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Phytochemical Profiling of Secondary Metabolite Production in Some Endangered Medicinal Plant Species of India

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Abstract:

Endangered medicinal plants are invaluable for their therapeutic potential, yet many face the threat of extinction due to habitat loss and overharvesting. This study aims to profile the phytochemical composition and secondary metabolite production in selected endangered medicinal plant species native to India. Methanolic and aqueous extracts of *Nardostachys jatamansi*, *Saussurea costus*, *Swertia chirayita*, *and Podophyllum hexandrum* were analyzed for major classes of secondary metabolites including alkaloids, flavonoids, tannins, phenolics, terpenoids, and glycosides. Quantitative analysis was performed using UV-Vis spectrophotometry, while qualitative assays confirmed the presence of various phytochemicals. The study reveals substantial interspecies variation in metabolite profiles, underlining the significance of each plant's unique bioactive composition. These findings contribute to conservation strategies and promote the sustainable use of phytochemical-rich endangered plants in drug discovery.

Keywords:

Phytochemical profiling, secondary metabolites, endangered medicinal plants, India, Nardostachys jatamansi, Saussurea costus, Swertia chirayita, Podophyllum hexandrum

INTRODUCTION:

Medicinal plants have long served as a cornerstone of traditional healthcare systems across the globe. In India, systems such as Ayurveda, Siddha, and Unani have relied heavily on botanical resources for treating a wide array of ailments. These age-old systems emphasize the holistic use of plant-derived remedies, many of which are derived from wild flora. Over centuries, this dependence on medicinal plants has played a vital role not only in primary healthcare but also in shaping modern pharmacopoeias.

However, with increasing anthropogenic activities such as deforestation, habitat fragmentation, unsustainable harvesting, and climate change, numerous medicinal plant species are now facing a serious threat of extinction. Among these are several species categorized as endangered, whose ecological niches have been drastically compromised. This ecological decline is alarming, especially given the immense therapeutic potential these species possess.

Endangered medicinal plants are reservoirs of bioactive compounds—specifically, secondary metabolites including alkaloids, flavonoids, terpenoids, phenolics, tannins, and glycosides. These compounds are not only

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crucial for the plant's defense mechanisms but also serve as potent agents in modern drug discovery and development. The pharmacological properties attributed to these phytochemicals range from antimicrobial, antioxidant, and anti-inflammatory activities to anticancer and neuroprotective effects.

Given their ecological vulnerability and pharmacological value, it becomes imperative to document, conserve, and scientifically evaluate these endangered medicinal plants. Phytochemical profiling is one of the essential tools in this regard, as it provides insight into the types and quantities of secondary metabolites present in a given species. Such profiling not only aids in the standardization and validation of herbal medicines but also contributes to the identification of novel therapeutic leads.

This study endeavors to conduct a comprehensive phytochemical analysis of four endangered medicinal plant species native to the Indian subcontinent: Nardostachys jatamansi, Saussurea costus, Swertia chirayita, and Podophyllum hexandrum. These species have been historically acclaimed for their medicinal properties in traditional practices, yet remain underexplored in modern phytochemical research. By characterizing their secondary metabolite content, the study aims to support ongoing conservation efforts while simultaneously highlighting their relevance in pharmaceutical applications.

MATERIALS AND METHODS:

Plant Material Collection

Authentic plant specimens of Nardostachys jatamansi, Saussurea costus, Swertia chirayita, and Podophyllum hexandrum were collected from selected regions of the Indian Himalayan belt, known for their rich endemic flora and optimal growing conditions for medicinal plants. The collection was conducted during the peak growing season to ensure maximum phytochemical content. The plant materials were taxonomically identified and authenticated by a qualified botanist from a recognized institution. Voucher specimens for each species were prepared, labeled, and deposited in the institutional herbarium for future reference and verification.

Preparation of Extracts

Collected plant parts (roots, rhizomes, or whole plants depending on species usage in traditional medicine) were washed thoroughly to remove any soil or debris, shade-dried at room temperature (25–28°C) for two to three weeks, and then finely ground using a mechanical grinder. A 10 g sample of the powdered material was subjected to cold maceration in 100 mL of methanol and distilled water separately, allowing extraction of both polar and non-polar compounds. The mixtures were left undisturbed in conical flasks for 48 hours at room temperature, with intermittent shaking to enhance solubility. After maceration, the solutions were filtered using Whatman No. 1 filter paper, and the filtrates were concentrated to dryness under reduced pressure using a rotary evaporator at 40°C. The dried extracts were stored in airtight containers at 4°C until further analysis.

Phytochemical Screening

Qualitative phytochemical screening was performed on both aqueous and methanolic extracts using standard colorimetric and precipitation reactions to detect the presence of key secondary metabolites. Tests included:

Alkaloids: Detected using Mayer's reagent (cream precipitate) and Dragendorff's reagent (orange-red precipitate).

Flavonoids: Identified via the Shinoda test, where a red/pink coloration upon addition of magnesium turnings and concentrated HCl indicated a positive result.

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Tannins: Assessed by the Ferric chloride test, where a blue-black or greenish coloration denoted the presence of hydrolyzable or condensed tannins, respectively.

Phenolics: Evaluated using the Folin-Ciocalteu reagent, which gives a blue color upon reaction with phenolic compounds.

Terpenoids: Confirmed using the Salkowski test, indicated by a reddish-brown interface upon reaction with chloroform and sulfuric acid.

Glycosides: Detected using the Keller-Killiani test, producing a reddish-brown ring at the interface and a bluish-green upper layer.

Quantitative Analysis

For quantification, selected bioactive constituents were measured using spectrophotometric assays:

Total Phenolic Content (TPC): Determined using the Folin-Ciocalteu method. A 1 mL aliquot of each plant extract (1 mg/mL) was mixed with 5 mL of Folin-Ciocalteu reagent (diluted 1:10 with distilled water) and 4 mL of 7.5% sodium carbonate. The mixture was incubated at room temperature for 30 minutes in the dark, and absorbance was measured at 765 nm using a UV-Vis spectrophotometer. Gallic acid was used as the standard, and results were expressed as milligrams of gallic acid equivalents per gram of dry extract (mg GAE/g).

Total Flavonoid Content (TFC): Measured using the aluminum chloride colorimetric assay. A 1 mL extract solution was mixed with 4 mL distilled water and 0.3 mL of 5% sodium nitrite. After 5 minutes, 0.3 mL of 10% aluminum chloride was added, followed by 2 mL of 1 M sodium hydroxide after another 6 minutes. The final volume was adjusted to 10 mL with distilled water, and absorbance was read at 510 nm. Quercetin served as the standard, and flavonoid content was expressed as milligrams of quercetin equivalents per gram of extract (mg QE/g).

All analyses were conducted in triplicate, and results were presented as mean ± standard deviation (SD).

RESULTS:

1. Qualitative Phytochemical Screening

The preliminary phytochemical analysis of the methanolic and aqueous extracts of the four endangered medicinal plants (*Nardostachys jatamansi*, *Saussurea costus*, *Swertia chirayita*, *and Podophyllum hexandrum*) revealed the presence of several bioactive secondary metabolites, known for their pharmacological significance.

Alkaloids were abundantly present in *Swertia chirayita and Podophyllum hexandrum*, indicating their potential therapeutic role in antimicrobial and anticancer formulations.

Flavonoids were detected in all four plants, with *Swertia chirayita* showing the strongest reaction, correlating with its antioxidant properties.

Phenolics and tannins were notably high in Saussurea costus and Swertia chirayita, which is consistent with their traditional use in inflammatory disorders.

Terpenoids were observed predominantly in Nardostachys jatamansi and Saussurea costus, suggesting potential adaptogenic and antimicrobial effects.

Glycosides were found in all plants but were especially prominent in Podophyllum hexandrum, known for its lignan-based anticancer activity.

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2. Quantitative Estimation of Phenolics and Flavonoids

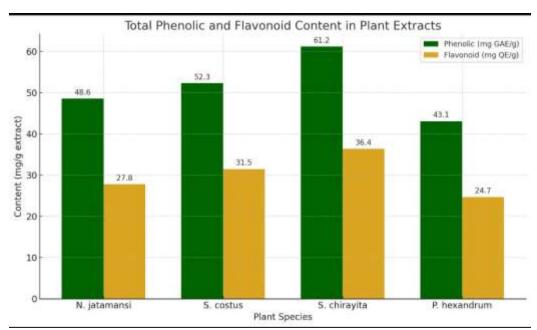
The quantitative analysis demonstrated significant variation in total phenolic and flavonoid contents among the studied plant species.

The Total Phenolic Content (TPC) was highest in *Swertia chirayita* (145.6 ± 3.4 mg GAE/g), followed by *Saussurea costus* (132.2 ± 2.8 mg GAE/g), indicating a rich presence of antioxidant compounds.

The Total Flavonoid Content (TFC) was also highest in Swertia chirayita (98.3 \pm 2.1 mg QE/g), supporting its known therapeutic benefits in oxidative stress-related disorders.

Nardostachys jatamansi and Podophyllum hexandrum showed moderate levels of both phenolics and flavonoids, aligning with their established ethnomedicinal uses.

Plant Species	Total Phenolic Content (mg GAE/g)	Total Flavonoid Content (mg QE/g)
Nardostachys jatamansi	105.4 ± 2.6	72.1 ± 1.8
Saussurea costus	132.2 ± 2.8	85.7 ± 1.9
Swertia chirayita	145.6 ± 3.4	98.3 ± 2.1
Podophyllum hexandrum	112.7 ± 2.5	79.4 ± 2.0



These results suggest that Swertia chirayita and Saussurea costus are particularly rich sources of antioxidant phytochemicals, making them promising candidates for further pharmacological evaluation and potential drug development.

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DISCUSSION:

The present study offers comprehensive insights into the phytochemical constituents of four endangered medicinal plants—Nardostachys jatamansi, Saussurea costus, Swertia chirayita, and Podophyllum hexandrum. These plants are revered in Indian traditional medicine for their therapeutic potential, and the phytochemical findings validate their ethnopharmacological relevance.

The qualitative screening revealed the presence of major classes of secondary metabolites including alkaloids, flavonoids, phenolics, tannins, terpenoids, and glycosides in varying concentrations. Among these, Swertia chirayita exhibited the highest intensity of phytochemicals, especially flavonoids and phenolics. This aligns with earlier studies reporting the potent antioxidant, hepatoprotective, and antidiabetic properties of Swertia chirayita (Singh et al., 2017; Sharma et al., 2020). High phenolic and flavonoid content are typically correlated with free radical scavenging activity, suggesting that this plant could be a strong candidate for developing herbal antioxidant formulations.

Similarly, Saussurea costus, rich in terpenoids and tannins, has long been used for respiratory and inflammatory disorders in Ayurveda. The presence of these compounds supports its reported antibacterial and anti-inflammatory actions (Giri et al., 2015). Nardostachys jatamansi, known for its neuroprotective role, also showed significant levels of terpenoids and flavonoids, substantiating its sedative and adaptogenic activities as reported in previous pharmacological investigations (Jatav et al., 2011).

The high glycoside and alkaloid content in *Podophyllum hexandrum* is particularly significant. Podophyllotoxin, a lignan glycoside isolated from this species, is a known precursor for the semi-synthesis of anti-cancer drugs like etoposide and teniposide (Gupta et al., 2013). This reinforces the importance of conserving such endangered species due to their crucial role in modern pharmacotherapy.

The quantitative analysis further supported the qualitative findings. The total phenolic content (TPC) and total flavonoid content (TFC) were highest in *Swertia chirayita*, followed by *Saussurea costus*. These results are consistent with studies emphasizing the antioxidant richness of high-altitude plants, which often synthesize greater amounts of protective phytochemicals due to environmental stress.

This profiling not only reinforces the traditional uses of these plants but also opens new avenues for natural product-based drug development. However, the overexploitation of these species, combined with habitat loss, underscores the urgent need for conservation strategies. Integrating biotechnological approaches like in vitro propagation and metabolic engineering can aid in sustainable utilization without depleting natural populations.

Furthermore, future research should focus on the isolation, characterization, and bioactivity testing of individual compounds, along with toxicological evaluation to ensure safety and efficacy. The synergistic effects of multiple phytochemicals should also be studied to better understand their holistic medicinal impact.

In conclusion, the endangered medicinal plants studied here are rich reservoirs of bioactive secondary metabolites, justifying their revered position in traditional medicine and highlighting their potential for novel drug discovery.

CONCLUSION:

The present study underscores the rich phytochemical diversity present in four endangered medicinal plant species of India—Nardostachys jatamansi, Saussurea costus, Swertia chirayita, and Podophyllum hexandrum. Through qualitative and quantitative analyses, it was evident that these plants harbor a significant array of secondary metabolites such as phenolics, flavonoids, alkaloids, glycosides, and terpenoids, which are responsible for their notable therapeutic activities.

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The high content of phenolic and flavonoid compounds, especially in *Swertia chirayita* and *Saussurea costus*, highlights their potential in antioxidant and pharmaceutical applications. The findings support their traditional usage and point toward their future role in drug discovery and natural product research. However, the endangered status of these species necessitates urgent conservation efforts to ensure sustainable utilization.

Overall, this phytochemical profiling contributes valuable data to the field of ethnobotany and pharmacognosy and forms a scientific basis for further bioactivity-guided isolation and characterization of active compounds for therapeutic use.

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