Potential of Cyanobacterial Compounds as Antibacterial Inhibitors Against the Protein Marker 1T2P and 2W9S in Staphylococcus Aureus: An in Silico Approach

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Abstract**—***In order to combat the growing antibiotic resistance in Staphylococcus aureus, new antimicrobial agents have to be found. An extensive variety of bioactive secondary metabolites with possible antibacterial activity are known to be produced by cyanobacteria. Through the use of molecular docking techniques, this work sought to assess the binding affinity of specific cyanobacterial compounds against important S. aureus protein targets (1T2P and 2W9S) and compare their efficacy with widely used synthetic antibiotics like Penicillin G and Penicillin V.Calothrixin and Cyanobacterin, are cyanobacterial compounds that were molecularly docked against the S. aureus protein targets 1T2P and 2W9S utilizing AutoDock, Cygwin, and Biovia Discovery Studio Client.. These natural compounds binding affinities were contrasted with those of Penicillin G and Penicillin V. Penicillin G (-5.24 kcal/mol) and Penicillin V (-6.25 kcal/mol) showed weaker binding affinities than Calothrixin (-7.82 kcal/mol) and Cyanobacterin (-7.45 kcal/mol) for the 1T2P target. In comparison to Penicillin G (-8.59 kcal/mol) and Penicillin V (-8.26 kcal/mol), Cyanobacterin (-8.64 kcal/mol) also demonstrated higher binding affinities for the 2W9S target. According to the docking data, cyanobacterial bioactive compounds show interesting binding interactions with important S. aureus proteins, namely Cyanobacterin, and Calothrixin. These results suggest that they may have antibacterial properties. To confirm the antibacterial effectiveness of these cyanobacterial extracts and investigate their mode of action, more in vitro research is necessary.*

Keywords—Cyanobacteria; Bioactive compounds; Antimicrobial activity; Staphylococcus aureus; Molecular docking

I. INTRODUCTION

The improvement of human life quality and the reduction of human diseases are greatly dependent on therapeutics research. Pathogenic organisms are the cause of a huge number of diseases. Microorganisms that are detrimental to the human body are known as pathogens. Pathogens include bacteria, viruses, fungi, prion, protozoa, and viruses, among others. Even though the human body can fight off possible infections, multidrug-resistant microbes cause a significant increase in microbial illnesses in living things (Laxminarayan, R. et al, 2016; Sarmah, P. et al, 2018). One of the major issues facing the contemporary healthcare system is the proliferation of multidrug-resistant (MDR) bacteria, which has been caused by the overuse of antibiotics in recent decades. Due to increased resistance, effective treatment becomes more and more complicated with the available, common antibiotics. Therefore, new treatments have to be brought onto the market, discovering new antibacterial substances, a key factor in the fight against the widespread of MDR bacteria (Bharadwaj, A et al, 2022).

Even though the pharmaceutical industry has made great advances in synthetic chemistry regarding the development of new, bioactive substances against a wide variety of pathogens, this technology still has its limitations: many natural products have highly complex structures that are too complicated and too expensive to produce on an industrial scale. In addition, natural sources offer a high diversity of substances, from which only a small part has been discovered so far. Therefore, the screening and isolation of bioactive compounds as new therapeutic substances remains an important aspect of research (Ahmad & Aqil, 2020; Lahlou et al., 2013). In terms of bioactive compounds, cyanobacteria are a promising source of new, undiscovered substances. Cyanobacteria are photoautotrophic microorganisms that occur in many different environments, such as freshwater, seawater, and fields, leading to a high chemodiversity of secondary metabolites (Garcia-Pichel et al., 2003; Swain et al., 2017).
They produce a wide variety of bioactive compounds like proteins, lipids, polysaccharides, fatty acids, alkaloids, and polyketides, which are considered to have a variety of properties like antifungal, antiviral, antibacterial, algicidal, and anti-inflammatory activity (Demay et al., 2019).

A number of studies addressing isolated chemicals have been published in recent years due to the promising potential of cyanobacteria as manufacturers of novel bioactive compounds (Levasseur&Pozzobon, 2020; Swain et al., 2017; Xue et al., 2018). This study notably focuses on screening that results in the identification of antibacterial chemicals from cyanobacteria. Among these multidrug-resistant microbes is S. aureus. It is a spherical bacterium belonging to the Firmicutes family (Ogston A et al., 1984). This species has strong, amorphous cell walls. Peptidoglycan makes up 50% of the cell wall's bulk and is its main constituent. Teichoic acids make up the remaining 40% of the mass of the cell wall, with exoproteins making up the remaining 10%, surface proteins, and peptidoglycan hydrolases.

Naturally, this bacterium is found in the nasopharynx of the human body and on the skin. S. aureus can cause infections of the nose, skin, vagina, urethra, and gastrointestinal tract (Kluytmans J, et al.,1997; Cole AM, et al., 2001;Llarrull LI et al.,2009) . They are non-sporing, non-motile, and few strains are encapsulated. Nearly 50% of the human population are carriers of S. aureus. Generally, beta (β)-lactum antibiotics such as Penicillin, Carbapenems, Monobactams, and Cephalosporins are used to heal infections caused by Staphylococcus aureus. These drugs bind with proteins in the bacteria and thereby inhibit the bacterial cell wall synthesis (LlarrullLI et al., 2009). But several studies showed that beta (β)-lactum antibiotics have a low binding affinity with Penicillin-binding proteins and are a major cause for less antibiotic activity (Sainsbury S., et al., 2011; Yoshida H et al, 2012). *In silico* is an expression which means a "performed on the computer or via computer simulation. In silico or computational techniques play an important role in investigating multi-target directed ligands (MTDLs) with cost and time benefits. Several *In silico* techniques have been evolved, which can be divided into two major application areas, i.e., ligand-based drug design and structure- based drug design. Ligand-based drug design (LBDD) techniques like quantitative structure- activity relationship (QSAR) rely on knowledge of diverse ligands that interact with the biological targets of interest. On the other hand, the structure-based drug design (SBDD) techniques like molecular docking and molecular dynamics, etc., rely on three-dimensional (3D) knowledge of the target protein structure and its binding site to investigate the key ligand-protein interactions as well as to interpret the binding energy. Structure-based and ligand-based drug design techniques together become a powerful tool for screening of large chemical libraries to identify potential ligands against panels of biological targets (Manoharan and Ghoshal, 2018). Thus, virtual screening employing both LBDD and SBDD techniques is highly effective and can play a vital role to investigate novel compounds as multi-target directed ligands (Castro, L. H., &Sant'Anna, C. M. R. 2022).The present study aimed to identify the novel naturally-derived antibacterial compounds from various cyanobacterial sources to understand the molecular interaction between associated markers of staphylococcus aureus with bioactive compounds.

Staphylococcus aureus **Sortase-A**

Sortases have a great role in the cell wall envelope assembly and bacterial pathogenicity. The natural habitat of Staphylococcus aureus in humans is the skin and nasopharynx. It can cause a wide variety of infections involving skin and soft tissues, endovascular sites and internal organs. *S. aureus* continues to be an important pathogen in the community and in hospitals, causing high morbidity and mortality. The organism can be disseminated from a superficial site via the bloodstream to internal organs where it can set up a metastatic focus of infection. Major sites of infection in hospital patients are surgical wounds and indwelling medical devices. In the latter the bacteria may colonize the implanted device causing local damage or it can disseminate. In addition, food poisoning can occur after ingestion of food contaminated with enterotoxins. *S. aureus* also causes the economically important ruminant mastitis. A major part of this chapter is devoted to reviewing

the structure and function of potential virulence factors and the evidence for their involvement in pathogenicity. (Pal, M. et al, 2021). *Staphylococcus aureus* is a bacterium commonly found on the skin and in the nasal passages of humans. While it is often harmless, it can cause a range of infections, from minor skin infections to more serious conditions like pneumonia, endocarditis, and sepsis. *Staphylococcus Aureus* is notorious for its ability to develop antibiotic resistance, posing a significant challenge in healthcare settings (Fayisa, W. O., &Tuli, N. F., 2023). Sortase A is an enzyme produced by Staphylococcus aureus and other gram-positive bacteria. It plays a crucial role in the anchoring of surface proteins to the bacterial cell wall. This process is essential for the virulence of *Staphylococcus Aureus*, as it allows the bacterium to adhere to host tissues and evade the host immune response. Research based on the 1T2P structure may focus on elucidating the catalytic mechanism of sortaseA, identifying potential inhibitors that disrupt its activity, and exploring the role of sortase-mediated protein anchoring in *Staphylococcus aureus pathogenesis*(Kudryavtsev, K. V. et al, 2021). Such studies contribute to our understanding of bacterial virulence mechanisms and facilitate the development of new therapeutic approaches to combat *Staphylococcus Aureus* infections.

Dihydrofolate reductase (DHFR)

Dihydrofolate reductase (DHFR) is an enzyme that catalyses the formation of tetrahydrofolate (THF) by the reduction of Dihydrofolate (DHF) in the presence of nicotinamide adenine dinucleotide phosphate (NADPH). Also, it has a great role in the synthesis of thymidylate, purines, methionine, and some other important metabolites (Sehrawat, R. et al, 2024). These enzymes are required for cell proliferation. Thus, inhibition of dihydrofolate reductase will results in the destruction of the intracellular tetrahydrofolate pool thereby preventing biosynthesis of RNA, DNA, thymidine, and protein. Due to the wide range of cellular functions, they are targets for anticancer and antimicrobial agents *Staphylococcus Aureus* is a Gram-positive bacterium known for its ability to cause a wide range of infections in humans, ranging from mild skin infections to severe conditions such as pneumonia, endocarditis, and sepsis. It is a significant public health concern due to its ability to develop antibiotic resistance, making treatment challenging. The extracellular adherence protein (Eap) is a multifunctional virulence factor produced by Staphylococcus Aureus. It plays a crucial role in the pathogenesis of Staphylococcus Aureus infections by promoting bacterial adhesion to host cells, modulating the host immune response, and facilitating biofilm formation. The crystal structure of Eap, represented by PDB entry 2W9S, provides valuable insights into the molecular architecture and function of this important virulence factor. Understanding the structure of Eap and its interactions with host molecules can help elucidate its role in *Staphylococcus Aureus* pathogenesis and may inform the development of new therapeutic strategies to combat Staphylococcus Aureus infections (Taj, Z., & Chattopadhyay I., .2024). Research based on the 2W9S structure may focus on elucidating the mechanisms by which Eap promotes bacterial adhesion and modulates host immune responses. Additionally, studies may explore the potential of targeting Eap as a novel therapeutic approach to disrupt *Staphylococcus Aureus* infections, either by inhibiting its activity or by interfering with its interactions with host cells and extracellular matrix components. Overall, the crystal structure of Eap provides a valuable foundation for further research aimed at understanding the molecular basis of Staphylococcus Aureus pathogenesis and developing new strategies for the prevention and treatment of *Staphylococcus Aureus* infections (Linz, M. S. ET AL., (2023).

II. Material and Methods

*InSilico*Approach

"*In silico*" refers to a process that is "conducted on the computational approach or via computer simulation." Investigating multi-target directed ligands (MTDLs) with time and cost advantages requires the use of in silico or computational approaches. Two main application areas of *in silico* techniques ligand-based drug design and structure-based drug design are the result of their evolution. Understanding the many ligands that interact with the biological targets of interest is essential for ligand-based drug design (LBDD) methodologies such as quantitative structure-activity relationship (QSAR). However, the structure-based drug design (SBDD) methods, such as molecular docking and molecular dynamics, depend on three-dimensional (3D) information on the structure of the target protein and its binding site in order to explore the important ligand-protein interactions as well as to interpret the binding energy. Structure-based and ligand-based drug design techniques together become a powerful tool for screening of large chemical libraries to identify potential ligands against panels of biological targets (Manoharan and Ghoshal, 2018). Thus, virtual screening employing both LBDD and SBDD techniques is highly effective and can play a vital role to investigate novel compounds as multi-target directed ligands (Passeri et al., 2018). A basic assumption of any *In silico* study is the correctness of the input data extracted from the literature or databases. However, one should always be concerned about the possibility of having an inferior qualityof both the chemical

(i.e., chemical structures) and biological (like experimental activity) data, especially when the data are extracted from an online database. Thus, curation of both chemical and biological data is critical for the success of any *in silico* study, especially when we are dealing with 'big data'(Fourches, Muratov, &Tropsha, 2010; Fourches, Muratov, &Tropsha, 2016). In simple terms, the chemical curation involves both the identification and correction of the structural errors for a set of chemicals, while the biological curation is required to verify the accuracy, consistency, and reproducibility of the reported experimental data.

DOCKING METHODOLOGY

PREPARATION OF TARGET PROTEIN STRUCTURE AND LIGANDS

Protein Data Bank (PDB) is a repository of 3-D structural data of macromolecules. The 3D crystal structure of 1T2P and 2W9S was retrieved from RCSB PDB. All the water molecules and crystallographic substructures from the target protein were eliminated and the necessary hydrogen atoms were added along with Gasteiger- Marsili charges. The minimization process was undergone and protein protocol was automatically generated and the final structure was visualized in Discovery Studio Visualizer 4.0. A total of 50 bioactive compounds from cyanobacterial strains and 2 synthetic drugs were selected by the literature survey for docking studies and the bioactive compound structures were downloaded from PubChem (Ahmad, S.et al., 2024). *Staphylococcus Aureus*(PDB ID: 1T2P)While not specifically designed to overcome MDR, their clinical approval established a crucial precedent and paved the way for more sophisticated systems.

Staphylococcus Aureus(PDB ID: 1T2P)

Sortases are extracellular transpeptidases found in gram-positive bacteria. As the name suggests, sortases are enzymes that separate proteins into the Gram-positive bacteriaL cell wall compartment. Sortases are essential for the formation of the cell wall membrane and the pathogenicity of bacteria. PDB ID 1T2P, which shows the 3D crystal structure of *Staphylococcus Aureus*sortaseA in association with a peptide substrate, offers important information about the molecular interactions that underlie sortase A enzymatic activity. New antibiotic approaches that target *Staphylococcus Aureus* infections can be developed with the help of an understanding of sortase A structure and interactions with substrates (Nitulescu, G.. ET AL, 2017).

Dihydrofolate reductase (DHFR) (PDB ID: 2W9S)

An enzyme called dihydrofolate reductase (DHFR) catalyzes the reduction of dihydrofolate (DHF) in the presence of nicotinamide adenine dinucleotide phosphate (NADPH) to produce tetrahydrofolate (THF). Furthermore,

Fig II: Dihydrofolate reductase (DHFR) (PDB ID: 2W9S)

it plays a significant part in the production of methionine, purines, thymidylate, and a few other critical metabolites. Cell growth requires these enzymes. As a result, blocking dihydrofolate reductase will deplete the intracellular tetrahydrofolate pool, which will stop RNA, DNA, thymidine, and protein synthesis from occurring. Their diverse spectrum of biological functions makes them candidates for antibacterial and anticancer drugs (Polshakov, V. I., 2001)

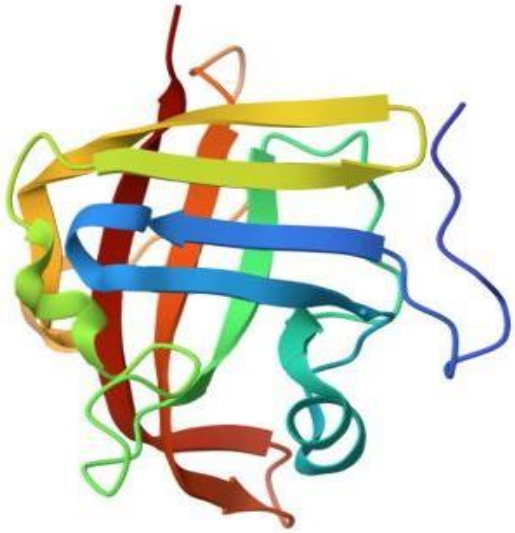


Fig I:*StaphylococcusAureus*(PDBID:1T2P)

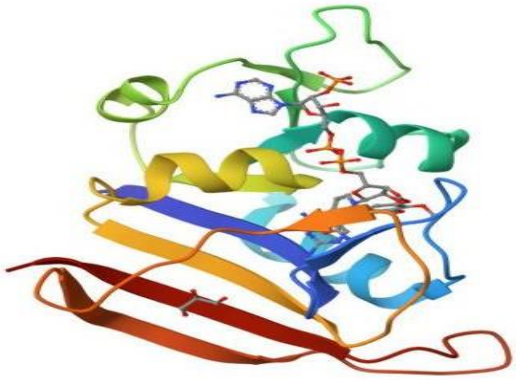


Table I. ADMET Profile of cyanobacterial compounds and control compounds

S.No	Compound	Skin Permeability	Buffer Solubility	PPB	BBB	CaCO2	MDCK	HIA	Ames_Test	Algae_at	Carcino_mouse	Carcino_Rat
1	Cyanobacterin	-2.73105	1.08503	94.063554	0.109392	30.7663	0.263006	96.397566	non-mutagen	0.0073038 1	positive	negative
2	Nostolactone4	-2.97384	1.29961	100.000000	2.25622	20.7534	0.043885 9	90.240239	non-mutagen	0.0030 4167	negative	negative
3	UlongamideA	-2.69646	89.9207	88.449392	0.0584 408	38.1904	0.043415 5*	95.046316	non-mutagen	0.0013 359	negative	negative
4	Kalkitoxin	-0.901813	209.714	98.178461	0.897038	49.691	0.277258	97.662934	mutagen	0.0046 9583	positive	negative
5	Anatoxin A	-3.13135	2595.2	22.306845	0.612474	30.5172	21.5161	95.398255	mutagen	0.100 819	positive	negative
6	LyngbyatoxinA	-2.48401	0.201294	88.537281	7.74389	39.1752	69.8067	92.069048	non-mutagen	0.0034346 8	negative	negative
7	Kanamienamide	-2.35562	56.1889	90.214284	0.061603	52.0874	0.051340 4	98.842114	mutagen	0.0065 5343	positive	negative
8	Axisonitrile3	-0.833635**	2139.08**	100.000000**	10.0928	23.6358	82.8608**	100.0000	non-mutagen	0.0175 511**	positive	negative
9	Neosaxitoxin	-5.16417	179.126	27.741376	0.0396 268	18.7571	0.565055	1.379886	mutagen	0.1175 35	negative	negative
10	Cylindrospermopsin	-4.27657	102702**	25.574380	0.0422 016	12.9846	0.58379	23.393858	mutagen	0.1082 88	negative	negative
11	27-DeoxylyngbyabellinA	-4.02443*	13.3947	89.777114	0.192504	33.4073	0.043415 5*	95.427245	non-mutagen	7.3219 4e005	negative	negative
12	MuscorideA	-2.82074	2015.67	81.715997	0.0160 974	51.8626	0.044078 2	97.594733	mutagen	0.0048 6011	negative	negative
13	Nostocarboline	-4.52453	143.851	17.913312	5.18456	16.7822	251.778	97.910575	mutagen	0.1151 55	positive	negative
14	Homoanatoxina	-2.64771	1121.24	68.857661	1.08338	38.3862	55.913	95.551189	mutagen	0.0514 568	negative	negative
15	LyngbyatoxinA	-2.48401	0.201294	88.537281	7.74389	39.1752	69.8067	92.069048	non-mutagen	0.0034 3468	negative	negative
16	AmbigolA	-1.90659	17.9086	100.000000	13.4989	46.5271	25.512*	95.546983	mutagen	0.0002 02861	negative	negative
17	Noscomin	-1.04916	139.603	100.000000	3.07873	20.8991	0.078209 9	94.233418	non-mutagen	0.0023 585	negative	negative
18	Anhydroaplysiatoxin	-1.99533	1.42522	90.491824	0.395191	40.446	0.043679 7	95.832614	non-mutagen	0.0011 2684	negative	negative

19	Banyascyclamide B	-5.10649	201.967	57.264065	0.223618	6.24238	0.0599412	45.208037	non-mutagen	0.0156897	negative	negative
20	Herbamide B	-2.44063	31.1525	100.000000	1.5733	41.6597	0.0544121	96.771275	mutagen	0.00563332	positive	negative
21	Scytonemin	-2.28657*	0.00394521	100.000000	0.224864	21.9848	0.0463535	97.117607	mutagen	0.00104286	negative	negative
22	Calothrixin B	-4.16192	4.90516	88.378420	1.60763	20.7093	132.056	94.769029	mutagen	0.0565641	negative	negative
23	Fischerellin A	-1.86648	5803.83	1.#IND00	1.49973	22.2815	204.401	100.000000	non-mutagen	0.002694	positive	negative
24	Lyngbic acid	-0.820175	6790.5	97.285692	0.309534	36.5579	56.7625	97.301378	mutagen	0.0122626	negative	negative
25	Abietane	-0.950175	93.0812	100.000000	17.4862	22.2014*	67.1758*	100.000000	non-mutagen	0.0046307	negative	negative
26	Hapalosin	-1.9145	38.5759	92.631712	0.284506	36.0072	0.10327	97.500022	non-mutagen	0.00326983	negative	negative
27	CuracinA	-1.11249	26.4918	99.687473	5.7525	35.2846	22.0034	97.546286	mutagen	0.0047236	positive	negative
28	PenicillinV	-4.60455	7676.97	86.308339	0.0817593	14.4602	0.153893	89.665389	mutagen	0.0570241	negative	negative
29	Cefixime	-4.81237	7632.27	52.983428	0.0399635	17.9882	0.10071	20.926075	non-mutagen	0.0510886	positive	negative
30	PenicillinG	-4.54685	14898.5	82.585730	0.0932861	11.859	3.13642	92.750136	mutagen	0.067168	negative	negative

III. Result

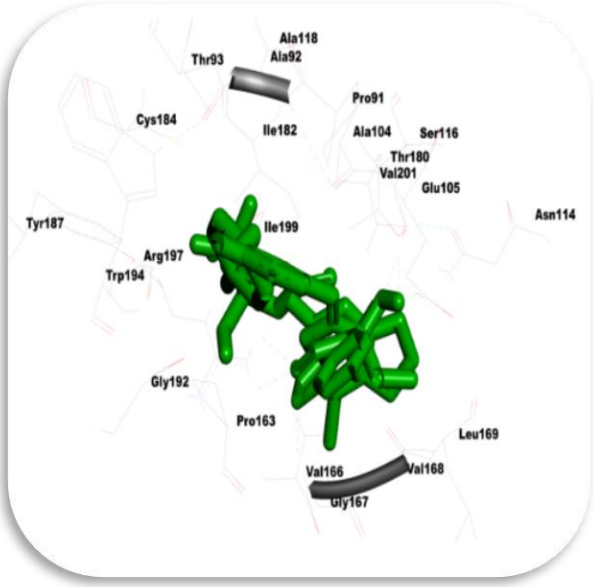
Table II. Molecular interaction result of Synthetic compounds along with Cyanobacterial bioactive compounds against 1T2P.

S.NO.	Ligand	BindingEnergy kcal/mol	Inhibition constant (Ki)
1.	Penicillin V	-6.25	6.1µM
2.	Penicillin G	-5.24	143.37µM
3.	CalothrixinB	-7.82	1.86µM
4.	Cyanobacterin	-7.45	3.45µM

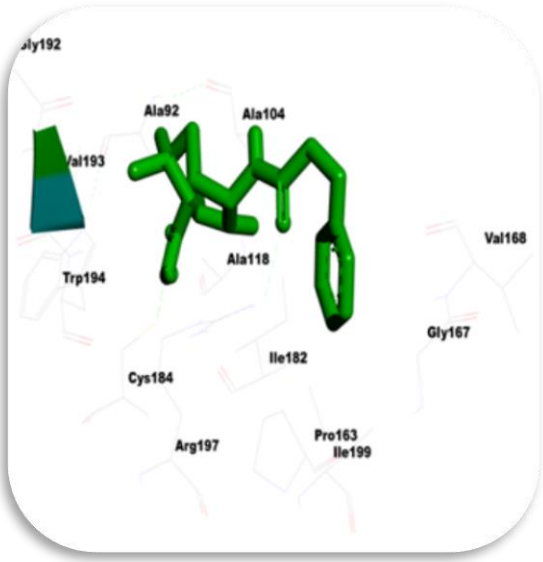
Table III: Molecular interaction result with interacted amino acid residues of Synthetic compounds along with Cyanobacterial bioactive compounds against 1T2P

S.No	Compound Name	Binding Energy kcal/mol	AminoAcid	Inhibition constant (Ki)
1.	PenicillinG	-5.24	Gly192, Val193, Ala92, Ala104, Ala118, Trp194, Arg197, Ile182, Cys184,Pro163,Ile199,Gly167, Val168.	143.37µM
2.	PenicillinV	-6.25	Glu108,Gln64,Ser109,Ile65,Pro63, Leu110, Lys62, Asp111.	26.1µM
3.	Calothrixin B	-7.82	Lys175, Gln172, Val168, Gly167, Val166, Gln178, Ile158, Val201, Ile199, Thr180, Ile182, Ser116, Arg197,Ala104,Glu105,Cys184.	1.86µM
4.	Cyanobacterin	-7.45	Tyr187,Cys184,Thr93,Ala118, Ala92, Ile182, Ala104, Pro91, Ser116,Thr180,Val201,Glu105,Trp194, Arg197, Ile199, Gly192,Pro163,Val166,Gly167,Val168, Leu169,Asn114.	3.45µM

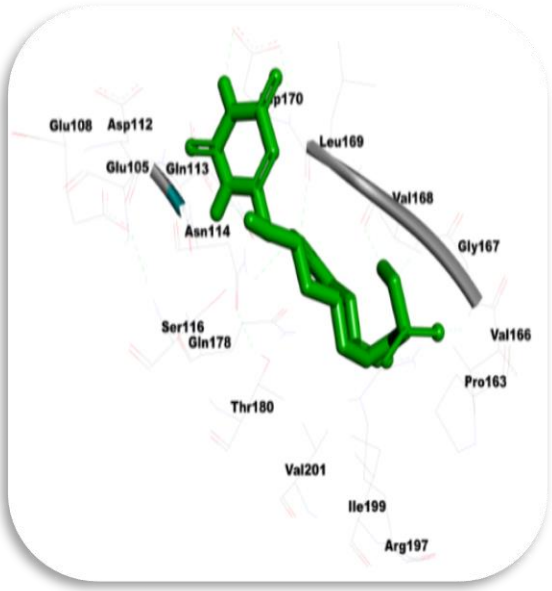
Complex Calothrixin B with 1T2P



Complex of Cyanobacterin with 1T2P



Complex of Penicillin G with 1T2P



Complex of Penicillin V with 1T2P

Fig III: Complexes of synthetic and cyanobacterial ligand with 1T2P

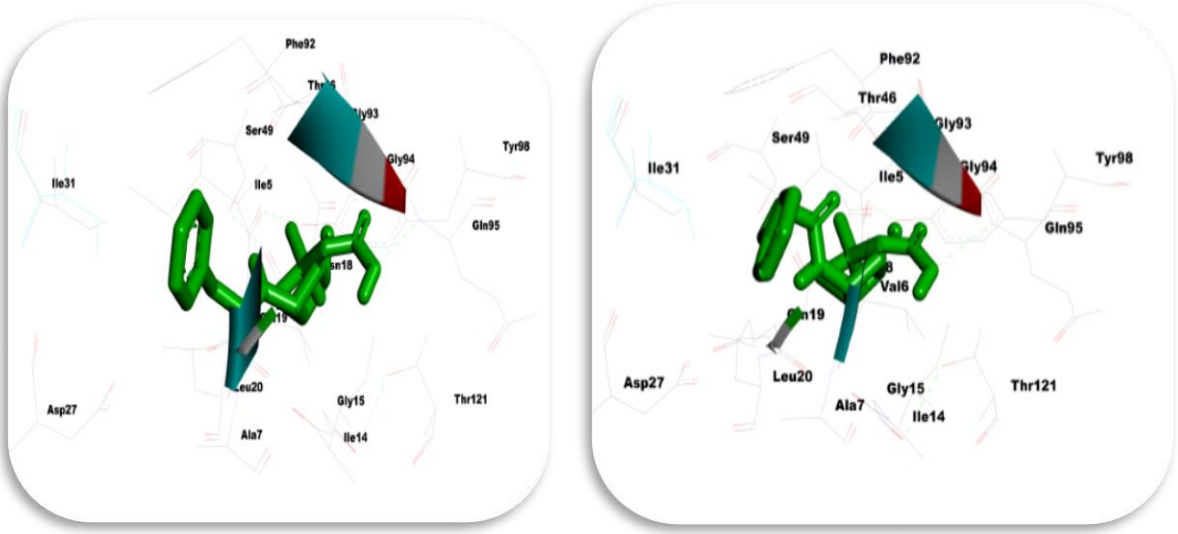
Table IV: Molecular interaction result of Synthetic compounds along with Cyanobacterial bioactive compounds against 2W9S

S.No.	Ligands	Binding Energy kcal/mol	Inhibition constant (Ki)
1.	Penicillin G	-8.59	502.32nM
2.	Penicillin V	-8.26	884.95nM
3.	Cyanobacterin	-8.64	468.06nM

Table V: Molecular interaction result with interacted amino acid residues of Synthetic compounds along with Cyanobacterial bioactive compounds against 2W9S

S.No	Compound Name	Binding Energy kcal/mol	Amino Acid	Inhibition constant (Ki)
1.	Penicillin G	-8.59	Ile31, Ile5, Asp27, Val6, Ala7, Leu20, Gln19, Gly15, Ile14, Thr121, Asn18, Gln95, Gly94, Tyr98, Gly93, Ser49, Thr46, Phe92	502.32nM
2.	Penicillin V	-8.26	Ile31, Ile5, Asp27, Ala7, Val6, Leu20, Gln19, Asn18, Gly15, Ile14, Thr121, Gln95, Gly94, Tyr98, Thr46, Phe92, Ser49, Gly93.	884.95nM
3.	Cyanobacterin	-8.64	Leu54, Lys52, Ile31, Leu28, Ile50, Phe92, Ser49, Thr46, Lys45, Gly93, Gly94, Tyr98, Asn18, Gln19, Gln95, Gln17, Leu20, Gly15, Ile14, Tyr16, Thr121, Asp120.	468.06nM

Complex of Cyanobacterin with 2W9S



Complex of Penicillin G with 2W9S **Complex of Penicillin V with 2W9S**

Fig IV: Complexes of synthetic and cyanobacterial ligand with 2W9S

IV. Discussion

The docked ligands of cyanobacterial compounds displayed acceptable binding energy values for 1T2P i.e. Calothrixin (-7.82) and Cyanobacterin (-7.45) having good binding affinity in comparison of synthetic compound which is Penicillin G (-5.24) and Penicillin V (-6.25) and also Cyanobacterin (-8.64) showed the good binding energy in comparison of synthetic compound i.e. Penicillin G (-8.59) and Penicillin V (-8.26) against the second one target which is 2W9S. These 2 cyanobacterial compounds out of 50 having more efficient result. These results suggest that Cyanobacterin and Calothrixinthese Cyanobacterial Bioactive compounds could serve as antibacterial compounds for the inhibition of proteins markers 1T2P and 2W9S in *Staphylococcus aureus*.

V. Conclusion

There is a necessity for designing drugs for *Staphylococcus aureus* infections caused in human as it has remained an opportunistic pathogen which causes significant number of the serious and deadly diseases in human and has been found resistant against various antibiotics present in the market. The docked ligands displayed acceptable binding energy values for 1T2P and 2W9S. These results suggest that Cyanobacterial Bioactive compounds such as Cyanobacterin, Calothrixin, could serve as inhibitor for proteins 1T2P and 2W9S in Staphyllococcus aureus. So these Cyanobacterial bioactive compounds can act as inhibitors of *Staphyllococcus aureus* infections. By further in vitro studies, that the extract of natural bioactive compounds from cyanobacterial strains can be examined as a potential antibacterial agent

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Competing interests
The authors declare no conflict of interest

Ethical approval
Not Applicable

Consent to participate
Not Applicable

Consent to publish
Not Applicable

VI. References

[1]. Laxminarayan, R., Matsoso, P., Pant, S., Brower, C., Røttingen, J. A., Klugman, K., & Davies, S. (2016). Access to effective antimicrobials: a worldwide challenge. The Lancet, 387(10014), 168-175

[2]. Sarmah, P., Dan, M. M., Adapa, D., & Sarangi, T. K. (2018). A review on common pathogenic microorganisms and their impact on human health. Electronic Journal of Biology, 14(1), 50-58.

[3]. Bharadwaj, A., Rastogi, A., Pandey, S., Gupta, S., &Sohal, J. S. (2022). Multidrug-resistant bacteria: their mechanism of action and prophylaxis. BioMed research international, 2022(1), 5419874.

[4]. Ahmad, I., & Aqil, F. (2020). New strategies combating bacterial infection. Wiley. https://www.wiley.com/enus/New%2BStrategies%2BCombating%2BBacterial%2BInfection-p-9783527322060

[5]. Lahlou, M. (2013). The success of natural products in drug discovery. Pharmacology & Pharmacy,4(3A).https://doi.org/10.4236/pp.2013. 43A003

[6]. Garcia-Pichel, F., Belnap, J., Neuer, S., &Schanz, F. (2003). Estimates of global cyanobacterial biomass and its distribution. Algological Studies, 109(1), 213–227.

[7]. Swain, S. S., Paidesetty, S. K., &Padhy, R. N. (2017). Antibacterial, antifungal and antimycobacterial compounds from cyanobacteria. Biomedicine & Pharmacotherapy, 90, 760–776.

[8]. Demay, J., Bernard, C., Reinhardt, A., & Marie, B. (2019). Natural products from cyanobacteria: Focus on beneficial activities. Marine Drugs, 17(6), 320.

[9]. Levasseur, P., &Pozzobon. (2020). A review of high value-added molecules production by microalgae in light of the classification. Biotechnology Advances, 41, 107545.

[10]. Xue, Y., Zhao, P., Quan, C., Zhao, Z., Gao, W., Li, J., Zu, X., Fu, D., Feng, S., Bai, X., Zuo, Y., & Li, P. (2018). Cyanobacteria-derived peptide antibiotics discovered since 2000. Peptides, 107, 17–24.

[11]. Ogston A (1984) Classics in infectious diseases. Rev Infect Dis 6(1):122–128. https://doi.org/10.1093/clinids/6.1.122

[12]. Kluytmans J, Belkum AV, Herbrugh V (1997) Nasal carriage of Staphylococcus aureus:N epidemiology, underlying mechanisms, and associated risks. ClinMicrobiol Rev 10(3):505–520. https://doi.org/10.1128/CMR.10.3.505

[13]. Cole AM, Tahk S, Oren A, Yoshioka D, Kim YH, Park A, Ganz T (2001) Determinants of Staphylococcus aureus nasal carriage. ClinDiagn Lab Immunol 8(6):1064–1069. https://doi.org/10.1128/CDLI.8.6.1064-1069.2001

[14]. Llarrull LI, Fisher JF, Mobashery S (2009) Molecular basis and phenotype of methicillin resistance in Staphylococcus aureus and insights into new β- lactams that meet the challenge. Antimicrob Agents Chemother 53(10): 4051–4063. https://doi.org/10.1128/AAC.00084-09

[15]. Sainsbury S, Bird L, Rao V, Shepherd SM, Stuart DI, Hunter WN, Owens RJ, Ren J (2011) Crystal structures of penicillin-binding protein 3 from Pseudomonas aeruginosa: comparison of native and antibiotic-bound forms. J MolBiol 405(1):173–184. https://doi.org/10.1016/j.jmb.2010.10.024

[16]. Yoshida H, Kawai F, Obayashi E, Akashi S, Roper DI, Tame JR, Park S-Y (2012) In silico study on penicillin derivatives and cephalosporins for upper respiratory tract bacterial pathogens. J MolBiol 423(3):351–364. https://doi. org/10.1016/j.jmb.2012.07.012

[17]. Manoharan P, Ghoshal N. Fragment-based virtual screening approach and molecular dynamics simulation studies for identification of BACE1 inhibitor leads. Journal of Biomolecular Structure and Dynamics. 2018 May 19;36(7):1878-92.

[18]. Castro, L. H., &Sant'Anna, C. M. R. (2022). Molecular Modeling Techniques Applied to the Design of Multitarget Drugs: Methods and Applications. Current Topics in Medicinal Chemistry, 22(5), 333-346

[19]. Pal, M., Gutama, K. P., &Koliopoulos, T. (2021). Staphylococcus aureus, an important pathogen of public health and economic importance: A comprehensive review. Journal of Emerging Environmental Technologies and Health Protection, 4(2), 17-32.

[20]. Fayisa, W. O., &Tuli, N. F. (2023). Review on Staphylococcus aureus. Int. J. Nurs. Care Res, 1, 1-8.

[21]. Kudryavtsev, K. V., Fedotcheva, T. A., &Shimanovsky, N. L. (2021). Inhibitors of sortases of gram-positive bacteria and their role in the treatment of infectious diseases. Pharmaceutical Chemistry Journal, 55(8), 751-756.

[22]. Sehrawat, R., Rathee, P., Khatkar, S., Akkol, E., Khayatkashani, M., Nabavi, S. M., &Khatkar, A. (2024). Dihydrofolate reductase (DHFR) inhibitors: a comprehensive review. Current Medicinal Chemistry, 31(7), 799-824.

[23]. Taj, Z., & Chattopadhyay, I. (2024). Staphylococcus aureus virulence factors and biofilm components: synthesis, structure, function and inhibitors. In ESKAPE pathogens: detection, mechanisms and treatment strategies (pp. 227-270). Singapore: Springer Nature Singapore.

[24]. Linz, M. S., Mattappallil, A., Finkel, D., & Parker, D. (2023). Clinical impact of Staphylococcus aureus skin and soft tissue infections. Antibiotics, 12(3), 557.

[25]. Ahmad, S., Akhtar, S., &Farooqui, A. (2024). In Silico Validation Studies of Cyanobacterial Bioactive Compounds Against A-amylase and A-glucosidase Markers in Type 2 Diabetes Mellitus. The Open Bioinformatics Journal, 17(1).

[26]. Nitulescu, G., Nicorescu, I. M., Olaru, O. T., Ungurianu, A., Mihai, D. P., Zanfirescu, A., ...&Margina, D. (2017). Molecular docking and screening studies of new natural sortaseA inhibitors. International Journal of Molecular Sciences, 18(10), 2217.

[27]. Polshakov, V. I. (2001). Dihydrofolate reductase: structural aspects of mechanisms of enzyme catalysis and inhibition. Russian chemical bulletin, 50(10), 1733-1751.