In silico screening for seed compounds as potential inhibitors of quorum sensing regulator lasR of *Pseudomonas aeruginosa*

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Abstract:
Introduction: Quorum sensing (QS) is a mechanism used by many bacteria to synchronize their collective behavior when reaching a sufficient high cell density,QS inhibitors (QSIs) aim at disabling the QS molecular signaling machinery within a bacterial pathogen, effectively rendering cells incapable of sensing the neighboring cell and consequently modifying the regulation of genes As a consequence, QSI modifies the regulation of genes such as biofilm formation, the production of secondary metabolites, and the expression of disease-causing virulence factors. The aim of this study is to identify the QS and biofilm inhibitors through computational analysis of activity compounds against *Pseudomonas aeruginosa.*
Methods: The molecular studies of LasR of *Pseudomonas aeruginosa*, lasR pseudomonas aeruginosa and the compounds were carried out in Schrodinger maestro software version 9.2. The binding affinity of the compound was retrieved from PubChem. Similarly, LasR proteins were retrieved from the Data Bank. In the crystal structure, missing residues are added and water molecules around the receptor were removed using the protein preparation wizard. The crystal structure, active site, grinds generation were adjusted with co-crystallized compounds.
Result: In silico docking study was performed to screen the interaction and binding affinity of 10, 12-dacasadiyndioc acid with QS signal receptor of LasR protein. The docking study revealed that the 10, 12-Docasadiyndioic acid potentially binds with LasR protein and a finding affinity of -8.1 Kcal/mol.
Conclusion:10, 12- Docasadiyndioic acid with highest binding affinity for LasR protein possessing strong inhibitory binding interaction. Hence, we concluded that 10, 12- Docasadiyndioic acid could serve as anti-QS and anti-biofilm molecules for treating Pseudomonas aeruginosa infection the 10, 12- Docasadiyndioic acid analogs will be further experimentally validated therapeutic strategies in near

Kew words: *Pseudomonas aeruginosa, Virulence, Docking study, In silico, LasR, Environment, innovative technique, eco friendly.*

Introduction:

Quorum sensing (QS) is a mechanism in which bacteria ability to synchronize their collective behavior when reaching the threshold concentration. Signaling machinery within a bacterial pathogen, effectively rendering cells incapable of sensing the neighboring cell and consequently modifying the regulation of genes (Ding *et al.*, 2011). Quorum sensing is regulate and control the biofilm formation, the production of secondary metabolites, and the expression of disease-causing virulence factors (McKenna, 2013),The major issue is that while the QS inhibition does not directly kill bacteria or stop bacteria from growing like conventional broad-spectrum antibiotics it does, however, alter the behavior of targeted pathogens by modifying the expression levels of QS-regulated genes. These changes are likely to influence the intra- and inter-strain interactions. As a result, QS inhibition can introduce changes into the microbiome by redistributing the competitive advantage during the development of a complex community.(Miller and Bassler, 2001; McKenna, 2013), *Pseudomonas aeruginosa*, is a common human pathogen that can cause infections in cystic fibrosis (CF) patients. In *P. aeruginosa*, the QSI pressure can lead to the selection of QS-negative strains. In patients with CF, loss of QS signaling is associated with chronic infection and increased growth rates(Passador *et al.*, 1993). QS-regulated virulence determinants such as rhamnolipids and alginate play a key role in establishing *P. aeruginosa* colonization in the CF lung.(Bjarnsholt *et al.*, 2005), Bacteria are social organisms and they interact with each other, which according to some researchers are similar to those performed by insects, vertebrates and humans. The term QS is used to describe bacterial cell to cell communication process, which involves the production, detection and response to extracellular signalling molecules called AIs. Once increases the threshold concentration of these signalling molecules are directly proportional to the population of bacteria as a result in turn use this information to alter a number of factors. (Junker and Clardy, 2007).

Virulence of *P. aeruginosa* is regulated by QS to overcome activation of the host's immunity, by deleting the production of virulence factors until the bacteria population raises sufficiently to cause infection. The cellular processes regulated by QS are symbiosis, transfer of conjugative plasmids, sporulation, antimicrobial peptide synthesis, regulation of virulence, and biofilm formation.(Wu *et al.*, 2004; Junker and Clardy, 2007). Biofilm formation is controlled and synchronized by various environmental and genetic factors. Most of the Gram negatives use QS systems which control the bacterial mobility, cell membrane proteins, and extracellular polysaccharides (Ishida *et al.*, 2007). Our team has extensive knowledge and research experience that has translate into high quality publications(Krishnamurthy *et al.*, 2009; Abdul Wahab *et al.*, 2017; Eapen, Baig and Avinash, 2017; Ravindiran and Praveenkumar, 2018; Subramaniam and Muthukrishnan, 2019; Anita *et al.*, 2020; Kumar *et al.*, 2020; Rajasekaran *et al.*, 2020; Arumugam, George and Jayaseelan, 2021; Dhanraj and Rajeshkumar, 2021)

The aim of study is to identify the QS and biofilm inhibitors through computational analysis of activity compounds against *Pseudomonas aeruginosa.*

Materials and method:

The molecular studies of LasR of Pseudomonas aeruginosa, lasR pseudomonas aeruginosa and the compounds were carried out in Schrodinger maestro software version 9.2. The binding affinity of the compound was retrieved from PubChem. Similarly, LasR proteins were retrieved from Data Bank. In the crystal structure, missing residues are added and water molecules around the receptor were removed using the protein preparation wizard. The crystal structure, active site, grinds generation were adjusted with co-crystallized compounds. The chemical compounds energy minimizing, rotatable bonds and hydrogen bond optimization was executed with OPLS force field. Interaction of receptor and ligands were prepared using mastero Schrodinger suite. Docking of all compounds with a receptor was consequently performed using a glide module to analyze the conformations of the protein-ligand complex.

Results and Discussion

LasR, *P.aeruginosa* are the target genes which are responsible for the QS signaling mechanism and biofilm formation. In silico docking study was performed to screen the interaction and binding affinity of 10, 12- dacasadiyndioc acid with QS signal receptor of LasR protein. The docking study revealed that the 10, 12- Docasadiyndioic acid potentially binds with LasR protein and a finding affinity of -8.1 Kcal/mol (Table 1). The CviR and compound interaction was presented as a 3D image (Fig. 1). (Venkatramanan *et al.*, 2020) reported that the bioactive compounds were docked and had the highest binding affinity of CviR receptors. In the similar study aromatic aldehyde, 5-hydroxymethylfurfural (5-HMF) at sub-inhibitory concentration was capable of modulating quorum sensing regulated phenotypes and inhibited biofilm formation in the *P. aeruginosa* PA01 as determined by *in silico* analysis.(Rajkumari *et al.*, 2019), Similarly, Annapoorani et, al (2012) have reported that rosmarinic acid and mangiferin were docked with lasR receptors. The bioactive compound inhibited biofilm formation and docking analysis showed that selected compounds bind with lasR protein(Annapoorani *et al.*, 2012).

In the recent study Quorum inhibitory potential of hydrocinnamic acid (HCA) has not been reported earlier and hence is the first report. HCA has potentially inhibited the quorum sensing signaling molecules and suppresses QS receptor genes of *P. aeruginosa*. *In silico* analysis showed the competitive binding nature of HCA against natural autoinducer molecules suggesting this natural molecule could have far-reaching application in the management of pathogenesis in humans.(Sharma *et al.*, 2019). In the similar study The *in vitro* results inferred impairment in the production of LasA protease, LasB elastase, chitinase, pyocyanin, exopolysaccharides and rhamnolipids. In addition, motility and biofilm formation by *P. aeruginosa* PAO1 was significantly altered.(Pattnaik *et al.*, 2018)

Table:1 Predicted docking score indicating the binding affinity of LasR protein with bioactive compound of 10,12- docasadiyndioic acid.

S.No.	Ligand	Name of the Compound	Binding affinity
1	lasr_544138	10,12- Docasdiyndioic acid	-8.1

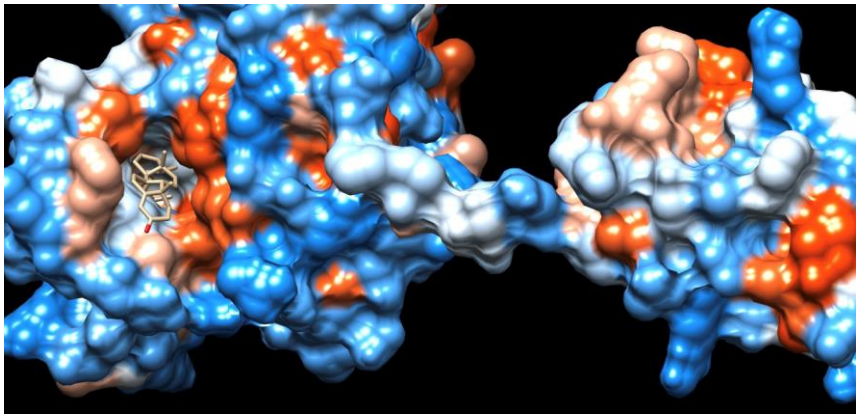

Figure 1: Schematic 3D image showing the binding of 10, 12-Dodecanedioic acid to the LasR receptor.

Conclusion:

10, 12- Docasadiyndioic acid with highest binding affinity for LasR protein possessing strong inhibitory binding interaction. Hence, we concluded that 10, 12- Docasadiyndioic acid could serve as anti-QS and anti-biofilm molecules for treating Pseudomonas aeruginosa infection the 10, 12-Docasadiyndioic acid analogs will be further experimentally validated therapeutic strategies in near.

Authors Contribution:

PSG did the study design, PSG and TMNP did *in silico* study, data collection was done by TMNP, TMNP wrote the manuscript and it was edited and revised by PSC, ASSG and JVP approved the submission of the manuscript.

Conflict of interest:

The author declares there is no conflict of interest.

Acknowledgement:

Author thanks Dr. Deepak Nalaawamy, Director, Saveetha Dental College and Hospital, Saveetha institute of medical and Technical science (SIMATS), for providing facilities and ideas to carry out this work.

SOURCE OF FUNDING: The present study was supported by the following agencies
Saveetha Dental College,
Technical science,
Saveetha University
Ravi Nursing Home

Reference:

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