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Screening of Traditional Chinese Medicine Library Against Penicillin-binding Protein 2a for Methicillin-resistant *Staphylococcus aureus* by Molecular Docking, Dynamics Simulation and *In vitro* Antimicrobial Activity

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The methicillin-resistant Staphylococcus aureus (MRSA) is considered a public health threat that can increase both treatment duration and cost. Currently, the MRSA strain has developed resistance to many penicillins and cephalosporins due to the expression of insensitive penicillin-binding protein 2a (PBP2a). This transpeptidase variant, PBP2a, has a low binding affinity toward the ß-lactam ring. As such, the active site of the PBP2a enzyme is now regarded as a potential molecular target for developing new anti-MRSA therapeutics. Many of the available antibiotics were developed from microbial sources but the herbal sources are still to be explored. Therefore, the aim of this in silico study is to virtually screen a library of Traditional Chinese Medicine (TCM) compounds against the active site of PBP2a to identify possible anti-MRSA phytomedicines. For this purpose, both molecular docking and dynamics simulation were employed in this study. Moreover, the agar well diffusion method was used to assess the in vitro antimicrobial activity of final hit compounds against MRSA cultured colonies. The results of the molecular dynamics (MD) study indicate that both sciadopitysin and plantamajoside can maintain a close proximity to the PBP2a active site during 50 nanoseconds simulation. Additionally, the most preferred molecular mechanics-Poisson Boltzmann surface area (MM-PBSA) binding energy was reported to be -24.09 Kcal/ mol for sciadopitysin. While docking study results point to the possible hydrogen bond interaction of sciadopitysin and plantamajoside with Serine 403 active site residue. Further, the agar well diffusion study refers to the fact that both sciadopitysin and plantamajoside are effective in inhibiting the growth of MRSA culture with a measured zone of inhibition: 12.5 ± 0.7 and 9.0 ± 1.4 mm respectively. In conclusion, it is predicted that the phenolic compounds sciadopitysin and plantamajoside from the TCM library are potential inhibitors of the MRSA PBP2a enzyme.

Keywords: Antimicrobial Activity; Docking; Dynamics Simulation; Methicillin-resistant Staphylococcus aureus; Penicillin-binding protein 2a; Phytochemical compounds.

Staphylococcus aureus (*S. aureus*) is a gram-positive coccoid bacterium that is found in about 30% of the human population as part of skin and nasal normal flora.[1,2] Clinically, *S. aureus* has an adverse impact on human health as it can cause a wide spectrum of infective diseases like endocarditis, bacteremia, pneumonia and osteomyelitis.[3] Fortunately, these infective diseases were greatly controlled with the advent of penicillin antibiotics. However, the wide use of penicillin

antibiotics had subsequently forced some strains of *S. aureus* to produce penicillinase enzyme. The production of this hydrolytic enzyme enables *S. aureus* to resist various penicillin antibiotics by targeting â-lactam ring.[4] Then, methicillin was introduced as a semi-synthetic penicillin antibiotic that resists hydrolysis by penicillinase enzyme and thereby can control infections by these resistant *S. aureus* strains.[5] Eventually, the development of resistance against this antibiotic led to the emergence of methicillin-resistant *S. aureus* (MRSA) strain.[6]

Currently, MRSA is considered a severe threat to public health that can result in both community and hospital-acquired resistant *S. aureus* infections.[7] Based on a recent meta-analysis study, the global prevalence of MRSA in elderly care centers was 14.69% but the infection rate is believed to be gradually increasing.[8,9] The treatment of MRSA infection is challenging because this strain is resistant to many penicillin and cephalosporin antibiotics, and even the last resort antibiotics like vancomycin are no longer applicable.[10] As such, MRSA infection is usually associated with longer hospitalization, more treatment costs and higher hospital mortality.[11]

When considering the molecular mechanism of resistance acquired by MRSA, it has been observed that this stain can evade â-lactam antibiotics inhibitory effect through expression of â-lactam insensitive penicillin-binding protein 2a (PBP2a). The PBP2a is a transpeptidase enzyme encoded by *mecA* gene, this gene is believed to be acquired from a non-*S. aureus* source.[12] The penicillin-binding proteins (PBPs), also known as transpeptidases, are enzymes located on bacterial cell membrane. In *S. aureus*, four types of these transpeptidases were identified and these are: PBP1, PBP2, PBP3 and PBP4. Moreover, MRSA strain can express another variant known as PBP2a. The catalytic role of these PBPs is essential for the integrity of bacterial cell walls by crosslinking the peptide side chains of adjacent glycan strands.[13,14] Thus, the bactericidal effect of â-lactam antibiotics is produced by the irreversible binding of â-lactam ring with the active site in PBPs. As a result, the inactivated PBPs will no longer able to crosslink peptidoglycan in the cell wall leading to bacterial lysis.[15] Unfortunately, the affinity of â-lactam antibiotics towards PBP2a is very low. As a result, MRSA is considered insensitive to the bactericidal effect of many penicillin and cephalosporin antibiotics.[16] It is worth to mention that new broad spectrum cephalosporins were introduced to the clinical application with a significant activity against MRSA. These new cephalosporins, ceftaroline and ceftobiprole, can bind and inhibit PBP2a.[17,18]

Despite the introduction of novel antimicrobials, the rate of new antibiotics development is still slower than the speed by which bacterial resistance is emerging to pharmacotherapy. In this regard, most of the available antibiotics were derived and developed from microbial sources but herbal sources have not yet been explored.[19] Therefore, the aim of this *in silico* study is to virtually screen a library of Traditional Chinese Medicine (TCM) against the active site of PBP2a crystal. For this purpose, both molecular docking and dynamics simulation were implemented in the screening process to identify new phytomedicines capable of inhibiting MRSA. Then, the agar well diffusion method was used to evaluate the antimicrobial activity of final hits against MRSA cultured colonies.

MATERIALS AND METHODS

Setting up virtual screening outlines

The main stages of this computational project are summarized in Figure 1. In brief, a library of Traditional Chinese Medicine (TCM) was first screened by docking approach against target crystal of MRSA PBP2a. Then, the best ten phytomedicines with least docking energy were subjected to a prediction of the chemical, pharmacokinetics and toxicity characteristics. After that, the selected ten phytomedicines were submitted to molecular dynamics simulation. In this step, both ligand proximity to PBP2a active site and binding energy were reported as a function of simulation duration. Finally, only those hit compounds with best dynamics simulation results were evaluated *in vitro* for their antimicrobial activity against cultured MRSA.

Molecular docking

For this step, the DrugRep server was employed to carry out molecular docking of the TCM library of compounds against MRSA PBP2a enzyme.[20] The DrugRep online server

harnesses both AutoDock Vina version 1.1.2 and AutoDockTools (ADT) version 1.5.6 to perform docking operations.[21,22] It is also worth noting that the used TCM library contains 2,390 compounds from about 800 traditional Chinese medicines.[20] These TCM compounds were screened against only chain A of PBP2a crystal with PDB code 4DKI,[15] UCSF Chimera version 1.18 was used to extract only chain A and remove co-crystalized ligand from target crystal.[23] During the docking process, the following coordinates of the target active site were applied: X = 27, Y = 28 and Z = 81. While the docking grid box size was 20*20*20 Angstrom. After the complete of docking step, only the best ten phytomedicines with least energy of binding were picked for further evaluation. For each docking complex of these ten phytomedicines, the orientation with the least binding energy pose was visualized and assessed by the protein-ligand interaction profiler (PLIP) web tool.[24] Additionally, the following web-based servers: Molsoft L.L.C, SwissADME, pkCSM and ProTox 3.0 were used to predict several chemical, pharmacokinetics and toxicological properties for the best ten phytochemical compounds.[25–28]

Molecular dynamics (MD) simulation

In this step, the best ten phytochemical compounds were subjected into two consecutive runs of MD simulation for durations of 25 and 50 nanoseconds respectively. For this purpose, YASARA Dynamics version 20.12.24 was used to execute these two runs of MD study.[29] During these MD simulations, the ligand proximity to PBP2a active site was estimated as Root Mean Square Deviation (RMSD). In the first MD run, each docking complex with least binding energy pose was subjected to simulation for 25 nanoseconds. Then, only these phytomedicine-PBP2a complexes with mean ligand proximity RMSD value of no more than 4 Angstrom were submitted to the second MD run. Throughout the second MD simulation of 50 nanoseconds, the cutoff value of no more than 4 Angstrom for mean ligand proximity RMSD was used again to choose the final hit compounds. Additionally, in the second MD simulation, average Molecular Mechanics-Poisson Boltzmann Surface Area (MM-PBSA) binding energy was computed by employing AMBER14 force field.[30] The detailed procedure and options employed to execute these two MD simulations are the same as what we have applied in previously published *in silico* studies.[31–33] Briefly, NaCl was used during this simulation with a concentration of 0.9% and an extra amount of either sodium or chloride ions was applied to ensure the neutralization of phytomedicine-PBP2a complex. Also, the hydrogen bonding network was optimized to increase solute stability. Then, pKa anticipation was applied to fine-tune the protonation state of protein residues at physiological pH of 7.4. For this MD study, AMBER14 forcefield was used for the solute while AM1BCC and GAFF2 forcefields were applied for the ligand. Moreover, water molecules were subjected to TIP3P forcefield.[30,34,35]

***In vitro* antimicrobial activity**

Only those hits, with the best MD simulation results, were then subjected to the *in vitro* assessment of their antimicrobial activity against cultured MRSA. In this step, the agar well diffusion approach was used to evaluate the *in vitro* anti-MRSA activity for the final hits.[36] For this purpose, a standard strain of MRSA (ATCC 33591) was cultivated first in Mueller-Hinton II broth (Sigma-Aldrich, India) and incubated overnight at a temperature of 37 °C. After that, the turbidity of the growth was set to 0.5 McFarland standard. Then, 100 µL of the MRSA growth was transferred and spread into a 150 mm Petri dish filled with Mueller-Hinton II agar (Sigma-Aldrich, India). By using a metal cork-borer, five wells of 6 mm diameter were made in the agar.

A 0.5 mg/ mL solution of each selected hit compound was prepared by using dimethyl sulfoxide (DMSO) solvent. These selected compounds were purchased from MedChemExpress web store. Then, 100 µL of each prepared solution was filled into one of the wells and incubated overnight at 37 °C. For this test, the solvent DMSO was employed as a negative control and this test was repeated as a duplicate.

RESULTS

In Table 1, the top ten hits are listed for the structure-based virtual screening of TCM library against MRSA PBP2a monomer. These best hits, in Table 1, are ranked based on their least energy of binding to PBP2a chain A. As seen in this table, the natural sources and pharmacological effects for each hit compound are enlisted.

Then, various chemical characteristics were predicted for these ten phytomedicines. The predicted chemical properties for these ten compounds along with their docking energy are presented in Table 2. As can be noted from the chemical formula in Table 2, the two phytomedicines, daurisoline and isoliensinine, are alkaloids while other compounds are polyphenolic with no nitrogen-based structure. Moreover, the only hit compound that comply with Lipinski's rule of five is silydianin. However, the anticipated polar surface area (PSA) for silydianin is still greater than 140 squared Angstrom.

After that, several pharmacokinetics and toxicological features were anticipated for these top phytomedicines. These predicted features are presented along with drug-likeness score for each compound in Table 3. According to Table 3, all these compounds have good drug-likeness score except sciadopitysin, mirificin, plantamajoside. Also, all the listed hits have good or moderate water solubility with the exception of daurisoline, sciadopitysin and isoliensinine. As such, these three compounds with poor water solubility are also predicted to have high intestinal absorption but low volume of distribution. Finally, the only hit that may have a mutagenic potential in Table 3 is the alkaloid daurisoline according to AMES toxicity; while the lowest median lethal dose (LD_{50}) was reported for mirificin and lithospermic acid. Consequently, these three hit compounds: daurisoline, mirificin and lithospermic acid are expected to be unsafe.

The results of MD simulation study are shown in Table 4 for the top ten compounds. These tabulated results are reported for the two runs of 25 and 50 nanoseconds intervals. When considering the ligand proximity to PBP2a active site, only four phytomedicines were able to record mean ligand movement RMSD value of less than 4 Angstrom during 25 nanoseconds interval. These four phytomedicines with low mean ligand movement RMSD are: sciadopitysin, mirificin, plantamajoside, lithospermic acid. Then, only these four compounds were subjected to the second MD run of 50 nanoseconds. During this extended simulation run, only three compounds were able to maintain mean ligand movement RMSD value that

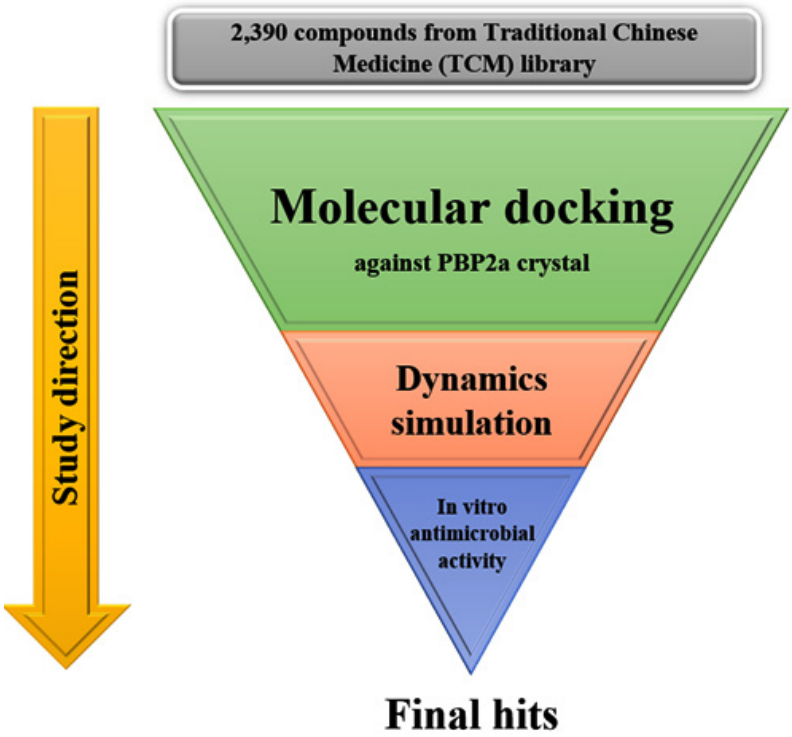

Fig. 1. A schematic representation of main stages of this *in silico* study

didn't exceed 4 Angstrom. These three compounds are: sciadopitysin, plantamajoside and lithospermic acid as seen in Table 4. Interestingly, the best average MM-PBSA binding energy was computed for the compound sciadopitysin of -24.09 Kcal/ mol during 50 nanoseconds interval.

A detailed plot can be seen in Figure 2 for the ligand movement RMSD as a function of 50 nanoseconds simulation duration. It is very clear from this plot that both sciadopitysin and plantamajoside were able to record a close proximity to PBP2a active site during simulation, as compared to the other two compounds. Also, it is evident from the plot in Figure 2 that only plantamajoside was able to keep a ligand movement RMSD that did never exceed 4 Angstrom value.

A close view for the binding orientation and possible interactions for both sciadopitysin and plantamajoside with PBP2a active site can be seen in Figure 3 for the docking complexes. The

Table 1. A list for natural sources and pharmacological activities of the top hit compounds in this *in silico* screening study. These hits were ranked based on their minimum docking energy to enzyme crystal

No.	Hit name	Natural source	Pharmacological activity
1	Daurisoline	*Menispermum dauricum, Rhizoma Menispermi*	Antiarrhythmic, anticancer.[37,38]
2	Eriocitrin	*Citrus limon, Citrus sulcate, Citrus reticulata*	Antioxidant, anticancer.[39,40]
3	Sciadopitysin	*Ginkgo biloba, Taxus cuspidata*	Antioxidant, anti-inflammatory.[41]
4	Verbascoside	*Acanthus mollis, Arrabidaea pulchra, Buddleja brasiliensis*	Antioxidant, anti-inflammatory, neuroprotective.[42]
5	Narirutin	*Citrus sulcata, Citrus reticulata*	Antioxidant, anti-inflammatory, antituberculosis.[43,44]
6	Silydianin	*Silybium marianum*	Antioxidant, anticancer.[45,46]
7	Mirificin	*Puerariae Lobatae*	Potential tyrosinase inhibitor.[47]
8	Plantamajoside	*Plantago asiatica, Rehmannia glutinosa*	Anti-inflammatory, anticancer.[48,49]
9	Lithospermic acid	*Salvia miltiorrhiza*	Antioxidant, hepatoprotective.[50]
10	Isoliensinine	*Nelumbo nucifera*	Antioxidant, anti-inflammatory, anticancer.[51]

Table 2. Chemical features for the best ten compounds generated by *in silico* screening of traditional Chinese medicine (TCM) library against chain A of penicillin-binding protein 2a (PBP2a). These hits were arranged based on their least docking energy to the transpeptidase crystal

No.	Compound name	Chemical formula	Docking score (Kcal/ mol)	M.W. (g/mol)	HBD	HBA	Log P	TPSA (Å^2)
1	Daurisoline	$C_{37}H_{42}N_2O_6$	-9.8	610.7	2	8	5.14	83.86
2	Eriocitrin	$C_{27}H_{32}O_{15}$	-9.3	596.5	9	15	-1.28	245.29
3	Sciadopitysin	$C_{33}H_{24}O_{10}$	-9.1	580.5	3	10	4.76	148.80
4	Verbascoside	$C_{29}H_{36}O_{15}$	-9.0	624.6	9	15	-0.60	245.29
5	Narirutin	$C_{27}H_{32}O_{14}$	-8.8	580.5	8	14	-1.06	225.06
6	Silydianin	$C_{25}H_{22}O_{10}$	-8.5	482.4	5	10	0.88	162.98
7	Mirificin	$C_{26}H_{28}O_{13}$	-8.3	548.5	8	13	-1.10	219.74
8	Plantamajoside	$C_{29}H_{36}O_{16}$	-8.2	640.6	10	16	-1.34	265.52
9	Lithospermic acid	$C_{27}H_{22}O_{12}$	-8.2	538.5	7	12	1.62	211.28
10	Isoliensinine	$C_{37}H_{42}N_2O_6$	-7.8	610.7	2	8	5.16	83.86

M.W.: molecular weight; HBD: hydrogen bond donor; HBA: hydrogen bond acceptor; Log P: logarithm of partition coefficient; TPSA: topological polar surface area; Å: angstrom.

careful evaluation of docking images can be helpful in explaining any difference in the *in silico* and *in vitro* activities of sciadopitysin and plantamajoside against PBP2a.

Finally, the *in vitro* anti-MRSA activity evaluation showed that both sciadopitysin and plantamajoside were effective during the agar well diffusion testing as presented in Figure 4. In this *in vitro* analysis, the measured zone of inhibition diameter was higher for sciadopitysin (12.5 ± 0.7 mm) as compared to plantamajoside (9.0 ± 1.4 mm).

DISCUSSION

The wide and incorrect use of antibiotics like penicillin compounds is driving the development of bacterial resistance to these drugs. Of these resistant strains, MRSA is considered a real public health threat that can increase both the

Table 3. A summary of drug-likeness score, pharmacokinetics, and toxicity characteristics for the top hit compounds. These hit compounds were listed according to their least docking energy to the PBP2a crystal

No.	Hit name	Drug-likeness	Pharmacokinetics			Toxicity	
			Water solubility (mg/ml)	Intestinal absorption (%)	VDss (L/Kg)	AMES toxicity	LD_{50} (mg/ Kg)
1	Daurisoline	1.68	7.13e-06 (poor)	89.92	0.17	Yes	1,180
2	Eriocitrin	1.13	2.98e-01 (soluble)	26.97	33.81	No	12,000
3	Sciadopitysin	0.13	6.93e-07 (poor)	98.32	0.05	No	4,000
4	Verbascoside	0.51	4.09e-02 (moderate)	32.12	179.89	No	5,000
5	Narirutin	1.06	3.26e-01 (soluble)	36.63	19.72	No	2,300
6	Silydianin	0.93	7.58e-02 (soluble)	91.34	16.98	No	10,000
7	Mirificin	0.20	2.57e+00 (soluble)	42.60	42.46	No	832
8	Plantamajoside	0.36	5.21e-02 (moderate)	14.69	9.66	No	5,000
9	Lithospermic acid	0.64	1.13e-02 (moderate)	13.32	0.78	No	25
10	Isoliensinine	1.71	7.13e-06 (poor)	89.94	0.17	No	1,180

VDss: steady state volume of distribution; LD_{50}: median lethal dose.

Table 4. A tabular summary for molecular dynamics (MD) simulation results of the top ten compounds

No.	Compound name	MD simulation interval		
		25 nanoseconds	50 nanoseconds	
		Mean ligand movement RMSD (Å)	Mean ligand movement RMSD (Å)	Average MM-PBSA binding energy (Kcal/ mol)
1	Daurisoline	5.82	-	-
2	Eriocitrin	4.04	-	-
3	Sciadopitysin	3.25	3.17	-24.09
4	Verbascoside	4.16	-	-
5	Narirutin	4.75	-	-
6	Silydianin	4.71	-	-
7	Mirificin	3.87	4.15	-40.74
8	Plantamajoside	2.88	3.00	-49.77
9	Lithospermic acid	3.08	3.77	-68.66
10	Isoliensinine	7.23	-	-

MD: Molecular dynamics; RMSD: Root mean square deviation; Å: Angstrom; MM-PBSA: Molecular mechanics-Poisson Boltzmann surface area.

length and cost of infection treatment.[11] Despite this health threat, the speed of developing new and effective antibiotics is still behind the rate of bacterial resistance emergence.[19] The mechanism behind MRSA resistance appears to be related to the bacterial expression of a membrane protein called PBP2a that has a low affinity to â-lactam antibiotics.[12] Consequently, the active site of the PBP2a enzyme represents a valuable molecular target towards the development of novel antibiotics against MRSA strain. Therefore, it was of our interest to computationally screen a library of herbal compounds against PBP2a monomer to identify potential inhibitors. Then, these identified hits were evaluated *in vitro* for their antimicrobial activity against MRSA.

When considering the pharmacological activities of the top hit compounds in Table 1 of the screening results, it is easy to note that many of the hit compounds have antioxidant, anti-inflammatory and anticancer effects. Also, it is expected that all the listed phytomedicines in Table 2 do have poor oral bioavailability due to the violation of the predicted chemical characteristics to Lipinski's rule of five and/ or Veber's rule.[52,53] Moreover, the predicted features in both Table 2 and Table 3 point to the fact that these top hits are hydrophilic with the exception of daurisoline, sciadopitysin and isoliensinine. The hydrophilic nature of these hit compounds can be deduced by the low partition coefficient and high polar surface area as predicted in Table 2. As a result, this predicted hydrophilicity will lead to better water solubility, higher volume of distribution and poor penetration through biological barriers for these hits as seen in Table 3.

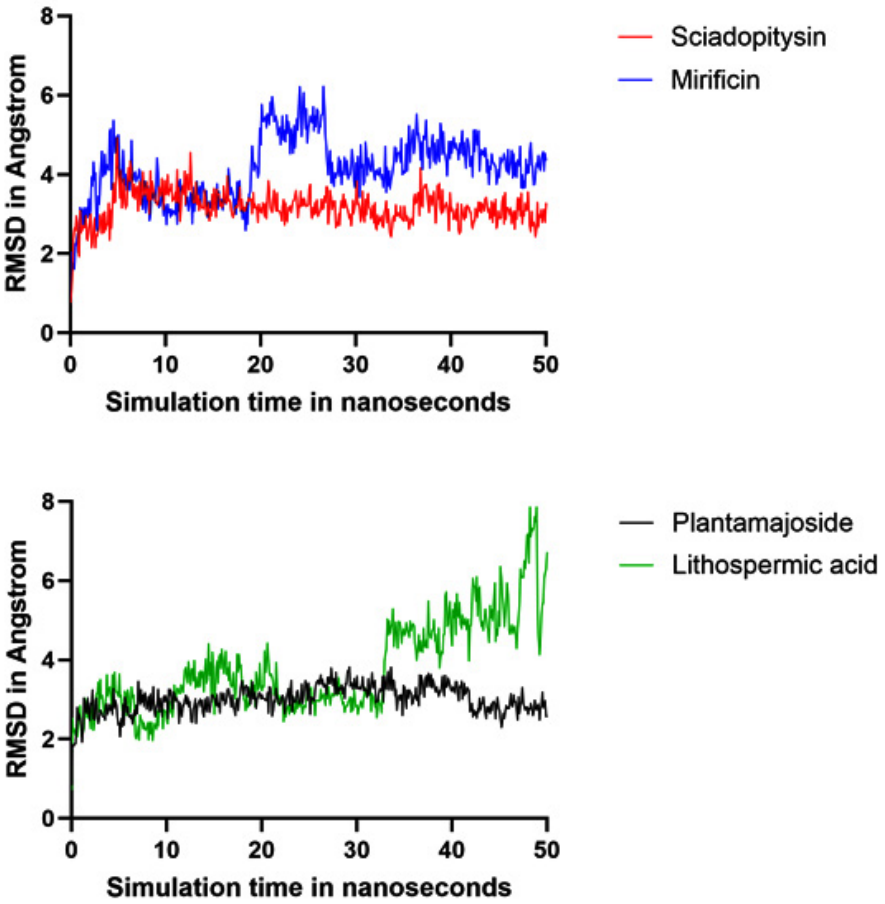

Fig. 2. A detailed plot of ligand movement RMSD versus MD simulation interval

It is well-known that low ligand movement RMSD during simulation study usually refer to closer proximity of the ligand to enzyme active site, thus stronger binding can be concluded.[54] Also, according to YASARA dynamics guideline, the more positive MM-PBSA binding energy points to stronger interaction between ligand and target crystal.[29] As such, both sciadopitysin and plantamajoside are expected to be potential inhibitors of PBP2a when considering ligand proximity and binding energy parameters in both Table 4 and Figure 2 throughout simulation interval.

Analysis of docking images in Figure 3 refers to the possibility that the two compounds, sciadopitysin and plantamajoside, occupied different locations within PBP2a active site but both of them were able to interact with the nucleophile Serine 403 residue by a hydrogen bond interaction. As noted from Figure 3, the compound plantamajoside is involved in higher number of hydrogen bond interactions with PBP2a active site

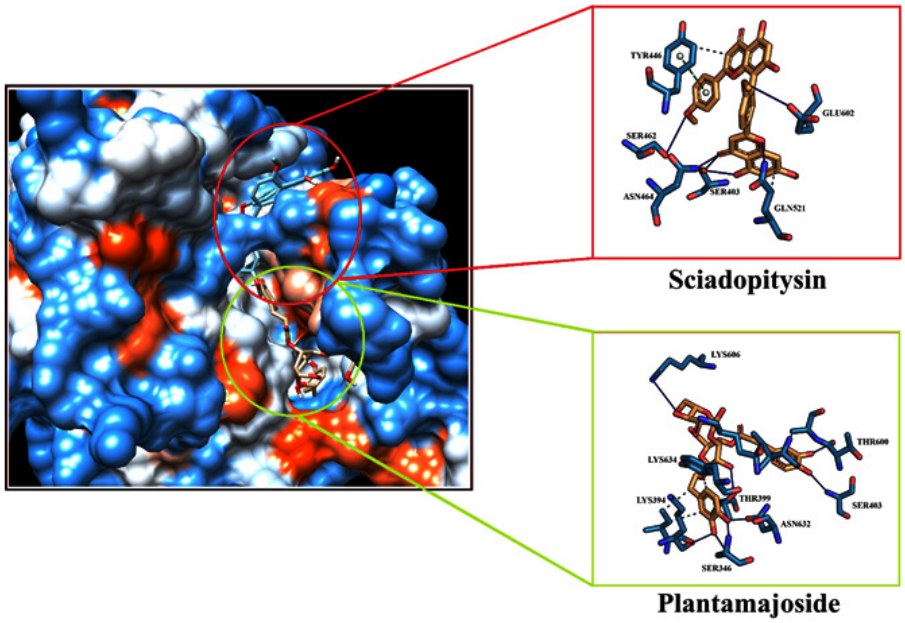

Fig. 3. An illustration for the binding orientation and potential interactions for both sciadopitysin and plantamajoside with PBP2a active site

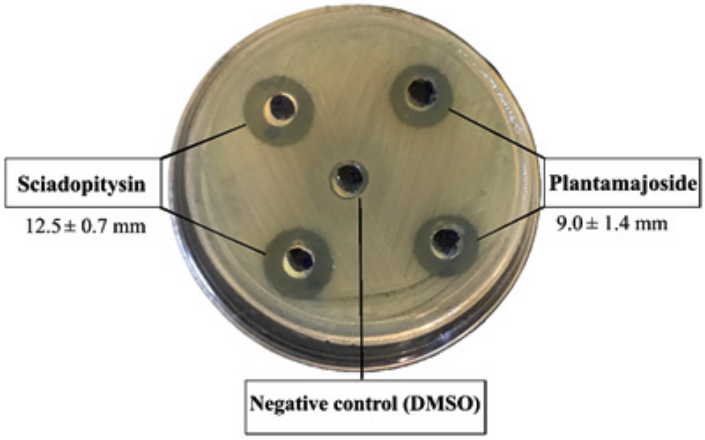

Fig. 4. Agar well diffusion assessment for sciadopitysin and plantamajoside against cultured MRSA

residues as compared to sciadopitysin. However, the binding location of sciadopitysin seems to be very similar to the co-crystalized ceftobiprole antibiotic. Also, the compound sciadopitysin is engaged in two hydrogen bond interactions with Serine 403 active site residue as compared to only one interaction in case of plantamajoside. Moreover, the length of these hydrogen bonds between sciadopitysin and Serine 403 active site residue is shorter than that predicted for plantamajoside (2.28 and 2.75 versus 3.27 Angstrom). Thus, despite the fact that plantamajoside can form many hydrogen bond interactions with transpeptidase active site residues but the binding location, orientation and length of the hydrogen bonds in case of sciadopitysin looks to be more favorable.

These *in silico* findings were then further confirmed *in vitro* by measuring the zone of inhibition diameter produced by these two phytomedicines against cultured MRSA. Records from agar well-diffusion experiment in Figure 4 points to the fact that the compound sciadopitysin can produce larger zone of inhibition as compared to plantamajoside.

CONCLUSION

In this *in silico* study, the phenolic compounds sciadopitysin and plantamajoside are predicted to be potential inhibitors of penicillin-binding protein 2a (PBP2a). Thus, these two phytomedicines can be used toward the development of new antibiotics against methicillin-resistant *Staphylococcus aureus* (MRSA). Docking study indicates that the energy of binding to PBP2a active site is -9.1 and -8.2 Kcal/ mol for sciadopitysin and plantamajoside respectively. As compared to sciadopitysin docking complex, the glycoside plantamajoside is involved in a higher number of hydrogen bonds with PBP2a active site residues. However, the flavonoid sciadopitysin is engaged in two hydrogen bonds with Serine 403 active site residue while plantamajoside is involved in only one hydrogen bond with this residue. Also, the length of these two hydrogen bonds formed by sciadopitysin is shorter when compared to plantamajoside. Then, molecular dynamics (MD) simulation points to the fact that these two compounds can maintain a close proximity to PBP2a active site throughout 50 nanoseconds interval. Further, the best MM-PBSA binding energy was calculated for sciadopitysin during simulation analysis. Finally, agar well diffusion study showed that both compounds are effective in inhibiting the growth of MRSA culture and the zone of inhibition was larger for sciadopitysin. However, the physicochemical properties of both sciadopitysin and plantamajoside should be modified to enhance compounds' drug-likeness score and pharmacokinetics.

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Conflict of interest

The author(s) do not have any conflict of interest.

Data Availability

This statement does not apply to this article.

Ethics Statement

This research did not involve human participants, animal subjects, or any material that requires ethical approval.

Informed Consent Statement

This study did not involve human participants, and therefore, informed consent was not required.

Clinical Trial Registration

This research does not involve any clinical trials

Authors' Contribution

Hasanain Abdulhameed Odhar: Conceptualization, Methodology, Data collection, Analysis, Writing – Original Draft; Zanan Abdulhameed Odhar: Writing – Review & Editing, Supervision; Mustafa Raed Muhi: Writing – Review, Supervision.

REFERENCES

1. Lee AS, De Lencastre H, Garau J, et al. Methicillin-resistant Staphylococcus aureus. *Nat Rev Dis Prim*. 2018;4.
2. Wertheim HFL, Melles DC, Vos MC, et al. The

role of nasal carriage in Staphylococcus aureus infections. *Lancet Infect Dis*. 2005;5(12):751-762.
3. Tong SYC, Davis JS, Eichenberger E, Holland TL, Fowler VG. Staphylococcus aureus Infections: Epidemiology, Pathophysiology, Clinical Manifestations, and Management. *Clin Microbiol Rev*. 2015;28(3):603.
4. Bush K, Bradford PA. Epidemiology of â-Lactamase-Producing Pathogens. *Clin Microbiol Rev*. 2020;33(2).
5. Bouiller K, Bertrand X, Hocquet D, Chirouze C. Human Infection of Methicillin-Susceptible Staphylococcus aureus CC398: A Review. *Microorganisms*. 2020;8(11):1-19.
6. Jaradat ZW, Ababneh QO, Sha'aban ST, Alkofahi AA, Assaleh D, Al Shara A. Methicillin Resistant Staphylococcus aureus and public fomites: a review. *Pathog Glob Health*. 2020;114(8):426-450.
7. Lakhundi S, Zhang K. Methicillin-Resistant Staphylococcus aureus: Molecular Characterization, Evolution, and Epidemiology. *Clin Microbiol Rev*. 2018;31(4).
8. Hasanpour AH, Sepidarkish M, Mollalo A, et al. The global prevalence of methicillin-resistant Staphylococcus aureus colonization in residents of elderly care centers: a systematic review and meta-analysis. *Antimicrob Resist Infect Control*. 2023;12(1).
9. Lv N, Kong Q, Zhang H, Li J. Discovery of novel Staphylococcus aureus penicillin binding protein 2a inhibitors by multistep virtual screening and biological evaluation. *Bioorg Med Chem Lett*. 2021;41.
10. Foster TJ. Antibiotic resistance in Staphylococcus aureus. Current status and future prospects. *FEMS Microbiol Rev*. 2017;41(3):430-449.
11. Zhen X, Lundborg CS, Zhang M, et al. Clinical and economic impact of methicillin-resistant Staphylococcus aureus: a multicentre study in China. *Sci Rep*. 2020;10(1).
12. Fishovitz J, Hermoso JA, Chang M, Mobashery S. Penicillin-Binding Protein 2a of Methicillin-Resistant Staphylococcus aureus. *IUBMB Life*. 2014;66(8):572.
13. Miyachiro MM, Contreras-Martel C, Dessen A. Penicillin-Binding Proteins (PBPs) and Bacterial Cell Wall Elongation Complexes. *Subcell Biochem*. 2019;93:273-289.
14. Wada A, Watanabe H. Penicillin-Binding Protein 1 of Staphylococcus aureus Is Essential for Growth. *J Bacteriol*. 1998;180(10):2759.
15. Lovering AL, Gretes MC, Safadi SS, et al. Structural Insights into the Anti-methicillin-resistant Staphylococcus aureus (MRSA) Activity of Ceftobiprole. *J Biol Chem*. 2012;287(38):32096.
16. Shalaby MAW, Dokla EME, Serya RAT, Abouzid KAM. Penicillin binding protein 2a: An overview and a medicinal chemistry perspective. *Eur J Med Chem*. 2020;199.
17. Steed ME, Rybak MJ. Ceftaroline: a new cephalosporin with activity against resistant gram-positive pathogens. *Pharmacotherapy*. 2010;30(4):375-389.
18. Kisgen J, Whitney D. Ceftobiprole, a Broad-Spectrum Cephalosporin With Activity against Methicillin-Resistant Staphylococcus aureus (MRSA). *Pharm Ther*. 2008;33(11):631.
19. Alhadrami HA, Hamed AA, Hassan HM, Belbahri L, Rateb ME, Sayed AM. Flavonoids as Potential anti-MRSA Agents through Modulation of PBP2a: A Computational and Experimental Study. *Antibiot (Basel, Switzerland)*. 2020;9(9):1-16.
20. Gan J hong, Liu J xiang, Liu Y, et al. DrugRep: an automatic virtual screening server for drug repurposing. *Acta Pharmacol Sin 2022 444*. 2022;44(4):888-896.
21. Trott O, Olson AJ. AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization and multithreading. *J Comput Chem*. 2010;31(2):455.
22. Morris GM, Ruth H, Lindstrom W, et al. AutoDock4 and AutoDockTools4: Automated Docking with Selective Receptor Flexibility. *J Comput Chem*. 2009;30(16):2785.
23. Pettersen EF, Goddard TD, Huang CC, et al. UCSF Chimera—a visualization system for exploratory research and analysis. *J Comput Chem*. 2004;25(13):1605-1612.
24. Adasme MF, Linnemann KL, Bolz SN, et al. PLIP 2021: expanding the scope of the protein-ligand interaction profiler to DNA and RNA. *Nucleic Acids Res*. 2021;49(W1):W530-W534.
25. Molsoft L.L.C.: Drug-Likeness and molecular property prediction. Accessed August 24, 2024. https://molsoft.com/mprop/
26. Daina A, Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Sci Rep*. 2017;7.
27. pkCSM. Accessed August 24, 2024. https://biosig.lab.uq.edu.au/pkcsm/prediction
28. Banerjee P, Kemmler E, Dunkel M, Preissner R. ProTox 3.0: a webserver for the prediction of toxicity of chemicals. *Nucleic Acids Res*. 2024;52(W1):W513-W520.
29. Krieger E, Vriend G. YASARA View - molecular graphics for all devices - from smartphones to workstations. *Bioinformatics*. 2014;30(20):2981-

2982.
30. Wang J, Wolf RM, Caldwell JW, Kollman PA, Case DA. Development and testing of a general amber force field. *J Comput Chem*. 2004;25(9):1157-1174.
31. Odhar HA, Hashim AF, Ahjel SW, Humadi SS. Molecular docking and dynamics simulation analysis of the human FXIIa with compounds from the Mcule database. *Bioinformation*. 2023;19(2):160-166.
32. Odhar HA, Hashim AF, Humadi SS, Ahjel SW. Ligand-Based Virtual Screening Of Fda-Approved Drugs To Identify New Inhibitors Against Lactate Dehydrogenase Enzyme Of Malaria Parasites. *Int J Appl Pharm*. 2024;16(1):255-260.
33. Odhar HA, Hashim AF, Ahjel SW, Humadi SS. Virtual Screening Of Fda-Approved Drugs By Molecular Docking And Dynamics Simulation To Recognize Potential Inhibitors Against Mycobacterium Tuberculosis Enoyl-Acyl Carrier Protein Reductase Enzyme. *Int J Appl Pharm*. 2024;16(1):261-266.
34. Maier JA, Martinez C, Kasavajhala K, Wickstrom L, Hauser KE, Simmerling C. ff14SB: Improving the Accuracy of Protein Side Chain and Backbone Parameters from ff99SB. *J Chem Theory Comput*. 2015;11(8):3696-3713.
35. Jakalian A, Jack DB, Bayly CI. Fast, efficient generation of high-quality atomic charges. AM1-BCC model: II. Parameterization and validation. *J Comput Chem*. 2002;23(16):1623-1641.
36. Balouiri M, Sadiki M, Ibnsouda SK. Methods for in vitro evaluating antimicrobial activity: A review. *J Pharm Anal*. 2016;6(2):71.
37. Liu Q, Mao X, Zeng F, Jin S, Yang X. Effect of daurisoline on HERG channel electrophysiological function and protein expression. *J Nat Prod*. 2012;75(9):1539-1545.
38. Huang K, Chen Q, Deng L, Zou Q, Min S. Daurisoline Inhibiting Tumor Angiogenesis and Epithelial-Mesenchymal Transition in Bladder Cancer by Mediating HAKAI Protein Stability. *Iran J Pharm Res IJPR*. 2022;21(1):129798.
39. Xu J, Ma L, Fu P. Eriocitrin attenuates ischemia reperfusion-induced oxidative stress and inflammation in rats with acute kidney injury by regulating the dual-specificity phosphatase 14 (DUSP14)-mediated Nrf2 and nuclear factor-êB (NF-êB) pathways. *Ann Transl Med*. 2021;9(4):350-350.
40. Wang Z, Zhang H, Zhou J, et al. Eriocitrin from lemon suppresses the proliferation of human hepatocellular carcinoma cells through inducing apoptosis and arresting cell cycle. *Cancer Chemother Pharmacol*. 2016;78(6):1143-1150.
41. Ijaz MU, Qamer M, Hamza A, et al. Sciadopitysin mitigates spermatological and testicular damage instigated by paraquat administration in male albino rats. *Sci Rep*. 2023;13(1).
42. Alipieva K, Korkina L, Orhan IE, Georgiev MI. Verbascoside—a review of its occurrence, (bio) synthesis and pharmacological significance. *Biotechnol Adv*. 2014;32(6):1065-1076.
43. Funaguchi N, Ohno Y, La BLB, et al. Narirutin inhibits airway inflammation in an allergic mouse model. *Clin Exp Pharmacol Physiol*. 2007;34(8):766-770.
44. Sahu PK, Mohapatra PK, Rajani DP, Raval MK. Structure-based Discovery of Narirutin as a Shikimate kinase Inhibitor with Anti-tubercular Potency. *Curr Comput Aided Drug Des*. 2020;16(5):523-529.
45. Vostálová J, Tinková E, Biedermann D, Kosina P, Ulrichová J, Svobodová AR. Skin Protective Activity of Silymarin and its Flavonolignans. *Molecules*. 2019;24(6).
46. Koltai T, Fliegel L. Role of Silymarin in Cancer Treatment: Facts, Hypotheses, and Questions. *J Evidence-based Integr Med*. 2022;27.
47. Liu H, Zhu Y, Wang T, Qi J, Liu X. Enzyme-Site Blocking Combined with Optimization of Molecular Docking for Efficient Discovery of Potential Tyrosinase Specific Inhibitors from Puerariae lobatae Radix. *Molecules*. 2018;23(10).
48. Wu H, Zhao G, Jiang K, et al. Plantamajoside ameliorates lipopolysaccharide-induced acute lung injury via suppressing NF-êB and MAPK activation. *Int Immunopharmacol*. 2016;35:315-322.
49. Zuo X, Li L, Sun L. Plantamajoside inhibits hypoxia-induced migration and invasion of human cervical cancer cells through the NF-êB and PI3K/akt pathways. *J Recept Signal Transduct Res*. 2021;41(4):339-348.
50. Chan KWK, Ho WS. Anti-oxidative and hepatoprotective effects of lithospermic acid against carbon tetrachloride-induced liver oxidative damage in vitro and in vivo. *Oncol Rep*. 2015;34(2):673-680.
51. Cheng Y, Li HL, Zhou ZW, et al. Isoliensinine: A Natural Compound with "Drug-Like" Potential. *Front Pharmacol*. 2021;12.
52. Lipinski CA, Lombardo F, Dominy BW, Feeney PJ. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Adv Drug Deliv Rev*. 2001;46(1-3):3-26.
53. Veber DF, Johnson SR, Cheng HY, Smith BR, Ward KW, Kopple KD. Molecular properties that influence the oral bioavailability of drug candidates. *J Med Chem*. 2002;45(12):2615-

2623.
54. Odhar HA, Ahjel SW, Albeer AAMA, Hashim AF, Rayshan AM, Humadi SS. Molecular docking and dynamics simulation of FDA approved drugs with the main protease from 2019 novel coronavirus. *Bioinformation*. 2020;16(3):236-244.