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**Mohammad Abu Bin Nyeem**  
Assistant Professor, Department  
of Unani Medicine, Hamdard  
University, Bangladesh

**Md. Abdul Mannan**  
Faculty of Unani & Ayurvedic  
Medicine, Department of Unani  
Medicine, Hamdard University,  
Bangladesh

## Rubia cordifolia-phytochemical and Pharmacological evaluation of indigenous medicinal plant: A review

**Mohammad Abu Bin Nyeem and Md. Abdul Mannan**

### Abstract

*Rubia cordifolia* (Manjistha), often known as common Madder is a species of flowering plant in the coffee family, Rubiaceae. It has been cultivated for a red pigment derived from roots. It is a perennial, prickly climber with a stem, growing up to 12 m long. Leaves are highly variable, ovate lanceolate, 5-7 nerved, 2-10 cm long and 2-5 cm broad, occurring in whorls of 4-6. Flowers are fragrant, minute, whitish or greenish yellow. Fruit is minute, glabrous, 1-2 seeded, dark purplish or blackish when mature. During August to October plant carries flower and fruit. Roots are perennial, long, cylindrical, and rusty brown in color. Manjistha is probably the best alternative or blood-purifying herb in indigenous system of medicine. It cools and detoxifies the blood, dissolves obstructions in blood flow and removes stagnant blood. It is having healing action, thus helpful in erosions of gums and bleeding gums. In indigenous medicine, it is used as an immune regulator. Its antioxidant properties are also being investigated. Its role in supporting heart health is evidenced by studies that show that it regulates blood pressure, blood vessel constriction and the tendency of blood to form clots.

Many research teams have suggested that *Rubia cordifolia* has wound healing, antibacterial, antioxidant, anticancer, anti-inflammatory and analgesic, hepatoprotective, anti-platelet activating factor & anti-acute activity.

**Keywords:** *Rubia cordifolia*, manjistha, pharmacological activity

### Introduction

In the last few years there has been an exponential growth in the field of herbal medicine and these drugs are gaining popularity both in developing and developed countries because of their natural origin and less side effects. Many traditional medicines in use are derived from medicinal plants, minerals and organic matter. Over the last 2500 years, there have been very strong traditional systems of medicine such as Chinese, Ayurvedic, and the Unani, born and practiced, more in the eastern continent. These traditions are still flourishing, since; approximately 80% of the people in the developing countries rely on these systems of medicine for their primary health care needs [1]. These plants contain substances that can be used for therapeutic purposes, of which are precursors for the synthesis of drugs [2]. A lot of research work has been carried out on some medicinal herbs and they have been found to have definite action on the nervous, circulatory, respiratory, digestive and urinary systems; as well as the sexual organs, the skin, vision, hearing and taste [3].

*Rubia cordifolia*, often known as common Madder or Indian Madder, is a species of flowering plant in the coffee family, Rubiaceae [4]. Its stem is slender, more or less cylindrical, slightly flattened, wiry, about 0.5 cm thick, brown to purple colored; surface scabrous, stiff and grooved with longitudinal cracks; prickles present in the immature stem; nodes distinct having two leaf scars, one on either side; fracture, short. Mature stem shows exfoliating cork, ruptured at places, forming dome-shaped structure, consisting of 3-12 or more layered, squarish and tangentially elongated, thin-walled cells, appearing polygonal in surface view; secondary cortex 3-5 layered consisting of tangentially elongated, thin-walled cells, some of which contain acicular crystals of calcium oxalate as isolated or in bundles; a few cells contain sandy crystals as black granular masses; secondary phloem, a wide zone of reddish color, composed of sieve elements and phloem parenchyma, fibers absent; phloem parenchyma smaller towards inner side gradually becoming larger and tangentially elongated towards periphery, a few cells

### Correspondence

**Mohammad Abu Bin Nyeem**  
Assistant Professor, Department  
of Unani Medicine, Hamdard  
University, Bangladesh

contain sandy crystals of calcium oxalate; secondary xylem forms a continuous cylinder of reddish color, composed of vessels, tracheids, fibers and xylem parenchyma; vessels numerous, distributed uniformly throughout xylem, larger towards outer side and smaller towards centre; in macerated preparation, vessels show great variation in shape and size having lignified walls and pitted thickening; xylem fibres thick-walled, long and short, longer ones have narrow lumen while shorter ones have wide lumen with pitted thickenings; xylem parenchyma also vary in shape and size having pitted or reticulate thickening; centre occupied by narrow pith consisting of thin walled, parenchymatous cells, a few cells contain sandy crystals of calcium oxalate [5].



**Fig 1:** *Rubia cordifolia*

### Scientific Classification of *Rubia*

Kingdom: Plantae  
 Class: Dicotyledoneae  
 Subclass: Sympetalae  
 Order: Rubiales  
 Family: Rubiaceae  
 Genus: *Rubia*  
 Species: *cordifolia* [6].

### Chemical constituents of *Rubia cordifolia*

Different classes of bioactive compounds such as anthraquinones and their glycosides, naphthoquinones and glycosides, terpenes, bicyclic hexapeptides, iridoids [7], carboxylic acids and saccharides were isolated from various parts of *R. cordifolia*. The roots contain a mixture of purpurin, munjistin, small amounts of xanthopurpurin and pseudopurpurin. Alizarin (1, 3-dihydroxy-2-ethoxymethyl, 10-anthraquinone), mollugin (1-hydroxy-2-methyl-9, 10-anthraquinone), 1, 3, 6-trihydroxy-2-methyl-9, 10-anthra-

quinone-3-O-(6'-O-acetyl)- $\alpha$ -L-rhamnosyl (1 $\rightarrow$ 2)- $\beta$ -D-glucoside, 1, 3, 6-tri hydroxy-2-methyl-9, 10-anthraquinone-3-O- $\beta$ -L-rhamnosyl (1 $\rightarrow$ 2)- $\beta$ -D-glucoside, 1, 3, 6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'O-acetyl)- $\beta$ -D-glucoside, 2-carbomethoxy+++3-prenyl-1, 4-naphthohydroquinone di- $\beta$ -D-glucoside, rubimallin,  $\beta$ -sitosterol and daucosterol were also isolated from roots [8]. Several naphthoquinones and hydroxy anthraquinones and their glycosides were also isolated [9]. 1-hydroxy 2-methyl anthraquinone, nordamnacanthal, physcion and 1, 4-dihydroxy 6-methyl-anthraquinone [10], rubiatril [11], 1-hydroxy 2-methoxy anthraquinone; 1, 4dihydroxy 2-methyl 5-methoxy anthraquinone; 1, 3-dimethoxy 2-carboxy anthraquinone, rubiadin [12], naphthaquinones [13], 1, 4-dihydroxy 2methylanthraquinone and 1, 5-dihydroxy 2methylanthraquinone and 3-prenyl methoxy 1, 4naphthoquinone [14] and an anti-inflammatory compound rubimallin and antibacterial agents like  $\alpha$ -sitosterol and daucosterol, [15] rubicoumaric acid and rubifolic acid were identified and their structure was elucidated spectrometrically [16]. Other quinones isolated were 4dihydroxy 2-methylanthraquinone, 1, 5-dihydroxy 2methyl anthraquinone, 3-prenyl methoxy 1, 4naphthoquinone, lucidin primeveroside, ruberythric acid anthraquinones, 2-methyl-1, 3, 6-trihydroxy-9, 10anthraquinone and 2-methyl-1, 3, 6-trihydroxy-9, 10anthraquinone 3-O $\alpha$ -rhamnosyl (1 $\rightarrow$ 2)- $\beta$ -glucoside. 6Methoxy geniposidic acid (iridoids) is found along with manjistin, garancin and alizarin [17]. Oleananes such as rupiprasin A, B and C along with arborane triterpenoids including rubiarbonol A, B, C, D, E and F have been isolate [18-19]. Naphtho hydroquinones, furomollugin, mollugin, rubilactone [20], nordamnacanthal, naphthohydroquinone anhydride were isolated from root extracts [21]. The dried roots contain mollugin, furomollugin, eugenol and (E) anethole as chief component in the essential oil and eugenol, geraniol and geranyl acetate were the most aroma compounds [22]. A new anthraquinone, rubiacordone A [23], naphthoic acid esters like rubilactone, dihydromollugin were identified based on the physicochemical properties and spectrometric analyses [24]. Cordifoliol and cordifodiol also have been isolated from the roots [25-26]. Epoxymollugin, 31 1-hydroxy-2, 7-dimethylanthraquinone, 2-hydroxy-6methylanthraquinone and 2, 6-methylanthraquinone, nnonadecane, n-heptadecane, 8-hydroxy-n-pentadecanyl decan-4en-1-oate and noctaosanyl octa-1-oate were isolated from the roots [27]. Recently, the chloroform soluble fraction of the chloroform-methanol extract of the dried roots led to isolation of one new naphtho hydroquinone dimer and 3- $\alpha$ -friedelinol, atraric acid, vanillic acid and D-3-O-methoxy-chiro-inositol [28].

Bicyclic peptide of RA-series was elucidated from spectroscopic and chemical evidences from the roots of *R. cordifolia*. The bicyclic hexapeptides RA-I and RA-II have been isolated from chloroform/methanol extract. RA-III to RAVII has been obtained from the benzene-soluble fraction of methanol extracts [29-30]. The structures of antitumor bicyclic hexapeptides RA-VI, RA-VIII [31], RA-VII and RA-X were identified [32]. RA-VII is an inhibitor of protein biosynthesis *in vitro* and *in vivo* [33]. Ala 2 modified RA-VII derivative had been synthesized from RA-X methyl ester and evaluated for cytotoxicity to P388 leukemia and KB cells *in vitro* [34]. The conformational analysis of RA-VII, derived from RA-VII by hepatic microsomal biotransformation revealed a restricted conformational state [35]. The bicyclic hexapeptides glucosides RA-IX and -X, [36] RAXI, RA-XII, XIII, RA-IV [37], RA-XV and -XVI possess antitumour

activity [38]. RAXVII was another antitumour agent, which showed little effect on the conformation of the molecule [39]. The spectroscopic studies revealed the structure of hexapeptides like RA-XV, RAXVI, RA-XII [40], RA-XVII, RA-XIX, -XX, -XXI and -XXII. RAXVIII is a hydroxylated derivative of RA-VII by the semisynthesis from deoxybouvardin, showed cytotoxicity against P-388 cells [41]. The structure of RA-XXIII and RAXXIV [42] and RA-dimer A, a dimeric antitumor bicyclic hexapeptides, were also identified from the root extracts [43]. Recently two new bicyclic hexapeptides, alloRA-V and neo-RA-V, and one cyclic hexapeptide, O-secoRA-V were isolated [44].

## Pharmacological Activity

### Antibacterial Activity

The antibacterial activity of the extracts of *Ventilago madraspatana* stem-bark, *Rubia cordifolia* root and *Lantana camara* root-bark, prepared with solvents of different polarity, was evaluated by the agar-well diffusion method. Twelve bacteria, six each of gram-positive and gram-negative strains, were used in this study. Chloroform and methanol extracts of *R. cordifolia* and *L. camara* was found to be more specific towards the gram-positive strains, although gramnegative *P. aeruginosa* was also inhibited by the methanol extracts of both these plants in a dose dependent manner. *R. cordifolia* was significantly active against *B. subtilis* and *S. aureus* compared with streptomycin and penicillin G used as standards [45].

### Wound Healing Activity

Assessment of Wound Healing of a polyherbal formulation containing *Rubia cordifolia* was done. Cream formulation of the herbal drug combination of *R. cordifolia*, *C. asiatica*, *T. belerica*, *P. zeylanica*, and *W. somnifera* was formulated. Animals were inspected daily up to 20th days and healing was assessed based on physical parameter namely, wound contraction, period of epithelization and histological study. It promotes contraction and epithelization of excision wound [46].

Several drugs of plant, mineral and animal origin are described in the Ayurveda for their wound healing properties under the term 'vranaropaka'. *R. cordifolia* was also found to be effective in experimental models [47].

Psoriasis is skin disorder characterized by hyperproliferation and aberrant differentiation of epidermal keratinocytes. Ethyl acetate (EA) fraction of *Radix Rubiae* inhibits cell growth and promotes terminal differentiation in cultured human keratinocytes which strongly suggest its antipsoriatic activity. Evaluation is done by cornified envelope (CE) formation assay showed that EA fraction of *Radix Rubiae* significantly accentuated the CE formation, a well-recognized marker of terminal differentiation, in cultured HEK and HaCaT cells in a dose and time dependent manner [48].

### Antioxidant Activity

*R. cordifolia* extracts were also evaluated for antioxidant and lipid peroxidation inhibitory activity by 1, 1-diphenyl-2-picryl-hydrazyl and TBARs Thiobarbituric acid reactive substances method respectively. Extract of *R. cordifolia* showed a significant inhibitory activity against *Propioni bacterium acnes* standardized culture. The evaluation was carried out by broth dilution method; suggested MIC of *R. cordifolia* extract was 600µg/ml. The methanolic extract of *R. cordifolia* showed significant lipid peroxidation inhibitory activity. The IC<sub>50</sub> value of 138µg/ml and R<sub>2</sub> was 0.9921. The result was compared with curcumin as standard (IC<sub>50</sub>

50µg/ml, R<sub>2</sub> 0.9469). These investigations have revealed *R. cordifolia* as a promising anti-acne agent because it inhibits the proliferation of *Propioni bacterium acnes* and hence prevents its consequences [49].

### Anticancer activity

Cancer is the most devastating disease and leading cause of death throughout the world. Natural drugs are under investigation for their selective cytotoxicity to cancer cells. Methanol fraction of *Rubia cordifolia* extract exhibited potent inhibition of Human cervical cancer cell line and Human larynx carcinoma cell line while was found to be less cytotoxic against normal human kidney cells displaying safety for normal cells. *Rubia cordifolia* can be a source of potent pharmacophore for treatment of disease like cancer [50].

### Anti-inflammatory and Analgesic activity

The present study was aimed to investigate the analgesic and anti-inflammatory effect of the methanolic extract of root of *Rubia cordifolia* in rats. *Rubia cordifolia* (100-300 mg/kg, p. o.) was evaluated for its antiinflammatory activity by carrageenan induced rat paw edema and *Rubia cordifolia* (200-400 mg/kg) for its analgesic activity by tail flick method. *Rubia cordifolia* (100-300 mg/kg, p. o.) showed significant ( $P<0.05$ ) reduction in the paw edema produced by the carrageenan and significant ( $P<0.05$ ) increased reaction time in tail flick test [51].

*R. cordifolia* is considered to be traditionally useful as an analgesic, astringent, external application in inflammations, ulcers and skin diseases [52]. (Khalid, 1995). The plant is also claimed to relieve the symptoms of pruritus, burning and exudation from skin [53]. (Nadkarni, 1976). During studies in patients with eczema, the topical application of the plant showed a 50% reduction in the severity score within 4 days, the oedema, exudation and itching being significantly relieved [54]. (Bapalal, 1965). *R. cordifolia* was studied for the anti-inflammatory effect in rats with carrageenan paw oedema. The plant showed significant anti-inflammatory activity at a dose of 10 and 20 ml/kg of the water extracts. The activity was comparable to that of phenylbutazone (100 mg/kg) [55] (Antarkar *et al.* 1983). *R. cordifolia* inhibited the lipoxygenase enzyme pathway and the production of cumene hydroperoxides. The lipoxygenase pathway catalyses the production of various inflammatory mediators such as the leukotrienes which are involved in asthma, arthritis, and other inflammatory disorders [56].

### Hepatoprotective Activity

The hepatoprotective activity of an aqueousmethanol extract of *R. cordifolia* was investigated against acetaminophen and CCl<sub>4</sub>-induced hepatic damage. Acetaminophen produced 100% mortality at a dose of 1 g/kg in mice while pretreatment of animals with *R. cordifolia* extract reduced mortality to 30%. Acetaminophen at a dose of 640 mg/kg produced liver damage in rats as manifested by the rise in serum levels of glutamic oxaloacetic transaminase (SGOT) and glutamate pyruvate transaminase (SGPT). Pretreatment of rats with *R. cordifolia* extract lowered significantly the SGOT and SGPT levels. Similarly, hepatotoxic dose of CCl<sub>4</sub> raised the SGOT and SGPT levels respectively compared with respective control. The same dose of *R. cordifolia* was able to prevent significantly the CCl<sub>4</sub>-induced rise in serum enzymes and the estimated values of SGOT and SGPT. Moreover, it prevented CCl<sub>4</sub>-induced prolongation in pentobarbital-induced sleep confirming the hepatoprotective effects of the extract [57].

### Anti-platelet activating factor activity

*R. cordifolia* is clinically used for the purification of blood by the physicians of the Indian Systems of Medicine. The effect of the partially purified fraction of this whole plant had been studied on rabbit platelets. It inhibited the platelet aggregation induced by PAF (platelet activating factor) but not thrombin. PAF (platelet activating factor) is a phospholipids involved in thrombosis, allergy and nervous disorders. *R. cordifolia* extract also inhibited the binding of <sup>3</sup>H labeled-PAF to the platelets in the dose-dependent manner. Thus it appears that *R. cordifolia* inhibits action of PAF at its receptor level either by its blocking or by desensitization <sup>[58]</sup>.

### Anti-acne property

*Propionibacterium acnes*, an anaerobic pathogen, plays an important role in the pathogenesis of acne by inducing certain inflammatory mediators. These mediators include reactive oxygen species (ROS) and pro-inflammatory cytokines. In the study, ROS, interleukin-8 (IL-8) and tumor necrosis factor- $\gamma$  (TNF- $\gamma$ ) were used as the major criteria for the evaluation of anti-inflammatory activity. The polymorphonuclear leukocytes (PMNL) and monocytes were treated with culture supernatant of *P. acnes* in the presence or absence of herb. It was found that *R. cordifolia* caused a statistically significant suppression of ROS from PMNL. Thus, *R. cordifolia* showed anti-inflammatory activity by suppressing the capacity of *P. acnes*-induced ROS and proinflammatory cytokines, the two important inflammatory mediators in acne pathogenesis <sup>[59]</sup>.

### Anxiolytic Activity

Mice treated with triterpenes isolated from the petroleum ether extract of *R. cordifolia* exhibited anxiogenic activity by remaining for most of the time in the closed arm 38. Whereas, the ethanolic extract exhibited anxiolytic activity as indicated by a significant increase in open arm occupancy <sup>[60]</sup>.

### Anti-allergic Activity

Alcoholic extract of *R. cordifolia* inhibited passive cutaneous anaphylaxis (PCA) in the mouse and rat <sup>[61]</sup>.

### Radio Protective Property

Radio protective potential of alcoholic extract of root of Manjistha showed a significant radiation protection (67%) as assessed by increased animal survival when *R. cordifolia* extract was administered intraperitoneally before radiation exposure. Results suggest the alcoholic root extract provides protection against radiation-induced lipid peroxidation, hemopoietic injury and genotoxicity <sup>[62]</sup>.

### Anti-HIV Activity

Assessment of anti-HIV activity of various extracts prepared from Indian medicinal plants. The plants were chosen on the basis of similarity of chemical constituents with reported anti-HIV compounds or on the basis of their traditional usage as immunomodulators. Different extracts were prepared by Soxhlet extraction and liquid-liquid partitioning. Ninety-two extracts were prepared from 23 plants. Anti-HIV activity was measured in a human CD4<sup>+</sup> T-cell line, CEM-GFP cells infected with HIV-1NL4.3. Nine extracts of 8 different plants significantly reduced viral production in CEM-GFP cells infected with HIV-1NL4.3. *Aegle marmelos*, *Argemone mexicana*, *Asparagus racemosus*, *Coleus forskohlii*, and *Rubia cordifolia* demonstrated promising anti-HIV potential <sup>[63]</sup>.

### Conclusions

*Rubia cordifolia* commonly known as Manjistha or Indian madder is a rich source of anthraquinones responsible for its traditional, phytochemical and pharmacological activities. Today clinical investigations of herbal formulations and their market preparations, both are on demanding because of better safety and efficacy without or minimal side effects. Manjistha stem describes as cure for snake bite and scorpion sting. It is also effective on non-healing diabetic foot ulcer. Manjistha having cooling effect in the body and therefore, traditionally used for chronic pyrexia and puerperal fever. It is a popular remedy for the relief of heat and itching in eczema, psoriasis, herpes, and scabies and also reported successful in treatment of vitiligo when given with honey. Manjistha has been reported for the presence of glycosides, saponins, anthraquinones, tannins, hexapeptides, quinones, and triterpenoids. *R. cordifolia* is an important medicinal plant commonly used in the traditional system of medicine for treatment of different ailments. This review illustrates its major constituents, pharmacological actions substantiating the claims made about this plant in the traditional system of medicine and its clinical applications.

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