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A COMPREHENSIVE REVIEW ON THE PHYTOCHEMICAL, PHARMACOLOGICAL, AND ETHNOBOTANICAL PROFILE OF *Boerhavia diffusa* Linn. WITH SPECIAL REFERENCE TO INDEPTH HEPATOPROTECTIVE MECHANISMS

Aswath T., Kabilesh P. V., Madhu Sri A., Sujitha R., Sudhikshaa Devi M. S., Dhinesh Kumar S., Rajasekaran A., *Arivukkarasu R.

KMCH College of Pharmacy, Coimbatore, Tamil Nadu, India -641048. Affiliated by Dr M.G.R Medical University.

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***Corresponding Author**

Arivukkarasu R.

Professor, Department of Pharmacognosy, KMCH College of Pharmacy, Coimbatore, Tamil Nadu, India -641048. Affiliated by Dr M.G.R Medical University.

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ABSTRACT

This comprehensive review combines ancient ethnobotanical tradition with modern pharmacological validation, to explore the therapeutic profile of *Boerhaavia diffusa Linn.* (Punarnava). The paper highlights the global geographical distribution of the plant, its unique macroscopic morphology and rich phytochemical matrix consisting of active chemical markers such as punarnavine, boeravinone B and liriodendrin. The paper focuses on the multi-targeted hepatoprotective mechanisms of the plant and how its bioactives alleviate the hepatic stress. These pathways include restoration of endogenous antioxidant defenses, suppression of NF-κB-mediated inflammatory cascades, regulation of Cytochrome P450 enzymes, and modulation of Nrf2/ARE signaling pathway to promote cell survival and hepatocyte membrane stabilization.

KEYWORDS: *Boerhaavia diffusa,* Hepatoprotective, molecular docking, Punarnava, Punarnanvine, Boeravinone B.

INTRODUCTION

Boerhavia diffusa Linn. (Syn. Boerhaavia repens Linn., Family: Nyctaginaceae) as 'Punarnava' in the traditional Indian Sanskrit literature is a critically important perennial

creeping herb that is widely celebrated in ancient ethno-medical traditions. The literal etymological derivation of its vernacular name ‘Punarnava’- where ‘Puna’ means ‘again’ and ‘Nava’ means ‘new’—profoundly reflects its acclaimed clinical capacity to rejuvenate, regenerate and restore the vital operational architecture of compromised bodily organs, most notably the hepatic and renal organ networks.[1,2] The taxon has been classified for millennia in the elite ‘Rasayana’ class of therapeutic agents in classical Ayurvedic texts such as Charaka Samhita and Sushruta Samhita, owing to its multi-systemic physiological adaptogenic, immunomodulatory and anti-aging capabilities.[3] Beyond the geographical confines of the Indian subcontinent, indigenous civilisations in Africa, Central and South America and South-East Asia have independently incorporated this botanical resource into their empirical health care systems to address a wide spectral range of metabolic, inflammatory, infectious and degenerative pathology.[4] B. diffusa is scientifically well validated in modern phytotherapeutic paradigms and researchers are trying to validate its structural-functional mechanisms of action using modern analytical platforms, isolating unique bioactive scaffolds with remarkable antioxidant, anti-inflammatory, immunomodulatory, chemopreventive and tissue-regenerative qualities.[5]

Geographical Distribution

Boerhavia diffusa is a highly resistant, pantropical and subtropical weed, with an extensive, worldwide geographical matrix. It is naturally distributed and abundant in the vast terrestrial area of the Indian subcontinent, from the lower structural levels of the southern Himalayan ranges, through the extensive Indo-Gangetic plains, to the southernmost tip of the peninsular region, attaining heights of nearly 2,000 m above mean sea level.[6] Its geographical distribution outside India extends to Pakistan, Nepal, Bangladesh, Sri Lanka and Myanmar among other countries. Outside South Asia, it is widely distributed in tropical Africa, native to both western and eastern regional ecological zones, the southern United States (especially Florida and Texas), the Caribbean basin and equatorial South America, including Brazil, Colombia and Venezuela.[7] The plant is thriving with extraordinary adaptive plasticity as a ruderal weed, populating open and highly disturbed habitats such as roadsides, agricultural fallow fields, waste dump sites, railway embankments and pasture clearings, adapting easily to poor, sandy, stony or nitrogenous soil beds under diverse climate configurations ranging from highly arid to hyper-humid.[8]

Description of Genus

The genus *Boerhaavia* (sometimes spelled *Boerhaavia* in older classical literature) was named by Carl Linnaeus in honor of the famous Dutch physician, botanist and academician Hermann Boerhaave (1668–1738). Taxonomically, the genus is included in the family Nyctaginaceae (popularly known as the Four-O'Clock family), which has about 40 to 50 valid species distributed in tropical, subtropical and warm temperate regions of the Old and New Worlds.[9] Members of the genus *Boerhaavia* are morphologically characterized as annual or perennial herbs, sometimes subshrubby, with a frequent prostrate, ascending or diffuse habit. The genus is characterized by an unusual pattern of secondary growth in the matrices of both stem and root, which is manifested in the form of successive concentric rings of collateral vascular bundles in thick, highly lignified conjunctive tissue.[10] The inflorescence architecture is usually terminal or axillary paniculate cymes, umbels or capitula. The flowers are small, bisexual, actinomorphic and usually monochlamydeous, lacking a true corolla. The calyx is petaloid, tubular or campanulate, and constricted mid-way, the lower portion of the calyx persisting around the developing ovary, and hardening into a specialized, often glandular, anthocarp (fruit). This anthocarp is a family-specific structure with 3–5 prominent longitudinal ribs that often secrete sticky mucilaginous compounds from specialized capitate trichomes, facilitating zoochorous seed dispersal.[11]

Morphology of *Boerhavia Diffusa*

Boerhavia diffusa displays a very distinct and highly branched macroscopic morphology that facilitates its aggressive prostrate colonization of terrestrial substrates. Root system consists of a huge, woody, fusiform and elongated taproot, very succelent and fleshy when fresh, 15–30 cm long and up to 3-5 cm in diameter. The outer surface of the root-bark is pale brown to soft greyish yellow with fine longitudinal wrinkles and a fibrous inner structure that breaks with a characteristic earthy odour and a bitter acrid taste.[12] Stems cylindrical, highly branched, prostrate or ascending up to 1-1.5 m long, distinctly purplish-red to green, markedly swollen or jointed at the nodal junctions, glabrous to finely puberulent or sticky-glandular along younger growth axes.[13] The leaf is simple, distinctly opposite and explicitly unequal in each pairs (anisophylly). The larger leaf of the pair is about 2.5-4 cm long and the smaller counterpart is 1-2 cm. The leaf blades are broadly ovate, oblong-obovate or subcordate with an entire, often wavy (undulate) margin with a characteristic purplish coloration along the margin. The adaxial (upper) surface is deep green and glabrous, while the abaxial (lower) surface has a distinct silvery-white, glaucous appearance due to

microscopic scales and fine puberulent hairs.[14] The inflorescences are small, capillary, umbellate clusters of 4–10 small flowers on long-peduncled axillary or terminal panicles. The perianth is bright pink, magenta or rarely white with an infundibuliform tube 2 mm long and a 5-lobed limb. Stamens 2 to 3, slender, distinctly exserted beyond perianth tube. The fruit is a small, dry, clavate, 5-ribbed, 3 mm long, one-seeded anthocarp, densely covered with sticky glandular capitate hairs.[15]

Fig. 1.

Fig. 2.

Fig. 1: It represent the root morphology of *Boerhaavia diffusa* & Fig.2- represent the whole morphology of *Boerhaavia diffusa.*

Ethnobotanical Uses

The ethnobotanical uses of *Boerhavia diffusa* are surprisingly extensive and diverse, reflecting a surprising confluence of traditional medical knowledge across different indigenous cultures worldwide. In classical Ayurvedic medicine, the whole plant and particularly the succulent taproot is formulated as 'Punarnavadi Kwath' or 'Punarnavasava' to serve as a primary therapy for generalized anasarca, ascites and dropsy due to early stage hepatic cirrhosis or advanced chronic kidney diseases.[1,16] In the rural areas of India, traditional healers treat acute jaundice with decoctions of fresh roots which stimulate the flow of bile and resolve painful splenomegaly. Roots have been described as a powerful diuretic, stomachic, expectorant and purgative having excellent clinical resolution of internal abdominal swellings, tumors and dyspeptic disorders in historical compendiums like Kirtikar and Basu's Indian Medicinal Plants.[17] Leaves are regularly collected by the indigenous tribal groups in West Bengal and Assam and boiled as a curative pot-herb to combat nutritional anemia and to relieve acute asthmatic dyspnea. In ethno-ophthalmology, a topical wash of fresh leaf sap mixed with honey is instilled into the eyes for the treatment of night blindness,

corneal opacity and acute conjunctivitis.[18] In the traditional African medicine systems, especially in Nigeria and Ghana, the plant is employed as a powerful spasmolytic and anticonvulsant to manage epilepsy and febrile convulsions in infants. The paste of the whole herb is used externally as a poultice to draw out venom from snakebites, scorpion stings and to resolve chronic abscesses and skin diseases like scabies and eczema.[19] In Central and South American ethnomedicine (e.g., Brazil), it is valued under the name 'Erva-tostao' as an elite lithontriptic agent to expel renal calculi, clear urinary tract infections, and reduce hepatic congestion.[20]

Phytochemical Review

The mass phytochemical screening of *B. diffusa* has revealed a diversified chemical matrix which led to a rich collection of secondary metabolites in all the tissues of the plant. The most chemically distinct group of compounds isolated from this taxon is a unique family of rotenoids known as 'Boeravinones'. To date, the characterization of boeravinones A, B, C, D, E, F, G, H, I and J has been carried out using sophisticated spectroscopic techniques, with particular scientific interest in Boeravinone B and Boeravinone G due to their extraordinary multi-targeted biological activities.[21] The plant also synthesized a unique purine nucleoside derivative 'Punarnavine' (an alkaloid) in roots in a highly concentrated form, which is a major chemical marker for standardization.[22] Besides these rotenoid scaffolds, the plant also synthesized a unique purine nucleoside derivative. The plant is also rich in a range of polyphenolic and flavonoid compounds such as quercetin, kaempferol, myricetin and isorhamnetin-3-O-robinobioside, which provide a strong baseline antioxidant capacity.[23] The lipid fraction contains a high level of phytosterols, especially beta-sitosterol, stigmasterol and campesterol. Furthermore the roots contain a complex mixture of non-structural carbohydrates, specialised glycoproteins, lignans (e.g. liriodendrin, syringaresinol mono-beta-D-glucoside) and a large amount of potassium nitrate (KNO3) which synergizes with the organic components to promote therapeutic diuresis.[24, 25]

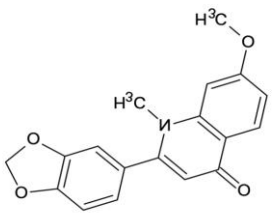

Fig. 3: Punarnavine.

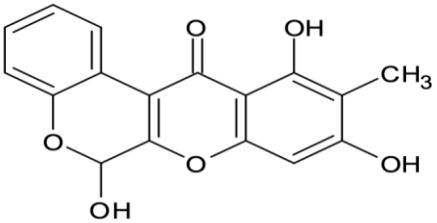

Fig. 4: Boeravinone B.

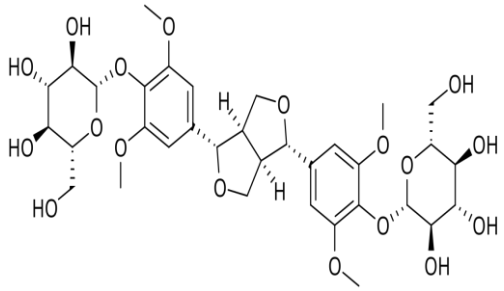

Fig. 5: Liriodendrin.

Pharmacological Properties

Boerhavia diffusa has wide pharmacological profile which has been well documented with modern scientific approaches in vitro and in vivo. The plant shows broad-spectrum antimicrobial activity against pathogenic bacterial and fungal isolates, impressive anti-inflammatory activity by inhibiting cyclooxygenase (COX-1 and COX-2) and 5-lipoxygenase (5-LOX) enzymes and potent immunomodulatory activity characterized by activation of macrophages and regulation of specific pro-inflammatory cytokines.[2,26] It also showed appreciable antiurolithic, antidiabetic, anticonvulsant and nephroprotective effects.[27]

Indepth Review of Hepatoprotective Mechanisms

The most studied and mechanistically understood pharmacological property of *B. diffusa* is its hepatoprotective potential. Pre-clinical studies in different animal models such as rodents challenged with hepatotoxins such as carbon tetrachloride (CCl4), paracetamol (acetaminophen), rifampicin, isoniazid, thioacetamide and heavy metals have shown that pre-administration or post-treatment of *B. diffusa* root extracts (especially methanolic and aqueous fractions) prevents severe architecture damage to hepatocytes.[28,29] At serum biochemical level, the treatment with *B. diffusa* significantly prevents the pathological leakage of intracellular hepatic enzymes, leading to a marked decrease of the elevated Alanine Aminotransferase (ALT), Aspartate Aminotransferase (AST), Alkaline Phosphatase (ALP) and Total Bilirubin levels and simultaneously preserving the serum albumin and total protein biosynthesis.[30]

Mechanistically, the hepatoprotective action is driven by several distinct molecular pathways:

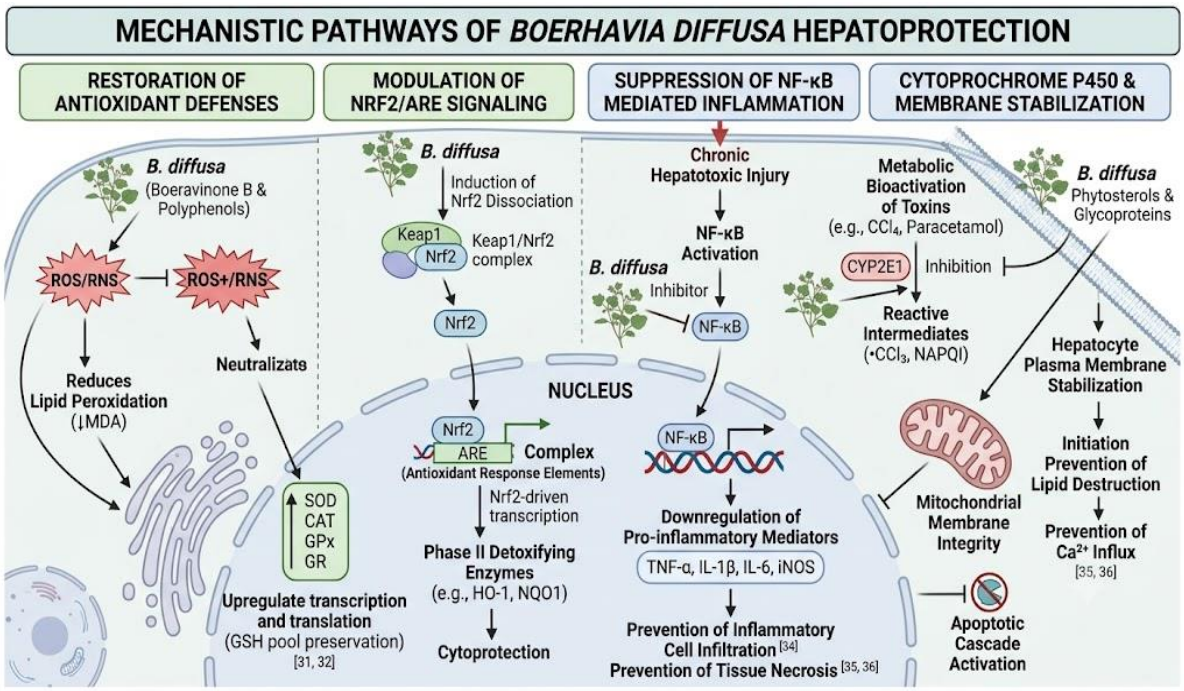

Fig. 6: A Schematic diagram represents mechanism of hepatoprotective action for *Boerhaavia diffusa.*

Restoration of Endogenous Antioxidant Defenses: *B. diffusa* extracts rich in Boeravinone B and polyphenols, directly scavenges reactive oxygen species (ROS) and reactive nitrogen species (RNS). This significantly reduces lipid peroxidation as indicated by a reduction in malondialdehyde (MDA) formation and upregulates the transcription and translation of endogenous antioxidant enzymes such as Superoxide Dismutase (SOD), Catalase (CAT), Glutathione Peroxidase (GPx), and Glutathione Reductase (GR), thereby preserving the cellular pool of reduced Glutathione (GSH).[31, 32]

Modulation of the Nrf2/ARE Signaling Pathway: Molecular assays indicate that active fractions of *B. diffusa* induce dissociation of Nuclear Factor Erythroid 2-Related Factor 2 (Nrf2) from its cytosolic repressor Keap1. After being released, Nrf2 moves into the nucleus, where it attaches to Antioxidant Response Elements (ARE) to stimulate the transcription of Phase II detoxifying enzymes and cytoprotective proteins such as Heme Oxygenase-1 (HO-1) and NAD(P)H Quinone Dehydrogenase 1 (NQO1).[33]

Suppression of NF-kB Mediated Inflammation: *B. diffusa* prevents the activation and nuclear translocation of Nuclear Factor Kappa B (NF-kB) in chronic hepatotoxic injury. It downregulates the transcription of major pro-inflammatory mediators such as Tumor Necrosis Factor-alpha (TNF-a), Interleukin-1 beta (IL-1b), Interleukin-6 (IL-6), and Inducible

Nitric Oxide Synthase (iNOS) thus preventing widespread inflammatory cell infiltration and subsequent hepatic tissue necrosis.[34]

Regulation of Cytochrome P450 Enzymes and Membrane Stabilization: *B. diffusa* regulates the activity of specific Cytochrome P450 isoenzymes (in particular CYP2E1) which are involved in the metabolic bioactivation of toxins such as CCl4 and paracetamol into highly reactive, destructive intermediate radicals (e.g. trichloromethyl radicals or NAPQI). It inhibits these isoenzymes in a moderate way, and stabilizes the hepatocyte plasma membrane owing to its phytosterol and glycoprotein components, thus preventing the initiation of radical-induced lipid destruction and preventing the influx of calcium inside the cell, maintaining the integrity of the mitochondrial membrane and preventing the activation of apoptotic cascades.[35, 36]

***Insilico* molecular Docking studies**

An *Insilico* molecular docking study was performed using **CB-DOCK2** to evaluate and validate the hepatoprotective potential of natural alkaloids **punarnavine** and **boeravinone B** against a panel of seven targeted liver-associated therapeutic protein targets (PDB IDs: *8BHE,5LDO, 2R3X, 3NXU, 4IQK, 2C6U and 2HI4*). The binding affinities, orientation and molecular interactions of these phytochemical ligands in the catalytic pockets of enzymes were systematically investigated for comparison with standard synthetic hepatoprotective drugs namely Ursodeoxycholic acid, N-Acetylcysteine, Metdoxine & Bifendate, which are used as hepatoprotective. In this context, the present comparative study aims to elucidate the exact molecular mechanisms, receptor binding preferences, and structural efficacy of punarnavine and Boeravinone B in alleviating hepatic stress and improving liver function by comparing the docking scores and hydrogen bonding profiles of the natural compounds with the established synthetic controls.

Table No. 1: Binding affinity for phytochemicals with target enzymes.

Target Enzymes	Binding Affinity (KJ/Mol)					
	Punarnavine	Boeravinone-B	Ursodeoxycholic Acid	N-Acetylcysteine	Metadoxine	Bifendate
8BHE	-9.9	-9.4	-9.5	-5.0	-5.5	-7.6
5LDO	-6.7	-7.5	-7.1	-4.8	-5.7	-7.0
2R3X	-9.9	-9.8	-9.7	-4.9	-5.8	-7.3
3NXU	-8.3	-8.9	-9.2	-4.8	-5.5	-6.7
4IQK	-9.3	-9.9	-9.7	-4.8	-6.4	-7.6
2C6U	-7.0	-7.4	-7.9	-4.3	-5.4	-5.8
2HI4	-10.9	-11.6	-7.1	-4.9	-6.6	-7.6

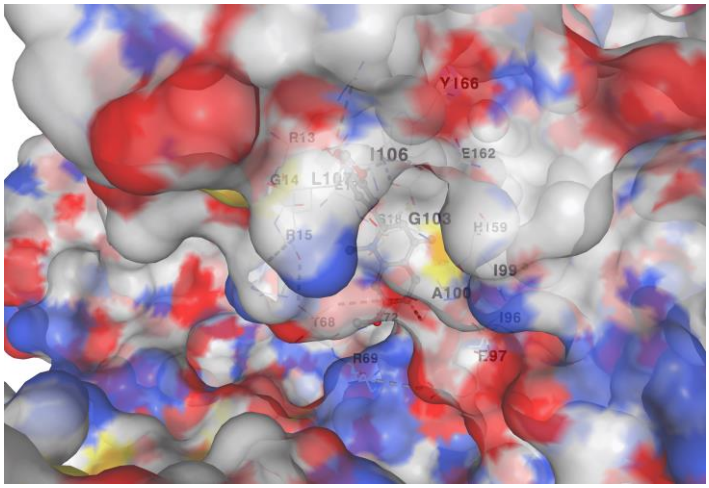

Fig. 7: Represent the Binding affinity of punarnavine in 8BHE Target enzymes.

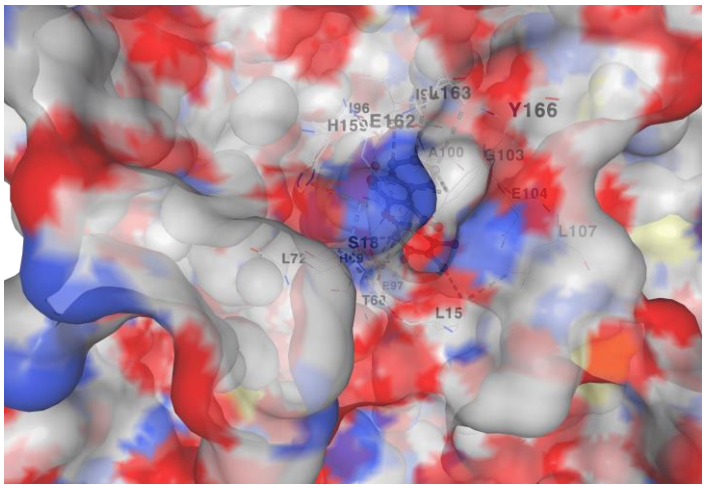

Fig. 8: Represent the Binding affinity of Boeravinone B in 2R3X Target enzymes.

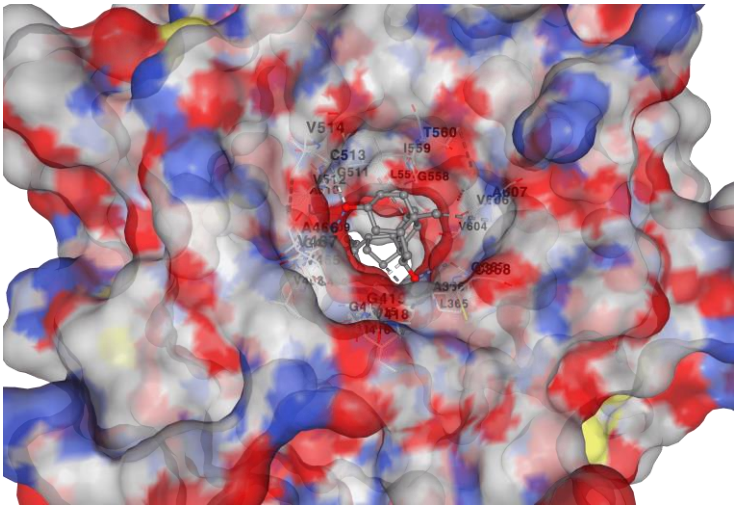

Fig. 9: Represent the Binding affinity of ursodeoxycholic acid in 4IQKTarget enzymes.

The molecular docking studies revealed that the natural alkaloid (Punarnavine) and the natural rotenoid (Boeravinone-B) showed better binding affinities (more negative values)

against almost all the seven target enzymes than the known synthetic controls N-Acetylcysteine, Metadoxine and Bifendate. Besides, both natural compounds are very competitive with the best synthetic control, Ursodeoxycholic Acid, in the dataset. Punarnavine and Boeravinone-B are either equal or far better than the synthetic control on some targets like 8BHE, 2R3X and 2HI4 (where Boeravinone-B reaches the highest affinity of -11.6 KJ/Mol). In general, the binding strength of the Rotenoid (Boeravinone-B) is marginally higher than the Alkaloid (Punarnavine) in relation to most of the targets, but both the natural molecules show excellent and scientifically validated potential which is comparable or more than the standard synthetic drugs at the molecular level which strongly supports the hepatoprotective profile of the plant.

CONCLUSION

This review is on *Boerhavia diffusa* Linn. as an important therapeutic herb by the successful integration of ancient ethnobotanical traditions and contemporary pharmacological validation. The plant has a complex chemical composition and contains bioactive markers such as punarnavine, liriodendrin and a unique group of rotenoids called boeravinones. The plant has wide-ranging antimicrobial, anti-inflammatory and nephroprotective properties. Most interestingly, it performs significant hepatoprotective potential via a complex, multi-targeted molecular network that reestablishes endogenous antioxidant defenses, modulates Nrf2/ARE pathways to activate cytoprotective proteins, inhibits NF-κB-driven inflammatory cascades and modulates Cytochrome P450 enzymes to stabilize hepatocyte membranes. Additional validation for this mechanical pathway is provided by molecular docking data suggesting that isolated compounds such as Punarnavine and Boeravinone B show binding affinities equal to standard synthetic hepatoprotective drugs. These findings ultimately validate the traditional classification of *B. diffusa* as a rejuvenative agent and emphasize its enormous potential as a scientifically validated blueprint for the development of novel, targeted therapies for metabolic, Inflammatory and Hepatic disorders.

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