

EVALUATION OF PHYTOCHEMICAL POTENTIAL OF *AVICENNIA MARINA* BY HISTOCHEMISTRY

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ABSTRACT

Avicennia marina (Forssk.) Vierh. is commonly known as the grey or white mangrove. It is a vital species within mangrove ecosystems, renowned for its ecological significance and potential medicinal applications. *A. marina* has been traditionally utilized in various cultures, it has been employed in Persian folk medicine for treating smallpox lesions. Scientific investigations of methanol extracts of *A. marina* demonstrated significant inhibitory activity against Herpes Simplex Virus (HSV) and moderate activity against Human immunodeficiency Virus (HIV). Additionally, the plant exhibits anti-glycation properties, which are beneficial in managing diabetes and Alzheimer's disease. Ecologically, *A. marina* plays a crucial role in coastal protection, sediment stabilization, and serving as a nursery for various marine species. For current research work, leaves of the *Avicennia marina* plant were collected from the Thane Creek area which is presently facing a high degree of pollution caused by domestic as well as industrial waste. Therefore detailed phytochemical and heavy metal detection had been carried out by histochemical study of leaves. The study revealed the presence of almost all phytochemicals and its storage site i.e. different leaf tissue. Safety to use leaf tissue as medicine was also ensured by absence of heavy metals. The presence of different secondary metabolites in the said plant indicates the high medicinal potential of leaves of the plant for prospective drug development.

KEYWORDS: *Avicennia marina*, histochemical tests, secondary metabolites, heavy metals.

INTRODUCTION

Mangrove forests found in the inter-tidal zones of the tropical and subtropical regions of the world represent a rich biodiversity of microorganisms, plants and animals. Among these, plants play a vital role by synthesizing essential

nutrients to support ecosystem; acting as breeding ground for various animals and minimizing coastal erosion effect.^[1] Very often, secondary metabolites which are non-essential for primary metabolism, are abundantly produced by mangrove plants due to harsh conditions such as high salinity and tidal fluctuation. These metabolites includes phenolics (serve as antioxidants and antimicrobials), terpenes (antimicrobial and antifungal compounds), alkaloids (exhibit antimicrobial and insecticidal activities and flavonoids (provide antioxidant and anti-inflammatory benefits).^[2,3,4]

Avicennia marina (family - Acanthaceae) is one of the most widespread mangroves across the Indo-West Pacific and has long-standing ethnomedicinal use. It is medium size tree which produces distinctive system of pneumatophore or breathing roots. The leaves show presence of salt glands; excrete excess of salts from plants. Modern studies indicate presence of diverse phytochemical classes found in the species (e.g., phenolics, flavonoids, terpenoids, alkaloids, steroids/triterpenes). Recent review of pharmacology encompasses antimicrobial, antioxidant, anti-inflammatory, antidiabetic and anticancer domains.^[5,6,7] Complementing its medicinal promise, *A. marina* is increasingly recognized as a bioindicator and phytoremediator in polluted coastal environments. Studies in Visakhapatnam, India, have reported that leaf tissues of *A. marina* accumulate heavy metals—such as Cr, Pb, and Cd—resulting in anatomical modifications including thicker cuticles and expanded mesophyll regions, especially in areas impacted by industrial pollution.^[8] In parallel, investigations along Egypt's Red Sea coast found that plants growing in metal-contaminated zones display elevated concentrations of antioxidant enzymes (e.g., catalase, peroxidase) and secondary metabolites (phenols, flavonoids, tannins), suggesting an adaptive ecological response to oxidative stress.^[9]

Phytochemical research reinforces the medicinal promise of *A. marina*. Ethanolic leaf extracts have tested positive for a wide range of secondary metabolites—including alkaloids, flavonoids, phenolics, saponins, tannins, glycosides, and triterpenoids—confirmed via qualitative screening and GC-MS profiling; these compounds often underlie its antimicrobial, antioxidant, anti-inflammatory, and hepatoprotective activities.^[10] A more recent study from Pichavaram, India, leveraging LC-MS, NMR, and GC-MS techniques, identified 52 distinct compounds (e.g., pentanoic acid, thiazolidinones, arabinopyranosides), with diverse bioactivities ranging from antioxidative and anticancer to anti-aging effects.^[11]

Although many studies confirm the presence of beneficial secondary metabolites and the ecological resilience of *A. marina*, specific histochemical investigations on leaf tissue—especially regarding the localization of phytochemicals in leaf anatomy and heavy metal presence or absence remain limited. This gap is especially pertinent in polluted sites like Thane Creek, Mumbai, where domestic and industrial effluents threaten mangrove health.

In light of these insights and gaps, the current study focuses on leaves of *A. marina* collected from the Thane Creek area, subject to significant contamination. It employs histochemical methods to elucidate the presence and tissue localization of phytochemicals and to assess heavy metal contamination in leaf tissues. The study's findings—indicating both widespread phytochemical presence and absence of heavy metals—provide compelling evidence of the medicinal potential and safety of *A. marina* leaves, reinforcing the species's dual ecological and therapeutic value.

MATERIALS AND METHODS

The plant of *Avicennia marina* was first authenticated from standard herbarium available at Blatter Herbarium at St. Xavier's College, Fort. Fresh leaves of *Avicennia marina* brought from mature plants growing along banks of Thane

creek. Leaves were washed thoroughly and dried at room temperature. Macroscopical characters were listed and further leaves were subjected to microscopy. Different histochemical tests and heavy metal tests were done by following standard procedures and chemical reagents.^[12, 13,14,15] The observations are depicted in following tables.

OBSERVATIONS

Table 1: Macroscopy of Leaf.

Sr. No	Parameters	<i>A. marina</i>
1.	Type	Simple
2.	Phyllotaxy	Opposite, Decussate
3.	Size Length in cm Breadth in cm	7.7 3.5
4.	Petiole	Cylindrical, solid
6.	Stipule	Exstipulate
7.	Peculiarities	Fleshy, lower surface pubescent



Photoplate 1: Leaves of *Avicennia marina*.



Photoplate 2: Adaxial & Abaxial surface of leaf.

Microscopy of Leaf Lamina *A. marina*

- **Upper epidermis** – It is uniseriate; cuticularized, cuticle smooth and moderately thick; composed of tangentially elongated cells devoid of epidermal hair.
- **Mesophyll** - is made up of 3-4 layers of palisade cells and 9-11 layers of spongy tissue with intermittent vascular bundles which has xylem on top side and phloem on bottom.
- **Lower Epidermis** - It is single layered; cuticularized; consists of isodiametric to tangentially elongated cells with unicellular trichomes and stomata.

Microscopy of Leaf Midrib *A. marina*

- **Upper epidermis** – It is biseriate; cuticularized, cuticle smooth and moderately thick; composed of tangentially elongated cells with epidermal hairs.
- **Mesophyll** - is made up of 3-4 layers of palisade cells and 9-11 layers of spongy tissue with intermittent vascular bundles which has xylem on top side and phloem on bottom.
- **Vascular tissue** - at centre; composed of xylem on top side and phloem on bottom.
- **Ground tissue** - present towards lower epidermis; made up of 8-10 layers of parenchyma.
- **Lower Epidermis** - It is single layered; cuticularized; consists of isodiametric to tangentially elongated cells with unicellular trichomes and stomata.

Table 2: Histochemistry of Leaf.

Sr. No.	Plant constituent test	<i>A. marina</i>	Region	Tissue
1.	Test for Cellulose	+	Midrib	Parenchyma
2.	Test for Starch	+	Midrib & Lamina	Parenchyma
3.	Test for Pectin	+	Lamina	Parenchyma
4.	Test for Lignin	+	Lamina	Parenchyma
5.	Test for Proteins a) Biuret test	+	Midrib	Parenchyma
6.	Test for Lipids	+	Lamina	Parenchyma
7.	Test for Alkaloids a) Mayer's reagent	-	-	-
8.	Test for Tannins a) FeCl ₃ test	+	Midrib	Parenchyma
9.	Test for Glucosides	+	Midrib	Parenchyma
10.	Test for Saponins	+	Midrib	Parenchyma
11.	Test for Mucilage	+	Lamina	Parenchyma
12.	Test for Calcium-oxalate crystals a) H ₂ SO ₄ test	+	Midrib	Parenchyma
13.	Test for Diosmin	+	Midrib	Parenchyma

Key: '+' Present, '-' Absent

Table 3: Heavy Metal Studies.

Sr. No.	Heavy Metal test	<i>A. marina</i>
1.	Test for Lead	-
2.	Test for Cadmium	-
3.	Test for Mercury	-
4.	Test for Arsenic	-

Key: '+' Present, '-' Absent

RESULTS AND CONCLUSION

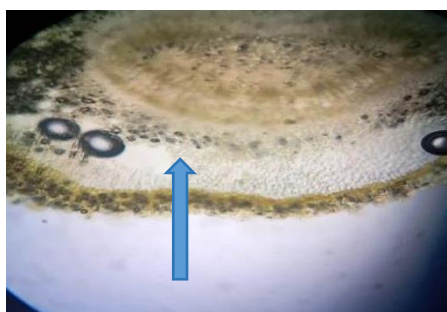
Avicennia marina plant was found to be storehouse of various phytochemicals. The study revealed histochemical localization of various secondary metabolites. Phytochemicals are prevalently stored in parenchymatous tissues of leaf midrib. Diosmin which is a derivative of flavonoid was major highlight of investigation. Creek water pollution does not lead to accumulation of heavy metals in leaf tissue. In microscopy of leaf, abnormal anatomical feature was not observed as result of pollution stress or adaptive strategy by plant. However detail quantitative phytochemistry can be undertaken as further study so that leaf tissue can be used in preparation of herbal remedies.



Photoplate 3: T.S. of Leaf Midrib.



Photoplate 4: T.S. of Leaf Lamina.



Photoplate 5: Calcium Oxalate Crystals.

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Conflicts of Interest - The authors declare that there are no conflict of interests.

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