive inflammation that leads to tissue damage and impaired organ function. These diseases may result from infections, autoimmune reactions, allergic responses, genetic factors, or environmental triggers. Common inflammatory diseases include rheumatoid arthritis, in which chronic inflammation affects the joints causing pain, swelling, and deformity; inflammatory bowel disease (IBD), including Crohn's disease and ulcerative colitis, which causes inflammation of the gastrointestinal tract; asthma, characterized by chronic inflammation of the airways; and psoriasis, an inflammatory skin disorder marked by red, scaly patches. Other examples include systemic lupus erythematosus (SLE), chronic obstructive pulmonary disease (COPD), hepatitis, and atherosclerosis, where inflammation contributes to blood vessel damage and cardiovascular disease. Chronic inflammatory diseases can significantly affect quality of life and may lead to long-term complications if not properly managed. Early diagnosis and appropriate treatment are essential to control inflammation, prevent tissue damage, and improve patient outcomes.

Standard Drugs for Inflammation and Their Side Effects

Several standard drugs are used to treat inflammation by reducing pain, swelling, and tissue damage. The most commonly used anti-inflammatory agents include nonsteroidal anti-inflammatory drugs (NSAIDs), corticosteroids, and disease-modifying anti-rheumatic drugs (DMARDs). NSAIDs such as aspirin, ibuprofen, diclofenac, naproxen, and indomethacin act by inhibiting cyclooxygenase (COX) enzymes, thereby reducing prostaglandin synthesis and inflammation. However, prolonged use of NSAIDs may cause side effects such as gastric irritation, peptic ulcers, gastrointestinal bleeding, kidney dysfunction, and an increased risk of cardiovascular events. Corticosteroids, including prednisolone, dexamethasone, and hydrocortisone, are potent anti-inflammatory agents that suppress immune and inflammatory responses. Their long-term use can lead to adverse effects such as osteoporosis, hypertension, hyperglycemia, weight gain, increased susceptibility to infections, cataracts, and adrenal suppression. DMARDs, such as methotrexate and sulfasalazine, are mainly used in chronic inflammatory and autoimmune diseases like rheumatoid arthritis, but they may cause hepatotoxicity, bone marrow suppression, gastrointestinal disturbances, and increased risk of infections. Although these drugs are effective in controlling inflammation, their potential side effects highlight the need for safer and more effective therapeutic alternatives [17].

Three major groups of drugs are commonly used in the treatment of inflammatory diseases: corticosteroids, disease-modifying antirheumatic drugs (DMARDs), and non-steroidal anti-inflammatory drugs (NSAIDs) such as aspirin, ibuprofen, and naproxen. These agents are widely prescribed and have proven efficacy in the management of various inflammatory disorders. Among them, NSAIDs are the most frequently used drugs for the treatment of inflammation. They exert their therapeutic effects by inhibiting cyclooxygenase (COX) enzymes, thereby reducing the synthesis of prostaglandins (PGs), which are key mediators of pain, fever, and inflammation. More than fifty NSAIDs are currently available for clinical use, and they possess three principal

pharmacological actions: anti-inflammatory, analgesic, and antipyretic effects. The anti-inflammatory action of NSAIDs makes them useful in the treatment of conditions such as rheumatoid arthritis, osteoarthritis, musculoskeletal disorders, and pericarditis. Their analgesic activity provides relief from mild to moderate pain without producing dependence, unlike opioid analgesics. In addition, their antipyretic action reduces fever by inhibiting the synthesis of prostaglandins in the hypothalamus, which is stimulated by endogenous pyrogens released from monocytes and macrophages during infection or inflammation.

NSAIDs act by blocking COX enzymes and consequently reducing prostaglandin production throughout the body. This mechanism alleviates inflammation, pain, and fever associated with both acute and chronic inflammatory conditions. NSAIDs have been extensively used in the management of chronic inflammatory diseases such as arthritis, psoriasis, and asthma. Commonly prescribed NSAIDs include aspirin, indomethacin, ibuprofen, naproxen, and diclofenac, with diclofenac being one of the most widely used NSAIDs worldwide. Most COX inhibitors possess analgesic, antipyretic, anti-inflammatory, and antiplatelet properties. NSAIDs function by inhibiting the active site of cyclooxygenase isozymes, thereby preventing the formation of inflammatory prostaglandins.

Based on their selectivity toward cyclooxygenase enzymes, NSAIDs are classified into two major groups: COX-2 selective inhibitors (COXIBs) and non-selective NSAIDs. They may also be categorized according to their chemical structure and pharmacokinetic characteristics. Although NSAIDs are highly effective therapeutic agents, their long-term use is associated with several adverse effects. Prolonged administration may cause gastrointestinal irritation, peptic ulceration, gastrointestinal bleeding, platelet dysfunction, and damage to renal and cardiovascular systems. Corticosteroids are potent anti-inflammatory agents that suppress immune and inflammatory responses. However, long-term corticosteroid therapy may result in serious side effects, including hyperglycemia, insulin resistance, diabetes mellitus, osteoporosis, anxiety, hypertension, and increased susceptibility to infections. Furthermore, prolonged NSAID use may impair immune function, which can have significant consequences in children, elderly individuals, and immunocompromised patients. Despite their effectiveness, currently available synthetic anti-inflammatory drugs are limited by their toxicity and the recurrence of symptoms following discontinuation of therapy. Therefore, the search for safer and more effective anti-inflammatory agents, particularly from natural sources, remains an important area of pharmaceutical research [18].

5. MATERIAL AND METHODS:

Collection and Identification of Plant Material

The fresh Ipomea Hederifolia are collected and identified was purchased from local market Pradesh, India, and authenticated by Dr. K. Madhava Chetty, Assistant Professor, Department of Botany, S.V University, Tirupati.

Preparation of the Extract

The Collected plant was wasted, air dried at room temp 7-14 days. were pulverized with the help of electric grinder the powder was further Sieved to get a fine powder and Stored in air tight Container. The powder was extracted using Soxhlet apparatus.

Extraction procedure

Coarsely powdered Ipomea Hederifolia were used for extraction with Ethanol by using Soxhlet method for 6-8 hours. 50g of dried powder was weighed and 500mL of solvent Ethanol is used for the extraction. The extracts were evaporated by using a rotary evaporator and dried at room temperature. The obtained crude extracts were weighed and stored at 4^{o}C for further analysis.

Acute toxicity study

The acute toxicity study of *Ipomea Hederifolia* extract is conducted to assess its safety and determine the highest dose that can be administered without causing harmful effects. Healthy laboratory animals, such as Wistar rats or Swiss albino mice, are divided into groups and given single oral doses of the extract at varying concentrations, typically ranging from 300 mg/kg to 2000 mg/kg body weight. Observations are made for signs of toxicity, behavioral changes, alterations in food and water intake, and any mortality over a period of 24 hours and then daily for up to 14 days. At the end of the study, a gross necropsy is performed to examine vital organs such as the liver, kidney, and heart for any pathological changes. The study is carried out according to internationally accepted guidelines like OECD 423, ensuring ethical treatment of animals under Institutional Animal Ethics Committee approval. The outcome helps establish the safe dose for further pharmacological studies and provides important information on the toxicological profile of the extract, thereby supporting its potential use as a therapeutic agent.

Induction of Inflammation

The induction of inflammation in experimental animals is an essential step for evaluating the anti-inflammatory activity of *Ipomea Hederifolia* extract. In this study, inflammation is typically induced by injecting an inflammatory agent such as carrageenan into the sub-plantar region of the rat's hind paw. Carrageenan, a sulfated polysaccharide, triggers acute inflammation by promoting the

release of histamine, serotonin, bradykinin, and prostaglandins, leading to edema and swelling. The animals are divided into groups, including a control group, a standard drug group (treated with an anti-inflammatory agent like diclofenac), and test groups treated with different doses of the extract. Paw volume is measured at regular intervals after induction, using a Plethysmometer or vernier caliper, to assess the extent of inflammation and the efficacy of the treatment. This model reliably simulates acute inflammatory conditions and allows for the observation of the anti-inflammatory potential of the plant extract under controlled experimental conditions.

ANIMALS

Wistar albino rats of both sexes (180-220 g) were used for the study. The animals were obtained from the animal house Jeeva life sciences All the rats were kept in standard plastic rat cages with stainless steel coverlids and wheat straw was used as bedding material. The animals were kept at the animal house The animals were facilitated with standard environmental condition of photoperiod (12:12 h dark: light cycle) and temperature (25 ± 2°C). They were provided with commercial rat and mice feed and water given ad libitum. The use of these animals and the study protocols were approved by CPCSEA recognized local ethical committee.

Pre-treatment

Before conducting the anti-inflammatory study, healthy adult Wistar albino rats (180–220 g) were selected and acclimatized to laboratory conditions for at least seven days with free access to standard diet and water. The animals were randomly divided into groups including normal control, negative control, standard (e.g., indomethacin), and test groups receiving different doses of the ethanolic extract. The test extract was prepared using an appropriate vehicle such as 1% carboxymethylcellulose (CMC) and administered orally using a gavage needle. The animals were fasted overnight prior to treatment, with water allowed ad libitum, to ensure accurate absorption of the test drug. Pre-treatment was done 30 to 60 minutes before the induction of inflammation, depending on the specific experimental model. Ethical guidelines were strictly followed throughout the study, and prior approval was obtained from the Institutional Animal Ethics Committee (IAEC). Care was taken to minimize stress to the animals by gentle handling during the dosing and treatment process.

Selection of the Doses for Animal Study

Oral acute toxicity study will be done as per standard procedure (OECD 404) to select the dose for this study. Acute toxicity studies were performed by using OECD guide lines (Organization of Economic Cooperation and Development) 423. It's a step wise procedure with 3 female animals per step. Depending upon the mortality and/or morbidity status of animals, the average of 2-3 steps are necessary to allow judgment on the acute toxicity of the test substance. It results in the use of minimum number of animals while allowing for acceptable data based scientific conclusion. The procedure uses defined doses (2000 mg/kg body weight) and the result allows a test substance to be ranked and classified according to the Globally Harmonized System (GHS) for the classification of chemicals which cause acute toxicity.

Experimental design [19]

Group I Normal control
Group II Negative control
Group III Standard diclofenac 10 mg/kg
Group IV Ipomea Hederifolia extract (low dose 200 mg/kg)
Group V Ipomea Hederifolia extract (high dose 500 mg/kg)

Induction of Inflammation

Inflammation in experimental animals can be induced using various chemical and physical agents to evaluate the anti-inflammatory activity of test drugs or plant extracts. The choice of inducer depends on whether acute or chronic inflammation is being studied.

Carrageenan-Induced Paw Edema [20]

Carrageenan-induced paw edema is the most widely used model for acute inflammation. A 0.1 mL suspension of 1% carrageenan is injected into the subplantar region of the rat hind paw, resulting in edema formation. The inflammatory response is biphasic: the early phase (0–2 h) involves the release of histamine, serotonin, and bradykinin, while the late phase (3–5 h) is mediated by prostaglandins and leukotrienes. The degree of paw swelling is measured using a plethysmometer.

The chronic anti-inflammatory activity of the ethanolic extract of *Ipomoea hederifolia* was evaluated using the cotton pellet-induced granuloma model in Wistar albino rats. Sterile cotton pellets weighing 10 ± 1 mg were prepared and autoclaved before implantation. Under light anesthesia, a small incision was made in the axillary region of each rat, and a sterile cotton pellet was implanted subcutaneously. The incision was then sutured, and the animals were allowed to recover. The rats were divided into different groups and treated orally with vehicle, standard drug (Diclofenac sodium, 10 mg/kg), or the ethanolic extract of *Ipomoea hederifolia* at selected doses once daily for seven consecutive days. On the eighth day, the animals were sacrificed, and the

cotton pellets along with the surrounding granulomatous tissue were carefully removed. The pellets were weighed to determine the wet weight and then dried in a hot air oven at 60°C for 24 hours to obtain a constant dry weight. The increase in dry weight of the pellets was considered an index of granuloma formation. The anti-inflammatory activity of the extract was assessed by comparing the reduction in granuloma weight and percentage inhibition of granuloma formation with those of the control and standard-treated groups.

Cotton Pellet-Induced [21]

The cotton pellet-induced chronic inflammation model is commonly used to evaluate the anti-inflammatory activity of drugs and plant extracts during the proliferative phase of inflammation. In this method, sterile cotton pellets weighing approximately 10 ± 1 mg are implanted subcutaneously into Wistar albino rats under light anesthesia. The implanted pellets act as foreign bodies, triggering a chronic inflammatory response characterized by fibroblast proliferation, collagen deposition, and infiltration of inflammatory cells. The test extract and standard drug are administered orally for seven consecutive days. At the end of the treatment period, the cotton pellets are removed, dried to a constant weight, and weighed. The increase in pellet weight reflects the extent of inflammatory tissue formation. A significant reduction in pellet weight in the treated groups compared with the control group indicates the anti-inflammatory potential of the test substance. This model is widely employed for assessing the efficacy of compounds against chronic inflammatory conditions.

Catalase (CAT) Assay [22]

Catalase activity was estimated by measuring the decomposition of hydrogen peroxide (H_2O_2). Briefly, tissue homogenate was prepared in ice-cold phosphate buffer (pH 7.0) and centrifuged to obtain the supernatant. The reaction mixture contained phosphate buffer, hydrogen peroxide solution, and the tissue supernatant. The decrease in absorbance due to the decomposition of hydrogen peroxide was measured spectrophotometrically at 240 nm at regular intervals. Catalase activity was expressed as units of H_2O_2 decomposed per minute per milligram of protein.

Reduced Glutathione (GSH)

Reduced glutathione levels were estimated using Ellman's reagent [5,5'-dithiobis-(2-nitrobenzoic acid), DTNB]. Tissue homogenate was prepared in phosphate buffer and precipitated with trichloroacetic acid (TCA). After centrifugation, the supernatant was collected and mixed with DTNB reagent and phosphate buffer. The yellow-colored chromogen formed due to the reaction between DTNB and sulfhydryl groups of glutathione was measured spectrophotometrically at 412 nm. The GSH concentration was calculated using a standard curve and expressed as µmol/g tissue.

Lipid Peroxidation (LPO) [23]

Lipid peroxidation was assessed by measuring malondialdehyde (MDA) levels using the Thiobarbituric Acid Reactive Substances (TBARS) method. Tissue homogenate was mixed with thiobarbituric acid (TBA), trichloroacetic acid (TCA), and hydrochloric acid (HCl). The reaction mixture was heated in a boiling water bath for 15 minutes and then cooled to room temperature. After centrifugation, the absorbance of the pink-colored chromogen formed by the reaction of MDA with TBA was measured spectrophotometrically at 532 nm. The MDA concentration was expressed as nmol of MDA formed per gram of tissue and served as an index of lipid peroxidation and oxidative stress.

Antioxidant Parameters [24]

Antioxidant parameters were estimated in tissue homogenates prepared from the excised inflammatory tissue. The tissues were washed with ice-cold normal saline, weighed, and homogenized in phosphate buffer (0.1 M, pH 7.4). The homogenate was centrifuged at 10,000 rpm for 15 minutes at 4°C, and the resulting supernatant was used for the estimation of antioxidant enzymes and oxidative stress markers.

Superoxide Dismutase (SOD) [25]

Superoxide dismutase activity was determined based on its ability to inhibit the auto-oxidation of pyrogallol. The reaction mixture consisted of Tris-HCl buffer, pyrogallol solution, and tissue supernatant. The change in absorbance was measured spectrophotometrically at 420 nm. One unit of SOD activity was defined as the amount of enzyme required to inhibit pyrogallol auto-oxidation by 50%, and the results were expressed as U/mg protein.

Catalase (CAT) [26]

Catalase activity was estimated by measuring the rate of decomposition of hydrogen peroxide (H_2O_2). The reaction mixture containing phosphate buffer, hydrogen peroxide, and tissue supernatant was incubated, and the decrease in absorbance was recorded at 240 nm using a UV-visible spectrophotometer. Catalase activity was expressed as µmoles of H_2O_2 decomposed per minute per milligram of protein.

Reduced Glutathione (GSH) [27]

Reduced glutathione levels were determined using Ellman's reagent [5,5′-dithiobis-(2-nitrobenzoic acid), DTNB]. The tissue homogenate was treated with trichloroacetic acid (TCA) and centrifuged. The supernatant was mixed with DTNB and phosphate buffer, producing a yellow-colored complex. The absorbance was measured at 412 nm, and GSH levels were expressed as µmol/g tissue.

Lipid Peroxidation (LPO/MDA) [28-32]
Lipid peroxidation was assessed by estimating malondialdehyde (MDA) levels using the Thiobarbituric Acid Reactive Substances (TBARS) method. Tissue homogenate was mixed with thiobarbituric acid (TBA), trichloroacetic acid (TCA), and hydrochloric acid (HCl), followed by heating in a boiling water bath for 15 minutes. After cooling and centrifugation, the absorbance of the resulting pink chromogen was measured at 532 nm. The MDA concentration was expressed as nmol MDA/g tissue and used as an indicator of oxidative stress.

STATIS ICAL ANALYSIS
Statistical analysis was performed using GraphPad Prism version 9.0 for windows (GraphPad Software, San digicams, and Microsoft excel 2016.Raw data obtained from different wound models are expressed as mean ±SEM. Values less than P< 0.05 were statistically significant. The data compared by one way ANOVA followed by Dunnet's test.

RESULTS:

Percentage of yield

$$Percentage\ of\ yield = \frac{Weight\ of\ crude\ extract}{Weight\ of\ dried\ material} \times 100$$

$$Percentage\ of\ yield = \frac{10g}{50g} \times 100 = 20\%$$

50gms of drug dissolved in 200ml of solvent in 1:4 Ratio

Table Preliminary qualitative phytochemical analysis

Sl. No.	Phytoconstituents	Test result
1	Alkaloid	-ve
2	Glycosides	-ve
3	Carbohydrate	-ve
4	Protein	-ve
5	Amino acid	+ve
6	Steroids	-ve
7	Flavonoids	+ve
8	Terpenoids	+ve
9	Phenols	+ve
10	Saponins	-ve
11	Tannin	+ve

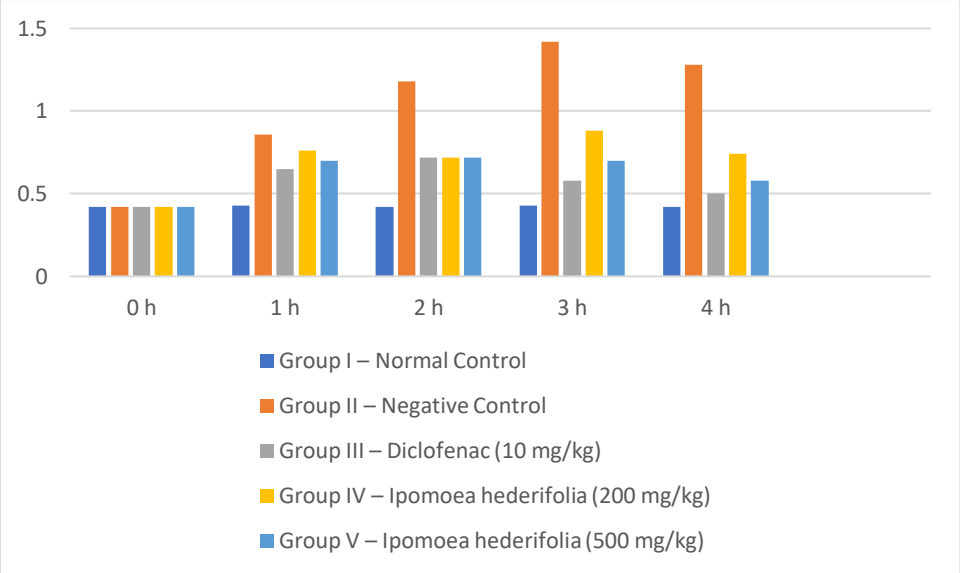
+ve: Present; -ve: Absent

Carrageenan-Induced Paw Edema

Groups	0 h	1 h	2 h	3 h	4 h
Group I – Normal Control	0.42 ± 0.01	0.43 ± 0.01	0.42 ± 0.01	0.43 ± 0.01	0.42 ± 0.01
Group II – Negative Control	0.42 ± 0.01	0.86 ± 0.03	1.18 ± 0.04	1.42 ± 0.05	1.28 ± 0.04
Group III – Diclofenac (10 mg/kg)	0.42 ± 0.01	0.65 ± 0.02***	0.72 ± 0.03***	0.58 ± 0.02***	0.50 ± 0.02***
Group IV – Ipomoea hederifolia (200 mg/kg)	0.42 ± 0.01	0.76 ± 0.03*	0.96 ± 0.03**	0.88 ± 0.03**	0.74 ± 0.03**
Group V – Ipomoea hederifolia (500 mg/kg)	0.42 ± 0.01	0.70 ± 0.02**	0.84 ± 0.03***	0.70 ± 0.02***	0.58 ± 0.02***

Carrageenan administration produced a significant increase in paw edema in the negative control group, reaching a maximum paw volume of 1.42 ± 0.05 mL at 3 h. Treatment with diclofenac sodium (10 mg/kg) significantly reduced paw swelling throughout the experimental period, showing 60.94% inhibition of edema at 4 h. The ethanolic extract of *Ipomoea hederifolia* exhibited dose-dependent anti-inflammatory activity. The low dose (200 mg/kg) reduced paw edema with 42.19%

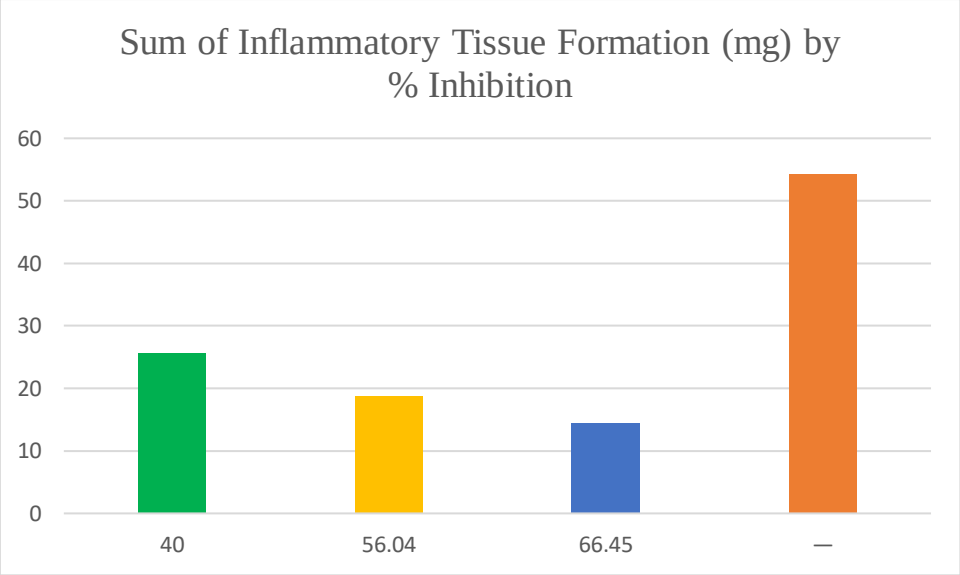
inhibition, while the high dose (500 mg/kg) showed a greater effect with 54.69% inhibition at 4 h. The results indicate that the extract possesses significant anti-inflammatory activity comparable to the standard drug, particularly at the higher dose. The observed effect may be attributed to the presence of flavonoids, phenolic compounds, and other bioactive constituents capable of inhibiting inflammatory mediator release.



Cotton Pellet-Induced

Groups	Initial Pellet Weight (mg)	Final Dry Weight (mg)	Inflammatory Tissue Formation (mg)	% Inhibition
Group I – Normal Control	10.00 ± 0.00	21.45 ± 0.78	11.45 ± 0.78	—
Group II – Negative Control	10.00 ± 0.00	52.86 ± 1.45	42.86 ± 1.45	—
Group III – Diclofenac (10 mg/kg)	10.00 ± 0.00	24.38 ± 0.92***	14.38 ± 0.92***	66.45
Group IV – Ipomoea hederifolia (200 mg/kg)	10.00 ± 0.00	35.72 ± 1.16**	25.72 ± 1.16**	40
Group V – Ipomoea hederifolia (500 mg/kg)	10.00 ± 0.00	28.84 ± 1.04***	18.84 ± 1.04***	56.04

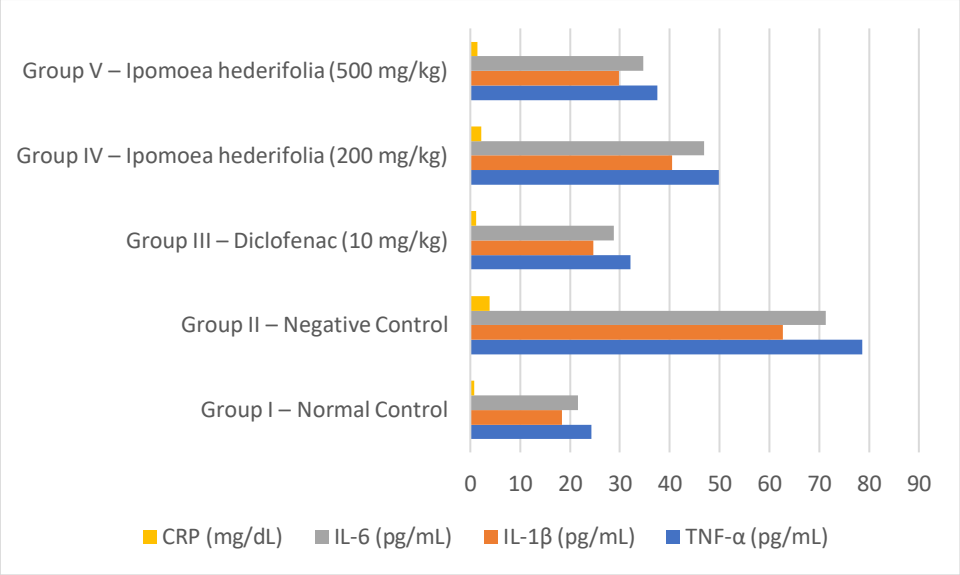
The cotton pellet-induced chronic inflammation model produced a marked increase in inflammatory tissue formation in the negative control group, with a final dry pellet weight of 52.86 ± 1.45 mg and inflammatory tissue formation of 42.86 ± 1.45 mg. Treatment with diclofenac sodium (10 mg/kg) significantly reduced inflammatory tissue formation to 14.38 ± 0.92 mg, corresponding to 66.45% inhibition. The ethanolic extract of *Ipomoea hederifolia* exhibited significant dose-dependent anti-inflammatory activity. The low dose (200 mg/kg) reduced inflammatory tissue formation to 25.72 ± 1.16 mg, showing 40.00% inhibition, whereas the high dose (500 mg/kg) further reduced tissue formation to 18.84 ± 1.04 mg, resulting in 56.04% inhibition. The higher dose demonstrated anti-inflammatory activity approaching that of the standard drug. These findings suggest that the ethanolic extract of *Ipomoea hederifolia* effectively suppresses chronic inflammatory responses and connective tissue proliferation, possibly due to the presence of flavonoids, phenolic compounds, and other bioactive phytoconstituents.



Biochemical parameters

Groups	TNF-α (pg/mL)	IL-1β (pg/mL)	IL-6 (pg/mL)	CRP (mg/dL)
Group I – Normal Control	24.35 ± 1.28	18.42 ± 0.96	21.58 ± 1.14	0.84 ± 0.05
Group II – Negative Control	78.64 ± 2.45	62.75 ± 2.08	71.32 ± 2.36	3.92 ± 0.12
Group III – Diclofenac (10 mg/kg)	32.18 ± 1.42***	24.64 ± 1.18***	28.73 ± 1.34***	1.18 ± 0.07***
Group IV – Ipomoea hederifolia (200 mg/kg)	49.82 ± 1.86**	40.46 ± 1.65**	46.88 ± 1.82**	2.14 ± 0.09**
Group V – Ipomoea hederifolia (500 mg/kg)	37.56 ± 1.54***	29.82 ± 1.32***	34.75 ± 1.48***	1.46 ± 0.08***

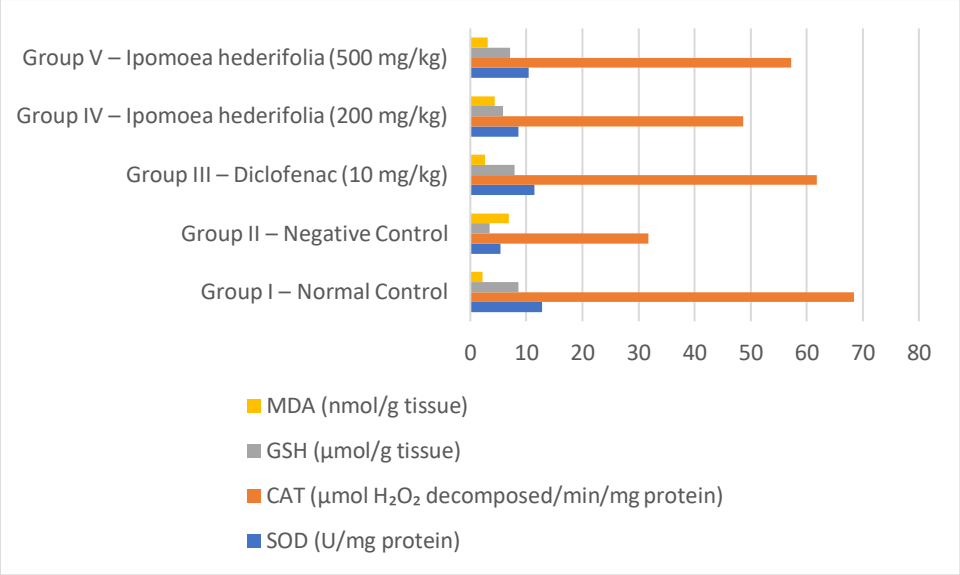
The negative control group exhibited a significant elevation in serum inflammatory biomarkers, including TNF-α, IL-1β, IL-6, and CRP levels, indicating the establishment of chronic inflammation. Treatment with diclofenac sodium (10 mg/kg) significantly reduced all biochemical parameters compared with the negative control group. The ethanolic extract of *Ipomoea hederifolia* produced a dose-dependent reduction in inflammatory cytokines and CRP levels. At 200 mg/kg, the extract significantly decreased TNF-α, IL-1β, IL-6, and CRP levels, while the 500 mg/kg dose produced a more pronounced effect, approaching that of the standard drug. The reduction in pro-inflammatory cytokines suggests that the extract suppresses inflammatory mediator release and modulates the inflammatory response. These findings support the anti-inflammatory potential of *Ipomoea hederifolia* in chronic inflammatory conditions.



Antioxidant

Groups	SOD (U/mg protein)	CAT (µmol H_2O_2 decomposed/min/mg protein)	GSH (µmol/g tissue)	MDA (nmol/g tissue)
Group I – Normal Control	12.84 ± 0.42	68.42 ± 2.15	8.62 ± 0.31	2.14 ± 0.11
Group II – Negative Control	5.36 ± 0.28	31.74 ± 1.42	3.42 ± 0.18	6.88 ± 0.24
Group III – Diclofenac (10 mg/kg)	11.42 ± 0.38***	61.85 ± 1.94***	7.86 ± 0.28***	2.68 ± 0.13***
Group IV – Ipomoea hederifolia (200 mg/kg)	8.54 ± 0.32**	48.62 ± 1.76**	5.84 ± 0.24**	4.32 ± 0.18**
Group V – Ipomoea hederifolia (500 mg/kg)	10.46 ± 0.36***	57.24 ± 1.88***	7.12 ± 0.26***	3.12 ± 0.15***

The negative control group showed a significant reduction in endogenous antioxidant enzymes, including superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH), along with a marked increase in malondialdehyde (MDA) levels, indicating enhanced oxidative stress and lipid peroxidation. Treatment with diclofenac sodium (10 mg/kg) significantly restored antioxidant enzyme activities and reduced MDA levels toward normal values. Administration of the ethanolic extract of *Ipomoea hederifolia* produced a dose-dependent improvement in antioxidant status. The low dose (200 mg/kg) significantly increased SOD, CAT, and GSH levels while decreasing MDA levels compared with the negative control group. The high dose (500 mg/kg) exhibited a more pronounced effect, closely approaching the activity of the standard drug. These results suggest that the anti-inflammatory activity of *Ipomoea hederifolia* may be partly mediated through its antioxidant properties, which help protect tissues against oxidative damage associated with chronic inflammation.



DISCUSSION:

The present study was undertaken to evaluate the anti-inflammatory activity of the ethanolic extract of *Ipomoea hederifolia* using carrageenan-induced paw edema and cotton pellet-induced chronic inflammation models in rats. The results demonstrated that the extract possesses significant anti-inflammatory activity in both acute and chronic phases of inflammation, with the higher dose (500 mg/kg) showing greater efficacy than the lower dose (200 mg/kg).

In the carrageenan-induced paw edema model, carrageenan administration produced a marked increase in paw volume in the negative control group, indicating the development of acute inflammation. Carrageenan-induced inflammation is a biphasic process, where the early phase is mediated by histamine, serotonin, and bradykinin, while the late phase is primarily associated with prostaglandin synthesis and neutrophil infiltration. Treatment with the ethanolic extract of *Ipomoea hederifolia* significantly reduced paw edema in a dose-dependent manner. The higher dose exhibited substantial inhibition of edema formation, comparable to the standard drug diclofenac. This reduction suggests that the extract may inhibit the release or action of inflammatory mediators, particularly prostaglandins, which play a crucial role in the progression of acute inflammation.

The cotton pellet-induced chronic inflammation model was employed to assess the effect of the extract on the proliferative phase of inflammation. The negative control group showed significant inflammatory tissue formation, whereas treatment with the ethanolic extract significantly reduced inflammatory tissue development. The reduction was more pronounced at the dose of 500 mg/kg, indicating its ability to suppress fibroblast proliferation, collagen deposition, and inflammatory cell infiltration. These findings suggest that the extract is effective not only in controlling exudative inflammation but also in inhibiting chronic inflammatory processes associated with tissue proliferation.

Biochemical analysis further supported the anti-inflammatory activity of the extract. Elevated levels of pro-inflammatory cytokines such as TNF-α, IL-1β, IL-6, and CRP were observed in the negative control group, confirming the inflammatory state. Administration of the ethanolic extract significantly reduced these inflammatory biomarkers in a dose-dependent manner. The suppression of these cytokines indicates that the extract may modulate inflammatory signaling pathways and reduce the production of mediators responsible for the maintenance and progression of inflammation.

Oxidative stress is known to contribute significantly to inflammatory tissue damage. In the present study, chronic inflammation resulted in decreased levels of endogenous antioxidants, including superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH), along with increased malondialdehyde (MDA) levels, a marker of lipid peroxidation. Treatment with the ethanolic extract restored antioxidant enzyme levels and significantly reduced MDA concentrations. These findings indicate that the extract possesses potent antioxidant activity, which may contribute to its anti-inflammatory effects by scavenging free radicals and protecting tissues from oxidative damage.

The observed pharmacological effects of *Ipomoea hederifolia* may be attributed to the presence of bioactive phytoconstituents such as flavonoids, phenolic compounds, tannins, alkaloids, and terpenoids. Flavonoids and phenolic compounds

are known to inhibit cyclooxygenase and lipoxygenase pathways, suppress cytokine production, and neutralize reactive oxygen species. Therefore, the anti-inflammatory activity observed in this study is likely the result of the combined action of these phytochemicals.

Overall, the findings of the present investigation demonstrate that the ethanolic extract of *Ipomoea hederifolia* possesses significant anti-inflammatory and antioxidant activities in experimental models of acute and chronic inflammation. The higher dose (500 mg/kg) showed greater efficacy and produced effects comparable to those of diclofenac sodium. These results provide scientific evidence supporting the traditional use of *Ipomoea hederifolia* in inflammatory conditions and suggest its potential as a source of novel anti-inflammatory agents. Further studies are warranted to isolate and characterize the active constituents responsible for these pharmacological effects and to elucidate their precise mechanisms of action.

CONCLUSION:

The present study demonstrated that the ethanolic extract of *Ipomoea hederifolia* possesses significant anti-inflammatory activity in both acute and chronic experimental models of inflammation. In the carrageenan-induced paw edema model, the extract effectively reduced paw swelling in a dose-dependent manner, indicating its ability to suppress acute inflammatory responses. Similarly, in the cotton pellet-induced chronic inflammation model, the extract significantly decreased inflammatory tissue formation, suggesting inhibition of the proliferative phase of inflammation.

Biochemical investigations revealed that treatment with the extract significantly reduced the levels of pro-inflammatory cytokines such as TNF-α, IL-1β, IL-6, and C-reactive protein (CRP). Furthermore, the extract enhanced endogenous antioxidant defenses by increasing superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH) levels while decreasing malondialdehyde (MDA), a marker of lipid peroxidation. These findings indicate that the anti-inflammatory effect of *Ipomoea hederifolia* is associated with both the suppression of inflammatory mediators and the attenuation of oxidative stress. Among the tested doses, the higher dose (500 mg/kg) exhibited greater efficacy and produced effects comparable to those of the standard drug diclofenac sodium. The observed pharmacological activities may be attributed to the presence of bioactive phytoconstituents such as flavonoids, phenolic compounds, tannins, and terpenoids present in the ethanolic extract. In conclusion, the ethanolic extract of *Ipomoea hederifolia* possesses potent anti-inflammatory and antioxidant properties and may serve as a promising natural therapeutic agent for the management of inflammatory disorders. Further studies are required to isolate the active constituents, elucidate their molecular mechanisms, and establish their safety and efficacy through clinical investigations.

REFERENCES:

1. Medzhitov R. Origin and physiological roles of inflammation. *Nature*. 2008;454(7203):428–35.
2. Nathan C. Points of control in inflammation. *Nature*. 2002;420(6917):846–52.
3. Ferrero-Miliani L, Nielsen OH, Andersen PS, Girardin SE. Chronic inflammation: importance of NOD2 and NALP3 in interleukin-1β generation. *Clin Exp Immunol*. 2007;147(2):227–35.
4. Libby P. Inflammatory mechanisms: the molecular basis of inflammation and disease. *Nutr Rev*. 2007;65(12 Pt 2):S140–6.
5. Chen L, Deng H, Cui H, Fang J, Zuo Z, Deng J, et al. Inflammatory responses and inflammation-associated diseases in organs. *Oncotarget*. 2018;9(6):7204–18.
6. Serhan CN, Savill J. Resolution of inflammation: the beginning programs the end. *Nat Immunol*. 2005;6(12):1191–7.
7. Vane JR, Botting RM. Mechanism of action of anti-inflammatory drugs. *Int J Tissue React*. 1998;20(1):3–15.
8. Ricciotti E, FitzGerald GA. Prostaglandins and inflammation. *Arterioscler Thromb Vasc Biol*. 2011;31(5):986–1000.
9. Dinarello CA. Proinflammatory cytokines. *Chest*. 2000;118(2):503–8.
10. Lawrence T. The nuclear factor NF-κB pathway in inflammation. *Cold Spring Harb Perspect Biol*. 2009;1(6):a001651.
11. Gilroy DW, Lawrence T, Perretti M, Rossi AG. Inflammatory resolution: new opportunities for drug discovery. *Nat Rev Drug Discov*. 2004;3(5):401–16.
12. Barnes PJ. Anti-inflammatory actions of glucocorticoids: molecular mechanisms. *Clin Sci (Lond)*. 1998;94(6):557–72.
13. Vane JR. Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nat New Biol*. 1971;231(25):232–5.
14. Botting RM. Mechanism of action of aspirin-like drugs. *Postgrad Med J*. 2000;76(895):242–4.
15. Wallace JL. Prostaglandins, NSAIDs, and gastric mucosal protection: why doesn't the stomach digest itself? *Physiol Rev*. 2008;88(4):1547–65.
16. Grosser T, Smyth E, FitzGerald GA. Anti-inflammatory, antipyretic, and analgesic agents; pharmacotherapy of gout. In: Brunton LL, editor. *Goodman & Gilman's The Pharmacological Basis of Therapeutics*. 13th ed. New York: McGraw-Hill; 2018.

17. Calder PC, Ahluwalia N, Brouns F, Buetler T, Clement K, Cunningham K, et al. Dietary factors and low-grade inflammation in relation to overweight and obesity. *Br J Nutr*. 2011;106(S3):S5–78.
18. Henson PM. Dampening inflammation. *Nat Immunol*. 2005;6(12):1179–81.
19. Gęgotek A, Skrzydlewska E. Antioxidative and anti-inflammatory activity of ascorbic acid. Antioxidants. 2022 Oct 7;11(10):1993.
20. Singh T, Singh P, Pandey VK, Singh R, Dar AH. A literature review on bioactive properties of betel leaf (Piper betel L.) and its applications in food industry. Food chemistry advances. 2023 Dec 1; 3:100536.
21. Darmawan A, Yusuf S, Tahir T, Syahriyani S. Betel leaf extract efficacy on wound healing: a systematic review. STRADA: Jurnal Ilmiah Kesehatan. 2021 May 2;10(1):526-36.
22. Winter CA, Risley EA, Nuss GW. Carrageenin-induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. *Proc Soc Exp Biol Med*. 1962;111(3):544-547.
23. Vinegar R, Schreiber W, Hugo R. Biphasic development of carrageenin edema in rats. *J Pharmacol Exp Ther*. 1969;166(1):96-103.
24. Swingle KF, Shideman FE. Phases of the inflammatory response to subcutaneous implantation of cotton pellet and their modification by certain anti-inflammatory agents. *J Pharmacol Exp Ther*. 1972;183(1):226-234.
25. D'Arcy PF, Howard EM, Muggleton PW, Townsend SB. The anti-inflammatory action of griseofulvin in experimental animals. *J Pharm Pharmacol*. 1960;12:659-665.
26. Bradley JR. TNF-mediated inflammatory disease. *J Pathol*. 2008;214(2):149-160.
27. Aggarwal BB. Signalling pathways of the TNF superfamily: a double-edged sword. *Nat Rev Immunol*. 2003;3(9):745-756.
28. Dinarello CA. Overview of the IL-1 family in innate inflammation and acquired immunity. *Immunol Rev*. 2018;281(1):8-27.
29. Marklund S, Marklund G. Involvement of the superoxide anion radical in the auto-oxidation of pyrogallol and a convenient assay for superoxide dismutase. *Eur J Biochem*. 1974;47(3):469-474.
30. McCord JM, Fridovich I. Superoxide dismutase: an enzymatic function for erythrocuprein. *J Biol Chem*. 1969;244(22):6049-6055.
31. llman GL. Tissue sulfhydryl groups. *Arch Biochem Biophys*. 1959;82(1):70-77.
32. Sedlak J, Lindsay RH. Estimation of total, protein-bound, and non-protein sulfhydryl groups in tissue with Ellman's reagent. *Anal Biochem*. 1968;25:192-205.