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Gut-brain axis modulation of *Prosopis juliflora* pods: Integrated *in silico* and *in vivo* approach for neuroprotective and gut restorative therapeutics

Preyenga R[1]*, A Anita Margret[1], R Gayathri[2], Sujidha S[1] & Lokeshwaran S[1]

[1]Bishop Heber College (Autonomous), Department of Biotechnology, Affiliated to Bharathidasan University, Tiruchirappalli-620 017, Tamil Nadu, India

[2]Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical sciences, Chennai-600 077, Tamil Nadu, India

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The invasive spread and ecological burden of *Prosopis juliflora*, especially its adverse effects on groundwater resources, underscore the urgent need for sustainable ways to utilize this species. This study aimed to valorize *P. juliflora* by exploring the gut health - promoting, metabolic, and therapeutic potential of its pod extracts. The phytochemical constituents of the aqueous pod extract were characterized using Gas Chromatography-Mass Spectrometry (GC-MS) and Fourier Transform Infrared (FTIR) spectroscopy. Network pharmacology software tools were used to identify the therapeutic significance and gene-level interactions of key bioactive compounds. The zebrafish (*Danio rerio*) model served as an *in vivo* system to evaluate toxicity and gut restoration effects. Analytical characterization using GC-MS and FTIR confirmed the presence of important phenolic compounds, alkaloids, flavonoids, and glycosides in the extract. Histopathological evaluation revealed a dose-dependent toxic response, mainly associated with phenolic and flavonoid constituents, with a calculated probit LC_{50} of 15.20 ppm. Network pharmacology analysis revealed strong interactions between the extract's bioactive compounds and several neuroprotective and gut-related targets, including GPR30, ESR1, and PPARG. These interactions suggest significant therapeutic potential for supporting neurological health and modulating inflammatory bowel conditions. The *in vivo* findings further underscore the importance of dose optimization to achieve the best therapeutic outcomes with minimal toxicity, indicating the biomedical potential of *P. juliflora* in treating gut-brain axis related disorders.

Keywords: Estrogen receptors, GPCR receptors, Gut microbiome, Inflammatory bowel disease, Neuroprotective agents, Phytochemicals, Plant extracts, Zebrafish

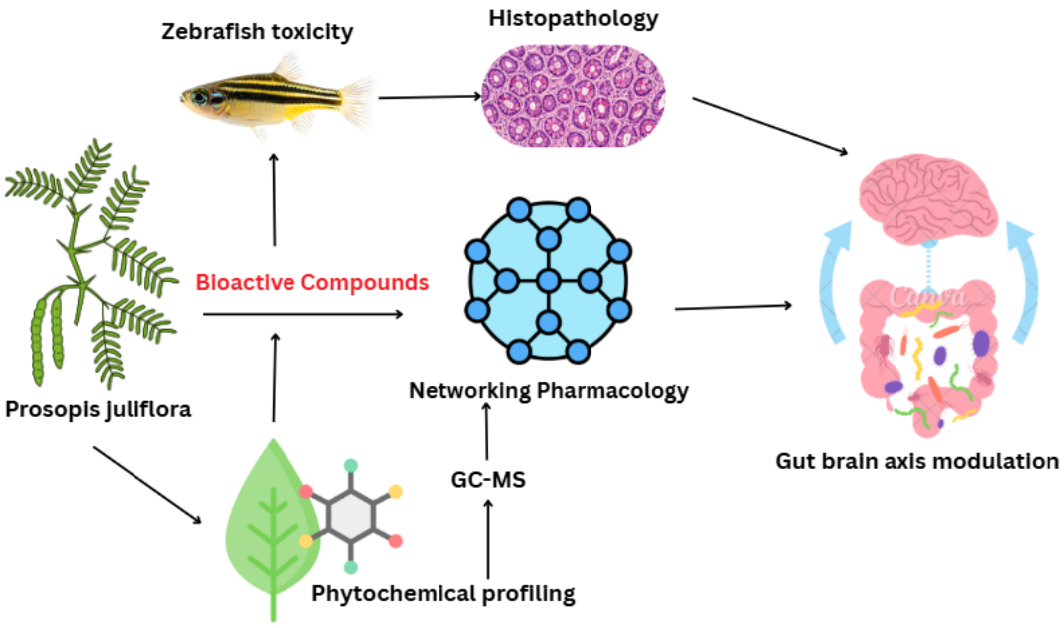
Prosopis juliflora, despite its deleterious environmental impact, *Prosopis juliflora* has the potential to help manage a variety of health issues[1-3]. Studies have demonstrated that the bioactivities of the plant include antioxidant and anti-inflammatory activities that play a crucial role in the prevention of Parkinson's and Alzheimer's disease. These properties contribute to the use of pods in the treatment of neurodegenerative disorders[4]. Thus, it is imperative to evaluate the safety and effectiveness of *Prosopis juliflora*[5,6].

*Correspondence:
E-mail: preyengaramesh@gmail.com

Abbreviations: AChE, Acetylcholine Esterase; ESR1, Estrogen receptor; FTIR, Fourier Transform Infrared Spectroscopy; GCMS, Gas Chromatography Mass spectroscopy; GPR30, G-Protein Coupled receptor; HDAC4, Histone Deacetylase 4; KEGG, Kyota Encyclopedia of Genes and Genomes; NFKB1, Nuclear Factor Kappa B subunit; NQO1, NAD (P)H Quinone Dehydrogenase 1; OECD, Organization Economic Co-operation and Development; PPARG, Peroxisome proliferator – Activated receptor Gamma; SIRT1, Sirtuin 1; TLR4, Toll-Like Receptor 4; YWHAB, Tyrosine 3-Monooxygenase/Tryptophan 5-Monooxygenase Activation Protein Zeta

Prosopis juliflora uncover their significance because of their proficient use and various nutritional aspects. These pods are also rich in protein, fiber, and amino acids, and where they are consumed as food for animals, they boost their health and production, making them healthy for animal diets[7]. In conventional medicine, the pods are believed to have therapeutic uses or to treat disorders of the digestive system and infections, as they possess antimicrobial and anti-inflammatory properties. The succulent tasty edible pulp of pods is also used in preparing food products such as flour and drinks, which is highly beneficial to food security in arid zones[8-10]. In addition, investigations of the pod have focused on nutrient composition and possible beneficial effects, including antioxidant and hypoglycemic activity. In conclusion, *P. juliflora* are potential products for agricultural and biomedical applications[11].

The pharmacological importance of plant-based products for the treatment of health issues has attracted attention because of their versatile bioactive compounds and use in ethnopharmacology. The *Prosopis juliflora* plant has been extensively documented in traditional



Graphical abstract

medicine; it has potential as a therapeutic agent spanning all crucial domains of human health[12]. This study aimed to assess the treatment potential of *Prosopis juliflora* pods by using a combined approach of *in silico* molecular profiling and *in vivo* assessment.

Here, the network pharmacology approach with molecular modeling helps elucidate how *P. juliflora* constituents influence the target biological network. The bioactive compounds related to gut health were analyzed using Cytoscape and information was gathered from PubChem, Binding DB, KEGG, and Disgenet[13]. Animal models were used for *in vivo* analysis to confirm the safety and efficacy of *P. juliflora* pods as medicines. In the present study, zebrafish were used to investigate the effects of aqueous extracts on the gut. The toxicity of the extract, behavioural changes, and histological evaluations of zebrafish have been studied[14].

An effective way to evaluate the efficacy of *Prosopis juliflora* pods was made possible by a combination of *in silico* and *in vivo* research. It is crucial to position them within the framework of coordination with GI health maintenance, and to clarify how they might serve as the foundation for future research and therapies.

Materials and Methods

Plant sample collection

Healthy, mature *Prosopis juliflora* pods were harvested from agricultural land in Woraiyur, Trichy (latitude 10.8308° N, Longitude 78.6799°E), Tamil Nadu, India. After removing the pods, they were completely washed to eliminate dust particles and air-dried for 2–3 days. Subsequently, the dried pods were homogenized into a fine powder and stored in an airtight container.

Preparation of pod extract

Aqueous and ethanolic extracts from various powdered plant parts were obtained using the Soxhlet extraction method, conducted at boiling points of 100°C for water and 70°C for ethanol, over 5–6 continuous cycles[15]. Required amount of plant powder was weighed and placed inside the thimble in Soxhlet extractor. Using water or ethanol as solvents, repeated evaporation and condensation cycles enabled efficient phytochemical extraction. The resulting extracts were then concentrated and stored for further analysis.

Phytochemical profiling- qualitative analysis

Phytochemical screening is a preliminary test used to identify bioactive compounds in samples extracted from bark, flowers, pods, and leaves, using standard protocols[16].

Characterization of plant extract

FTIR analysis

The presence of functional groups in the potential extract of *Prosopis juliflora* extract was identified using Fourier Transform Infrared Spectroscopy (FTIR) with a Thermo Nicolet Avatar 370. Spectroscopic analysis was conducted in the KBr mode at a ratio of 1:100, scanning across a range of 400–4000 cm^{-1} with 4 cm^{-1} resolution[17].

GC-MS analysis

The extract was analyzed for the phytochemicals present in the *Prosopis juliflora* pods using Gas Chromatography-Mass Spectrometry (GC-MS) on SHIMADZU QP 2020 using an SH-rxi-5sil ms column with the carrier gas being an inert one like helium neon and argon. This setup included an AOC-20i auto sampler and a GC connected to a mass spectrometer. The operating parameters were as follows: Among the conditions that may be preset include injector flow rate of 1.2 mL/min for the injector, a temperature for the injector at 250°C and temperature of ion source at 200°C. The thermal regime was fixed at a temperature of 50°C for 2 min, then the temperature slope raised to 280°C at a rate of 20°C/s, and the sample was kept at 280°C for 2 min[18]. MS spectra were obtained at a scan time of 0.3 sec in the m/z range of 50-500 for fragmented ions. The compound was identified based on GC-MS compared to mass spectra using the WILEY8LIB online database[19].

Network pharmacology

Network pharmacology evaluates a drug's effectiveness by using data from Cytoscape to display the effect of the drug on certain metabolic pathways or nodes, as well as the network of communications between the drug and the target molecules. Ideally, drugs permit binding only to the target location within the molecule. This was accomplished by obtaining data on the identified bioactive chemicals from the PubMed and PubChem databases, identifying the target receptors from the Binding Database, and converting UniProt identities to the Kyoto Encyclopedia of Genes and Genomes (KEGG) to ascertain their function in metabolism. The DISGENET database was used to obtain disease-gene connections. These data were processed and stored in an Excel file and then transferred into Cytoscape, where a network diagram was built and used to improve the effectiveness of the drug and enhance treatment strategies[13].

In vivo studies - Zebrafish model for probit analysis

The Ethical clearance for using zebrafish (*Danio rerio*) was obtained (No: IHEC/SDC/FACULTY/24/BIOCHEM/049) from the Institutional Animal Ethics Committee. An Adult zebrafish (Age: 1-2 months) were obtained from a local supplier and habituated to the test conditions for approximately two weeks. To perform the probit analysis, the zebrafish were treated with *P. juliflora* pod extract as the treated group, and another group not treated with the extract served as a control. Toxicity was observed over 72 h following the Organization for Economic Co-operation and Development (OECD) guidelines (Test no: 236, 2013)[20]. Pod extracts (120 µL, 160 µL, and 240 µL) containing 12, 16, and 24 ppm, respectively, were introduced into the fish habitat. The fish were observed for 72 h for mortality and behavioural alterations. Dead fish were autopsied, and their brains, along with their abdominal viscera (gut), were kept in 1 % formalin solution for histopathological studies[14].

Results

Fresh bark, leaves, flowers, and pods of *Prosopis juliflora* were collected from Woraiyur, Tiruchirappalli, Tamil Nadu, and shade-dried for 5–10 days. Ethanol and aqueous extracts were prepared using Soxhlet extraction (Fig. 1) and analysed for phytochemicals using standard qualitative methods. The results are depicted in the heat map (Fig. 2).

Qualitative heatmap analysis of aqueous and ethanol extracts from different *Prosopis juliflora* parts

Fig. 1 — Soxhlet extraction

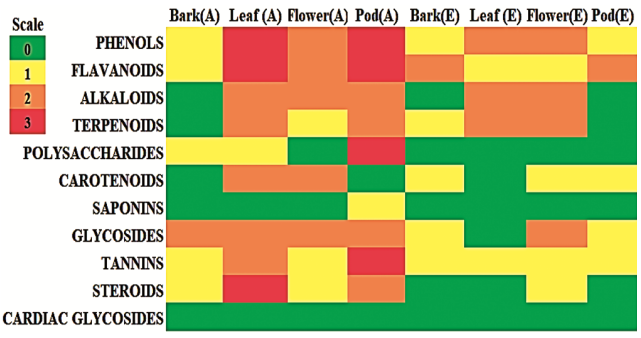


Fig. 2 — Heat map demonstrating bioactive profiling of different parts of *P. juliflora*

revealed distinct phytochemical profiles. Aqueous extracts showed high levels of phenols, flavonoids, and tannins in leaves and pods, with moderate alkaloids and polysaccharides. Ethanolic extracts contained moderate terpenoids and carotenoids, especially in leaves and flowers. Cardiac glycosides were found to be absent in all samples. The aqueous extract of pods exhibited the abundant phytochemical content and were employed for further research study.

GC-MS analysis of the aqueous extract of *P. juliflora* pods established 20 different bioactive compounds (Fig. 3). Key bioactives included 2-Keto butyric acid, 4-Allyl-2-methoxyphenol, 4',5,7-Trihydroxyflavone and Hexadecanoic acid, well known for their therapeutic and pharmaceutical applications. The detailed list of the bioactive compounds is provided in (Table 1).

FTIR analysis (Fig. 4) revealed functional groups corresponding to phenols, alcohols, carboxylic acids, alkanes, ethers, and various amines. Preliminary qualitative phytochemical screening of *P. juliflora* pod extract identified the presence of flavonoids, alkaloids, tannins, terpenoids, saponins, anthraquinones, and glycosides.

FTIR spectrum of aqueous *P. juliflora* pod extract showed notable key absorption peaks demonstrating active functional groups. The broad band appearing at 3434.99 cm^{-1} corresponds to O-H stretching (alcohols), whereas peaks between 2981.61-2904.41 cm^{-1} indicate the CHO-aldehyde groups. The prominent peak at 2095.24 cm^{-1} revealed silicon-containing compounds. The peak appeared at 1639.44 cm^{-1} was associated with stretching corresponding to C=C or C=N groups. The peaks approaching from 1405.16 - 452.39 cm^{-1} reflect C-O, C-O-C, and C-OH bonds, whereas spectral bands showing at 877.34 cm^{-1} and 666.22 cm^{-1} represent -CH bending and C-Cl stretching (alkyl halides), respectively.

The interactions between *P. juliflora* bioactive compounds and key biological targets for

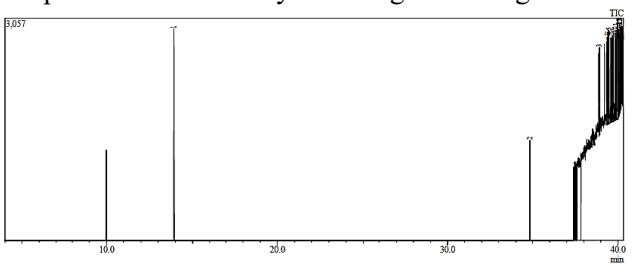


Fig. 3 — GCMS spectrum of *Prosopis juliflora* pods -Aqueous extract

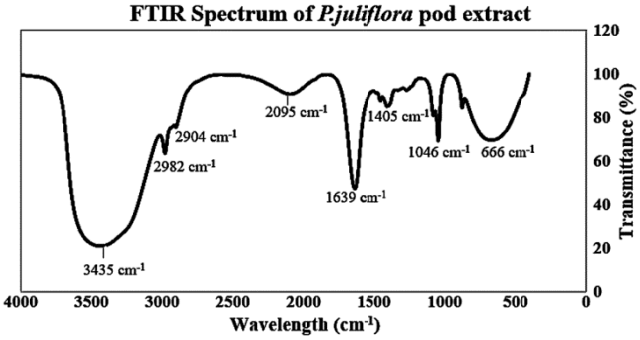


Fig. 4 — FTIR Spectrum of *Prosopis juliflora* pod extract

Table 1 — GCMS Analysis of aqueous extract of *Prosopis juliflora* pods

S. No	Compound name	Retention time	Molecular formula	Molecular weight (g/mol)
1.	2 – Keto butyric acid	4.764	$C_4H_6O_3$	102.09
2.	1-Methyl-4-(1-methylethenyl)-cyclohexene	6.391	$C_{10}H_{16}$	136.23
3.	1,3-Dioxolane, 2-[2-(4-benzyloxy) phenylethyl]-	6.745	$C_{18}H_{20}O_3$	284.3
4.	Cyclopentasiloxane, Decamethyl-	6.823	$C_{10}H_{12}O_2$	164.2
5.	4',5,7-Trihydroxyflavone	9.142	$C_{15}H_{10}O_5$	270.24
6.	4-Allyl-2-methoxyphenol	9.949	$C_{10}H_{30}O_5Si_5$	370.77
7.	Cyclohexasiloxane, dodecamethyl-	13.869	$C_{12}H_{36}O_6Si_6$	444.92
8.	Cycloheptasiloxane, tetradecamethyl-	17.48	$C_{14}H_{42}O_7Si_7$	519.07
9.	Benzoic acid, 2,4-bis(trimethylsiloxy)-, trimethylsilyl ester	20.714	$C_{16}H_{30}O_4Si_3$	370.66
10.	9Z,12Z)-Octadeca-9,12-dienoic acid	23.511	$C_{18}H_{32}O_2$	280.4
11.	1,3-diphenyl-1-((trimethylsilyl)oxy)-1(z)-heptene	25.998	$C_{22}H_{30}OSi$	338.6
12.	Hexadecanoic acid	28.267	$C_{16}H_{32}O_2$	256.42
13.	Ethyl 2,3,4,6-tetra-o-acetyl-1-thio-beta-d-glucopyranoside	36.316	$C_{16}H_{24}O_9S$	392.4
14.	(9Z)-Octadec-9-enoic acid	38.021	$C_{18}H_{34}O_2$	282.5
15.	(9Z)-Octadec-9-enoic acid	38.155	$C_{18}H_{34}O_2$	282.5
16.	N-(T-butyl)-2-benzoylbenzamide	38.193	$C_{18}H_{19}NO_2$	281.3
17.	3,4,5-Trihydroxybenzoic acid	38.286	$C_7H_6O_5$	170.12
18.	1-Methyl-4-(1-methylethenyl)-cyclohexene	39.045	$C_{10}H_{16}$	136.23
19.	1-Methyl-4-(1-methylethenyl)-cyclohexene	39.675	$C_{10}H_{16}$	136.23
20.	Ethanone, 1-[4-[(1,1-dimethylethoxy)methyl]phenyl]-	40.292	$C_{13}H_{18}O_2$	206.28

neuroprotection and gut dysbiosis was elucidated in network pharmacological studies (Fig. 5). The green nodes of network indicate GC-MS bioactive compounds, while orange nodes suggest target proteins involved in neurological and gut-related functions. Remarkable targets having UniProt IDs such as Q9Y5P9 - GPR30, P03372 - ESR1, and P63104 - 14-3-3 protein, were found to be linked with neuroprotection, and other receptors like P35354 - CB1 receptor, P55011- PPARG, and Q96P20 - GPR55, were associated with gut microbiome health. Therefore, the pharmacological network underscores the therapeutic relevance of the bioactive compounds for major diseases like diabetes, inflammation, cancer, and microbial infections.

Figure 6 represents the gene-disease associations linking to neuro-protective and gut restoration of pod bioactives by network interactions

. Significant neuroprotective genes like PPARG, SIRT1, NQO1, and HDAC4, were involved in Alzheimer's, Parkinson's, and neuroinflammation pathways. Inflammatory conditions are linked to TLR4 and NFKB1, which play evident roles in immune response and metabolic disorders. Overall, the network underscores the potential of these genes in combating oxidative stress and inflammation in both neurological and gut-related diseases.

Histopathological analysis of zebrafish treated with antibiotics and *P. juliflora* aqueous pod extract was performed using hematoxylin and eosin staining. Control zebrafish gut and brain tissues, without pod extract treatment, are shown in (Figs. 7a & 8a), respectively.

Figure 7 shows histological changes in zebrafish gut tissue. The control (Fig. 7a) displays intact villi and glial cell distribution. Treatment with a low concentration of *P. juliflora* pod extract (15–20 ppm, Fig. 7b) resulted in effective restoration of gut architecture. Figure 7c indicates the cytotoxic effects of treating zebrafish at higher pod concentrations which markedly shows structural damages such as mucosal layer disruption, distortion of epithelial layer and disorganised villi structure. While employing lower concentration, the extract showed beneficial effects in gut restoration by supporting mucosal integrity and retaining villi

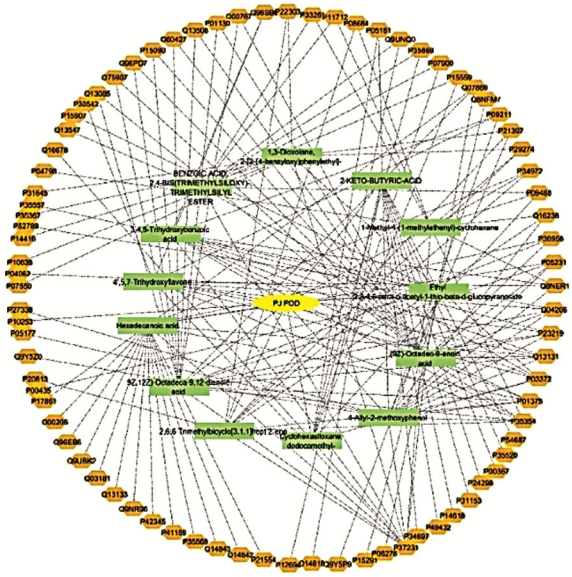


Fig. 5 — Pharmacological networking of bioactives and targets of *P. juliflora* pod extract

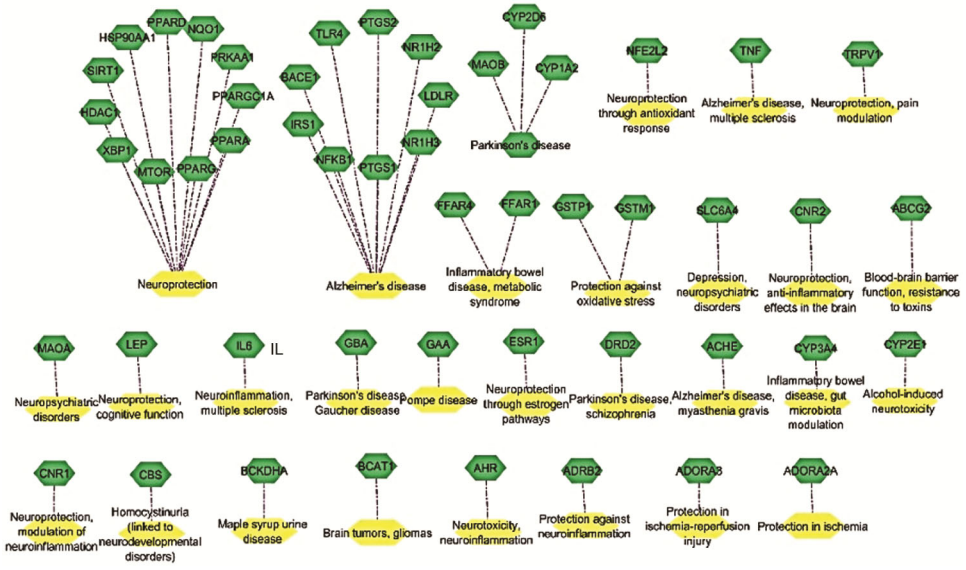


Fig. 6 — Networking of gene-disease associations of *P. juliflora* pod extract

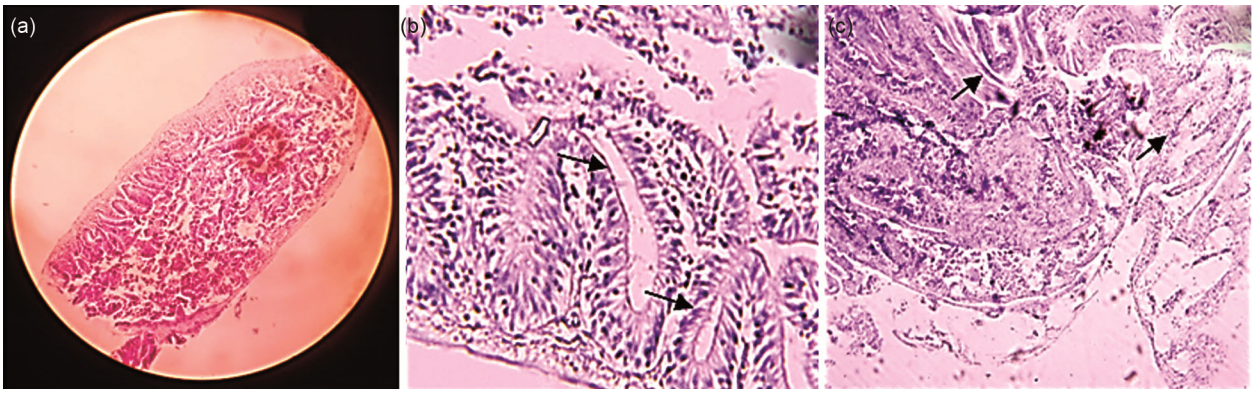


Fig. 7 — Histopathology results of Zebra fish (a) Gut Control; (b) Gut treated at low concentration of Pod extract; and (c) Gut treated at high concentration of Pod extract

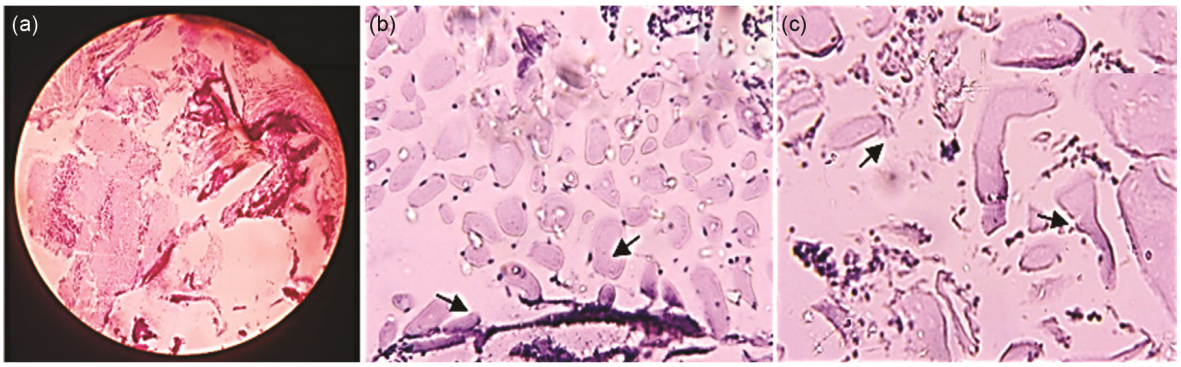


Fig. 8 — Histopathology results of Zebra fish (a) Control - Brain; (b) Brain treated at low concentration of Pod extract; and (c) Brain treated at high concentration of Pod extract

morphology. These findings denote that the lower concentrations of extractions were found to be safe and effective for treating gut dysbiosis whereas the higher dosages show detrimental effects to gut tissue.

Figure 8b illustrates the histopathological examination of brain tissues treated with low-dosage of pod extracts which revealed restoration of neural architecture whereas treating with higher doses of extract altered disorganization of neural tissue and vacuolization, suggesting neurotoxic effects (Fig. 8c). The LC_{50} value 15.20 ppm of the pod extract showed beneficial and positive effects on gut villi and mucosal barrier. Conclusively, the *P. juiliflora* pod extract having low-dosage when administered at optimal concentrations exert potential therapeutic benefits in supporting gut-brain axis modulation.

Discussion

The phytochemical profiling of aqueous pod extract of *Prosopis juliflora* retains versatile bioactives including alkaloids, flavonoids, tannins, saponins, terpenoids, and glycosides, each exerting pharmacological activities including free-radical scavenging property, anti-inflammatory, and protective efficacy against toxic agents[21,22]. The gas-chromatography analysis revealed bioactives of pod extract and FTIR spectral analysis further confirmed key functional groups including alcohols, aldehydes, esters, hydroxyls, and carboxylic acids[23]. The protein-target interaction studies of pod extract showed significant relevance to gut-brain axis. The previous literature works showed that *Juliflorine* bioactive and many flavonoid and phenolic compounds of *P. juliflora* reduced acetylcholinesterase (AChE) activity in a dose-dependent manner[24]. The pharmacological network analysis further shows evidences of interactions involving GPR30, ESR1 and YWHAB which are involved in pathways associated with neurodegenerative conditions such as Alzheimer's and Parkinson's disease[25,26]. Additionally, protein targets like PPARG, CNR1, and GPR55 were found to be linked with gut microbiome health and inflammation[27]. Genes like PPARG, SIRT1, and NFKB1 emerged as central key regulators in controlling oxidative stress and inflammatory responses, helping to manage both neurological and gastrointestinal diseases[28,29].

Histopathological examination of zebrafish (*Danio rerio*) provided important *in vivo* evidence supporting the extract's ability to influence both the gut and brain. At lower concentrations of the aqueous pod extract supported tissue regeneration supporting restorative potential. However, higher doses led to marked cytotoxic effects, including mucosal damage and neurodegenerative changes[30]. The lethal concentration LC_{50} value was calculated to be 15.20 ppm, indicating the threshold for safer dosage administration. These findings underscore the need for dose optimization to balance pharmacological efficacy and safety regulations[31].In summary, the *P. juliflora* pod extract demonstrates clear and promising biological activity, showing potential therapeutic benefits for both gut and brain health. However, further work is needed to standardize and optimize the dosage to enhance its efficacy while reducing possible side effects.

Conclusion

This research work underscores the importance of *Prosopis juliflora*, an invasive plant, as a source of abundant bioactive compounds with therapeutic efficacy by phytochemical profiling. GC-MS and FTIR further confirmed the presence of various key metabolites including phenols, flavonoids, tannins, glycosides, and alkaloids. The protein targets GPR30, ESR1, PPARG associated with neuroprotection and gut health restoration were identified through *in silico* network pharmacology approach. *In vivo* toxicity testing using *Danio rerio* revealed dose-dependent effects, with lower concentration restoring mucosal integrity and neural architecture and higher concentrations causing gut and brain damage. Since this plant holds potential as lignocellulosic biomass, future studies should explore nanolignocellulose-based delivery systems to enhance safety, minimise toxicity, and support gut-brain axis restoration.

Limitations and Future Directions

While this study offers valuable insights of abundant phytochemical profiling with polar and less polar solvents, network pharmacology associations of gut-brain, and *in vivo* histopathological effects of *Prosopis juliflora* pod extract, it did not include direct *in vitro* neuroprotective or gut-restorative assays. Future studies would incorporate *in vitro* validation, such as neuronal cell viability assays, oxidative stress assessments, and gut epithelial integrity tests, to substantiate/validate the mechanistic pathways involved in gut-brain axis regulation. These approaches would further strengthen the therapeutic relevance and efficacy of *P. juliflora* pods for gut-brain axis modulation.

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Conflict of interests

All authors declare no conflict of interests.

References

1 Mishra K, An overview of *Prosopis juliflora*'s pharmacologic aspects. *Int J Pharmacogn Pharm*, 4 (2023) 121.
2 Mungoche J, Wasonga OV, Ikiror D, Akala H, Gachuiri C & Gitau G, *Prosopis juliflora* (Sw.) DC in the drylands: A review of invasion, impacts and management in Eastern Africa. *Sustain Environ*, 11 (2025) 2521946. .
3 Kumar V, Mahajan N, Kumar V & Sharma A, Valorization of *Prosopis juliflora* pods as livestock feed and functional food ingredient: A review. *Plant Arch*, 20 (2020) 1130.
4 Ukande MD, Shaikh S, Murthy K & Shete R, Review on pharmacological potentials of *Prosopis juliflora*. *J Drug Deliv Ther*, 9 (2019) 755.
5 Mahmoud DA, Sharaf AM & Saleh NA, Exploration of the antimicrobial and antioxidant potential of *Prosopis* species: Towards novel therapeutic applications. *S Afr J Bot*, 139 (2021) 325.
6 Shoaib S, Ansari MA, Fatease AA, Safhi AY, Hani U, Jahan R & Islam N, Plant-derived bioactive compounds in the management of neurodegenerative disorders: Challenges, future directions and molecular mechanisms involved in neuroprotection. *Pharmaceutics*, 15 (2023) 749.
7 Zhong J, Lu P, Wu H, Liu Z, Sharifi-Rad J, Setzer WN & Suleria H, Current insights into phytochemistry, nutritional and pharmacological properties of *Prosopis* plants. *Evid Based Complement Altern Med*, 1 (2022) 2218029.
8 Chaudhary LB & Shukla RP, Reassessing the ecological and medicinal significance of invasive *Prosopis juliflora*. *Ecol Process*, 11 (2022) 45.
9 Jain S, Prajapati R & Verma A, Nutritional and therapeutic profiling of *Prosopis juliflora* pod extract: An *in vitro* and *in vivo* approach. *Biocatal Agric Biotechnol*, 51 (2023) 102504.
10 Sharifi-Rad J, Kobarfard F, Ata A, Ayatollahi SA, Khosravi-Dehaghi N, Jugran AK & Martins N, *Prosopis* plant chemical composition and pharmacological attributes: Targeting clinical studies from preclinical evidence. *Biomolecules*, 9 (2019) 777.
11 Henciya S, Seturaman P, James AR, Tsai YH, Nikam R, Wu YC & Chang FR, Biopharmaceutical potentials of *Prosopis* sp. (Mimosaceae, Leguminosae). *J Food Drug Anal*, 25 (2017) 187.

12 deLemos ABS, Chaves G, Ribeiro PPC & daSilva Chaves Damasceno KSF, *Prosopis juliflora*: Nutritional value, bioactive activity and potential application in human nutrition. *J Sci Food Agric*, 103 (2023) 5659.
13 Sekaran K, Karthik A, Varghese RP, Sathiyarajeswaran P, Devi MS, Siva R & Doss CGP, *In silico* network pharmacology study on *Glycyrrhiza glabra*: Analyzing immune-boosting phytochemical properties against COVID-19. *Adv Protein Chem Struct Biol*, 138 (2024) 233.
14 Chahardehi AM, Arsad H & Lim V, Zebrafish as a successful animal model for screening toxicity of medicinal plants. *Plants*, 9 (2020) 1345.
15 ZambariI F, Hafid SRA & Muhamad NA, Optimisation of extraction method and phytochemical compounds of *Christia vespertilionis* leaves using GC-MS. *Int J Pharm Sci Rev Res*, 70 (2021) 1.
16 Evans WC, *Trease and Evans' Pharmacognosy*. Elsevier Health Sciences, London, (2009) 1.
17 Malik SK, Ahmed M & Khan F, Identification of novel anticancer terpenoids from *Prosopis juliflora* pods. *Trop J Pharm Res*, 17 (2018) 661.
18 Sundhararajan R, Sabarisenthil Salam NA, Rilvan MF, Sathish B, Kumar RS, Ahmed HS & Jena D, GC-MS analysis of chemical substances from ethanolic extract of *Prosopis juliflora* leaves. *J Phytopharmacol*, 12 (2023) 253.
19 Fiehn O, Metabolomics by gas chromatography–mass spectrometry: Combined targeted and untargeted profiling. *Curr Protoc Mol Biol*, 114 (2016) 30.
20 OECD, Test no. 236: Fish embryo acute toxicity (FET) test. *OECD Guidel Test Chem*, 2 (2013) 1.
21 Patnaik P, Abbasi T & Abbasi SA, *Prosopis juliflora*: Blessing and bane. *Trop Ecol*, 58 (2017) 455.
22 Naik NM, Krishnaveni M & Mahadevswamy M, Characterization of phytocomponents with antimicrobial traits in *Prosopis juliflora* leaves using GC-MS. *Sci Rep*, 13 (2023) 4064.
23 Berwal R, Vasudeva N, Sharma S & Das S, Investigation of biomolecules in ethanol extract of *Prosopis juliflora* fruits using GC-MS. *J Herbs Spices Med Plants*, 25 (2019) 172.
24 Choudhary MI, Nawaz SA, Azim MK, Ghayur MN, Lodhi MA, Jalil S & Ahmad VU, Juliflorine: A potent acetylcholinesterase inhibitor with calcium-channel blocking potential. *Biochem Biophys Res Commun*, 332 (2005) 1171.
25 Safe S, Kim K & Lee SO, Transcriptional activation of genes involved in neuroprotection by estrogen receptor signaling. *Endocrinology*, 149 (2008) 2798.
26 Zhang L & Zhang YQ, GPR30 and neuroprotection: Implications for neurodegenerative diseases. *Front Aging Neurosci*, 10 (2018) 1.
27 Peyrin-Biroulet L, Beaulieu DB, Iacucci M & Somers M, Role of PPARγ in inflammatory bowel disease. *Gut*, 67 (2018) 1.
28 Cario E, Toll-like receptors in inflammatory bowel diseases: A decade later. *Inflamm Bowel Dis*, 16 (2010) 1583.
29 Wang Y & Sun Y, The PPARG signaling pathway in inflammatory bowel disease. *Front Immunol*, 11 (2020) 1.
30 Menke AL, Spitsbergen JM, Wolterbeek AP & Woutersen RA, Normal anatomy and histology of the adult zebrafish. *Toxicol Pathol*, 39 (2011) 759.
31 Henciya S, Seturaman P, James AR, Tsai YH, Nikam R, Wu YC & Chang FR, Biopharmaceutical potentials of *Prosopis* sp. *J Food Drug Anal*, 25 (2017) 187.