

SCREENING OF ACTIVE PHYTOCHEMICALS IN LEAVES AND STEM OF *MEYNA SPINOSA* ROXB. EX. LINK.

Biological Science

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ABSTRACT

BACKGROUND: *Meyna spinosa* Roxb. Ex. Link. is a spinous, armed small tree or large shrub. The different plant parts of *Meyna spinosa* has been consumed as traditional or folk medicine to treat different health-related problems such as diabetes, diphtheria, stomach pain, headache, liver problem, indigestion, throbbing urination, and skin problems such as pimples and acne problems. The important phytochemicals of *Meyna spinosa* include phenolic compounds, tannins, flavonoids, alkaloids, saponins, phytosterols, terpenoids, triterpenoids, oleanic acid which are used in the treatment of various chronic diseases such as hepatic disorder, cancer, inflammation, diabetes, microbial infection, and also exhibit potent anti-oxidant activity.

AIM: To study the active phytochemicals.

OBJECTIVE: To investigate the active principles of this plant with characteristic functional groups.

METHODS: Stem and leaves of *Meyna spinosa* was extracted using methanol as solvent, and its dry powder was subjected to screening of active phytochemicals employing FTIR analysis. The tests were performed to check the presence or absence of bioactive compounds.

RESULTS: FTIR analysis of leaves gave four major peaks. The first major peak had the value of 2916 belongs to the functional group of aromatic (C-H). The second major peak had the value of 2848, which belonged to the primary, secondary amines and amides (N-H) group. The third major peak of 1735 belonged to the esters and saturated aliphatic group (C=O) and the last peak 1242 belonged to the group of aromatic amines (C-N). Methanol extract of leaves of *Meyna spinosa* revealed the presence of 12 bioactive compounds. The stem of *Meyna spinosa* revealed the presence of sterols, triterpenoids (Lanosterol) and alkaloids, tannins etc.

CONCLUSIONS: The present study contributes valuable information of active phytochemicals with their relevant functional groups in stem and leaves of *Meyna spinosa*. The dry powder of methanol extract of stem and leaves had all the active principles like flavonoids, glycosides, saponins, phenols, steroids, tannins and terpenoids.

KEYWORDS

Meyna spinosa, Active phytochemicals, Leaves, Stem, FTIR

INTRODUCTION

Meyna spinosa Roxb. Ex. Link. Syn. *Vangueria spinosa* Hook. F. belongs to family Rubiaceae of Dicotyledonous angiosperm. This plant is a spinous, armed small tree or large shrub about 5-8 m tall. Branches are bushy, bark dark-colored, nearly smooth. Spines are axillary or supra axillary, straight, sharp, 5-40 mm. Leaves are membranous, ovate or elliptic-oblong, 4-15 cm × 2.5-5 cm, slightly pubescent above, densely pubescent beneath, apex bluntly or obtusely acuminate, margin entire, base rounded or acute; secondary nerves 7-9 pairs, tertiary nerves prominent below; petiole to 1 cm, densely hairy; stipules short, broad, very obtuse, caducuous. Flowers are crowded into fascicles and have shorter pedicels and petioles. Flowers are small, 4-5 merous, pale green or greenish white with faintly fragrant on short pedunculate axillary cymes, calyx 5 toothed corolla 5 lobes. Fruits are yellowish, subglobose drupe, smooth with persistent calyx lobes. Flowering time is in the month of April and May and while fruits appear in August to December (Bora and Kumar¹, Pullaiah², Sen³).

Medicinal plants are always identified by presence of rich sources of bioactive compounds widely used as potential drug for the treatment of various disease and WHO (World Health Organization) has also stated that the traditional medicines are safe remedies for the treatment of various health related problems (Divya and Mukesh⁴, WHO⁵). Now a day's the pharmaceutical companies are moving towards the formulation of the natural drugs from plant extracts because of side effect associated with synthetic drugs.

Meyna spinosa Roxb is a significant ethno medicinal plant widely distributed in India, Bangladesh, China, and Myanmar. The plant parts of *Meyna spinosa* has been consumed as traditional or folk medicine to treat different health-related problems such as diabetes, diphtheria, stomach pain, headache, liver problem, indigestion, throbbing urination, and skin problems such as pimples and acne problems.

The important phytochemicals of *Meyna spinosa* include phenolic compounds, tannins, flavonoids, alkaloids, saponins, phytosterols,

terpenoids, triterpenoids, oleanic acid (Chatterjee *et al.*⁶, Buragohain⁷, Gogoi and Sarma^{8,9}) which is used in the treatment of chronic diseases such as hepatic disorder, cancer, inflammation, diabetes, microbial infection, and also exhibit potent anti-oxidant activity (Ayelesoet *al.*¹⁰).

MATERIALS AND METHODS

The fresh stem and leaves of *Meyna spinosa* were collected from earthen pot grown plants and brought to the laboratory, washed with water and air-dried at room temperature for 7 days, and then oven-dried at 40°C to remove the residual moisture. The dried parts were powdered using a mixer grinder and stored in air-tight container for future use. Methanol was used as a solvent for extraction of plant parts. About 1 gm of the plant samples were added into the test tubes containing 5 ml of methanol, and were extracted at room temperature. The dry powder of the methanol extract was subjected to screening of active phytochemicals employing FTIR analysis. The tests were performed to check the presence or absence of bioactive compounds such as carbohydrate, protein, amino acids, phenol, tannins, saponins, and terpenoids.

The Fourier transform Infrared Spectrum (FTIR) analysis was performed (Perkin Elmer Spectrophotometer system, UK) for the methanol extracts of leaves and stem of *Meyna spinosa*. This technique was used to perceive the typical peaks and their functional groups. FTIR spectrophotometry is plausibly the most controlling tools for recognizing the kinds of chemical bonds (functional groups) present in compounds. The wavelength of light absorbed is unique to the chemical bond can be seen in the annotated spectrum. The chemical bonds of a molecule can be determined using the infrared absorption spectrum. The plant constituents of dried powder sample of methanol extract were used for FTIR investigation. For instrumentation analysis, dried powdered sample was used. To make translucent sample, 8 mg of methanol extract dried sample was condensed in 100 mg of KBr pellet. The active phytochemicals were identified by spectroscopy techniques and the results obtained have been presented in Figure-1, 2, 3 and 4; Table-1.

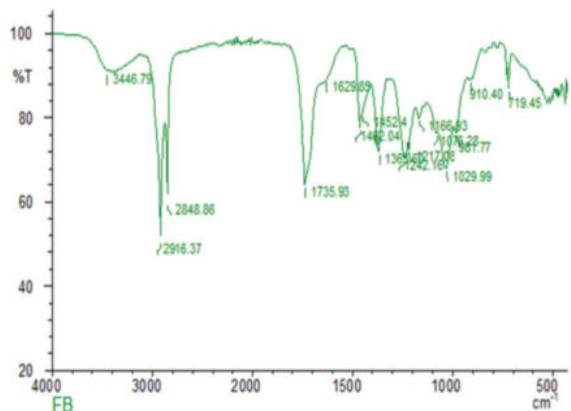


Figure-1: Fourier transform infrared spectrum of methanol extract of leaves of *Meyna spinosa*

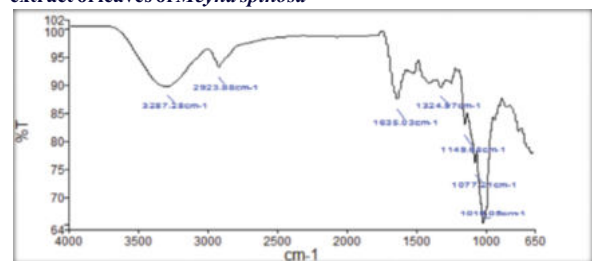


Figure-2: FTIR analysis of methanol extract of stem of *Meyna spinosa*

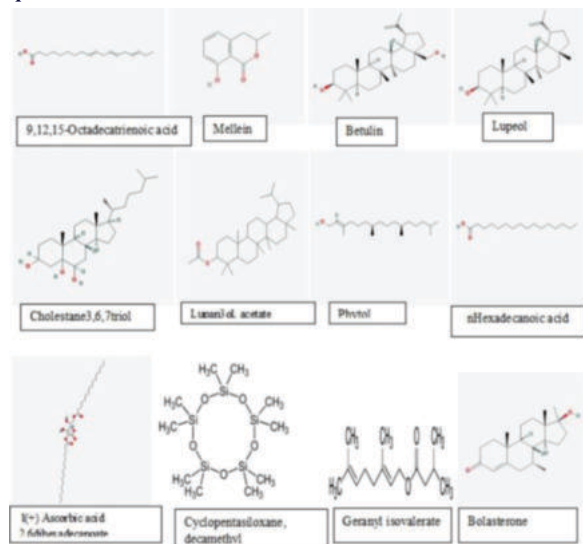


Figure-3: Molecular Structure of active phytochemicals identified in methanol extract of leaves of *Meyna spinosa*

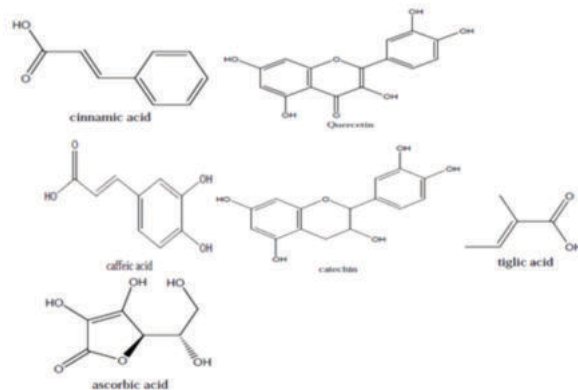
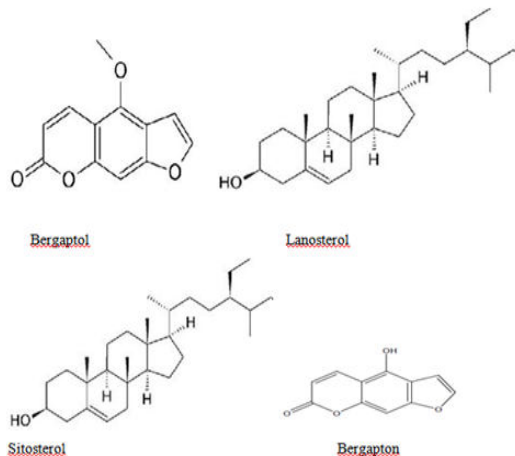


Figure-4: Phytochemicals detected in methanol extract of stem of *Meyna spinosa*

Table-1: Bioactive compounds detected in methanol extract of leaves and stem of *Meyna spinosa* and their known biological activities

Methanol extract of leaves				
Name of Compound	Nature of Compound	Retenti onTime	Molecular Formula	Biological Activities
9,12,15-Octadecatrienoic acid	Fatty acid	4.56	C ₂₇ H ₅₂ O ₄	Essential oil
Mellein	Phenol	14.65	C ₁₀ H ₁₀ O ₃	Asthma, coughs, tuberculosis, and related respiratory problems
Betulin	Triterpene	29.31	C ₃₀ H ₅₀ O ₂	Anti-inflammatory and anti-bacterial activity
Lupeol	Triterpene	29.31	C ₃₀ H ₅₀ O	Antiprotozoal, antimicrobial, antiinflammatory, antitumor and chemopreventive properties
Cholestane3,6,7triol,	Cholestone	27.80	C ₂₇ H ₄₈ O ₃	Anti-bacterial activity
Bolasterone	Steroids	25.53	C ₂₁ H ₃₂ O ₂	Muscle strength and mass
Oleic acid, 3(octadecyloxy)propyl ester	Fatty acid	25.89	C ₃₉ H ₇₆ O ₃	Restored the heart's
Phytol	Diterpenes	20.88	C ₂₀ H ₄₀ O	Antioxidant and anticancer
nHexadecanoic acid	Fatty acid	19.43	C ₁₆ H ₃₂ O ₂	Anti-inflammation
l(+)- Ascorbic acid		19.43	C ₃₈ H ₆₈ O ₈	Anti-bacterial activity
2,6dihexadecanoate				
Cyclopentasiloxane, decamethyl		19.84	C ₁₀ H ₃₀ O ₅ Si ₅	Biomedical application
Geranyl isovalerate	Monoterpe	16.79	C ₁₅ H ₂₆ O ₂	Anti-inflammatory, antioxidant and anti-viral activities
Methanol extract of stem				
Name of compounds				
Silicic acid, diethyl bis (trimethylsilyl) ester	Ester	24.28	C ₁₀ H ₂₈ O ₄ Si ₃	Not known

Cyclotrisiloxane, (hexamethylsilyl) ester	Ester	29.90	C ₆ H ₁₈ O ₃ Si ₃	Antibacterial activity
Gluanol acetate	Triterpin			
Tiglic acid esters of taraxasterol	Ester			
Lupeol acetate	Ester			
Friedelin (Hentriacontane)	Ester			
β-sitosterol	Ester			
Stigmasterol	Ester			
Aspartic protease	Peptide			
Lupeol ceryl behenate				
α-amyrin				
Lupenol		29.31		
Leucodyanidin-3-O-β-D-glucopyranoside				
Leucopelargonidin				
Leucopelargonidin-3-O-α-L-rhamnopyranoside				

RESULTS AND DISCUSSION

In the present investigation FTIR spectrum was used to analyze the functional groups of the phytochemical compounds present in the leaves and stem of *Meyna spinosa* (Fig-1). The results revealed the presence of four major peaks. The first major peak had the value of 2916 belongs to the functional group of aromatic (C-H). The second major peak had the value of 2848, which belongs to the primary, secondary amines and amides (N-H) group. The third major peak of 1735 belonged to the esters and saturated aliphatic group (C=O) and the last peak 1242 belonged to the group of aromatic amines (C-N).

When the methanol extract of stem of *Meyna spinosa* was passed into the FT-IR, the functional groups of the components were separated based on its peaks ratio. The results of FT-IR analysis confirmed the presence of N-H, O-H, C=C, C-H, C-O and CH₃ functional groups (Fig-2). The molecular structures of the identified phytochemicals in the methanol extract of leaves and stem of *Meyna spinosa* have been illustrated in Figure-3 and 4.

Methanol extract of leaves of *Meyna spinosa* revealed the presence of 12 bioactive compounds viz. 9,12,15-Octadecatrienoic acid, Mellein, Betulin, Lupeol, Cholestane3,6,7-triol, Bolasterone, Oleic acid, 3 (Octadecyloxy) propyl ester, Phytol, n Hexadecanoic acid, 1 (+) Ascorbic acid 2, 6 dihexadecanoate, Cyclopentasiloxane, decamethyl, Geranyl isovalerate (Table-1). The mass spectrometer analyses the compounds eluted at different times to identify the nature and structure of the compounds. These mass spectra are the fingerprint of that compounds giving rise to appearance of peaks at different m/z ratio, which can be identified from the library search (Kalimthi and Prabakaran¹¹).

The stem of *Meyna spinosa* revealed the presence of sterols, triterpenoids (Lanosterol) and alkaloids, tannins and falconoid, gluanol acetate, β-sitosterol, leucodyanidin-3-O-β-D-glucopyranoside, leucopelargonidin, leucopelargonidin-O-α-L-rhamnopyranoside, lupeol ceryl behenate, lupeol acetate and α-amyrin acetate, lupenol, β-sitosterol and stigmasterol, gluanol acetate, glucose, tiglic acid esters of taraxasterol, lupeol acetate, friedelin higher hydrocarbons (Hentriacontane) and other phytosterols. A new tetra cyclic triterpene gluanol acetate was isolated from the leaves. An unusual thermos table aspartic protease was isolated from stem extract of *Meyna spinosa* (Table-1). The preliminary phytochemical analysis of *Meyna spinosa* was also tested and showed the presence of various bioactive compounds according to the standard procedure (Arun Kumar *et al.*¹², Trease and Evans¹³, Yadav and Agarwal¹⁴, Pashupati *et*

*al.*¹⁵). The present study contributes valuable information of bioactive compounds in stem and leaves of *Meyna spinosa*. The present findings gain support from the work of Rekha bora *et al.*¹⁶, Senguttuvan *et al.*¹⁷, Wang¹⁸, Teixeira *et al.*¹⁹ and Pashupati Yadav *et al.*⁵ who also observed more or less similar active phytochemicals in leaves and stem of *Meyna spinosa*.

The presences of active biological molecules in methanol extracts of leaves and stem of *Meyna spinosa* have the potential anti-inflammation, antimicrobial and anti-oxidant, anti ulcer, anti cancer, free radical scavenging activity, hepato protective, cytotoxicity, anti melanogenic, anti-helminthic, wound healing and anticonvulsant activities.

CONCLUSIONS:

From the present observation it can be concluded that the knowledge and usage of conventional medicine for the treatment of various diseases among the rural and urban communities is still a major part of their life and culture. People of Indo-Gangetic plain have a strong faith in the efficacy and success of traditional medicine and the results of the present study provide evidence that the active phytochemicals of *Meyna spinosa* continued to play a vital role in the healthcare system.

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Conflict of Interest: Authors declare no conflict of interest directly or indirectly.

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