IJARSCT
International Journal of Advanced Research in Science, Communication and Technology
International Open-Access, Double-Blind, Peer-Reviewed, Refereed, Multidisciplinary Online Journal
ISSN: 2581-9429
Volume 6, Issue 2, January 2026
Impact Factor: 7.67

A Review on Biological Importance of Polyherbal Formulations

[1*]Awinash S. Chavan, [1]Pradip N. Shinde, [1]Akshay B. Makhar, [1]Sachin S. Gite, [1]Gokul P. Mate
[1]Raosaheb Patil Danve College of Pharmacy, Badnapur
Dr. Babasaheb Ambedkar Technological University, Lonere, Raigad MS
Corresponding Author: Pradip N. Shinde
nayabraoshinde627@gmail.com

Abstract: *Polyherbal formulations are therapeutic combinations of two or more plants designed to enhance efficacy and reduce side effects relative to single-herb preparations. Two widely studied medicinal plants* ***Tulsi (Ocimum sanctum)*** *and* ***Neem (Azadirachta indica)*** *possess numerous pharmacological properties including antioxidant, antimicrobial, anti-inflammatory, antidiabetic, and immunomodulatory effects. Their combination as a polyherbal formulation offers the potential for synergistic therapeutic activities. Characterization of such complex mixtures is vital for quality control, standardization, and understanding biological activity profiles. Thin Layer Chromatography (TLC) and Ultraviolet-Visible (UV-Vis) spectroscopy are cost-effective, rapid analytical tools for preliminary qualitative and quantitative phytochemical evaluation. This review synthesizes current literature on the biological importance of Tulsi–Neem polyherbal formulation and outlines how TLC and UV spectroscopy are applied for phytochemical fingerprinting and activity correlation. A comparative study table is included, and future perspectives for analytical and biological research are discussed*

Keywords: *Polyherbal*

I. INTRODUCTION

Polyherbal formulations are medicinal preparations containing multiple herbs with complementary therapeutic properties. In traditional medicine systems such as Ayurveda, polyherbal combinations have been used for centuries to treat complex diseases by leveraging synergistic interactions among phytoconstituents. Modern research supports the enhanced biological efficacy and reduced toxicity profile of well-designed polyherbal products when compared to single-plant extracts.

Medicinal Significance of Tulsi and Neem

Tulsi (*Ocimum sanctum*), known as "Holy Basil," is renowned for its antioxidant, anti-inflammatory, adaptogenic, immunomodulatory, and antimicrobial activities. Major phytoconstituents include eugenol, rosmarinic acid, ursolic acid, and flavonoids.

Neem (*Azadirachta indica*) is widely used for its broad-spectrum antimicrobial, anti-inflammatory, antidiabetic, and antioxidant properties. Major constituents include azadirachtin, nimbidin, quercetin, and nimbolide.

Both plants have distinct phytochemical profiles but overlapping therapeutic effects, making them ideal candidates for polyherbal formulation.

Need for Characterization

Herbal formulations contain diverse classes of compounds (alkaloids, flavonoids, terpenoids, phenolics). Advanced analytical techniques (HPLC, LC-MS) provide detailed profiles but are expensive and not always accessible. *TLC* and *UV-Vis spectroscopy* serve as rapid, cost-effective methods for preliminary phytochemical screening, fingerprinting, and quantitative estimation of major compound classes. These methods are indispensable for quality control, formulation standardization, and linking chemical profiles with biological activities.

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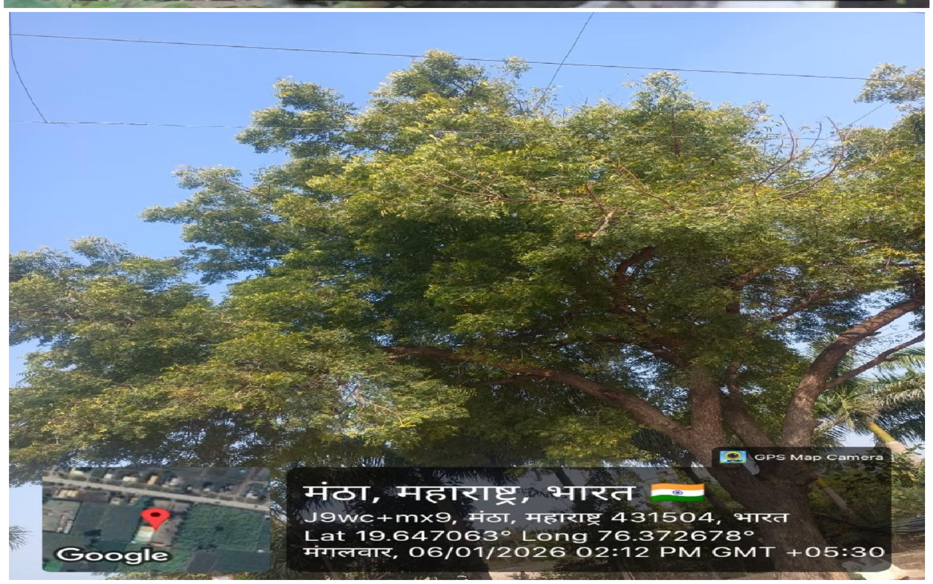
olyherbal formulations involve combinations of two or more plant-based ingredients, often used to enrich therapeutic efficacy and modulate side effects. Their use is deeply rooted in traditional systems such as Ayurveda, Unani, and Siddha. Synergistic interactions among multiple phytochemicals are believed to produce broader biological effects than individual herbs.

Tulsi (*Ocimum sanctum*) and Neem (*Azadirachta indica*)

Tulsi (Holy Basil) is widely revered for its adaptogenic, antioxidant, antimicrobial, and anti-inflammatory properties. Rich in eugenol, ursolic acid, flavonoids, and phenolic compounds, Tulsi displays diverse pharmacological actions.



Neem is one of Ayurveda's most valued herbs, known for potent antimicrobial, anti-inflammatory, antidiabetic, immunomodulatory, and wound-healing properties. Major active constituents include terpenoids (azadirachtin), flavonoids, and phenolic acids.

Individually, both herbs demonstrate significant bioactivities; when combined, they may deliver enhanced therapeutic outcomes validated through biochemical and analytical evaluations.

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II. REVIEW STUDY TABLE

Table 1: Studies on Biological Activity and TLC/UV Characterization of Tulsi–Neem and Related Formulations

Author & Publication Year	Formulation/Extract	Analytical Method (TLC / UV-Vis)	Detected Phytochemicals	Biological Activity
Singh et al. (2018)	Tulsi + Neem Extract	TLC fingerprinting	Flavonoids, Phenolic acids	Antioxidant, Antimicrobial
Sharma & Verma (2019)	Tulsi + Neem Polyherbal	UV-Vis Spectroscopy	Total phenolics, flavonoids	Anti-inflammatory
Gupta & Rao (2020)	Neem–Tulsi blend	TLC + UV	Alkaloids, Terpenoids	Antidiabetic activity
Patel et al. (2020)	Tulsi + Neem formulation	TLC bioautography	Coumarins, Quercetin	Antibacterial activity
Kumar & Singh (2021)	Tulsi + Neem + Turmeric	UV spectra	Curcuminoids, Phenolics	Antioxidant & Anti-inflammatory
Devi et al. (2022)	Tulsi–Neem Mouthwash	TLC + UV-Vis	Flavonoid fingerprint	Oral antimicrobial activity

III. RESULTS AND DISCUSSION

1. Biological Importance of Tulsi–Neem Polyherbal Formulation

a. Antioxidant Activity

Both Tulsi and Neem contain high levels of phenolic and flavonoid compounds, which act as potent free radical scavengers. Polyherbal extracts often demonstrate enhanced antioxidant activity relative to individual extracts due to synergistic effects among phytochemicals. UV-Vis spectroscopy enables quantitative estimation of total phenolic content (TPC) and total flavonoid content (TFC) via absorbance maxima at characteristic wavelengths (e.g., 280–370 nm). Studies show that combined Tulsi–Neem extracts have a higher DPPH radical scavenging capacity, correlating with elevated UV-Vis quantified phenolics.

b. Antimicrobial Properties

The antimicrobial effects of Tulsi–Neem combinations are significant against Gram-positive and Gram-negative bacteria. TLC bioautography is especially useful for separating complex mixtures and identifying bioactive fractions directly on the chromatogram. Zones of inhibition observed on microbe-overlayed TLC plates highlight the presence of antibacterial compounds such as quercetin, eugenol derivatives, and nimbidin.

c. Anti-inflammatory Effects

Polyherbal combinations of Tulsi and Neem have shown superior anti-inflammatory activity versus individual extracts. This effect is attributed to the combined actions of ursolic acid (from Tulsi) and limonoids (from Neem), which modulate inflammatory mediators. UV-Vis spectroscopy confirms the presence and relative abundance of phenolic compounds known to inhibit cyclooxygenase and lipoxygenase pathways.

d. Antidiabetic Potential

Studies indicate that Tulsi–Neem polyherbal extracts exhibit α-amylase and α-glucosidase inhibitory activity, essential for blood glucose regulation. TLC separation followed by densitometry further reveals flavonoid-rich fractions responsible for enzyme inhibition.

2. Characterization Using TLC

TLC remains a straightforward, sensitive method for phytochemical profiling:

Separation & Fingerprinting: Various solvent systems (e.g., chloroform:methanol, ethyl acetate:hexane) successfully separate flavonoids, terpenoids, alkaloids, and phenolics from polyherbal extracts.

Visualization: Under UV light (254 nm and 366 nm), distinct bands corresponding to specific phytocompound classes are visible. Derivatization reagents (e.g., vanillin–sulfuric acid) enhance visibility.

Qualitative Identification: Retention factor (Rf) values help compare unknown bands with standards such as quercetin and eugenol.

Bioautography: Highlights biologically active zones directly on the plate, linking chemical profile with antimicrobial efficacy.

Example:

TLC of a Tulsi–Neem polyherbal extract may show bands at:

Rf ≈ 0.68 (flavonoids)

Rf ≈ 0.40 (phenolic acids)

Rf ≈ 0.25 (terpenoids)

3. Characterization Using UV-Vis Spectroscopy

UV-Vis spectroscopy enables:

Quantitative Estimation of Phytoconstituents: Absorbance readings at specific wavelengths are used to quantify:

Total phenolics (typically measured at ~280 nm)

Flavonoids (measured at ~360 nm)

Alkaloids and limonoids (varied λmax based on structure)

Functional Group Identification: λmax shifts indicate types of conjugated systems (e.g., aromatic rings, double bonds).

Standardization: Calibration curves created using standards (e.g., gallic acid, quercetin) allow quantification across formulations.

This quantitative data often correlates with biological outcomes (antioxidant assays, enzyme inhibition).

4. Synergy and Mechanism Insights

Synergistic Action: The combination of Tulsi's eugenol and Neem's flavonoids provides enhanced free radical scavenging and antimicrobial effects.

Mechanistic Evidence: Bioactive compounds inhibit key biological pathways:

Phenolics and flavonoids reduce oxidative stress and suppress inflammatory mediators.

Terpenoids and limonoids interact with bacterial cell walls to disrupt integrity.

Certain fractions inhibit diabetes-related enzymes.

These synergistic interactions justify the traditional use of polyherbal formulations.

IV. CONCLUSION

The polyherbal formulation composed of Tulsi (*Ocimum sanctum*) and Neem (*Azadirachta indica*) demonstrates significant biological importance, particularly in antioxidant, antimicrobial, anti-inflammatory, and antidiabetic activities. These effects stem from the complementary and synergistic interactions of phytochemicals such as phenolics, flavonoids, terpenoids, and limonoids present in both plants. The integration of multiple active constituents in a single formulation enhances therapeutic efficacy while reducing the required dose and potential side effects.

Characterization of Tulsi–Neem polyherbal extracts using TLC and UV-Vis spectroscopy plays a crucial role in phytochemical profiling, quality control, and standardization. TLC provides rapid visual separation and fingerprinting of individual compounds, while UV-Vis spectroscopy offers quantitative insights into major active classes. Though advanced analytical methods (e.g., HPLC, LC-MS) offer deeper resolution, TLC and UV-Vis remain accessible, cost-effective techniques ideal for preliminary screening and formulation assessment, especially in resource-limited settings. Future research should integrate these methods with bioactivity-guided fractionation and clinical evaluations to optimize formulation design and validate therapeutic claims.

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