Pharmacognostical And Preliminary Phytochemical Studies of Nothapodytes Nimmoniana

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Nothapodytes nimmoniana (Amruta Tree / Stinking Tree)

Nothapodytes nimmoniana flower cluster

***Abstract*—Nothapodytes nimmoniana (J. Graham) Mabb., commonly known as the Amruta Tree or Stinking Tree, is a medicinally significant small-to-medium-sized tree native to the Western Ghats of India and classified under the family Icacinaceae. It is recognized as one of the richest natural plant sources of camptothecin (CPT), a pentacyclic monoterpene indole alkaloid with potent anticancer properties. This review synthesizes findings from five peer-reviewed research publications to present a comprehensive evaluation of the plant's phytochemistry, pharmacological activities, distribution of bioactive compounds, biotechnological production strategies, and conservation status.**

Studies reviewed herein confirm that CPT is unevenly distributed across plant tissues, with roots and shoots demonstrating the highest concentrations (up to 1.87 mg/g dry weight). Endophytic fungi isolated from N. nimmoniana, particularly Alternaria alstroemeriae and Alternaria burnsii, have shown extraordinary potential for sustainable CPT production without harvesting the endangered plant. The principal mechanism of CPT involves selective inhibition of DNA topoisomerase I, inducing irreversible DNA strand breaks in cancer cells. Multiple pharmacological activities beyond anti-cancer effects — including anti-microbial, antioxidant, anti-malarial, anti-helminthic, and anti-inflammatory properties — have also been documented. The overexploitation of this plant has led to a minimum 20% population decline, necessitating urgent conservation and sustainable production strategies. Future research directions include genomic exploration, metabolic engineering, and nanoparticle-based drug delivery systems.

***Index Terms*—Nothapodytes nimmoniana, Camptothecin, Topoisomerase I Inhibitor, Endophytes, Western Ghats, Anticancer Alkaloid, Phytochemistry, Conservation**

I. INTRODUCTION

Plants have historically served as the foundation of medicinal systems across the globe, and continue to yield pharmacologically significant compounds that form the basis of modern oncology drugs. Nothapodytes nimmoniana (J. Graham) Mabb., belonging to the family Icacinaceae and order Icacinales, is a small-to-medium deciduous tree found predominantly in the shola forests of the Western Ghats of India, particularly in the states of Karnataka, Kerala, Maharashtra, Tamil Nadu, and parts of Andhra Pradesh. The plant is locally known as 'Amruta' in Kannada — a name meaning 'nectar of immortality' — and 'Narkya' in Sanskrit, owing to its extraordinary medicinal properties.

The plant gained global scientific interest following the discovery that it contains camptothecin (CPT), one of the most commercially valuable anticancer alkaloids of the 21st century, ranking third in demand

after Taxol and Vinca alkaloids (Mohinudeen et al., 2021). CPT was originally isolated in 1966 from the Chinese tree Camptotheca acuminata by Wall and colleagues, and subsequently identified in N. nimmoniana — which has since been established to carry the highest CPT content of any plant species, reaching approximately 0.3% in some populations (Uma Shaanker et al., 2008). Two semi-synthetic water-soluble derivatives of CPT — Topotecan and Irinotecan (CPT-11) — are approved by the United States Food and Drug Administration (USFDA) for the clinical treatment of ovarian, colorectal, and lung cancers, as well as pediatric brain tumors.

Despite its immense pharmaceutical significance, N. nimmoniana faces severe ecological threat. Driven by the exponential global demand for CPT, the tree is heavily exploited, leading to a documented population decline of at least 20% in recent decades (Uma Shaanker et al., 2008). The International Union for Conservation of Nature (IUCN) has consequently categorized the species as 'Vulnerable,' placing it on the conservation radar. This review critically appraises five key published research papers that collectively address the phytochemical profiling, tissue-specific distribution of bioactive compounds, endophytic fungal production systems, comprehensive pharmacological activities, and conservation strategies associated with N. nimmoniana.

II. MOLECULAR PROPERTIES OF KEY BIOACTIVE COMPOUNDS

Before examining the literature, it is essential to understand the key molecular entities studied in the context of N. nimmoniana. Camptothecin (CPT) is the primary pharmacologically active compound isolated from this plant. It is a pentacyclic lactone alkaloid structurally characterized by a planar fused ring system composed of five rings: A (benzene), B (pyridine), C (alpha-hydroxy-lactone), D (pyridone), and E (the critical lactone ring essential for biological activity).

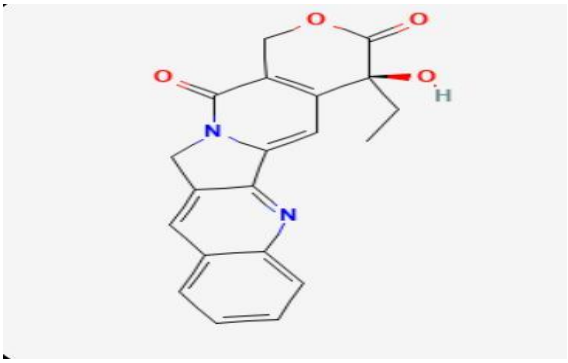
The alpha-hydroxy lactone moiety of the E ring is specifically responsible for the inhibitory interaction with the DNA–topoisomerase I complex. The presence of an intact, closed lactone ring at physiological or acidic pH is an absolute prerequisite for biological activity. At neutral or basic pH, the lactone undergoes reversible hydrolytic ring-opening to form the carboxylate form, which has significantly reduced cytotoxicity. This structural sensitivity has been a major challenge in pharmaceutical development of CPT-based drugs.

Table 1: Molecular Properties of Key Bioactive Compounds

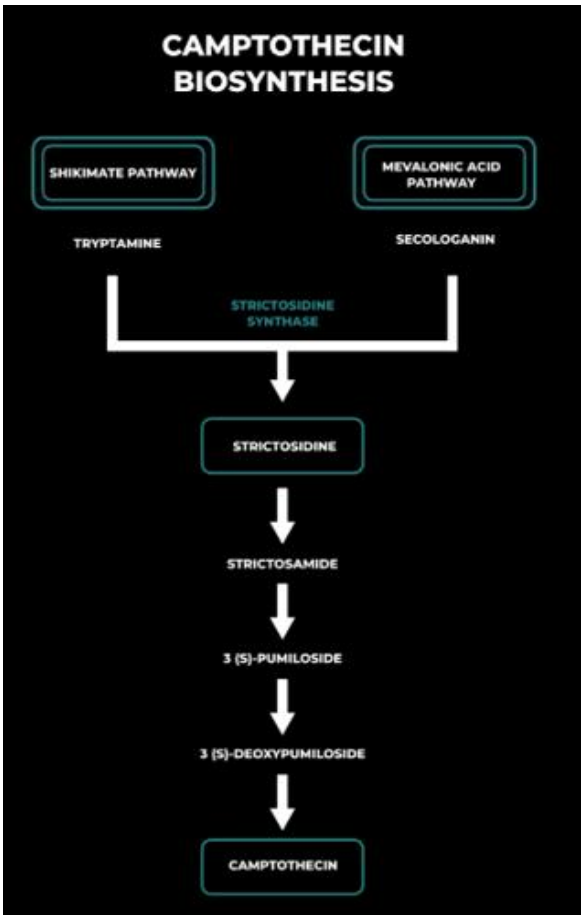
Compound	Molecular Formula	Molecular Weight	CAS No.
Camptothecin (CPT)	$C_{20}H_{16}N_2O_4$	348.35 g/mol	7689-03-4
9-Methoxycamptothecin	$C_{21}H_{18}N_2O_5$	378.38 g/mol	78287-27-1
Topotecan (CPT derivative)	$C_{23}H_{23}N_3O_5$	421.45 g/mol	123948-87-8
Irinotecan (CPT-11)	$C_{33}H_{38}N_4O_6$	586.68 g/mol	97682-44-5

Table 1: Molecular weights

In addition to CPT, N. nimmoniana also contains 9-methoxycamptothecin (9-Me-CPT), a monomethylated derivative with enhanced lipophilicity. Research reviewed by Gayakwad et al. (2024) highlights that this compound too exhibits cytotoxic properties and is co-extracted alongside CPT during standard extraction protocols. The presence of multiple alkaloids underscores the complex phytochemical richness of this plant species.



Molecular Structure of Camptothecin

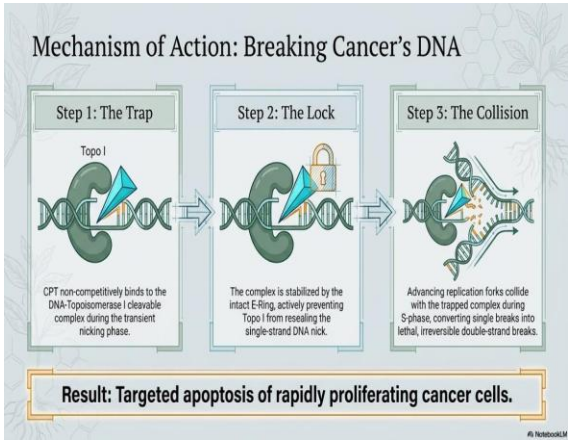


Biosynthesis of Camptothecin

III. MECHANISM OF ACTION OF CAMPTOTHECIN

The primary pharmacological mechanism of camptothecin (CPT) and its derivatives involves selective and potent inhibition of the nuclear enzyme DNA topoisomerase I (Topo I). Topoisomerase I is an essential cellular enzyme responsible for relieving torsional stress in the DNA double helix during DNA replication and transcription. Under normal cellular conditions, Topo I creates transient single-strand breaks in DNA, allows the DNA to rotate around the intact strand to relieve supercoiling, and then reseals the nick — a process collectively described as the 'catalytic cycle.'

CPT functions as a non-competitive inhibitor of topoisomerase I by intercalating into the ternary cleavable complex formed between Topo I and the DNA strand during the transient nicking phase. Specifically, CPT stabilizes this normally transient DNA–Topo I covalent complex, thereby preventing the relegation step. The result is an accumulation of persistent single-strand DNA breaks that, upon collision with advancing replication forks during S-phase of the cell cycle, are converted into lethal double-strand DNA breaks. These irreversible double-strand breaks trigger apoptotic cell death pathways, making CPT specifically cytotoxic to actively dividing (rapidly proliferating) cancer cells while exhibiting relatively less toxicity toward quiescent normal cells.



Mechanism of Action: Breaking Cancer's DNA

The requirement for an intact alpha-hydroxy lactone E-ring (closed lactone conformation) for this ternary complex formation explains why the carboxylate (open) form of CPT — predominant at physiological pH — has significantly reduced anticancer potency. This structural insight has driven the development of semi-synthetic analogues. Irinotecan (CPT-11) is a prodrug that is enzymatically converted to its active metabolite SN-38 by carboxylesterases in vivo; SN-38 is 100 to 1,000 times more potent than irinotecan itself as a Topo I inhibitor. Topotecan, on the other hand, carries a basic dimethylaminomethyl group at position 9 that enhances water solubility while retaining potent Topo I inhibitory activity.

Beyond CPT, the biosynthetic pathway in N. nimmoniana proceeds through the general monoterpene indole alkaloid (MIA) pathway. The precursors tryptamine and secologanin condense under the action of strictosidine synthase to form strictosidine — the universal MIA precursor. Subsequent enzymatic transformations, including the action of strictosamide epoxidase and other enzymes

yet to be fully characterized, ultimately yield CPT. A landmark 2025 genomic study (Wang et al., 2025) of N. nimmoniana's chromosome-level genome revealed that the plant is an allotetraploid with a 5 Gb genome encoding 92,630 genes, and that gene duplication events from allotetraploidization likely contribute to enhanced CPT production in this species.

IV. COMPARATIVE ANALYSIS OF RESEARCH PAPERS

A systematic comparative analysis of the five reviewed studies reveals convergent conclusions alongside important methodological and contextual differences, each contributing a unique perspective to the overall understanding of N. nimmoniana.

4.1 Similarities Across Studies

All five studies uniformly affirm that camptothecin is the primary bioactive compound of pharmacological significance in N. nimmoniana, and that the plant is under imminent ecological threat due to its overexploitation. There is consistent agreement across the literature that sustainable production alternatives — whether tissue culture, endophyte cultivation, or targeted harvesting of elite individuals — are urgently required. The quantification of CPT using HPLC or related chromatographic techniques is the universally adopted analytical standard across studies, reflecting methodological consistency.

4.2 Key Differences

The studies diverge significantly in their methodological approaches and focal scales. Mingzhang et al. (2011) focus at the intra-plant (tissue level) scale, while Pai et al. (2015) operate at the inter-individual and population level, and Uma Shaanker et al. (2008) scale up further to the regional landscape level through ecological niche modelling. Mohinudeen et al. (2021) take an entirely different approach — shifting the focus from the plant entirely to its associated endophytic microbiome as an alternative production platform. Gayakwad et al. (2024) provide the broadest scope, covering not just CPT but the full pharmacological spectrum of the plant.

Table 2: Comparative Analysis of Reviewed Studies on Nothapodytes nimmoniana

Study	Focus Area	Key Method	CPT Yield/ Finding	Approach
Mingzhang et al., 2011	CPT distribution in plant parts	HPLC	1.87 mg/g (roots); 1.07 mg/g (shoots)	Extraction-based
Mohinudeen et al., 2021	Endophytic fungi as CPT source	LC-MS, ^{13}C labelling	426.7 µg/g DW (Alternaria sp.)	Biotechnological
Pai et al., 2015	UFLC-based population screening	UFLC-PDA	Inter-individual variability in CPT	Phytochemical profiling
Gayakwad et al., 2024	Pharmacological & extraction review	Literature synthesis	Multi-activity (antitumor, anti-microbial, antioxidant)	Comprehensive review
Uma Shaanker et al., 2008	Chemical profiling & sustainable production	Ecological niche modelling	Highest CPT ≈0.3%; 20% population decline	Conservation-focused

Table 2: A structured comparison of the five key studies reviewed in this report, highlighting focal areas, methods, key findings, and strategic approaches.

4.3 Effectiveness of Approaches

From a conservation perspective, the endophytic fungal production approach pioneered by Mohinudeen et al. (2021) is arguably the most impactful, as it potentially eliminates the need for plant extraction altogether. From a practical extraction standpoint, Mingzhang et al. (2011) offer the most immediately actionable guidance. For long-term sustainable forest management, the population-level elite individual identification methodology of Pai et al. (2015) and the ecological niche modelling approach of Uma Shaanker et al. (2008) offer complementary frameworks. Gayakwad et al. (2024) provide the most comprehensive pharmacological evidence base for the plant's medicinal utility beyond anti-cancer applications.

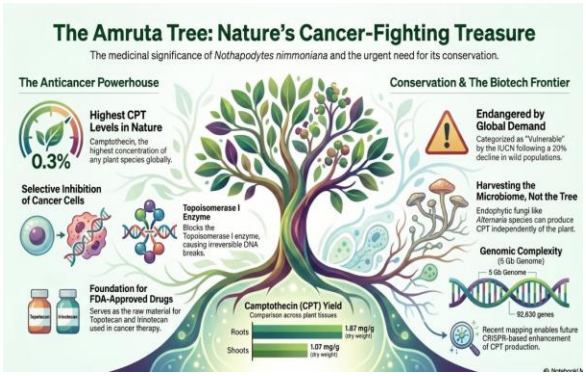
V. RESULTS AND DISCUSSION

The combined findings of the five reviewed studies paint a detailed and scientifically rigorous picture of N. nimmoniana as an extraordinarily valuable medicinal plant facing an unprecedented conservation crisis.

In terms of CPT yield, the data across studies demonstrates considerable variability depending on the tissue type, developmental stage of the plant, geographic origin, and extraction methodology. The highest plant-derived yields recorded are in roots (1.87 mg/g; Mingzhang et al., 2011), while the highest endophyte-derived yields are from Alternaria alstroemeriae (426.7 µg/g DW; Mohinudeen et al., 2021). The population-level studies of Uma Shaanker et al. (2008) and Pai et al. (2015) establish that high-yielding elite individuals do exist within wild populations and can be identified through systematic chemical profiling — a finding of direct relevance to domestication and clonal propagation programs.

The pharmacological profile documented by Gayakwad et al. (2024) extends the medicinal relevance of N. nimmoniana far beyond anti-cancer applications. The documented anti-malarial activity is particularly noteworthy given the global health burden of malaria and the growing problem of multi-drug resistance. The antioxidant properties suggest potential applications in managing oxidative stress-related diseases. These multi-target pharmacological properties suggest that N. nimmoniana extracts may have applications in polypharmacological therapeutic strategies.

The mechanistic understanding of CPT action — selective stabilization of the DNA–Topoisomerase I covalent complex leading to replication fork-mediated double-strand breaks — provides a robust molecular rationale for the observed anti-cancer activity. The cell-cycle specificity of CPT (predominantly S-phase active) is consistent with its high potency against rapidly proliferating cancer cells. The cytotoxic activity confirmed in fungal extract experiments (Mohinudeen et al., 2021) against multiple cancer cell lines validates endophyte-derived CPT as pharmacologically equivalent to plant-derived material.



The Amruta Tree: Nature's Cancer-Fighting Treasure

Taken together, these findings support the conclusion that N. nimmoniana is a pharmacological treasure house whose sustainable exploitation requires an integrated strategy combining: (1) tissue-specific, shoot-targeted extraction from wild plants; (2) development of elite clonal orchards based on high-CPT individuals; (3) endophytic fungal fermentation systems for industrial CPT production; and (4) stringent conservation regulations and enforcement.

VI. LIMITATIONS OF REVIEWED STUDIES

Each of the reviewed studies, while scientifically rigorous within its scope, presents certain limitations that should be acknowledged for a complete critical appraisal.

Mingzhang et al. (2011) collected plant specimens from a limited geographic range, which may not fully represent the CPT content variability observed across the broader distributional range of N. nimmoniana in

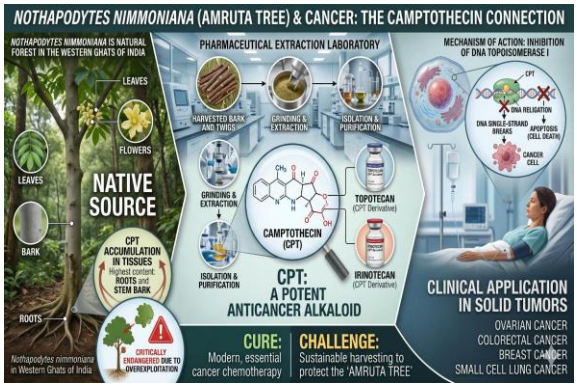
the Western Ghats. The study's findings are therefore most applicable to populations from the specific collection sites, and extrapolation to other regions must be done with caution.

Mohinudeen et al. (2021), while demonstrating breakthrough CPT production levels in endophytic Alternaria species, acknowledge that only suspension culture conditions were studied. Scale-up to industrial bioreactor conditions was not addressed, and the long-term genetic stability of the high-producing strains beyond the subcultures tested remains to be fully established. The mechanisms governing CPT retention in axenic culture also require further elucidation.

The study by Pai et al. (2015) was limited to three localities in the North-Central corridor of the Western Ghats, potentially missing populations in the Southern and Eastern fringes of the species' range. Additionally, the Content Range Chart method, while innovative, has not yet been validated in independent studies.

Gayakwad et al. (2024), being a review article, is dependent on the quality and completeness of primary studies included. The study acknowledges that the precise molecular mechanisms underlying the anti-malarial, anti-helminthic, and anti-inflammatory activities of N. nimmoniana extracts remain inadequately characterized at the receptor/enzyme level.

Uma Shaanker et al. (2008) relied on ecological niche modelling predictions for identifying CPT chemical hotspots. While innovative, the predictions require empirical field validation across the predicted hotspot zones. Additionally, the study was conducted prior to the widespread availability of high-resolution genomic data on N. nimmoniana, which could have provided additional molecular insights.



Nothapodytes nimmoniana (Amruta Tree) & Cancer: The Camptothecin Connection

VII. FUTURE SCOPE AND RESEARCH DIRECTIONS

The reviewed studies collectively identify several significant research gaps and future directions that merit prioritized investigation.

The most immediate and impactful future priority is the scale-up of endophytic CPT production to industrial bioreactor conditions. While Mohinudeen et al. (2021) demonstrated exceptional laboratory-scale yields from Alternaria spp., the transition to large-scale fermentation requires optimization of growth media, aeration, pH control, and downstream processing. Additionally, metabolic engineering of these endophytic strains — through genetic approaches targeting rate-limiting enzymes in the CPT biosynthetic pathway — could further enhance titers.

The recent elucidation of the N. nimmoniana chromosome-level allotetraploid genome (Wang et al., 2025) opens transformative possibilities for CRISPR-Cas9 based gene editing of key CPT biosynthetic enzymes, including strictosidine synthase and strictosamide epoxidase. Overexpression of rate-limiting biosynthetic genes in plant cell cultures or hairy root systems could significantly enhance CPT yield.

Nanoparticle-based encapsulation of CPT and its derivatives represents another promising frontier. CPT's poor water solubility and pH-dependent lactone ring instability have historically limited its clinical application. Nanoformulations — including liposomal systems, polymeric nanoparticles, and cyclodextrin inclusion complexes — could improve CPT bioavailability, tumor targeting, and therapeutic index.

From a conservation biology perspective, establishment of ex situ conservation banks of elite N. nimmoniana genotypes, combined with community-based in situ conservation programs and regulatory enforcement against illegal harvesting, represents an urgent societal need. Ethnobotanical documentation of traditional uses of N. nimmoniana in Ayurvedic and folk medicine systems of the Western Ghats region also warrants systematic study, as it may reveal additional pharmacological applications not yet captured in the scientific literature.

VIII. CONCLUSION

Nothapodytes nimmoniana stands as one of the most pharmacologically significant medicinal plants of the Indian subcontinent. This review, grounded in five rigorously selected peer-reviewed research publications, confirms that N. nimmoniana is the richest known plant source of camptothecin — a monoterpene indole alkaloid that selectively inhibits DNA topoisomerase I, producing lethal double-strand DNA breaks in rapidly proliferating cancer cells.

The studies reviewed herein collectively demonstrate that CPT distribution within the plant is tissue-specific, with roots exhibiting the highest concentrations but shoots offering the most sustainable harvest tissue. Population-level variability in CPT content is significant and geographically correlated, enabling the identification of elite high-yielding individuals suitable for clonal propagation. Endophytic fungi — particularly Alternaria alstroemeriae and Alternaria burnsii — have emerged as viable, sustainable, and plant-independent production platforms for CPT, with demonstrated cytotoxic activity against multiple cancer cell lines.

Beyond its anticancer properties, N. nimmoniana exhibits a broad pharmacological profile encompassing anti-microbial, antioxidant, anti-malarial, anti-helminthic, and anti-inflammatory activities, underscoring its multi-therapeutic potential. However, the species is under severe ecological stress due to overexploitation, with documented population declines of at least 20% across the Western Ghats.

An integrated strategy combining tissue-specific sustainable extraction, elite individual-based clonal orcharding, endophytic biotechnology, genomic-assisted metabolic engineering, and stringent conservation policy represents the path forward for ensuring that this botanical treasure continues to serve human health without being driven to extinction.

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