MOLECULAR DOCKING STUDIES OF NOVEL AMINOALKYLNAPHTHOLS AS POTENT INHIBITORS OF UDP-3-O-[R-3-HYDROXYMYRISOYL]-N-ACETYLGLUCOSAMINE DEACTYLASE (LPXC) OF P. AERUGINOSA AND PEPTIDE DEFORMYLASE (PDF) OF S. AUREUS

Imtiaz Khalid Mohammed[a]*, Waheedah Abdul Hakeem[b], Nurkhalida Kamal[c], Noorazura Ashikeen Ahmad[d]

Article History: **Received:** 23.04.2022 **Revised:** 17.08.2022 **Accepted:** 04.10.2022

Abstract: The spawn of antimicrobial resistance (AMR) era has started ever since the past few decades, and it has caused a great concern especially by healthcare providers. The importance of a new class of novel antibiotics is very much needed to overcome this AMR. Fortunately, through computer aided drug design (CADD) and various other *in silico* methods, it allows scientists to create new potent ligands as potential drugs. **Methodologies:** Herein, 156 novel aminoalkylnaphthols are examined for the first time in this molecular docking study against two metalloenzymes; UDP-3-O-[R-3-hydroxymyrisoyl]-N-acetylglusomaine deactylase of *pseudomonas aeruginosa* (PaLpxC) and peptide deformylase of *staphylococcus aureus* (SaPDF). **Results:** The novel ligands are reported to have satisfactory inhibition constant (K_i). The best docked ligand against PaLpxC is 4y with K_i of 1.71 and 4u with a K_i of 14.73 against SaPDF. This indicates the potency of the novel ligands. Besides that, after further graphical analyses, the novel aminoalkylnaphthols are found to bind at a new binding site in PaLpxC as compared to reported inhibitors in literatures, which might suggest a new promising binding site has been found. **Conclusion:** Therefore, this new class of novel ligands are promising potent inhibitors against these two important enzymes.

[a]. Department of Pharmaceutical Sciences, Faculty of Pharmacy, University of Cyberjaya, Cyberjaya, 63000, Malaysia.
[b]. Department of Pharmaceutical Sciences, Faculty of Pharmacy, University of Cyberjaya, Cyberjaya, 63000, Malaysia.
[c]. Department of Pharmaceutical Sciences, Faculty of Pharmacy, University of Cyberjaya, Cyberjaya, 63000, Malaysia.
[d]. Department of Pharmaceutical Sciences, Faculty of Pharmacy, University of Cyberjaya, Cyberjaya, 63000, Malaysia.

***Corresponding Author**

E-mail: imtiaz@cyberjaya.edu.my

DOI: 10.31838/ecb/2022.11.09.008

INTRODUCTION

Antimicrobial resistance (AMR) has caused a great fear in the healthcare setting and it has increased at an alarming rate over the past decade (Laxminarayan et al., 2013). It is responsible for many deaths worldwide and the need for a new novel class of antibiotic is very much in need[1]. This is supported by the global action plan as mentioned by World Health Organization (WHO) in fighting against AMR in which they stressed about acquiring new knowledge on potentially potent drugs which might fight known bacteria with AMR (WHO, 2015). Two out of many bacteria which possess this AMR includes *p. aeruginosa* and *s. aureus* (Frieden, T. 2013). There are many ongoing researches to find new inhibitors to fight with these bacteria and the few promising targets includes targeting the UDP-3-O-[R-3-hydroxymyrisoyl]-N-acetylglusomaine deactylase of *pseudomonas aeruginosa* (PaLpxC) and peptide deformylase of *staphylococcus aureus* (SaPDF) (Palamthodi, Gaikwad, Ghasghase, & Patil, 2011; Verma et al., 2011).

PDF is a unique metalloenzyme which is important in the protein syntheses in bacteria. It works by removing the removing the formyl group in N-methylmethionyl peptide in which subsequent product will be the precursor to a mature protein[5]. LpxC is another essential metalloamidase found to be important in the biosynthesis of lipid A of Gram-negative bacteria. Lipid A is crucial in protecting the bacteria against any external agents (e.g. antibiotics) (Angelica & Fong, 2008). Another specialty about LpxC is that it can only be found in Gram-negative bacteria as it does not have any similar sequence to other mammalian protein, which makes it a unique target (Barb & Zhou, 2008). Therefore, highly specific inhibitors can be designed to halt the processes of these important enzymes and may subsequently inhibit the growth of the bacteria.

Actinonin, an inhibitor of PDF was discovered as early as in 1962 which has been identified to have a K_i of 0.28 nM. PDF inhibitor contains a metal chelating group that is suggested to be the main functional group and the crucial aspect in the PDF inhibition (Chen et al, 2000). Meanwhile, clinically useful antibiotics are not yet available that specifically targets LpxC or in general the biosynthesis of lipid A (Angelica & Fong, 2008)[6].

Aminoalkylnaphthols were firstly synthesised as early as 1900s by Mario Betti and they were called as *Betti Base* (Mukhopadhyay, Rana, & Butcher, 2012). These novel compounds are found to have an analgesic effect as well as the ability to combat numerous diseases that includes migraines, asthma, cardiovascular diseases and many more (Doyle, Dale, Choi, & City, 2012). It was until recently that these novel aminoalkylnaphthols was found to have prominent biological properties including cytotoxic activity, antioxidant, antifungal and antibacterial properties (Raghunath & Mathada, 2014). It was thought that these aminoalkylnaphthols possesses chelating centres that can inhibit metalloenzymes; the two enzymes mentioned previously (Mukhopadhyay et al., 2012)[9].

Currently, there has been a lot of molecular docking studies which targets these enzymes however no literatures are available for the docking studies between these targets and the novel aminoalkylnaphthols. Herein in this study, the novel property of these compound will be discussed by knowing its binding interaction. It is hoped that this study will provide a new window for research in advancing a potent ligand into a potent drug for the use of patients in the future.

METHODOLOGIES

Preparation of ligands

156 aminoalkylnaphthol compounds were sketched in 2D structures by using ChemSketch Software before proceeding to converting into 3D structures, adding hydrogens and structure minimization by using BIOVIA Discovery Studio Client 2016. Out of 156, 13 structures were extracted from available literature (Mukhopadhyay et al., 2012). The rest of the compounds were made by obtaining all possible combinations of starting materials from the 13 structures. All files were then saved in PDB format. Positive controls in this study are LPC-040, in which the structure was obtained by extracting it from an available crystal complex (PDB entry: 5DRQ) found at RCSB Protein Data Bank (PDB) and actinonin as the PDF inhibitor which is drawn similarly like those of the steps used in preparing the individual ligands. All ligands were converted from PDB format into PDBQT by using AutoDock 4.2

Target Crystal Structures or Enzymes

The X-ray crystal structures of PaLpxC (PDB entry: 5DRQ) and SaPDF (PDB entry: 1LMH) were obtained from RCSB protein databank. Water molecules were removed from the crystal structures.

Molecular Docking

All molecular dockings were done by using the AutoDock 4.2. X, Y and Z directions for Grid parameter file (GPF) of both enzymes were set to 110 x 110 x 110. For PaLpxC, the grid box was positioned at 31.624, 18.264 and 41.406 in x, y and z coordinates respectively while the grid box of SaPDF was centred at 17.412, 4.725 and 6.230 in x, y and z directions respectively. All GPF files were created by using AutoDockTools (ADT).

In this study, Lamarkian Genetic Algorithm (LGA) was chosen for all docking stimulations. The Genetic Algorithm (GA) parameters were set to default settings except for the number of GA runs which was set to 50 runs. The population size (150), maximum number of energy evaluation performed (2500000), the maximum number of generations simulated during each LGA run (27000) were all set to default.

The novel ligands were ranked based on their inhibition constants (K_i) against the target proteins PaLpxC and SaPDF. 20 top ligands of each target protein were considered as hits. Graphical analyses were done for the top 2 hits of each target using BIOVIA Discovery Studio Client 2016 to get a more detailed view of the possible interaction involved.

Validation of Molecular Docking

To validate the docking method used in this study, prior molecular docking study was done to the crystal structure of PaLpxC and its inhibitor LPC-040 with similar docking parameters as mentioned above. This was done to ensure that the ligand is binding in the similar binding pocket at the crystallographic structure. The conformation of the docked structure of LPC-040 was then compared against the crystallographic structure to test and validate the molecular docking.

RESULTS AND DISCUSSION

Validation of Molecular Docking

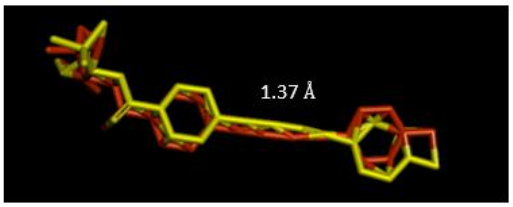


Figure 1: Superimposition of crystallographic structure of LPC-040 shown in yellow and the docked structupare of LPC-040 in red

As stated previously, for validation the docking process, LPC-040 was docked against its crystal structure PaLpxC and the superimposition of the crystal ligand structure and the docked conformation is shown in figure 1. This is to ensure that the docking process of the novel ligands and the target structure is in the same binding pocket. The redocking of the ligand showed a root mean square deviation (RMSD) of 1.37 Å. It is generally accepted, and it is considered successful if the redocking of the ligand to its complex is under 3 Å. This means that the difference of confirmation between the original crystallographic structure and the redocked ligand is only by the distance of 1.37 Å. The docking method in this study is therefore considered to be reliable.

Docking Results

The top 20 docked results are shown in table 1 for novel aminoalkylnaphthols against PaLpxC and table 2 for the same compounds against SaPDF.

Table 1 show the top 20 derivatives that have free energy of binding (FEB) ranging from -11.96 to -10.91 kcal/mol. This has lesser FEB than the positive control LPC-040 having a FEB of -9.77 kcal/mol. This show that the newly designed derivatives are potentially more potent than the validated inhibitor itself.

Top 3 derivatives are 4y, 4u and 4t and have a FEB of -11.96, -11.89 and -11.68 kcal/mol respectively.

Table 2 show the top 20 derivatives that has been docked to PDF of *S. aureus*. They have FEB ranging between -10.68 to -9.89 kcal/mol. Newly designed top hits have a preferable interaction between ligand-protein complex better than the validated PDF inhibitor, actinonin which has a FEB of -7.47 kcal/mol. 3 new derivatives are 4ae, 4ac and 4ab with FEB of -10.68, -10.66 and -10.56 kcal/mol respectively. Our best docked model has more favourable FEB compared to 3 out of 4 models that was studied to inhibit SaPDF (Gao et al., 2013).

The inhibition constant (Ki) is one of the most important data and it indicates the potency of a compound. In molecular docking that correlates directly with the FEB (Autodock, 2010; Iman, Sasdabadi, & Davood, 2015):

Table 1 show the top 20 docked derivatives against LpxC of *p. aeruginosa* that have K_i ranging from 1.71 to 10.02 nM. LPC-040 (positive control) however was found to have a K_i of 68.41 nM which is significantly higher than the new derivatives. This means that the new derivatives are much more potent than the validated inhibitor. Top 3 docked aminoalkylnaphthol derivatives against LpxC have K_i of 1.71, 1.92 and 2.68 nM for 4y, 4u and 4t respectively.

The success of the new derivatives can also be seen upon docking to PDF of *s. aureus* as per table 2. The top 20 docked inhibitors have K_i ranging from 14.73 nM to 56.41 nM which compared to the validated inhibitor actinonin has K_i of 3370 nM. The difference in K_i between the validated inhibitor and novel aminoalkylnaphthol derivatives is outstanding which implies the higher potency of the derivatives compared to actinonin. The top 3 docked aminoalkylnaphthol derivatives against PDF are 4ae, 4ac and 4ab which has K_i of 14.73, 15.41 and 18.17 nM respectively.

Overall, all docked derivatives show good correlation between FEB and Ki towards the target site. The top docked derivatives have shown better FEB and Ki as being compared to their respective positive controls. The top 2 best docked derivative against LpxC of p. aeruginosa (4y and 4u) and PDF of s. aureus (4ae and 4ac) were further analysed by investigating the overall ligand-protein interaction to compare with their respective positive controls. This is to understand the possible interactions that contributed to the total FEB.

Table 1. Top 20 aminoalkylnaphthol derivatives docked with LpxC of p. aeruginosa

Entry	FEB[a]	K_i[b]	Cluster[c]
Positive Control			
LPC-040	-9.77	68.41	42
Aminoalkylnaphthol Derivatives			
4y	-11.96	1.71	6
4u	-11.89	1.92	5
4t	-11.69	2.68	15
4m	-11.59	3.21	2
4z	-11.42	4.23	5
4aa	-11.32	5.07	3
4do	-11.31	5.13	2
4ds	-11.26	5.55	6
4ag	-11.25	5.63	1
4af	-11.23	5.89	2
4dn	-11.21	6.03	5
4dp	-11.19	6.25	5
4dr	-11.14	6.82	4
4eo	-11.13	6.96	2
4cj	-11.11	7.22	6
4q	-11.09	7.46	7
4cg	-11.05	7.90	3
4bt	-11.03	8.24	2
4v	-10.92	9.89	1
4l	-10.91	10.02	2

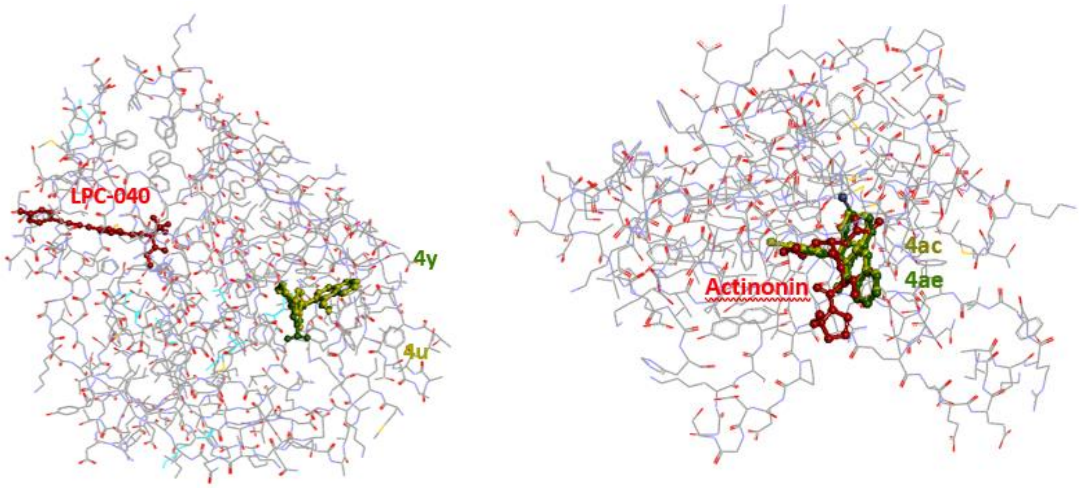
[a] Free Energy of Binding in kcal.mol^{-1}
[b] Inhibition constant in nano molar concentration
[c] Number of conformations in a cluster

Table 2. Top 20 aminoalkylnaphthol derivatives docked with peptide deformylase (PDF) of S. aureus

Entry	FEB[a]	K_i[b]	Cluster[c]
Positive Control			
Actinonin	-7.47	3370	3
Aminoalkylnaphthol Derivatives			
4ae	-10.68	14.73	20
4ac	-10.66	15.41	22
4ab	-10.56	18.17	21
4dz	-10.56	18.17	12
4bq	-10.35	25.92	1
4be	-10.35	26.03	3
4ef	-10.32	27.11	1
4bh	-10.31	27.55	12
4dp	-10.27	29.63	4
4bd	-10.25	30.77	13
4ai	-10.21	32.86	17
4b	-10.21	32.90	2
4a	-10.19	34.05	8
4do	-10.18	34.40	39
4ad	-10.16	35.87	1
4bt	-10.06	42.51	12
4ec	-10.02	45.09	1
4k	-10.10	46.31	16
4dn	-10.00	46.98	16
4ap	-9.89	56.41	45

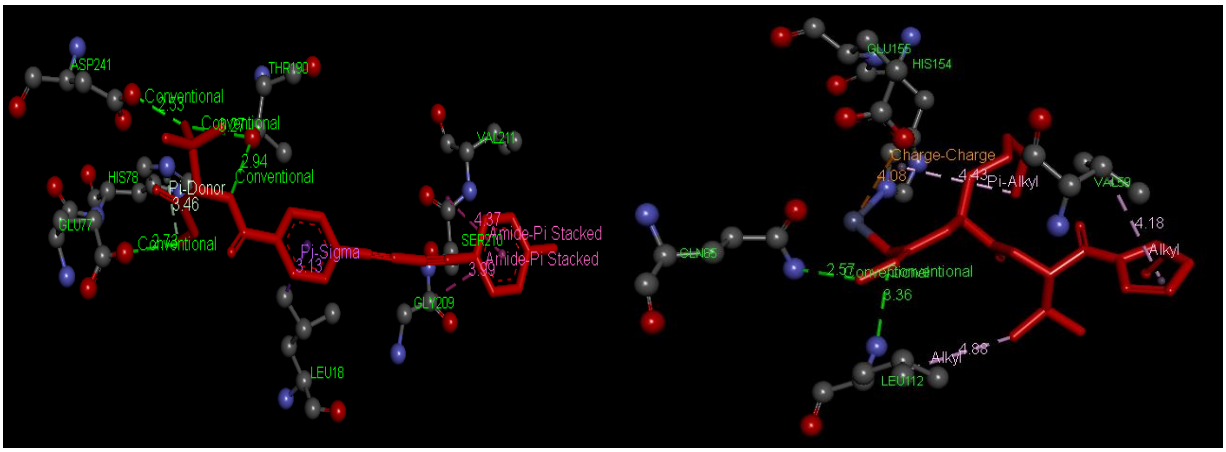
[a] Free Energy of Binding in kcal.mol^{-1}
[b] Inhibition constant in nano molar concentration
[c] Number of conformations in a cluster

Graphical Analyses



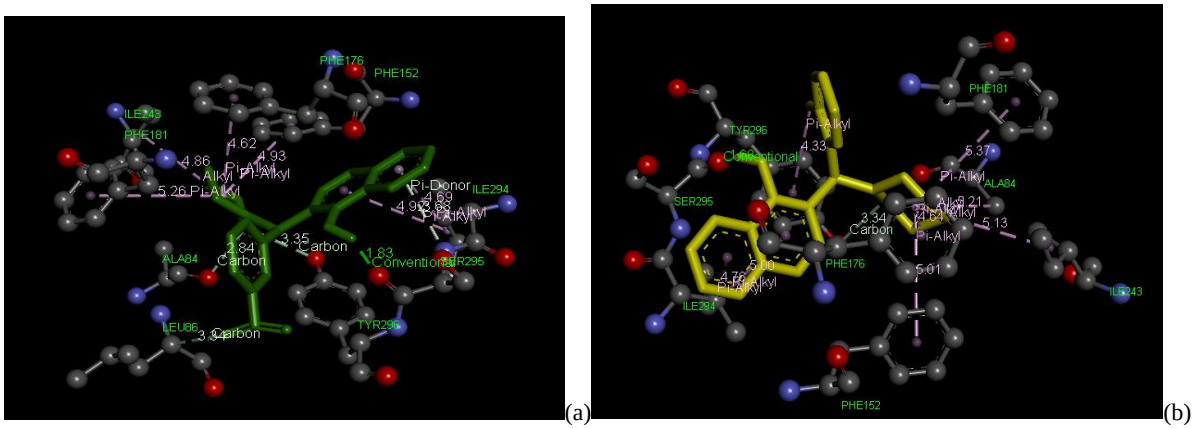
(a) (b)

Figure 2: Comparison of binding site and conformation of the novel ligands (green and yellow-coloured) and its respective positive control (red-coloured) against (a) PaLpxC and (b) SaPDF



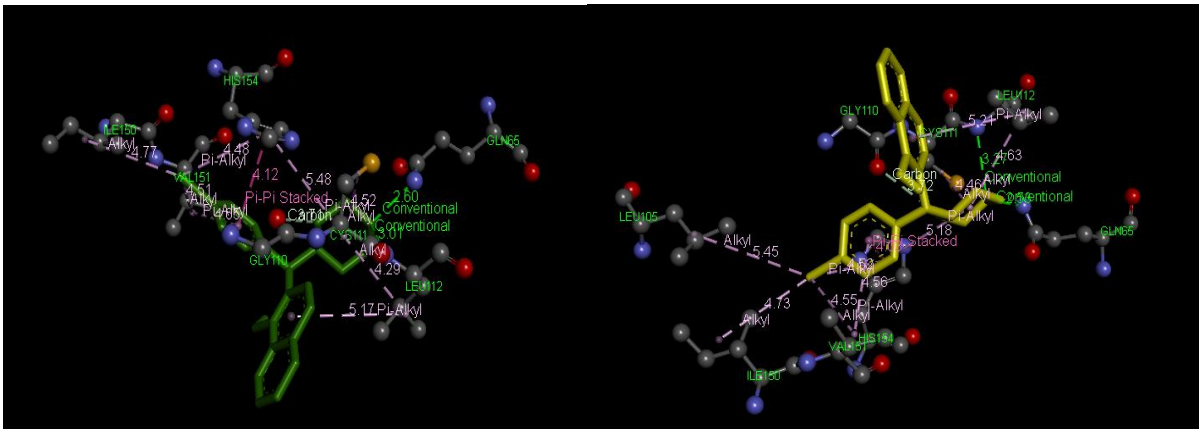
(a) (b)

Figure 3: Interaction between (a) LPC-040 and amino acid residues of PaLpxC and (b) actinonin and amino acid residues of SaPDF.



(a) (b)

Figure 4: Interaction of ligands (a) 4y and (b) 4u with amino acid residues of PaLpxC.



(a) (b)

Figure 5: Interaction of ligands (a) 4ae and (b) 4ac with amino acid residues of SaPDF.

Figure 2(a) show the binding site of LPC-040, the positive control and validated inhibitor of PaLpxC is different compared to 4y and 4u. Table 3 summarises the amino acid residues that show interaction with the ligands. There are no matching residues that interact with either ligand and therefore, this show a new potential binding site for LpxC. This is further supported by a recent study conducted by Pradhan *et al*, where it is reported that the amino acid residues that are involved in binding between the inhibitor BB-78485 and LpxC of *leptospirosa* includes Leu21, Asn65, Ala66, Glu80, His81, Thr191, Phe192, Gly193, Phe194, Asp197, Gly210, Ser211, Ala215, His237, Asp241, His264, etc. which are not similar to any amino acid residue reported with the docked ligand, 4y and 4u (Pradhan et al., 2014). In another molecular docking study between the potent inhibitor of LpxC for *E. Coli*, 1B1 and an inhibitor of LpxC for *P. aeruginosa*, 1B2 has found that the common residues involved in both of the active binding pocket includes Val174, Leu253, Gly85, Phe152, Ala84, Val182, Leu240, Val111, Val185, Leu243, Phe181 and Ser252 (Kadam, Shivange, & Roy, 2007). These amino acid residues did not match with any residues that were found in this study.

Furthermore, in figure 2(b), ligands were docked to a similar binding site in SaPDF. Table 3 show that three of amino acid residues in PDF have interacted with all three ligands and positive control (actionin and 4ae and 4ac). These residues are leu112 and his154. However, a study reported that the binding of actinonin to PDF of *s. aureus* includes the amino acid residue Cys111, which is seen in the binding mode between 4ae and 4ac against the target but not seen in the docked positive control actinonin and PDF (Yoon et al., 2004).

Table 3. Overall interaction between novel ligands and the amino acid residues of target sites which includes hydrogen bonds and hydrophobic bonds.

Protein	Entry	Residues Involved
P. aeruginosa LpxC	LPC-040	Leu18, Glu77, His78, Thr190, Gly209, Ser210, Val211, Asp241,
	4y	Ala83, Leu86, Phe152, Phe176, Phe181, Ile243, Ile294, Ser295, Tyr296,
	4u	Ala84, Phe152, Phe176, Phe181, Ile243, Ile294, Ser295, Tyr296
S. aureus PDF	Actinonin	**Gln65**, **Leu112**, **His154**, Glu155, Val59
	4ae	Ile150, **His154**, Val151, Gly110, **Gln65**, Cys111, **Leu112**
	4ac	**Leu112**, **Gln65**, Cys111, Gly110, Val151, **His154**, Leu105, Ile150

Table 4. Specific interaction between novel ligands and the amino acid residues of target sites which includes the number of hydrogen bonds and hydrophobic bonds found.

Protein	Entry	FEB	H-bond	Residues Involved	H-phobic	Residues Involved
P. aeruginosa LpxC	4y	-11.96	1	Ser295	5	Ile294, Phe176, Ile294, Ile243, Phe152
	4u	-11.89	1	Ser295	7	Ile294, Ala84, Phe152, Phe181, Ile243, Phe176, Tyr296
S. aureus PDF	4ae	-10.68	2	Leu112, Gln65	6	Ile150, Val151, Cys111, His154
	4ac	-10.66	2	Leu112, Gln65	10	Ile150, Val151, Cys111, His154, Leu112, Leu105

Based on figure 4 and table 4, there is only one conventional hydrogen bond identified between both novel ligand 4y and 4u against PaLpxC. The interaction between Ser295 of the binding pocket and hydroxyl group of the ligand 4y has the strong interaction with distance of 2.90 Å however the hydrogen bond formed between Ser295 and hydroxyl group of 4u has a stronger interaction with a distance of 1.68 Å. Reported in a molecular docking study, a potent inhibitor of *P. aeruginosa*'s LpxC, 1t, was found to have a hydrogen-bonding interaction with Lys238 and have a distance of 3.20 Å (Brown et al., 2012). Ligand 4y and 4u forms a shorter hydrogen bond, than the reported literature which suggests a stronger bond. However, the interaction of BB-78485 inhibitor docked with LpxC of *P. aeruginosa* have formed a total of 5 hydrogen bonds out of which 2 of the bonds were formed with a distance of 3.0 Å and 3.3 Å respectively is slightly weaker than the hydrogen bonds

formed of the best docked derivative, 4y (Mochalkin, Knafels, & Lightle, 2008). The other 3 bonds have a range of distance between 2.5–2.8 Å. Ligand 4u forms only one bond but has a shorter distance compared to all 5 of BB-78485 hydrogen bonds.

Only two conventional hydrogen bonds are identified between both ligand 4ae and 4ac against amino acid residues in binding site of SaPDF as seen in figure 5. The bond formed between nitrogen of Gln65 and Leu112 and the oxygen of phenyl group of 4ae has 2.60 Å and 3.01 Å distance respectively. However, the hydrogen bond form between the oxygen of phenyl of ligand 4ac and nitrogen of Gln165 is stronger compared to 4ae with 2.53 Å distance. Interaction between nitrogen Leu112 and oxygen of phenyl for 4ac has 3.27 Å distance. LBM415, the first candidate that entered phase I clinical trial as the PDF inhibitor was also found to have a hydrogen bond with residues Leu112 and Val59 (Lv et al., 2016). 4ae and 4ac also forms hydrogen bond with Leu112.

It has been highlighted in the previously that the binding pocket of our novel ligand against PaLpxC is at a new binding site compared to the positive control, LPC-040. It is consistently reported in a study that the interaction between various inhibitors of LpxC of *p. aeruginosa* commonly involves amino acid residues His78 and Glu77 among many that forms hydrogen bond with the inhibitors[18]. These residues were not identified to be involved in the interaction with 4y, instead, both residues were found to be involved with the positive control ligand, LPC-040 as shown previously in table 4 Therefore, this further suggests that a potentially new binding site is discovered.

Referring to the docking between ligand and PDF of *s. aureus*, a study reported that macrolactin N, an inhibitor of PDF of *s. aureus* formed an interaction with the amino acid residue Gly110, which found to be similar with both 4ae and 4ac (Gao et al, 2013). Glyn110 and 4ae as well as 4ac forms a carbon hydrogen bond together instead of conventional hydrogen bond.

Table 4 summarises the hydrophobic interaction between amino acid residues of the targets and the ligand. Both ligand 4u and 4ac have a higher number of hydrophobic interactions as compared to the best docked novel ligand of each target protein which is 4y and 4ae respectively.

A study reported that LpxC inhibitors; S-15192 and 49536 were found to have an interaction with the amino acid residues Thr190 and Asp241 respectively, which was seen in the positive control, LPC-040 but was not seen in 4y and 4u (Kadham et al, 2007).

Table 4 show amino acid residues of SaPDF that are involved in the hydrophobic interaction. The interactions are anticipated as reported literature have suggested that Cys111, His154 and His158 residues are important in the binding process between actinonin and SaPDF[17]. Gao *et al* have also reported the important amino acid residues that participates in the vdW interaction within PDF of *s.* aureus and macrolactin N includes Arg56, Gly59, Val59, Leu61, Leu105, Gly110, Cys111, Leu112, Val151 and His154[13]. His154 and Val151 was also seen in the binding between actinonin, 4ae and 4ac.

CONCLUSION

In this first in its class molecular docking study, we have concluded that out of 156 novel aminoalkylnaphthols that was tested for docking against PaLpxC and SaPDF, the top 20 novel ligands were found to be more potent than their respective positive control, which was decided based on its inhibition constant, K_i. Further graphical analyses have suggested that novel ligand 4ae and 4ac are bound to a new binding site, which was not reported in literature thus far. Thus, this gives us a new information that new novel and potent ligands has the capability to inhibit the important enzymes which in return, can inhibit the growth of the bacteria *p. aeruginosa* and *s. aureus.* Further study is needed to prove the potency of this drug through synthetisation and *in vitro* studies.

Funding: This research was funded by CUCMS Research grant.

Acknowledgments: We would like to acknowledge staffs of UOC laboratory and MGI for their invaluable assistance in handling the chemicals and instruments.

Conflicts of Interest: The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

REFERENCES

i. Laxminarayan, R., Duse, A., Wattal, C., Zaidi, A. K. M., Wertheim, H. F. L., Sumpradit, N., Cars, O. (2013). Antibiotic resistance — the need for global solutions, 3099(13).

ii. WHO/World Health. (2015). Global Action Plan on Antimicrobial Resistance.

iii. Frieden, T. (2013) Antibiotic Resistance Threats in the United States. Diane Publishing Company, Collingdale, PA.

iv. Palamthodi, S. M., Gaikwad, V. J., Ghasghase, N. V, & Patil, S. S. (2011). Antibacterial targets in pseudomonas aeruginosa. *International Journal of Pharmaceutical Application*, *2*(3), 159–164.

v. Verma, S. K., Jat, R. K., Nagar, N., Saharan, R., Sharma, V., Pandey, S., & Bansal, K. (2011). A Novel Antibacterial Target: Peptide Deformylase, *2*(2), 114–123.

vi. Angelica, M. D., & Fong, Y. (2008). Mechanism and Inhibition of LpxC: an essential zinc-dependent deacetylase of bacterial lipid A synthesis. *October*, *141*(4), 520–529.

vii. Mukhopadhyay, C., Rana, S., & Butcher, R. J. (2012). Catalyst-Free, One-Pot, Expeditious Synthesis of Aminoalkylnaphthols at Room Temperature. *Synthetic Communications*, *42*(20), 3077–3088.

viii. Doyle, I. M. P., Dale, G., Choi, H., & City, B. (2012). (12) UnIted States Patent, *2*(12), 6071–6075.

ix. Raghunath, S. A., & Mathada, K. N. (2014). Synthesis and biological evaluation of aminonaphthols incorporated indole derivatives. *International Journal of Medicinal Chemistry, 12 pages*.

x. Gao, J., Cheng, Y., Cui, W., Zhang, F., Zhang, H.,

Du, Y., & Ji, M. (2013). Prediction of the binding modes between macrolactin N and peptide deformylase from Staphylococcus aureus by molecular docking and molecular dynamics simulations. *Medicinal Chemistry Research*, *22*(6), 2889–2901.

xi. AutoDock. (2010.). Retrieved December 20, 2020, from http://autodock.scripps.edu/faqs-help/faq/how-autodock-4-converts-binding-energy-kcal-mol-into-ki

xii. Iman, M., Saadabadi, A., & Davood, A. (2015). Molecular docking analysis and molecular dynamics simulation study of ameltolide analogous as a sodium channel blocker. *Turkish Journal of Chemistry*, *39*(2), 306–316.

xiii. Pradhan, D., Priyadarshini, V., Munikumar, M., Swargam, S., Umamaheswari, A., & Bitla, A. (2014). Para-(benzoyl)-phenylalanine as a potential inhibitor against LpxC of Leptospira spp.: homology modeling, docking, and molecular dynamics study. *Journal of Biomolecular Structure & Dynamics*, *32*(2), 171–85.

xiv. Kadam, R. U., Shivange, A. V., & Roy, N. (2007). Escherichia coli versus Pseudomonas aeruginosa deacetylase LpxC inhibitors selectivity: Surface and cavity-depth-based analysis. *Journal of Chemical Information and Modeling*, *47*(3), 1215–1224.

xv. Brown, M. F., Reilly, U., Abramite, J. A., Arcari, J. T., Oliver, R., Barham, R. A., … Wishka, D. G. (2012). Potent inhibitors of LpxC for the treatment of gram-negative infections. *Journal of Medicinal Chemistry*, *55*(2), 914–923.

xvi. Mochalkin, I., Knafels, J. D., & Lightle, S. (2008). Crystal structure of LpxC from Pseudomonas aeruginosa complexed with the potent BB-78485 inhibitor. *Protein Science : A Publication of the Protein Society*, *17*(3), 450–457.

xvii. Lv, F., Chen, C., Tang, Y., Wei, J., Zhu, T., & Hu, W. (2016). New peptide deformylase inhibitors design, synthesis and pharmacokinetic assessment. *Bioorganic and Medicinal Chemistry Letters*, *26*(15), 3714–3718.

xviii. Yoon, H. J., Hye, L. K., Lee, S. K., Kim, H. W., Kim, H. W., Jae, Y. L., … Se, W. S. (2004). Crystal structure of peptide deformylase from Staphylococcus aureus in complex with actinonin, a naturally occurring antibacterial agent. *Proteins: Structure, Function and Genetics*, *57*(3), 639–642.

xix. Kadam, R. U., Garg, D., Chavan, A., & Roy, N. (2007). Evaluation of Pseudomonas aeruginosa deacetylase LpxC inhibitory activity of dual PDE4-TNF inhibitors: A multiscreening approach. *Journal of Chemical Information and Modeling*, *47*(3), 1188–1195.