



Plant-derived phytochemicals as potential antibacterial agents for foodborne pathogen, *Campylobacter jejuni* targeting *guaB*-encoded IMPDH

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Abstract

Aim: This study aimed to identify novel plant-derived inhibitors targeting the *guaB*-encoded inosine monophosphate dehydrogenase (IMPDH, inosine 5'-monophosphate dehydrogenase) of the foodborne pathogen *Campylobacter jejuni* to address the rising global threat of multidrug-resistant infections.

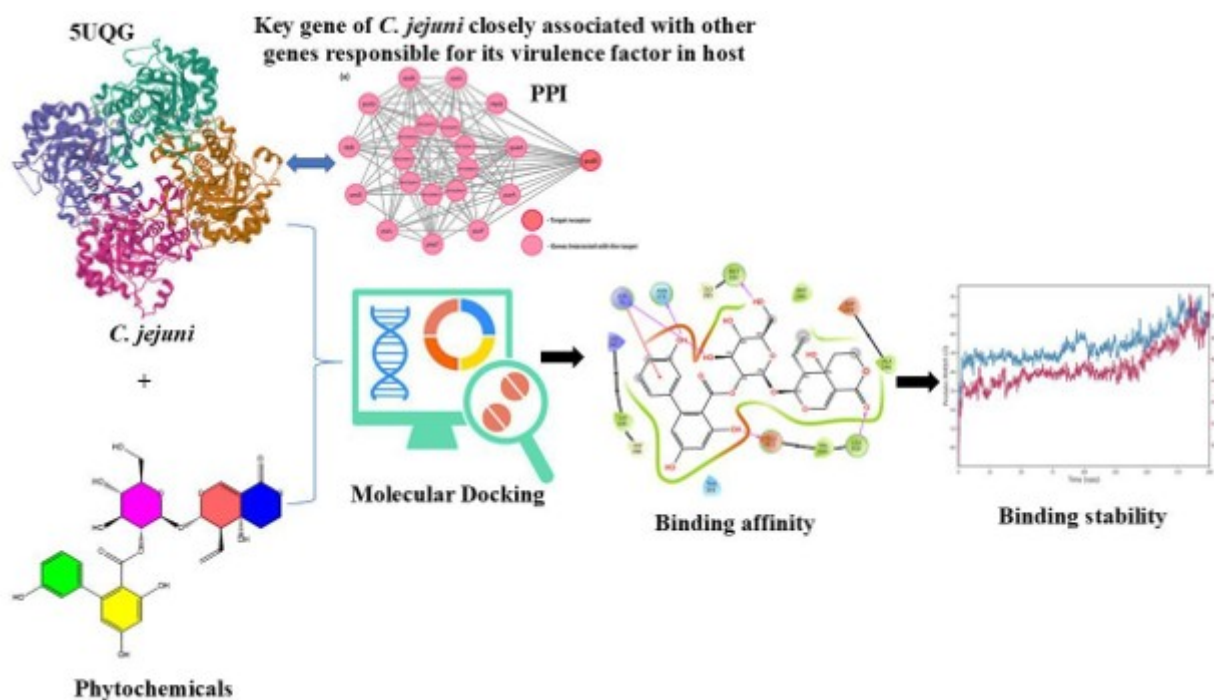
Method: The present research aimed to identify a novel candidate from medicinal plants using various drug discovery approaches, including network pharmacology, molecular docking, molecular dynamics (MD) simulations, and ADMET (absorption, distribution, metabolism, excretion, and toxicity) profiling to assess pharmacokinetic properties.

Results: Molecular docking studies revealed that amaroswerin and eriocitrin had notable interactions with IMPDH, with docking scores of -8.612 and -8.488 kcal/mol, respectively. MD simulations for 200 ns showed that the amaroswerin-IMPDH complex remained stable until 99 ns, with a root mean square



deviation (RMSD) of $< 2.8 \text{ \AA}$. However, this RMSD gradually increased and reached $4.5 \text{ \AA}$ by the end of the 200 ns trajectory. This indicates that complex underwent conformational adjustment. Amaroswerin showed a significant intestinal absorption rate of 49.29% and a notable permeability rate in Caco-2 cells. These data suggest it may be absorbed into the bloodstream more readily after oral administration. In terms of excretion, amaroswerin had a higher total clearance value of 0.465, indicating that it can be easily excreted from the body. ADMET analysis predicted moderate oral bioavailability and relatively favorable profiles across several toxicity endpoints, supporting amaroswerin as a promising candidate for further investigation. Nevertheless, these predictions are preliminary and require experimental validation of their pharmacokinetic and safety profiles. The organ toxicity and toxicity endpoints revealed that amaroswerin had higher inactive probability rates for hepatotoxicity (0.78), neurotoxicity (0.80), carcinogenicity (0.75), mutagenicity (0.66), and cytotoxicity (0.74), supporting its prioritization for further experimental evaluation.

Conclusions: Based on the current computational findings, amaroswerin may be considered a promising lead compound for further investigation as an inhibitor of *C. jejuni* IMPDH. However, these findings do not establish antimicrobial efficacy or therapeutic suitability. Therefore, comprehensive in vitro and in vivo studies are required to experimentally validate IMPDH inhibition, antimicrobial activity, pharmacokinetic behavior, and safety before any consideration of clinical application.



Graphical abstract. Integrated computational workflow for identifying potential anti-*C. jejuni* phytochemicals. The graphical abstract illustrates the integrated computational approach for identifying potential anti-*C. jejuni* agents. The three-dimensional structure of the target protein (5UQG) and key virulence-associated genes were analyzed through protein-protein interaction (PPI) networks. Selected phytochemicals were subjected to molecular docking against the target protein to evaluate their binding interactions. The best-performing compounds were further analyzed for binding affinity through interaction profiling and assessed for stability using molecular dynamics simulations. This workflow highlights the identification of promising phytochemical candidates with potential inhibitory activity against *C. jejuni*.

Keywords

Campylobacter jejuni, amaroswerin, docking, molecular dynamics, toxicity

Introduction

Campylobacter jejuni is the principal causative agent of acute bacterial gastroenteritis in humans. It accounts for approximately 400–500 million cases of diarrheal diseases each year [1, 2]. According to recorded food infection-related clinical cases, nearly 80% of microbial transmissions are caused by the consumption of infected chicken meat [1, 3]. This condition ultimately results in severe health complications and mortality among older adults [3, 4]. However, multidrug-resistant strains have rapidly emerged, presenting a major obstacle to the treatment of campylobacteriosis [2, 5]. They have resistance to ciprofloxacin (31/35, 88.6%) and tetracycline (30/35, 85.7%) [6]. Consequently, *C. jejuni* is a global public health concern regarding food safety. This underscores the need for novel antimicrobial agents that are effective against multidrug-resistant strains.

Many bacterial pathogens, including *Staphylococcus aureus*, *Escherichia coli*, *Acinetobacter baumannii*, and *C. jejuni*, require inosine 5'-monophosphate dehydrogenase (IMPDH) for survival, making this enzyme an attractive target for antibacterial drug development [7–9]. IMPDH catalyzes the rate-limiting conversion of inosine monophosphate (IMP) to xanthosine monophosphate (XMP), a key step in de novo guanine nucleotide biosynthesis required for DNA and RNA synthesis, energy metabolism, and bacterial proliferation [8]. Inhibition of IMPDH depletes the intracellular guanine nucleotide pools, thereby suppressing bacterial growth and survival [9]. Unlike several conventional antimicrobial targets, such as DNA gyrase, topoisomerase IV, and ribosomal proteins, which are increasingly compromised by widespread resistance mechanisms, IMPDH in *C. jejuni* remains underexplored and is a metabolically essential enzyme that faces lower selective pressure from currently used antibiotics. Furthermore, structural differences between bacterial and human IMPDH, particularly in the NAD-binding region, offer opportunities to develop selective inhibitors with reduced host toxicity [9]. These characteristics make IMPDH a promising alternative target for overcoming multidrug resistance in *C. jejuni*.

Plant-derived chemicals have gained significant attention owing to their wide spectrum of pharmacological properties, including antioxidant and anti-virulence effects against pathogenic microorganisms. Numerous studies have documented that plant metabolites can interact with key enzymes and metabolic pathways in microbes, affecting replication, survival, energy metabolism, and pathogenicity [10]. For instance, plant-derived phytochemicals such as baicalein, carvacrol, chrysin, cinnamaldehyde, curcumin, epicatechin, fisetin, honokiol, hydroxytyrosol, kaempferol, luteolin, morin, piceatannol, quercetin, resveratrol, rosmarinic acid, safranal, silymarin, and thymoquinone have been reported to be effective in controlling pathogenic microorganisms by targeting the key enzymatic pathways responsible for survival, replication, pathogenicity, and virulence factors (Table 1) [11–18].

Table 1. Plant-derived phytochemicals are reported to be effective in the inhibition of pathogenic organisms by disturbing various pathways involved in survival, replication, pathogenicity, and virulence factors.

S. No.	Name of the phytochemicals	Scientific name of source plants and common names	Targeting microbial enzyme	Role of microbial enzymes	Literature
1.	Baicalein	<i>Scutellaria baicalensis</i> (Skullcap)	ATP synthase (<i>E. coli</i>)	Inhibit ATP synthesis for cellular energy	[11]
2.	Carvacrol	<i>Origanum vulgare</i> (Oregano)	ATPase activity	Enhances energy disruption	[12]
3.	Chrysin	<i>Passiflora caerulea</i>	Biofilm formation enzymes	Bacterial aggregation	[13]
4.	Cinnamaldehyde	<i>Cinnamomum verum</i> (Cinnamon)	DNA synthesis enzymes	Enhance bactericidal action	[12]
5.	Curcumin	<i>Curcuma longa</i> (Turmeric)	Bacterial enzymes (e.g., membrane-associated)	Inhibit enzymatic activity responsible for growth and virulence	[14]
6.	Epicatechin	<i>Camellia sinensis</i> (Green tea)	ATP synthase	Inhibits energy production	[11]
7.	Fisetin	<i>Fragaria vesca</i> (Strawberry)	Biofilm enzymes (<i>S. aureus</i>)	Inhibit colony growth	[13]

Table 1. Plant-derived phytochemicals are reported to be effective in the inhibition of pathogenic organisms by disturbing various pathways involved in survival, replication, pathogenicity, and virulence factors. (continued)

S. No.	Name of the phytochemicals	Scientific name of source plants and common names	Targeting microbial enzyme	Role of microbial enzymes	Literature
8.	Honokiol	<i>Magnolia officinalis</i>	Multiple membrane enzymes	ESKAPE pathogen resistance	[15]
9.	Hydroxytyrosol	<i>Olea europaea</i> (Olive)	ATP synthase (<i>E. coli</i>)	Membrane-bound energy generation (~60% inhibition)	[16]
10.	Kaempferol	Propolis/Tea	Biofilm enzymes	<i>S. aureus</i> adhesion	[13]
11.	Luteolin	Red wine plants (e.g., <i>Arachis hypogaea</i>)	<i>S. aureus</i> biofilm enzymes	Pathogen inhibition	[13]
12.	Morin	<i>Morus alba</i> (Mulberry) or <i>Maclura tinctoria</i>	ATP synthase (<i>E. coli</i>)	Oxidative phosphorylation	[11]
13.	Piceatannol	<i>Vitis vinifera</i> (Grape) or Rheum species	ATP synthase	Proton translocation and ATP formation	[16]
14.	Quercetin	Quercetin-rich plants (onions, apples, <i>Psidium guajava</i>)	Nucleic acid synthesis enzymes	Disrupt protein synthesis and DNA replication	[17]
15.	Resveratrol	<i>Vitis vinifera</i> (Grape)	Agr-mediated QS enzymes (<i>S. aureus</i>)	Inhibit toxin production and biofilm formation	[18]
16.	Rosmarinic acid	<i>Rosmarinus officinalis</i> (Rosemary)	Quorum sensing enzymes (LuxI-type)	Inhibit the virulence factor's ability	[18]
17.	Safranal	<i>Crocus sativus</i> (Saffron)	ATP synthase	Disrupt energy metabolism in bacteria	[16]
18.	Silymarin	<i>Silybum marianum</i> (Milk thistle)	ATP synthase	Cellular respiration and growth	[11]
19.	Thymoquinone	<i>Nigella sativa</i> (Black cumin)	ATP synthase	ATP production and energy generation	[16]

The six phytochemicals evaluated in this study (amaroswerin, eriocitrin, peganine, afzelechin, amarogentin, and naringenin) were selected because they represent structurally diverse classes of plant-derived bioactive compounds with previously reported antimicrobial, antioxidant, or anti-inflammatory activities. Yet, their inhibitory potential against *C. jejuni* IMPDH has not been systematically investigated. Their structural diversity, spanning flavonoids, secoiridoid glycosides, and alkaloids, enables broader exploration of chemical scaffolds that can interact with the NAD-binding pocket of IMPDH. This rational selection increases the likelihood of identifying novel binding modes and lead molecules for further antimicrobial development, rather than limiting the investigation to commonly studied polyphenols alone. Although numerous computational studies have screened phytochemicals against various bacterial proteins, none have systematically investigated amaroswerin, eriocitrin, peganine, afzelechin, amarogentin, and naringenin as potential inhibitors of the *guaB*-encoded IMPDH from *C. jejuni*, an essential yet comparatively underexplored antimicrobial target. Moreover, previous *in silico* studies have largely focused on identifying high-affinity ligands through molecular docking alone or have evaluated limited computational endpoints. In contrast, the present study integrates molecular docking, molecular dynamics (MD) simulations, density functional theory, absorption, distribution, metabolism, excretion, and toxicity (ADMET) prediction, and network pharmacology to comprehensively evaluate binding stability, electronic properties, pharmacokinetic behavior, and potential biological mechanisms. This multi-level computational framework provides mechanistic insights into ligand-IMPDH interactions and prioritizes promising lead compounds for future experimental validation. Therefore, this study aims to identify structurally diverse phytochemicals with the potential to inhibit *C. jejuni* IMPDH and to provide a comprehensive computational basis for developing novel anti-*Campylobacter* agents.

Materials and methods

Hardware and software

To perform the *in silico* research, a high-end Dell workstation (Precision 7865) equipped with a dual NVIDIA RTX A2000 graphics card was used. Molecular docking was performed using Maestro V14.5 (Schrödinger Suite). Cytoscape V3.10.4 was used to construct the network.

Construction of protein-protein interaction networks (PPINs)

PPINs help to reveal, how proteins cooperate in biological systems and identify key interaction points that serve as potential drug targets [19, 20]. In this study, a PPIN associated with the IMPDH gene (*guaB*) of *C. jejuni* was constructed to explore its functional interaction partners. Initially, draft PPINs were constructed using the STRING v12.0 [21] server with a high confidence score of 0.7 (i.e., interaction confidence > 70%). The final network was designed and validated using Cytoscape V3.10.4 [22]. Network features were calculated using the Network Analyzer module in Cytoscape.

Phytochemical selection

The phytochemicals included in this study were selected through a comprehensive literature survey of plant-derived compounds with reported antimicrobial and pharmacological activities. The selection was based on the following criteria: (i) evidence of antimicrobial, anti-inflammatory, or antioxidant properties reported in previous studies; (ii) structural diversity to represent different classes of natural products; (iii) availability of chemical structure information for computational analysis; and (iv) limited or no prior reports investigating their inhibitory activity against *C. jejuni* inosine monophosphate dehydrogenase.

Based on these criteria, six structurally diverse phytochemicals—afzelechin, amarogentin, amaroswerin, eriocitrin, naringenin, and peganine—were selected for further investigation (Figure 1 and Table 2). These compounds span multiple phytochemical classes, including flavonoids, secoiridoid glycosides, and alkaloids, and have been reported to exhibit promising biological activities. However, their potential as inhibitors of *C. jejuni* IMPDH has not been systematically explored. Therefore, these compounds were selected to evaluate their binding affinity, interaction stability, and drug-like properties against IMPDH using an integrated computational approach.

Table 2. Names, nature, source, and key structural features of unexplored phytochemicals used in the present study.

S. No.	Compound	Subclass/Group	Key structural features	Primary plant sources	Literature
1.	Afzelechin	Flavan-3-ol (Catechin)	5-dehydrocatechin; A, B, C rings with OH groups	<i>Bergenia ligulata</i>	[23]
2.	Amarogentin	Secoiridoid glycoside	Bicyclic structure with a glycosidic moiety	<i>Swertia chirayita</i>	[24]
3.	Amaroswerin	Secoiridoid	Structurally related to amarogentin	<i>Swertia chirayita</i>	[24]
4.	Eriocitrin	Flavanone glycoside	7-O-rutinoside of eriodictyol	<i>Citrus limon</i> (lemon peel)	[25]
5.	Naringenin	Flavanone (Flavonoid)	4',5,7-trihydroxyflavanone; aglycone form	<i>Citrus fruits</i> (grapefruit, bergamot), tomatoes	[26]
6.	Peganine	Quinoline alkaloid	Nitrogen-containing heterocyclic ring	<i>Peganum harmala</i> (Syrian rue)	[27]

Protein preparation

The crystal structure of the catalytic domain of inosine monophosphate dehydrogenase (IMPDH) from *C. jejuni* in complex with the inhibitor p200 was obtained from the Protein Data Bank (PDB) [28]. The deposition code for this protein is 5UQG. It contains 406 amino acid residues and is a key enzyme involved in guanine nucleotide biosynthesis, which is essential for bacterial growth, virulence, and pathogenicity [28]. The protein retrieved from the database was prepared using the Protein Preparation Wizard in Maestro V14.5 (Schrödinger, LLC). During preparation, the water molecules and heteroatoms were removed from the structure. Following optimization, minimization was continued until the average root mean square deviation (RMSD) of the non-hydrogen atoms reached 0.3 Å. This procedure alleviated steric clashes and achieved a stable protein conformation suitable for subsequent computational analyses [29].

Active site

The protein retrieved from the databank has a co-crystallized ligand (p200). Hence, the co-crystallized ligand site was selected as the active binding pocket for docking the phytochemicals [30]. Subsequently, a grid was constructed around the co-crystallized ligand to stabilize the protein binding site [31].

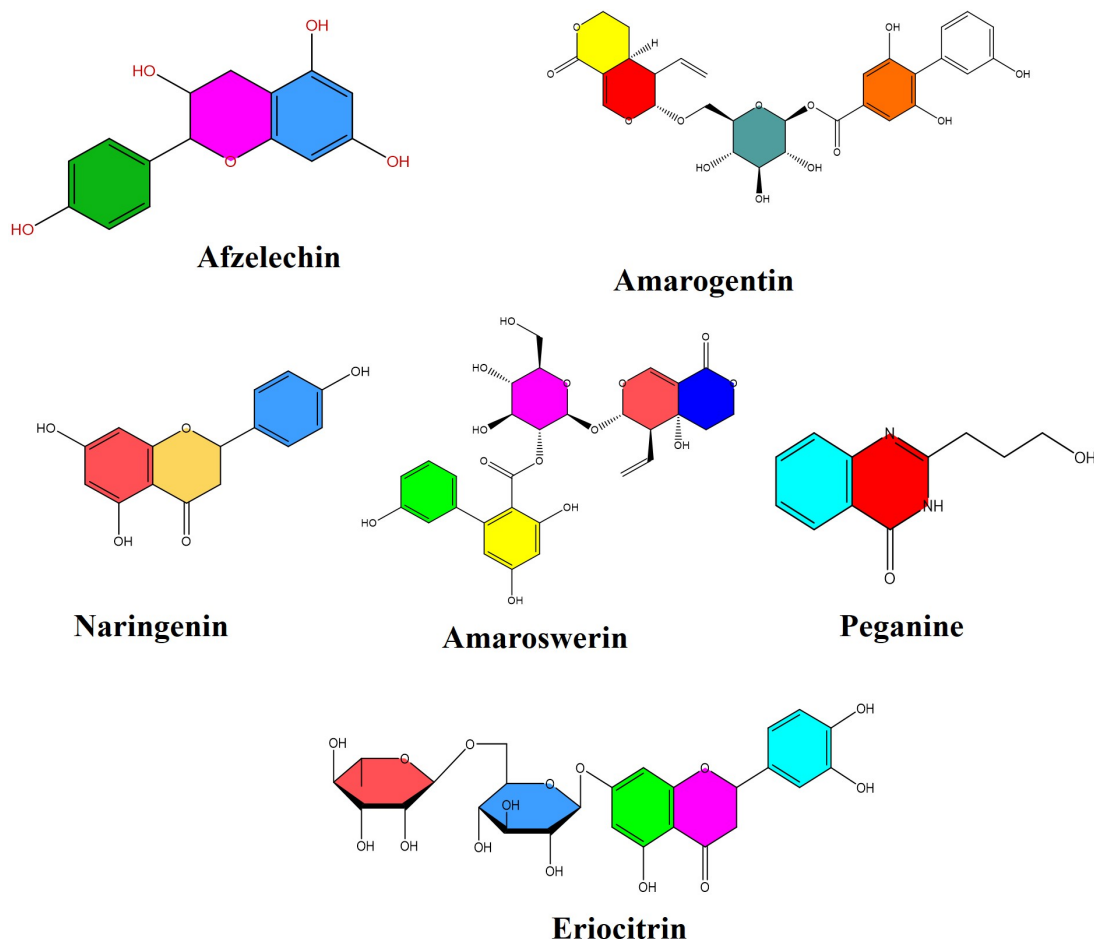


Figure 1. Chemical structures of phytochemicals used in the present research.

Ligand preparation

The physical complexity of the plant metabolites was reduced using the LigPrep module of Maestro V14.5. This module employs two phases: tautomerism and stereoisomerism. The ligand molecules were energy-minimized using the Optimized Potentials for Liquid Simulations (OPLS) 5 force field to ensure optimal shape and stability. To preserve the specified chirality of the ligand molecules, a maximum of 32 stereoisomers were generated for each ligand molecule. According to Lipinski's rule of five, ligands containing more than 500 Da were excluded from the ligand preparation. Furthermore, during the preparation process, the ionization states were consistently maintained at pH 7.0 $\pm$ 2.0 using Epik to ensure accurate molecular configuration [32].

Molecular docking

To assess the antimicrobial potential of the selected phytochemicals, the prepared ligands and grid-generated protein models were subjected to molecular docking using the ligand docking module in Maestro V14.5. The binding affinities of the ligands and protein molecules were measured using various parameters, including Glide docking score, Glide energy, Glide eModel, hydrogen bond contacts, pi-pi stacking, and pi-cation. The docked complex was explored using the Maestro interface to measure hydrogen-bond contacts and their distances. The interaction plot module was used for each phytochemical complex to screen the types of contacts that existed at the IMPDH binding site [29].

Docking protocol validation

To validate the molecular docking protocol, the co-crystallized ligand was extracted from the crystal structure of *C. jejuni* inosine monophosphate dehydrogenase, and all water molecules and other heteroatoms were removed. The native ligand was then redocked into the original binding pocket using the

same docking parameters as for all phytochemical ligands. In addition, the interactions of the redocked ligand with the active-site residues were compared with those of the crystal ligand to confirm conservation of the binding pattern.

MD simulations

To further assess the binding stability of the amaroswerin-IMPDH complex, MD simulations were performed using the Desmond module (Schrödinger). It was run for 200 ns. The molecular simulation system was built by placing the protein-ligand complex inside an orthorhombic periodic boundary box solvated with Transferable Intermolecular Potential with 3 Points (TIP3P) water molecules. This widely used explicit solvent model accurately captures hydrogen bonding and solvent dynamics in biomolecular simulations. A minimum buffer distance of 10 Å was maintained between the protein surface and the box boundary to prevent artificial interactions across periodic images. The system was neutralized by adding suitable counterions and supplemented with 0.15 M NaCl to mimic the physiological ionic strength. The OPLS_2005 force field was applied to model the molecular interactions of both protein and ligand atoms, offering reliable parameters for biomolecular simulations and it has been extensively validated in protein-ligand interaction studies. Before the production run, the system was energy-minimized using the steepest-descent algorithm to eliminate unfavorable steric contacts and relax the structures. Equilibration and production simulations were performed in the NPT ensemble [constant number of particles, pressure, and temperature (ensemble)]. The Nosé-Hoover chain thermostat maintained the system temperature at 300 K, and the Martyna-Tobias-Klein barostat maintained a pressure of 1 atm, ensuring accurate thermodynamic sampling [33]. Covalent bonds involving hydrogen atoms were constrained using the SHAKE algorithm, which enabled a 2-fs integration time step to improve computational efficiency while preserving stability.

ADMET properties

The pharmacokinetic profiles of the hit phytochemicals were evaluated using two online databases, pkCSM and ProTox-v3.0. The SMILES representations of the phytochemicals were used to evaluate their potential for ADMET in the body using the pkCSM server. Furthermore, organ toxicity, including hepatotoxicity, neurotoxicity, nephrotoxicity, respiratory toxicity, and cardiotoxicity, as well as toxicity endpoints such as carcinogenicity, immunotoxicity, mutagenicity, cytotoxicity, blood-brain barrier (BBB) permeability, ecotoxicity, and clinical toxicity, were assessed using the ProTox-v3.0 server.

Results

Analysis of PPINs

PPINs revealed that the target receptor *guaB* interacts with several key proteins, including *purA*, *purF*, *purL*, *purQ*, *guaA*, *pheT*, *proS*, *ribB*, *rpoB*, *rpoC*, and *htpG*. The connectivity among these proteins highlights the central role of *guaB* as a hub in metabolic and regulatory functions in purine biosynthetic metabolism (Figure 2a).

The predicted network features and functional descriptions of the proteins are presented in Tables 3 and 4. Gene Ontology (GO) enrichment analysis was performed for both biological processes and metabolic pathways using the constructed PPINs.

Table 3. Predicted network features of the target receptor (*guaB*) and other interacting genes.

Network features	
# of nodes	21
# of edges	98
Average node degree	9.33
Average local clustering coefficient	0.698
Expected number of edges	60
PPI enrichment <i>p</i> -value	3.03E-06

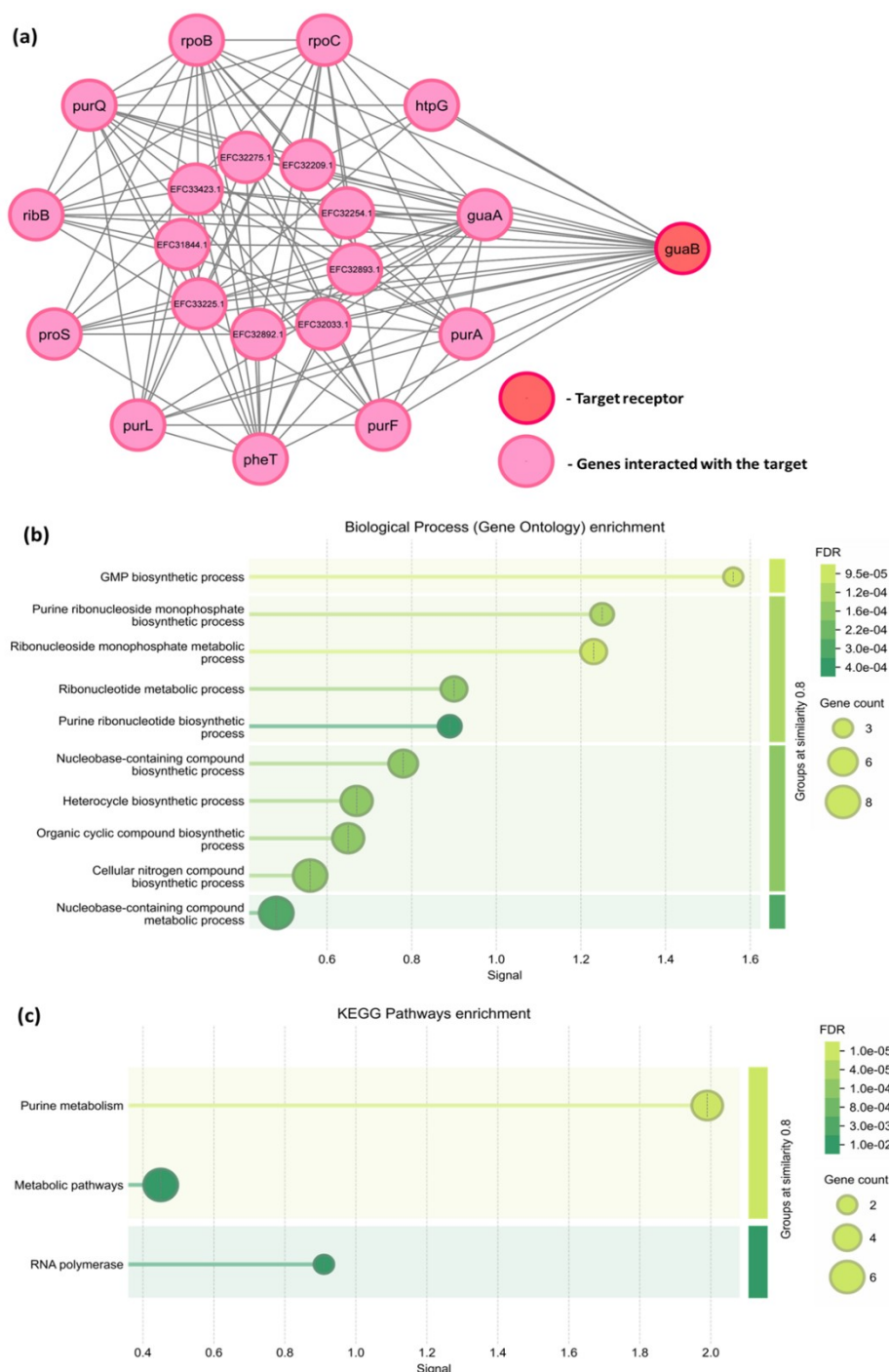


Figure 2. Protein–protein interaction network and functional pathway prediction analysis of *guaB* in *Campylobacter jejuni*. (a) Protein-protein interaction network (PPIN) of *guaB* gene in *C. jejuni*; (b) predicted biological process based on Gene Ontology (GO); and (c) predicted KEGG pathway enrichment analysis.

Table 4. Functional description of the genes that interact with the target receptor IMPDH (*guaB*).

Gene symbol	Functional description
<i>guaB</i>	Inosine-5'-monophosphate dehydrogenase
<i>guaA</i>	Bifunctional GMP synthase/glutamine amidotransferase protein
<i>purA</i>	Adenylosuccinate synthetase
<i>purF</i>	Amidophosphoribosyltransferase
<i>purL</i>	Phosphoribosylformylglycinamide synthase
<i>purQ</i>	Phosphoribosylformylglycinamide synthetase I
<i>rpoC</i>	DNA-directed RNA polymerase, beta subunit

Table 4. Functional description of the genes that interact with the target receptor IMPDH (*guaB*). (continued)

Gene symbol	Functional description
<i>rpoB</i>	DNA-directed RNA polymerase beta chain
<i>pheT</i>	Phenylalanyl-tRNA synthetase, beta subunit
<i>htpG</i>	Hsp90 family of heat shock proteins
<i>ribB</i>	3,4-Dihydroxy-2-butanone 4-phosphate synthase
<i>proS</i>	Prolyl-tRNA synthetase
EFC31844.1	Putative flagellar motor switch protein
EFC32033.1	Acid phosphatase SurE
EFC32254.1	Nucleotide phosphoribosyltransferase
EFC32893.1	valyl-tRNA synthetase
EFC32892.1	valyl-tRNA synthetase
EFC32275.1	Adenylosuccinate lyase
EFC33423.1	Adenylosuccinate lyase
EFC33225.1	ATP-dependent chaperone protein ClpB
EFC32209.1	Transformation system protein

Proteins were predominantly involved in purine-containing compound biosynthetic processes, nucleoside monophosphate biosynthesis, and ribonucleotide biosynthesis, as shown in [Figure 2b](#). The enrichment based on Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway showed that most proteins are involved in ligase activity ($1.4\text{e-}04$) and magnesium ion binding ($6.9\text{e-}04$), as depicted in [Figure S1](#). GO enrichment analysis of cellular compartments revealed that the network proteins were primarily associated with the cytoplasm (15 proteins) and intracellular compartments (17 proteins), as shown in [Figure S2](#). The KEGG pathway enrichment analysis showed that the network proteins were primarily involved in purine metabolism, followed by riboflavin metabolism and RNA polymerase-related pathways, as depicted in [Figure 2c](#). These results suggest that *guaB* and its partners are crucial for maintaining nucleotide synthesis, transcriptional activity, and cofactor metabolism ([Tables 3 and 4](#)).

Amaroswerin

Amaroswerin had a docking score of -8.612 kcal/mol ([Table 5](#)), with six interactions comprising five hydrogen bond contacts and one pi-cation contact ([Table 6](#)). In this case, ASN 273 (1.69 Å), GLU 411 (1.61 Å), LEU 408 (2.17 Å), MET 384 (2.20 Å), and LYS 71 (2.41 Å) were the key residues that formed hydrogen-bond contacts with amaroswerin. Notably, LYS 71 had two contacts: a π -cation interaction and a hydrogen bond. The two-dimensional interaction plot shows that, except for LEU 408, all hydrogen-bond contacts were established in the hydroxyl region of amaroswerin ([Figure 3a and 3b](#)).

Table 5. Docking scores and other energy parameters of selected phytochemicals for *C. jejuni* targeting inosine-5'-monophosphate dehydrogenase (IMPDH).

S. No.	Phytoconstituents	Glide docking score (kcal/mol)	Glide energy (kcal/mol)	Glide eModel (kcal/mol)
1.	Amaroswerin	-8.612	-65.739	-95.867
2.	Eriocitrin	-8.488	-66.644	-91.809
3.	Peganine	-7.999	-32.527	-50.657
4.	Afzelechin	-7.739	-33.513	-49.204
5.	Amarogentin	-7.635	-39.897	-57.791
6.	Naringenin	-7.619	-61.57	-83.247

Table 6. Active site residues of the inosine-5'-monophosphate dehydrogenase enzyme that established contacts with the phytochemicals.

S. No.	Phytoconstituents	Amino acid residue interactions and their distance	Types of bond formation	
			H-bond back chain	H-bond side chain
1.	Amaroswerin	GLU 411 (1.61), ASN 273 (1.69), LEU 408 (2.17), MET 384 (2.20), LYS 71 (2.41)	ASN 273, MET384, GLU411	LEU408, LYS71
2.	Eriocitrin	VAL 409 (1.82), GLY 385 (2.03), LEU 408 (2.04), GLU 411 (2.19), GLY 296 (2.25)	GLY 296, LEU 408	VAL 409, GLY 385, GLU 411
3.	Peganine	GLU 411, GLY 296, THR 303	GLY 296	GLU 411, THR 303
4.	Afzelechin	GLU 411 (1.61), ALA 47 (1.93), ASN 273 (2.69)	-	GLU 411, ALA 47, ASN 273
5.	Amarogentin	GLU 411 (1.85), MET 384 (1.94), LEU 408 (2.01), LYS 71 (2.39), GLY 294 (2.77)	LEU 408, LYS 71	-
6.	Naringenin	GLY 411, ALA 47, LYS 71, GLY 294	-	GLY411, ALA47, LYS71, GLY294

Eriocitrin

Eriocitrin ranked second among metabolites for inhibiting IMPDH. This complex had six hydrogen bond contacts and a docking score of -8.488 kcal/mol (Table 5). In the binding site, GLY 296, GLU 411, VAL 409, LEU 408, and GLY 385 were key residues that established hydrogen bond contacts with eriocitrin at distances of 2.25, 2.19, 1.94, 1.82, and 2.03 Å, respectively (Figure 3c and 3d). Notably, the hydroxyl regions of eriocitrin were identified as key sites that formed hydrogen-bond contacts with residues in the target-binding site (Table 6).

Afzelechin

Afzelechin had a docking score of -7.739 kcal/mol with three interactions with the target protein (Table 5). In this case, GLU 411, ALA 47, and ASN 273 were the key residues that interacted with afzelechin. The distances of the hydrogen bond contacts for GLU 411, ALA 47, and ASN 273 were 1.61, 1.93, and 2.69 Å, respectively, indicating that afzelechin has a good binding affinity for IMPDH (Table 6). The interaction plot indicated that the hydroxyl moieties of afzelechin were the primary sites of interaction with IMPDH (Figure 3e and 3f).

Amarogentin

Amarogentin had a docking score of -7.635 kcal/mol (Table 5). It has a similar interaction profile to amaroswerin and eriocitrin, indicating strong binding affinity for IMPDH. The complex comprises five hydrogen-bond contacts and a π -cation contact. The active-site residues GLY 294, GLU 411, LEU 408, MET 384, and LYS 71 formed hydrogen-bond contacts at distances of 2.77 Å, 1.85 Å, 2.01 Å, 1.94 Å, and 2.39 Å, respectively (Figure 3g and 3h). Among these contacts, except for LEU 408, all residues formed hydrogen bonds with the hydroxyl moieties of amarogentin (Table 6).

Peganine and naringenin

Peganine had a docking score of -7.999 kcal/mol (Table 5). This complex has three interactions: GLU 411, THR 303, and GLY 296 were the active-site residues that formed hydrogen-bonding contacts with peganine (Figure 4a). In this case, GLU 411 established a hydrogen bond with the ammonium moiety of peganine, whereas the other two residues, THR 303 and GLY 296, established a hydrogen bond with the hydroxyl moiety of peganine. Naringenin had a docking score of -7.619 kcal/mol (Table 5). Despite this molecule having a lower docking score than peganine, it formed better interactions with IMPDH, including four hydrogen-bond contacts and a π -cation contact (Table 6). The active site residues GLU 411, GLY 294, ALA 47, and LYS 71 interacted with naringenin (Figure 4b).

Docking validation: comparison of crystal poses and redocked pose

To assess the accuracy of the docking protocol, the co-crystallized ligand was redocked into its crystallized site (Figure 5). The redocked ligand occupied the native binding pocket and formed notable interactions

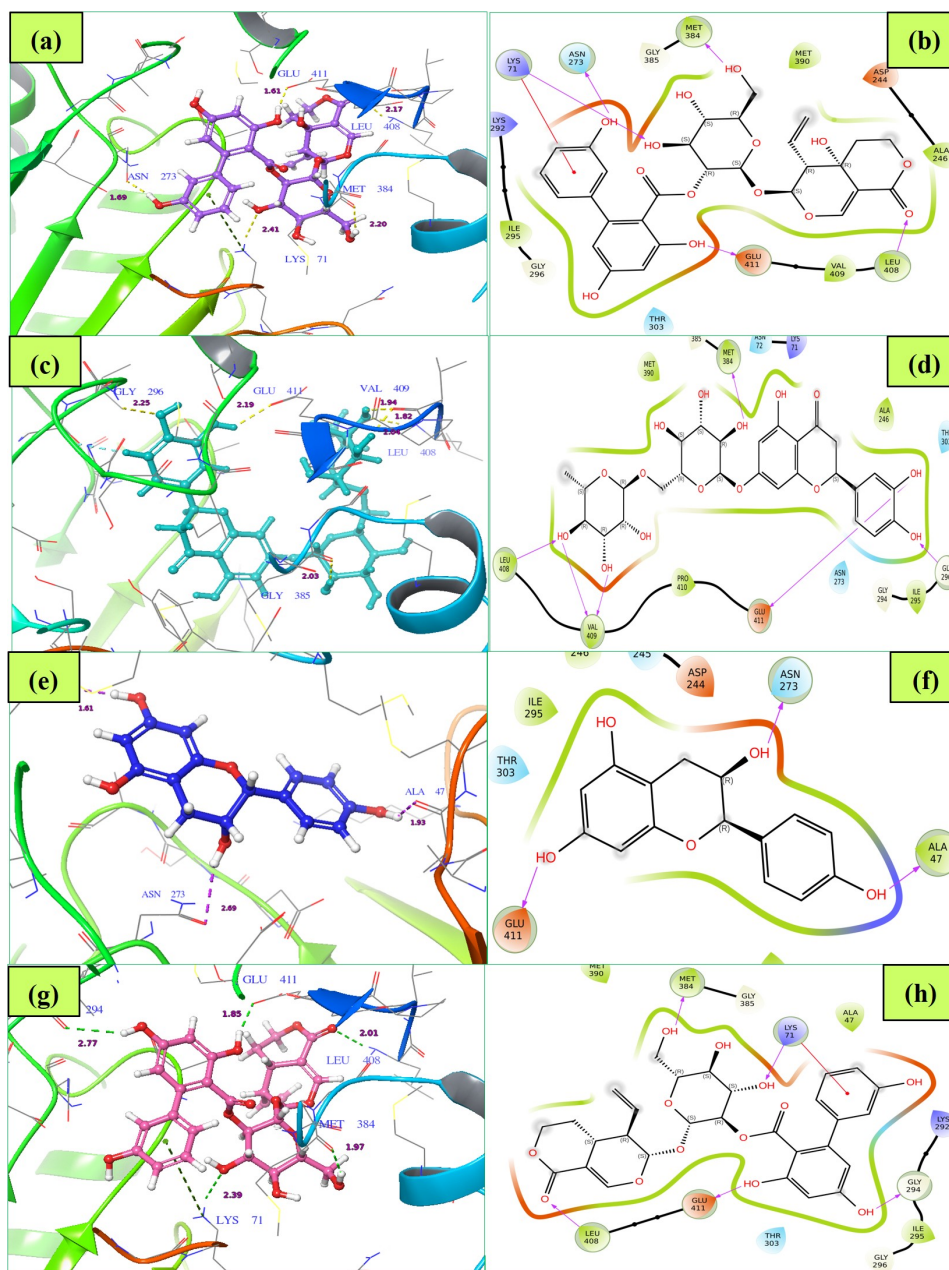


Figure 3. Binding affinities of phytochemicals with IMPDH. (a and b) Amaroswerin; (c and d) eriocitrin; (e and f) afzelechin; and (g and h) amarogentin.

with the active-site residues of IMPDH. These findings validate the docking procedure and increase confidence in the predicted binding poses of the selected phytochemicals (Figure 6).

Binding stability of amaroswerin at the predicted active site of IMPDH

To assess the dynamic behavior and binding stability of the amaroswerin–IMPDH complex, MD simulations were performed for 200 ns. During the initial phase of the trajectory (0–85 ns), the C α RMSD remained below approximately 2.8 Å. The RMSD gradually increased to approximately 4.5 Å at 180 ns, then stabilized toward the end of the 200 ns simulation. This pattern suggests that the complex underwent conformational adjustment during the simulation while retaining its overall structural association. The most fluctuating residual areas in this protein were ASN 1, ALA 0, MET 1, LYS 392, LEU 408, SER 307, LYS 2, GLY 308, GLN 391, VAL 309, and VAL 409, which fluctuated with RMSF ranges of 9.259, 7.802, 6.321, 5.663, 5.414, 5.145, 4.999, 4.862, 4.753, 4.681, and 4.642 Å, respectively. ASP 334 had the highest interaction fraction of 2.03 Å, involving a water-bridge-assisted hydrogen bond. Notably, ASP 334 (94% and 80%), GLU 411 (72%), ALA 47 (54%), ASP 49 (53%), ASP 244 (52% and 32%), LYS 21 (35%), and LYS 292 (32%) were the principal

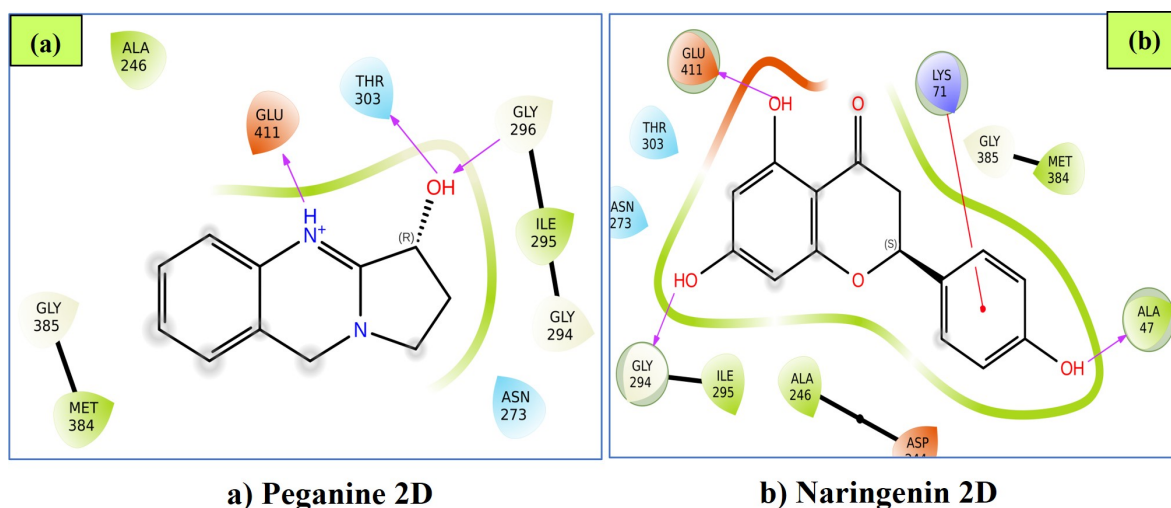


Figure 4. 2D template shows the interactions of (a) peganine and (b) naringenin with IMPDH.

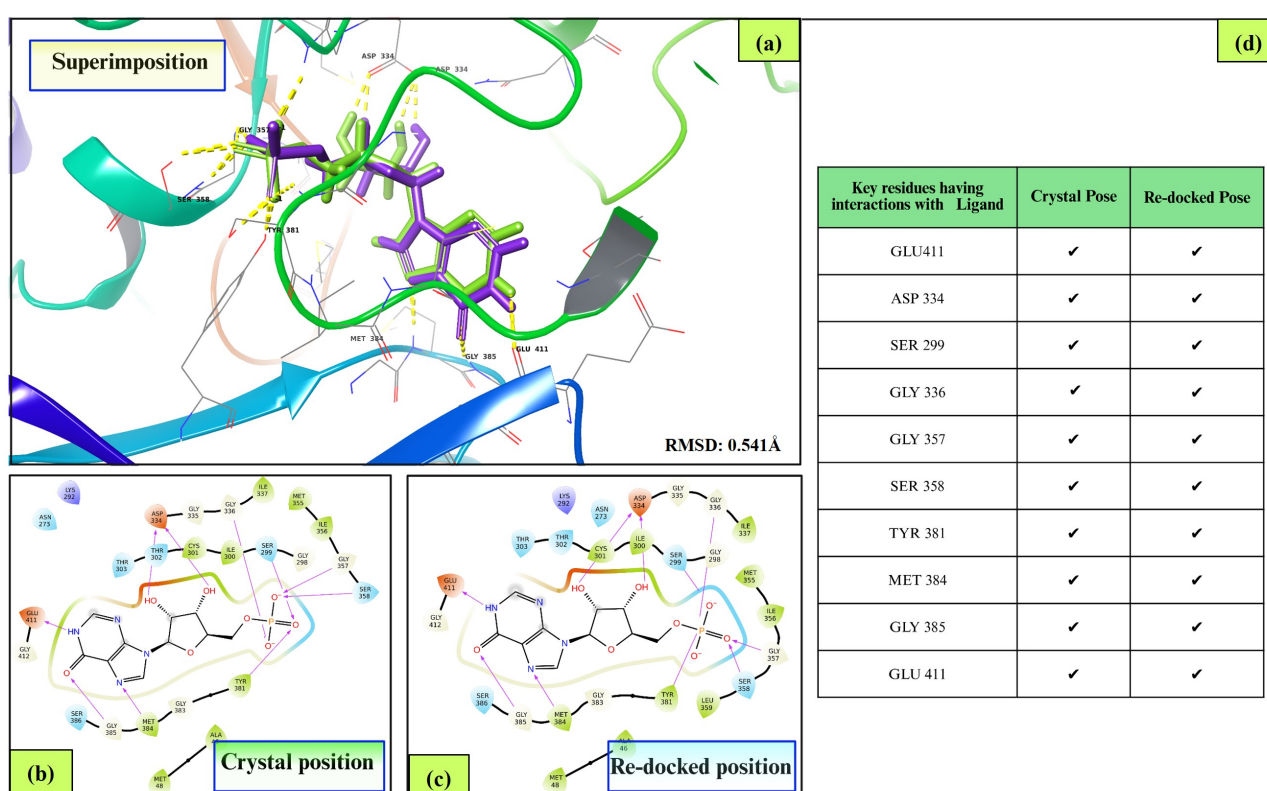


Figure 5. Docking validation of inosinic acid with IMPDH. (a) Superimposition of crystal pose (violet) and redocked pose (green) (ligand pose RMSD: 0.541 Å); (b) 2D interaction plot of crystal pose; (c) 2D interaction plot of redocked pose; and (d) comparison of interactions.

residues that facilitated the stability of the amaroswerin-IMPDH complex through robust hydrogen bond interactions (Figure 7). The binding stability was maintained by an average of 12 contacts over 200 ns.

Binding stability of amaroswerin at the co-crystal site of IMPDH

A 150 ns MD simulation was performed to evaluate the dynamic stability of the amaroswerin-IMPDH complex beyond molecular docking. The stability of the complex structure was evaluated by RMSD, root mean square fluctuation (RMSF), residue interaction, and protein-ligand contact profiling (Figure 8). RMSD of the protein backbone initially rose during the first 20–30 ns of the simulation to allow for structural relaxation from the docked state, and then tended to stabilize around 3.0–4.2 Å over the rest of the simulation (Figure 8a). Both RMSD and RMSF values of the ligand were relatively stable after initial equilibration, ranging from 4.5–5.2 Å and 11.0–11.1 Å, respectively, suggesting that amaroswerin was

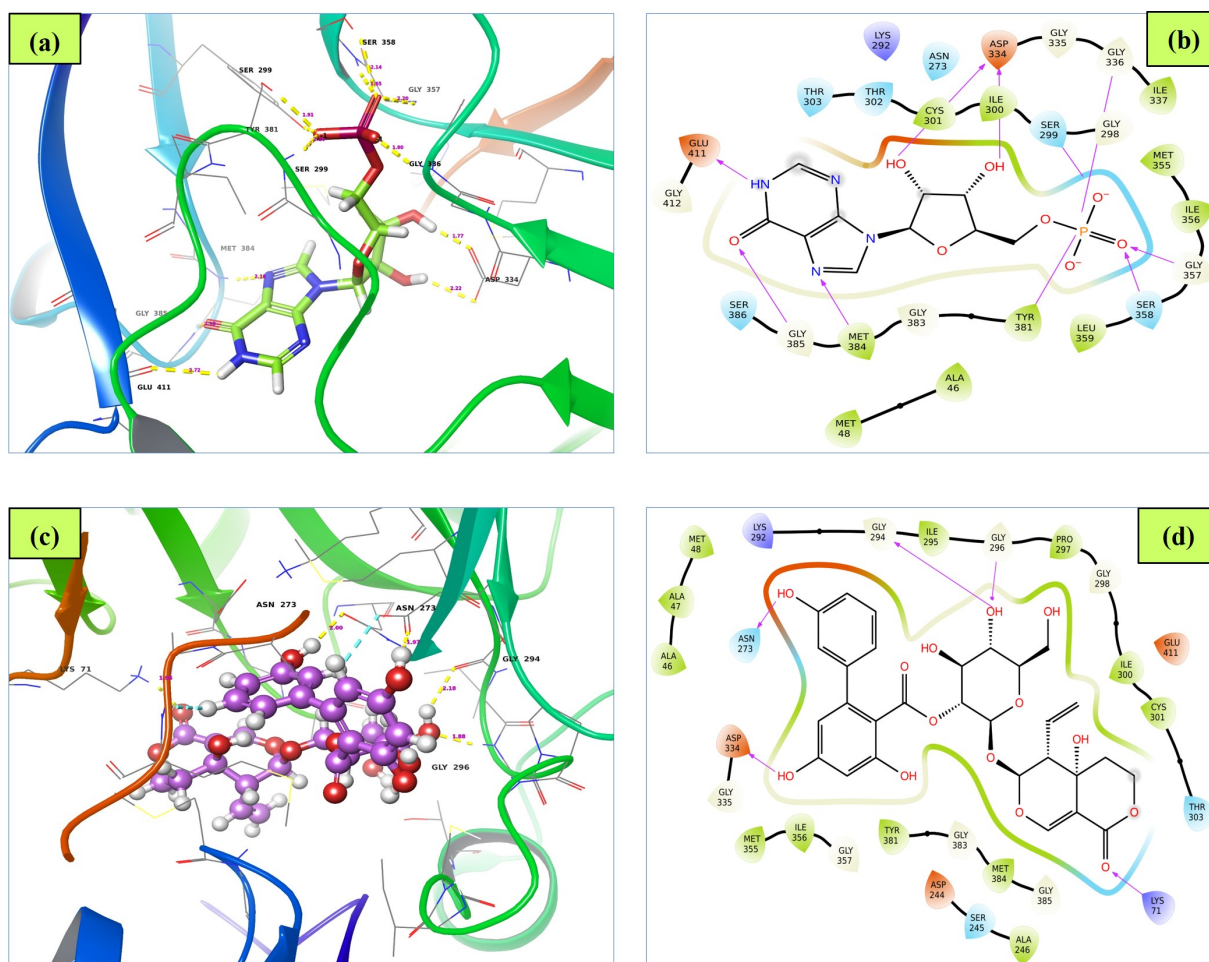


Figure 6. Binding orientation of the re-docked reference ligand and amaroswerin at the co-crystallized ligand-binding site of the target protein. (a and b) Three-dimensional and two-dimensional interaction plots of inosinic acid; and (c and d) three-dimensional and two-dimensional interaction plots of amaroswerin.

bound in the same orientation in the active site, with only minor conformational changes. No large deviations were observed during the production phase, indicating that the protein-ligand complex equilibrated and remained structurally stable throughout the simulation. The fluctuations in amino acid residues, as measured by RMSF, showed that most residues were within 1.5 Å of their mean positions, indicating the overall rigidity of the protein structure (Figure 8b). The elevation of fluctuations was mainly found in the terminal regions and a few loops at residues 90, 180–190, 275–285, and the C-terminal region, which are typical of flexible solvent-exposed loops. Importantly, the residues that form the ligand-binding pocket showed relatively small fluctuations, which would help to preserve the integrity of the active site when the ligands are bound.

Amaroswerin formed stable interactions with several active-site residues during the simulation (Figure 8c), as confirmed by the protein-ligand interaction fraction analysis. Of these, GLU 411 had the highest interaction occupancy, being in contact for almost the entire simulation. Other residues, ASP 334, MET 384, THR 303, LYS 292, and ASP 244, also contributed to ligand stabilization through hydrogen bonding, hydrophobic interactions, and water-mediated interactions. The interaction profile indicates that multiple residues contribute to stabilizing the ligand within the catalytic pocket. The two-dimensional interaction map also showed that amaroswerin has a large hydrogen-bonding network with important catalytic residues (Figure 8d). It was determined that the residues GLU 411, MET 384, and LYS 292 form direct hydrogen bonds, and additional water-supported hydrogen bonds were detected between ASP 334 and ASP 244. Hydrophobic contacts with residues are further strengthened ligand binding. These complementary interactions helped maintain the ligand in an optimal conformation throughout the simulation. The protein-ligand contact timeline showed that Glu411 had the highest number of contacts

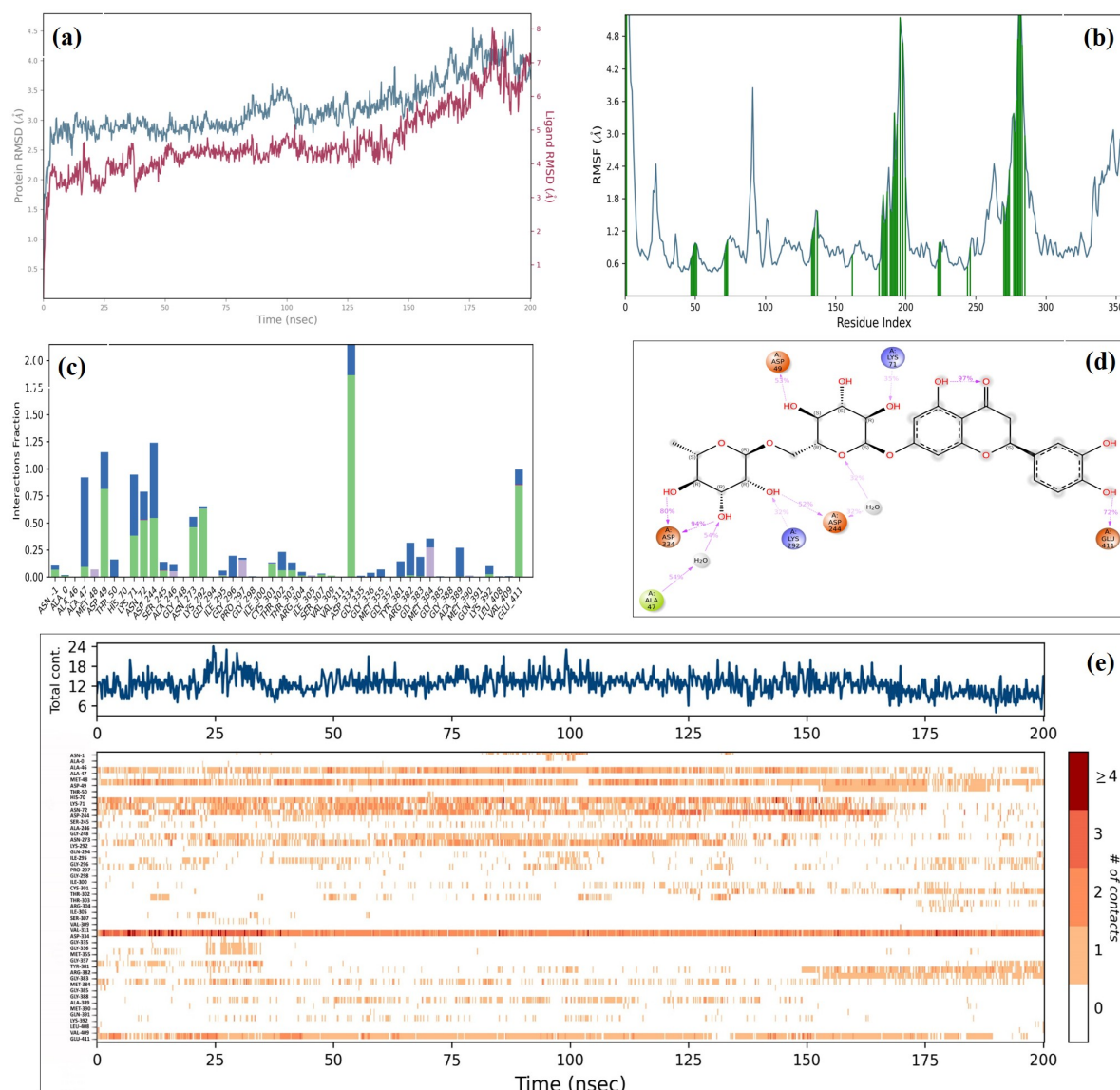


Figure 7. Simulation plots represent the binding stability of amaroswerin with the *Campylobacter jejuni* IMPDH protein. (a) RMSD; (b) RMSF; (c) interaction fraction plot; (d) contact summary; and (e) amaroswerin contacts plot with the residues of the *Campylobacter jejuni* IMPDH protein. RMSF: root mean square fluctuation.

that persisted throughout most of the simulation (Figure 8e), along with Met384, Thr303, Lys292, and Asp244. Transient interactions with a few residues were observed sporadically, but the overall stability of these core interactions suggests that amaroswerin remained tightly bound to the IMPDH active site.

Co-crystal

To ensure the stability of the native ligand in the active site of *C. jejuni* inosine-5'-monophosphate dehydrogenase (IMPDH), a 100 ns MD simulation was performed. The structural stability and interaction profile of the co-crystal complex were assessed using RMSD, RMSF, interaction fraction, and protein-ligand contact analysis (Figure 9). The initial 10–20 ns showed a large increase in the protein backbone RMSD, corresponding to structural equilibration, and the RMSD then remained relatively constant between 2.5 and 3.3 Å throughout the rest of the simulation (Figure 9a). The ligand RMSD fluctuated moderately during the equilibration phase and converged to a steady value of 2.0–3.0 Å, suggesting a stable orientation of the co-crystal ligand within the catalytic pocket throughout the simulation. There is no significant conformational drift, indicating that the protein-ligand complex has equilibrated and remained stable during the 100 ns simulation.

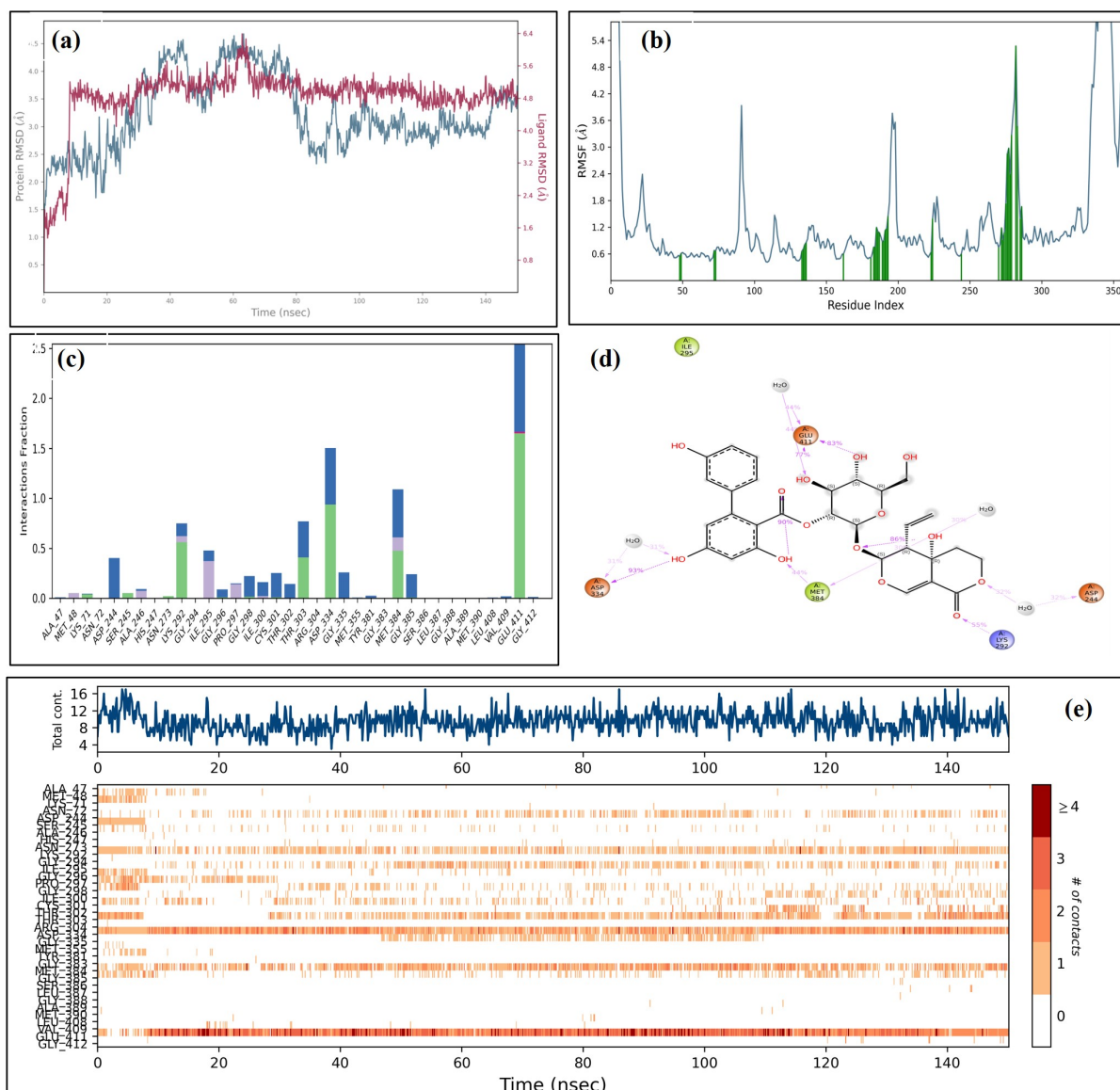


Figure 8. Simulation plots represent the binding stability of amaroswerin with the *Campylobacter jejuni* IMPDH protein. (a) RMSD; (b) RMSF; (c) interaction fraction plot; (d) contact summary; and (e) amaroswerin contacts plot with the residues of the *Campylobacter jejuni* IMPDH protein. RMSF: root mean square fluctuation.

In the RMSF profile, most protein residues showed fluctuations less than 1.5 Å, suggesting limited flexibility in the protein structure (Figure 9b). The general trend of higher fluctuations was observed in the flexible loops around residues 85–90, 190–200, and 275–285, as well as in the C-terminal residues, which are not close to the catalytic core. Residues lining the ligand-binding pocket, on the other hand, showed relatively small variations, indicating that ligand binding helped preserve the structural stability of the active site. To investigate the dynamics of the interaction between the co-crystal ligand and the active site residues, data from the interaction fraction analysis were used to simulate the interaction (Figure 9c). High interaction occupancies are observed for PRO 297, SER 299, ASP 334, GLY 336, and ILE 337, with the latter showing the highest occupancy. The interactions were mainly hydrogen bonding, water-mediated bridging, and hydrophobic interactions, collectively supporting the ligand within the binding cavity.

The two-dimensional representation of the interaction map also revealed numerous hydrogen bonds between the ligand and surrounding residues (Figure 9d). Stable hydrogen bonds were established with ASP 334, SER 299, SER 358, GLY 357, MET 384, and GLY 385 residues, and hydrophobic interactions with ILE 300, ILE 337, TYR 381, and PRO 297 also contributed to ligand binding. Several other hydrogen-bond interactions were also mediated by water molecules, underscoring the importance of solvent bridges in maintaining the protein-ligand complex. The protein-ligand contact timeline (Figure 9e) shows that most

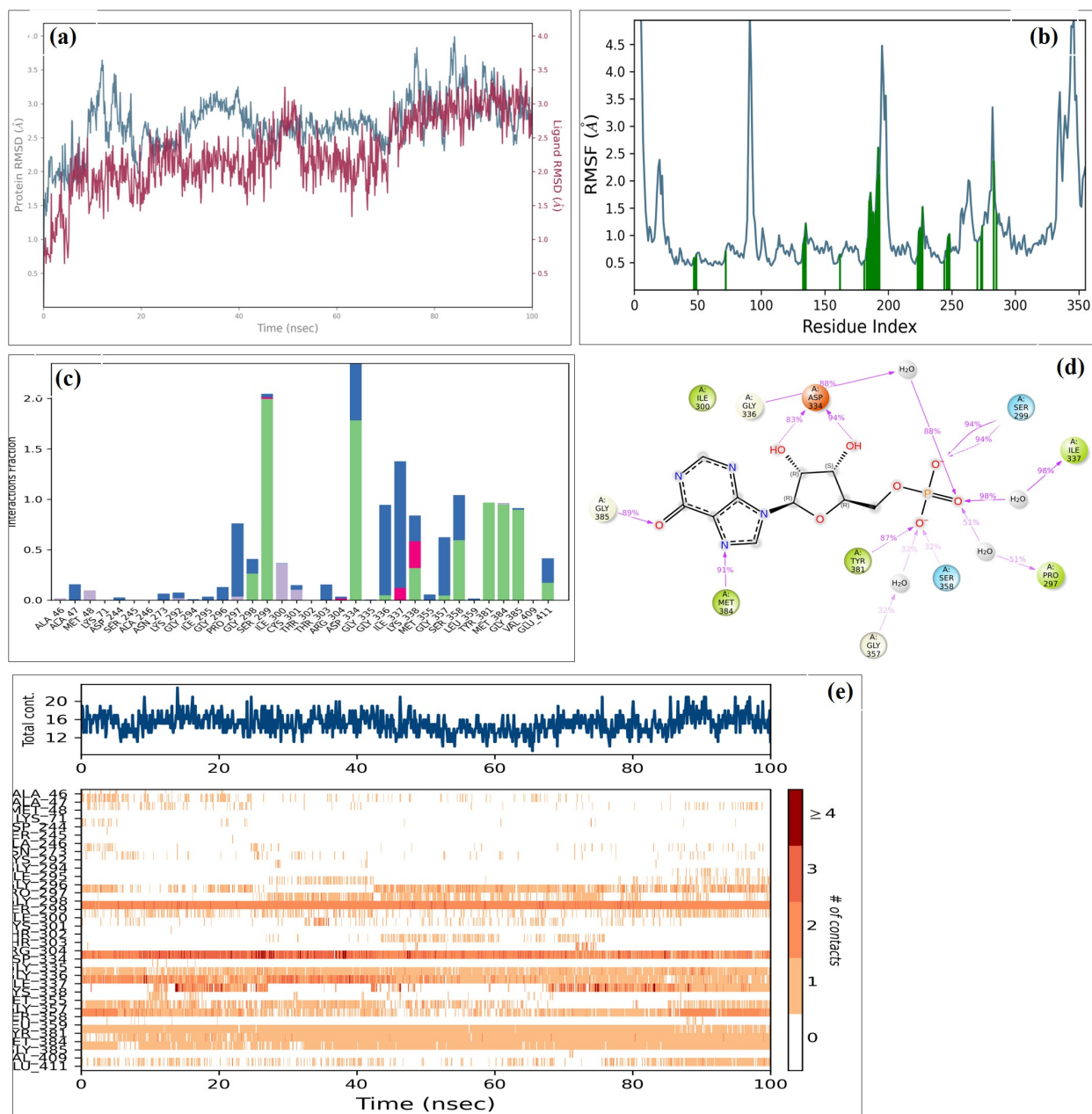


Figure 9. Simulation plots represent the binding stability of the co-crystal with the *Campylobacter jejuni* IMPDH protein. (a) RMSD; (b) RMSF; (c) interaction fraction plot; (d) contact summary; and (e) co-crystal contacts plot with the residues of the *Campylobacter jejuni* IMPDH protein. RMSF: root mean square fluctuation.

contacts were maintained for a large fraction of the 100 ns simulation. Contacts were maintained throughout the trajectory, and other residues were in intermittent contact, contributing to the overall stability of the complex. The same interaction pattern throughout the simulation suggests that the co-crystal ligand was well bound in the active site.

Pharmacokinetics analysis

The ADMET profiles of the hit metabolites showed that afzelechin had the highest intestinal absorption (91.48%), followed by amarogentin (50.44%) and amaroswerin (49.294%). In terms of Caco-2 permeability, amaroswerin (−0.592) and eriocitrin (−1.101) had the lowest values, indicating low permeability potential. Interestingly, all the hit metabolites were found to act as substrates for P-glycoprotein, indicating possible transporter-mediated efflux. The probability distribution rates of these phytochemicals ranged from −1.406 to 1.529, with eriocitrin showing the highest distribution potential. In terms of BBB permeability, all phytochemicals showed a negative rate, indicating poor penetration across the BBB. Likewise, the central nervous system (CNS) permeability values for these chemicals are low,

indicating a lower risk of neurotoxicity. The metabolic potential of these chemicals revealed that none were substrates or inhibitors of major cytochrome P450 (CYP450) enzymes, including CYP1A2, CYP2C19, CYP2C9, CYP2D6, and CYP3A4. The absence of significant CYP inhibition indicates that the phytochemicals are less likely to cause adverse drug-drug interactions and are metabolically compatible. In terms of excretion, amaroswerin and amarogentin have relatively high clearance rates. Furthermore, none of these metabolites were found to be substrates for renal OCT2 (organic cation transporter 2), indicating lower renal transport-associated toxicity. The phytochemicals were found to be non-hepatotoxic and non-sensitizing to the skin, whereas the oral chronic rat toxicity values ranged from 2.215 to 3.33, indicating an acceptable toxicity range. Additionally, the toxicity values for *Tetrahymena pyriformis* and the fathead minnow indicate that these substances are relatively safe for the environment and living organisms. Together, the ADMET profiles indicated that the molecules have favorable pharmacokinetic and acceptable safety profiles, supporting their potential as promising candidates for *C. jejuni* targeting of IMPDH (Table 7).

Table 7. Pharmacokinetic profiling of hit phytochemicals.

Absorption	Amaroswerin	Eriocitrin	Afzelechin	Amarogentin
Water solubility	−3.868	−2.941	−3.254	−3.11
Caco-2 permeability	−0.592	−1.101	1.077	0.271
Intestinal absorption (human)	49.294	26.972	91.482	50.444
Skin permeability	−2.754	2.735	−2.735	−2.735
P-glycoprotein substrate	Yes	Yes	Yes	Yes
P-glycoprotein I inhibitor	Yes	No	No	No
P-glycoprotein II inhibitor	Yes	No	No	No
Distribution				
VDss (human)	−1.406	1.529	0.562	1.115
Fraction unbound (human)	0.313	0.148	0.194	0.094
BBB permeability	−2.048	−1.831	−0.818	−1.811
CNS permeability	−4.382	−4.859	−2.473	−4.448
Metabolism				
CYP2D6 substrate	No	No	No	No
CYP3A4 substrate	Yes	No	No	Yes
CYP1A2 inhibitor	No	No	No	No
CYP2C19 inhibitor	No	No	No	No
CYP2C9 inhibitor	No	No	No	No
CYP2D6 inhibitor	No	No	No	No
CYP3A4 inhibitor	No	No	No	No
Excretion				
Total clearance	0.465	0.115	0.255	0.419
Renal OCT2 substrate	No	No	No	No
Toxicity				
Oral rat chronic toxicity (LOAEL)	2.427	2.514	2.215	3.33
Hepatotoxicity	No	No	No	No
Skin sensitization	No	No	No	No
<i>T. pyriformis</i> toxicity	0.285	0.285	0.519	0.285
Minnow toxicity	2.391	7.621	2.75	3.384

LOAEL: lowest-observed-adverse-effect level.

An online toxicity prediction platform was used to evaluate the potential organ toxicity and toxicity endpoints of the selected phytochemicals. Amaroswerin, eriocitrin, afzelechin, and amarogentin showed higher predicted probabilities of inactivity for hepatotoxicity and neurotoxicity, suggesting a lower computational likelihood of liver and neuronal toxicity. However, eriocitrin, afzelechin, and amarogentin exhibited relatively higher predicted probabilities for nephrotoxicity and respiratory toxicity (0.77, 0.75, and 0.80, respectively), indicating that these potential toxicities should be considered in future

experimental investigations. Similarly, the higher predicted inactive probabilities for carcinogenicity (0.75, 0.91, 0.70, and 0.77, respectively), together with the low predicted probabilities for mutagenicity, cytotoxicity, blood–brain barrier penetration, ecotoxicity, and clinical toxicity, suggest a favorable preliminary computational safety profile (Table 8). Nevertheless, these predictions are generated using *in silico* models and should not be interpreted as definitive evidence of safety. Therefore, comprehensive *in vitro* and *in vivo* toxicological studies are required to validate these computational findings before considering further drug development.

Table 8. Predicted organ toxicity and toxicity endpoints of the phytochemicals identified as possible inhibitors of inosine-5'-monophosphate dehydrogenase.

Classification	Target	Name of the phytochemicals			
		Amaroswerin	Eriocitrin	Afzelechin	Amarogentin
Organ toxicity	Hepatotoxicity	Inactive: 0.78	Inactive: 0.80	Inactive: 0.71	Inactive: 0.75
	Neurotoxicity	Inactive: 0.80	Inactive: 0.89	Inactive: 0.90	Inactive: 0.80
	Nephrotoxicity	Active: 0.66	Active: 0.77	Active: 0.62	Active: 0.75
	Respiratory toxicity	Active: 0.61	Active: 0.63	Active: 0.80	Active: 0.56
	Cardiotoxicity	Active: 0.68	Inactive: 0.98	Inactive: 0.76	Active: 0.65
Toxicity endpoints	Carcinogenicity	Inactive: 0.75	Inactive: 0.91	Inactive: 0.70	Inactive: 0.77
	Immunotoxicity	Active: 0.98	Active: 0.99	Inactive: 0.98	Active: 0.98
	Mutagenicity	Inactive: 0.66	Inactive: 0.88	Inactive: 0.56	Inactive: 0.66
	Cytotoxicity	Inactive: 0.74	Inactive: 0.64	Inactive: 0.84	Inactive: 0.75
	BBB-barrier	Inactive: 0.52	Inactive: 0.75	Active: 0.58	Inactive: 0.52
	Ecotoxicity	Inactive: 0.67	Inactive: 0.60	Inactive: 0.56	Inactive: 0.68
	Clinical toxicity	Active: 0.66	Active: 0.52	Active: 0.51	Active: 0.65
	Nutritional toxicity	Active: 0.73	Active: 0.54	Active: 0.66	Active: 0.66
Total number of organ toxicity probability		3	2	2	3
Total number of toxicity endpoints probability		4	3	4	4

Notes: Green denotes inactive probability > 0.7; pink denotes inactive probability < 0.7; red denotes active probability > 0.7; yellow denotes active probability < 0.7.

Discussion

Currently, many diseases are caused by foodborne pathogens, with *C. jejuni* often responsible for illness [34]. More than 200 types of foodborne diseases have been identified [35]. The most severe cases typically occur in both older and younger individuals with weakened immune systems, as well as in healthy people who are exposed to a very high dose of an organism [34]. Traditionally, people have used plants to treat foodborne diseases caused by contaminated food, as well as other digestion-related issues [36]. In particular, in developing countries worldwide, approximately 70–80% of the population relies on traditional herbal remedies for their primary health care needs [37]. Therefore, this study aimed to identify prospective plant candidates for the management of *C. jejuni*, which is implicated in gastroenteritis, using various computational drug discovery approaches. Computer-aided drug discovery (CADD) methods contribute significantly to drug development by improving discovery efficiency at lower cost and time while reducing the practical complications of clinical trials. These methods have transformed from a complementary tool to a core component of drug discovery pipelines. In particular, computational tools have been integrated with traditional pharmacological methods to accelerate the identification of novel therapeutic agents for targeted diseases [38]. In this study, the phytochemicals were identified as active candidates for inhibiting *C. jejuni* by targeting the essential IMPDH-mediated guanine nucleotide biosynthetic pathway, which is responsible for bacterial survival, multiplication, and pathogenicity. The docking scores, ranging from –8.612 to –7.619 kcal/mol, indicate favorable interactions with the target protein, suggesting that these phytochemicals represent potential lead compounds for further development.

Amaroswerin is a secoiridoid glycoside predominantly isolated from *Swertia chirayita* (flowers: 0.741%) and *Swertia mussotii* [24]. It has previously been reported to exhibit exceptional anti-inflammatory potency, with nitric oxide (NO) inhibition (IC_{50} : 5.42 μ g/mL) in lipopolysaccharide (LPS)-stimulated RAW264.7 macrophages, suppressing inducible NO synthase (iNOS), tumor necrosis factor- α (TNF- α), interleukin-6 (IL-6), and IL-1 β expression at both the protein and mRNA levels via transcriptional downregulation [39]. In addition, it has been reported to possess various therapeutic properties, including gastroprotective [40] and anti-inflammatory activities [39].

Eriocitrin is a flavanone glycoside found in *Citrus limon* [25]. It has been reported to exert strong anti-inflammatory effects by targeting multiple inflammatory pathways. Previous research reported that mixtures of eriocitrin and resveratrol, at doses of 12.5 μ M and 50 mg/g, and 400 μ M and 10 mg/kg, effectively inhibited key proinflammatory mediators, including NO, IL-1 β , TNF- α , NF- κ B (nuclear factor kappa-light-chain-enhancer of activated B cells), and MAPK, in LPS-induced RAW264.7 cells. It has also reduced edema and inflammation in the subcutaneous tissues of mouse models [41]. In dextran sulfate sodium (DSS)-induced colitis in male mice, the administration of eriocitrin (30 mg/kg) effectively reduced MPO production and modulated MMP-9 and NF- κ B signaling. In this study, it suppressed pro-inflammatory mediators, including iNOS and cyclooxygenase-2 (COX-2), thereby protecting the colon from inflammation [42]. Furthermore, in a middle cerebral artery occlusion (MCAO)/reperfusion model in male Sprague-Dawley rats, eriocitrin (8, 16, and 32 mg/kg) significantly decreased pro-inflammatory cytokines TNF- α and IL-6, whereas it enhanced anti-inflammatory and antioxidant markers such as IL-10, Nrf2, HO-1, and NQO1 [43]. In Caco-2 cell in vitro models, a dose of 25 ng/mL reduced the release of IL-6, IL-8, and NO, indicating its potential to control intestinal inflammation [44]. Additionally, in OGD/R-induced HK-2 cells and AKI models, the various doses (10, 30, and 60 mg/kg) significantly upregulated DUSP14 and Nrf2 expression and inhibited NF- κ B phosphorylation. It also attenuated inflammation and oxidative stress in rats [45]. In LPS-induced periodontal disease models in BALB/c male mice, eriocitrin (25 and 50 mg/kg) significantly suppressed IL-1 β and TNF- α levels in gingival tissues and increased IL-10 production, thereby protecting oral tissues from inflammatory damage [46].

Afzelechin is a flavan-3-ol catechin derivative that is abundant in *Bergenian ligulata* [23]. Lee and Bae [23] reported that it has potent anti-inflammatory activity and protective effects against LPS-induced inflammatory injury by modulating NF- κ B, STAT-1, and Nrf2 signaling pathways. These findings suggest that AZC may be a promising candidate for the development of novel anti-inflammatory therapies, warranting further investigation. Cho et al. [47] reported that it may alleviate sepsis-induced acute lung injury by reducing inflammatory cytokines and modulating the TLR4/NF- κ B, PI3K/Akt, Hippo, and Rho signaling pathways. These findings suggest that AZC may be a promising therapeutic candidate for sepsis-induced lung injury and warrant further investigation.

Amarogentin is a secoiridoid glycoside mainly extracted from *Swertia chirayita* [24]. It is structurally similar to amaroswerin. Zhang et al. [48] reported that their investigations in the mast cell line LAD-2 and keratinocyte studies revealed anti-inflammatory and immunomodulatory properties. At micromolar concentrations ranging from ~10–100 μ M, it inhibited the de novo synthesis of TNF- α in mast cells without affecting their degranulation. Ultimately, TNF- α suppression reduces the levels of downstream inflammatory mediators in keratinocytes, including IL-8, MMP-1, IL-6, and nerve growth factor (NGF). Furthermore, it directly inhibited the release of IL-8 and MMP-1 in response to stimulation with histamine and TNF- α . Mechanistically, these effects are mediated by the activation of bitter taste receptors (TAS2Rs), particularly TAS2R1 and related subtypes, indicating a receptor-dependent pathway for controlling inflammation [48]. Administration of amarogentin at various doses in fibrosis-induced mice protected the liver from damage, decreased the levels of alanine aminotransferase, aspartate aminotransferase, malondialdehyde, and hydroxyproline, and increased the levels of albumin, cyclic guanosine monophosphate, glutathione peroxidase, and superoxide dismutase. Based on their findings, Zhang et al. [49] documented that it has hepatoprotective effects against carbon tetrachloride-induced liver fibrosis and suppresses the mitogen-activated protein kinase signaling pathway.

Peganine, also known as vasicine, is a quinazoline alkaloid found in *Peganum harmala* (Syrian rue) and *Justicia adhatoda* (Malabar nut) [27]. It acts as a potent bronchodilator by inhibiting PDE-IV and elevating cAMP levels, thereby promoting airway relaxation, a mechanism comparable to that of theophylline. Peganine has demonstrated promising antileishmanial activity against both promastigote and intracellular amastigote forms of *Leishmania donovani*, with its activity associated with apoptosis-like cell death and inhibition of DNA topoisomerase I [50]. Numerous studies have reported that it has anti-allergic and anti-Alzheimer effects by inhibiting acetylcholinesterase, regulating neurotransmitters, and exerting antioxidative and gastroprotective effects [51–53].

Likewise, various doses (5–50 μM) of naringenin exhibited significant cytotoxic effects in prostate cancer. According to Lim et al. [54], it effectively inhibits the proliferation and migration of cancer cells, induces apoptosis and reactive oxygen species (ROS) production, and disrupts the mitochondrial membrane, with an increased Bax/Bcl-2 ratio, in PC3 and LNCaP cells. Another study reported that it was effective against melanoma cells, exhibiting antiproliferative effects at various doses of 25–100 μM [55]. Furthermore, it exhibited significant lethal effects on glioma cells (U-118 MG) at a dose of 211 μM [56]. Further supporting the previous findings, Zhang et al. [57] reported that 200 mg/kg naringenin significantly reduced TGF- β 1 secretion and intracellular accumulation in breast cancer cells and inhibited their transport from the trans-Golgi network. In addition to its anticancer effects, it has been reported to have antiviral potential against chikungunya and dengue viruses. An in vitro experiment on infected hamster kidney (BHK-21) cells showed antiviral activity against chikungunya virus at a dose of 6.818 μM [58]. Likewise, dengue viral research conducted in Huh75 human hepatoma cells showed that it can prevent dengue viral infection at a concentration of 250 μM [59].

The docking score obtained for amaroswerin indicates a favorable binding affinity toward the active site of *C. jejuni* IMPDH and is comparable to the binding pattern observed for the co-crystallized ligand. Nevertheless, docking scores should be interpreted with caution because they represent approximate scoring functions rather than absolute measures of binding affinity. They are primarily useful for ranking candidate ligands within the same computational framework and do not directly predict experimental inhibitory activity, such as IC_{50} or K_i values. Therefore, the docking results were complemented with 200 ns MD simulations to evaluate the stability of the protein-ligand complex over time. Although the combined docking and MD analyses support amaroswerin as a promising lead compound, its inhibitory activity against IMPDH must ultimately be confirmed through in vitro enzyme inhibition assays and microbiological studies. This integrated interpretation provides a more realistic assessment of the computational findings while acknowledging the inherent limitations of docking-based predictions.

In conclusion, among the investigated phytochemicals, amaroswerin showed the most favorable computational performance against *C. jejuni* inosine monophosphate dehydrogenase. Molecular docking and 200 ns MD simulations indicated strong binding affinity and stable interactions with the target protein. In addition, ADMET and toxicity predictions suggested favorable pharmacokinetic characteristics and a low probability of major toxicity endpoints. These findings identify amaroswerin as a promising computational lead compound for the development of novel IMPDH inhibitors against *C. jejuni*. However, the present results are based entirely on computational analyses, and the predicted antimicrobial activity, binding efficacy, pharmacokinetic properties, and safety require validation through in vitro enzyme inhibition assays, microbiological studies, in vivo investigations, and ultimately clinical studies before any therapeutic application can be considered.

Abbreviations

ADMET: absorption, distribution, metabolism, excretion, and toxicity

BBB: blood-brain barrier

C. jejuni: *Campylobacter jejuni*

CADD: computer-aided drug discovery

CNS: central nervous system
COX-2: cyclooxygenase-2
CYP450: cytochrome P450
GO: Gene Ontology
IL-6: interleukin-6
IMPDH: inosine 5'-monophosphate dehydrogenase
iNOS: inducible nitric oxide synthase
KEGG: Kyoto Encyclopedia of Genes and Genomes
LPS: lipopolysaccharide
MD: molecular dynamics
NF- κ B: nuclear factor kappa-light-chain-enhancer of activated B cells
NO: nitric oxide
NPT: constant number of particles, pressure, and temperature (ensemble)
OCT2: organic cation transporter 2
OPLS: Optimized Potentials for Liquid Simulations
PDB: Protein Data Bank
PPINs: protein-protein interaction networks
RMSD: root mean square deviation
RMSF: root mean square fluctuation
ROS: reactive oxygen species
TIP3P: Transferable Intermolecular Potential with 3 Points
TNF- α : tumor necrosis factor-alpha

Supplementary materials

The supplementary tables for this article are available at: https://www.explorationpub.com/uploads/Article/file/1010196_sup_1.pdf.

Declarations

Acknowledgments

Rengarajan Murugesan sincerely thanks the management of Rajagiri College of Social Sciences for providing the opportunity and necessary support to prepare the manuscript. The author(s) declared that generative AI was used in the creation of this manuscript. During manuscript preparation, the authors used Grammarly Premium to correct language errors. This AI-assisted tool checked spelling, grammar, punctuation, clarity, and plagiarism, suggesting corrections where needed. This collaboration enhanced the manuscript's quality by combining human expertise with AI efficiency, and Turnitin was used to detect plagiarism and AI-generated content.

Author contributions

RM: Methodology, Investigation, Project administration, Supervision, Data curation, Validation, Writing—original draft, Writing—review & editing. KK: Conceptualization, Data curation, Formal analysis. RP: Investigation, Methodology, Software, Writing—review & editing. SJ: Conceptualization, Project administration. KD: Resources, Validation. BK: Visualization, Writing—review & editing. SAC: Data curation, Formal analysis, Software. SP: Methodology, Investigation, Project administration, Supervision, Data

curation, Validation, Writing—original draft, Writing—review & editing. SS: Validation, Visualization, Writing—original draft, Writing—review & editing. All authors read and approved the submitted version.

Conflicts of interest

The authors declare no conflicts of interest.

Ethical approval

Not applicable.

Consent to participate

Not applicable.

Consent to publication

Not applicable.

Availability of data and materials

The data supporting the findings of this study are available within the article and its Supporting Information. Additional datasets generated during the current study are available from the corresponding author upon reasonable request.

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References

1. Luangtongkum T, Jeon B, Han J, Plummer P, Logue CM, Zhang Q. Antibiotic Resistance in *Campylobacter*: Emergence, Transmission and Persistence. *Future Microbiol.* 2009;4:189–200. [DOI] [PubMed] [PMC]
2. Tang Y, Fang L, Xu C, Zhang Q. Antibiotic resistance trends and mechanisms in the foodborne pathogen, *Campylobacter*. *Anim Health Res Rev.* 2017;18:87–98. [DOI] [PubMed]
3. Tumulty M, Di Bari C, Devleeschauwer B, Pires SM, Kabir Z. A systematic review of the methodological considerations in *Campylobacter* burden of disease studies. *PLOS Negl Trop Dis.* 2025;19:e0012681. [DOI] [PubMed] [PMC]
4. Jia S, Xu X, Qu M, Pei Y, Sun S, Liu Y, et al. Longitudinal Trends and Drivers of Antimicrobial Resistance in *Campylobacter* Worldwide (1954–2023). *Zoonoses.* 2025;5. [DOI]
5. Deforet F, Jehanne Q, Bénéjat L, Aptel J, Prat R, Desbiolles C, et al. Combined genomic-proteomic approach in the identification of *Campylobacter coli* amoxicillin-clavulanic acid resistance mechanism in clinical isolates. *Front Microbiol.* 2023;14:1285236. [DOI] [PubMed] [PMC]
6. Rossi DA, Dumont CF, Santos ACS, Vaz MEL, Prado RR, Monteiro GP, et al. Antibiotic Resistance in the Alternative Lifestyles of *Campylobacter jejuni*. *Front Cell Infect Microbiol.* 2021;11:535757. [DOI] [PubMed] [PMC]

7. Peng Y, Moffat JG, DuPai C, Kofoed EM, Skippington E, Modrusan Z, et al. Differential effects of inosine monophosphate dehydrogenase (IMPDH/GuaB) inhibition in *Acinetobacter baumannii* and *Escherichia coli*. *J Bacteriol*. 2024;206:e0010224. [DOI] [PubMed] [PMC]
8. Modi G, Marqus GM, Vippila MR, Gollapalli DR, Kim Y, Manna AC, et al. The Enzymatic Activity of Inosine 5'-Monophosphate Dehydrogenase May Not Be a Vulnerable Target for *Staphylococcus aureus* Infections. *ACS Infect Dis*. 2021;7:3062–76. [DOI] [PubMed] [PMC]
9. Wang X, Rosenberg MM, Kim Y, Maltseva N, Cuny GD, Joachimiak A, et al. Role of the Mobile Active Site Flap in IMP Dehydrogenase Inhibitor Binding. *ACS Infect Dis*. 2025;11:442–52. [DOI] [PubMed] [PMC]
10. Su Y, Wang J, Gao W, Wang R, Yang W, Zhang H, et al. Dynamic metabolites: A bridge between plants and microbes. *Sci Total Environ*. 2023;899:165612. [DOI] [PubMed]
11. Chinnam N, Dadi PK, Sabri SA, Ahmad M, Kabir MA, Ahmad Z. Dietary bioflavonoids inhibit *Escherichia coli* ATP synthase in a differential manner. *Int J Biol Macromol*. 2010;46:478–86. [DOI] [PubMed] [PMC]
12. Huang W, Wang Y, Tian W, Cui X, Tu P, Li J, et al. Biosynthesis Investigations of Terpenoid, Alkaloid, and Flavonoid Antimicrobial Agents Derived from Medicinal Plants. *Antibiotics*. 2022;11:1380. [DOI] [PubMed] [PMC]
13. Xu W, Lin Z, Cortez-Jugo C, Qiao GG, Caruso F. Antimicrobial Phenolic Materials: From Assembly to Function. *Angew Chem Int Ed*. 2025;64:e202423654. [DOI] [PubMed]
14. Zheng D, Huang C, Huang H, Zhao Y, Khan MRU, Zhao H, et al. Antibacterial Mechanism of Curcumin: A Review. *Chem Biodivers*. 2020;17:e2000171. [DOI] [PubMed]
15. Jadimurthy R, Jagadish S, Nayak SC, Kumar S, Mohan CD, Rangappa KS. Phytochemicals as Invaluable Sources of Potent Antimicrobial Agents to Combat Antibiotic Resistance. *Life*. 2023;13:948. [DOI] [PubMed] [PMC]
16. Ahmad Z, Hassan SS, Azim S. A Therapeutic Connection between Dietary Phytochemicals and ATP Synthase. *Curr Med Chem*. 2017;24:3894–906. [DOI] [PubMed] [PMC]
17. Qi W, Qi W, Xiong D, Long M. Quercetin: Its Antioxidant Mechanism, Antibacterial Properties and Potential Application in Prevention and Control of Toxipathy. *Molecules*. 2022;27:6545. [DOI] [PubMed] [PMC]
18. Papaneophytou C. Phytochemical Quorum-Sensing Inhibitors Against Bacterial Pathogens: Mechanisms of Action and Translational Challenges. *Curr Issues Mol Biol*. 2026;48:214. [DOI] [PubMed] [PMC]
19. Prathviraj R, Adithya K, Rajeev R, Khan RT, Hassan S, Selvin J, et al. Alleviation of migraine through gut microbiota-brain axis and dietary interventions: Coupling epigenetic network information with critical literary survey. *Trends Food Sci Technol*. 2023;141:104174. [DOI]
20. Vinothkanna A, Chen L, Prathviraj R, Prabhu S, Thiruvengadam R, Sivakumar TR. Ashwagandharishta compounds possess affinity interactions against diabetes-linked liver cancer: Implications from network pharmacology, molecular docking and dynamics simulations. *S Afr J Bot*. 2025;177:26–49. [DOI]
21. Szklarczyk D, Kirsch R, Koutrouli M, Nastou K, Mehryary F, Hachilif R, et al. The STITCH database in 2023: protein–protein association networks and functional enrichment analyses for any sequenced genome of interest. *Nucleic Acids Res*. 2022;51:D638–46. [DOI] [PubMed] [PMC]
22. Shannon P, Markiel A, Ozier O, Baliga NS, Wang JT, Ramage D, et al. Cytoscape: A Software Environment for Integrated Models of Biomolecular Interaction Networks. *Genome Res*. 2003;13:2498–504. [DOI] [PubMed] [PMC]
23. Lee IC, Bae JS. Anti-Inflammatory Activities of (+)-Afzelechin against Lipopolysaccharide-Induced Inflammation. *Biomol Ther (Seoul)*. 2024;32:467–73. [DOI] [PubMed] [PMC]
24. Sharma S, Sharma YP, Bhardwaj C, Mehta A. Quantification of Amaraswerin and Amarogentin in Different Parts of *Swertia chirayita* by Chromatographic Analysis. *Int Res J Pure Appl Chem*. 2019;17:1–7. [DOI]

25. Patel K, Patel DK. Medicinal Importance and Therapeutic Benefit of Bioactive Flavonoid Eriocitrin: An Update on Pharmacological Activity and Analytical Aspects. *Nat Prod J*. 2024;14. [DOI]
26. Uçar K, Göktaş Z. Biological activities of naringenin: A narrative review based on in vitro and in vivo studies. *Nutr Res*. 2023;119:43–55. [DOI] [PubMed]
27. Prashanth D, John S. Antibacterial activity of *Peganum harmala*. *Fitoterapia*. 1999;70:438–9. [DOI]
28. Kim Y, Maltseva N, Makowska-Grzyska M, Gu M, Gollapalli D, Hedstrom L, et al. Crystal Structure of the Catalytic Domain of the Inosine Monophosphate Dehydrogenase from *Campylobacter jejuni* in the complex with inhibitor p200 [Internet]. [cited 2026 Apr 16]. Available from: <https://doi.org/10.2210/pdb5uqg/pdb>
29. Samy CRA, Karunanithi K, Sheshadhri J, Rengarajan M, Srinivasan P, Cherian P. (R)-(+)-Rosmarinic Acid as an Inhibitor of Herpes and Dengue Virus Replication: an In Silico Assessment. *Rev Bras Farmacogn*. 2023;33:543–50. [DOI] [PubMed] [PMC]
30. Vijaya Prabhu S, Suman S, Chellapandi A, Baskaradoss P, Padma Priya S, Pandurangan T, et al. Targeting Quorum Sensing LsrR Protein in *E. coli*: A Computational Approach to Screen the Plant Bioactive Compounds as Inhibitors of Biofilm Formation in Urinary Tract Infections. *ASSAY Drug Dev Technol*. 2026;24:517–37. [DOI] [PubMed]
31. Sureshkumar J, Prabhu S, Mariraj M, Amalraj S, Kadaikunnan S, Murugesan R, et al. Chemical profiling, proximate composition, and antibacterial, antioxidant, anti-diabetic, and anti-rheumatoid arthritis properties of wild fern rhizome (*Drynaria quercifolia* (L.) J. Sm): In vitro and drug discovery approaches. *Microb Pathog*. 2026;214:108369. [DOI] [PubMed]
32. Sastry GM, Adzhigirey M, Day T, Annabhimoju R, Sherman W. Protein and ligand preparation: parameters, protocols, and influence on virtual screening enrichments. *J Comput-Aided Mol Des*. 2013;27:221–34. [DOI] [PubMed]
33. Priya SP, Chaitanya NC, Padmanabhan V, Sridhar SB, Sivaswamy V, Amalraj S, et al. Targeting calcineurin to combat fungal infections: Computational insights into phytochemicals as promising antifungal agents. *Lett Drug Des Discov*. 2025;22:100100. [DOI]
34. Bintsis T. Foodborne pathogens. *AIMS Microbiol*. 2017;3:529–63. [DOI] [PubMed] [PMC]
35. Mead PS, Slutsker L, Dietz V, McCaig LF, Bresee JS, Shapiro C, et al. Food-Related Illness and Death in the United States. *Emerg Infect Dis*. 1999;5:607–25. [DOI] [PubMed] [PMC]
36. Omukisa KK. Investigating Medicinal Plants' Efficacy in Treating Foodborne Diarrhea. *NEWPORT INT J OF RES IN MED SCI*. 2025;6:137–46. [DOI]
37. Smith-Hall C, Larsen HO, Pouliot M. People, plants and health: a conceptual framework for assessing changes in medicinal plant consumption. *J Ethnobiol Ethnomed*. 2012;8:43. [DOI] [PubMed] [PMC]
38. Bajorath J. Computer-aided drug discovery. *F1000Res*. 2015;4:F1000 Faculty Rev-630. [DOI] [PubMed] [PMC]
39. Xiang Y, Haixia W, Zenggen L, Yanduo T. Anti-inflammatory activity of compounds isolated from *Swertia mussotii*. *Nat Prod Res*. 2019;33:598–601. [DOI] [PubMed]
40. Niiho Y, Yamazaki T, Nakajima Y, Yamamoto T, Ando H, Hirai Y, et al. Gastroprotective effects of bitter principles isolated from Gentian root and *Swertia* herb on experimentally-induced gastric lesions in rats. *J Nat Med*. 2006;60:82–8. [DOI]
41. Liu J, Huang H, Huang Z, Ma Y, Zhang L, He Y, et al. Eriocitrin in combination with resveratrol ameliorates LPS-induced inflammation in RAW264.7 cells and relieves TPA-induced mouse ear edema. *J Funct Foods*. 2019;56:321–32. [DOI]
42. Guo G, Shi W, Shi F, Gong W, Li F, Zhou G, et al. Anti-inflammatory effects of eriocitrin against the dextran sulfate sodium-induced experimental colitis in murine model. *J Biochem Mol Toxicol*. 2019;33:e22400. [DOI] [PubMed]

43. He J, Zhou D, Yan B. Eriocitrin alleviates oxidative stress and inflammatory response in cerebral ischemia reperfusion rats by regulating phosphorylation levels of Nrf2/NQO-1/HO-1/NF- κ B p65 proteins. *Ann Transl Med.* 2020;8:757. [DOI] [PubMed] [PMC]
44. Denaro M, Smeriglio A, Trombetta D. Antioxidant and Anti-Inflammatory Activity of Citrus Flavanones Mix and Its Stability after In Vitro Simulated Digestion. *Antioxidants.* 2021;10:140. [DOI] [PubMed] [PMC]
45. Xu J, Ma L, Fu P. Eriocitrin attenuates ischemia reperfusion-induced oxidative stress and inflammation in rats with acute kidney injury by regulating the dual-specificity phosphatase 14 (DUSP14)-mediated Nrf2 and nuclear factor- κ B (NF- κ B) pathways. *Ann Transl Med.* 2021;9:350. [DOI] [PubMed] [PMC]
46. Carvalho JS, Ramadan D, de Paiva Gonçalves V, Maquera-Huacho PM, Assis RP, Lima TFO, et al. Impact of citrus flavonoid supplementation on inflammation in lipopolysaccharide-induced periodontal disease in mice. *Food Funct.* 2021;12:5007–17. [DOI] [PubMed]
47. Cho S, Park YJ, Kim E, Bae JS. The Therapeutic Potential of (+)-Afzelechin for Alleviating Sepsis-Associated Pulmonary Injury. *J Med Food.* 2024;27:12–21. [DOI] [PubMed]
48. Zhang Q, Wang H, Ran C, Lyu Y, Li F, Yao Y, et al. Anti-inflammatory effects of amarogentin on 2,4-dinitrochlorobenzene-induced atopic dermatitis-like mice and in HaCat cells. *Anim Models Exp Med.* 2022;6:255–65. [DOI] [PubMed] [PMC]
49. Zhang Y, Zhao H, Li H, Cao W, Wang F, Zhang T, et al. Protective Effects of Amarogentin against Carbon Tetrachloride-Induced Liver Fibrosis in Mice. *Molecules.* 2017;22:754. [DOI] [PubMed] [PMC]
50. Khaliq T, Misra P, Gupta S, Reddy KP, Kant R, Maulik PR, et al. Peganine hydrochloride dihydrate an orally active antileishmanial agent. *Bioorg Med Chem Lett.* 2009;19:2585–6. [DOI] [PubMed]
51. Liu D, Zhang L, Duan LX, Wu JJ, Hu M, Liu ZQ, et al. Potential of herb-drug / herb interactions between substrates and inhibitors of UGTs derived from herbal medicines. *Pharmacol Res.* 2019;150:104510. [DOI] [PubMed]
52. Deng G, Wu C, Rong X, Li S, Ju Z, Wang Y, et al. Ameliorative effect of deoxyvasicine on scopolamine-induced cognitive dysfunction by restoration of cholinergic function in mice. *Phytomedicine.* 2019;63:153007. [DOI] [PubMed]
53. Singh VK, Mishra V, Tiwari S, Khaliq T, Barthwal MK, Pandey HP, et al. Anti-secretory and cytoprotective effects of peganine hydrochloride isolated from the seeds of *Peganum harmala* on gastric ulcers. *Phytomedicine.* 2013;20:1180–5. [DOI] [PubMed]
54. Lim W, Park S, Bazer FW, Song G. Naringenin-Induced Apoptotic Cell Death in Prostate Cancer Cells Is Mediated via the PI3K/AKT and MAPK Signaling Pathways. *J Cell Biochem.* 2017;118:1118–31. [DOI] [PubMed]
55. Nasr Bouzaiene N, Chaabane F, Sassi A, Chekir-Ghedira L, Ghedira K. Effect of apigenin-7-glucoside, genkwanin and naringenin on tyrosinase activity and melanin synthesis in B16F10 melanoma cells. *Life Sci.* 2016;144:80–5. [DOI] [PubMed]
56. Stompor M, Uram Ł, Podgórski R. In Vitro Effect of 8-Prenylnaringenin and Naringenin on Fibroblasts and Glioblastoma Cells-Cellular Accumulation and Cytotoxicity. *Molecules.* 2017;22:1092. [DOI] [PubMed] [PMC]
57. Zhang F, Dong W, Zeng W, Zhang L, Zhang C, Qiu Y, et al. Naringenin prevents TGF- β 1 secretion from breast cancer and suppresses pulmonary metastasis by inhibiting PKC activation. *Breast Cancer Res.* 2016;18:38. [DOI] [PubMed] [PMC]
58. Ahmadi A, Hassandarvish P, Lani R, Yadollahi P, Jokar A, Bakar SA, et al. Inhibition of chikungunya virus replication by hesperetin and naringenin. *RSC Adv.* 2016;6:69421–30. [DOI]
59. Frabasile S, Koishi AC, Kuczera D, Silveira GF, Verri WA Jr, Duarte Dos Santos CN, et al. The citrus flavanone naringenin impairs dengue virus replication in human cells. *Sci Rep.* 2017;7:41864. [DOI] [PubMed] [PMC]