

Exploring the Anti-Inflammatory Properties of *Nyctanthes Arbor-Tristis*: Formulation Approaches and Therapeutic Potential

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ABSTRACT

Inflammation is a complex physiological response underlying a wide range of acute and chronic diseases. Conventional anti-inflammatory therapies, including NSAIDs and corticosteroids, are effective but associated with adverse effects and long-term complications. This review explores the potential of *Nyctanthes arbor-tristis* (NAT), a traditional medicinal plant widely used in Ayurveda, as a natural alternative for anti-inflammatory treatment. NAT contains diverse phytoconstituents such as iridoid glycosides (e.g., arbortristoid-A), flavonoids (quercetin, astragalin), phenolic acids, tannins, and essential oils that exhibit anti-inflammatory, antioxidant, and immunomodulatory properties. These compounds modulate key inflammatory pathways by inhibiting COX and LOX enzymes, suppressing pro-inflammatory cytokines (e.g., IL-6, TNF- α), and scavenging reactive oxygen species. Formulation strategies including gels, tablets, and nanoformulations have been explored to enhance the solubility, bioavailability, and stability of NAT extracts. Evaluation methods—such as physicochemical assessments (pH, viscosity, spreadability), in vitro drug release, stability studies, and animal models—confirm the therapeutic efficacy and safety of NAT-based preparations. While preclinical studies demonstrate promising dose-dependent anti-inflammatory activity comparable to standard drugs, clinical data remain limited. Challenges such as batch variability, lack of phytochemical standardization, and limited regulatory recognition need to be addressed. The future of NAT lies in integrating advanced drug delivery systems (e.g., nanoparticles, transdermal patches), synergistic herbal-synthetic combinations, and clinical trials to validate its use as a modern phytopharmaceutical. This review provides a foundation for further research and development toward evidence-based utilization of *Nyctanthes arbor-tristis* in anti-inflammatory therapy.

Keywords: *Nyctanthes arbor-tristis*, anti-inflammatory, herbal gel, Phytopharmaceuticals, cyclooxygenase inhibition, nano formulation, traditional medicine

INTRODUCTION

Inflammation is a complex biological response of body tissues to harmful stimuli such as pathogens, damaged cells, or irritants. It serves as a protective mechanism intended to remove injurious stimuli and initiate the healing process. However, chronic or excessive inflammation contributes to the pathogenesis of numerous disorders including

rheumatoid arthritis, atherosclerosis, and cancer [1,2]. Nonsteroidal anti-inflammatory drugs (NSAIDs) and corticosteroids are commonly used to manage inflammation. Despite their efficacy, long-term use of these agents is associated with a spectrum of adverse effects such as gastrointestinal ulcers, renal dysfunction, cardiovascular risks, and immunosuppression [3]. The development of drug resistance and contraindications in certain populations

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necessitates safer alternatives. Herbal medicines have garnered global interest due to their minimal side effects, bioavailability, and cost-effectiveness. Phytochemicals such as flavonoids, terpenes, and phenolic compounds are known to exhibit significant anti-inflammatory properties through various mechanisms including inhibition of cyclooxygenase enzymes (COX), suppression of pro-inflammatory cytokines, and free radical scavenging [4]. Commonly referred to as “night-flowering jasmine” or “Parijat,” *Nyctanthes arbor-tristis* (NAT) belongs to the Oleaceae family and is distributed across India and Southeast Asia. Traditionally used in Ayurvedic

medicine, NAT has demonstrated anti-inflammatory, antioxidant, antimicrobial, antipyretic, and hepatoprotective effects [5,6]. These properties are attributed to its rich phytochemical composition, which includes flavonoids, iridoid glycosides, tannins, and saponins [7]. This review aims to comprehensively summarize the phytochemical profile, pharmacological properties, formulation, and evaluation of anti-inflammatory gel derived from *Nyctanthes arbor-tristis*. Special focus is placed on the preparation and effectiveness of NAT-based herbal gel as an alternative to conventional topical anti-inflammatory agents.



Fig.01: *Nyctanthes arbor-tristis* Plant

2. Botanical Description of *Nyctanthes Arbor-Tristis*

2.1 Taxonomical Classification

Table 1: Taxonomical Classification of *Nyctanthes arbor-tristis*

Taxonomic Rank	Classification
Kingdom	Plantae
Subkingdom	Viridiplantae
Infrakingdom	Streptophyta
Superdivision	Embryophyta
Division	Tracheophyta
Subdivision	Spermatophytina
Order	Lamiales
Family	Oleaceae
Genus	<i>Nyctanthes</i>
Species	<i>Nyctanthes arbor-tristis</i> L.

2.2 Geographical Distribution

Nyctanthes arbor-tristis is native to the **Indian subcontinent** and is extensively found in **South Asia** and **Southeast Asia**, including countries such as India, Nepal, Bangladesh, Sri Lanka, Thailand, and parts of Malaysia [9]. It thrives in warm, humid

climates and is often found in gardens, temples, and near dwellings due to its fragrant and culturally significant flowers. It grows well in loamy and clayey soils and is relatively drought-resistant. The plant prefers semi-shade but can tolerate full sunlight and is typically grown between altitudes of **10–1500 meters** above sea level [10].

2.3 Morphological Features

Nyctanthes arbor-tristis is a shrub or small tree that can grow up to 10 meters in height. It is characterized by gray, flaky bark and stiff, slightly drooping branches. The leaves are arranged oppositely on the stem and are simple in structure, with an ovate-lanceolate shape measuring between 6 to 12 centimeters in length. The upper surface of the leaves is rough, while the underside is pale. The plant bears white, fragrant flowers that grow in small clusters known as cymes. A distinguishing feature of these flowers is their bright orange corolla tube. They bloom during the night and typically fall off by early morning, which has earned the plant the poetic name "tree of sorrow." The fruit of *Nyctanthes arbor-tristis* is a flattened capsule, ranging in color from brown to green, usually two-lobed and containing a single seed in each lobe [11,12]. The unique floral morphology contributes to its cultural significance and easy identification, while the leaves and bark are most commonly utilized in traditional and herbal medicinal preparations.

2.4 Traditional Uses in Ayurveda and Folk Medicine

Nyctanthes arbor-tristis holds a prominent place in Ayurvedic and folk medicine due to its diverse therapeutic applications. In classical Ayurvedic literature, it is known to balance the Vata and Kapha doshas. Its taste, or *rasa*, is described as *tikta* (bitter) and *kashaya* (astringent), while its qualities (*guna*) are *laghu* (light) and *ruksha* (dry). The plant is considered to have *ushna virya* (hot potency) and exhibits a *katu vipaka* (pungent post-digestive effect), making it suitable for a range of pathological conditions. Traditionally, various parts of the plant have been employed for multiple medicinal purposes. The leaves and bark are known for their anti-inflammatory properties and are used to treat conditions such as arthritis, gout, and localized swellings. A decoction made from the leaves is commonly used as an antipyretic to reduce fever. Both seeds and leaves serve as effective anthelmintic agents to expel intestinal parasites. The juice extracted from the leaves is traditionally administered for its hepatoprotective benefits, particularly in managing jaundice and liver disorders. *Nyctanthes arbor-tristis* also finds use in the treatment of respiratory ailments

including chronic cough, bronchitis, and asthma. For dermatological conditions, pastes made from the leaves are applied externally to manage skin eruptions, wounds, and boils. Additionally, the plant is used as an adjuvant in the traditional treatment of malaria and intermittent fevers. These traditional uses have been increasingly supported by modern pharmacological research, which has confirmed the plant's significant anti-inflammatory, immunomodulatory, and antioxidant properties [13–16].

3. Phytochemical Constituents of *Nyctanthes Arbor-Tristis*

Nyctanthes arbor-tristis (NAT), known commonly as night-flowering jasmine or "Parijat," is a phytochemically rich medicinal plant used extensively in Ayurvedic, Siddha, and folk medicine systems. Its anti-inflammatory activity is attributed to a broad spectrum of bioactive compounds, notably iridoid glycosides, flavonoids, phenolic compounds, tannins, and essential oils. These compounds exhibit significant pharmacological effects, including the inhibition of pro-inflammatory enzymes and cytokines, reduction of oxidative stress, and promotion of tissue repair [17,18].

3.1 Overview of Major Bioactive Compounds

3.1.1 Iridoid glycosides

Iridoid glycosides, a class of monoterpenoid compounds, are among the major bioactive constituents of *Nyctanthes arbor-tristis* and are recognized for their strong anti-inflammatory, antipyretic, and analgesic properties. One such compound is nyctanthin, a carotenoid-based glycoside primarily found in the flower petals of the plant. Nyctanthin exhibits notable antioxidant and immune-modulatory activities, contributing to the plant's therapeutic effects. Another key iridoid glycoside is arbortristoside-A, which is isolated from the seeds of *Nyctanthes arbor-tristis*. This compound has shown significant anti-inflammatory activity in experimental models, particularly through its ability to inhibit edema and inflammatory exudate formation. The mechanism of action involves the suppression of various inflammatory mediators, including histamine, serotonin, and prostaglandins. These iridoid

glycosides exert a dual-phase inhibitory effect in inflammation models, effectively targeting both the early vascular phase and the later cellular phase of the inflammatory response, thereby demonstrating comprehensive anti-inflammatory potential [19,20].

3.1.2 Flavonoids

Flavonoids are a class of plant-derived phenolic compounds widely recognized for their potent antioxidant and anti-inflammatory activities. In *Nyctanthes arbor-tristis* (NAT), several key flavonoids contribute to its therapeutic effects. Quercetin, one of the major flavonoids present, functions by inhibiting pro-inflammatory enzymes such as lipoxygenase and cyclooxygenase-2 (COX-2), thereby suppressing the synthesis of inflammatory mediators. Another important flavonoid, kaempferol, exerts its anti-inflammatory action by inhibiting the nuclear factor kappa B (NF- κ B) signaling pathway, which plays a central role in regulating immune and inflammatory responses. Additionally, compounds such as astragalin and nicotiflorin have been shown to reduce the expression of pro-inflammatory cytokines and protect tissues from oxidative damage. Collectively, these flavonoids act by scavenging reactive oxygen species (ROS), thereby minimizing tissue inflammation and preserving endothelial function. Their ability to modulate oxidative stress and inflammatory signaling makes them particularly beneficial in managing chronic inflammatory conditions such as arthritis [21,22].

3.1.3 Phenolic Compounds

Phenolic compounds play a significant role in contributing to the antioxidant and antimicrobial properties of *Nyctanthes arbor-tristis* (NAT), as well as in modulating inflammatory pathways. These compounds assist in the downregulation of key inflammatory markers such as interleukins and tumor necrosis factor-alpha (TNF- α), which are central to the inflammatory response. Among the primary phenolic constituents identified in NAT are nyctanthic acid, ascorbic acid, and oleanolic acid. These molecules are known to inhibit lipid peroxidation, a process that contributes to cellular damage during inflammation, and help stabilize cell membranes, thereby protecting tissues from inflammation-induced injury [23]. Additionally, their

lipophilic nature enhances their ability to penetrate the skin, making them particularly effective in topical or gel-based formulations by increasing local bioavailability at the site of application.

3.1.4 Tannins

Tannins, a group of polyphenolic compounds found in the leaves and bark of *Nyctanthes arbor-tristis* (NAT), are chiefly responsible for the plant's astringent and protective properties. These compounds exert notable pharmacological effects by inhibiting key enzymes such as inducible nitric oxide synthase (iNOS) and cyclooxygenase (COX), both of which play central roles in the inflammatory cascade. Through this mechanism, tannins help suppress the production of pro-inflammatory mediators. Additionally, they promote vasoconstriction and reduce vascular permeability, effectively preventing the formation of edema in inflamed tissues [24]. Beyond their anti-inflammatory functions, the tannins in NAT also exhibit strong antimicrobial activity, making them valuable as natural preservatives in various herbal formulations. Their dual role in both therapeutic action and formulation stability underscores their importance in traditional medicine and phytopharmaceutical applications.

3.1.5 Essential Oils

Although present in smaller concentrations compared to other phytochemicals, the essential oils extracted from the flowers and leaves of *Nyctanthes arbor-tristis* (NAT) exhibit noteworthy bioactivity. These oils contain key components such as linalool, β -caryophyllene, and geraniol, which are known for their antimicrobial, anti-inflammatory, and mood-stabilizing properties [25]. These volatile compounds act in synergy with flavonoids and phenolic constituents to enhance the modulation of inflammatory responses, making them particularly effective in topical applications such as skin formulations. Furthermore, the pleasant aroma and volatile nature of these essential oils contribute to improved user compliance, especially when incorporated into gels or lotions for therapeutic or cosmetic use.

3.2 Extraction Techniques and Solvents Used

The efficacy of a plant-based formulation is highly dependent on the extraction method and solvent system employed, as these factors influence the yield, purity, and biological potency of the extracted bioactive compounds. In the case of *Nyctanthes arbor-tristis* (NAT), various extraction techniques have been utilized to isolate specific phytoconstituents for medicinal applications. One of the traditional methods is maceration, wherein dried and powdered leaves or flowers of NAT are soaked in solvent mixtures at room temperature for several days with intermittent agitation. Common solvents used in this process include ethanol, methanol, water, and hydroalcoholic mixtures (typically a 70:30 ethanol-water ratio), which are particularly effective in extracting flavonoids and glycosides [26]. Soxhlet extraction, a continuous hot extraction method, is another widely used technique. In this process, the solvent is repeatedly evaporated and condensed over the powdered plant material, allowing efficient extraction of phenolics, tannins, and iridoid glycosides. Solvents such as ethanol, methanol, and acetone are typically preferred for this method. Ultrasound-assisted extraction (UAE) represents a more modern and environmentally friendly approach. This technique employs ultrasonic waves to rupture plant cell walls, enhancing solvent penetration and improving phytochemical yield. UAE is especially suitable for extracting thermolabile compounds like essential oils and certain flavonoids without causing degradation [27]. Supercritical fluid extraction (SFE) utilizes supercritical carbon dioxide (CO₂) to extract non-polar essential oils. This advanced method ensures high purity and minimal thermal damage, making it ideal for isolating volatile oil fractions from NAT [28]. Ultimately, the choice of extraction technique depends on the target compound and the intended application, whether for topical gels or oral formulations. Hydroalcoholic extracts are most frequently used in anti-inflammatory herbal gels due to their ability to capture a broad spectrum of active constituents.

4. Mechanism Of Anti-Inflammatory Action Of *Nyctanthes Arbor-Tristis*

The anti-inflammatory properties of *Nyctanthes arbor-tristis* (NAT) are primarily attributed to its diverse array of phytoconstituents, including iridoid glycosides, flavonoids, phenolic compounds, and

tannins. These constituents interact with molecular targets involved in the inflammatory cascade, including pro-inflammatory cytokines, enzymes like cyclooxygenase (COX) and lipoxygenase (LOX), and reactive oxygen species (ROS) [29,30]. Their combined actions result in the attenuation of inflammation through multiple pathways.

4.1 Role of Phytoconstituents in Inflammation Modulation

The phytoconstituents present in *Nyctanthes arbor-tristis* (NAT) exert their anti-inflammatory effects through synergistic and multi-targeted mechanisms, making the plant a potent modulator of both acute and chronic inflammation. Iridoid glycosides, particularly arbortristiside-A, play a key role by modulating the activity of immune cells such as macrophages and lymphocytes, thereby reducing the formation of inflammatory exudates and limiting tissue damage [31]. Flavonoids, including quercetin and astragalin, contribute to the anti-inflammatory response by stabilizing cellular membranes, inhibiting neutrophil degranulation, and decreasing vascular permeability, which collectively help to control edema and prevent further inflammatory cascades [32]. Phenolic compounds enhance this action by suppressing oxidative stress and preventing the activation of nuclear factor-kappa B (NF- κ B), a pivotal transcription factor involved in the upregulation of pro-inflammatory cytokines [33]. Additionally, tannins provide astringent effects that help reduce local swelling and maintain tissue integrity by forming protective complexes with proteins on the surface of inflamed tissues [34]. Together, these bioactive constituents interact across multiple inflammatory pathways to provide a broad-spectrum, balanced modulation of the body's inflammatory response.

4.2 Inhibition of Cyclooxygenase (COX) and Lipoxygenase (LOX) Pathways

One of the central mechanisms underlying inflammation is the metabolism of arachidonic acid through the cyclooxygenase (COX) and lipoxygenase (LOX) enzyme pathways, which leads to the production of key pro-inflammatory mediators such as prostaglandins and leukotrienes. Extracts from *Nyctanthes arbor-tristis* (NAT) have been shown to

inhibit the activity of COX-2, resulting in a significant reduction in the synthesis of prostaglandin E2 (PGE2). This prostaglandin is a major contributor to inflammation-associated symptoms such as fever, pain, and vasodilation [35]. Concurrently, NAT also inhibits the LOX pathway, leading to decreased levels of leukotriene B4, a potent chemotactic agent responsible for recruiting neutrophils to the site of inflammation [36]. By targeting both COX and LOX pathways, NAT mimics the dual inhibition strategy employed by certain modern nonsteroidal anti-inflammatory drugs (NSAIDs), providing a more comprehensive anti-inflammatory response while potentially minimizing the gastrointestinal and cardiovascular side effects typically associated with selective COX inhibition.

4.3 Modulation of Pro-inflammatory Cytokines (IL-6, TNF- α)

Pro-inflammatory cytokines such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF- α) play a central role in the pathogenesis of chronic inflammation and autoimmune disorders. *Nyctanthes arbor-tristis* (NAT) has demonstrated significant potential in modulating these key inflammatory mediators. Studies conducted both in vitro and in animal models have shown that NAT extracts markedly reduce the levels of IL-6 and TNF- α , thereby attenuating the inflammatory response [37]. This effect is largely attributed to the presence of flavonoids in NAT, which inhibit the activation of nuclear factor-kappa B (NF- κ B), a critical transcription factor responsible for the expression of various pro-inflammatory cytokines. Additionally, iridoid glycosides present in the plant influence the mitogen-activated protein kinase (MAPK) signaling

pathway, leading to the downregulation of cytokine-related gene expression [38]. Through these combined mechanisms, NAT effectively suppresses both systemic and localized inflammation, reinforcing its value as a natural anti-inflammatory agent in managing chronic inflammatory conditions.

4.4 Antioxidant Activity Contributing to Anti-inflammatory Effects

Oxidative stress plays a crucial role in the development and persistence of inflammation, as reactive oxygen species (ROS) can activate redox-sensitive transcription factors such as NF- κ B and AP-1. These transcription factors, in turn, promote the release of pro-inflammatory cytokines and the infiltration of immune cells into affected tissues. *Nyctanthes arbor-tristis* (NAT) exhibits strong antioxidant properties that contribute significantly to its anti-inflammatory activity. It is rich in compounds such as ascorbic acid, quercetin, and oleanolic acid, all of which are known for their ability to neutralize free radicals and protect tissues from oxidative damage [39]. In addition to directly scavenging ROS, these bioactive molecules also enhance the body's endogenous antioxidant defense system by upregulating enzymes like superoxide dismutase (SOD) and glutathione peroxidase (GPx), which play vital roles in maintaining redox balance [40]. By mitigating oxidative stress, NAT helps suppress inflammatory signaling pathways, thus providing therapeutic benefits in conditions characterized by chronic or autoimmune inflammation. The plant's antioxidant potential, therefore, represents a key mechanism in its broad-spectrum anti-inflammatory action.

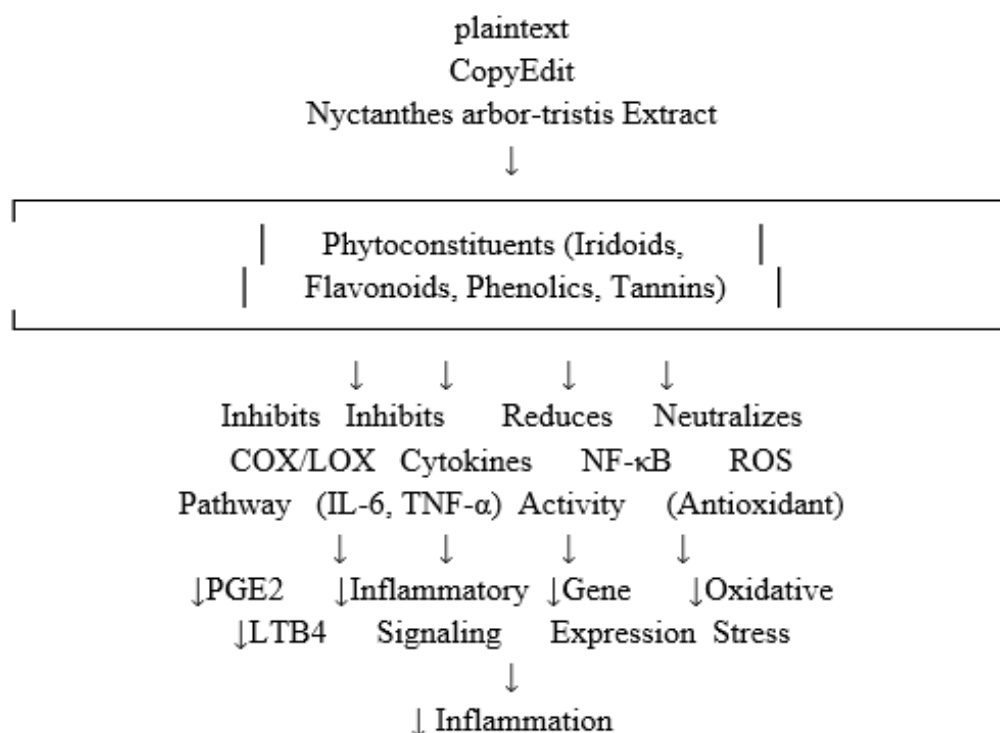


Fig.02: Summary Diagram of Anti-inflammatory Mechanism

5. Preclinical and Clinical Studies on The Anti-Inflammatory Activity of *Nyctanthes Arbor-Tristis*

The therapeutic potential of *Nyctanthes arbor-tristis* (NAT) as an anti-inflammatory agent has been widely explored in **preclinical in vitro and in vivo models**, with emerging interest in human clinical studies. These studies demonstrate not only the mechanistic underpinnings of NAT's bioactivity but also validate its **dose-dependent effects, comparative efficacy**, and **safety** profiles.

5.1 Summary of In Vitro and In Vivo Studies Demonstrating Anti-inflammatory Activity

Numerous in vitro and in vivo studies have demonstrated the significant anti-inflammatory potential of *Nyctanthes arbor-tristis* (NAT). In vitro experiments have revealed that NAT extracts effectively inhibit key pro-inflammatory mediators. For instance, Siddiqui et al. (2023) identified iridoid glycosides in NAT leaves that suppress the activity of cyclooxygenase-2 (COX-2) and lipoxygenase (LOX) in RAW 264.7 macrophage cells. This inhibition led to a marked reduction in the production of prostaglandin E2 and leukotrienes, both of which are central to the inflammatory process [41]. Similarly, Goswami et al. (2017) reported a downregulation of

TNF- α , IL-1 β , and COX-2 expression in lipopolysaccharide (LPS)-induced inflammation models treated with NAT flower extract, confirming the plant's multi-targeted action within the inflammatory cascade [42]. In vivo studies further support these findings. Saxena et al. (1987) demonstrated that NAT leaf extract significantly inhibited carrageenan-induced paw edema and cotton pellet granuloma formation in rats, with anti-inflammatory effects comparable to those of indomethacin [43]. More recently, Saini et al. (2021) isolated ursolic acid from NAT and showed its efficacy in reducing inflammation in a mouse model of oxidative stress-induced arthritis, thereby confirming its systemic anti-inflammatory activity [44]. Additionally, Naik et al. (2020) formulated a phytosomal gel containing NAT and *Tagetes patula*, which effectively reduced D-galactose-induced skin inflammation in mice by modulating inflammatory markers and oxidative enzymes [45]. Together, these in vitro and in vivo findings provide strong evidence that NAT possesses both acute and chronic anti-inflammatory properties.

5.2 Dose-dependent Effects

Preclinical studies have consistently demonstrated that the anti-inflammatory effects of *Nyctanthes*

arbor-tristis (NAT) extracts are dose-dependent. In a carrageenan-induced paw edema model, the ethanolic extract of NAT administered at doses of 200 mg/kg and 400 mg/kg resulted in 48% and 71% inhibition of inflammation, respectively, within four hours. In contrast, a lower dose of 100 mg/kg produced only minimal effects, indicating a clear dose-response relationship [43]. Similarly, arbortristoxide-A, a major iridoid glycoside isolated from NAT, showed significant anti-inflammatory activity when administered to mice at doses ranging from 25 to 100 mg/kg. The most effective reduction in histamine-induced inflammation was observed at an optimal dose of 75 mg/kg [46]. Across various rodent models, the effective oral dose range for NAT extracts is generally between 200 and 400 mg/kg, while topical formulations typically contain NAT at concentrations of 5–10% w/w. Importantly, these doses have not been associated with any significant toxicity, supporting the safety and therapeutic potential of NAT in both systemic and topical applications.

5.3 Comparative Efficacy with Standard Anti-inflammatory Agents

Comparative studies have indicated that extracts of *Nyctanthes arbor-tristis* (NAT) exhibit anti-inflammatory efficacy comparable to, and in some cases even exceeding, that of conventional synthetic drugs in specific experimental settings. In a study conducted by Saxena et al., an ethanolic extract of NAT administered at a dose of 400 mg/kg produced an anti-inflammatory effect in rats that was on par with indomethacin administered at 10 mg/kg, highlighting its potential as a natural alternative to standard NSAIDs [43]. Similarly, Siddiqui et al. reported that iridoid glycosides isolated from NAT demonstrated COX inhibitory activity nearly equivalent to that of diclofenac in biochemical assays, but with notably lower cytotoxicity, suggesting a better safety profile [41]. In another comparative study, Naik et al. (2020) evaluated a gel formulation containing NAT against hydrocortisone cream in a model of dermal inflammation. The results revealed that the NAT-based gel was equally effective in reducing inflammation, yet did not cause adverse effects such as skin irritation, which are commonly associated with corticosteroid use [45]. These findings collectively suggest that NAT possesses substantial therapeutic potential as an anti-

inflammatory agent and may serve as a safer alternative to NSAIDs and corticosteroids, particularly in topical formulations or for patients with sensitivities to conventional pharmaceuticals.

5.4 Any Available Human Clinical Trials

Currently, human clinical trials evaluating *Nyctanthes arbor-tristis* (NAT) as a standardized anti-inflammatory formulation are limited. However, some preliminary studies provide insights into its potential therapeutic applications. In a study conducted by Godse et al. (2018), NAT extracts were administered as adjunct therapy to malaria patients. Although the primary focus was not on inflammatory biomarkers, the treatment group exhibited notable improvements in secondary outcomes such as reduced fever, joint inflammation, and spleen enlargement, suggesting systemic anti-inflammatory activity [47]. Additionally, Naik et al. (2020) evaluated the dermatological safety of a NAT-based phytosomal gel in human volunteers. Over a 7-day period, the topical application of the gel resulted in no signs of skin irritation or hypersensitivity, supporting its safety profile for topical use [45]. Despite these encouraging findings, there remains a need for well-designed randomized controlled trials to evaluate the clinical efficacy, pharmacokinetics, and long-term safety of NAT in humans, particularly in the context of inflammatory disorders.

6. Formulation Strategies Using *Nyctanthes arbor-tristis* (NAT)

The formulation of herbal products containing *Nyctanthes arbor-tristis* (NAT) involves multiple considerations, including **delivery route**, **bioavailability**, **solubility**, and **phytochemical stability**. Advances in herbal technology have expanded NAT formulation approaches beyond traditional decoctions to modern **gels**, **capsules**, **nanoemulsions**, and **transdermal systems**, enhancing therapeutic efficacy and patient compliance.

6.1 Herbal Formulation Approaches

Herbal formulation approaches involving *Nyctanthes arbor-tristis* (NAT) have evolved to accommodate both traditional and modern delivery systems aimed

at maximizing its anti-inflammatory potential. Among these, gels and ointments have gained particular attention due to their suitability for localized therapy. The incorporation of NAT leaf and flower extracts into gel formulations using carbopol 940 as a gelling agent has resulted in stable, easily spreadable products. For instance, Londhe et al. (2025) developed a topical gel combining NAT with *Azadirachta indica*, which exhibited significant anti-inflammatory and antimicrobial activity without causing skin irritation [48]. Gels are advantageous for their ease of application, rapid onset of action, and ability to bypass first-pass metabolism, making them ideal for treating localized inflammatory conditions. In addition to topical applications, NAT has been formulated into oral dosage forms such as tablets and capsules. These preparations, often containing powdered leaves or ethanolic extracts, are widely used in Ayurvedic medicine for the treatment of chronic arthritis, fevers, and liver disorders. However, formulation challenges persist due to the limited aqueous solubility and poor oral bioavailability of some iridoid glycosides present in NAT, which can affect therapeutic efficacy [49]. To overcome these limitations, advanced nanoformulation strategies have been explored. Jamdagni et al. (2011) synthesized zinc oxide nanoparticles using NAT flower extracts, which demonstrated enhanced antifungal and anti-inflammatory properties [50]. Additionally, Naik et al. (2020) developed a phytosome-based gel containing carotenoids from NAT, significantly improving skin retention and transdermal absorption [51]. These innovative systems offer controlled release, targeted delivery, and improved stability of phytoconstituents, thereby enhancing the overall therapeutic value of NAT formulations.

6.2 Solubility, Bioavailability, and Stability Considerations

Formulating herbal preparations with *Nyctanthes arbor-tristis* (NAT) requires addressing several physicochemical challenges, particularly in terms of solubility, bioavailability, and stability. One of the major issues is the poor water solubility of several key bioactive constituents, such as oleanolic acid and arbortristosiide-A, which are hydrophobic in nature. To enhance their solubility, hydroalcoholic solvent systems or surfactants are commonly employed during extraction and formulation processes [52].

Another significant challenge is the limited oral bioavailability of many active compounds due to their poor intestinal permeability and susceptibility to enzymatic degradation in the gastrointestinal tract. Advanced formulation strategies, such as encapsulation within liposomes or phytosomes, have been shown to improve absorption by protecting these compounds and facilitating their transport across biological membranes [53]. Stability is also a critical concern, as NAT phytochemicals are sensitive to environmental factors like light exposure, temperature fluctuations, and pH changes. To ensure product integrity and prolong shelf life, formulations often include protective excipients such as polymers, chelating agents, and antioxidants like vitamin E [54]. Additionally, proper packaging and storage—particularly in amber-colored containers and under refrigerated conditions—further help to preserve the phytochemical profile and therapeutic efficacy of NAT-based preparations over time.

6.3 Role of Excipients and Delivery Systems

Excipients play a vital role in enhancing the delivery, stability, and overall efficacy of *Nyctanthes arbor-tristis* (NAT)-based formulations. Gelling agents such as Carbopol 940 and methylcellulose are commonly used to provide the desired gel consistency and ensure effective adherence to the skin, which is essential for topical applications. To facilitate transdermal absorption of the active constituents, penetration enhancers like propylene glycol and glycerin are incorporated, allowing for improved delivery of phytochemicals through the skin barrier. Maintaining an appropriate pH is also crucial for ensuring skin compatibility; triethanolamine is frequently employed to regulate the pH of formulations, typically maintaining it within a skin-friendly range of 5.5 to 7. In order to prevent microbial contamination and extend shelf life, natural preservatives such as neem extract and ascorbic acid are used due to their antimicrobial and antioxidant properties [55,56]. Beyond traditional excipients, the adoption of advanced delivery systems has further improved the performance of NAT formulations. Systems such as hydrogels, emulgels, and lipid-based nanoparticles offer improved bioadhesion, controlled release, and enhanced retention of the active compounds at the target site. These modern delivery platforms are particularly beneficial in dermatological applications,

where localized and sustained anti-inflammatory action is desired. By integrating appropriate excipients with innovative delivery technologies, NAT-based formulations can achieve greater therapeutic efficacy and patient compliance.

6.4 Examples of Existing NAT-based Formulations

A few studied and marketed formulations (mainly in India and Southeast Asia) demonstrate NAT's application in both traditional and modern formats:

Formulation Type	Example/Product	Key Use	Reference
Topical Herbal Gel	NAT + Neem Gel (Londhe et al., 2025)	Skin inflammation, wounds	[48]
Phytosomal Anti-aging Gel	NAT + <i>Tagetes patula</i> (Naik et al., 2020)	Anti-aging, oxidative stress-induced skin	[51]
Ayurvedic Capsule	NAT + Ashwagandha (various manufacturers)	Joint inflammation, fever	[56]
ZnO-Nanoparticles	NAT flower-based nanoparticle	Antimicrobial, anti-inflammatory	[50]

These examples highlight the versatility of NAT in **both traditional remedies and contemporary pharmaceutical formulations**.

7. Evaluation of Nat-Based Anti-Inflammatory Formulations

For any herbal formulation to be therapeutically effective and clinically viable, it must undergo rigorous evaluation to ensure its **quality, stability, bioavailability, and pharmacological efficacy**. Formulations containing *Nyctanthes arbor-tristis* (NAT), particularly herbal gels, have been tested using both **physicochemical** and **pharmacodynamic** parameters. This section outlines the standard techniques used for assessing NAT-based anti-inflammatory products.

7.1 Physicochemical Evaluations

The evaluation of physicochemical parameters is essential for assessing the quality and effectiveness of topical formulations containing *Nyctanthes arbor-tristis* (NAT). One of the most critical attributes is pH, which directly affects skin compatibility. Ideally, the pH of a topical product should range between 5.5 and 7.0 to match the skin's natural acidic environment. In a study by Londhe et al. (2025), a pH value of 6.8 was reported for a NAT-based herbal gel, confirming its suitability for dermal application with minimal risk of irritation [57]. Viscosity is another key parameter, as it influences the formulation's spreadability, drug release profile, and overall patient acceptability. It is typically measured using a Brookfield viscometer. The optimal viscosity ensures that the gel maintains

structural stability while remaining sufficiently fluid to allow easy application and absorption [58]. Spreadability, which is commonly assessed using the slip and drag method, determines how well the formulation can be applied across the skin. NAT-based gel formulations demonstrated spreadability values in the range of 6.1 to 6.5 g·cm/sec, indicating favorable spreading characteristics that enhance patient compliance and ensure uniform drug distribution [57]. In terms of homogeneity and appearance, a high-quality gel should exhibit a smooth, lump-free texture with no signs of phase separation. NAT-based formulations were observed to be homogeneous, non-greasy, and aesthetically pleasing, with a pleasant aroma attributed to the inclusion of essential oils. These attributes collectively contribute to the formulation's efficacy, stability, and user acceptability [59].

7.2 Drug Release Studies

Drug release studies, also known as in vitro diffusion studies, are critical for evaluating how effectively active phytoconstituents are released from a gel base and become bioavailable at the site of inflammation. These studies help in understanding the release kinetics and potential therapeutic performance of the formulation. The Franz diffusion cell is the most commonly employed apparatus for such evaluations, utilizing either a cellophane membrane or excised animal skin to simulate the skin barrier. In a recent study by Londhe et al. (2025), the NAT-based herbal gel demonstrated a controlled and sustained release of key bioactive compounds, including arbortristiside-A and flavonoids, over a period of six hours [57]. The

cumulative percentage of drug released is typically plotted against time to analyze the release pattern, which can follow various kinetic models such as zero-order, first-order, or the Higuchi diffusion model. These data provide essential insights into the formulation's ability to deliver therapeutic concentrations of active constituents at the site of action over a desired time frame, supporting its use in managing localized inflammatory conditions.

7.3 Stability Studies

Stability studies are essential to determine whether a formulation retains its efficacy, physical appearance, pH, viscosity, and microbial purity over time when subjected to various storage conditions. These assessments help ensure product safety, performance, and shelf life. Two primary types of stability tests are commonly conducted: accelerated and long-term studies. Accelerated stability testing involves storing the formulation at elevated conditions, typically $40 \pm 2^\circ\text{C}$ and $75 \pm 5\%$ relative humidity (RH) for a duration of three months. In contrast, long-term stability testing is performed under more standard conditions, usually at $25 \pm 2^\circ\text{C}$ and $60 \pm 5\%$ RH, and may extend from six to twelve months. Key parameters monitored during these studies include pH stability, where acceptable drift is limited to ± 0.5 units to ensure skin compatibility. Visual assessments are also conducted to check for color changes or phase separation, both of which would indicate formulation instability. Microbial purity is assessed to confirm the absence of microbial growth, thereby validating the effectiveness of preservatives used in the formulation. In a study conducted by Londhe et al. (2025), the NAT-based herbal gel demonstrated excellent physical and chemical stability under accelerated conditions over a three-month period. The formulation maintained its pH within the acceptable range, exhibited no significant changes in color or texture, showed no phase separation, and remained free from microbial contamination. These findings support the product's stability and suitability for long-term use [57].

7.4 Anti-inflammatory Activity Testing

The anti-inflammatory activity of *Nyctanthes arbor-tristis* (NAT) formulations has been extensively validated through both in vivo and in vitro models. In

vivo, rodent models are commonly used to evaluate the efficacy of NAT in acute and chronic inflammatory conditions. One of the most widely accepted models for assessing acute inflammation is carrageenan-induced paw edema in rats. In a study conducted by Londhe et al. (2025), the application of a NAT-based gel significantly reduced paw swelling by 58.3% after four hours, a result that was comparable to the reduction observed with diclofenac gel, which achieved a 61.4% decrease in edema [57]. For evaluating chronic inflammation, the cotton pellet-induced granuloma model is employed, which measures the proliferative phase of inflammation. In this model, ethanolic extracts of NAT incorporated into gel formulations led to a statistically significant reduction in granuloma mass ($p < 0.01$) when compared to the placebo group, indicating potent anti-proliferative and anti-inflammatory effects [60]. In vitro studies further support these findings by examining the molecular mechanisms underlying NAT's anti-inflammatory activity. Lipopolysaccharide (LPS)-stimulated RAW 264.7 macrophage cells are frequently used to assess the suppression of inflammatory mediators such as nitric oxide (NO), tumor necrosis factor-alpha (TNF- α), interleukin-6 (IL-6), and cyclooxygenase-2 (COX-2). Siddiqui et al. (2023) reported that iridoid glycosides extracted from NAT inhibited NO production by 65% and suppressed COX-2 expression by over 70% in these cells [61]. These findings confirm that NAT is capable of modulating both enzymatic and cytokine-mediated inflammatory pathways, demonstrating its potential as a broad-spectrum anti-inflammatory agent.

8. Toxicological and Safety Profile

An essential aspect of herbal drug development is the evaluation of safety and toxicity. Despite its long-standing use in traditional medicine, the toxicological profiling of *Nyctanthes arbor-tristis* (NAT) is critical to establish its **therapeutic window, dosage limits, and regulatory acceptability** for modern formulations. The available **in vivo and in vitro studies** support a favorable safety profile, although high-dose or chronic use may present specific concerns.

8.1 Acute and Chronic Toxicity Studies



Acute and chronic toxicity studies have provided valuable insights into the safety profile of *Nyctanthes arbor-tristis* (NAT). In terms of acute toxicity, several preclinical investigations have demonstrated that NAT extracts are well-tolerated at high doses. For instance, Sharma et al. (2012) reported that oral administration of NAT leaf extract at doses up to 2,000 mg/kg in Wistar rats did not result in any signs of toxicity, mortality, or behavioral changes over a 14-day observation period, indicating that the plant is non-toxic at therapeutic levels [62]. Similarly, Naik et al. (2020) assessed the dermal safety of a NAT-based phytosomal gel and found no evidence of skin irritation, redness, or hypersensitivity reactions in both rabbit models and human volunteers, supporting its suitability for topical use [63]. Chronic toxicity studies, although limited, are beginning to emerge. Saini et al. (2021) conducted a 28-day subchronic toxicity study in which rats received daily doses of NAT ethanolic extract ranging from 100 to 400 mg/kg. The results showed no significant changes in liver and kidney function tests, hematological parameters, or organ weights, suggesting a favorable safety profile for long-term use [64]. Moreover, histopathological examinations of major organs—including the liver, kidney, heart, and lungs—revealed no cellular abnormalities, further confirming the non-toxic nature of NAT when administered at moderate doses over extended periods. Collectively, these findings support the safe therapeutic use of NAT in both topical and oral formulations.

8.2 LD₅₀ Data

The median lethal dose (LD₅₀) is a critical parameter used to assess the acute toxicity of a substance, representing the dose required to cause death in 50% of a test population. While specific LD₅₀ data for humans are not available, preclinical studies have provided valuable estimates in animal models for *Nyctanthes arbor-tristis* (NAT). In rodent studies, the oral LD₅₀ for the ethanolic extract of NAT has been determined to be greater than 2,000 mg/kg, which classifies the plant extract as having low toxicity under the OECD 423 guidelines, placing it in Category 5 [65]. This categorization suggests a wide margin of safety when used at therapeutic doses. Although a precise LD₅₀ value for topical application has not been established, formulations such as herbal gels and ointments containing NAT have been

deemed safe at commonly used concentrations, with no reported adverse dermal effects. These findings support the classification of NAT as Generally Recognized as Safe (GRAS) in traditional and modern herbal formulations, reinforcing its suitability for both oral and topical use in medicinal applications.

8.3 Reported Side Effects or Contraindications

Nyctanthes arbor-tristis (NAT) is generally well-tolerated, but some mild side effects and contraindications have been reported. Mild gastrointestinal irritation may occur when NAT decoctions or tablets are consumed on an empty stomach and in large quantities, which suggests the importance of adhering to recommended dosages and administration guidelines [66]. In preclinical studies, high doses exceeding 500 mg/kg have occasionally resulted in mild lethargy or drowsiness in rodents, likely due to the presence of sedative alkaloids. Regarding contraindications, NAT is not recommended for use during pregnancy because of its potential uterotonic activity, although this effect has not been confirmed in human clinical trials [67]. Individuals with hypotension or known sensitivities to herbal products are also advised to use NAT cautiously until more comprehensive safety data become available. Despite these considerations, the side effects associated with NAT are typically minimal and self-limiting, particularly when the plant is used in topical formulations such as gels, where systemic absorption is limited. This favorable safety profile supports its continued use in herbal medicine, especially for localized anti-inflammatory applications.

8.4 Regulatory Status

Nyctanthes arbor-tristis holds a recognized position in traditional medical systems such as Ayurveda, Unani, and Siddha, and is officially listed in the Ayurvedic Pharmacopoeia of India (API) [68]. Within India's regulatory framework, the Ministry of AYUSH has approved NAT for medicinal use in herbal formulations, and a comprehensive monograph is available in the API, detailing its therapeutic applications and preparation standards. In the United States, while NAT is not currently approved by the Food and Drug Administration (FDA) as a botanical drug, it may be incorporated into dietary supplements

under the Dietary Supplement Health and Education Act (DSHEA), provided it is accompanied by appropriate safety data and complies with labeling requirements. In Europe, NAT is regarded as a traditional herbal substance; however, no official monograph has yet been published by the European Medicines Agency (EMA). Regardless of the jurisdiction, formulations containing NAT must adhere to general pharmaceutical standards, including Good Manufacturing Practices (GMP), proper labeling, validated safety testing, and stability and microbial quality assessments. The absence of significant regulatory barriers, particularly for topical use, makes NAT an attractive candidate for incorporation into modern phytopharmaceutical and cosmeceutical products. Its established traditional use, coupled with favorable safety data, supports its potential for broader integration into evidence-based herbal medicine.

CHALLENGES AND LIMITATIONS

Despite the promising pharmacological and formulation-based evidence supporting the anti-inflammatory potential of *Nyctanthes arbor-tristis* (NAT), several **critical challenges and limitations** impede its advancement into mainstream therapeutics. These issues largely pertain to the **standardization, quality control, solubility, phytochemical variability**, and the **lack of high-quality clinical trials**. Addressing these challenges is essential to ensure reproducibility, regulatory compliance, and clinical efficacy.

9.1 Standardization and Quality Control of Herbal Extracts

One of the major challenges in the development of *Nyctanthes arbor-tristis* (NAT)-based formulations is the standardization of herbal extracts, which is essential for ensuring consistent identity, potency, and purity across different production batches. NAT contains a diverse range of active phytoconstituents, including arbortristoxide-A, nyctanthin, and quercetin. The relative concentrations of these bioactive compounds can vary significantly depending on several factors such as geographical origin, climatic conditions, harvesting season, the specific plant part used, and the method of extraction employed [69,70]. This inherent variability

complicates the formulation of products with predictable therapeutic efficacy. Furthermore, the lack of validated analytical markers and established reference standards makes it difficult to perform accurate quantitative assessments of the active compounds. This limitation hampers the ability to define dose-response relationships and undermines quality assurance processes. Another contributing factor to inconsistency is the insufficient documentation and implementation of Good Agricultural and Collection Practices (GACP), which leads to significant batch-to-batch variability and limits the scalability of NAT-based products for commercial use [71]. To address these challenges, advanced analytical and molecular techniques are increasingly being recognized as essential. High-performance liquid chromatography (HPLC), liquid chromatography–mass spectrometry (LC-MS/MS), and DNA barcoding offer precise methods for identity verification, quantification of bioactive compounds, and detection of adulterants. The adoption of these technologies is critical for improving the reproducibility, reliability, and regulatory compliance of NAT formulations intended for therapeutic applications.

9.2 Poor Solubility or Bioavailability

One of the primary pharmacokinetic challenges associated with *Nyctanthes arbor-tristis* (NAT)-derived compounds is their poor aqueous solubility and limited oral bioavailability. Key constituents such as iridoid glycosides and flavonoids are inherently hydrophobic, which hampers their dissolution and absorption within the gastrointestinal tract [72]. As a result, systemic availability of these compounds is significantly reduced when administered orally, often necessitating higher doses or repeated dosing to achieve therapeutic effects. Although topical formulations offer an advantage by bypassing first-pass metabolism, they are not without limitations, particularly in terms of skin penetration and sustained retention at the target site. These barriers can compromise the overall efficacy of NAT-based treatments. To address these issues, recent advancements in drug delivery systems have shown considerable promise. Nanoformulations—including phytosomes, liposomes, and polymeric nanoparticles—have been developed to enhance the solubility, permeability, and bioavailability of NAT

phytoconstituents [73]. These systems improve target-site delivery by increasing the surface area for absorption, protecting active compounds from degradation, and facilitating controlled release. Such innovations represent a significant step forward in optimizing the therapeutic potential of NAT and expanding its clinical utility.

9.3 Variability in Phytoconstituent Concentrations

The concentration of phytoconstituents in *Nyctanthes arbor-tristis* (NAT) is subject to significant variability, influenced by both intrinsic and extrinsic factors. Intrinsic factors include the age of the plant, the specific part of the plant used, and genetic differences among plant populations. Extrinsic factors encompass environmental conditions such as soil composition, irrigation practices, methods of drying, and the choice of extraction solvents [74]. This variability presents a substantial challenge in the standardization of herbal formulations, as it complicates accurate dosage determination, toxicological risk assessment, and cross-study comparison of pharmacological outcomes. For instance, studies have shown that the quercetin content in NAT leaves can vary widely, ranging from 0.3% to 1.2% w/w depending on the geographical location of the collection site [75]. Such inconsistency poses difficulties in meeting the stringent specifications required by regulatory bodies for herbal medicinal products, which demand precise quantification and quality control of active constituents. To address these issues, the integration of chemometric modeling and batch fingerprinting techniques is becoming increasingly essential. These advanced analytical tools can help monitor and manage phytochemical variability, ensuring consistency, reproducibility, and regulatory compliance in the development of NAT-based therapeutic products.

9.4 Lack of Clinical Data

Although preclinical studies have consistently demonstrated significant anti-inflammatory activity of *Nyctanthes arbor-tristis* (NAT) in both in vitro and in vivo models, there is a notable lack of robust clinical data to support its therapeutic use in humans. Only a handful of exploratory clinical studies have been conducted, primarily focusing on NAT's effects

in conditions such as malaria and skin inflammation. These studies have been limited by small sample sizes, poor methodological rigor, and the absence of standardized formulations, making it difficult to draw definitive conclusions about efficacy and safety [76]. Critically, there is a complete absence of randomized controlled trials (RCTs), dose-ranging investigations, and pharmacokinetic evaluations, all of which are essential for the clinical translation of NAT-based formulations. This gap in clinical research severely hampers efforts to validate the therapeutic potential of NAT and obstructs its pathway toward regulatory approval as a standardized anti-inflammatory agent. Furthermore, the long-term safety of NAT, especially when used systemically, has not been thoroughly investigated. The potential for drug-herb interactions, which could impact the pharmacodynamics or pharmacokinetics of co-administered medications, also remains largely unexplored. In the absence of well-designed human trials and regulatory-grade safety data, integrating NAT-based anti-inflammatory formulations into mainstream clinical practice remains a significant challenge. Advancing this promising botanical into evidence-based medicine will require strategic investment in rigorous clinical research to support its efficacy, safety, and therapeutic positioning.

10. Future Prospects and Research Directions

The transition of *Nyctanthes arbor-tristis* (NAT) from traditional medicine to mainstream anti-inflammatory therapeutics requires **multidisciplinary research** spanning pharmacognosy, nanotechnology, pharmacology, and clinical sciences. Although preclinical evidence is robust, future investigations should focus on **formulation innovation, synergistic combinations, clinical validation, and integration with existing therapies** to fully harness NAT's potential in modern medicine.

10.1 Use of Advanced Delivery Systems

Modern drug delivery systems offer innovative approaches to overcome the inherent limitations of *Nyctanthes arbor-tristis* (NAT), particularly its poor solubility, low stability, and limited bioavailability. Among these, nanoparticle-based and nanoemulsion delivery systems have shown considerable promise in enhancing the pharmacological performance of

NAT's active constituents. Nanocarriers such as polymeric nanoparticles, nanostructured lipid carriers (NLCs), and nanoemulsions are capable of improving the solubility of hydrophobic compounds and facilitating better skin permeation. These systems not only increase the surface area for absorption but also provide controlled and sustained release, which can significantly enhance therapeutic outcomes. In a notable example, Jamdagni et al. (2011) synthesized zinc oxide (ZnO) nanoparticles using flower extracts of NAT and demonstrated a marked increase in antimicrobial and antioxidant activity compared to conventional formulations. This study highlights how nanotechnology can potentiate the bioefficacy of plant-derived compounds by enhancing their stability and biological interaction at the cellular level [78]. Overall, the integration of advanced delivery systems into NAT-based formulations represents a critical step forward in optimizing its clinical utility and expanding its application in modern phytomedicine.

10.2 Combination with Other Herbal or Synthetic Agents

Combining *Nyctanthes arbor-tristis* (NAT) with other herbal or synthetic agents offers a promising approach to enhance therapeutic efficacy through synergistic mechanisms. In the realm of herbal medicine, NAT has been investigated in combination with botanicals such as *Tagetes patula*, *Azadirachta indica*, and *Curcuma longa*. These combinations have been shown to improve the overall antioxidant and anti-inflammatory potential of formulations, leveraging the complementary phytochemical profiles and bioactivities of each component [80]. In addition to herbal pairings, NAT has also been explored in conjunction with synthetic drugs, particularly nonsteroidal anti-inflammatory drugs (NSAIDs) such as diclofenac. Co-formulating NAT with NSAIDs may allow for dose reduction of the synthetic agent, potentially minimizing common adverse effects like gastrointestinal irritation, without compromising therapeutic efficacy [81]. However, such multi-component formulations necessitate rigorous compatibility studies to ensure there are no negative interactions between the constituents. Standardization of the ingredient ratios is also essential to maintain consistent efficacy, safety, and formulation stability across batches. By carefully designing and validating these combination therapies, NAT can be effectively

integrated into more potent and safer treatment regimens for inflammatory and oxidative stress-related conditions.

10.3 Clinical Trials for Human Validation

Despite the promising results observed in preclinical studies, there remains a significant gap in clinical validation for *Nyctanthes arbor-tristis* (NAT) formulations. Human trials are currently limited, and rigorous clinical research is essential to establish the safety, efficacy, and therapeutic applicability of NAT in inflammatory conditions. Future investigations should prioritize well-designed randomized controlled trials (RCTs) with adequate sample sizes and appropriate blinding to evaluate the effectiveness of NAT-based formulations in conditions such as osteoarthritis, eczema, and rheumatoid arthritis. These trials would help determine therapeutic outcomes and guide clinical use. In addition to efficacy trials, pharmacokinetic and pharmacodynamic studies are needed to better understand the absorption, distribution, metabolism, and elimination of NAT's active constituents in the human body. Such data are critical for defining optimal dosing regimens and ensuring systemic safety. Furthermore, specific dermal safety trials should be conducted to assess the effects of NAT-based gel formulations under repeated-dose conditions and long-term exposure, particularly for chronic inflammatory skin disorders. While preliminary evidence from an exploratory study by Godse et al. (2018) indicated that NAT extract helped reduce inflammatory symptoms in patients with malaria-induced splenomegaly, no large-scale or controlled clinical trials have been conducted since that time [82]. This underscores the urgent need for structured human studies to validate the translational potential of NAT and facilitate its integration into evidence-based phytotherapeutic protocols.

10.4 Integration in Modern Anti-inflammatory Therapy

With the growing demand for natural, non-toxic, and sustainable therapeutic options, *Nyctanthes arbor-tristis* (NAT) emerges as a highly promising phytomedicinal candidate for integration into modern anti-inflammatory therapy. Its broad spectrum of anti-inflammatory, antioxidant, and antimicrobial

activities positions it well for inclusion in a variety of healthcare applications. One of the most compelling avenues for integration lies in the field of cosmeceuticals. NAT-based gels and creams have shown potential in managing inflammatory skin conditions such as acne, eczema, psoriasis, and even signs of skin aging, offering a gentle yet effective alternative to synthetic corticosteroids and chemical-based formulations. In the realm of nutraceuticals, standardized extracts of NAT can be formulated into oral capsules aimed at managing chronic, low-grade systemic inflammation, which is implicated in conditions like metabolic syndrome, cardiovascular disease, and arthritis. Furthermore, NAT has potential as an adjunct therapy when used alongside conventional anti-inflammatory medications. In particular, it could provide added benefit in patients who are at risk for gastrointestinal or renal complications associated with prolonged NSAID use, thereby allowing for lower doses or reduced dependency on synthetic agents [83]. To fully realize these applications, regulatory frameworks and pharmacovigilance systems must adapt to support the safe and standardized use of plant-based therapies. NAT can serve as a model for the integration of botanicals into evidence-based, personalized, and holistic treatment strategies, bridging the gap between traditional medicine and modern clinical practice.

CONCLUSION

Nyctanthes arbor-tristis (NAT) has emerged as a potent candidate in the realm of herbal anti-inflammatory therapeutics. Rich in iridoid glycosides, flavonoids, tannins, and phenolic compounds, NAT exhibits multi-targeted pharmacological actions including inhibition of COX and LOX enzymes, suppression of pro-inflammatory cytokines, and strong antioxidant activity. These mechanisms collectively support its effectiveness in mitigating both acute and chronic inflammatory conditions. Modern formulation techniques—such as topical gels, capsules, and nanoformulations—have enhanced the solubility, stability, and bioavailability of NAT extracts. Evaluation studies confirm the physicochemical integrity, controlled drug release, dermal safety, and significant anti-inflammatory efficacy of NAT-based formulations, particularly in topical applications. Importantly, NAT has demonstrated favorable toxicological profiles in both

acute and chronic settings, with high LD₅₀ values and minimal adverse effects, affirming its safety for therapeutic use. However, challenges remain, including variability in phytoconstituent content, lack of standardized extraction protocols, and insufficient clinical trial data. These gaps limit the scalability and regulatory acceptance of NAT-based products. Addressing these issues through robust quality control, advanced delivery systems (e.g., nanoparticles, transdermal patches), and well-designed human trials is imperative. Future integration of NAT into evidence-based medicine can position it as a complementary or even alternative approach to conventional anti-inflammatory drugs, especially in populations seeking safer, plant-based remedies. Overall, this comprehensive review underscores the therapeutic promise of *Nyctanthes arbor-tristis* and provides a foundational platform for future translational research and clinical innovation.

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