RESEARCH PAPER

Computational Docking of Pyocyanin for Antimicrobial Efficacy Against Bacterial and Fungal Pathogens

Vaishnavi Joshi[1], Akshada Jagdale[2], Dr. Sunita Dange[3]*, Dr. Parimita Jadhav[4], Prof. Rajkumar Deshmukh[5]

[1,2,3,4] :Department of Microbiology, Shardabai Pawar Mahila Arts, Commerce and Science college, Baramati, Pune, Maharashtra, India ,413115

[5] Department of Botany, Shardabai Pawar Mahila Arts, Commerce and Science college, Baramati, Pune, Maharashtra, India ,413115

***Corresponding author**- Dr.Sunita Dange

sunitadange@gmail.com

Abstract: The present study investigates pyocyanin, a pigment produced by *Pseudomonas aeruginosa*, for its potential as an antimicrobial agent. Using in-silico techniques like Drug likeness, ADME analysis and molecular docking using Swiss ADME and Mcule 1-click docking platform respectively, interactions were explored with key proteins from *E. coli*, *S. aureus*, *P. aeruginosa*, and *C. albicans*. Our findings aim to establish pyocyanin as a promising lead for developing antibacterial and antifungal therapies. The findings highlighted the antimicrobial potential of pyocyanin and it's prospects for diverse pharmaceutical applications, paving the way for further exploration in drug discovery and development.

Keywords: Pyocyanin, Molecular Docking, Drug Likeness, ADME, Pathogens

How to cite this article: Joshi V, Jagdale A, Dange S, Jadhav P, Deshmukh R. Computational Docking of Pyocyanin for Antimicrobial Efficacy Against Bacterial and Fungal Pathogens. Int J Drug Deliv Technol. 2026;16(48s): 133-143. DOI: 10.25258/ijddt.16.48s.14

Introduction:

Pseudomonas aeruginosa is a Gram-negative, motile, non-fermenting, aerobic rod bacterium belonging to the Pseudomonadaceae family [1]. This bacterium produces a variety of pigments as secondary metabolites, which, although not essential for its basic growth, are crucial for its pathogenicity and environmental interactions [2]. *P. aeruginosa* synthesizes at least six distinct pigments: fluorescein [3], pyoverdine [4], pyomelanin [5], aeruginosin A, aeruginosin B [6], and pyocyanin [7]. Pyocyanin, a chloroform-soluble and water-diffusible compound, is optimally produced at pH levels between 7.4 and 8.4, with production significantly decreasing below pH 6.0 or above pH 9.0 [8]. This pigment is distinguished by its vivid blue color, which exhibits pH-dependent spectral absorption properties [9,10]. Previous research has demonstrated pyocyanin's antimicrobial efficacy against *E. coli, Staphylococcus aureus, Candida albicans,* and *Pseudomonas aeruginosa* itself [11, 12]. To explore pyocyanin's potential as a therapeutic lead compound, comprehensive in-silico studies targeting various essential proteins of pathogenic organisms are warranted. Prior investigations have employed molecular docking to analyze pyocyanin's binding interactions with the AgrA protein of *S. aureus* and its inhibitory effects on the 5-lipoxygenase enzyme [13, 14]. In silico methods, encompassing molecular docking, virtual screening, and ADME/pharmacokinetic predictions, enable the examination of molecular interactions, prediction of binding affinities, and assessment of pharmacokinetic properties through computer simulations [15].

Molecular docking is a crucial technique that predicts the optimal orientation of a ligand when bound to a protein, thereby elucidating intermolecular complex structures. Essential tools for these studies include Zdock, FRED, Surflex, FLOG, EUDOC, LigandFit, DOCK, and Mcule 1-click docking [16,17]. These in-silico approaches facilitate the rapid and cost-effective exploration of drug-target interactions, guiding the design and optimization of therapeutic agents and accelerating the drug discovery process [18]. Pathogens such as *E. coli, S. aureus, C. albicans, and P. aeruginosa* are significant contributors to nosocomial infections [19]. Previous molecular docking studies have targeted proteins like β-ketoacyl-acyl carrier protein synthase I (FabB) and cell division protein FtsZ in *E. coli* for antibacterial agent development [20,21]. In *Staphylococcus aureus*, docking studies have focused on cell division protein FtsA and DNA ligase protein [22,23]. Proteins such as LasR and RhlR receptors in *Pseudomonas aeruginosa* have been targeted to inhibit its quorum sensing mechanism [24]. In *Candida albicans,* the inhibition of targets including Cst20, Cek1, and Hst7 has been examined to develop antifungal therapeutics [25]. In-silico studies thus play a pivotal role in advancing pharmaceutical innovation and enhancing patient outcomes [18]. This study undertakes an in-silico exploration of pyocyanin's interactions with various proteins from bacterial and fungal pathogens, including *E. coli, S. aureus, P. aeruginosa*, and *C. albicans*, providing insights into its broad-spectrum activity. This investigation aims to establish pyocyanin as a potential lead compound for the development of antibacterial and antifungal therapeutics.

*Author for Correspondence: sunitadange@gmail.com

Materials and Methods:

1. Drug likeness prediction:

The Swiss ADME explorer uses chemical structure and calculate on – the – fly

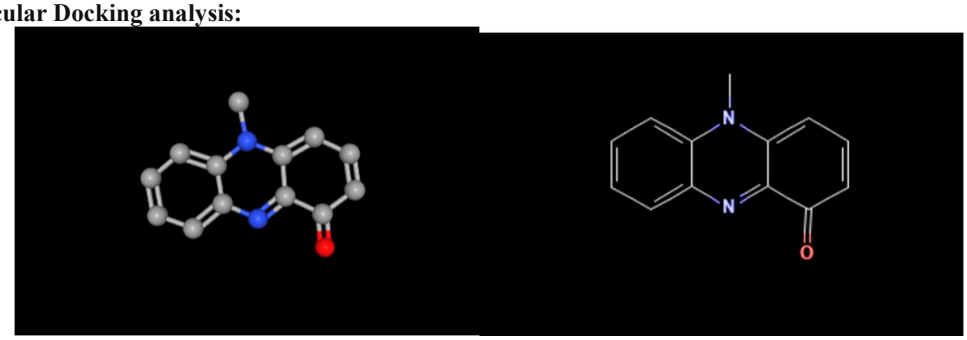
Fig. 2 D structure of Pyocyanin **Fig.** 3 D structure of pyocyanin

various drug – relevant properties whenever a structure is valid. Prediction results are valued and colour coded. Properties analysed are TPSA, *c* log P calculation, log *S* calculation, molecular weight, fragment-based drug – likeness and drug score.

2. ADME analysis:

ADME properties of a compound deal with its absorption, distribution, metabolism, excretion and toxicity through human body. ADME, which constitutes the pharmacokinetics profile of a drug molecule, is very essential in evaluating its pharmacodynamics activities. Nowadays a lot of online tools and offline software programs are available which helps us in the predicting the behaviour of the drug candidate. In this study, we have used the Swiss ADME prediction tool (SwissADME).

3. Molecular Docking analysis:



To investigate the binding studies, the molecular docking of the ligand (pyocyanin) molecule with the different target proteins of pathogenic microorganisms was done by using 1- Click Docking Mcule software platform [26].

The rise of multidrug-resistant pathogens significantly compromises the effectiveness of antibacterial treatments. Consequently, the design and development of new drugs have become increasingly crucial. In this study, we employed computer-based analyses to discover novel drug compounds, presenting promising candidates for subsequent experimental validation.

Result and Discussion:

1.1 Drug likeness prediction

Table 1. A drug likeness result of pyocyanin

Ligand	Drug likeness					Bioavailability score
	Lipinski	Ghose	Veber	Egan	Muegge	
pyocyanin	Yes; 0 violation	Yes	Yes	Yes	Yes	0.55

According to Lipinski's rule, an active drug typically meets specific criteria: it has a molecular weight (MW) of 500 or less, a LogP value of 5 or lower, no more than 10 hydrogen bond acceptors, and no more than 5 hydrogen bond donors. Veber's rules suggest that an active drug should have a total of 12 or fewer hydrogen bonds, no more than 10 rotatable bonds, and a polar surface area (PSA) of less than or equal to 140, which often indicates oral bioavailability of 20% or more. Lastly, according to Ghose's rules, an active drug typically has a Log P value between -0.4 and 5.6, a molar refractivity (MR) between 40 and 150, a molecular weight (MW) between 160 and 480, a number of atoms between 20 and 70, and a polar surface area (PSA) of less than 140 [27]. Based on the drug likeness analysis, the compound pyocyanin was found by the Lipinski's, Verber rule and Ghose's rule.

1.2 ADMET prediction

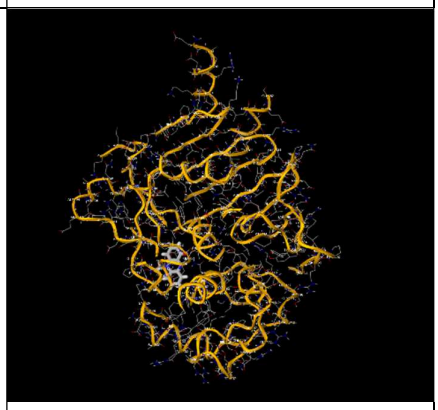
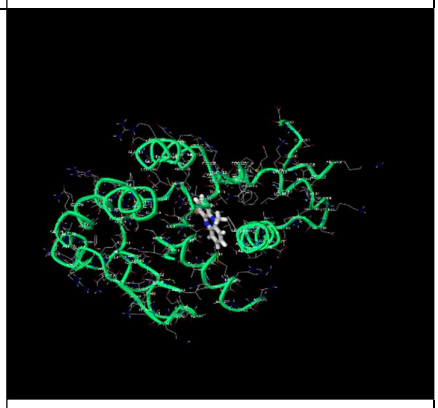
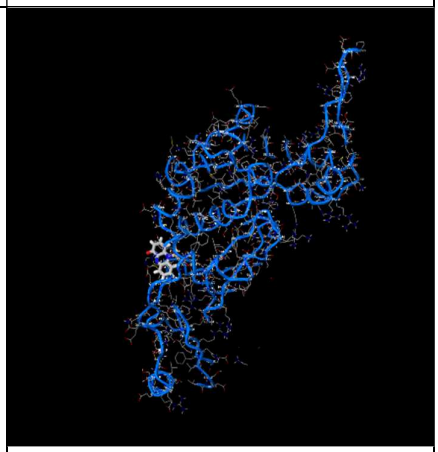
In present work predicted data of these compound were obtained by applying computer-based studies. The absorption, distribution, metabolism, elimination, and toxicity (ADMET) analysis have a big importance in drug discovery studies. In silico ADMET predictions have been designed to evaluate the pharmacokinetic and toxicity properties. In present work, human intestinal absorption, aqueous solubility levels, BBB penetration

levels, CYP inhibition, hepatotoxicity, etc. of pyocyanin were determined. ADMET analysis shows that most of the compounds are predicted good human intestinal absorption, no toxicity, and water solubility.

1.3 Molecular Docking analysis

Till date, the in detail computational study of pigment pyocyanin has not been done yet. In present research work computational analysis of pyocyanin compound against 4 common pathogens have been explored to check its potential efficacy to act as an active compound. Here, we aimed at the discovery of new potential drug compounds with computer-based analyses and presented an opportunity for further experimental analysis.

Table 2. Target organism: *Pseudomonas aeruginosa* (Total target: 57)

Target protein	PDB ID	Function of target protein	Binding Energy	Ligand-Target binding image
Beta-lactamase	2wzx	Beta-lactamase in *Pseudomonas aeruginosa* breaks down beta-lactam antibiotics, rendering them ineffective and contributing to antibiotic resistance [28].	-7.1 Kcal/mol	
Cell division protein ftsZ	1ofu	In *Pseudomonas aeruginosa*, the cell division protein FtsZ plays a fundamental role in bacterial cell division. FtsZ is a key component of the divisome, which is a multiprotein complex responsible for orchestrating cell division in bacteria [29].	-6.8 Kcal/mol	
DNA traslocase ftsK	2iuu	The DNA translocase FtsK plays a crucial role in chromosome segregation and cell division. FtsK is involved in ensuring the accurate and timely separation of replicated chromosomes during cell division [30].	-6.4 Kcal/mol	

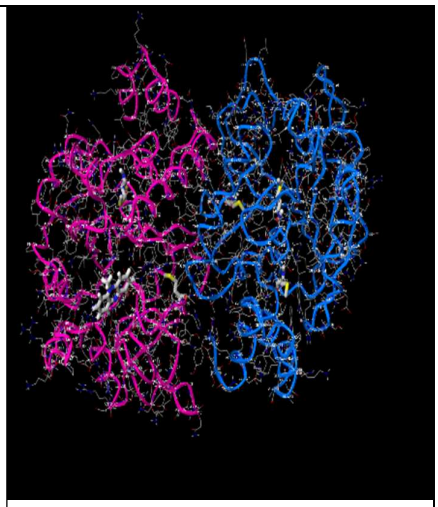
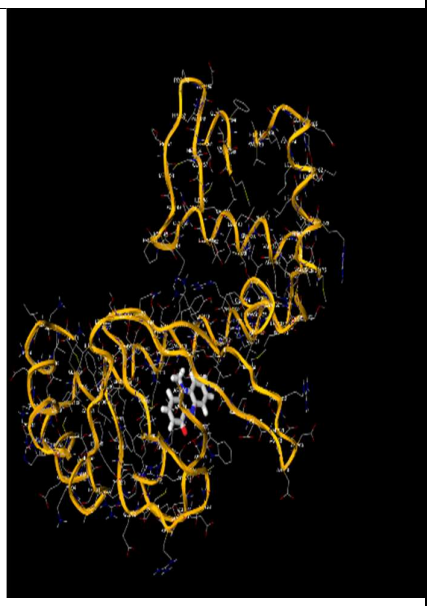
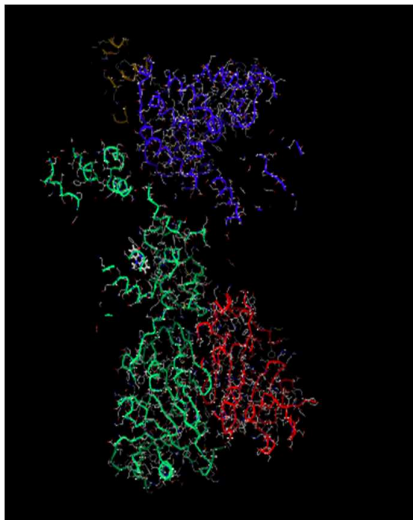
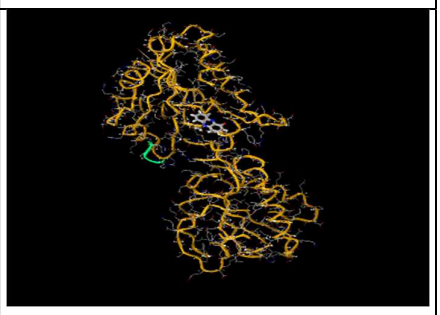
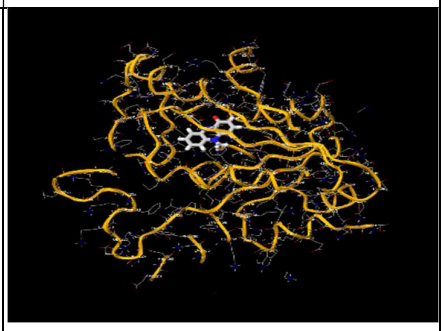
Glucose-1-phosphate thymidyly transferase	1g23	It plays a critical role in the biosynthesis of the cell surface polysaccharide called alginate. Alginate is a key component of the extracellular matrix produced by *Pseudomonas aeruginosa*, particularly in mucoid strains associated with chronic infections, such as those found in cystic fibrosis patients [31].	-7.6 Kcal/mol	
Cyclic AMP receptor like protein	2oz6	CRP homologs in *Pseudomonas aeruginosa* typically function as transcriptional regulators by binding to specific DNA sequences called CRP binding sites or cAMP receptor protein binding sites. When bound to these sites, CRP homologs can either activate or repress the transcription of nearby genes, depending on the specific regulatory context and the presence of other factors [32].	-7.3 Kcal/mol	

Table 3. Target organism: *Staphylococcus aureus* (Total target: 93)

Target protein	PDB ID	Function of target protein	Docking score	Ligand-Target binding image
yruvate carboxylase	3ho8	The primary function of pyruvate carboxylase in *Staphylococcus aureus* is to replenish the tricarboxylic acid (TCA) cycle with oxaloacetate. Oxaloacetate serves as a precursor for the synthesis of various biomolecules, including amino acids, nucleotides, and certain intermediates required for energy metabolism [33].	-6.3 Kcal/mol	
Cap 50	3ojl	Cap50 protein in *Staphylococcus aureus* plays a critical role in capsular polysaccharide biosynthesis, which is essential for bacterial virulence and pathogenesis [34].	-6.6 Kcal/mol	
Tyrosyl-tRNA synthetase	1jik	TyrRS is essential for the accurate and efficient translation of mRNA into protein. Proper functioning of TyrRS is crucial for the synthesis of functional proteins necessary for bacterial growth, metabolism, and virulence [35].	-8.8 Kcal/mol	

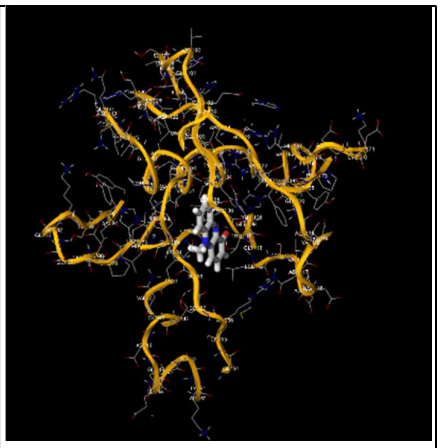
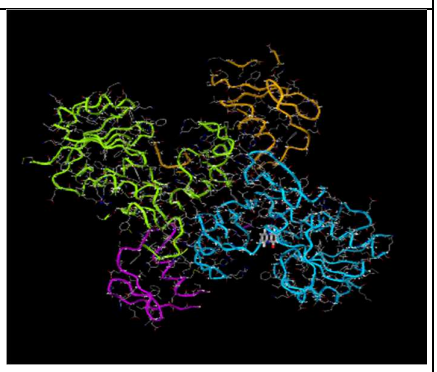
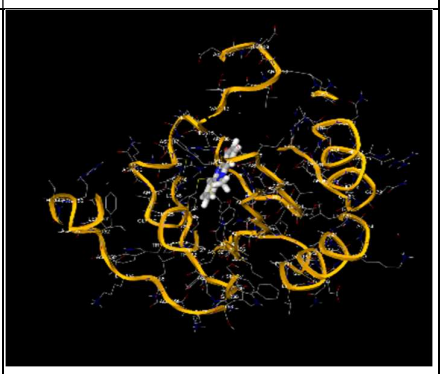
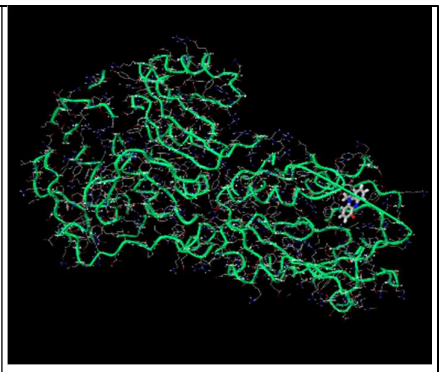
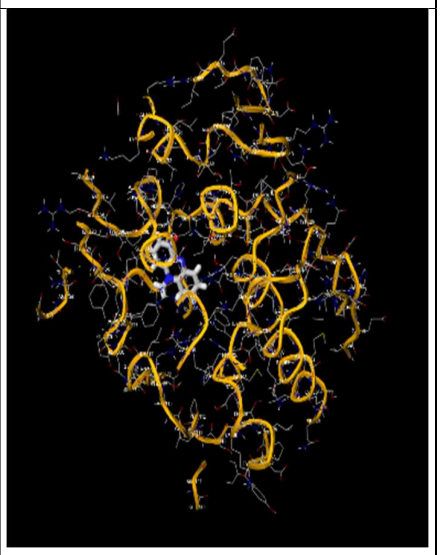
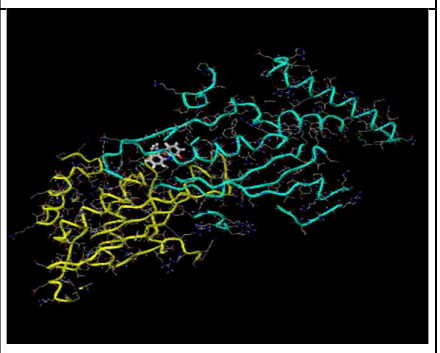
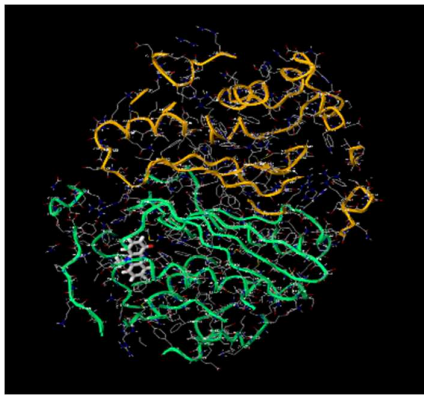
Peptide deformylase	1q1y	Peptide deformylase is essential for bacterial growth and viability, as it is involved in the processing of virtually all newly synthesized proteins. Therefore, peptide deformylase represents a potential target for the development of antibacterial agents aimed at disrupting protein synthesis and inhibiting bacterial growth [36].	-7.0 Kcal/mol	
Glyceraldehyde-3-phosphate dehydrogenase-1	3k73	GAPDH1 is a crucial enzyme in *Staphylococcus aureus* metabolism, playing a central role in glycolysis and contributing to the bacterium's ability to generate energy and sustain growth and survival in various environments [37].	-6.9 Kcal/mol	

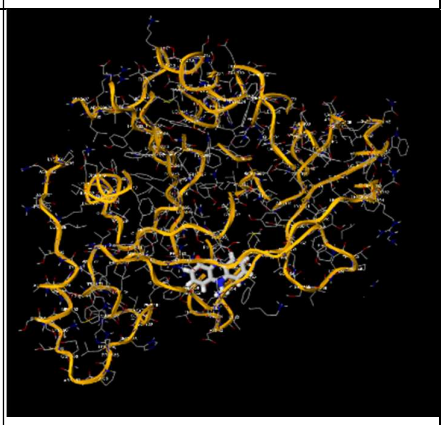
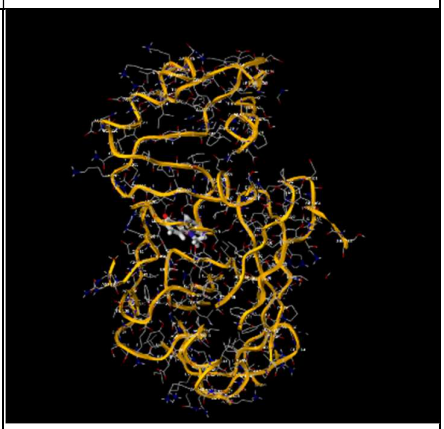
Table 4. Target organism: *Escherichia coli* (Total target: 659)

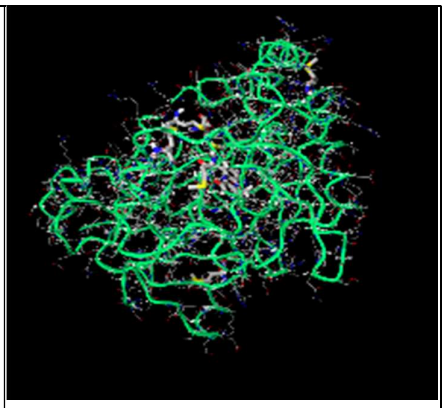
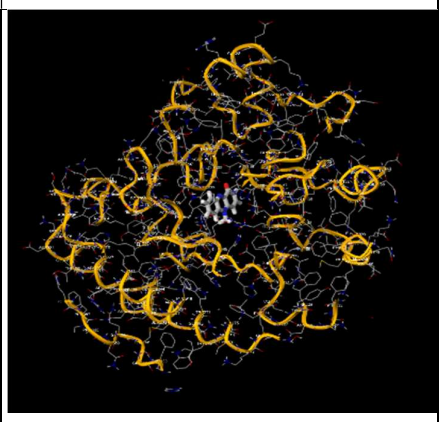
Target protein	PDB ID	Function of target protein	Docking score	Ligand-Target binding image
2-amino-4-hydroxy-6-hydroxyl methyl dehydropteridine pyrophosphate	3ipo	The function of HMDP in *E. coli* is to contribute to the synthesis of tetrahydrofolate (THF), which is essential for various metabolic processes, including DNA synthesis, amino acid metabolism, and purine and pyrimidine biosynthesis [38].	-7.3 Kcal/mol	

Chondroitin synthase	2z87	This is enzyme involved in the production of polysaccharides such as cellulose, colanic acid, and lipopolysaccharides (LPS). These polysaccharides have essential roles in the bacterial cell wall structure, biofilm formation, and interaction with the host organism or environment [39].	-7.5 Kcal/mol	
UDP-N-acetylglycosamine-1-carboxyvinyl transferase	1a69	It catalyzes the first step in the synthesis of peptidoglycan, which involves the transfer of a carboxyvinyl group from phosphoenolpyruvate (PEP) to UDP-N-acetylglucosamine (UDP-GlcNAc). This reaction results in the formation of UDP-N-acetylmuramic acid (UDP-MurNAc), a key precursor in peptidoglycan biosynthesis [40].	-7.7 Kcal/mol	
GTP cyclohydrolase 1	1a8r	GTP cyclohydrolase 1 plays a critical role in *E. coli* by initiating the pathway for the biosynthesis of BH4, which is essential for various metabolic processes and neurotransmitter synthesis [41].	-7.3 Kcal/mol	
Thymidylate synthase	2vfo	The function of thymidylate synthase in *E. coli* is to catalyze the conversion of deoxyuridine monophosphate (dUMP) to deoxythymidine monophosphate (dTMP) by transferring a methyl group from 5,10-methylenetetrahydrofolate (CH2-THF) to the uracil ring of dUMP. This methylation reaction is essential for DNA replication and repair because it generates the	-7.3 Kcal/mol	

		thymidine nucleotide required for DNA synthesis [42].		

Table 5. Target organism: *Candida albicans* (Total target: 9)

Target protein	PDB ID	Function of target protein	Docking score	Ligand-Target binding image
Glycylpeptide N-tetradecanoyl transferase	1iyk	Glycylpeptide N-tetradecanoyltransferase in *Candida albicans* adds a myristoyl group to proteins, crucial for their function in signaling, membrane anchoring, and virulence [43].	-6.9 Kcal/mol	
Candidapepsin-2	1zap	Candidapepsin2 in *Candida albicans* is a protease enzyme that plays a key role in the virulence of the fungus. It is involved in the degradation of host proteins, facilitating tissue invasion and evasion of host immune responses. Additionally, candidapepsin2 contributes to nutrient acquisition and biofilm formation, essential for the pathogenicity of *Candida albicans* [44].	-7.0 Kcal/mol	

mRNA-capping enzyme subunit alpha	1p16	mRNA capping enzyme subunit alpha in *Candida albicans* adds 5' cap to mRNA, essential for stability and translation, ensuring accurate gene expression and adaptation [45].	-8.2 Kcal/mol	
Glucan 1,3-beta-glucosidase	2pb1	Glucan 1,3-beta-glucosidase in *Candida albicans* breaks down beta-glucans, aiding in cell wall remodeling and nutrient acquisition for fungal growth [46].	-7.8 Kcal/mol	

The essential nature of the targeted proteins across the 4 pathogens underscores their pivotal roles in pathogen growth and survival. Hence, the binding of pyocyanin to these proteins could disrupt their normal functioning, potentially impeding pathogen growth or inducing pathogen death. This observation highlights pyocyanin's promise as a candidate for inhibiting pathogenic organisms via targeted protein interactions, offering valuable insights into its therapeutic potential. Understanding the molecular mechanisms underlying pyocyanin's binding to these essential proteins not only enhances our knowledge of pathogen survival but also lays the groundwork for developing innovative strategies to combat infectious diseases. Targeting essential proteins in pathogens emerges as a promising avenue for discovering and designing effective antimicrobial agents.

In present molecular docking analysis, we observed that a negative docking score corresponds to a higher binding affinity between the ligand and the target molecule. This implies a stronger interaction between the two entities, suggesting a potentially more effective binding of the ligand to the target site. Moreover, our results underscore the importance of considering docking scores alongside other parameters such as binding energy and conformational stability to comprehensively evaluate the potential efficacy of ligand-target interactions. By integrating these factors, researchers can better assess the likelihood of successful binding and thus streamline the process of drug development and optimization.

Conclusion:

In conclusion, our in-silico investigation demonstrates the potential of pyocyanin as a versatile antimicrobial agent targeting essential proteins in *E. coli, S. aureus, P. aeruginosa,* and *C. albicans*. Through molecular docking and virtual screening, we have elucidated pyocyanin's binding interactions and predicted its pharmacokinetic properties, highlighting its broad-spectrum antimicrobial activity. These findings support the further development of pyocyanin as a promising lead compound for novel antibacterial and antifungal therapeutics, underscoring its significance in pharmaceutical innovation and potential impact on improving patient outcomes.

References:

1. Wu, M., & Li, X. (2015). *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*. *In Molecular medical microbiology* (pp. 1547-1564).
2. Rani, A., & Azmi, W. (2019). An overview on biosynthesis and applications of extracellular pyocyanin pigment and its role in *Pseudomonas aeruginosa* pathogenesis. Ann Phytomed, 10.
3. Allydice-Francis, K., & Brown, P. D. (2012). Diversity of antimicrobial resistance and virulence determinants in Pseudomonas aeruginosa associated with fresh vegetables. *International journal of microbiology*, *2012*.
4. Yin, Y., Papavasiliou, G., Zaborina, O. Y., Alverdy, J. C., & Teymour, F. (2017). De novo

synthesis and functional analysis of polyphosphate-loaded poly (ethylene) glycol hydrogel nanoparticles targeting pyocyanin and pyoverdin production in *Pseudomonas aeruginosa* as a model intestinal pathogen. *Annals of biomedical engineering*, *45*, 1058-1068.

5. Ferguson, D., Cahill, O. J., & Quilty, B. (2007). Phenotypic, molecular and antibiotic profiling of nosocomial *Pseudomonas aeruginosa* strains isolated from two Irish Hospitals. *J. Med*, *1*(1).
6. Abu, E. A., Su, S., Sallans, L., Boissy, R. E., Greatens, A., Heineman, W. R., & Hassett, D. J. (2013). Cyclic voltammetric, fluorescence and biological analysis of purified aeruginosin A, a secreted red pigment of *Pseudomonas aeruginosa* PAO1. *Microbiology*, *159*(Pt_8), 1736-1747.
7. Viana, A. A. G., Martins, R. X., Ferreira, G. F., Zenaide-Neto, H., Amaral, I. P. G., & Vasconcelos, U. (2017). *Pseudomonas aeruginosa* and pyocyanin negatively act on the establishment of Enterobacteriaceae biofilm on a ceramic surface. *IJERA*, *7*, 23-30.
8. Kurachi, M. (1958). Studies on the biosynthesis of pyocyanine.(I): On the cultural condition for pyocyanine formation. *Bulletin of the Institute for Chemical Research, Kyoto University*, *36*(6), 163-173.
9. Moayedi, A., Nowroozi, J., & Sepahy, A. A. (2017). Effect of fetal and adult bovine serum on pyocyanin production in *Pseudomonas aeruginosa* isolated from clinical and soil samples. *Iranian Journal of Basic Medical Sciences*, *20*(12), 1331.
10. Muller, M. (2011). Glutathione modulates the toxicity of, but is not a biologically relevant reductant for, the *Pseudomonas aeruginosa* redox toxin pyocyanin. *Free Radical Biology and Medicine*, *50*(8), 971-977.
11. Devnath, P., Uddin, M. K., & Aha, F. (2017). Extraction, purification and charact *Pseudomonas aeruginosa*. *Extraction*, *6*(5), 1-9.
12. Hamad, M. N., Marrez, D. A., & El-Sherbieny, S. M. (2020). Toxicity evaluation and antimicrobial activity of purified pyocyanin from *Pseudomonas aeruginosa*. *Biointerface Res Appl Chem*, *10*(6), 6974-90.
13. Abdelaziz, A. A., Kamer, A. M. A., Al-Monofy, K. B., & Al-Madboly, L. A. (2023). *Pseudomonas aeruginosa's* greenish-blue pigment pyocyanin: its production and biological activities. *Microbial Cell Factories*, *22*(1), 110.
14. Santha, S. S. R., & Vishwanathan, A. S. (2022). Mechanistic insights into 5-lipoxygenase inhibition by pyocyanin: a molecular docking and molecular dynamics study. Journal of Biomolecular Structure and Dynamics, 40(20), 9752-9760.
15. Abdullahi, M., & Adeniji, S. E. (2020). In-silico Molecular Docking and ADME/Pharmacokinetic Prediction Studies of Some Novel Carboxamide Derivatives as Anti-tubercular Agents. *Chemistry Africa,* 3, 989–1000.
16. Adelusi, T. I., Oyedele, A. Q. K., Boyenle, I. D., Ogunlana, A. T., Adeyemi, R. O., Ukachi, C. D., ... & Abdul-Hammed, M. (2022). Molecular modeling in drug discovery. *Informatics in Medicine Unlocked,* 29, 100880.
17. Odhar, H. A., Hashim, A. F., Ahjel, S. W., & Humadi, S. S. (2023). Molecular docking and dynamics simulation analysis of the human FXIIa with compounds from the Mcule database. *Bioinformation*, 19(2), 160.
18. Terstappen, G. C., & Reggiani, A. (2001). In silico research in drug discovery. *Trends in pharmacological sciences*, *22*(1), 23-26.
19. Jarvis, W. R., & Martone, W. J. (1992). Predominant pathogens in hospital infections. Journal of antimicrobial chemotherapy, 29(suppl_A), 19-24.
20. Sabbagh, G., & Berakdar, N. (2015). Docking studies of flavonoid compounds as inhibitors of β-ketoacyl acyl carrier protein synthase I (Kas I) of Escherichia coli. Journal of Molecular Graphics and Modelling, 61, 214-223.
21. Domadia, P. N., Bhunia, A., Sivaraman, J., Swarup, S., & Dasgupta, D. (2008). Berberine targets assembly of Escherichia coli cell division protein FtsZ. Biochemistry, 47(10), 3225-3234.
22. Ragunathan, A., Malathi, K., Ramaiah, S., & Anbarasu, A. (2019). FtsA as a cidal target for Staphylococcus aureus: Molecular docking and dynamics studies. Journal of Cellular Biochemistry, 120(5), 7751-7758.
23. Vijayalakshmi, P., & Daisy, P. (2015). Effective interaction studies for inhibition of DNA ligase protein from Staphylococcus aureus. Journal of Receptors and Signal Transduction, 35(1), 15-25.
24. Annapoorani, A., Umamageswaran, V., Parameswari, R., Pandian, S. K., & Ravi, A. V. (2012). Computational discovery of putative quorum sensing inhibitors against LasR and RhlR receptor proteins of Pseudomonas aeruginosa. Journal of computer-aided molecular design, 26, 1067-1077.
25. Jha, A., Vimal, A., Bakht, A., & Kumar, A. (2019). Inhibitors of CPH1-MAP kinase pathway: ascertaining potential ligands as multi-target drug candidate in Candida albicans. International Journal of Peptide Research and Therapeutics, 25, 997-1010.
26. Shaukat, A., Hussain, K., & Shehzadi, N. (2023). Molecular Docking Studies of Phenolic Markers of Standardized Remedy Against Anti-Gout and Anti-Inflammatory Protein Targets. *RADS Journal of Pharmacy and Allied Health Sciences,* 1(3), 94-100.
27. Yalcin, S. (2020). Molecular docking, drug likeness, and ADMET analyses of passiflora compounds as P-glycoprotein (P-gp) inhibitor for the treatment of cancer. *Current Pharmacology Reports*, *6*(6), 429-440.
28. Hong, D. J., Bae, I. K., Jang, I. H., Jeong, S. H., Kang, H. K., & Lee, K. (2015). Epidemiology and

characteristics of metallo-β-lactamase-producing *Pseudomonas aeruginosa*. *Infection & chemotherapy*, *47*(2), 81.

29. Long, Y., Fu, W., Li, S., Ren, H., Li, M., Liu, C., ... & Wu, W. (2019). Identification of novel genes that promote persister formation by repressing transcription and cell division in *Pseudomonas aeruginosa. Journal of Antimicrobial Chemotherapy*, *74*(9), 2575-2587.
30. Crozat, E., & Grainge, I. (2010). FtsK DNA translocase: the fast motor that knows where it's going. *Chembiochem*, *11*(16), 2232-2243.
31. Alphey, M. S., Pirrie, L., Torrie, L. S., Boulkeroua, W. A., Gardiner, M., Sarkar, A., ... & Naismith, J. H. (2013). Allosteric competitive inhibitors of the glucose-1-phosphate thymidylyltransferase (RmlA) from *Pseudomonas aeruginosa*. *ACS chemical biology*, *8*(2), 387-396.
32. Endoh, T., & Engel, J. N. (2009). CbpA: a polarly localized novel cyclic AMP-binding protein in *Pseudomonas aeruginosa*. *Journal of bacteriology*, *191*(23), 7193-7205.
33. Xiang, S., & Tong, L. (2008). Crystal structures of human and *Staphylococcus aureus* pyruvate carboxylase and molecular insights into the carboxyltransfer reaction. *Nature structural & molecular biology*, *15*(3), 295-302.
34. Tokumitsu, H., Mizutani, A., Minami, H., Kobayashi, R., & Hidaka, H. (1992). A calcyclin-associated protein is a newly identified member of the Ca2+/phospholipid-binding proteins, annexin family. *Journal of Biological Chemistry*, *267*(13), 8919-8924.
35. Qiu, X., Janson, C. A., Smith, W. W., Green, S. M., McDevitt, P., Johanson, K., ... & Jarvest, R. L. (2001). Crystal structure of *Staphylococcus aureus* tyrosyl-tRNA synthetase in complex with a class of potent and specific inhibitors. *Protein Science*, *10*(10), 2008-2016.
36. Margolis, P. S., Hackbarth, C. J., Young, D. C., Wang, W., Chen, D., Yuan, Z., ... & Trias, J. (2000). Peptide deformylase in *Staphylococcus aureus*: resistance to inhibition is mediated by mutations in the formyltransferase gene. *Antimicrobial agents and chemotherapy*, *44*(7), 1825-1831.
37. Bermingham, A., Bottomley, J. R., Primrose, W. U., & Derrick, J. P. (2000). Equilibrium and Kinetic Studies of Substrate Binding to 6-Hydroxymethyl-7, 8-dihydropterin Pyrophosphokinase from *Escherichia coli*. *Journal of Biological Chemistry*, *275*(24), 17962-17967.
38. Badri, A., Williams, A., Awofiranye, A., Datta, P., Xia, K., He, W., ... & Koffas, M. A. (2021). Complete biosynthesis of a sulfated chondroitin in *Escherichia coli*. *Nature Communications*, *12*(1), 1389.
39. Zhu, J. Y., Yang, Y., Han, H., Betzi, S., Olesen, S. H., Marsilio, F., & Schönbrunn, E. (2012). Functional consequence of covalent reaction of phosphoenolpyruvate with UDP-N-acetylglucosamine 1-carboxyvinyltransferase (MurA). *Journal of Biological Chemistry*, *287*(16), 12657-12667.
40. Sharma, R., Lambu, M. R., Jamwal, U., Rani, C., Chib, R., Wazir, P., ... & Khan, I. A. (2016). Escherichia coli N-acetylglucosamine-1-phosphate-uridyltransferase/glucosamine-1-phosphate-acetyltransferase (GlmU) inhibitory activity of terreic acid isolated from *Aspergillus terreus*. *Journal of biomolecular screening*, *21*(4), 342-353.
41. Schoedon, G., Redweik, U., Frank, G., Cotton, R. G., & Blau, N. (1992). Allosteric characteristics of GTP cyclohydrolase I from *Escherichia coli. European journal of biochemistry*, *210*(2), 561-568.
42. Belfort, M., Maley, G., Pedersen-Lane, J., & Maley, F. (1983). Primary structure of the *Escherichia coli* thyA gene and its thymidylate synthase product. *Proceedings of the National Academy of Sciences*, *80*(16), 4914-4918.
43. Kim, Y., Islam, N., Moss, B. J., Nandakumar, M. P., & Marten, M. R. (2011). Autophagy induced by rapamycin and carbon-starvation have distinct proteome profiles in Aspergillus nidulans. *Biotechnology and bioengineering*, *108*(11), 2705-2715.
44. Manjoor, A. S., Padmanabha Reddy, Y., & Chandrasekhar, K. B. Journal of Pharmaceutical and Scientific Innovation.
45. Schwer, B., Lehman, K., Saha, N., & Shuman, S. (2001). Characterization of the mRNA capping apparatus of Candida albicans. *Journal of Biological Chemistry*, *276*(3), 1857-1864.
46. Jiang, C., Song, J., Cong, H., Zhang, J., & Yang, Q. (2017). Expression and characterization of a novel antifungal exo-β-1, 3-glucanase from Chaetomium cupreum. *Applied Biochemistry and Biotechnology*, *182*, 261-275.