

Pharmacognostic and Pharmacological Profile of *Albizia amara roxb* and *Achyranthes aspera linn*

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Abstract

Albizia amara and *Achyranthes aspera* are widely recognized in traditional medicine for their therapeutic properties. *Albizia amara*, belonging to the Fabaceae family, exhibits anti-inflammatory, antimicrobial, antioxidant, and hepatoprotective activities, whereas *Achyranthes aspera*, a member of Amaranthaceae, is reported for its diuretic, antidiabetic, wound-healing, and immunomodulatory effects. This chapter provides a comprehensive pharmacognostic and pharmacological analysis of these two medicinal plants, integrating modern scientific approaches and novel findings. Detailed botanical descriptions, including macro- and microscopic characteristics, physicochemical parameters, and powder microscopy, establish reliable identification criteria. Phytochemical profiling reveals a rich diversity of bioactive secondary metabolites such as flavonoids, saponins, alkaloids, triterpenoids, and phenolic acids, which are correlated with their pharmacological activities. Advanced extraction and chromatographic techniques and standardization of major bioactive constituents.

Pharmacological studies underscore the therapeutic potential of both species. *Albizia amara* demonstrates significant antioxidant activity through free radical scavenging and lipid peroxidation inhibition, alongside anti-inflammatory effects mediated via COX and NF-κB pathway modulation. Its antimicrobial efficacy spans Gram-positive and Gram-negative bacteria, and it exhibits hepatoprotective effects in preclinical models of chemically induced liver damage. *Achyranthes aspera* shows hypoglycemic activity through modulation of insulin secretion and glucose uptake, wound-healing potential via collagen synthesis enhancement, and immunomodulatory effects by regulating cytokine expression. Mechanistic insights highlight molecular targets, signaling pathways, and structure–activity relationships that underpin these pharmacological outcomes. Toxicological evaluations confirm their relative safety at therapeutic doses, though further chronic and clinical investigations are warranted.

The chapter also emphasizes ethnobotanical significance, tracing traditional uses across Ayurveda, Siddha, and folk medicine systems, and compares them with modern pharmacological

evidence. Novel perspectives include exploration of synergistic effects with other herbal constituents, potential for nanoformulation-based delivery, and implications for drug development and nutraceutical applications. Research gaps are identified in clinical translation, standardization, bioavailability enhancement, and sustainable sourcing. By integrating pharmacognostic validation with contemporary pharmacological studies, this work provides a robust scientific foundation for future research and therapeutic development of *Albizia amara* and *Achyranthes aspera*.

Keywords

Albizia amara, *Achyranthes aspera*, phytochemistry, flavonoids, pharmacological activity, anti-inflammatory, antioxidant, antimicrobial, hepatoprotective, toxicology, ethnomedicine.

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Introduction

Medicinal plants have been used across cultures for many centuries. Their role has been preserved through traditional systems, household practices, and community knowledge. Healthcare has been supported by these plants in regions where clinical services are limited. Remedies have been prepared through simple methods. Leaves, roots, bark, and flowers have been selected based on symptoms and local experience. Such knowledge has been transferred orally across generations. Regional healing systems such as Ayurveda, Siddha, and Unani have been shaped by these plants. Their pharmacological potential has been recognized through long-term observation. *Albizia amara* and *Achyranthes aspera* have been placed among important species due to wide use and broad activity. Their parts have been applied for inflammatory disorders, digestive problems, respiratory issues, wounds, and infections.

A large portion of the global population has been served by traditional medicine. Medicinal plants have been used because they are accessible, inexpensive, and familiar. They have been trusted for primary healthcare. Their therapeutic effects have been reported repeatedly across regions. Preparations have been made with simple tools and techniques. Decoctions, powders, pastes, and juices have been prepared in households. Plant-based management has been

practiced for fever, cough, pain, digestive issues, skin diseases, and chronic disorders. Equivalent synthetic drugs have not always been available. Plant-based care has filled this gap.

Scientific evaluation has been expanded to confirm these practices. Bioactive molecules have been isolated from herbal sources. Diverse structures have been observed. These structures have been linked with antioxidant, antimicrobial, anti-inflammatory, analgesic, hepatoprotective, and immunomodulatory effects. The study of *Albizia amara* and *Achyranthes aspera* has been increased due to their ethnomedicinal importance. Both species have been examined through phytochemical screening, pharmacological studies, and toxicity evaluation. Their traditional claims have been supported by experimental data.

Medicinal plants have been integrated into national and regional healthcare programs. Standardized extracts have been prepared. Quality control parameters have been established through pharmacognostic studies. Identification features have been noted to prevent adulteration. Regulatory agencies have required safety and efficacy data. Many plants have been documented for official pharmacopoeias. *Albizia amara* and *Achyranthes aspera* have been included in several ethnobotanical surveys. Their therapeutic uses have been reported in villages across India and neighboring countries.

The role of plants in chronic disease management has been strengthened. Oxidative stress and inflammation have been recognized as major contributors to many diseases. Natural antioxidants have been explored extensively. Plant-derived polyphenols, flavonoids, alkaloids, glycosides, tannins, and saponins have been analyzed. *Albizia amara* and *Achyranthes aspera* have been shown to contain such compounds. Their extracts have been tested in vitro and in vivo. Reduction of oxidative markers has been reported. Inhibition of inflammatory mediators has been observed. These activities have been linked with their traditional use for wounds, joint pain, gastric irritation, and infections.

Albizia amara belongs to the Fabaceae family. It has been distributed widely across dry and semi-arid regions. Its bark, leaves, and flowers have been used in traditional therapeutics. Decoctions of its bark have been administered for diarrhea, ulcers, respiratory symptoms, and inflammation. Leaves have been applied on skin lesions and wounds. Saponins, flavonoids, tannins, and triterpenoids have been isolated from various parts. Antioxidant and antimicrobial activity has been reported through laboratory studies. Gastric mucosal protection has been confirmed through animal models.

Achyranthes aspera belongs to the *Amaranthaceae* family. It has been found near roadsides, fields, and open lands. It has been used for cough, asthma, wounds, kidney stones, digestive problems, skin infections, and joint pain. Roots, leaves, seeds, and inflorescences have been employed in different regions. Ecdysterone, alkaloids, steroids, saponins, and phenolic compounds have been reported. Diuretic, antimicrobial, anti-inflammatory, and wound healing effects have been documented. Many formulations have included this plant for respiratory and renal support.

The dependence on medicinal plants has been increased due to resistance to synthetic antibiotics. Pathogens have developed tolerance to many standard drugs. Plant extracts have been evaluated for integrated antimicrobial strategies. *Albizia amara* extracts have been shown to inhibit *Staphylococcus aureus*, *Escherichia coli*, and several fungal species. *Achyranthes aspera* extracts have been reported to act against bacteria and fungi relevant to skin and wound infections. These findings have been linked with traditional use in rural health systems.

Many chronic diseases have been associated with inflammatory processes. Edema models, paw inflammation models, and gastric inflammation models have been used to evaluate anti-inflammatory effects. *Albizia amara* bark extract has been shown to reduce inflammation in several models. *Achyranthes aspera* extracts have been shown to inhibit inflammatory pathways. Tissue swelling has been reduced. Pain markers have been lowered. Such results have reinforced the traditional use of both species for arthritis, wounds, and respiratory problems.

Oxidative stress has been implicated in various diseases. Plant antioxidants have been examined for protective effects. *Albizia amara* extract has been reported to reduce free radicals through multiple assays. *Achyranthes aspera* has been shown to provide similar antioxidant protection. These effects have supported their use in wound healing, gastric protection, liver support, and general health improvement.

Ethnobotanical importance of both plants has been established in many surveys. Healers have recognized them easily through morphological features. Fresh leaves, bark pieces, and roots have been collected in specific seasons. Their preparation techniques have been preserved. Community-based healing practices have used them for acute and chronic disorders. Many local communities have depended on these plants when clinical facilities are unavailable. Their availability has increased their use.

Pharmacognostic characterization has been performed to ensure correct identification. *Albizia amara* bark has been described through microscopic features such as fibers, sclerenchyma, and saponin cells. *Achyranthes aspera* leaves and roots have been described through trichomes, epidermal patterns, calcium oxalate crystals, and anatomical structures. These features have been used by researchers, manufacturers, and regulatory bodies to verify plant material.

Safety studies have been performed on both species. Acute and subacute toxicity tests have been conducted. *Albizia amara* extracts have shown wide safety margins at traditional doses. *Achyranthes aspera* extracts have shown minimal toxicity during laboratory evaluation. Mild irritation or toxicity has been observed at higher doses. Safety standards have been established for herbal formulations.

The link between traditional knowledge and modern science has been strengthened. Claims regarding treatment of ulcers, wounds, respiratory issues, digestive problems, and infections have been supported by experimental results. Anti-ulcer activity of *Albizia amara* bark extract has been proven in ethanol and stress-induced ulcer models. *Achyranthes aspera* has shown effects in reducing inflammatory edema. These results have validated cultural practices. Wound healing activity has been documented for both species. *Albizia amara* leaves have been applied on wounds. Faster epithelialization has been reported. *Achyranthes aspera* extracts have been shown to increase collagen content. Such activity has matched traditional first-aid practices in rural areas. Increased tensile strength, reduced healing time, and improved granulation tissue formation have been reported.

Hepatoprotective activity has been noted for *Albizia amara*. Reduction of liver enzyme levels has been reported during toxin-induced liver damage. Protection of hepatocytes from oxidative injury has been observed. *Achyranthes aspera* has shown renal protective activity. Increase in urine output has been noted. Reduction in kidney stone formation has been reported. These activities have made them suitable for nutraceutical use.

Anti-diabetic activity has been observed. *Albizia amara* extracts have reduced blood glucose levels in experimental animals. Glucose metabolism has been influenced. Insulin sensitivity has been improved. *Achyranthes aspera* extracts have also shown hypoglycemic effects. These findings have gained importance due to rising diabetes prevalence. Herbal supplements have been used alongside clinical treatment.

Gastrointestinal activity has been reported. *Albizia amara* extracts have shown anti-diarrheal effects. Gut motility and secretion have been reduced. *Achyranthes aspera* extracts have shown anti-emetic effects. Digestive support has been noted. These effects have aligned with the traditional use of both plants in digestive complaints.

Respiratory activity has been examined. *Albizia amara* and *Achyranthes aspera* have been used traditionally for cough and asthma. Laboratory models have confirmed bronchodilatory and anti-inflammatory effects. Mucus secretion has been regulated. Airway inflammation has been reduced. These properties have made them important for respiratory formulations.

Veterinary uses have been observed. *Albizia amara* bark decoctions have been used in livestock for digestive issues. *Achyranthes aspera* has been applied for wounds and infections in animals. These uses have expanded the value of these plants.

Cultivation and conservation needs have been highlighted. Habitat loss and overharvesting have affected many medicinal plants. Sustainable harvesting methods have been encouraged. *Albizia amara* grows well in degraded lands. It has been used in reforestation programs. *Achyranthes aspera* grows easily without fertilizers. It has been integrated into low-input cultivation systems. Their adaptability has reduced pressure on wild populations.

Quality control has been emphasized. Plant age, season of collection, drying method, storage, and extraction method have been standardized. *Albizia amara* bark collected during the dry season has shown higher saponin content. *Achyranthes aspera* leaves harvested during early growth have contained higher phenolics. Controlled drying and storage have preserved active compounds.

Polyherbal formulations have been developed using these plants. *Albizia amara* has been combined with anti-ulcer and anti-inflammatory herbs. *Achyranthes aspera* has been included in respiratory and diuretic formulations. Synergistic effects have been observed. Multi-target activity has been improved. Such combinations have been based on traditional practices and verified through research.

Economic importance has been recognized. Herbal markets have used these plants for extracts, powders, capsules, and dried raw material. Employment opportunities have been created. Farmers have cultivated them for commercial supply. Industries have used them for herbal supplements. Their wide availability and diverse activity have increased demand.

Phytochemical analysis has identified key compounds. *Albizia amara* has been shown to contain betulinic acid, oleanolic acid, tannins, flavonoids, and saponins. *Achyranthes aspera* has been shown to contain ecdysterone, achyranthine, steroids, alkaloids, phenolic acids, and saponins. These compounds have been linked with pharmacological effects. Their isolation has allowed further development of therapeutic agents.

Modern analytical tools have been used to characterize extracts. HPLC, GC-MS, FTIR, and UV spectrophotometry have been applied for identification and quantification. Such methods have been used to standardize formulations. Consistent quality has been ensured for clinical applications.

Research has been expanded to understand molecular mechanisms. Anti-inflammatory pathways have been studied through cytokine inhibition. Antioxidant effects have been measured through free radical scavenging. Antimicrobial activity has been studied through zone inhibition and MIC tests. Wound healing has been analyzed through collagen content and epithelialization rate. These studies have validated traditional uses with measurable parameters.

Albizia amara and *Achyranthes aspera* have gained attention due to broad therapeutic actions. Their presence in traditional medicine, ease of availability, and phytochemical richness have made them important. Their potential for future drug development has been recognized. They have been widely used in rural communities and studied in research laboratories.

Botanical Description of *Albizia amara* and *Achyranthes aspera*

The botanical identity of medicinal plants has been considered essential for correct use, quality control, and pharmacognostic evaluation. *Albizia amara* and *Achyranthes aspera* have been classified under two different families. Their morphology, microscopic features, and vernacular names have been recorded across regions for validation and reference. Their structures have been studied for proper authentication and to prevent substitution or adulteration in raw drug supply. Accurate knowledge of plant characters has been maintained in traditional systems. These details have been recorded in many botanical surveys, herbaria, and pharmacognostic manuals. Their diagnostic features have been documented to guide researchers, students, cultivators, and manufacturers. The following account presents a wide and detailed botanical description in passive voice.

Taxonomical Classification of *Albizia amara* and *Achyranthes aspera*

Albizia amara has been placed under the Fabaceae family. This species has been positioned within the Mimosoideae subfamily. Its classification has been standardized for scientific documentation. The following hierarchy has been accepted in botanical references. The kingdom has been listed as Plantae. The division has been placed under Tracheophyta. The class has been listed as Magnoliopsida. The order has been grouped under Fabales. The family has been assigned as Fabaceae. The genus has been designated *Albizia*. The species has been recorded as *Albizia amara*. This classification has been used in pharmacognostic texts and floras describing dry tropical vegetation. Several synonyms have been mentioned in earlier literature, but *Albizia amara* has been recognized as the accepted name in current taxonomic databases. Variations in regional floras have been reported because the species grows across large dry zones.

Achyranthes aspera has been placed under the Amaranthaceae family. The plant has been classified under the Caryophyllales order. Its taxonomic structure has been recognized consistently across floras. The kingdom has been listed as Plantae. The division has been assigned as Tracheophyta. The class has been included in Magnoliopsida. The order has been placed under Caryophyllales. The family has been listed as Amaranthaceae. The genus has been designated as *Achyranthes*. The species name has been accepted as *Achyranthes aspera*. This structure has been used in many scientific articles and identification guides. The species has been known for wide geographical distribution, and several varieties have been described in tropical and subtropical regions. Its classification has remained stable through revisions.

Vernacular Names of *Albizia amara* and *Achyranthes aspera*

Many vernacular names have been recorded for *Albizia amara*. These names have been derived from regional languages. They have been used by local communities, healers, and traditional practitioners. The plant has been known as Arappu in Tamil. It has been called Chilla or Chilla-pala in some northern regions. In Kannada, the tree has been referred to as Aralu. In Telugu, the plant has been known as Chiruthaku. In Malayalam, the plant has been described as Arappa. In Sinhala, the plant has been called Halamba. In English references, it has been sometimes labeled as Bitter *Albizia*. These names have been used for trade as well as documentation. Their variations have been influenced by local usage and dialects. Knowledge of vernacular names has been considered essential for field identification.

Achyranthes aspera has been known by many names across regions. In Hindi, the plant has been called Apamarg. In Sanskrit, it has been referred to as Apamarga. In Tamil, it has been called Nayuruvi. In Telugu, the plant has been known as Uttareni. In Kannada, it has been referred to as Uttarani. In Malayalam, the plant has been called Kadaladi. In Bengali, it has been referred to as Apang. In English sources, it has been sometimes called Prickly Chaff Flower. Vernacular names have been used for rural prescriptions, ethnomedicinal records, and herb market identification. The accuracy of these names has been important to prevent confusion with related species.

Morphological Features of *Albizia amara* and *Achyranthes aspera*

Albizia amara has been described as a deciduous tree. It has been observed in dry deciduous forests, scrublands, and semi-arid regions. The height has been recorded between 6 and 12 meters. The trunk has been moderately straight. The bark has been grey or brown. The bark surface has been rough with vertical cracks. Exudation has not been prominent. The branches have been seen to spread widely, giving a divided crown. The leaves have been bipinnate. Leaflets have been arranged in pairs. Small secondary leaflets have been present. Leaflets have been oblong, smooth, and pale green. The surface has been glabrous. The petiole has been slender. The leaves have closed during night hours, and this feature has been reported in many surveys. The inflorescence has been arranged in heads or clusters. The flowers have been small, whitish, and fragrant. Numerous stamens have been present. The calyx has been short. The corolla has been tubular. The stamens have been long and free. The flowers have been characteristic of the Mimosoideae group. The pods have been flat, narrow, and elongated. The colour has been brown when mature. Seeds have been arranged in rows. The seeds have been small, hard, and brownish. The flowering season has been observed during dry months. Fruiting has been reported soon after flowering. The root system has been deep. Taproots have been observed with numerous lateral branches. Root bark has been tough. The wood has been pale and moderately hard. The tree has been resistant to drought. The leaves and bark have been used in traditional medicine. The plant has been recognized easily in dry landscapes due to its bipinnate leaves and distinctive pods. *Achyranthes aspera* has been described as an erect or semi-erect herb. The plant has reached a height of 30 to 100 centimeters. The stem has been simple or branched. The surface has been rough because of fine hairs. The stem has been angular. The colour has

been green. The leaves have been simple, opposite, and petiolate. The leaf margin has been entire. The venation has been prominent. The leaf shape has been ovate or elliptic. The surface has been slightly hairy. The size has varied with environmental conditions. The inflorescence has been arranged in long spikes. The spikes have been erect. Numerous small flowers have been attached closely. Persistent bracts have given the spike a rough touch. The flowers have been greenish. The perianth has been small. The utricle fruit has been one-seeded. The seed has been ovoid and reddish brown. The flowering period has been observed across many months, depending on climate. The root has been cylindrical. It has been long and tough. It has extended deep into the soil. The root surface has been pale. The cross-section has shown central xylem and peripheral phloem. The plant has been distributed widely across plains, hillsides, roadsides, and uncultivated fields. It has been adapted to disturbed habitats. Its hardy nature has allowed rapid growth during rainy seasons. The entire plant has been harvested for medicinal use.

Microscopic Characteristics of *Albizia amara* and *Achyranthes aspera*

Microscopic features of *Albizia amara* have been documented for bark, leaves, and wood. The bark has shown stratified layers. The outer bark has shown cork cells arranged in radial rows. The cork has been multilayered. Secondary cortex has been present. Parenchyma cells have been observed with tannin content. Sclerenchyma fibers have been distributed irregularly. Stone cells have been present in some sections. The phloem has shown sieve tubes and companion cells. Phloem parenchyma has been abundant. Starch grains have been noted in many cells. Calcium oxalate crystals have been detected occasionally. In leaf microscopy, the epidermis has been single-layered. The upper epidermis has been covered with a smooth cuticle. The lower epidermis has contained stomata. The stomatal type has been paracytic. Palisade cells have been arranged in one layer. Spongy tissue has been loosely arranged. Vascular bundles have been present along the midrib. Bundle sheath cells have been observed. Calcium oxalate crystals have been found in some tissues. In wood sections, vessels have been present in diffuse and radial arrangement. Fibers have been thick-walled. Parenchyma cells have been present in axial rows. Medullary rays have been narrow. The wood structure has been consistent with the Fabaceae family. Microscopic features have supported taxonomic identification and pharmacognostic analysis.

Microscopic features of *Achyranthes aspera* have been reported for root, stem, and leaf. The root has shown a multilayered cork. Cork cells have been tangentially elongated. Secondary

cortex has been broad. Parenchyma cells have contained starch. Calcium oxalate crystals have been abundant. Phloem has shown sieve tubes, companion cells, and phloem parenchyma. Xylem has been arranged in radial pattern. Vessels have been large. Wood fibers have been thick-walled. Medullary rays have been uniseriate. The overall structure has been characteristic of dicot roots.

The stem has shown an epidermis with unicellular hairs. Collenchyma has been present beneath the epidermis. Cortex has been composed of parenchyma. Vascular bundles have been arranged in a ring. Xylem vessels have been lignified. Phloem has been present external to xylem. Pith has been large and parenchymatous. Crystals have been scattered throughout tissues. Leaf microscopy has shown a thin cuticle. The epidermis has been single-layered. Trichomes have been present on both surfaces. Stomata have been found on lower surfaces. Mesophyll has been differentiated into palisade and spongy tissue. Vascular bundles have been arranged along the midrib and veins. Collenchyma has been present near midrib. Calcium oxalate crystals have been visible across the mesophyll and epidermal layers. These microscopic features have been used to confirm the identity of *Achyranthes aspera* in crude drug samples. Diagnostic characters have been considered stable and useful for quality control. Further structural details have been documented during field studies. Young twigs have been slender. Leaflets have been arranged in two to four pairs. Each pinna has carried several pairs of small leaflets. Leaflets have shown a smooth margin. The apex has been rounded. The base has been asymmetrical. Glands have been present on the petiole, as reported in some descriptions. The rachis has shown slight hairiness in young stages. The inflorescence has resembled small pom-poms. Colour has been pale cream. The pods have been thin. Each pod has contained several seeds. The seeds have been disc-shaped. The kernel has been hard. Pod dehiscence has been observed during dry periods. Detailed observations have reported rough stems because of persistent trichomes. The nodes have been enlarged. The internodes have been short. The leaves have shown variation depending on moisture availability. Larger leaves have been observed during rainy seasons. Smaller leaves have been seen in dry periods. The inflorescence spike has been long, cylindrical, and stiff. The bracts have been spiny at maturity. These bracts have been responsible for the unique roughness of the spike. Flowers have been inconspicuous. The ovary has been unilocular. The stigma has been capitate. The utricle fruit has been small and enclosed by hardened perianth segments. Seeds have been smooth, polished, and located inside the utricle. Transverse sections of the bark

have shown distinct cork, cortex, and phloem. Cork formation has increased with age. Cortex cells have shown dark brown contents due to tannin. Secondary phloem has displayed stratification. Fiber patches have been interspersed with parenchyma. Some parenchyma cells have contained saponin content. These features have been important for confirming species identity during pharmacognostic evaluation. Leaf sections have shown dorsiventral structure. The midrib has been prominent. Xylem has been found internal. Phloem has been positioned externally. The spongy tissue has been more prominent near lower epidermis. The presence of abundant trichomes has been considered a key microscopic character. The trichomes have been unicellular or multicellular. Some have been hooked. Crystals have been frequent in root and stem tissues. They have been mostly calcium oxalate. Druses, prismatic forms, and small clusters have been recorded. Parenchymatous tissues have been dominant in cortex and pith. In leaf sections, the palisade tissue has been single layered. Spongy tissue has been loosely arranged. Vascular bundles have been collateral. The midrib has shown a broad vascular bundle. Supporting tissues have included collenchyma near the epidermis. Such features have strengthened botanical authentication.

Phytochemical Constituents

A comprehensive evaluation of the phytochemical makeup of *Albizia amara* and *Achyranthes aspera* has been carried out through extensive experimental, chromatographic, and spectroscopic studies, and the chemical diversity of both species has been described in a wide range of phytochemical investigations. The chemical profiles of these medicinal plants have been viewed as central to their biological activity, and the identification of their major groups of secondary metabolites has been performed by various laboratories using standardized and modified methodologies. The following text presents a detailed account of the major phytochemicals reported from *Albizia amara* and *Achyranthes aspera*, supported by a description of phytochemical screening procedures and a comparative analytical discussion of their active constituents. All statements are delivered in passive voice as requested.

Major Chemical Compounds in *Albizia amara* and *Achyranthes aspera*

The phytochemical profile of *Albizia amara* has been documented across several decades, and extensive chemical characterization has been performed using thin-layer chromatography, high-performance liquid chromatography, gas chromatography-mass spectrometry, UV-visible spectrometry, liquid chromatography-mass spectrometry, and nuclear magnetic resonance

spectroscopy. A rich variety of compounds have been reported, and the plant is recognized for producing a wide group of bioactive constituents. Significant attention has been given to flavonoids, tannins, triterpenoids, glycosides, saponins, alkaloids, and phenolic derivatives, and these have been isolated from the leaves, bark, stem, pods, seeds, and root tissues. The most frequently documented flavonoids from *Albizia amara* include quercetin, rutin, catechin, kaempferol, apigenin, and their various glycosylated derivatives, and the presence of flavonol-type compounds has been confirmed through several spectroscopic studies. Saponins have been identified as one of the dominant classes, and they have been isolated in both crude and purified forms; their biological relevance has been emphasized due to their surfactant properties and immunomodulatory effects. A series of triterpenoid saponins, including amarin, amarosaponin, and oleanane-type constituents, have been widely described, and their structures have been elucidated using NMR and MS fragmentation analysis. Tannins are also abundant, and a range of condensed and hydrolysable tannins has been reported, with particular emphasis on leucoanthocyanidins and proanthocyanidins. Phenolic acids, including gallic, ferulic, caffeic, syringic, and p-coumaric acids, have been quantified in various plant fractions, and these compounds have been related to the plant's antioxidant properties. Several alkaloids have been mentioned in literature, although their concentrations have been documented as relatively lower compared with flavonoids and saponins. The bark extract has been described as rich in polyphenols, while the leaf extract has been noted for containing a higher proportion of flavonoids. Seeds have been shown to carry lipid-soluble compounds, sterols, fatty acids, and minor quantities of fixed oils. Phytosterols such as β -sitosterol, stigmasterol, and campesterol have been reported. The gums and mucilage fractions of the plant have been characterized for polysaccharides, and monosaccharide composition analysis has identified rhamnose, arabinose, galactose, and glucose. A number of glycosides including triterpenoid glycosides, flavonoid glycosides, and phenolic glycosides have also been detailed. The overall chemical complexity of *Albizia amara* has been interpreted as evidence of its long-standing medicinal use in traditional systems.

The chemical constituents of *Achyranthes aspera* have been examined in equal breadth, and extensive phytochemical exploration has revealed that the plant produces a wide spectrum of alkaloids, saponins, sterols, flavonoids, phenolics, lignans, diterpenoids, triterpenoids, ecdysteroids, and polysaccharides. Analytical reports have described the isolation of several

unique compounds that are rarely found in other medicinal plants. Among the most studied constituents, the alkaloid group has been identified as one of the key classes, and a variety of alkaloidal compounds have been isolated, including achyranthine, betaine, and 4-hydroxy-2-methylquinoline. Saponins have been reported in high quantity, and several oleanolic acid-based saponins have been described. Ecdysteroids such as ecdysterone (20-hydroxyecdysone) and polypodine B have been identified in root and leaf tissues, and these compounds have been studied for their anabolic and adaptogenic effects. Flavonoids including quercetin, rutin, and apigenin have been reported, and phenolic acids such as gallic, vanillic, ferulic, and caffeic acids have been detected. Diterpenoids including clerodane-type and labdane-type structures have been described. A variety of triterpenoids such as oleanolic acid, ursolic acid, and amyrins have been isolated and structurally characterized. Lignans such as medioresinol and syringaresinol have been recorded in some fractions. Sterols including β -sitosterol, stigmasterol, and brassicasterol have been found in leaf and seed tissues. The fixed oil content of the seeds has been analyzed for fatty acids, and linoleic, oleic, palmitic, and stearic acids have been documented as the major components. Polysaccharides extracted from aerial and root portions have been described as complex heteropolymers containing arabinose, xylose, mannose, galactose, and glucose. Aqueous, ethanolic, methanolic, chloroform, petroleum ether, and hydroalcoholic extracts have been subjected to qualitative and quantitative phytochemical analysis, and extensive variation in chemical profile has been observed depending on solvent polarity. The presence of betaine has been repeatedly confirmed through chemical tests, infrared spectroscopy, and chromatographic analysis. Ashoka-type glycosides, anthraquinones, and minor alkaloids have been mentioned in a small number of studies. Overall, *Achyranthes aspera* has been reported as chemically diverse and rich in secondary metabolites with significant therapeutic value.

Methods of Phytochemical Screening

Phytochemical screening of *Albizia amara* and *Achyranthes aspera* has been performed using a set of standardized qualitative and quantitative protocols adopted from pharmacognostic and phytochemical manuals. The general workflow involves extraction, preliminary screening, chromatographic separation, spectroscopic identification, and quantification. Hot and cold extraction techniques have been employed, and solvents including water, ethanol, methanol, acetone, ethyl acetate, petroleum ether, chloroform, and hydroalcoholic mixtures have been used

to obtain selective fractions. Soxhlet extraction has been widely adopted for exhaustive extraction, while maceration and infusion have been used for temperature-sensitive constituents. Ultrasound-assisted extraction and microwave-assisted extraction have also been employed in modern studies.

Qualitative phytochemical tests have been performed using classical methods. Alkaloids have been screened using Dragendorff, Wagner, Mayer, and Hager reagents. Flavonoids have been detected through alkaline reagent test, Shinoda test, and lead acetate test. Saponins have been tested using froth formation method and emulsification index method. Tannins have been identified using ferric chloride, lead acetate, and gelatin tests. Sterols have been characterized using Salkowski and Liebermann–Burchard reactions. Terpenoids have been identified through Salkowski test. Phenolic compounds have been screened using ferric chloride coloration. Carbohydrates have been detected through Molisch and Benedict tests, and proteins have been identified using Biuret and Millon tests.

Chromatographic methods have been extensively used in phytochemical profiling. Thin-layer chromatography has been performed for preliminary fraction separation, and specific spray reagents have been used to visualize flavonoids, alkaloids, and saponins. High-performance liquid chromatography and high-performance thin-layer chromatography have been used for quantification of polyphenols, flavonoids, and saponins. Gas chromatography-mass spectrometry has been applied for volatile constituents and fatty acids, while liquid chromatography-mass spectrometry has been employed for non-volatile secondary metabolites. Nuclear magnetic resonance spectroscopy has been used for full structural elucidation. Fourier transform infrared spectroscopy has been used for functional group identification.

Quantitative phytochemical analyses have included total phenolic content using Folin–Ciocalteu assay, total flavonoid content using aluminum chloride method, tannin content estimation using Folin–Denis method, saponin content estimation through gravimetric or spectrophotometric assays, and alkaloid content determination using acid-base titration or gravimetric precipitation. These methods have produced reproducible results that have been widely utilized to characterize the bioactive potential of both plants.

Comparative Analysis of Active Constituents

Comparative phytochemical assessment of *Albizia amara* and *Achyranthes aspera* has been performed by multiple research groups, and interesting similarities and differences have been

observed. Both species have been shown to contain rich profiles of flavonoids, phenolics, saponins, sterols, and triterpenoids, yet the specific identity and structural types of these compounds differ.

Flavonoids such as quercetin and rutin have been reported in both plants, but apigenin is more prominently documented in *Albizia amara*, while ecdysteroids have been found exclusively in *Achyranthes aspera*. Saponins occur in high concentration in both species, yet their aglycone structures differ substantially; *Albizia amara* produces mainly oleanane-type triterpenoid saponins, while *Achyranthes aspera* has yielded both oleanolic acid-based and ecdysteroid-related saponins.

Phenolic acids are present in both plants, but *Albizia amara* has shown higher levels of gallic and syringic acid, while *Achyranthes aspera* has shown higher concentrations of caffeic and vanillic acids. The alkaloid fraction is more pronounced in *Achyranthes aspera*, supported by the presence of achyranthine and betaine. In contrast, *Albizia amara* contains only minor alkaloid traces. Sterol composition shows some overlap, as both species produce β -sitosterol and stigmasterol. Triterpenoids show partial similarity, with oleanolic acid and amyryns present in both, yet *Achyranthes aspera* features a larger number of triterpenoid derivatives.

Polysaccharides from both species have been shown to contain similar monosaccharide units, although their proportions differ. Lignans are more characteristic of *Achyranthes aspera* and have rarely been reported in *Albizia amara*. The presence of ecdysteroids in *Achyranthes aspera* represents a major differentiating feature because such compounds are absent in *Albizia amara* and are known for unique biochemical properties.

Pharmacognostic Evaluation

Macroscopic and microscopic evaluation of *Albizia amara* and *Achyranthes aspera* has been performed extensively in pharmacognostic studies, and both plants have been subjected to standard identification procedures recommended in pharmacopoeial guidelines. Macroscopic identification has been carried out by examining fresh and dried plant materials, and diagnostic features have been recorded for leaves, stems, roots, bark, flowers, and seeds. The leaves of *Albizia amara* have been described as bipinnate, with numerous small leaflets arranged alternately, and the lamina has been noted for its smooth texture and entire margins. The size of the leaflets has been reported as small and oblong, and a pale green coloration has been observed in fresh specimens. The bark has been described as rough, fissured, and dark brown externally,

while the internal bark region has displayed yellowish brown tones. The pods have been documented as thin, flat, and elongated with multiple seeds embedded in the inner tissue. The macroscopic features of *Achyranthes aspera* have been described as equally distinctive. The plant has been recognized for its erect habit, opposite leaves, and stiff stems covered with fine hairs. The leaves have been recorded as ovate, coarse, and rough with a greenish grey appearance. The inflorescence has been described as a long spike lined with persistent bracts and hooked seeds that adhere to cloth or skin. Stems have been found to display quadrangular outlines in young portions that become cylindrical with age. Seeds have been described as lenticular and brownish, embedded within bracteates that contribute to the characteristic prickly nature of the plant.

Microscopic identification has been performed using transverse sections and powdered materials of both species. In *Albizia amara*, the transverse section of the leaf has revealed a dorsiventral arrangement with upper and lower epidermis, each bearing a single layer of cells. The mesophyll has been differentiated into palisade and spongy parenchyma. Collateral vascular bundles have been observed with xylem vessels, phloem tissue, and surrounding parenchymatous sheath. Calcium oxalate crystals have been recorded in the form of prisms and druses. The stem section has displayed a typical dicot configuration with epidermis, cortex, phloem, xylem, and large central pith. Fiber bundles have been identified within the cortex, and lignified xylem vessels have been observed with scalariform or pitted thickenings. Bark sections have shown stratified cork, cortical parenchyma, abundant tannin cells, and secondary phloem elements. Microscopic features of *Achyranthes aspera* have included thick-walled epidermal cells with warty cuticle, abundant cystolithic outgrowths in the epidermis, and prominent parenchyma in the cortex. Vascular bundles have been found to be collateral and arranged in a circular pattern. Calcium carbonate cystoliths have been observed in leaf epidermis, which has been considered a diagnostic feature. Root sections have shown secondary growth with well-developed xylem and phloem. Vessel elements with simple perforations, medullary rays, and lignified fibers have been consistently documented.

Powder microscopy has been conducted on the dried powdered leaf, stem, bark, and root portions. In *Albizia amara*, diagnostic characters such as trichomes, fragments of epidermis with stomata, fibers, xylem vessels with spiral or reticulate thickening, tannin-containing cells, prismatic calcium oxalate crystals, and mesophyll fragments have been reported. Powder of bark

has shown cork cells, parenchyma with brownish content, stone cells, and phloem fibers. Powder of *Achyranthes aspera* has been described as rich in cystolith fragments, epidermal cells with warty cuticle, parenchyma cells with crystalline inclusions, trichomes, vessels with annular thickenings, and abundant sclerenchymatous fibers. The presence of characteristic hooked bract fragments has been noted in the powdered inflorescence. Powder microscopy has been emphasized for its importance in identifying adulteration, as both plants are used in traditional medicine and commercial herbal markets.

Physicochemical parameters have been evaluated in accordance with WHO and Indian Pharmacopoeia guidelines. Parameters such as moisture content, total ash, acid-insoluble ash, sulphated ash, water-soluble ash, alcohol-soluble extractive value, water-soluble extractive value, loss on drying, swelling index, foaming index, and pH of aqueous solutions have been determined. *Albizia amara* bark has been shown to exhibit high total ash values due to its tannin and mineral content. The acid-insoluble ash has been observed as moderate, indicating minimal contamination with siliceous matter. Extractive values have varied depending on the solvent, with methanolic and hydroalcoholic extracts showing higher yields. For *Achyranthes aspera*, total ash values have been reported as relatively lower, whereas water-soluble ash values have been documented as higher due to the presence of water-soluble components. The foaming index has been found to be high in both species due to the abundance of saponins, and swelling index values have been found to reflect the polysaccharide content in root and stem tissues. These physicochemical parameters have been used as quality standards for raw drug authentication.

Fluorescence analysis has been performed under visible, ultraviolet short-wave, and ultraviolet long-wave light using powdered drug samples treated with various reagents. Distinctive fluorescence patterns of *Albizia amara* powder have been observed when treated with sodium hydroxide, nitric acid, iodine, and ammonia solutions. Green, brown, yellow, and fluorescent green hues have been produced under different wavelengths. *Achyranthes aspera* powder has shown characteristic bright yellow and green fluorescence when treated with alkaline reagents, and reddish brown fluorescence has been noted under acidic treatments. These fluorescence reactions have been standardized for pharmacognostic identification.

Extractive values and ash content have been quantified for both plants, and standardized ranges have been established. Extractive values have been used to estimate the amount of active constituents present in different solvent fractions. Methanol and ethanol extracts have shown the

highest extractive yields, followed by water extracts. Petroleum ether and chloroform extracts have shown low yields, indicating fewer non-polar constituents. Total ash content has reflected total mineral matter, while acid-insoluble ash has indicated earthy impurities. Sulphated ash values have provided insight into inorganic sulfate content. These parameters have been regarded as important metrics for quality control and standardization.

Phytochemical Isolation and Characterization

Extraction methods, chromatographic techniques, spectral identification, and structural elucidation have been applied extensively to isolate and characterize bioactive compounds from *Albizia amara* and *Achyranthes aspera*. Extraction has been performed using maceration, percolation, Soxhlet extraction, hot continuous extraction, cold extraction, ultrasound-assisted extraction, microwave-assisted extraction, and supercritical fluid extraction. Solvents ranging from non-polar to polar have been used to fractionate different classes of compounds. Petroleum ether, hexane, chloroform, ethyl acetate, acetone, methanol, ethanol, and water have been used either singly or sequentially. Crude extracts have been concentrated under reduced pressure using rotary evaporation, and fractions have been dried and stored for further analysis.

Chromatographic techniques have played a central role in phytochemical separation. Thin-layer chromatography has been used for preliminary screening of flavonoids, tannins, saponins, and phenolics. High-performance thin-layer chromatography has been utilized for quantification and fingerprinting. Column chromatography using silica gel, alumina, Sephadex LH-20, ion-exchange resins, and reversed-phase sorbents has been extensively applied to isolate pure compounds. Gradient elution using binary and ternary solvent systems has been utilized. Preparative HPLC has been adopted for high-purity isolation of flavonoids and saponins. Gas chromatography-mass spectrometry has been used for volatile constituents and fatty acids. Liquid chromatography-mass spectrometry has been used for profiling non-volatile phytochemicals and for dereplication of known structures. Flash chromatography, size-exclusion chromatography, and vacuum liquid chromatography have been used to speed up separation. Repeated chromatographic purification has been conducted to achieve high purity of compounds destined for spectroscopic identification.

Spectroscopic identification has been performed using UV-visible spectroscopy, infrared spectroscopy, nuclear magnetic resonance spectroscopy, and mass spectrometry. UV-visible spectroscopy has been used to identify flavonoid absorption maxima and phenolic band patterns.

Infrared spectroscopy has been used to identify functional groups such as hydroxyl, carbonyl, aromatic, glycosidic, and aliphatic moieties. Nuclear magnetic resonance spectroscopy, including ^1H -NMR, ^{13}C -NMR, DEPT, COSY, HSQC, and HMBC, has been employed for full characterization of carbon and hydrogen skeletons. Mass spectrometry has been used for determining molecular weights and fragmentation pathways. Electrospray ionization, atmospheric pressure chemical ionization, and electron impact ionization have been used depending on compound class. High-resolution MS has been used to determine accurate mass values.

Structural elucidation has been carried out through interpretation of spectroscopic data. Flavonoids such as quercetin, kaempferol, apigenin, and their glycosides have been elucidated using UV, NMR, and MS patterns. Saponins have been characterized by identifying sugar moieties through acid hydrolysis followed by chromatographic analysis of monosaccharides. Triterpenoids such as oleanolic acid, amyrins, and ursolic acid have been confirmed through distinctive NMR signals and MS peaks. Alkaloids from *Achyranthes aspera* have been structurally identified through proton and carbon shifts indicative of nitrogen-containing heterocycles. Ecdysteroids such as 20-hydroxyecdysone have been confirmed through characteristic mass fragmentation and NMR data. Phenolic acids have been elucidated using combined HPLC retention times, UV spectra, and MS fragmentation.

Pharmacological Activities of *Albizia amara*

Extensive pharmacological investigation has been conducted on *Albizia amara*, and numerous biological activities have been documented through in vitro, in vivo, and ex vivo assays. A broad spectrum of pharmacological relevance has been attributed to its diverse phytochemical composition, particularly triterpenoid saponins, flavonoids, tannins, alkaloids, phenolic acids, and glycosides. The following section provides a detailed, expanded, paragraph-based exposition of antioxidant, anti-inflammatory, antimicrobial, antidiabetic, anticancer, hepatoprotective, nephroprotective, and other reported pharmacological actions of *Albizia amara*. The discussion has been written in passive voice, and the depth has been extended to approximate 5000-word scientific detail for the combined *Albizia amara* and *Achyranthes aspera* output.

Antioxidant Activity of *Albizia amara*

Prominent antioxidant effects have been reported for *Albizia amara*, and significant free radical neutralization has been demonstrated through several in vitro assays. Extracts have been

evaluated through DPPH, ABTS, FRAP, nitric oxide scavenging, superoxide anion scavenging, and lipid peroxidation inhibition methods. High phenolic and flavonoid content has been attributed to the potent scavenging potential. It has been observed that methanolic and ethanolic extracts exhibit stronger antioxidant potential compared to aqueous extracts, suggesting that polar-organic solvents facilitate the extraction of flavonoids, tannins, and saponins responsible for electron-donating abilities. Free radical inhibition in cell-based assays has also been documented, and the protective effect against oxidative membrane damage has been linked to reduced intracellular ROS production. Antioxidant activity has been associated with quercetin derivatives, catechin, ellagic acid, gallic acid, and saponin complexes that stabilize reactive oxygen species and terminate chain reactions involved in lipid peroxidation. Enhanced mitochondrial protection has been observed in oxidative stress-induced models, and DNA protection assays have shown reduced oxidative fragmentation, indicating major significance in preventing degenerative cellular processes.

Anti-inflammatory and Analgesic Effects of *Albizia amara*

Strong anti-inflammatory activity has been attributed to *Albizia amara*, and extracts have been evaluated using carrageenan-induced paw edema, cotton pellet granuloma, albumin denaturation, RBC membrane stabilization, and nitric oxide inhibition assays. Acute and chronic inflammation models have demonstrated a measurable reduction in inflammatory mediators. Inhibition of prostaglandins, leukotrienes, and pro-inflammatory cytokines has been reported, and significant membrane stabilization has been attributed to the presence of saponins and triterpenoids. Analgesic effects have been confirmed through tail-flick and acetic acid-induced writhing tests. Inhibition of cyclooxygenase and lipoxygenase pathways has been cited as a major mechanism, and reduced edema formation has been linked to improved vascular permeability regulation. In addition, protein denaturation inhibition suggests that *Albizia amara* possesses significant potential in conditions involving chronic inflammatory tissue damage. The analgesic effect has been considered moderate to strong, and central and peripheral nociceptive control has been suggested based on reduced response in thermal and chemical models.

Antimicrobial Activity of *Albizia amara*

Antibacterial and antifungal potentials of *Albizia amara* have been documented through multiple in vitro assays. Zone of inhibition, MIC, and MBC tests have revealed notable activity against gram-positive and gram-negative bacteria, including *Staphylococcus aureus*, *Streptococcus*

pyogenes, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, and *Klebsiella pneumoniae*. Strong antifungal effects have been documented against *Candida albicans*, *Aspergillus niger*, *Aspergillus flavus*, and *Trichophyton* species. Phytoconstituents such as saponins, tannins, flavonoids, and alkaloids have been associated with membrane disruption, protein precipitation, and enzyme inhibition. Bactericidal activity has been linked to increased membrane permeability and leakage of intracellular content. Synergistic effects with synthetic antibiotics have also been reported, indicating that *Albizia amara* extracts may enhance antibiotic potency in resistant strains. Selected extracts have shown biofilm inhibition, suggesting potential relevance in chronic infections involving biofilm-forming pathogens. Overall, antimicrobial activity has been characterized as moderate to strong depending on concentration, plant part, and solvent system.

Antidiabetic Potential of *Albizia amara*

Significant antidiabetic effects have been attributed to *Albizia amara*, and various extracts have been tested in alloxan-induced and streptozotocin-induced diabetic rat models. A marked reduction in fasting blood glucose has been observed, and improvement in glucose tolerance tests has been documented. Enhanced insulin secretion from pancreatic β -cells and reduced oxidative stress in pancreatic tissue have been reported. Inhibition of alpha-amylase and alpha-glucosidase enzymes has been demonstrated in vitro, suggesting that carbohydrate digestion and absorption are slowed, leading to improved postprandial glucose control. Regeneration of pancreatic islets has been observed in histological studies, and glycogen storage enhancement in hepatic tissue has been reported. Reduced lipid parameters, including triglycerides, LDL, and VLDL, suggests additional antihyperlipidemic potential in diabetic conditions. Improved antioxidant status in diabetic tissues suggests that *Albizia amara* reduces secondary complications of diabetes involving oxidative stress and mitochondrial dysfunction.

Anticancer and Cytotoxic Studies of *Albizia amara*

Cytotoxicity and anticancer properties of *Albizia amara* have been reported in several cell-based assays. Extracts have shown growth inhibition in cancer lines such as MCF-7, HeLa, A549, HT-29, and HepG2. Mechanisms have been linked to apoptosis induction, mitochondrial membrane destabilization, caspase activation, DNA fragmentation, and oxidative stress imbalance in malignant cells. Flow cytometry studies have confirmed increased apoptotic cell population. Phenolic and triterpenoid compounds have been suggested to play a central role in suppressing

cancer cell proliferation. Anti-angiogenic effects have been reported in CAM assays, and inhibition of metastasis-related enzymes such as MMP-2 and MMP-9 has been documented. *Albizia amara* extracts have shown selective toxicity, with lower cytotoxicity toward normal cells. In vivo tumor reduction in murine models has also been recorded, indicating potential for future therapeutic applications and chemopreventive relevance.

Hepatoprotective and Nephroprotective Effects of *Albizia amara*

The hepatoprotective potential of *Albizia amara* has been documented in models of CCl₄-induced, paracetamol-induced, and ethanol-induced liver toxicity. Extracts have shown normalization of liver enzymes including AST, ALT, ALP, and bilirubin. Restoration of hepatic architecture has been observed in histopathological sections, and reduced lipid peroxidation in hepatic tissue suggests strong antioxidant protection against toxic damage. Increased GSH, SOD, and CAT activities indicate enhanced endogenous antioxidant defense systems. Nephroprotective activity has been recorded in gentamicin-induced kidney toxicity models, with improvement in creatinine, urea, uric acid, and electrolytes. Histopathological sections have shown restoration of glomerular and tubular structure. Protection has been attributed to phenolic antioxidants, saponins improving membrane integrity, and tannins reducing oxidative and inflammatory damage.

Other Pharmacological Activities of *Albizia amara*

Additional activities include antiulcer, antidiarrheal, wound healing, immunomodulatory, anthelmintic, anticonvulsant, and antihypertensive properties. Antiulcer effects have been demonstrated in ethanol- and pylorus-ligated models, with reduced ulcer index and increased mucosal protection. Antidiarrheal activity has been observed through decreased intestinal transit. Immunomodulatory activity has been linked to improved macrophage phagocytosis and increased antibody titers. Wound healing activity has been reported through enhanced epithelialization and collagen deposition. Anthelmintic effects have been attributed to paralysis and death of helminths in vitro. CNS studies have shown mild anticonvulsant effects in PTZ and MES models. Antihypertensive effects have been reported through vasorelaxation and ACE inhibition. These findings indicate broad pharmacological relevance.

Pharmacological Activities of *Achyranthes aspera*

Antioxidant Activity of *Achyranthes aspera*

Potent antioxidant activity has been attributed to *Achyranthes aspera* based on strong free radical scavenging in DPPH, ABTS, FRAP, nitric oxide, superoxide, and hydroxyl radical assays. High phenolic content, particularly caffeic acid, ferulic acid, rutin, quercetin, saponins, and alkaloids, has been identified as responsible for radical quenching and metal chelation. Lipid peroxidation reduction has been demonstrated in rat liver homogenate assays. DNA and protein protection from oxidative damage has been reported. Improved mitochondrial stability and decreased intracellular ROS generation suggest relevance against degenerative diseases and inflammatory oxidative cascades.

Anti-inflammatory and Analgesic Effects of *Achyranthes aspera*

Strong anti-inflammatory activity has been demonstrated in carrageenan-induced paw edema, cotton pellet granuloma, xylene-induced ear edema, and lipoxygenase inhibition assays. Inhibition of COX and LOX pathways, reduced TNF- α , IL-1 β , IL-6, and suppressed nitric oxide production have been reported. Analgesic effects have been observed in tail-flick, hot plate, and acetic acid writhing models. Membrane stabilization and protein denaturation inhibition provide evidence of strong anti-inflammatory and analgesic relevance. The presence of saponins and alkaloids plays a significant role.

Antimicrobial Activity of *Achyranthes aspera*

Broad-spectrum antimicrobial activity has been documented. Inhibition of bacteria such as *S. aureus*, *E. coli*, *Salmonella*, *Shigella*, *Bacillus subtilis*, *P. aeruginosa*, and *K. pneumoniae* has been reported. Strong antifungal activity has been recorded against *Candida*, *Aspergillus*, *Trichophyton*, and *Penicillium* species. Extracts have shown membrane disruption, protein coagulation, and interference with microbial enzymatic pathways. Antiviral activity against selected DNA and RNA viruses has been suggested, though limited data exist. Biofilm inhibition and quorum-sensing suppression have been documented.

Antidiabetic Potential of *Achyranthes aspera*

Strong antidiabetic activity has been demonstrated. STZ and alloxan diabetic models have shown reduced blood glucose, improved glucose tolerance, enhanced hepatic glycogen storage, and pancreatic β -cell regeneration. α -Glucosidase and α -amylase inhibition has been documented in vitro. Lipid profiles have been improved, showing reduced triglycerides, LDL, and improved

HDL levels. Improved antioxidant markers in pancreatic tissue support its role in preventing β -cell oxidative damage.

Anticancer and Cytotoxic Studies of *Achyranthes aspera*

Cytotoxic activity has been reported against MCF-7, HeLa, DU-145, HepG2, A549, and colon cancer cell lines. Apoptotic mechanisms include increased caspase 3 and 9 activity, mitochondrial depolarization, cell cycle arrest at G2/M, and inhibition of angiogenesis. Saponins have shown membrane lytic effects in malignant cells. Animal models have shown tumor volume reduction and improved survival. Selective cancer cytotoxicity has been reported, with limited toxicity toward normal cells.

Hepatoprotective and Nephroprotective Effects of *Achyranthes aspera*

Hepatoprotective activity has been established in CCl₄, paracetamol, and ethanol toxicity models. Liver enzymes have shown normalization. Protection has been associated with reduced lipid peroxidation and increased antioxidant enzymes. Nephroprotective activity has been reported in gentamicin- and cisplatin-induced renal injury models. Improvement in creatinine, BUN, and electrolyte balance has been observed. Histopathology has shown restoration of glomerular and tubular architecture.

Other Reported Activities of *Achyranthes aspera*

Additional activities include antiulcer, anti-obesity, anti-arthritic, cardioprotective, wound healing, immunomodulatory, antifertility, antihypertensive, anti-asthmatic, anthelmintic, and CNS-modulating properties. Antiulcer activity has been linked to enhanced mucus secretion. Anti-obesity action has been associated with inhibition of pancreatic lipase. Anti-arthritic effects have been demonstrated in Freund's adjuvant models. Cardioprotective activity has been attributed to improved lipid profiles and reduced oxidative stress in myocardial tissue. Wound healing activity has been linked to increased collagen deposition. Immunomodulatory activity has been linked to enhanced macrophage activity. Antifertility studies show reduced sperm count and motility in male models. CNS studies suggest mild anxiolytic and sedative effects.

Ethnobotanical and Traditional Uses

Extensive ethnobotanical documentation has been available for *Albizia amara* and *Achyranthes aspera*, and their uses have been traced across several centuries in different traditional medical systems. A long history of use has been recorded in Ayurveda, Siddha, Unani, tribal healing systems, and folk medicine. The importance of these species in traditional therapeutics has been

shaped by the availability of plant parts, the recognition of symptom-based relief, and the strong cultural reliance on plant-based healing in rural communities. Knowledge transmission has been carried through oral traditions, local healers, classical manuscripts, and regional pharmacopoeias, and many of these uses have now been validated by modern pharmacological research.

Historical Uses in Ayurveda, Siddha, and Folk Medicine for *Albizia amara* and *Achyranthes aspera*

In Ayurveda, *Albizia amara* has been referenced under its traditional name, and its bark, leaves, and pods have been used for several systemic disorders. Its bark has been used as an astringent agent for conditions involving diarrhea, intestinal ulceration, and inflammatory disorders of the digestive tract. Decoctions have been used for wound cleansing and skin infections. The stem bark has been applied as a paste for skin inflammation, abscesses, and chronic dermatitis. In Siddha medicine, the plant has been used for snakebite management, respiratory congestion, and chronic inflammatory diseases. Bark infusion has been administered for coughing episodes, chest congestion, and sore throat. The root extract has been used for fever, while the leaf extract has been relied upon for chronic ulcerative lesions. Folk practitioners in South India have used *Albizia amara* bark powder for dental pain, bleeding gums, and oral infections. The seed extract has been used in limited applications for intestinal worms, while the gum exudate has been used for wound healing and joint pain. Past records show that *Albizia amara* has also been used in postpartum recovery, where decoctions have been provided to restore strength and to reduce inflammation. These uses highlight the broad therapeutic importance attributed to different parts of the plant.

Long-standing ethnomedicinal relevance has been attributed to *Achyranthes aspera*, and extensive use has been documented across India, Nepal, Sri Lanka, and Southeast Asia. In Ayurveda, the plant has been described as a potent remedy for respiratory distress, inflammatory disorders, and digestive imbalance. The whole plant has been dried and pulverized for use in chronic cough, asthmatic symptoms, bronchitic episodes, and fever. The root has been used in urinary diseases, and its decoction has been given for renal gravel, urinary retention, and dysuria. In Siddha medicine, *Achyranthes aspera* has been used for arthritis, painful swellings, and infections involving the skin and mucous membranes. Poultices prepared from leaves have been applied to painful joints and muscle inflammation. In tribal medicine, the plant has been used as an antidote for insect stings, snakebites, and skin rashes. The fresh leaf juice has been applied to

wounds to accelerate healing and prevent infections. The seeds have been used traditionally as a contraceptive agent, and paste prepared from seeds has been administered for menstrual irregularities and abdominal discomfort. Powdered root has been used in rural communities for toothache, gingival inflammation, and parasitic infections. These practices illustrate its versatile position in traditional healing systems.

Comparative Traditional Uses of Both Plants

Shared uses have also been observed between *Albizia amara* and *Achyranthes aspera*, particularly in inflammation, wound healing, digestive disturbance, and respiratory ailments. Both plants have been used for skin infections, gastrointestinal irritation, and conditions involving fever and body ache. Their overlapping use in wound repair has been supported by their strong astringent and antimicrobial properties, which have been recognized by traditional healers even prior to scientific validation. *Albizia amara* has been favored for its bark-driven astringent and anti-inflammatory properties, while *Achyranthes aspera* has been chosen for its whole-plant versatility and easy availability across diverse geographic regions. In many rural practices, formulations combining both plants have been prepared, especially for chronic inflammatory conditions, wound care, and respiratory infections. In postpartum care, *Albizia amara* has been used for tissue strengthening, while *Achyranthes aspera* has been used for its cleansing and detoxifying properties. The ethnobotanical record consistently highlights the significance of these species as therapeutic resources long before modern scientific systems evaluated their pharmacological potential.

Recent Advances and Research Trends

Major advances have been made in scientific research involving *Albizia amara* and *Achyranthes aspera*, and the recent decade has produced extensive pharmacological validation and technological innovation. Nanotechnology, synergistic combinations, modern extraction strategies, preclinical investigations, and early clinical insights have shaped current research trends. A clear shift has been noted toward the development of standardized phytoconstituent-rich extracts and advanced drug delivery systems that enhance bioavailability and therapeutic efficiency.

Nanotechnological Formulations

Nanotechnological approaches have been applied to extracts of *Albizia amara* and *Achyranthes aspera* to improve stability, solubility, and targeted delivery. Nanoparticles, nanoemulsions,

nanosuspensions, liposomes, phytosomes, and polymeric nanocarriers have been explored to encapsulate phytochemicals such as saponins, flavonoids, alkaloids, and phenolic acids. Controlled release has been observed, and enhanced antioxidant and anti-inflammatory activity has been documented at lower doses due to improved absorption and cellular uptake. Silver nanoparticles synthesized using plant extracts have been investigated for antimicrobial and anticancer effects, and strong cytotoxicity toward cancer cell lines has been recorded. Gold and zinc oxide nanoparticles prepared from *Achyranthes aspera* extracts have shown potent antibacterial and ROS-modulating activity. For *Albizia amara*, nanoscale carriers have been designed to deliver saponin fractions for hepatoprotective and antidiabetic applications. Increased bioavailability and reduced systemic toxicity have been reported, indicating that nanotechnology offers opportunities for clinical translation.

Synergistic Studies with Other Herbs

Synergistic studies have gained interest in recent research, and several investigations have been conducted to examine combined effects of these plants with other medicinal herbs. *Albizia amara* has been evaluated in multi-herbal formulations with plants such as *Terminalia chebula*, *Azadirachta indica*, and *Aegle marmelos* for enhanced antimicrobial and anti-inflammatory action. Preparations combining *Albizia amara* with *Curcuma longa* have shown amplified wound healing and antioxidant efficacy due to overlapping mechanisms of tissue repair and ROS reduction. *Achyranthes aspera* has been studied in combination with *Tinospora cordifolia*, *Ocimum sanctum*, and *Boerhavia diffusa*, where enhanced immunomodulatory and anti-inflammatory outcomes have been recorded. Synergistic antidiabetic activity has been observed when *Achyranthes aspera* extracts have been blended with *Gymnema sylvestre* and *Syzygium cumini*. Polyherbal combinations have also demonstrated superior hepatoprotective action, as shown in CCl₄ toxicity models. Many of these outcomes have aligned with traditional formulations documented in Ayurveda, indicating that synergy-based plant therapeutics are gaining scientific support.

Clinical and Preclinical Studies

Preclinical studies on both species have expanded, and detailed toxicological investigations have been conducted to evaluate safety. *Albizia amara* extracts have been tested in acute and sub-chronic toxicity models, and safe profiles have been observed at therapeutic doses. Similar safety has been reported for *Achyranthes aspera*, with no severe behavioral or histopathological

abnormalities detected in controlled studies. Preclinical pharmacological evaluations have validated historical uses, especially regarding anti-inflammatory, antidiabetic, hepatoprotective, and antimicrobial effects. Clinical data remain limited, though initial human observational studies have reported symptomatic improvement in inflammatory joint pain, mild respiratory conditions, and digestive disturbances when traditional formulations containing these plants have been used. Controlled clinical trials are limited, and larger sample sizes are required to confirm therapeutic efficacy. However, early clinical findings suggest that both species offer promising activity with favorable safety, encouraging further research toward pharmaceutical standardization and formulation development.

Conclusion and Future Prospects

A comprehensive evaluation of *Albizia amara* and *Achyranthes aspera* has highlighted strong pharmacognostic and pharmacological significance. Extensive traditional usage has been validated through systematic scientific research, and the broad spectrum of bioactivities reflects the rich presence of phytochemicals. Both species have shown strong antioxidant, anti-inflammatory, antimicrobial, antidiabetic, hepatoprotective, and cytotoxic effects. The pharmacognostic identity of each plant has been well-documented through macroscopic, microscopic, physicochemical, and fluorescence analysis. Extractive values and ash content have further supported quality assessment. Phytochemical screening has revealed diverse metabolites with therapeutic relevance, and recent analytical advancements have allowed structural elucidation of several bioactive compounds. These insights support the integration of these plants into evidence-based herbal medicine.

Research Gaps and Future Directions

Several research gaps remain, and new studies are required to advance clinical applicability. Standardized extraction techniques and purification-based isolation methods need further refinement. Long-term toxicity studies and detailed pharmacokinetic profiles remain limited, and these must be addressed. Clinical trials involving large populations must be conducted to validate therapeutic efficacy. Nanotechnology-based formulations should be tested *in vivo* to evaluate long-term stability and controlled release. Synergistic polyherbal combinations require deeper mechanism-based investigations. Genomic and metabolomic studies should be conducted to map biosynthetic pathways of major phytoconstituents. Greater emphasis must be placed on developing sustainable cultivation, conservation, and quality control strategies to ensure

consistent pharmacological outcomes. These directions will shape the future research landscape and enhance the integration of *Albizia amara* and *Achyranthes aspera* into modern therapeutic systems.

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