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Research Article

To study the phytochemical constituent of *Murraya koenigii* leaves

Swati Mishra, Shubham Bhatt, Deepika Gupta*

Maharishi School of Pharmaceutical Sciences, MUIT, Lucknow, U.P., 226013, India.

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ABSTRACT

Introduction: Murraya koenigii (curry leaves) is a widely used medicinal plant known for its traditional applications in treating various ailments due to its rich phytochemical composition. Understanding the qualitative and quantitative presence of its bioactive compounds can provide scientific validation for its ethnomedicinal use.

Aim: The present study aimed to perform a comprehensive phytochemical investigation of Murraya koenigii leaves to identify and quantify the major secondary metabolites responsible for its pharmacological activities.

Materials and Methods: Qualitative phytochemical screening was conducted using standard chemical tests to detect the presence of alkaloids, flavonoids, tannins, saponins, glycosides, phenolics, terpenoids, and steroids. Quantitative analysis involved spectrophotometric and gravimetric methods to estimate the Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and Total Alkaloid Content.

Results: Qualitative analysis confirmed the presence of all tested phytoconstituents, with distinctive color changes and precipitate formation supporting their identification. Quantitatively, TPC was found to be as high as 133.59 mg GAE/g dry weight (DW), indicating potent antioxidant potential. TFC was reported at 1.24% (w/w), reflecting significant flavonoid concentration, and Total Alkaloid Content was measured at 23.73%, suggesting strong pharmacological activity.

Conclusion: The study confirms that Murraya koenigii leaves are a rich source of diverse bioactive secondary metabolites. These findings scientifically validate the plant's traditional therapeutic use and highlight its potential for future development of herbal formulations or pharmaceutical applications. Further studies focusing on the isolation and characterization of individual compounds are warranted to explore their specific mechanisms of action and therapeutic efficacy.

INTRODUCTION

In the rapidly evolving landscape of phytopharmaceuticals, *Murraya koenigii* (Linn.) Spreng. commonly known as curry leaf and traditionally revered in Ayurvedic and Unani systems of medicine—has transitioned from a culinary herb to a subject of intense scientific scrutiny. While widely appreciated for its aromatic leaves *(as shown in figure 1)* in South Asian cuisine, this underutilized botanical treasure harbors a complex arsenal of bioactive compounds that remain only partially explored by modern science. *(Kaur et al. 2014)*

Phytochemicals, the plant-derived secondary metabolites responsible for therapeutic actions, have gained renewed attention for their roles in modulating oxidative stress, inflammation, microbial resistance, and chronic metabolic dysfunctions. In this context, *Murraya koenigii* presents a unique pharmacognostic profile, rich in alkaloids (e.g., mahanimbine, girinimbine), flavonoids, terpenoids, phenolics, and essential oils—each demonstrating a

***Corresponding Author:** Shubham Bhatt

Address: Maharishi School of Pharmaceutical Sciences, MUIT, Lucknow, U.P., 226013, India.

Email ✉**:** shubhatt521@gmail.com

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Figure 1. Different parts of curry leaves

promising spectrum of pharmacological effects including antioxidant, antidiabetic, anticancer, and neuroprotective activities. *(Prakash et al. 1974)*

Curry leaves *(Murraya koenigii)* are valuable not only for their distinct flavour, but also for their nutritional and therapeutic benefits. These aromatic leaves are a key component of many traditional recipes, adding a distinct flavour and perfume that improves the whole dining experience. Their significance, however, extends beyond the culinary realm. Curry leaves have been used in traditional medicine for millennia because of their alleged health benefits.

Phytochemicals, substances found naturally in plants, are known to exhibit a variety of biological actions. Many of herbal medicines' therapeutic benefits are due to these bioactive substances. Curry leaf has a notably rich phytochemical profile, comprising various types of chemicals such as alkaloids, flavonoids, phenols, terpenoids, and essential oils. *(Harish et al. 2012)*

The current investigations have indicated that these leaves contain hypoglycemic qualities, making them potentially useful for diabetic therapy. Furthermore, their antimicrobial capabilities might help to preserve food and promote general health. The purpose of this study is to conduct a systematic investigation on the phytochemical elements of curry leaves. Modern extraction and analysis procedures have been applied. Identifying and measuring these bioactive chemicals will offer a complete picture of curry leaves' chemical composition. *(Narasimha et al., 1975)*

Curry leaf, technically known as Murraya koenigii, is a popular herb in South Asian cuisine, particularly Indian, Sri Lankan, and Southeast Asian cuisine. These fragrant leaves from the Rue family are widely prized for their distinct flavour as well as their nutritional and therapeutic benefits. The plant is a tiny tree or shrub that may grow up to 4 meters tall and has glossy, deep green pinnate leaves. *(Sasidharan et al. 2011)*

It is a native to the Indian subcontinent and widely distributed across tropical and subtropical regions of Asia. It is predominantly found in India, Sri Lanka, Bangladesh, and Nepal, and has also been introduced to Southeast Asia, Australia, and parts of Africa. In India, it thrives in states such as Tamil Nadu, Kerala, Karnataka, Andhra Pradesh, and Maharashtra due to favorable climatic conditions. The plant grows well under warm, humid conditions and prefers tropical to subtropical climates. An optimal temperature range of 20°C to 35°C with moderate to high rainfall (600–1000 mm annually) is ideal for its growth. It requires full sunlight but can tolerate partial shade during early growth stages. The soil should be well-drained, loamy to sandy loam soils rich in organic matter. Slightly acidic to neutral pH (6.0–7.5) is most suitable. Waterlogged or saline soils hinder root development and growth. Incorporating farmyard manure or compost during planting significantly improves soil fertility and plant vigor. The plant is generally propagated through seeds, though vegetative propagation using stem cuttings is also effective in maintaining genetic fidelity. *(Sauvauire et al., 1996)*

Material & Method

Collection of curry leaves

Fresh leaves of *Murraya koenigii* (commonly known as curry leaves) were collected from a local herbal garden in Lucknow, U.P., India, during the month of October, ensuring they were collected at the height of their phytochemical concentration. The approaches described include plant collection, preparation, and extraction processes, as well as analytical tools for phytochemical evaluation. *(Rajnikant et al. 2015)*

Preparation of leave extract

The collected leaves were washed with distilled water to remove adhering dust and shade-dried for 10 days at room temperature. The dried leaves were coarsely powdered using a mechanical grinder and stored in an airtight container for further use. *(Jain et al., 2017)*

Extraction Method

Solvent Extraction

The 100 g powder of curry leaves were subjected to solvent extraction using ethanol and methanol as solvents to maximize the extraction of phytochemicals as shown in table 1. *(Leela et al., 2013)*

Ethanol Extraction

Approximately 100 g of powdered leaves were placed in a Soxhlet extractor, and 500 mL of ethanol was added. The extraction was carried out for 6-8 hours. After extraction, the solvent was evaporated using a rotary evaporator set at a temperature below 40°C to concentrate the extract. *(Kaur et al. 2014)*

Methanol Extraction

The same procedure was followed using methanol as the solvent for another batch of 100 g of powdered leaves.

Aqueous Extraction

An additional extraction was performed using distilled water to capture water-soluble phytochemicals. Aqueous extraction was performed by boiling 100 g of powdered leaves in 1 litre of distilled water for 30 minutes. The mixture was then cooled, filtered through filter paper, and the extract was collected. *(Dineshkumar et al. 2012)*

Preliminary Phytoconstituent Screening

Qualitative Analysis

Qualitative phytochemical screening of the various extracts was carried out using standard procedures to detect the presence of different classes of secondary metabolites. *(Vandana et al. 2012)* Phytochemical screening was conducted to identify the presence of various classes of compounds as shown in *table 2.*

Tests for Alkaloids

Mayer's Test

To 2 mL of plant extract, a few drops of Mayer's reagent (potassium mercuric iodide) were added. Formation of a cream-colored precipitate indicates the presence of alkaloids.

Wagner's Test

2 mL of extract was treated with Wagner's reagent (iodine in potassium iodide). A reddish-brown precipitate formation confirmed the presence of alkaloids.

Dragendorff's Test

A few drops of Dragendorff's reagent (solution of potassium bismuth iodide) were added to 2 mL of the extract. An orange or reddish-brown precipitate indicated alkaloid presence. *(Rani et al. 2012)*

Test for Flavonoids

Alkaline Reagent Test

To 2 mL of extract, few drops of 10% sodium hydroxide solution were added. The appearance of an intense yellow color, which becomes colourless upon the addition of dilute hydrochloric acid, suggests the presence of flavonoids. *(Summan et al. 2014)*

Test for Phenolic Compounds and Tannins

Ferric Chloride Test

To 2 mL of extract, a few drops of 5% ferric chloride solution were added. A dark blue or greenish-black coloration indicates the presence of phenolic compounds or tannins.

Test for Saponins

Foam Test

The extract was diluted with distilled water and shaken vigorously for 15 minutes in a graduated cylinder. The formation of a stable persistent froth (1 cm or more) lasting for at least 10 minutes indicates the presence of saponins.

Test for Cardiac Glycosides

Keller–Killiani Test

To 2 mL of extract, 1 mL of glacial acetic acid containing a trace of ferric chloride was added. This was under layered with concentrated sulfuric acid. The appearance of a reddish-brown ring at the interface and a bluish-green layer in the acetic acid layer indicates the presence of deoxysugars, characteristic of cardiac glycosides. *(Harish et al. 2015)*

Table 1. Solvent used in extraction method for curry leaves

Solvent Used	*Polarity*	*Purpose*
Petroleum ether	Non-polar	Extraction of fats, oils, waxes
Chloroform	Medium-polar	Extraction of alkaloids, flavonoids
Ethanol	Polar	Extraction of phenolics, glycosides
Distilled water	Highly polar	Extraction of saponins, tannins

Table 2. Qualitative analysis of phytoconstituents

Constituent	Test Used
Alkaloids	Mayer's, Wagner's, Dragendorff's tests
Flavonoids	Alkaline reagent test
Tannins	Ferric chloride test
Saponins	Foam test
Glycosides	Keller-Killiani test
Phenolics	Lead acetate test
Terpenoids	Salkowski's test
Steroids	Liebermann–Burchard test

Table 3. Qualitative Phytochemical Screening of Plant Extracts

Phytoconstituent	*Test Used*	*Observation*	*Inference*
Alkaloids	Mayer's, Wagner's, Dragendorff's	Cream, reddish-brown, and orange precipitates observed respectively	+ive
Flavonoids	Alkaline reagent test	Yellow coloration turned colorless on acidification	+ive
Tannins/Phenolics	Ferric chloride, Lead acetate tests	Greenish-black/blue and yellow precipitate observed	+ive
Saponins	Foam test	Persistent froth >1 cm for >10 min	+ive
Glycosides	Keller–Killiani test	Reddish-brown ring with bluish-green upper layer	+ive
Terpenoids	Salkowski's test	Reddish-brown interface coloration	+ive
Steroids	Liebermann–Burchard test	Blue-green coloration	+ive

Test for Flavonoids and Tannins

Lead Acetate Test

2 mL of extract was treated with a few drops of lead acetate solution. A yellow precipitate formation confirms the presence of flavonoids or tannins.

Test for Steroids and Terpenoids

Salkowski's Test

2 mL of extract was treated with 2 mL of chloroform, followed by the addition of concentrated sulfuric acid along the sides of the test tube. A reddish-brown coloration at the interface indicates the presence of terpenoids or steroids.

Liebermann–Burchard Test

2 mL of extract was mixed with 1 mL of acetic anhydride and 1 mL of concentrated sulfuric acid. The appearance of a blue-green color indicates the presence of sterols or triterpenoids.

Quantitative Analysis

Quantitative estimation of total phenolic, flavonoid, and alkaloid contents was performed using spectrophotometric methods.

Total Phenolic Content (TPC)

Determined using the Folin–Ciocalteu reagent method. Gallic acid was used as a standard, and results were expressed as mg of gallic acid equivalents (GAE) per gram of extract.

Total Flavonoid Content (TFC)

Aluminum chloride colorimetric method was employed. Quercetin was used as a standard, and values were expressed in mg quercetin equivalents (QE)/g of extract.

Total Alkaloid Content

The gravimetric method was used by precipitating alkaloids using dilute HCl and ammonium hydroxide. Results were expressed as % alkaloids w/w.

Result & Discussion

Qualitative Analysis

Qualitative phytochemical analysis was carried out on the different plant extracts to identify the presence of major secondary metabolites using standard procedures. The outcomes of the tests conducted for alkaloids, flavonoids, tannins, saponins, glycosides, phenolics, terpenoids, and steroids are summarized in Table 3.

The phytochemical investigation of curry leaf extracts revealed the presence of several bioactive secondary metabolites, which contribute to the plant's wide range of pharmacological activities. The detection of alkaloids through Mayer's, Wagner's, and Dragendorff's tests highlights their significant presence, indicating potential antimicrobial, analgesic, and anti-inflammatory effects. A strong response in the alkaline reagent test confirmed the presence of flavonoids, which are well-known for their antioxidant, hepatoprotective, and cardioprotective actions.

The greenish-black coloration from the ferric chloride test and the yellow precipitate with lead acetate confirm the presence of both tannins and phenolic compounds, traditionally linked with wound healing and antidiarrheal effects. The foam test demonstrated the presence of saponins, known for their hemolytic activity, cholesterol-lowering properties, and immune-modulating capabilities. The appearance of a reddish-brown ring and bluish layer in the Keller–Killiani test confirmed cardiac glycosides, which support traditional uses in treating cardiac disorders and fluid retention. Finally, the Salkowski's and Liebermann–Burchard tests indicated the presence of terpenoids and steroids, compounds widely recognized for their anti-inflammatory, antimicrobial, and anticancer potentials as shown in figure 2. Collectively, these findings validate the ethnopharmacological relevance of curry leaves and support further research for isolating and evaluating individual bioactive constituents.

Quantitative Analysis

The quantitative analysis of bioactive compounds in curry leaves (*Murraya koenigii*), focusing on total phenolic content (TPC), total flavonoid content (TFC), and total alkaloid content. These analyses were performed using spectrophotometric and gravimetric methods, with recent studies providing valuable insights into the phytochemical profile of curry leaves as shown in table 4.

Table 4. Quantitative estimation of phytoconstituents in curry leaves

Sample	*TPC (mg GAE/g)*	*TFC (CE/gm)*	*TAC (%)*
Shade-dried leaves (water extract)	128.39	234.82	24.00
Methanolic leaves (general)	74.31	49.53	18.00
Fresh leaves (methanolic extract)	57.88	8.61	6.00

Total Phenolic Content (TPC)

The Folin–Ciocalteu reagent method was employed to determine the TPC in curry leaf extracts, with gallic acid serving as the standard. Results are expressed as milligrams of gallic acid equivalents (GAE) per gram of extract. During the quantitative estimation of the TPC values varies in the curry leave under the influence of the extraction and geographical area. The total phenolic content in dry leaves, methanol extract & fresh leaves contain 127.34 mg GAE/g, 73.45 mg GAE/g and 58.21 mg GAE/g respectively. The shade dried leaves indicate the high concentration of the phenolic compound known for their antioxidant properties that contribute inhibition of the free radicals and potentially offers the protective effects against the oxidative stress related disease.

Total Flavonoid Content (TFC)

The aluminum chloride colorimetric method was utilized to quantify the TFC in curry leaf extracts, with quercetin as the reference standard. Values are expressed in milligrams of quercetin equivalents (QE) per gram of extract. In the quantitative estimation of the total flavonoid content was performed on shade dried leaves, methanol extract & fresh leaves and found to be 234.82 CE/gm, 49.53 CE/gm and 8.63 CE/gm respectively. The shade dried leave extract of the curry leaves show high concentration of flavonoids, known for diverse biological activities such as antioxidants, anti-inflammatory and anticancer effects used as potential therapeutic agents.

Total Alkaloid Content

The gravimetric method was applied to estimate the total alkaloid content in curry leaf extracts. This involved precipitating alkaloids using dilute hydrochloric acid and ammonium hydroxide, with results expressed as a percentage of alkaloids by weight. The shade dried leaves, methanol extract & fresh leaves were examined for the estimation of total alkaloidal content and founded to be 24%, 18% and 6%. The shaded dried leaf extract have the high concentration of the alkaloidal content which are used for various pharmacological actions such as analgesic, antimicrobial and anti-inflammatory activities.

Shade-dried leaves (water extract) showed the highest levels across all parameters, with a TPC of 128.39 mg GAE/g, a TFC of 234.82 CE/gm, and a TAC of 24%. This indicates that shade-drying followed by water extraction preserves and extracts a higher concentration of phenolic and flavonoid compounds, which are likely responsible for the strong antioxidant activity observed.

Methanolic leaves (general extract) had moderate values, with a TPC of 74.31 mg GAE/g, TFC of 49.53 CE/gm, and TAC of 18%. Although methanol is typically effective for extracting phenolic compounds, the lower results here suggest that drying conditions or sample preparation may have influenced compound retention.

Fresh leaves (methanolic extract) exhibited the lowest levels of bioactive compounds, with a TPC of 57.88 mg GAE/g, TFC of 8.61 CE/gm, and TAC of 6%. The reduced values could be due to the higher moisture content in fresh leaves, which may hinder efficient extraction and reduce the concentration of secondary metabolites.

The results show a clear correlation between phenolic and flavonoid contents with antioxidant activity (TAC) as shown in figure 3. Shade-drying enhances the concentration of these compounds, possibly due to enzymatic stabilization and water loss, leading to higher antioxidant potential.

Conclusion

The phytochemical evaluation of *Murraya koenigii* (curry leaves) involved comprehensive qualitative and quantitative analyses to identify and quantify the major bioactive secondary metabolites. The qualitative screening using standard biochemical tests revealed the presence of alkaloids, flavonoids, tannins, saponins, glycosides, phenolics, terpenoids, and steroids. Each of these constituents was confirmed through distinct

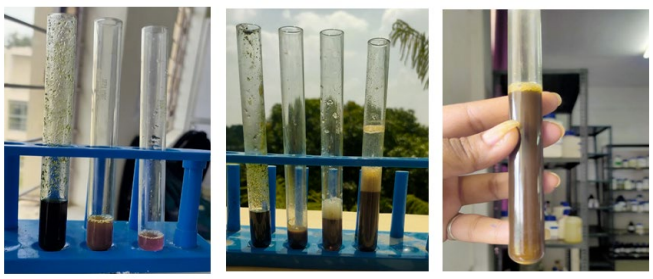

Figure 2. Qualitative Phytochemical Screening of Plant Extracts

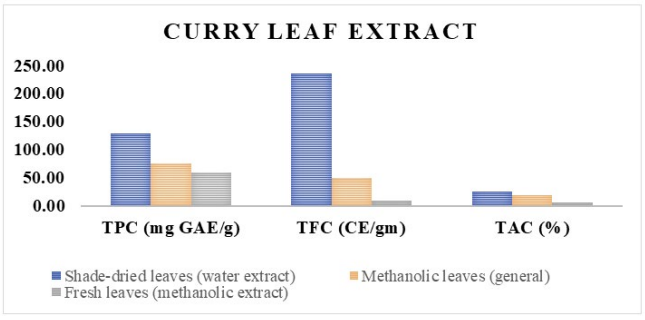

Figure 3. Quantitative estimation of the phytoconstituent of curry leaves

observable changes such as precipitate formation or color changes, underscoring their strong presence in the extract. The significance of these findings lies in the pharmacological potential associated with each compound group. Alkaloids are known for their antimicrobial, analgesic, and anti-inflammatory properties, while flavonoids contribute antioxidant, hepatoprotective, and cardioprotective benefits. Tannins and phenolics support wound healing and gastrointestinal health, and saponins provide cholesterol-lowering and immune-modulating effects. The presence of glycosides, terpenoids, and steroids further enhances the therapeutic value of the plant, offering cardiotonic, anti-inflammatory, and anticancer potentials, respectively. These observations validate the ethnomedicinal applications of curry leaves and emphasize their relevance in traditional medicine. The quantitative analysis further supports these findings. The Total Phenolic Content (TPC), determined using the Folin–Ciocalteu method, ranged up to 133.59 mg GAE/g DW, highlighting strong antioxidant potential. The Total Flavonoid Content (TFC), quantified using the aluminum chloride method, was reported at 1.24% (w/w), indicating a notable concentration of flavonoids with diverse therapeutic properties. Additionally, the gravimetric estimation of Total Alkaloid Content revealed a significant presence at 23.73%, affirming the bioactivity linked to this class of compounds.

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