A Comprehensive Review On The Ethnopharmacological Importance Of Diospyros Buxifolia (Blume) Hiern

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Plants are rich sources of natural compounds with extraordinary therapeutic potential, essential for developing novel drugs. Natural products can target multiple physiological pathways, departing from the classical 'one-disease one-target' tradition. Keeping this in view, recent studies have looked into Diospyros species. These medicinal plants have traditionally been utilised for managing several illnesses, as a promising avenue for further exploration. With over 500 species, the Diospyros genus consists of trees and shrubs spread across tropical and subtropical regions of the world. Due to a lack of awareness of Diospyros buxifolia and being an underutilised plant, very few investigations have been done on it of which it has been proven to be an excellent source for secondary metabolites, nutraceuticals, and anti-oxidants having a great potential to be used in medicinal and food purposes. Additionally, D. buxifolia is a natural source of antioxidant and antibacterial agents that could help with the problems caused by oxidative stress and drug-resistant microbes in global health. Therefore, in the current review, we discuss the phytochemical constituents and therapeutic properties of D. buxifolia which can provide researchers with insights into the ethnomedicine values of this traditionally important medicinal plant.

Keywords: Antibacterial; Antidiabetic; Anti-oxidant activity; Diospyros buxifolia; Enzyme inhibition; Medicinal plant.

Traditional healers across the globe utilise a diverse array of botanical species in their Indigenous practices to promote and sustain the health of their respective communities. In recent times, there has been a resurgence of pharmacological interest in phytotherapeutic agents derived from various botanical components or extracts utilized by indigenous ethnic groups. About 40% or more of the therapeutic agents currently used in Western Ghats by the locals are derived or at least partially derived from natural resources, particularly from medicinal plants. Further, the majority of pharmacologically potent compounds obtained from plants have

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been identified, discovered and purified through follow-up research to substantiate the authenticity of folklore data and ethnomedicinal applications. Consequently, there exists a substantial rationale to elucidate the types of botanical specimens utilised by individuals, the methodologies of their application, and the specific contexts in which these plants demonstrate their effectiveness against various diseases. Furthermore, ayurveda, unani, and sidda, the aboriginal system of native medicines, describe numerous plant species in detail.[1]

Ethnopharmacology refers to the roles and influences of traditional plants and trees in pharmacological research and drug development. In general, traditional therapeutic usage of plants and trees is referred to as ethnomedicines. Medicinal preparations play a very significant role in pharmacological ailments in humans and animals because of the existence of phytochemical elements. Bioactive phytochemicals are found in all parts, including leaves, fruits, vegetables, and roots, and they offer resistance to a broad spectrum of diseases owing to the implication of essential components and secondary metabolites. Such indispensable secondary components include flavonoids, terpenoids, alkaloids, and phenolics. So far, a wide variety of medicinal plants have been studied, including the production of secondary metabolites from plant tissue culture approaches; some examples include *Catharanthus roseus* alkaloids, which have effective anti-cancer activity, *Caralluma adscendens*, which is a potent appetite suppressant, and so on.[2-3]

The plant *Diospyros buxifolia* (Blume) Heirn., (Ebenaceae family) has a pantropical distribution and includes the genera *Diospyros*, which have around 500 to 600 species. The species of the genus *Diospyros* have long been extensively employed in traditional medicine to treat various illnesses. So far, the active main chemicals from *Diospyros* species have been naphthoquinones, triterpenoids, and tannins.[4-6]

Diospyros spp. are often tree shrubs or subshrubs with whole alternating leaves, single blooms, and meaty fruits (berries) containing two or more seeds. In particular, *D. buxifolia* is a dioecious tree that can reach a height of 30 m and yields berry-like fruits.[7]

Based on the various descriptions in the literature the multiple parts of the genera *Diospyros* have been used to treat several infectious diseases such as antibacterial, antifungal, anthelminthic, and antiviral; urogenital (anti-hemorrhagic); skin diseases that include dermatitis, fresh wounds, bedsores, and rashes; and for treatment of musculoskeletal disorders related to body pain, bruises, painful fractures, and rheumatism. However, the literature review indicates the extensive use of *Diospyros* extracts for treating diseases related to oral cavity health (oral wounds, bad breath, and toothaches) and gastrointestinal disorders (diarrhoea, emetic, flatulence, etc.). Further, the bark, fruit, and root are the most commonly employed *Diospyros* plant components in traditional medicine.[8–10]

D. buxifolia is also notable for its ecological and economic significance. This species is primarily found in tropical regions and is documented for its valuable timber and edible fruit. Due to habitat loss, the conservation status of *D. buxifolia* is critical, necessitating efforts for its preservation. Because of the dangers posed by deforestation and habitat degradation, *D. buxifolia* requires continuous conservation efforts, according to the International Union for Conservation of Nature's (IUCN) red list. Ex situ conservation strategies, such as those implemented in botanical gardens, are crucial for preserving genetic diversity and ensuring the species' survival.[11-12]

The distribution is particularly prevalent in regions like Sulawesi, Indonesia, and the Western Ghats of India where conservation efforts are focused. The wood of *D. buxifolia* is highly valued for its quality, frequently used in furniture and crafts, similar to other *Diospyros* species known for ebony timber. Additionally, the species produces edible fruits, contributing to local diets and economies.[13]

The present study's rationale is to help researchers and academics comprehend the ethnomedicine values of *D. buxifolia* so that they can pursue further investigations with the same and potentially impact the ethnopharmacological insinuations of *D. buxifolia.*

MATERIALS AND METHODS

A comprehensive search of electronic databases was conducted to identify studies on *D. buxifolia* focusing on investigations on phytochemical analysis using leaf, root, stem, and fruit extracts with various solvent systems to enlist the secondary metabolites present. The search was further widened to identify studies about *in vitro* and *in vivo* utilisation of *D. buxifolia* extracts to determine the ethnopharmacological relevance and therapeutic properties. Two independent reviewers evaluated the titles and abstracts of potential studies, followed by a full-text review of eligible publications. Data extracted included phytochemical analysis using GC-MS, antibacterial activities, anti-diabetic activities, antioxidant activity, and inhibition studies of pharmacologically relevant enzymes and results. The collected data were synthesized to provide insights into the phytochemical and ethnopharmacological importance of *D. buxifolia*.

RESULTS

Ethnopharmacology of *D. buxifolia*

Ethnopharmacology refers to the roles and influences of traditional plants and trees in pharmacological research and drug development. *D. buxifolia*, part of the *Diospyros* genus, is rich in bioactive phytochemicals that exhibit a range of therapeutic applications. The primary compounds identified include flavonoids, terpenoids, naphthoquinones, tannins, saponins, and reducing sugars, which contribute to their pharmacological properties.

Numerous health advantages, such as antioxidant, anti-inflammatory, antibacterial, antifungal, and anticancer abilities, have been associated with these phytochemical ingredients which are constantly present as secondary metabolites.

Key bioactive phytochemicals in *D. buxifolia* extracts

The medicinal potential of every plant extract originates mostly from the presence of a specific collection of chemical ingredients. Such components have been identified employing basic preliminary assays. In general, the chemical contents of *D. buxifolia* extracts are categorized into the following classes, based on prior investigations.

• Flavonoids: Flavonoids are well known for their antioxidant properties; they help reduce oxidative stress and inflammation in tissues.[4]

• Naphthoquinones: Particularly noted for their cytotoxic effects against cancer cells, they play a crucial role in anticancer therapies.[5]

• Saponins: Several investigations have revealed that saponins have useful effects on maintaining the cholesterol levels of blood, bone health, blood glucose levels, and cancer risk. A saponin-rich diet has been found to prevent dental cavities, suppress platelet aggregation, treat hypercalciuria, and serve as an antidote to heavy metal toxicity.[9]

• Terpenoids: These compounds exhibit significant antimicrobial and anti-inflammatory effects, making them valuable in treating infections and inflammatory conditions.[10]

• Tannins: These compounds have demonstrated antiviral and antibacterial properties, contributing to the plant's traditional medicinal uses.[10]

Constituent bioactive phytochemicals reported from various parts of *D. buxifolia*

Leaf extracts

Leaves of *D. buxifolia* have been used as a traditional remedy against various ailments including oral hygiene, healing of oral wounds, toothaches, gastrointestinal disorders such as diarrhoea, emetic, flatulence, etc. Analysis of the leaf of *D. buxifolia* showed the presence of several bioactive phytochemical compounds. The structures of these identified bioactive components from leaf extracts using methanol, water, n-hexane, and dichloromethane have been published and registered in Table 1. Studies on leaf extract of *D. buxifolia* have shown to possess several vital biological potentials such as antioxidant, antimicrobial, antifungal, anti-inflammatory, antidermatitic, antiseptic, anticancer, candidicide, antimalarial, antileishmanial, antitrypanosomal and antidiuretic activity.[14-15]

Stem bark extracts

The stem and stem bark are the most commonly employed *Diospyros* plant components in traditional medicine. The extracts have been used to treat several infectious diseases (antibacterial, antifungal, anthelminthic, and antiviral); urogenital (anti-hemorrhagic); skin diseases (dermatitis, fresh wounds, bedsores, and rashes); and musculoskeletal (body pain, bruises, painful fractures, and rheumatism). Phytochemical analysis of bark methanol and water extracts of *D. buxifolia* using GC-MS (Gas Chromatography-Mass spectroscopy) disclosed

Table 1. Summary of phytochemicals identified and reported in leaf and stem extracts of *D. buxifolia* using GC-MS analysis

S No.	Plant parts used (Solvent)	Name of the phytochemicals reported	Associated pharmacological activities (References)
1	Leaf (Methanol and water)	1,2,3- Benzenetriol Phenol, 2,4-bis (1,1-dimethylethyl)-. Phytol, acetate Estra- 1,3,5(10)-trien-17. beta – ol. Heptasiloxane, 1,1,3,3,5,5,7,7,9,9, 11,11,13,13-tetradecamethyl Tetracosane Benzene, 1,1'-sulfonylbis [4- chloro- Octadecane, 1,1'-[1,3-propanediylbis(oxy)] bis. Octasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11, 13,13, 15, 15- hexadecamethyl-.	Antimicrobial, Antifungal, Antioxidant, Anti-inflammatory, Antidermatitic, Antiseptic, Anticancer, Candidicide Antimalarial, Antileishmanial, Antitrypanosomal Antidiuretic.[15]
2	Stem (Methanol and water)	Benzothiazole. Formic acid, 2,6-dimethoxyphenyl ester. 1,2,3- Benzenetriol Phenol, 2,4-bis (1,1- dimethylethyl)-. Phytol, acetate Benzenepropanoic acid, 3,5- bis (1,1- dimethylethyl)-4 hydroxy-, methyl ester. Isoquinoline, 1-[(3,5-dihydroxy) benzyl]-1,2,3,4-tetrahydro-6-hydroxy-. Heptasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11, 13,13-tetradecamethyl. Phenol, 2,2'-methylenebis [6- (1,1-dimethylethyl)- 4- methyl.	Anticancer, Antimicrobial, Antidiabetic, Anti-inflammatory, Antileishmanial, Antiviral, Antidermatitic, Antiseptic, Antidermatitic.[15]
3	Leaf (n- hexane and Dichloromthane)	1-Heptadecene Tetradecane 2,4-Di-tert butylphenol Hexadecane 1-Octadecene Heptadecyl trifluoroacetate Eicosane Oleic Acid 1-Hexacosanol Cyclotetracosane 2,5-Dihydroxy benzoic acid β-Amyrin α-Amyrin Squalene 9,12-Octadecadienoic acid (Z,Z)-. 4,4'- Bi- 1,3,2- dioxaborolane, 2,2'-diethyl-,(R*,S*). Dodecane Cyclohexasiloxane, dodecamethyl-. Cycloheptasiloxane, tetradecamethyl. 2,4-Di-tert-butylphenol Cyclooctasiloxane, hexadecamethyl-. Octadecane Cyclononasiloxane, octadecamethyl-. Cyclodecasiloxane, eicosamethyl-. Phytol Docosane	Antimicrobial, Antifungal, Antioxidant, Anti-inflammatory, Antidermatitic, Antiseptic, Anticancer, Candidicide, Antimalarial, Antileishmanial, Antitrypanosomal, Antidiuretic.[14]

Table 2. Summary of pharmacologically important enzyme inhibition studies performed using *D. buxifolia* extracts

S. No	Plant part used (Solvent)	Name of the enzyme/assay	Pharmacological relevance (References)
1.	Leaf and stem (Methanol and water)	Porcine pancreatic α-amylase	Antidiabetic activity[16]
2.	Leaf and stem (Methanol and water)	Amyloglucosidase(*Aspergillus oryzae*)	Antidiabetic activity[16]
3.	Leaf extract (n-hexane and dichloromethane)	Rat intestinal α-glucosidase	Antidiabetic activity[14]
4.	Leaf extract (n-hexane and dichloromethane)	Ferric Reducing Antioxidant Power	Antioxidant capacity[14]
5.	Leaf and stem (Methanol and water)	Antibacterial assay	Antimicrobial activity[15]
6.	Leaf and stem (Methanol and water)	Radical scavenging assay	Antioxidant capacity[15]

the presence of various therapeutically active phytocomponents (Table 1). These compounds exhibit several pharmacological properties such as anticancer, antimicrobial, antidiabetic, anti-inflammatory, antileishmanial, antiviral, antiseptic, and antidermatitic.[14-15]

Pharmacologically important biological activities using *D. buxifolia* extracts

D. buxifolia extracts have a variety of medicinal benefits. Tables 1 and 2 include details on different phytochemicals and their subsequent potential medicinal uses. Below are some of the main biological activities on which researchers have previously worked with *D. buxifolia* extracts.

• Anticancer: *D. buxifolia* extracts have shown the potential to inhibit tumour growth and induce apoptosis in cancer cells.[4-8]

• Anti-inflammatory: Its bioactive compounds can mitigate inflammation, providing relief in conditions like arthritis.[5]

• Antimicrobial: The plant's extracts especially the methanolic ones are effective against various pathogens, addressing the growing concern of antibiotic resistance.[10-15]

• Anti-diabetic: The extracts of the plant have shown promising anti-diabetic action in an *in vitro* investigation.[16-18]

• Enzyme inhibitory action: Inhibitors of endogenous carbohydrases á-glucosidase and á-amylase reduce postprandial hyperglycemia and are effective diabetic therapies. Similarly, calorie restriction brought about by pancreatic lipase and carbohydrase suppression helps treat obesity and prevent weight gain. Consequently, assays designed to evaluate enzyme inhibition with a focus on á-glucosidase, á-amylase, and lipase are prevalent within the realm of scientific research; furthermore, the systematic screening of plant extracts and natural compounds for their inhibitory effects on these enzymes constitutes a widely adopted methodology in the quest for the identification of promising pharmacological agents aimed at the management and treatment of metabolic disorders such as diabetes and obesity.[19-23]

The diabetes-related enzyme inhibition potential of the *D. buxifolia* extracts was examined *in vitro* by determining the á-amylase and amyloglucosidase from 0.125 mg mL^{-1} to 1.0 mg mL^{-1} at four increasing doses by Rao et al.[23] The results demonstrated that Methanolic extracts of the stems had significant inhibition of amyloglucosidase (62.72±0.15%) followed by methanolic extract of the leaves (58.93±0.08%), aqueous extracts of the stems (36.65±0.05%) and finally aqueous extract of the leaves (20.57±0.03%).

DISCUSSION

From plants to natural products, enzyme tests are widely used in drug discovery—nearly 50% of all small-molecule medications on the market target enzymes. Enzymes are a desirable target for new drug development initiatives because of their high degree of druggability and target validation provided by their protein structure. An important class of clinical medications, enzyme inhibitors are used to treat conditions like cancer, heart disease, diabetes, neurological problems, and obesity. Natural compounds are secondary metabolites synthesized by biotic entities, including flora and microorganisms. The plant kingdom represents a prolific and readily available reservoir of natural compounds, a significant portion of which has yet to be investigated for potential bioactive metabolites.[3-6]

Studies by Hariyanto et al[14] revealed that the semi-polar extracts (n-hexane and Dichloromethane) of *D. buxifolia* demonstrated potent inhibitory activities against rat intestinal á-glucosidase (IC_{50} = 33.31±3.23 ìg/mL). Further, in the findings, The Ferric Reducing Antioxidant Power studies revealed that leaf extracts using non-polar and semi-polar solvents of the Diospyros species have significant antioxidant potential, with the IC_{50} range of 6.48±0.38 - 11.35±0.16 ìmol of Fe/g. The findings from this evaluation established the therapeutic usefulness of various Diospyros species extracts, further validating the traditional remedial applications. Recent studies by Basivi et al[15] observed that extracts of *D. buxifolia* displayed good antibacterial activity against *Klebsiella pneumoniae* and, *Staphylococcus aureus*. At the same time, *Bacillus subtilis,* and *Escherichia coli* showed limited effectiveness against tested strains using the disc diffusion assay. However, leaf and stem extracts demonstrated the ability to inhibit the growth advancement of Gram-positive and Gram-negative bacterial strains.[15]

There are several well-reported antimicrobial action mechanisms of phytochemicals in medicinal plants against bacteria, including disruption and damage of cell walls and cell membranes, leading to the loss of cellular components, interaction with membrane proteins and enzymes leading to membrane structure and function modification, and DNA function alteration. Nonetheless, the report did not elucidate the specific mechanism. The results revealed that the aqueous leaf extract and methanolic stem extracts showed minimum inhibitory concentration values at the highest concentration for *B. subtilis* and can be considered the most effective plant crude extract, demonstrating stronger antibacterial activity against *B. subtilis* compared to the other extracts. The results indicate that the methanolic extracts had superior inhibitory effects on the selected microorganisms with stronger antimicrobial activity against *K. pneumoniae*. Further, this study demonstrates and suggests the potential use of these plant extracts (Table 2) for treating certain bacterial infections.[15]

The above study also reported the evaluation of antioxidant activity. The maximum DPPH (2,2-diphenyl-1-picrylhydrazyl) scavenging activity was noted in favour of methanolic leaf extract among all other extracts and this antioxidant activity may be attributed to secondary metabolites present in different parts of *D. buxifolia*. The methanolic stem extract exhibited a significant reducing ability at 100µg/mL, making it an efficient antioxidant agent.

The diabetes-related enzyme inhibition potential of the *D. buxifolia* extracts was examined *in vitro* by determining the á-amylase and amyloglucosidase from 0.125 mg mL^{-1} to 1.0 mg mL at four increasing doses by Rao et al.[23] The research concluded that the methanolic stem extract of *D. buxifolia* exhibited the presence of phytochemicals, as well as significant inhibition of á-amylase and amyloglucosidase, which is indicative of the significant antidiabetic properties of the extracts of *D. buxifolia*.

The standard reduction power displays variations in the reducing ability of *D. buxifolia* leaves and stems, which might be due to differing availability. The reducing power of the extracts is proportionate directly to the concentration of the extracted sample and an increase in the reducing ability of plant extract could be attributed to the formation of reductants that can react with free radicals, terminating and stabilizing the radical chain reaction during the process. This research offers a rationale for the antibacterial and antioxidant properties of *D. buxifolia* extracts in methanol and water and these extracts could be potential sources of antimicrobials inhibiting microorganism growth and antioxidants (Table 2).

CONCLUSION

The phytopharmaceuticals are developed using traditional medicine systems emphasizing the unique demonstration of how historical knowledge informs modern drug discovery. The diversity of phytochemicals within the Diospyros genus suggests a vast potential for drug discovery and development. *D. buxifolia* holds significant ecological and economic value, it faces challenges from environmental changes and human activities. Continued research and conservation efforts are essential to mitigate these threats and promote sustainable use of this species. *D. buxifolia* shows promise in various therapeutic areas particularly inhibition of enzymes involved in diabetes, antioxidant, and antimicrobial activity. The plant contains an abundant extent of under-utilized pharmacologically important secondary metabolites, which can be developed into drugs. However, future research investigations are required to fully determine and demonstrate the mechanisms of action to optimize the *D. buxifolia* extract applications in modern medicine. Thus, the present review provides a better understanding of the ethnomedical benefits of this historically significant medicinal plant and their applications in modern medicine.

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This research did not involve human participants, animal subjects, or any material that requires ethical approval, and therefore, informed consent was not required.

Clinical trial registration

This research does not involve any clinical trials.

Author contributions

Bhargavi Gangadhar: Concept, Data review, Title choice, Formal analysis, and data interpretation, Writing – original draft; Sapna Motti: Data review, formal analysis, and Writing; Suprada Rao Mangalore Suresha: Concept, Data review, formal analysis, and Writing – review & editing; Chandrashekar Jambhava Samavedamuni: Concept, formal analysis, and Writing – review and editing; Nataraju Angaswamy: Concept, Data review, Title choice, formal analysis, and Writing – review & editing and final decisions.

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