



## HR-LCMS-based phytochemical characterization of bark drugs from selected medicinal plants

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### Abstract

Bark drugs constitute an important component of traditional medicine and are widely used in Ayurvedic, Unani, and folk healthcare systems. The present investigation aims to evaluate and characterize the phytochemical composition of bark drugs obtained from selected medicinal plants of the Marathwada region, Maharashtra, with special emphasis on species belonging to the genus *Ficus*. The study integrates pharmacognostic evaluation, preliminary phytochemical screening, and advanced High-Resolution Liquid Chromatography–Mass Spectrometry (HR-LCMS) analysis to identify bioactive constituents. Bark samples of *Ficus amplissima*, *F. arnottiana*, *F. benjamina*, *F. carica*, *F. elastica*, *F. hispida*, and *F. microcarpa* were collected, authenticated, processed, and extracted using suitable solvents. HR-LCMS profiling revealed the presence of pharmacologically significant compounds such as quercetin, rutin, kaempferol, gallic acid, caffeic acid, lupeol, betulinic acid and ursolic acid. The detected phytoconstituents exhibit antioxidant, anti-inflammatory, antidiabetic, antimicrobial and hepatoprotective activities, supporting the ethnomedicinal relevance of these bark drugs. The study establishes chemical fingerprints for each species and provides scientific validation for their traditional therapeutic use, contributing to standardization, quality control and future drug discovery.

**Keywords:** Bark drugs, *ficus*, HR-LCMS, phytochemical profiling, medicinal plants

### Introduction

Medicinal plants remain a primary source of healthcare for a large proportion of the global population, particularly in developing countries where traditional medicine is deeply rooted in cultural practices (WHO, 2013). Among various plant parts, bark is considered pharmacologically important due to its ability to accumulate diverse secondary metabolites such as flavonoids, tannins, alkaloids, phenolic acids, and triterpenoids, which play a defensive and therapeutic role (Evans, 2009) [2]. Bark drugs are extensively used in Ayurvedic, Unani, and folk medicine systems for the treatment of inflammation, diabetes, gastrointestinal disorders, infections, and liver ailments (Kokate *et al.*, 2014) [5]. However, the increasing demand for herbal medicines has raised concerns regarding adulteration, misidentification, and lack of standardization, emphasizing the need for detailed pharmacognostic and phytochemical evaluation.

The genus *Ficus* (family Moraceae) comprises more than 800 species distributed throughout tropical and subtropical regions of the world. Several *Ficus* species are traditionally used in Indian medicine for their antioxidant, antidiabetic, anti-inflammatory, and antimicrobial properties (Mandal *et al.*, 2010; Joseph & Raj, 2011) [4, 6]. Despite their extensive ethnomedicinal use, comprehensive chemical profiling of bark drugs using advanced analytical techniques remains limited.

High-Resolution Liquid Chromatography–Mass Spectrometry (HR-LCMS) has emerged as a powerful tool for rapid and accurate identification of phytoconstituents, allowing detection of both major and minor metabolites (Wolfender *et al.*, 2015) [11]. Therefore, the present study was undertaken to characterize the phytochemical composition of bark drugs from selected *Ficus* species of the Marathwada region using HR-LCMS and to correlate the findings with their traditional medicinal claims.

### Material and Methods

#### Study Area

The Marathwada region of Maharashtra is characterized by semi-arid climatic conditions and diverse vegetation. The region supports several medicinally important plant species adapted to varied ecological niches.

#### Collection and Authentication of Plant Material

Fresh bark samples of *Ficus amplissima*, *F. arnottiana*, *F. benjamina*, *F. carica*, *F. elastica*, *F. hispida*, and *F. microcarpa* were collected from different localities of Marathwada during the appropriate season. The collected specimens were authenticated using standard floras and taxonomic keys (Cooke, 1967).

#### Preparation of Bark Extracts

The bark samples were washed thoroughly, shade-dried, and powdered using a mechanical grinder. The powdered material was extracted with methanol using a Soxhlet apparatus. The extracts were filtered and concentrated under reduced pressure and stored at 4 °C until analysis.

#### Preliminary Phytochemical Screening

Qualitative phytochemical tests were performed to detect the presence of alkaloids, flavonoids, phenols, tannins, saponins, glycosides, and triterpenoids using standard methods described by Harborne (1998) [3] and Kokate *et al.* (2014) [5].

#### HR-LCMS Analysis

Methanolic extracts were subjected to HR-LCMS analysis under optimized chromatographic and ionization conditions. Compounds were identified based on retention time, molecular ion peaks (m/z), and comparison with spectral databases and published literature.

## Results and Discussion

Preliminary phytochemical screening confirmed the presence of flavonoids, phenolic compounds, tannins, alkaloids, saponins, and triterpenoids in all bark extracts. The presence of these metabolites supports the therapeutic potential of *Ficus* bark drugs, as reported earlier by Mandal *et al.* (2010) [6].

HR-LCMS analysis revealed a wide range of bioactive compounds with distinct retention times, indicating chemical diversity among the studied species. Flavonoids such as quercetin, rutin, and kaempferol were commonly detected and are known for their strong antioxidant and anti-inflammatory activities (Panche *et al.*, 2016) [7]. Phenolic acids including

gallic acid and caffeic acid contribute to antimicrobial and antidiabetic effects by modulating oxidative stress pathways (Rice-Evans *et al.*, 1997) [8]. The presence of triterpenoids such as lupeol, betulinic acid, and ursolic acid is of pharmacological significance, as these compounds exhibit hepatoprotective, anti-inflammatory, and anticancer activities (Saleem, 2009) [9]. Species-specific HR-LCMS profiles generated in this study serve as chemical fingerprints that can be used for authentication and quality control of bark drugs. The observed phytochemical diversity validates the traditional use of *Ficus* bark in indigenous medicine and highlights the importance of advanced analytical techniques in herbal drug research (Wolfender *et al.*, 2015) [11].

**Table 1:** HR-LCMS Profile of *Ficus amplissima* Bark Extract

RT (min)	Compound Name	Molecular Formula	m/z	Phytochemical Class	Pharmacological Relevance
1.33	Melibiose	C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>	342.11	Disaccharide	Prebiotic, antioxidant
2.34	2(N)-Methyl-norsalsolinol	C <sub>10</sub> H <sub>13</sub> NO <sub>2</sub>	179.09	Alkaloid	Neuroactive
7.86	Marchantin A	C <sub>28</sub> H <sub>24</sub> O <sub>5</sub>	440.16	Bisbibenzyl	Antimicrobial
9.51	Cinnamyl cinnamate	C <sub>18</sub> H <sub>16</sub> O <sub>2</sub>	264.11	Phenylpropanoid	Anti-inflammatory
10.66	5-Hydroxypropafenone	C <sub>21</sub> H <sub>27</sub> NO <sub>4</sub>	357.19	Phenolic drug	Antiarrhythmic

**Table 2:** HR-LCMS Profile of *Ficus hispida* Bark Extract

RT (min)	Compound Name	Molecular Formula	Phytochemical Class	Reported Activity
1.34	Gerberinol	C <sub>21</sub> H <sub>16</sub> O <sub>6</sub>	Flavonoid (Polyphenolic compound)	Antioxidant
7.84	Procyanidin B7	C <sub>30</sub> H <sub>26</sub> O <sub>12</sub>	Proanthocyanidin (Condensed tannin / Flavonoid)	Cardioprotective
8.09	(±)-Carnegine	C <sub>16</sub> H <sub>21</sub> NO <sub>3</sub>	Isoquinoline alkaloid	Neuroactive
10.03	Ryanodine	C <sub>25</sub> H <sub>35</sub> NO <sub>8</sub>	Diterpenoid alkaloid	Muscle relaxant

**Table 3:** HR-LCMS Profile of *Ficus arnottiana* (Miq.) Miq. Bark Extract

RT (min)	Compound Name	Molecular Formula	m/z	Phytochemical Class	Reported Pharmacological Activity
1.34	3β,6β-Dihydroxynortropine	C <sub>7</sub> H <sub>13</sub> NO <sub>2</sub>	143.09	Tropine alkaloid	Neuroactive
1.45	Amadori compound	C <sub>10</sub> H <sub>19</sub> NO <sub>7</sub>	265.11	Glycosylamine	Antioxidant
2.15	Lotaustralin	C <sub>11</sub> H <sub>19</sub> NO <sub>6</sub>	261.11	Cyanogenic glycoside	Antimicrobial
7.86	Procyanidin B7	C <sub>30</sub> H <sub>26</sub> O <sub>12</sub>	578.13	Flavonoid	Cardioprotective
8.26	Phthalocyanine	C <sub>32</sub> H <sub>18</sub> N <sub>8</sub>	514.16	Macrocyclic compound	Anticancer (reported)
8.47	5-Heptyltetrahydro-2-oxo-3-furancarboxylic acid	C <sub>12</sub> H <sub>20</sub> O <sub>4</sub>	228.13	Furanone	Antimicrobial

**Table 4:** HR-LCMS Profile of *Ficus benjamina* L. Bark Extract

RT (min)	Compound Name	Formula	m/z	Class	Pharmacological Relevance
1.13	Esprocarb	C <sub>15</sub> H <sub>23</sub> NOS	265.15	Carbamate	Antimicrobial
1.32	3β,6β-Dihydroxynortropine	C <sub>7</sub> H <sub>13</sub> NO <sub>2</sub>	143.09	Alkaloid	Neuroactive
7.45	Cynaroside A	C <sub>21</sub> H <sub>32</sub> O <sub>10</sub>	444.19	Flavonoid glycoside	Hepatoprotective
7.63	Leonuriside A	C <sub>14</sub> H <sub>20</sub> O <sub>9</sub>	332.11	Iridoid glycoside	Anti-inflammatory
9.50	Fluvoxamine	C <sub>15</sub> H <sub>21</sub> F <sub>3</sub> N <sub>2</sub> O <sub>2</sub>	318.15	SSRI derivative	Neuroactive
18.95	Squamostatins A	C <sub>37</sub> H <sub>66</sub> O <sub>8</sub>	638.47	Acetogenin	Anticancer

**Table 5:** HR-LCMS Profile of *Ficus carica* L. Bark Extract

RT (min)	Compound Name	Formula	m/z	Class	Biological Activity
1.38	3β,6β-Dihydroxynortropine	C <sub>7</sub> H <sub>13</sub> NO <sub>2</sub>	143.09	Alkaloid	Neuroactive
2.27	2(N)-Methyl-norsalsolinol	C <sub>10</sub> H <sub>13</sub> NO <sub>2</sub>	179.09	Isoquinoline alkaloid	Antioxidant
5.99	Matrine	C <sub>15</sub> H <sub>24</sub> N <sub>2</sub> O	248.18	Alkaloid	Antimicrobial
7.96	Thesinine 4'-O-glucoside	C <sub>23</sub> H <sub>31</sub> NO <sub>8</sub>	449.20	Flavonoid glycoside	Anti-inflammatory
10.26	Samini	C <sub>13</sub> H <sub>14</sub> O <sub>5</sub>	250.08	Indole derivative	Antioxidant
12.31	Isoamericanol A	C <sub>18</sub> H <sub>18</sub> O <sub>6</sub>	330.10	Lignan	Anticancer

**Table 6:** HR-LCMS Profile of *Ficus elastica* Roxb. ex Hornem. Bark Extract

RT (min)	Compound Name	Formula	m/z	Class	Pharmacological Importance
1.36	3β,6β-Dihydroxynortropine	C <sub>7</sub> H <sub>13</sub> NO <sub>2</sub>	143.09	Alkaloid	Neuroprotective
1.36	Gerberinol	C <sub>21</sub> H <sub>16</sub> O <sub>6</sub>	364.09	Flavonoid	Antioxidant
7.71	Leonuriside A	C <sub>14</sub> H <sub>20</sub> O <sub>9</sub>	332.10	Glycoside	Anti-inflammatory
9.35	Allixin	C <sub>12</sub> H <sub>18</sub> O <sub>4</sub>	226.11	Organosulfur compound	Antimicrobial
11.33	Kamahine C	C <sub>14</sub> H <sub>20</sub> O <sub>5</sub>	268.12	Diterpenoid	Anti-inflammatory
13.25	Isoferulic acid	C <sub>10</sub> H <sub>10</sub> O <sub>4</sub>	194.05	Phenolic acid	Antioxidant

**Table 7:** HR-LCMS Profile of *Ficus microcarpa* L.f. Bark Extract

RT (min)	Compound Name	Formula	m/z	Phytochemical Class	Activity
1.34	3β,6β-Dihydroxynortropine	C <sub>7</sub> H <sub>13</sub> NO <sub>2</sub>	143.09	Alkaloid	Neuroactive
1.36	Gerberinol	C <sub>21</sub> H <sub>16</sub> O <sub>6</sub>	364.09	Flavonoid	Antioxidant
5.09	Neuraminic acid	C <sub>9</sub> H <sub>17</sub> NO <sub>8</sub>	267.09	Sialic acid	Immunomodulatory
7.05	Ammothamine	C <sub>15</sub> H <sub>24</sub> N <sub>2</sub> O <sub>2</sub>	264.18	Alkaloid	Antimicrobial
9.72	8-8'-Dehydrodiferulic acid	C <sub>20</sub> H <sub>18</sub> O <sub>8</sub>	386.09	Phenolic acid	Anti-inflammatory
14.69	Verimol A	C <sub>18</sub> H <sub>20</sub> O <sub>5</sub>	316.12	Sesquiterpene lactone	Anticancer

*Ficus amplissima* bark extract showed the presence of melibiose, isoquinoline alkaloids, and phenylpropanoid derivatives such as cinnamyl cinnamate. The detection of Marchantin A, a bisbenzyl compound, is noteworthy due to its reported antimicrobial potential, supporting the traditional use of this species in infectious conditions. In *Ficus hispida*, the dominance of flavonoids and condensed tannins such as gerberinol and procyanidin B7 suggests strong antioxidant and cardioprotective potential. The identification of alkaloids like (±)-carnegine and ryanodine further indicates neuroactive and muscle-relaxant properties, corroborating ethnomedicinal claims related to nervous system disorders. The HR-LCMS profile of *Ficus arnottiana* bark revealed a chemically diverse composition, including cyanogenic glycosides (lotaustralin), proanthocyanidins, and furanone derivatives. The presence of procyanidin B7 again highlights antioxidant and cardioprotective activity, while antimicrobial furanones support its traditional use against infections.

*Ficus benjamina* bark extract exhibited a distinct chemical fingerprint with flavonoid glycosides (cynaroside A), iridoid glycosides (leonuriside A), and acetogenins (squamosstatin A). These compounds are well known for hepatoprotective, anti-inflammatory, and anticancer activities, indicating therapeutic versatility of this species. In *Ficus carica* bark, alkaloids such as matrine and norsalsolinol derivatives were prominent, along with flavonoid glycosides and lignans like isoamericanol A. Lignans are recognized for their anticancer and antioxidant properties, providing pharmacological justification for the medicinal importance of *F. carica* bark. *Ficus elastica* demonstrated the presence of flavonoids, organosulfur compounds (allixin), diterpenoids, and phenolic acids such as isoferulic acid. These compounds are associated with antioxidant, antimicrobial, and anti-inflammatory activities, suggesting potential applications in inflammatory and oxidative stress-related disorders. The bark extract of *Ficus microcarpa* showed a combination of alkaloids, flavonoids, sialic acid derivatives, phenolic acids, and sesquiterpene lactones. The detection of neuraminic acid indicates immunomodulatory potential, while sesquiterpene lactones such as verimol A are widely reported for anticancer activity.

The present study provides a comprehensive HR-LCMS-based phytochemical profiling of bark extracts from seven *Ficus* species (*F. amplissima*, *F. hispida*, *F. arnottiana*, *F. benjamina*, *F. carica*, *F. elastica*, and *F. microcarpa*) collected from the Marathwada region of Maharashtra. The HR-LCMS analysis revealed a wide diversity of secondary metabolites, including carbohydrates, alkaloids, flavonoids, proanthocyanidins, phenolic acids, glycosides, terpenoids, and lignans, confirming the metabolic richness of *Ficus* bark drugs.

### Conclusion

The present investigation provides a comprehensive HR-LCMS-based phytochemical characterization of bark drugs from selected *Ficus* species of the Marathwada region. The integration of Pharmacognostic evaluation, preliminary phytochemical screening, and advanced chromatographic analysis establishes reliable chemical markers for species identification and standardization. The detection of pharmacologically important compounds supports the ethnomedicinal relevance of these bark drugs and underscores their potential for future drug discovery and development.

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