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Phytochemical profiling and antimicrobial activity of *Morinda pubescens* Fruit

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Abstract

Morinda pubescens is a traditionally used medicinal plant from Southern Asia. This study evaluated its phytochemical profile, antimicrobial activity, and chemical composition via GC-MS analysis. Various solvent extracts revealed key bioactive compounds, including carbohydrates, alkaloids, and flavonoids. Antimicrobial assays demonstrated that the aqueous extract exhibited notable inhibition against *Escherichia coli* and *Staphylococcus aureus*. These findings substantiate the plant's traditional medicinal use and its potential for pharmaceutical development.

Keywords: *Morinda pubescens*, GC-MS, phytochemistry

Introduction

Due to the availability of certain bioactive components that are used by the pharmaceutical industry, plants are utilized as medicine in a variety of cultures and are a source of powerful pharmaceuticals.

Different phytochemicals, sometimes referred to as secondary metabolites, are found in plants. Because of their unique, complementary, or synergistic effects on health, phytochemicals can be helpful in the treatment of specific illnesses.

In the pharmaceutical sector, phytochemicals are essential for the creation of new medications and therapeutic agents.

The plant known as *Morinda pubescens* or *Morinda coria* was found by J.E. Smith and belongs to the Rubiaceae family. It is widely distributed throughout Southern Asia, showing a complete migration from the western to the eastern slopes of the Ghats, and it is primarily found in the dry regions of Southern Deccan and Southern Travancore.

Plantlist.org states that *Morinda pubescens* is the unresolved name for this plant, which was subsequently recognized as a synonym for *Morinda pubescens*. *Morinda tinctoria* (Roxb) and *Morinda tomentosa* (Roxb), among other scientific names, have been reduced to synonymy^[1].

Known by various names, such as Morinda tree, Brimstone tree, or wild ach root, this plant is commonly encountered. Indigenous people have traditionally used the entire plant, especially the leaves and roots, to treat a wide range of illnesses and ailments.

Topical applications for wounds, inflammation, gout, and stomach pains are common, as are oral administrations for conditions like diabetes, malaria, fever, rheumatism, and infectious diseases.

Numerous *in vitro* and *in vivo* investigations offered varied proof endorsing the majority of *Morinda pubescens*' conventional medicinal claims. Using *Morinda pubescens* extracts, bioassay-guided phytochemical screening revealed a wealth of bioactive compounds with notable medicinal potential, including flavonoids, alkaloids, and phenolic acids.

Apart from its widespread usage in traditional medicine, *Morinda pubescens* has many other potential uses, the well-researched of which are as an adsorbent for green dye and as green manure.

Materials and Methods

Chemicals

All the chemicals and reagents used for the research were of analytical grade.

Morinda pubescens plant was selected for the study (Figure 1). The *Morinda pubescens* was collected from the village Ralegan Siddhi, Tahsil Parner, Ahmednagar, Maharashtra, India.

The plant was authenticated as *Morinda pubescens* (Family: Rubiaceae) by the Botanical Survey of India, Western Regional Centre.



Fig 1: *Morinda pubescens* plant

Drying and Pulverizing

After obtaining the *Morinda pubescens*, they were dried for four weeks in the shade. To prevent microbiological contamination, ongoing monitoring was done. To create a fine powder, the dried plant materials were extracted and processed. To get even smaller particles, the powder was passed through a 2 mm sieve once more.

Until they were required for the investigation, the powdered samples were kept in a clean glass container at a low temperature.

Extraction of Plant Material

Aqueous Extract

Morinda pubescens was dried, ground, and placed in a 250 g sample container. The sample was macerated in water for seven days. Following that, the marc was removed by filtering, and then the marc was pressed. The solvent was evaporated using a rotating solvent evaporator. In the end, the extract was weighed, and its proportion of the plant material's air-dried weight was determined.

Hydroalcoholic Extract

Morinda pubescens fruit was dried, ground, and placed in a 250 g sample container. The sample was macerated in a water-ethanol mixture (6:4) for seven days. Following that, the marc was removed by filtering, and then the marc was pressed. The solvent was evaporated using a rotating solvent evaporator. In the end, the extract was weighed, and its proportion of the plant material's air-dried weight was determined [11].

Chloroform Extract

Morinda pubescens fruit was dried, ground, and placed in a 250 g sample container. The sample was macerated in chloroform for seven days. Following that, the marc was removed by filtering, and then the marc was pressed. The solvent was evaporated using a rotating solvent evaporator. In the end, the extract was weighed, and its proportion of the plant material's air-dried weight was determined.

Butanol Extract

Morinda pubescens fruit was dried, ground, and placed in a 250 g sample container. The sample was macerated in ethanol for seven days. Following that, the marc was removed by filtering, and then the marc was pressed. The

solvent was evaporated using a rotating solvent evaporator. In the end, the extract was weighed, and its proportion of the plant material's air-dried weight was determined [12].

Phytochemical Screening of Extract

The *Morinda pubescens* fruit extract was observed for the identification of various phytoconstituents by carrying out phytochemical tests.

Screening of Antimicrobial Activity of *Morinda pubescens* Extracts

The antimicrobial activity of *Morinda pubescens* aqueous, hydroalcoholic, chloroform, and butanol extracts was tested using bacterial strains *E. coli* and *S. aureus*. The pour plate assay was used. By autoclaving at 120°C (15 psi), the medium was made sterile. The zone of inhibition was checked for *Morinda pubescens* extract [13].

Sample Preparation

The lyophilized powder was made soluble in *n*-hexane and ethanol, and the GC-MS analysis of the compounds was performed.

1.0 g of the lyophilized powdered *Morinda pubescens* was soaked in 10 ml of ethanol and *n*-hexane separately for 12 hours. The extract was vortexed and filtered.

1.0 µl of this filtered solution was injected for GC-MS analysis.

Mass Spectrometry-Gas Chromatography (GC-MS) Analysis

GC-MS analysis of the extract of *Morinda pubescens* fruit was performed using a Thermo Scientific TRACE Ultra Gas Chromatograph interfaced to a TSQ Quantum Ultra™ Triple Quadrupole Mass Spectrometer (GC-MS) equipped with Trace GOLD TG-5MS GC Columns (5% diphenyl, 95% dimethylpolysiloxane) fused capillary column (30 × 0.25 µm ID × 0.25 µm df).

Helium (99.999%) was used as the carrier gas with a flow rate of 1 ml/min.

The splitless mode was used. An aliquot of 1 µl of the sample solution was injected into the column. The oven temperature was programmed from 80 °C (isothermal for 0.0 min), with an increase of 5.0 °C/min, to 300 °C, ending with an isothermal hold at 300°C for 2.0 min.

The injector and auxiliary temperatures were set at 300 °C and 280 °C, respectively. The ion source temperature was maintained at 225 °C.

The mass spectrum of compounds in the samples was obtained by electron ionization at 70 eV, and the detector was operated in scan mode from 35-500 m/z. The total running time was 46 minutes.

The relative percentage amount of each component was calculated by comparing its average peak area to the total peak areas [14].

Identification of Compounds

The National Institute of Standards and Technology (NIST) database was used to analyze the mass spectra and identify the components based on their retention indices. The database contains more than 62,000 patterns of recognized compounds.

The standard mass spectra of known components stored in the NIST library (NIST II) were compared to the acquired spectra of the unknown components in the *Morinda pubescens* fruit fraction.

Results and Discussion

Table 1: Preliminary Phytochemical Screening of various Extracts of *Morinda pubescens* fruit

Sr. No	Phytochemicals	Inference			
		Aqueous Extract of <i>Morinda pubescens</i> fruit	Hydro alcoholic Extract of <i>Morinda pubescens</i> fruit	Chloroform Extract of <i>Morinda pubescens</i> fruit	Butanol Extract of <i>Morinda pubescens</i> fruit
1.	Carbohydrates	+	-	+	+
2.	Proteins	+	-	-	-
3.	AminoAcids	-	+	-	+
4.	Flavonoids	+	+	-	-
5.	Alkaloids	+	+	+	-
6.	Terpenoids	+	-	-	-
7.	Saponins	-	-	-	+
8.	Tannins	+	+	-	-
9.	Glycoside	+	-	-	+

Preliminary Phytochemical Screening

Carbohydrates, proteins, alkaloids, flavonoids, glycosides, terpenoids, phenols, and tannins are all present in the aqueous extract of *Morinda pubescens* fruit, according to the preliminary phytochemical screening (Table 1). Amino acids and saponins are absent in the aqueous extract of *Morinda pubescens* fruit.

Amino acids, flavonoids, alkaloids, and tannins are present in the hydroalcoholic extract of *Morinda pubescens* fruit.

Carbohydrates and alkaloids are present in the chloroform extract of *Morinda pubescens* fruit.

Carbohydrates, amino acids, and saponins are present in the butanol extract of *Morinda pubescens* fruit (Table 1).

Preliminary Antimicrobial Activity of *Morinda pubescens* Extract

The antimicrobial activity of *Morinda pubescens* fruit extract was evaluated. Different extracts of *Morinda pubescens* fruit were tested for antimicrobial activity using the pour plate method.

The diameter of the zone of inhibition against various Gram-positive and Gram-negative bacteria was found to be 20 mm for *E. coli* and 16 mm for *S. aureus*.

The results indicate that all extracts of *Morinda pubescens* fruit have antimicrobial activity.

Table 2: Preliminary Antimicrobial Activity of Aqueous, Hydroalcoholic, Chloroform, and Butanol Extract of *Morinda pubescens* Fruit

Microorganisms	Zone of inhibition in mm				
	<i>Morinda coriea</i> Buch Ham fruit Aqueous extract 100 µg/ml	<i>Morinda coriea</i> Buch Ham fruit Hydroalcoholic extract 100 µg/ml	<i>Morinda coriea</i> Buch Ham fruit Chloroform extract 100 µg/ml	<i>Morinda coriea</i> Buch Ham fruit Butanol extract 100 µg/ml	Standard 100 µg/1ml
<i>E. coli</i>	22	15	16	07	21
<i>S. aureus</i>	16	18	17	15	18

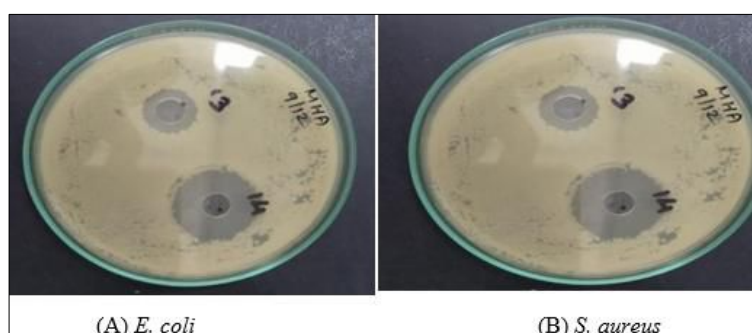


Fig 1: Antimicrobial Activity of Various Extracts of *Morinda pubescens* Fruit (A) *E. coli* (B) *S. aureus*

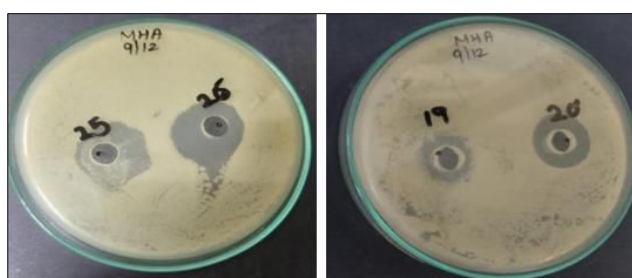
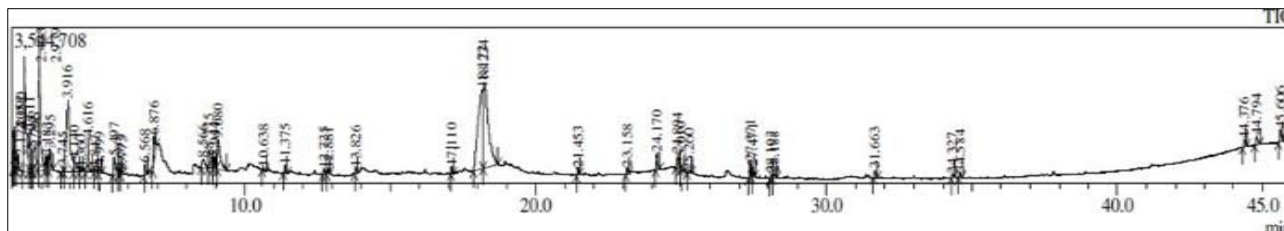


Fig 2: Antimicrobial Activity of Various Extracts of *Morinda pubescens* Fruit on *E. coli*

GC-MS Analysis

The GC-MS chromatogram of the standardized extract of *Morinda pubescens* fruit (Figure 2) showed various major peaks, indicating the presence of different phytochemical constituents.

On comparison of the mass spectra of the constituents with the NIST library, the different phytochemicals were characterized and identified. The spectrum of the unknown component was compared with the spectrum of the component stored in the NIST library, and the results are tabulated.



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