International Journal of Environmental Sciences
ISSN: 2229-7359
Vol. 11 No. 21s, 2025
https://theaspd.com/index.php

Structure-Based Discovery Of Marine Natural Inhibitors Against AMPC **B**-Lactamase Of Burkholderia Multivorans: An In Silico Pharmacological And Dynamic Simulation Study

P. R. Kiresee Saghana[1], S. Shanmugavani[2], R. Priya[3] and Hasifa Banu M[4]
[1,2]Assistant Professor Department of Bioinformatics, School of Life Sciences, Vels Institute of Science, Technology and Advanced Studies (VISTAS), Pallavaram, Chennai-600117, Tamil Nadu, India
[3]Assistant Professor Department of Bioinformatics, School of Life Sciences, Vels Institute of Science, Technology and Advanced Studies (VISTAS), Pallavaram, Chennai-600117, Tamil Nadu, India
[4]M.Sc.,Student Department of Bioinformatics, School of Life Sciences, Vels Institute of Science, Technology and Advanced Studies (VISTAS), Pallavaram, Chennai-600117, Tamil Nadu, India

Abstract: *The alarming rise of antibiotic resistance in Burkholderia multivorans has been closely linked to the expression of AmpC β-lactamase. This study aimed to identify potential marine-derived inhibitors targeting this enzyme through a comprehensive in silico approach. The functional role of AmpC was explored via KEGG and STRING databases, revealing its involvement in β-lactam resistance pathways and key protein–protein interactions. The crystal structure of AmpC (PDB ID: 3W4Q) was refined and validated before being subjected to site-directed virtual screening. A library of 60 filtered marine natural compounds from the CMNPD database was screened using AutoDock Vina, guided by CASTp-predicted active sites. Top hits were evaluated for binding affinity, drug-likeness, ADMET properties, and cardiotoxic risk using SwissADME and PRED-hERG tools. Among the shortlisted compounds, oplopanone and ellagic acid showed strong binding affinities, with oplopanone displaying the most stable interaction (−8.13 kcal/mol). Molecular dynamics simulations of the AmpC-oplopanone complex over 50 ns confirmed its dynamic stability and favorable interaction profile under physiological conditions. These results highlight oplopanone and ellagic acid as promising lead compounds for further in vitro and in vivo validation in the development of novel β-lactamase inhibitors.*

Keywords: *AmpC β-lactamase, Burkholderia multivorans, Marine natural products, Molecular docking and Molecular dynamics simulation*

INTRODUCTION

Antimicrobial resistance (AMR) is a growing global health crisis, threatening the efficacy of current antibiotic therapies and complicating the management of infectious diseases. A major contributor to this crisis is the widespread emergence of multidrug-resistant Gram-negative pathogens such as Escherichia coli, Pseudomonas aeruginosa, and Burkholderia cepacia complex (Bcc) species, including Burkholderia multivorans and Burkholderia cenocepacia [1–3]. These opportunistic bacteria are particularly concerning in immunocompromised individuals, especially cystic fibrosis (CF) patients, where they lead to persistent respiratory infections, rapid lung function deterioration, and, in some cases, fatal septicemia known as "cepacia syndrome" [4–6].

The Bcc, comprising over 20 closely related species, exhibit high genomic plasticity, inherent resistance to antibiotics, and adaptability to harsh environments, all of which contribute to their virulence and persistence in clinical settings [7,8]. In CF, defective mucociliary clearance due to mutations in the CFTR gene fosters the colonization of lung tissues by bacteria such as B. cenocepacia and B. multivorans, which are associated with accelerated lung function decline and post-transplant complications [9,10]. Infections caused by these organisms are particularly hard to treat due to their intrinsic resistance mechanisms and the lack of effective β-lactamase inhibitors.

A key resistance mechanism in these bacteria is the production of AmpC β-lactamases—class C serine hydrolases that hydrolyze a wide range of β-lactam antibiotics including cephalosporins and penicillins [11,12]. Unlike class A enzymes, AmpC β-lactamases are poorly inhibited by traditional β-lactamase inhibitors such as clavulanic acid and sulbactam [13]. Their chromosomal or plasmid-mediated expression, often triggered by antibiotic exposure, facilitates the horizontal transfer of resistance genes across bacterial populations, further complicating therapeutic strategies [14,15]. Despite their clinical significance, no FDA-approved inhibitor specifically targeting AmpC β-lactamases currently exists, underscoring an urgent need for novel drug candidates, this emerging evidence highlights that natural

products, particularly those derived from marine environments, offer a promising avenue for antibiotic discovery. [16,17]. Marine organisms, including bacteria, fungi, and algae, inhabit extreme and diverse ecological niches and produce a vast array of unique secondary metabolites with potent antimicrobial properties [18–20]. These bioactive compounds are structurally diverse and often exhibit novel mechanisms of action, making them attractive candidates for targeting resistant enzymes such as AmpC β-lactamases [21,22].

The Comprehensive Marine Natural Products Database (CMNPD) has catalogued over 47,000 such compounds, providing a rich resource for virtual screening and pharmacological exploration [23]. Many of these marine-derived molecules possess favourable drug-likeness profiles and unexplored pharmacophores that are underrepresented in conventional chemical libraries [24]. In the context of multidrug-resistant pathogens like B. multivorans, marine natural products represent a largely untapped reservoir of potential therapeutics.

The advancements in in silico methodologies, including Computer-Aided Drug Design (CADD), have revolutionized the drug discovery process by enabling rapid and cost-effective screening of large chemical libraries [25,26]. Techniques such as site-directed virtual screening (SDVS), molecular docking, and molecular dynamics (MD) simulations provide valuable insights into the binding interactions, stability, and pharmacokinetic behaviour of candidate inhibitors [27,28]. These computational strategies allow researchers to prioritize compounds with the highest therapeutic potential for further in vitro and in vivo validation.

In this study, we applied an integrative in silico approach to identify marine-derived inhibitors of AmpC β-lactamase from B. multivorans. The three-dimensional structure of the AmpC enzyme (PDB ID: 3W4Q) was retrieved, refined through energy minimization, and analyzed for active site characterization using CASTp[17]. Subsequently, a drug-likeness-filtered subset of CMNPD compounds was screened using AutoDock Vina. The top-ranked ligand-enzyme complexes were evaluated for their binding interactions, pharmacokinetic profiles using SwissADME, and potential cardiotoxicity via PRED-hERG screening. The binding stability and conformational dynamics, molecular dynamics simulations were conducted for 50 nanoseconds using the AMBER20 suite in an explicit solvent environment. Trajectory analyses, including root-mean-square deviation (RMSD) and hydrogen bond profiling, were performed with CPPTRAJ and VMD tools, respectively. The functional implications of AmpC inhibition were explored through KEGG pathway mapping and STRING-based protein-protein interaction (PPI) analysis.

Genomic and virulence landscape of B. cenocepacia and B. multivorans highlights the importance of developing novel therapeutic strategies. Genomic studies have revealed the presence of mobile genetic elements, including pathogenicity islands like the B. cepacia epidemic strain marker (BCESM), which encode key virulence factors such as quorum sensing systems, porins, and secretion proteins [29,30]. These features contribute to the pathogen's adaptability and resistance to oxidative stress, antibiotics, and host immune responses [31,32].

In quorum sensing systems, notably CepIR and CciIR, regulate a wide range of genes associated with virulence, motility, biofilm formation, and resistance [33]. The interplay between quorum sensing and β-lactamase expression suggests that targeting both systems simultaneously may yield synergistic therapeutic effects, the efflux pumps—such as those belonging to the RND, MFS, and ABC transporter families—confer multidrug resistance and further reduce the efficacy of conventional antibiotics [34], limitations of current treatments and the pathogen's genomic complexity, repurposing marine natural products through computational pipelines offers a promising strategy for combating β-lactamase-mediated resistance. Preclinical studies, involving in vivo pharmacokinetic and toxicity assessments, are essential to translate these in silico findings into viable clinical interventions.

MATERIALS AND METHODS

Data Identification and Retrieval

To explore the functional characteristics of the AmpC β-lactamase in Burkholderia multivorans, protein-protein interaction (PPI) analysis was initially performed using the STRING database (https://string-db.org). The three-dimensional crystal structure of AmpC β-lactamase from B. multivorans, resolved at a resolution of 2.37 Å with an R-value of 0.186, was retrieved from the Protein Data Bank (PDB ID: 3W4Q). Functional annotation was conducted via the KEGG pathway database using KO entry K01467, confirming the classification of the selected AmpC isoforms within the β-lactam resistance pathway. The

integration of STRING and KEGG analyses provided both functional and regulatory insights into AmpC activity.

Protein–Protein Interaction Analysis

The STRING database was used to construct a comprehensive PPI network for AmpC β-lactamase and its associated proteins. High-confidence interaction scores were set, and functional modules were analyzed. The resulting network revealed significant associations with proteins involved in peptidoglycan biosynthesis and β-lactam resistance, notably dacB, mtgA, and penR, indicating a regulatory network underpinning resistance phenotypes.

Target Protein Preparation

The AmpC structure was refined using UCSF Chimera v1.15. Co-crystallized ligands, extra chains, and water molecules were removed to prepare the structure for docking. Energy minimization was executed via 1000 steps using steepest descent and conjugate gradient algorithms with a step size of 0.02 Å. The AMBER ff14SB force field was applied to optimize both backbone and side chain residues. Ramachandran plot analysis was carried out using PDBSum to evaluate structural quality, comparing pre- and post-minimization conformations. Superimposition with the original structure confirmed structural stability.

Ligand Library Preparation from CMNPD

Marine-derived natural product structures were retrieved from the Comprehensive Marine Natural Products Database (CMNPD; https://www.cmnpd.org), comprising over 47,000 compounds from bacteria, fungi, and algae. The dataset was downloaded in .sdf format and subjected to filtering using FAFDrugs4 (http://fafdrugs4.mti.univ-paris-diderot.fr) to eliminate non-drug-like compounds, toxicophores, and PAINS using the Eli Lilly MedChem rules. Drug-like compounds were energy-minimized and converted to .pdbqt format using PyRx 0.8 for docking preparation.

Active Site Prediction

The Computed Atlas of Surface Topography of Proteins (CASTp 3.0; http://sts.bioe.uic.edu/castp) was used to identify the binding pockets on the AmpC enzyme. The analysis provided detailed measurements of solvent-accessible surface areas and volumes using Richards' and Connolly's models. This guided the selection of docking grid parameters for site-directed virtual screening.

Site-Directed Virtual Screening (SDVS)

The filtered CMNPD compounds were screened against the prepared AmpC structure using AutoDock Vina (version 1.1.2). A rigid docking protocol was employed. The docking grid was centered at the predicted active site, and 100 iterations were generated per compound. Vina scores were used to rank the compounds based on binding affinities. Top hits were visualized using UCSF Chimera and BIOVIA Discovery Studio Visualizer v21.1.0.20298 for interaction analysis.

Molecular Docking Protocol

Docking was carried out using AutoDock Vina (version 1.1.2) by first preparing the system wherein ligand and protein structures were converted to .pdbqt format using AutoDock Tools v1.5.6. Polar hydrogen atoms were added, Kollman charges were assigned to the protein, and torsion trees were defined to enable flexible docking for the ligands. The docking simulations were executed with an exhaustiveness parameter set to 32 to ensure thorough conformational sampling. Each ligand was subjected to 20 independent docking runs, generating up to 10 possible binding modes per run. AutoDock Vina is an open-source molecular docking software available at: https://vina.scripps.edu.

Visualization of Ligand–Protein Interactions

Ligand–receptor binding modes were analyzed using BIOVIA Discovery Studio Visualizer. Molecular interactions including conventional and non-conventional hydrogen bonds, π–π stacking, van der Waals forces, and electrostatic interactions were mapped for the top-ranked complexes. The visualizer is freely available at: https://discover.3ds.com/discovery-studio-visualizer-download.

Toxicity and ADMET Profiling

Physicochemical and pharmacokinetic profiles of the top hits were evaluated using SwissADME (http://www.swissadme.ch). This tool assessed water solubility, gastrointestinal absorption, blood–brain barrier permeability, skin permeability, synthetic accessibility, and PAINS alerts. Lipinski's Rule of Five and other medicinal chemistry filters were applied. Cardiac toxicity predictions were performed using the PRED-hERG server (https://predherg.labmol.com.br) to evaluate the potential of the molecules to

inhibit the hERG potassium channel, a known cause of QT prolongation.

Molecular Dynamics (MD) Simulations

Top-performing ligand–AmpC complexes were subjected to 50 ns MD simulations using AMBER20. The ff14SB force field was applied to the protein, while the GAFF force field was used for ligand parameterization. The systems were solvated in a TIP3P water box (12 Å) with appropriate counterions. Energy minimization was conducted for 1000 steps, followed by gradual heating to 310 K using Langevin dynamics for 20 ps. Equilibration was performed for 100 ps, and production was run for 100 ns. Stability analyses including root mean square deviation (RMSD) and hydrogen bonding were performed using the AMBER CPPTRAJ module. Visual Molecular Dynamics (VMD) was used for dynamic interaction visualization across simulation trajectories.

RESULT AND DISCUSSION

Protein-Protein Interaction (PPI) Network

A protein-protein interaction (PPI) network was constructed using the STRING database to explore the functional associations of AmpC β-lactamase and related proteins. The analysis revealed that **ampC-3** functions as a central hub, exhibiting strong interactions with several key proteins, including **ampC-2**, **penR**, **dacB**, **mtgA**, **BAG44514.1**, and **BAG45868.1**, as shown in **Figure 1**. The association with **penR**, a known transcriptional regulator, suggests a regulatory role in the expression of β-lactamase genes, while interactions with **dacB** and **mtgA** indicate involvement in peptidoglycan biosynthesis and cell wall maintenance. These interactions highlight the coordinated function of resistance mechanisms beyond enzyme activity, emphasizing the role of structural and regulatory proteins. Additionally, uncharacterized proteins such as **BAG44514.1** and **BAG45868.1** may represent novel contributors to β-lactam resistance, potentially assisting in the stabilization or secretion of AmpC enzymes. The interconnected nature of this network underscores the multifactorial basis of antibiotic resistance and suggests that ampC-3 plays a crucial role in maintaining resistance and cellular integrity under antibiotic stress.

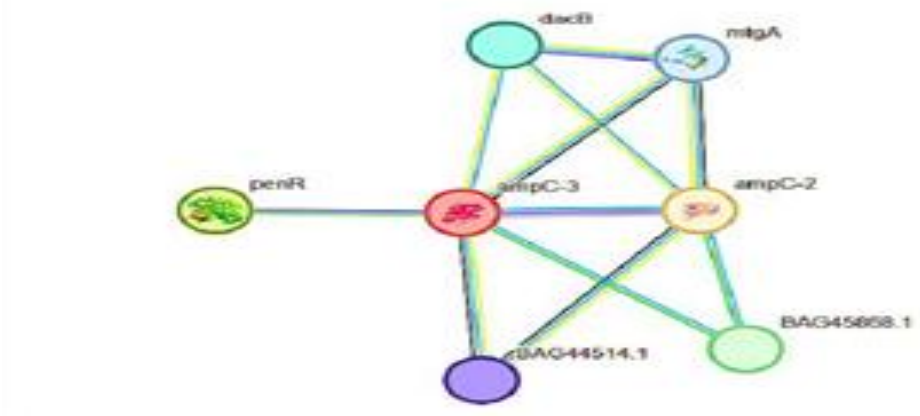


Figure 1 shows: Protein-Protein Interaction (PPI) Network

The mechanistic overview of β-lactam resistance pathways, illustrated in **Figure 2**, reveals the complex and multifaceted defense mechanisms employed by bacteria to counteract β-lactam antibiotics. The diagram highlights the role of multiple components, including β-lactamases (Class A–D), penicillin-binding proteins (PBPs), efflux pumps, porin alterations, and regulatory systems. Central to this resistance mechanism is the hydrolytic activity of β-lactamases such as AmpC, encoded by the ampC gene, which inactivates β-lactam antibiotics by breaking their β-lactam ring. Additionally, the downregulation or loss of porins like OmpF and OmpC reduces drug uptake, while efflux pumps such as the RND-type AbaQ actively expel antibiotics from the cell. Regulatory elements like AmpR, BlaR, and MarR families also play crucial roles in modulating gene expression under antibiotic stress. The interplay between these systems, including the overexpression of efflux genes and reduced binding affinity of altered PBPs, reinforces bacterial survival. These findings underline the necessity of targeting multiple resistance components simultaneously to develop more effective therapeutic strategies against β-lactam-resistant pathogens.

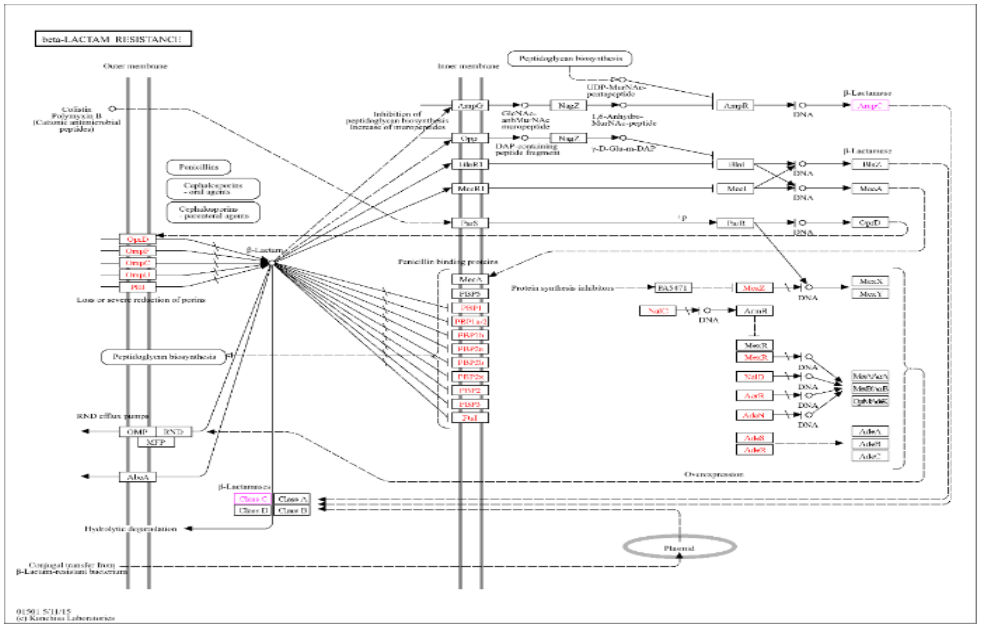
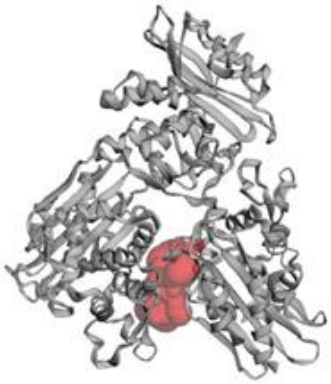


Figure 2 shows: Functional annotation drawn from KEGG pathway mapping

As shown in Fig. 3 , the G-factor values were close to zero for all parameters, including phi-psi and chi angles, with an overall average of –0.03, suggesting that the model geometry is well within expected ranges. Fig. 4 also supports these findings with a consistent G-factor distribution across different dihedral parameters. The three-dimensional structure of the modeled protein, depicted in Fig. 5, illustrates the active site pocket highlighted in red, revealing a well-defined and accessible ligand-binding region. This comprehensive validation indicates that the protein structure is reliable and suitable for downstream applications such as docking and dynamic simulation studies.dvadadv



2. G-Factors

Parameter	Score	Score
Dihedral amgles:=		
Phi-psi distribbution	−0.04	
Chi1-chi2 distribution	0.18	
Chi1 only	0.03	
Chi3 & chi4	0.56	−0.09
Omega	−0.48	
Main-chain covalent forces:=		
Main-chain bond lengths	0.37	0.01
Main-chain bond angles	−0.25	
OVERALL AVERAGE		−0.03

2. G-Factors

Parameter	Score	Average Score
Dihedral anggles:=		
Phi-psi distribbution	−0.03	
Chi1-chi2 distribution	0.21	
Chi1 only	0.04	
Chi3 & chi4	0.53	
Omega	−0.50*	−0.09
Main-chain covalent forces:=		
Main-chain bond lengths	0.37	
Main-chain bond angles	−0.24	0.01
OVERALL AVERAGE		−0.03

Figure 3 shows: 3w4q before and after minimization

To identify potential inhibitors of EPSP synthase, a curated dataset of approximately 47,000 compounds from the Comprehensive Marine Natural Products Database (CMNPD)—which includes molecules derived from marine bacteria, fungi, and algae—was employed. This database was chosen for its exceptional chemical diversity and the presence of bioactive molecules with drug-like characteristics. The CMNPD library, downloaded in .sdf format, was rigorously filtered using the FAFDrugs4 server, applying Lipinski's Rule of Five to ensure oral bioavailability and Eli Lilly's MedChem rules to exclude compounds with undesirable structural alerts. This screening yielded 59 high-confidence compounds, all sourced from the Plantae kingdom, making them well-suited for

Figure 5 shows: Active Site of 3w4q

interaction with eukaryotic targets. These filtered compounds were then prioritized for molecular docking against the AmpC enzyme, with the aim of identifying novel marine-derived, plant-compatible chemotypes as sustainable herbicide leads. Among the promising candidates were structurally diverse compounds such as Lenthionine, Chilenone, Grateloupine, Jacaranone, Carnosadine, Microthecin, gloiosiphone B, aconitate derivatives (A–F), maricyclohexene A, Vidalenolone, and Ellagic Acid. Detailed physicochemical evaluation of selected lead compounds revealed that (1E,5Z)-1,6-dichloro-2-methylhepta-1,5-dien-3-ol, trideca-3,6,9-trienoic acid, and 3-(hydroxyacetyl)indole possessed balanced hydrogen bonding capacity (2–4 donors/acceptors), suitable molecular weight (175–196 Da), and moderate topological polar surface area (TPSA 37.3–53.09 Å^2), indicating favorable bioavailability and interaction potential. Maricyclohexene A and oplopanone also demonstrated high saturation (Csp^3 values of 0.93 and 0.80 respectively), suggesting better 3D fit within target binding pockets. In contrast, debilone displayed high aromaticity and molar refractivity, accompanied by an elevated TPSA (141.3 Å^2), which may hinder its membrane permeability despite its structural complexity. Ellagic Acid similarly showed multiple hydrogen bond acceptors and donors, supporting its bioactive potential. Overall, this analysis highlights the drug-like nature and binding potential of marine-origin compounds, with particular promise shown by trideca-3,6,9-trienoic acid, 3-(hydroxyacetyl)indole, and maricyclohexene A as eco-compatible EPSP synthase inhibitors.

Table 1 shows: Physicochemical Properties

Compounds	Formula	MW	Heavy atoms	Aromatic heavy atoms	Fraction Csp3	Rotatable bonds	H-bond acceptors	H-bond donors	MR	TPSA
(1E,5Z)-1,6-dichloro-2-methylhepta-1,5-dien-3-ol	C11H18	150.26	11	0	0.45	4	0	0	53.57	0
trideca-3,6,9-trienoic acid	C12H18O2	194.27	14	0	0.42	7	2	1	60.15	37.3
3-(hydroxyacetyl)indole	C10H9NO2	175.18	13	9	0.1	2	2	2	49.66	53.09
Maricyclohexene A	C12H20O2	196.29	14	0	0.67	1	2	2	58.8	40.46
oplopanone	C15H26O2	238.37	17	0	0.93	2	2	1	71.39	37.3
debilone	C15H22O2	234.33	17	0	0.8	0	2	1	68.24	37.3
Ellagic Acid	C14H6O8	302.19	22	16	0	0	8	4	75.31	141.34

Fig 6 shows the lipophilicity profile of four selected marine-derived compounds is illustrated in the radial chart. Among them, (1E,5Z)-1,6-dichloro-2-methylhepta-1,5-dien-3-ol exhibits the highest lipophilicity values, indicating strong membrane permeability potential. 3-(hydroxyacetyl)indole and trideca-3,6,9-trienoic acid display moderate lipophilicity, suggesting balanced hydrophilic-lipophilic characteristics favorable for bioactivity. Maricyclohexene A shows the lowest lipophilicity among the group, potentially reflecting higher solubility and reduced membrane interaction.

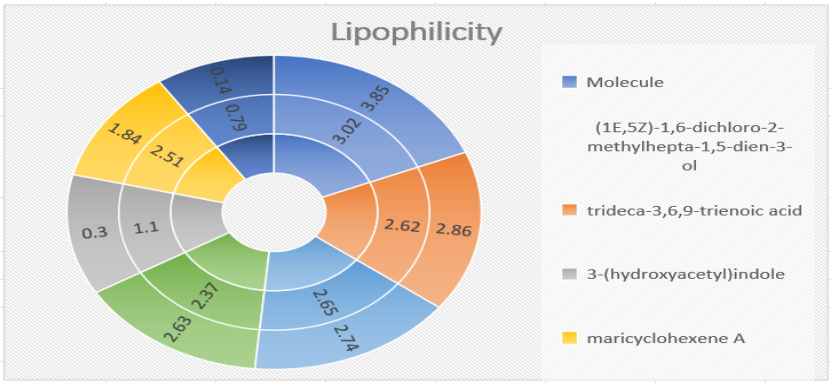


Figure 6: shows the Lipophilicity

The cardiac toxicity assessment of seven selected marine-derived compounds revealed that all were classified as non-blockers, indicating a low risk of hERG channel inhibition. This table 2 shows the property is crucial for ensuring cardiovascular safety in drug development. Notably, compounds such as (1E,5Z)-1,6-dichloro-2-methylhepta-1,5-dien-3-ol, trideca-3,6,9-trienoic acid, and ellagic acid showed favorable toxicity profiles. These results support their further evaluation as safe candidates for EPSP synthase inhibition.

Table 2 shows: Cardiac-Toxicity Profiling

S.NO	COMPOUND	CARDIAC-TOXICITY
1.	(1E,5Z)-1,6-dichloro-2-methylhepta-1,5-dien-3-ol	Non-blocker
2.	trideca-3,6,9-trienoic acid	Non-blocker
3.	3-(hydroxyacetyl)indole	Non-blocker
4.	maricyclohexene A	Non-blocker
5.	Oplopanone	Non-blocker
6.	Debilone	Non-blocker
7.	Ellagic Acid	Non-blocker

Figure 7 illustrates the optimal binding interaction between AmpC β-lactamase and the marine-derived compound Oplopanone. The 3D structure highlights Oplopanone securely positioned within the active site, characterized by α-helices, β-sheets, and loop regions, while the 2D interaction map reveals hydrogen bonding with TYR:D74 and THR:B206, along with Pi-alkyl interactions involving VAL:B185. These contacts suggest Oplopanone's favorable fit and stabilizing effect, marking it as a potential inhibitor candidate. Figure 8 further demonstrates the strong binding affinity between AmpC β-lactamase and another marine-derived compound, Debilone. The 3D visualization shows Debilone embedded within the active site, and the 2D interaction diagram reveals stabilizing forces such as hydrogen bonds, van der Waals contacts, and π-π stacking with residues like TYR B:192, ASN B:101, and THR B:206. Together, Figures 7 and 8 support the candidacy of Oplopanone and Debilone as effective inhibitors, providing valuable leads for the development of eco-friendly herbicides targeting AmpC β-lactamase.

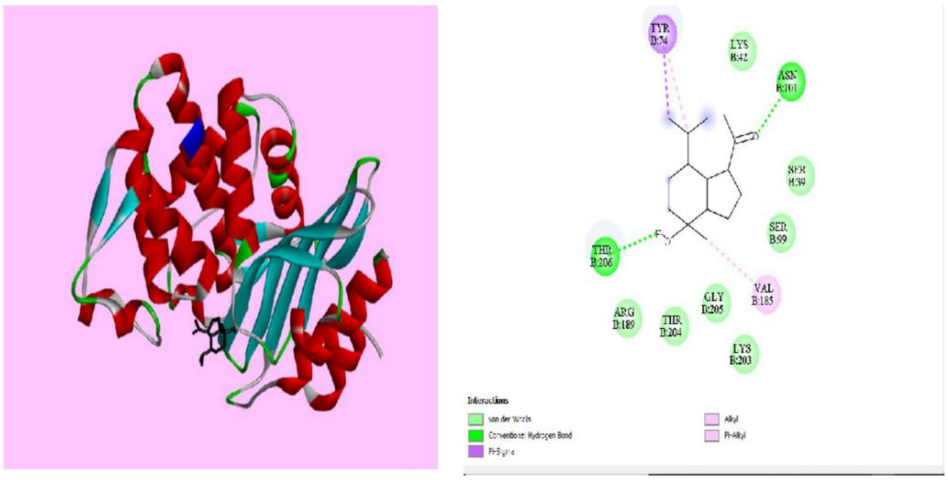


Figure 7: shows the 3D and 2D structures of the highest-affinity binding complex between AmpC β-lactamase and Oplopanone.

International Journal of Environmental Sciences
ISSN: 2229-7359
Vol. 11 No. 21s, 2025
https://theaspd.com/index.php

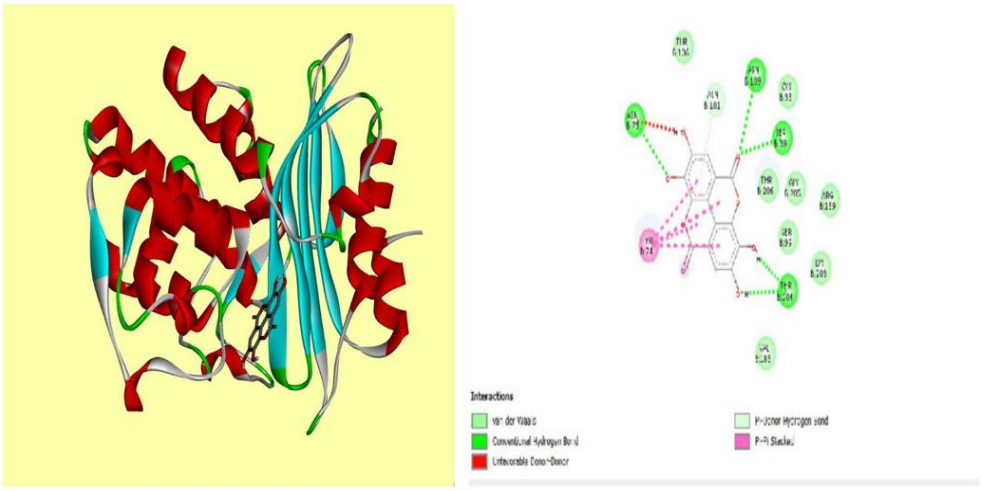


Figure 8: shows the 3D and 2D structures of the highest-affinity binding complex between AmpC β-lactamase and Ellagic Acid

The BOILED-Egg model analysis depicted in the images highlights the ADME properties of screened marine compounds, particularly focusing on gastrointestinal absorption (HIA) and blood-brain barrier permeability (BBB). Figure 8 shows that most of the compounds are positioned within the white and yellow ellipses, indicating favorable absorption and distribution characteristics. However, one molecule—Molecule 2—clearly falls outside the ideal physicochemical range, with a significantly high topological polar surface area (TPSA), suggesting poor permeability and reduced oral bioavailability. This observation implies that while the majority of candidates exhibit promising pharmacokinetic profiles, Molecule 2 may require structural optimization to enhance its drug-likeness.

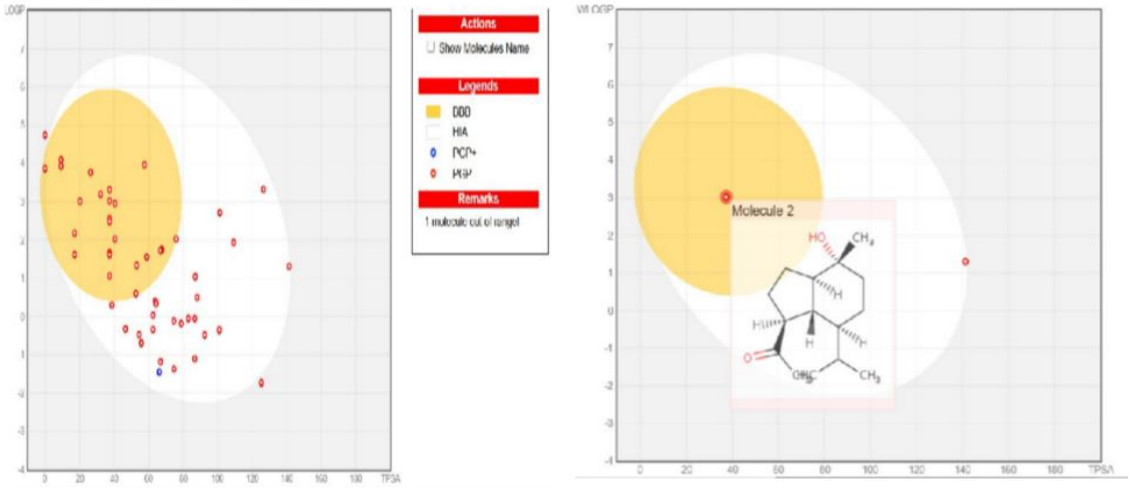


Figure 9 shows the BOILED-Egg plot predicting that most compounds exhibit favorable absorption and brain penetration, except Molecule 2, which falls outside the optimal range.

The PRED-hERG tool was used to evaluate the potential cardiac toxicity of Molecule 1 and Molecule 2. Both molecules were predicted to be non-blockers of the hERG channel, indicating a low risk of inducing cardiotoxic effects. Importantly, the predictions fall within the model's applicability domain, enhancing the reliability of the results. The consensus weighted confidence scores were 99.93% for Molecule 1 and 99.95% for Molecule 2, suggesting high prediction accuracy. Table 3 shows the cardiac toxicity prediction results obtained from the PRED-hERG tool

PRED- hERG	Consensus Weighted	Binary Prediction	Confiabilty %
Molecule1	Non- blocker	Non- blocker	99.93
Molecule2	Non- blocker	Non- blocker	99.95

Table 3 shows: The PRED-hERG tool prediction

The molecular dynamics (MD) simulation of the AmpC–Oplopanone complex over 50 ns revealed

notable stability and favorable interaction characteristics. The RMSD plot demonstrated that the ligand underwent initial conformational adjustments within the first 10 ns, followed by stabilization around ~0.4 nm, indicating that the complex reached equilibrium and maintained structural integrity throughout the simulation. RMSF analysis further confirmed the rigidity of most protein residues, with minimal fluctuations (<0.2 nm) across the structure, except for moderate flexibility near loop regions around residues ~50, ~250, and ~300. Importantly, residues in the active site showed minimal movement, suggesting strong and specific ligand binding. Additionally, hydrogen bond analysis showed consistent formation of 2–4 hydrogen bonds over time, with occasional fluctuations, underscoring the persistent and robust interactions between Oplopanone and key residues within the AmpC active site. Collectively, these dynamic properties highlight the structural stability and potential inhibitory efficacy of Oplopanone against AmpC β-lactamase

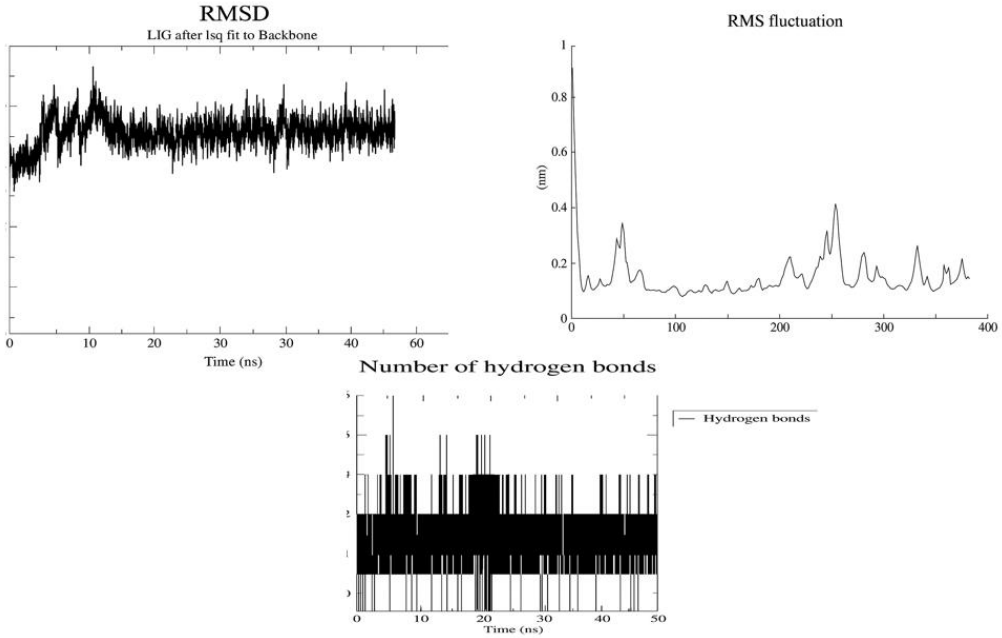


Figure 10: Molecular dynamics analysis of the AmpC–Oplopanone complex showing RMSD stabilization, limited RMSF fluctuations, and consistent hydrogen bonding, indicating structural stability and strong ligand binding.

CONCLUSION

This study focused on the clinically important AmpC β-lactamase enzyme, a key player in antibiotic resistance, and screened 60 marine-derived natural compounds to identify potential inhibitors. After evaluating toxicity, ADME properties, and cardiac safety (hERG), seven promising candidates were shortlisted. Among them, oplopanone and ellagic acid demonstrated strong binding to AmpC, with oplopanone showing the highest binding affinity (-8.13 kcal/mol), along with favorable drug-likeness and low cardiotoxic risk. Molecular interaction studies revealed stable hydrogen bonds and hydrophobic interactions with key active site residues, and molecular dynamics simulations confirmed the stability of the AmpC-oplopanone complex under physiological conditions. These findings suggest that the identified compounds, particularly oplopanone, hold strong potential for further in vitro and in vivo development as novel β-lactamase inhibitors.

REFERENCES

1. Rice, L.B. (2009). Emerging Issues in Antibiotic Resistance. Infect Control Hosp Epidemiol.
2. Mahenthiralingam, E., et al. (2005). Burkholderia cepacia complex infections in patients with cystic fibrosis. J Med Microbiol.
3. Sousa, S.A., et al. (2011). Burkholderia cepacia complex: emerging multihost pathogens equipped with a wide range of virulence factors and determinants. Int J Microbiol.
4. LiPuma, J.J. (2010). The changing microbial epidemiology in cystic fibrosis. Clin Microbiol Rev.
5. McClean, S., & Callaghan, M. (2009). Burkholderia cepacia complex: epithelial cell–pathogen confrontations and potential for therapeutic intervention. J Med Microbiol.
6. Govan, J.R., et al. (2007). Burkholderia cepacia: medical, taxonomic and ecological issues. J Med Microbiol.

7. Vandamme, P., et al. (2003). Burkholderia cepacia complex: taxonomic and epidemiological overview. Clin Microbiol Infect.
8. Holden, M.T.G., et al. (2009). Genomic plasticity of the Burkholderia cepacia complex and its implications for pathogenesis. Genome Res.
9. Cutting, G.R. (2015). Cystic fibrosis genetics: from molecular understanding to clinical application. Nat Rev Genet.
10. O'Sullivan, B.P., & Freedman, S.D. (2009). Cystic fibrosis. Lancet.
11. Jacoby, G.A. (2009). AmpC β-lactamases. Clin Microbiol Rev.
12. Bush, K. (2018). Past and present perspectives on β-lactamases. Antimicrob Agents Chemother.
13. Livermore, D.M. (1995). β-Lactamases in laboratory and clinical resistance. Clin Microbiol Rev.
14. Martínez, J.L. (2008). Antibiotics and antibiotic resistance genes in natural environments. Science.
15. Paterson, D.L., & Bonomo, R.A. (2005). Extended-spectrum β-lactamases: a clinical update. Clin Microbiol Rev.
16. Blunt, J.W., et al. (2018). Marine natural products. Nat Prod Rep.
17. V.S.Sai Durga Prasad M.S.Kumaran,P.R.Kiresee Saghana, Suseela Thankachy Hemalatha (2013). Molecular Docking Studies of Mage Inhibitors in the Treatment of Lung Cancer. Asian J Pharm Clin Res.vol 6:4,78-81
18. Molinski, T.F., et al. (2009). Drug development from marine natural products. Nat Rev Drug Discov.
19. Carroll, A.R., et al. (2001). Marine natural products. Nat Prod Rep.
20. Mayer, A.M.S., et al. (2010). Marine pharmacology in 2007–8: marine compounds with antibacterial, anticoagulant, antifungal, anti-inflammatory, antiplatelet, antiprotozoal, antituberculosis, and antiviral activities. Biochim Biophys Acta.
21. Leal, M.C., et al. (2012). Marine biodiversity and drug discovery: emerging research in the wake of the pharmacological potential of marine natural products. Biotechnol Adv.
22. Newman, D.J., & Cragg, G.M. (2020). Natural products as sources of new drugs over the nearly four decades from 01/1981 to 09/2019. J Nat Prod.
23. Butler, M.S., et al. (2014). The re-emergence of natural products for drug discovery in the genomics era. Nat Prod Rep.
24. CMNPD: Comprehensive Marine Natural Products Database. (2024).
25. Zhang, L., et al. (2011). Natural compound libraries for drug discovery. J Med Chem.
26. Lionta, E., et al. (2014). Molecular docking: introducing a "docking-based" screening strategy in drug discovery. Expert Opin Drug Discov.
27. Sliwoski, G., et al. (2014). Computational methods in drug discovery. Pharmacol Rev.
28. Trott, O., & Olson, A.J. (2010). AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. J Comput Chem.
29. Durrant, J.D., & McCammon, J.A. (2011). Molecular dynamics simulations and drug discovery. BMC Biol.
30. Ramos, J.L., et al. (2002). The expanding world of bacterial transcriptional regulators. FEMS Microbiol Rev.
31. Sass, A.M., et al. (2013). Genome-wide transcriptional responses of Burkholderia cenocepacia J2315 to polymyxin B. BMC Genomics.
32. Mahenthiralingam, E., et al. (2005). Quorum sensing control of virulence gene expression in Burkholderia cepacia complex bacteria. Curr Opin Microbiol.
33. Huber, B., et al. (2001). Genetic analysis of functions involved in the antimicrobial activity of Burkholderia cepacia H111. Arch Microbiol.
34. Venturi, V., et al. (2004). Quorum sensing in the Burkholderia cepacia complex. Res Microbiol.
35. Poole, K. (2005). Efflux-mediated antimicrobial resistance. J Antimicrob Chemother.