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RESEARCH ARTICLE OPEN ACCESS

Exploring the Ethnopharmacological Potential of *Holoptelea integrifolia*: Phytochemical and Pharmacological Perspectives

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Abstract:

Holoptelea integrifolia, prominently observed as an ethnomedicine, has gained a significant interest in research. The interaction between conventional knowledge with advanced research enhances its utility for developing safe, efficient, and sustainable therapeutics. Few characteristics of the plant are the unique anatomy of its leaves, bark, seeds and fruits along with diverse bioactive compounds including glycosides, flavonoids, phenols, saponins, triterpenoids, and phytosterols. The secondary metabolites gave the plant its alleged properties such as antibacterial, antifungal, antioxidant, anti-inflammatory, analgesic, antidiabetic, antiviral, anthelmintic, and wound healing activity. Recently employed analytical techniques have identified the key phytoconstituents such as β-amyrin, lupeol, friedelin, and β-sitosterol, highlighting their therapeutic significance. The evidence positions *Holoptelea integrifolia* as a valuable candidate for drug development and shows that it is a promising candidate after ensuring the safety, efficacy and standardisation. This review critically evaluates the pharmaceutical potential of *Holoptelea integrifolia*, highlighting it as a source for novel therapeutic agents including the comprehensive analysis of its morphological characteristics, phytochemical studies, ethnomedicinal uses, pharmacological activities and the way it can be used in the future.

Keywords — *Holoptelea integrifolia*, phytochemistry, ethnomedicinal uses, wound healing, anti-inflammatory, pharmacological activity.

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INTRODUCTION

Holoptelea integrifolia is a huge deciduous tree belonging to the Ulmaceae family, is commonly referred to as Indian Elm or "Chilbil Papri" [1]. It is found throughout the Northern Hemisphere's tropical and temperate regions, from the Indian Peninsula to Indo-China, Sri Lanka, and remote Pacific Islands, where *Holoptelea integrifolia* is thought to have originated [2].

The following is the Taxonomical classification:
Domain: Eukaryota
Kingdom: Plantae
Division: Magnoliophyta
Class: Magnoliopsida
Order: Urticales
Family: Ulmaceae
Genus: *Holoptelea*
Species: *integrifolia* [3].

It is a huge deciduous tree that may grow to a height of 25 to 35 meters. Its bark is smooth or peeling in corky scales, and it is greyish to whitish grey in thickness, measuring about 6 to 8 mm. When bark and crushed leaves are applied, it releases an unpleasant odour. Simple, elliptic-ovate to ovate, alternating, stipulate, acuminate, whole, and pinnately veined leaves are all present. The tiny

polygamous blooms, which range in colour from greenish yellow to greenish purple, are produced in axillary fascicles or short racemes. Bisexual flowers have five stamens, whereas male flowers have eight. The superior, unilocular, compact, and stigmatized ovaries are Bifid. A little kidney shaped seed is present in the light brown, indehiscent, one-seeded, winged samara fruits, which are roughly 2.5 to 3.5 cm in length and 1.5 to 2.5 cm in width [1,3].

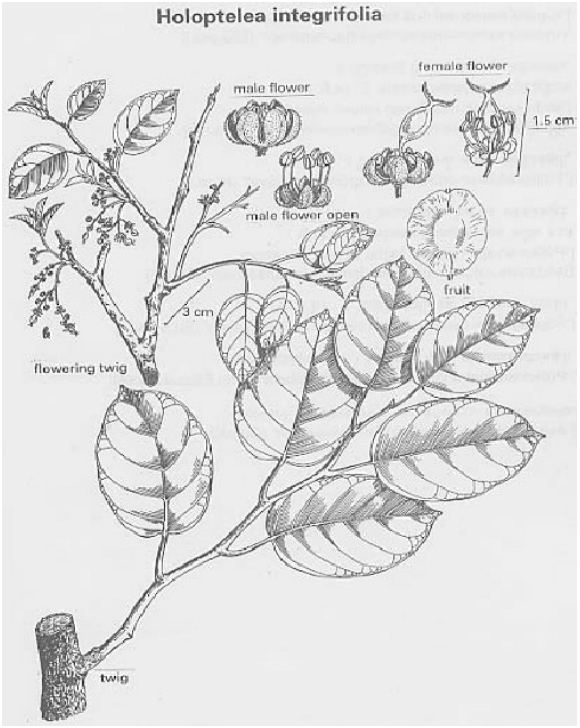


Fig. 1: A twig of *Holoptela integrifolia* showing significant different plant parts [1].

Fig. 2: J.M.Garg. (2008, February 2). *Holoptelea integrifolia W IMG 1284*

PHYTOCHEMICAL STUDY

TABLE I
PHYTOCHEMICAL CONSTITUENTS OF *HOLOPTELEA INTEGRIFOLIA*

Plant Part	Major Constituents	Reported Activities
Bark [6,8, 10, 17, 19]	Steroids/Terpenoids: Lupeol, Friedelin, β-sitosterol, Betulin, Betulinic acid Tannins in traces.	Anti-inflammatory; Antimicrobial; Wound healing; Antioxidant
Leaves [6, 12, 13, 14, 18, 19, 20,21 23, 24]	Flavonoids: Quercetin, Rutin Phenolic compounds: Tannins, Gallic acid Steroids/Terpenoids: β-sitosterol, Lupeol, Saponins	Antioxidant; Antibacterial; Analgesic; Anti-inflammatory
Seeds [3, 26, 28]	Fatty Acids: Linoleic acid, Oleic acid, Palmitic acid, Stearic acid Sterols: β Sitosterol, Campesterol, Stigmasterol	Antidiabetic; Anti-inflammatory; Antioxidant; Antibacterial
Roots [3, 16, 22]	Steroids/Terpenoids: β-Sitosterol, Triterpenoids Alkaloids	Antioxidant Hepatoprotective; Antimicrobial

1. Fruit and Seed:

The chemical compositions of the fruits and seeds of *Holoptelea integrifolia* varied substantially when subjected to physicochemical examination. The fruits contained a polar and nonpolar mixture that might account for their medicinal properties. The approaches used for extraction were by n-hexane, followed by ethanol and finally with an aqueous solution. The above content consisted of saponins, phenols, reducing sugars, tannins, and alkaloids [4]. The seeds and the seed oil were verified to attain a considerable amount of lipid content, primarily unsaturated oleic acid (47%) and saturated palmitic acid (37%), and also good protein, carbohydrates, fiber, and minerals, keeping the anti-nutritional factors in permissible limits. *Holoptelea integrifolia* seed oil contained 57.1% total unsaturated fatty acids (TUSFAs) and 42.9% total saturated fatty acid (TSFAs). The oil also possessed 14.8% malvalic acid, an unusual but important fatty acid in industry [5].

2. Root:
The phytochemical analysis of the plant, terpenoids, sterols, proteins, carbohydrates, alkaloids, phenols, flavonoids, glycosides, quinones, and saponins can be detected from the species. Many bioactive compounds, such as Holoptelin-A, Holoptelin-B, friedlin, epifriedlin, β-amyrin, stigmasterol, β-sitosterol, 1,4-naphthoquinone, betulin, betulinic acid, hexacosanol, and octacosanol, have been successfully isolated and described from this species [3].

3. Bark and Stem:
The young stem bark methanolic extract of *Holoptelea integrifolia* was subject to preliminary phytochemical screening and was found to contain some bioactive components such as carbohydrates, glycosides, steroids, triterpenoids, flavonoids, saponins, tannins, and phenolic compounds [6]. Plant material was authenticated and standardized by generating a unique fingerprint profile using HPTLC analysis of the hexane extract. Preliminary physicochemical analysis in UV and HPTLC profiling results will serve as biomarkers for authentication and quality control of raw drugs, therefore helping to prevent adulteration [7]. Similarly, ethanolic extracts from the stem bark gave positive results for the presence of alkaloids, tannins, glycosides, flavonoids, phenols, saponins, and reducing sugars, in addition to specific compounds such as holoptelin-A (epi-friedelinol palmitate), holoptelin-B (epi-friedelinol stearate), triterpenoid fatty acid esters, friedelin, and epi-friedelinol that may have therapeutic value in ailments such as rheumatic inflammation, fever, dysentery, convulsions, ulcers, and bladder cancer. Additionally, thin layer chromatographic (TLC) fingerprinting and histological analyses of the stem bark provided diagnostic characteristics useful for distinguishing the authentic drug from possible adulterants [8].

4. Leaves:
Holoptelea integrifolia leaves were screened for phytochemical analysis. Most bioactive compounds identified included proteins, carbohydrates, ketones, sugars, sterols, alkaloids, flavonoids, tannins, saponins, terpenoids, glycosides, and steroids, while anthraquinones were not found. Confirmed by preliminary phytochemical screening and HPTLC fingerprinting, these contents are responsible for the antioxidant and antimicrobial properties of the leaves [9,10,11]. Furthermore, these alcoholic extracts of leaves contained alkaloids, tannins, cardiac glycosides, saponin glycosides, cyanogenetic glycosides, and anthracene derivatives, with compounds like hexacosanol, octacosanol, β-sitosterol, and β-amyrin [11].

ETHNOMEDICINAL USES

Ethnomedicinally, the various parts of *Holoptelea integrifolia* are used by tribal people for the treatment of various ailments [3].

TABLE III
ETHNOMEDICINAL USES OF *HOLOPTELEA INTEGRIFOLIA*.

DESCRIPTION

Bark of H. integrifolia varies from greyish to dark brown and exfoliates in patches. It has a characteristic odor with a bitter taste. Bark is rough and uneven on the outside and has a fibrous fracture within. The basic configuration of leaves is spray-like with simple, alternative, and elliptic ovate shape measuring from 8 to 15 cm long. Flowers are small-sized, greenish-yellow, and unisexual whereas the fruit is flat samaras which has papery wings aiding wind dispersal [**Error! Bookmark not defined.**,**Error! Bookmark not defined.**,**Error! Bookmark not defined.**].
These morphological features with these distinct differences help organoleptic identification in raw drug evaluation.

Microscopic Characteristics:

Microscopic sections cut through transverse sections indicate:

- Multiple layers of cork cells that are tangentially elongated brown cells.
- The secondary phloem consists essentially of sieve elements, parenchyma, and stone cells.
- Presence of calcium oxalate crystals and starch grains scattered throughout the phloem tissues.
- Medullary rays running radially and having thick-walled lignified cells [1,7, **Error! Bookmark not defined.**].

T.S. of the leaf shows dorsiventral anatomy with:

- Upper and lower epidermis with thin cuticle.
- Mesophyll differentiated into palisade and spongy parenchyma.
- In midrib portion collateral vascular bundles.

Plant Part	Traditional Uses	Formulation/ Mode of Preparation
Leaves [12,13]	Used for the treatment of inflammation, diabetes, oxidative stress, redness of the skin, and microbial infections.	Generally used in decoctions.
Bark (fresh) [**Error! Bookmark not defined.**]	Treatment of rheumatic swellings and inflammation	Boiled to obtain mucilaginous extract; applied externally
Bark (dried) [**Error! Bookmark not defined.**]	Further reduces rheumatic pain and swelling	Powdered and applied topically after extraction
Stem bark [**Error! Bookmark not defined.**]	Anti-inflammatory for eye conditions Treatment of inflammatory lymph glands, fever, scabies, and ringworm	Topical Preparation
Seeds [**Error! Bookmark not defined.**]	Managing Inflammation and gastrointestinal disturbances.	Consumed
Fruit [**Error! Bookmark not defined.**]	Used in traditional remedies for analgesia	Applied in formulations for pain relief and inflammation
Heartwood [17]	Digestive ailments, and as a detoxifying agent	Incorporated in formulations aimed at treating obesity

- Anomocytic stomata and unicellular trichomes on the epidermis. [**Error! Bookmark not defined.**,**Error! Bookmark not defined.**].

These anatomical markers provide microscopic authentication parameters crucial for distinguishing H. integrifolia from adulterants or substitutes.

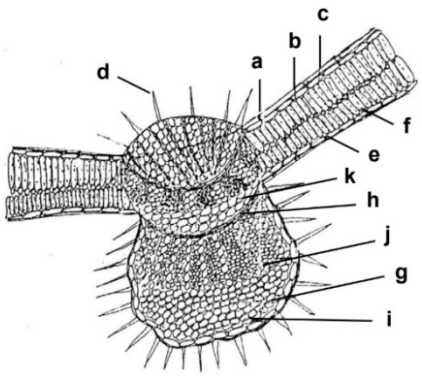
Fig no. 3: Transverse section of leaf of *Holoptelea Integrifolia* (Roxb) Planch. (a) Parasitic stomata with cuticle; (b) Upper eqidermis; (c) Lower eqidermis; (d) Upper epidermal trichome; (e) Upper palisade zone; (f) Lower epidermis; (g) Spongy Parenchyma; (h) Pericycle fibre; (i) Lower zone spongy parenchyma; (j) Xylem vessel; (k) Phloem vessel. **Error! Bookmark not defined.**

EXTRACTION TECHNIQUES

The [for instance, stems, leaves, and seeds] of *Holoptelea integrifolia* were well rinsed with tap water to get rid of fine dust and debris and the plant material was finally washed with distilled water. The cleansed plant material was shade-dried for nearly two weeks at room temperature to a constant weight [14,1]. The dried material was later coarsely powdered using a mechanical grinder. The powder was kept in an airtight container at 4°C for further use [11,3].

The extraction was performed using [Soxhlet extraction/maceration] method as described before with some modifications [11,17,5]. In brief, [generally around 500 g] dried powder was packed in a thimble and continuously extracted in a Soxhlet apparatus with [e.g., 1.5 L of methanol] as solvent [11,12]. The extraction lasted for [e.g., 48-72 hours] or until the solvent in the siphon tube became colorless, indicating the completion of extraction [11].

Maceration involved soaking the plant powder in an appropriate solvent (water, ethanol, etc.) kept in a closed vessel at room temperature for a 48 to 72-hour period with periodic shaking [20,21,15]. The mixture was then filtered, and the marc (residue) was re-extracted by repeating the process to ensure complete extraction [17]. The combined filtrates from either method were concentrated under reduced pressure using a rotary evaporator (Buchi, Switzerland) at a controlled temperature of [e.g., 40-50°C] to obtain a crude extract [11,27,25]. The percentage yield of the extract was determined as per the formula given and the resultant extract was stored at 4°C for future phytochemical and pharmacological investigations [11,17].



The following is the standard formula for calculation the Percentage yield obtained:

Percentage Yield (%) = (Weight of the extract obtained / Weight of the dried plant powder taken) × 100

TOXICITY STUDIES

1. Acute Toxicity Studies

Acute oral toxicity studies in Wistar albino rats and Swiss mice following OECD guidelines demonstrated that high doses of extracts of H. integrifolia bark and leaves were not toxic. Animals treated with single doses of as much as 2000 mg/kg showed:

- No signs of mortality or behavioural changes
- Significantly changed food intake, body weight, or locomotor activity.
- Nothing gross pathological changes and the specific vital organs were liver, kidney, heart, or spleen observed [17,3].
- These results therefore support their classification as plant extracts relatively safe, the LD_{50}-high indicates a relatively low acute toxicity [3].

2. Subacute and Subchronic Toxicity

Repeated-dose toxicity studies suggest an excellent safety profile. Hence, there were no significant hematological and biochemical abnormalities observed when rats were subjected to treatment for a long time with H. integrifolia extracts [3]. It was

observed that key liver enzymes (AST, ALT, ALP) and renal markers (creatinine, urea) were kept within normal physiological limits. Histopathological studies on tissues from the liver and kidneys revealed no cellular necrosis-a finding that confirms that repeated administration does not deprive the organ of significant injury [17]. These findings depict very good tolerability systemically and show that there is no cumulative toxicity from subchronic exposure.

3. Chronic and Reproductive Toxicity

Long-term administration of extracts obtained from H. integrifolia were reportedly safe in animal models without causing mortality or any significant systemic toxicity. Further include some specific bioactivities concerning reproduction. There is research evidence suggesting that at higher doses, the plant has temporary antifertility effects, which are reversible after the cessation of treatment [3]. Such phenomena are often attributed to phytoestrogenic or other hormonally active constituents and fit to those traditional uses and thus require further works for detailed mechanistic elucidation.

PHARMACOLOGICAL ACTIVITES

Fig. no. 4: Pharmacological activities of *Holoptelea integrifolia*

1.Anti-inflammatory:

Inflammation is the bodily response to an aggressive agent, together with a combination of vasodilation and movement of fluids and immune cells to the target area. Pain serves as one of the main features of inflammation, and it results from the direct activation of nociceptors or mediators of inflammation. Fundamentally, it is defensive in nature and seeks to eliminate the microorganisms, poisons, and their byproducts which make up necrotic tissue that cause harm [16].

Extracts from *Holoptelea integrifolia* leaves have shown strong anti-inflammatory properties in experimental studies. Leaf extracts in the following forms significantly reduced paw oedema in carrageenan-induced inflammation: aqueous (500 mg/kg), ethanolic (250 and 500 mg/kg), and methanolic (100 and 500 mg/kg). This suggests that these extracts may be used to regulate both acute and chronic inflammation [17]. The aq. extract demonstrates anti-inflammatory properties indicating its natural alternatives to NSAIDs [18].

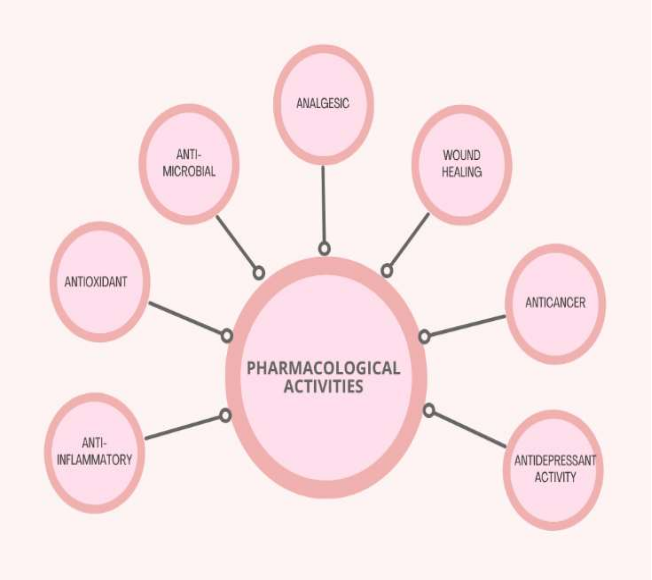
In another study, it was observed that after three hours of intervention, the aq. extracts have significantly reduced paw oedema without causing liver or renal damage, producing effects similar to those of diclofenac sodium [13]. This might be because of the steroids found in the leaves and bark found by the preliminary phytochemical investigation [19].

Additionally, both leaf powders and decoctions shown efficacy in carrageenan and formalin-induced models, although the powders performed better in sub-acute circumstances while the decoctions performed better in acute inflammation [20].

2. Anti-microbial:

The substance which kills or stops the growth of microbes is called as an Antimicrobial.

The aq. extract of *Holoptelea integrifolia* was observed to exert strong in-vitro antibacterial activity against Salmonella typhi, Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli, and Streptococcus pyogenes. Maximum sensitivity was found by S. pyogenes and E. coli. At higher



concentrations, the extract had good broad-spectrum antibacterial action against P. aeruginosa,

S. aureus, and S. typhi, indicating moderate to well-susceptibility. Terpenes, tannins, saponins, phenols, and other phytoconstituents present in the leaves of this plant are thought to be responsible for its antibacterial properties [21].

Another study reveals that hexadecane, which is present in several extracts, is very efficient against P. aeruginosa, while siloxanes, which are present in the methanolic leaf extract, exhibit antibacterial action. Fatty acids, terpenes, and other bioactive substances are more likely to be responsible for the antimicrobial properties of acetone root extract and ethyl acetate stem extract [22].

Various pathogens and solvent screenings were employed, and they revealed that, in terms of root and stem extracts were usually more effective than leaf extracts [6,24,23]

Strong antibacterial properties against Bacillus subtilis, P. aeruginosa, Micrococcus luteus, E. coli, and Proteus vulgaris were also discovered in ethanol extracts from leaves, bark, and fruits [24].

All the findings confirm to the use of *Holoptelea integrifolia* as a strong natural antimicrobial agent.

3. Antioxidant**:**

Living cells rely on the oxidation process. Oxidative damage to lipids, proteins and DNA can lead to several diseases associated with oxidative stress.

Compounds called antioxidants help neutralize the reactive oxygen species (ROS) and stop the harm they do. In addition to reducing power assays, ferric thiocyanate (FTC), thiobarbituric acid (TBA), DPPH (2,2-diphenyl-1-picrylhydrazyl), nitric oxide, and superoxide radical scavenging assays were used to evaluate the in-vitro antioxidant activity of ethanolic, methanolic, and aqueous extracts of *Holoptelea integrifolia* stem bark [**Error! Bookmark not defined.**].

In a study, it was observed that strong hydroxyl radical scavenging and total reduction properties were exhibited by petroleum ether (PEHI) and methanolic (MHI) leaf extracts, with PEHI exhibiting greater potency than MHI [**Error! Bookmark not defined.**].

The reason for this is that these metabolites contain steroids, flavonoids, and phytosterols that help quench free radicals and stop oxidation reaction chains [**Error! Bookmark not defined.**]. At a concentration of 250 µg/ml, ethanolic, acetonic, and aqueous extracts from the leaves and bark also shown strong antioxidant properties [**Error! Bookmark not defined.**].

4. Analgesic:

An analgesic is a drug that relieves pain. At 500 mg/kg, the crude ethanolic extract of *Holoptelea integrifolia* leaves produced a substantial ($P < 0.05$) analgesic effect [**Error! Bookmark not defined.**]. While n-butanol and water extracts showed only little analgesic action, with the greatest response occurring 150 minutes after injection, ethyl acetate extract was shown to be moderately efficacious. The presence of the bioactive component flavonoids may be the cause of the reported analgesic effect [25]. More research in this area is necessary.

5. Wound Healing:

After an injury, the biological process that seeks to repair the damaged tissue is called Wound Healing.

Using incision and excision wound models, the methanolic extracts of *Holoptelea integrifolia's* stem bark and leaves at a dosage of 50 mg/550 mm^2 demonstrated excellent wound-healing ability in albino rats, with a healing efficiency of over 90% [19].

Friedelin and lupeol are the main bioactive ingredients in the n-hexane extract of stem bark, according to phytochemical research. While lupeol is well-known for its anti-inflammatory, antioxidant, and wound-healing qualities, friedelin is a strong anti-inflammatory. Friedelin and lupeol, two of the fresh stem bark extract's separated constituents, seem to encourage re-epithelialization and MMP-9 activity, which improves tissue regeneration and repair. This data helps to know the reason as for why *Holoptelea integrifolia* show the activity of wound healing [26].

6. Anti-cancer:

Uncontrolled cell growth is known as Cancer and the drugs to cure cancer are known as Anti-cancer agents.

Based on the initial phytochemical screening, alkaloids, glycosides, sterols, flavonoids, tannins, and saponins were all found in the ethanolic leaf extract of *Holoptelea integrifolia* (EHI). In mice with Dalton's Ascitic Lymphoma (DAL), the anticancer impact resulted in a significant reduction in tumor volume and a dose-dependent increase in survival time. In order to demonstrate systemic recovery, EHI also returned a few abnormal hematological markers to levels that were close to normal [27].
Flavonoids, alkaloids, and terpenoids with antioxidant and chemopreventive qualities that can alter signal transduction by preventing angiogenesis and reducing cell proliferation are the primary sources of anticancer potential of the plant. This proves *Holopetela integrifolias* potential as a natural cancer treatment [8].

7. Antidepressant activity:
The use of both methanol and ether leaf extracts from *Holoptelea integrifolia* have confirmed the reduced inactivity periods in the Force Swim Test and Tail Suspension Test. As compared to the common medication of fluoxetine, the petroleum extract shows greater potent results. Modification of the adrenergic, dopaminergic, and serotonergic neurotransmission pathways may be the reason of the antidepressant effect [28].
The plant *Holoptelea integrifolia* must be studied extensively because it shows promising results as a form of natural antidepressants.

PHARMACOLOGICAL SIGNIFICANCE

The evidence collected so far indicates that *Holoptelea integrifolia* is a medicinal plant that has multiple therapeutic aspects, underpinning pharmacological significance by means of a prima facie rich repertoire of bioactive compounds. The plant continues to boast a range of activities including but not limited to anti-inflammatory, antioxidant, and antimicrobial [.3,18,13]. Most of these bioactivity properties stem from its richness in phytoconstituents such as triterpenoids, flavonoids, and other phenolic compounds, thus rationalizing the connection between traditional knowledge and modern pharmacology [14,3].

CONCLUSION

According to the findings, *Holoptelea integrifolia* is a notable medicinal plant with multiple promising uses because of its diverse morphological features and a rich distribution of phytochemicals spread across its leaves, bark, stem and fruits. Its pharmacological activities are supported by both traditional usage and modern medicine. The performed tests and analysis of the bioactive compounds give us the accurate information as for the specifications of the chemical constituents and their potential.
Ethnomedicinal data provides the information on the therapeutic potential while regarding the traditional knowledge. Further detailed investigations including clinical trials, toxicological assessments and various studies can help with the widespread adoption in modern medicine.
In conclusion, *Holoptelea integrifolia* stands as an ideal candidate and a promising drug in the future study of both drug discovery and sustainable development soon, combining the traditional knowledge and modern therapeutic science.

FUTURE OUTCOMES

Holoptelea integrifolia is a source of diverse phytomedicine with the potential against serious ailments, including cancer, malaria, and viral infections, as well as metabolic syndrome. Its traditional applications for treating inflammation, leprosy, eczema, and malaria are corroborated by modern pharmacological studies [19].
Further prospects are in developing bioactive compounds lupeol and friedelin into pharmaceutical formulations such as wound-healing cream and anti-inflammatory drugs [28]. Other possibilities include the development of nutraceuticals, screening for new antimicrobials and antivirals, and validating its efficacy through clinical trial processes [19].

According to the above article, we can tell that *Holoptelea integrifolia* is a promising drug for the future formulations.

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