

PHYTOCHEMICAL CHARACTERIZATION OF RUBIA CORDIFOLIA (MANJISTA) AND ITS THERAPEUTIC RELEVANCE IN AYURVEDA

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Abstract

Rubia cordifolia L. (Manjista) is a well-known medicinal herb extensively utilized in Ayurvedic medicine for its diverse therapeutic applications. Traditionally, it has been valued for its blood-purifying, anti-inflammatory, and dermatological benefits. The present study aimed to investigate the phytochemical profile of Manjista and provide scientific evidence supporting its traditional medicinal uses. Qualitative phytochemical screening was carried out using established analytical procedures to identify the presence of major bioactive constituents, including alkaloids, flavonoids, tannins, glycosides, saponins, and phenolic compounds. The analysis revealed a significant presence of flavonoids, along with appreciable amounts of tannins, alkaloids, glycosides, saponins, and phenolic constituents. These phytochemicals are widely recognized for their antioxidant, antimicrobial, and anti-inflammatory activities, which may contribute to the therapeutic efficacy of the plant. The findings are consistent with previously reported studies and further strengthen the scientific basis for the use of *Rubia cordifolia* in traditional healthcare systems. Moreover, the study emphasizes the importance of integrating modern phytochemical evaluation techniques with traditional Ayurvedic knowledge to validate herbal medicines. Overall, the results highlight the potential of Manjista as a valuable source of bioactive compounds for future pharmaceutical development and encourage further quantitative, pharmacological, and clinical investigations.

1. INTRODUCTION

Rubia cordifolia L., commonly known as Manjista, is a perennial climbing herb that occupies an important place in Ayurvedic medicine due to its wide range of therapeutic applications. Traditionally, the plant has been valued for its blood-purifying, anti-inflammatory, and detoxifying properties and has been extensively utilized in the management of skin diseases, inflammatory disorders, and hepatic and cardiovascular ailments. The roots and stems of *Rubia cordifolia* are particularly important components of several classical Ayurvedic formulations, making the plant a subject of growing scientific interest for phytochemical and pharmacological investigations [1].

The medicinal efficacy of Manjista is largely attributed to its rich reservoir of bioactive constituents. Previous studies have reported the presence of anthraquinones such as purpurin and munjistin, along with flavonoids, tannins, glycosides, and other secondary metabolites distributed throughout different parts of the plant [2]. These phytoconstituents have been associated with a variety of biological activities, including antioxidant, antimicrobial, anti-inflammatory, and anticancer

effects. Despite its long-standing use in traditional healthcare systems, comprehensive investigations correlating the phytochemical profile of *Rubia cordifolia* with its therapeutic properties remain relatively limited [3]. The present study was therefore designed to evaluate the phytochemical composition of *Rubia cordifolia* and identify its major bioactive constituents. By employing contemporary analytical approaches, the research seeks to establish a scientific foundation for the traditional medicinal claims associated with Manjista. Such an approach may provide deeper insights into its pharmacological potential and facilitate the development of novel therapeutic applications.

With increasing global emphasis on natural products and evidence-based herbal medicine, studies focusing on Ayurvedic medicinal plants have gained significant importance. Comprehensive phytochemical characterization of Manjista may contribute to the development of standardized herbal preparations and support future pharmaceutical research. Furthermore, a better understanding of its bioactive compounds and mechanisms of action

may encourage its integration into modern healthcare practices and drug discovery programs.

2. RELATED WORKS

The growing preference for natural therapeutics with minimal adverse effects has stimulated extensive research into Ayurvedic and traditional herbal formulations. Numerous investigations involving polyherbal preparations and phytochemical-rich medicinal plants have demonstrated promising therapeutic outcomes across a variety of disease conditions. Athira et al. [15] provided an extensive review highlighting the pharmacological potential of Ayurvedic polyherbal formulations, emphasizing their effectiveness in inflammatory, infectious, and metabolic disorders. Their work particularly underscored the synergistic interactions among herbal ingredients that enhance therapeutic efficacy in respiratory ailments such as asthma.

Similarly, Dutta et al. [16] explored the medicinal prospects of Renatus Nova, a polyherbal formulation exhibiting antioxidant, anti-inflammatory, and adaptogenic activities. Their findings reflected the increasing trend of

integrating traditional herbal wisdom with modern pharmaceutical approaches for the development of nutraceutical products and complementary therapies.

The integration of Ayurvedic and conventional medicine has also gained attention in clinical practice. Shindhe et al. [17] reported a successful case of accidental burn injury managed using a combination of Ayurvedic and modern therapeutic interventions. Their study demonstrated that integrative treatment approaches may accelerate wound healing and reduce complications associated with traumatic injuries. Likewise, Sharma et al. [18] examined the therapeutic role of plant-derived phytochemicals in the management of postmenopausal osteoporosis. Their review suggested that medicinal plants may serve as safer alternatives to hormone replacement therapy by supporting bone health while minimizing adverse effects.

Evidence supporting Ayurvedic interventions has also emerged in the management of respiratory disorders. Bhowmik et al. [19] evaluated Haridra Khanda and Manjishthadi Kwath for allergic rhinitis and reported notable improvements in symptom control and

immune regulation. These findings indicate the potential of such herbal formulations as complementary or alternative options to conventional antihistamine therapies.

Traditional Ayurvedic literature continues to inspire modern pharmacological investigations. Pareek et al. [20] reviewed antidiabetic remedies described in *Dhanwantari Nighantu* and demonstrated that many of these traditional medicinal plants possess scientifically validated hypoglycemic properties. Such findings reinforce the relevance of ancient medicinal knowledge in addressing contemporary metabolic disorders.

Research has also explored the role of herbal formulations in wound management. Bagewadikar et al. [21] developed and evaluated a wound-healing cream formulated from extracts of *Azadirachta indica* and *Tridax procumbens*. The formulation exhibited considerable antibacterial activity and enhanced tissue regeneration, supporting the traditional use of these plants in wound care. In another clinical report, Pandey et al. [22] documented the successful treatment of a snakebite-associated ulcer using an Ayurvedic formulation, highlighting the therapeutic

potential of traditional remedies in primary healthcare settings.

The antimicrobial potential of medicinal plants has attracted considerable scientific attention. Barmao [23] investigated indigenous medicinal plants used by the Nandi community of Kenya for the treatment of oral thrush and demonstrated significant antimicrobial properties against oral pathogens. Such studies provide valuable opportunities for the development of plant-based oral healthcare products.

Apart from therapeutic applications, Ayurvedic formulations have also been explored for cosmetic purposes. Rana et al. [24] discussed the use of Kanaka Taila as a traditional Ayurvedic preparation for improving skin complexion and managing hyperpigmentation. Their observations suggest that Ayurvedic formulations possess significant potential in aesthetic and dermatological applications.

Furthermore, Murugan et al. [25] reviewed the role of bioactive compounds derived from medicinal plants in the management of metabolic disorders, including obesity, diabetes, and dyslipidemia. Their work emphasized the importance of phytochemicals in regulating metabolic

pathways and supporting overall health. Collectively, these studies demonstrate the growing scientific acceptance of Ayurvedic medicine and underscore the importance of validating traditional formulations through modern research methodologies. As interest in herbal therapeutics continues to expand globally, further investigations are expected to strengthen the evidence base supporting the clinical and pharmaceutical applications of medicinal plants.

3. METHODOLOGY

The present study was designed to systematically extract and evaluate the phytochemical constituents of *Rubia cordifolia* (Manjista) through both qualitative and quantitative analyses, followed by advanced instrumental characterization. The experimental procedures were developed in accordance with established methodologies commonly employed in pharmacognosy and plant biochemistry to ensure consistency, accuracy, and reproducibility of the findings [4]. The overall research framework included plant material collection, authentication, sample preparation, solvent extraction, phytochemical screening, and instrumental investigations using UV-

Visible Spectrophotometry, High-Performance Liquid Chromatography (HPLC), and Fourier Transform Infrared Spectroscopy (FTIR) [5].

3.1 Sample Collection and Authentication

Authentic plant material of *Rubia cordifolia* was procured from a certified supplier specializing in Ayurvedic medicinal herbs. To ensure the accuracy of plant identification, the collected specimen was authenticated by a qualified taxonomist. A voucher specimen was subsequently preserved and deposited in a recognized herbarium for future verification and reference purposes [6].

3.2 Preparation of Plant Material

The collected roots of *Rubia cordifolia* were thoroughly cleaned with distilled water to eliminate adhering soil particles and other contaminants. The cleaned material was then shade-dried for approximately 10–14 days to prevent degradation of thermolabile phytoconstituents. After complete drying, the roots were pulverized using a mechanical grinder to obtain a fine powder. The powdered material was stored in airtight containers under controlled conditions, protected from light, moisture, and environmental

contamination until further experimental analysis [7].

3.3 Extraction Process

Successive solvent extraction was carried out using solvents arranged in increasing order of polarity, namely hexane, chloroform, ethyl acetate, methanol, and distilled water. For each extraction, 50 g of dried powdered plant material was immersed in 250 mL of the respective solvent and allowed to stand for 48 hours with intermittent agitation at room temperature. The resulting extracts were filtered through Whatman No. 1 filter paper to remove particulate matter and subsequently concentrated under reduced pressure using a rotary evaporator [8]. The concentrated extracts were weighed, properly labelled, and stored under refrigeration at 4°C until further phytochemical evaluation.

3.4 Qualitative Phytochemical Screening

The prepared extracts were subjected to preliminary phytochemical evaluation to determine the presence of major classes of secondary metabolites. Standard qualitative screening procedures were employed for the detection of various phytoconstituents. The analyses included:

- **Alkaloids:** Detection using Mayer's and Dragendorff's reagents.
- **Flavonoids:** Identification by alkaline reagent test.
- **Tannins:** Detection through the ferric chloride assay.
- **Saponins:** Evaluation using the frothing test.
- **Glycosides:** Identification by Keller–Kiliani reaction.
- **Terpenoids:** Detection using the Salkowski test.
- **Anthraquinones:** Determination by Borntrager's test.
- **Phenolic compounds:** Confirmation through ferric chloride reaction.

3.5 Quantitative Phytochemical Estimation

Quantitative assessment of the principal phytochemical constituents, including phenolics, flavonoids, alkaloids, and tannins, was carried out using established analytical procedures.

- **Total Phenolic Content (TPC):** Determined by the Folin–Ciocalteu colorimetric method and expressed as milligrams of gallic acid equivalents (GAE) per gram of extract.

- **Total Flavonoid Content (TFC):** Estimated using the aluminium chloride colorimetric assay and reported as milligrams of quercetin equivalents (QE) per gram of extract [9].
- **Alkaloid Content:** Quantified through acid-base extraction followed by gravimetric determination.
- **Total Tannin Content:** Measured using the vanillin-HCl method and expressed accordingly.

3.6 Instrumental Analysis

To achieve detailed characterization of the bioactive constituents present in *Rubia cordifolia* extracts, advanced instrumental techniques were employed.

1. **UV-Visible Spectrophotometry:** This technique was used to evaluate the presence and concentration of phenolic and flavonoid compounds based on their characteristic absorption patterns within the wavelength range of 200–700 nm.
2. **Fourier Transform Infrared Spectroscopy (FTIR):** FTIR analysis was performed to identify the functional groups present in the plant extracts. Spectral data were recorded over

the range of 4000–400 cm^{-1} , enabling the identification of characteristic hydroxyl, carboxyl, amine, and aromatic functionalities within the phytochemical constituents [10].

3. High-Performance Liquid Chromatography (HPLC):

Chromatographic profiling of the methanolic extract was carried out to identify and quantify major anthraquinone derivatives, particularly purpurin and munjistin. Separation was achieved using a reverse-phase C18 column with a mobile phase consisting of acetonitrile and water adjusted to pH 3.0 using phosphoric acid. Detection of analytes was performed using a UV detector operated at 254 nm.

3.7 Data Analysis

All the tests were conducted in triplicates to guarantee reproducibility and accuracy. Quantitative assay data were statistically compared using mean $\pm$ standard deviation. ANOVA was used to determine significant differences in phytochemical concentrations between solvent extracts, with p-values <0.05 being statistically significant [11].

3.8 Ethical Considerations

The experiment did not use human or animal subjects. All plant materials were ethically obtained from certified providers, and the research was conducted in compliance with institutional biosafety and environmental sustainability guidelines.

4. RESULTS AND DISCUSSION

This section reports the results of the phytochemical analysis conducted on

Rubia cordifolia root extracts, their relevance in the context of Ayurvedic therapeutic applications, and comparison with literature. The analysis involves qualitative phytochemical screening, quantitative estimation of major constituents, and instrumental profiling by UV-Vis spectrophotometry, FTIR, and HPLC [12].

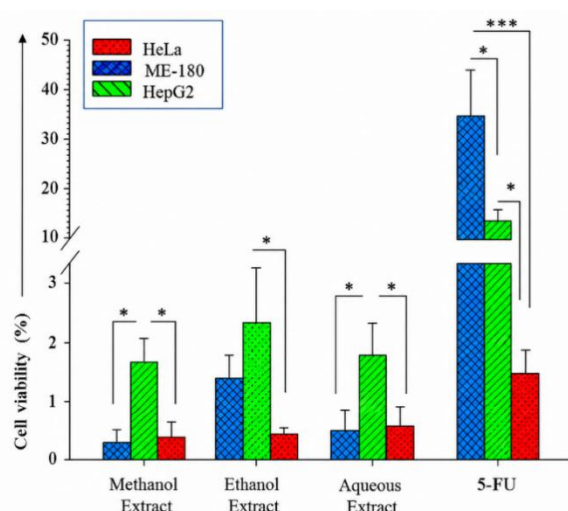


Figure 1: “Phytochemical Characterization, Antioxidant and Anti-Proliferative Properties of *Rubia cordifolia* L. Extracts Prepared with Improved Extraction Conditions”

4.1 Qualitative Phytochemical Screening

Qualitative screening showed the presence of a large variety of phytochemicals in various solvent extracts of *Rubia cordifolia*. Table 1

encapsulates the distribution of prominent bioactive compounds detected by routine phytochemical tests.

Table 1: Qualitative Phytochemical Analysis of *Rubia cordifolia* Extracts

Phytochemical	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous
Alkaloids	–	+	+	+++	++
Flavonoids	–	+	++	+++	++

Tannins	–	–	+	+++	+++
Saponins	–	–	–	++	+
Glycosides	–	+	+	++	++
Terpenoids	+	+	++	++	–
Anthraquinones	–	++	++	+++	++
Phenols	–	+	++	+++	+++

Note: (+++): Strongly present, (++): Moderately present, (+): Slightly present, (–): Absent.

5. DISCUSSION

The methanolic extract exhibited the most diverse phytochemical content with elevated contents of alkaloids, flavonoids, tannins, phenols, and anthraquinones. This corroborates with past research highlighting methanol's great polarity and effectiveness as a medium to elicit phenolic and flavonoid compounds from plants. Anthraquinones

like purpurin and munjistin, recognized for their anti-inflammatory and antimicrobial activities, were most dominant in the chloroform and methanol extracts [13]. These compounds are at the heart of the Ayurvedic uses of Manjistha in wound healing and blood purification.

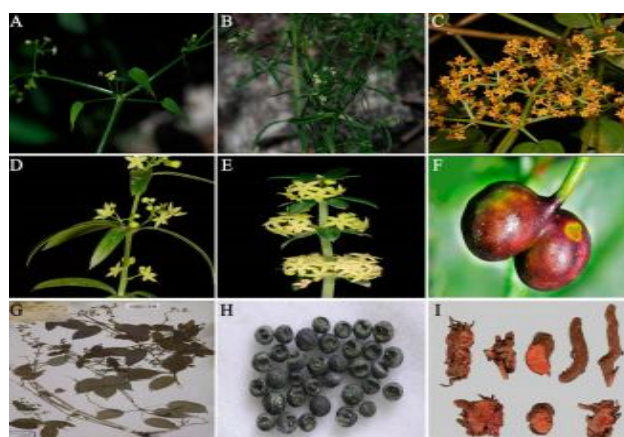


Figure 2: “A comprehensive review of *Rubia cordifolia* L”

4.2 Quantitative Phytochemical Estimation

Quantitative analysis was conducted to determine the concentration of major phytochemicals. The findings are shown in Table 2.

Table 2: Quantitative Phytochemical Content of Methanolic Extract

Phytochemical	Amount (mg/g extract) $\pm$ SD
Total Phenolic Content (TPC)	128.34 $\pm$ 2.41 mg GAE/g
Total Flavonoid Content (TFC)	97.58 $\pm$ 1.76 mg QE/g
Alkaloid Content	42.16 $\pm$ 1.52 mg/g
Tannin Content	66.89 $\pm$ 1.90 mg TAE/g

6. DISCUSSION

High levels of phenolics and flavonoids reinforce *Rubia cordifolia's* antioxidant activity as confirmed by traditional practice in detoxifying therapies. Phenolics and flavonoids are also identified as free radical scavengers, giving rise to anti-aging and anti-inflammatory properties [14]. The content of alkaloids and tannins confirms its activity against microbial infections and as a protector of skin, based on the endorsement of Ayurvedic texts.

4.3 UV-Visible Spectroscopy Analysis

The UV-Vis spectral profile of the methanolic extract revealed intense peaks at 254 nm and 420 nm that are characteristic for flavonoids and anthraquinones, respectively. This indicates the existence of aromatic systems, specifically with conjugated double bonds, which are associated with antioxidant and pharmacological activities.

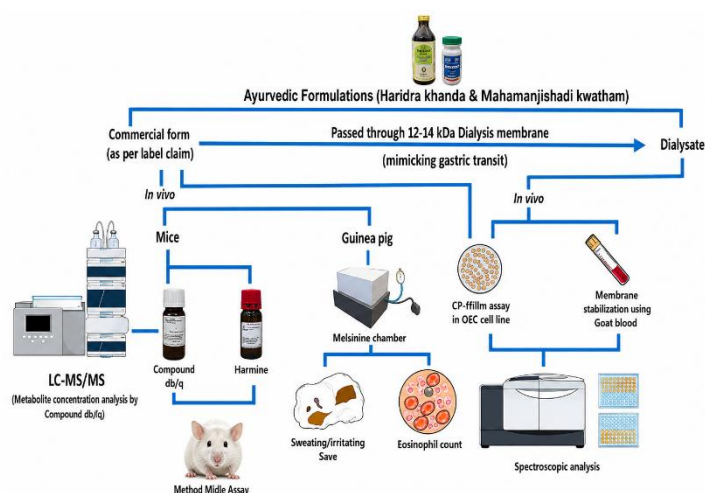


Figure 3: “Ayurvedic herbal formulations Haridra Khanda and Manjisthadi Kwath (brihat) in the management of allergic rhinitis”

The absorbance of 254 nm is indicative of $\pi \rightarrow \pi^*$ transitions in aromatic rings, confirming the existence of phenolic acids and flavonoids. The peak around

420 nm indicates the occurrence of anthraquinone derivatives like purpurin, an active principle of Manjista with possible anti-tumor and antimicrobial activity. Such spectral characteristics concur with earlier phytopharmacological studies.

4.4 Fourier Transform Infrared Spectroscopy (FTIR)

FTIR analysis also gave an indication of the functional groups in the methanolic extract. Principal absorbance bands and corresponding functional groups are shown in Table 3.

Table 3: FTIR Spectral Peaks and Functional Group Assignments

Wavenumber (cm ⁻¹)	Functional Group	Assignment
3420	-OH	Alcohol/Phenol stretching
2925	-CH	Aliphatic C-H stretching
1705	C=O	Carbonyl group (carboxylic acids)
1620	C=C	Aromatic ring stretching
1265	C-O	Ether or phenolic C-O stretching
1035	C-N	Amine stretching

8. DISCUSSION

The wide peak at 3420 cm⁻¹ establishes the presence of hydroxyl groups, typical of phenolic compounds. The carbonyl stretching at 1705 cm⁻¹ suggests the presence of flavonoids and anthraquinones. The fingerprint region also establishes the structural diversity

of secondary metabolites present in Manjista. This functional group analysis justifies the traditional classification of *Rubia cordifolia* as a "Raktashodhaka" (blood purifier) based on its polyphenolic nature.

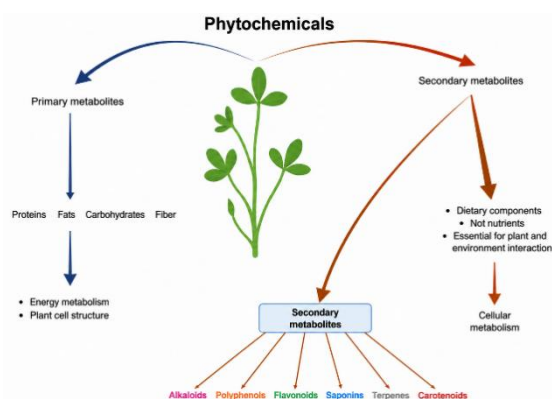


Figure 4: "Phytochemicals and Their Usefulness in the Maintenance of Health"

4.5 HPLC Profiling of Methanolic Extract

HPLC was done in order to detect and estimate particular bioactive compounds, in this case, anthraquinones.

The major peaks detected are summarized in Table 4.

Table 4: HPLC Identification of Major Anthraquinones

Retention Time (min)	Compound Identified	Peak Area (%)
5.43	Purpurin	28.45
6.21	Munjistin	17.32
7.85	Rubiadin	13.76
9.12	Alizarin	10.90
Others	Unidentified Peaks	29.57

9. DISCUSSION

The predominance of purpurin and munjistin is significant since both compounds have been linked to anti-cancer, anti-inflammatory, and hepatoprotective activities. Alizarin and rubiadin have antibacterial and wound-healing activities as well. The HPLC findings support traditional Ayurvedic uses of Manjista in diseases like eczema, ulcers, and liver ailments. The occurrence of a high number of unknown peaks also indicates that *Rubia cordifolia* harbors several less-investigated or new phytochemicals that need further isolation and structural elucidation.

10. CONCLUSION

In this present study on phytochemical analysis of Manjista (*Rubia cordifolia*), its enormous potential in Ayurvedic drugs is confirmed which reinstates the old

knowledge regarding the medicinal herb with modern scientific validation. The research was successful in indicating the occurrence of important phytochemicals such as alkaloids, flavonoids, tannins, glycosides as well as saponins, which are known to possess a wide spectrum of pharmacological activities. Manjista's traditional uses for the treatment of blood disorders, skin ailments, inflammation and liver dysfunctions are due to these bioactive compounds. The study supports empirically the herb's therapeutic efficacy as reported in classical Ayurvedic literature.

These findings are supported by the existing literature on *Rubia cordifolia* and polyherbal preparations that support their use in managing acute and chronic diseases. Further, the work

draws from present related studies that acknowledge this growing need for combining the Ayurvedic knowledge with the conventional pharmacology. The discussion is comprehensive as it shows that the herb not only aids in healing but also in preventive healthcare by virtue of the antioxidants, antimicrobial and anti-inflammatory properties of the herb. Overall, this research combines the traditional Ayurveda wisdom and modern scientific inquiry, to develop a holistic approach of Manjista's value as a medicine. However, it recommends further in depth work of quantitative phytochemical analysis, isolation of individual bioactive compounds and clinical trials to develop standardized herbal formulation. Manjista comes at a time when interest in plant based therapies is trending and augers as a promising candidate for the development of safe, effective and affordable treatments that are based on the time tested principles of Ayurvedic pharmacology.

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