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ORIGINAL ARTICLE

Anti-quorum sensing activity of *Forsythia suspense* extract against *Chromobacterium violaceum* by targeting CviR receptor

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Abstract

The increasing incidence of antimicrobial-resistant bacterial pathogens has focused researchers on quorum sensing inhibition strategies instead of those conventional approaches to fight bacterial infections. Anti-quorum sensing (QS) activity of aqueous extract from *Forsythia suspense* (FSE) was assessed, and its potential QS inhibition mechanisms were also analyzed. The minimal inhibitory concentration (MIC) of FSE to *Chromobacterium violaceum* 12472 is 0.5 mg mL^{-1}. Inhibition of QS-regulated violacein production and biofilm formation in *C. violaceum* 12472 by FSE occurred in a concentration-dependent manner at sub-MIC, with > 70.12 and > 85.31% inhibition at 0.25 mg mL^{-1}, respectively. *N*-Acyl homoserine lactones (AHLs) extracted from cultures of *C. violaceum* 31532 grown in the presence of FSE could not change the violacein production in *C. violaceum* 026, which indicated that FSE did not inhibit AHL synthesis. We also found that FSE cannot degrade AHLs. Finally, in silico molecular docking was conducted. The computed binding energy data suggested that components of *F. suspense* have a tendency to inhibit CviR with varying binding affinities and the energy score of Pinoresinol (− 26.02 kcal/mol) is higher than that of C6-HSL (− 16.09 kcal mol^{-1}). We concluded that FSE acts as an antagonist of bacterial quorum sensing by competing with AHL receptor binding site.

Keywords *Chromobacterium violaceum* · CviR · Docking · *Forsythia suspense* · Quorum sensing inhibitor·

Introduction

Antibiotics have been considered to be the most efficacious drugs for curing bacterial infectious diseases. As the quantity of antibiotics applied in human clinical and animal husbandry has increased over the past decades, numerous multiple drug-resistant bacterial strains have been isolated (Piddock 2017). The antibiotic-resistant bacteria population renders the antibiotics ineffective. Now, researchers are trying their best to find alternative approaches to avoid and treat bacterial infections (Rattanaumpawan et al. 2010), in which the quorum sensing (QS) inhibition pathway might be the novelty to solve the issue.

Microbes communicate with each other by creating and sensing diffusible, low-molecular weight, chemical-signaling molecules, auto-inducers (AI) which are termed as "quorum sensing," and these molecules enable gene regulation as a function of population density. QS frameworks direct physiological procedures, including the generation of virulence factors that are vital for pathogenic contamination, colonization, biofilm development, antimicrobial creation, motility, and furthermore interspecies microbial connections (Bassler and Losick 2006; Smith et al. 2006). The discovery of the QS system and its critical role in pathogenicity and survival has revealed a new target—a novel non-antibiotic way to control bacterial infections. It has been proposed that inactivating the QS arrangement of a pathogen can bring about a critical diminishing in harmfulness factor creation (Tang and Zhang 2014). Phytochemicals are one of the most used alternative therapeutic agents which can inhibit bacterial quorum sensing, thus controlling bacterial diseases without empowering the presence of safe strains; examples of such phytochemicals are epigallocatechin gallate from green tea, catechin and naringenin from *Combretum albiflorum*, allicin and ajoene from *Allium sativum*, and methyl eugenol from *Cuminum cyminum* (Ta and Arnason 2015; Borges et al. 2016; Subramaniyan et al. 2016; Musthafa et al. 2017).

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We have previously reported that *Forsythia suspense* extracts can inhibit the QS-controlled violacein production in *Chromobacterium violaceum* (Zhang and Chu 2017). The motivation behind this investigation was to assess the antimicrobial and antagonism effects QS has on *Forsythia suspense* and, moreover, to survey its component impact on microscopic organism correspondence. The discoveries of this examination could make ready for additionally itemized investigation of *Forsythia suspense* and for its more extensive application as QS inhibitor.

Materials and methods

Preparation of crude *Forsythia suspense* extracts

The dried fruits of *Forsythia suspense* were purchased from Tong Ren Tang Pharmaceutical Store (Nanjing, China). We utilized an altered convention to get aqueous concentrates ready as described by Hozumi et al. (1993), Luo and Zhang (2013), and Zhang et al. (2017). One hundred grams of the *F. suspense* fruits were grounded to powder and were extracted by maceration in 100 mL ultra-pure water at 100 °C for 2 h. The extract was centrifuged at 3000 rpm for 10 min, and the supernatant was filtered through a Whatman no. 1 filter paper and then concentrated on a rotary vacuum evaporator. The aqueous extract was then filtered with a 0.22-μm (pore size) filter and freeze-dried by cryodesiccation. The cryodesiccation powder was put away at − 70 °C and broke up in refined water to make supplies of 20 mg/mL before tests.

Strains and culture conditions

Chromobacterium violaceum bio-sensor framework strains were utilized as part of this study. The wild-sort strain of *C. violaceum* ATCC 12472 can create violacein, a purple shade, which is under the control of the QS framework. *C. violaceum* ATCC 12472 is utilized as a bio-sensor strain to distinguish potential majority detecting inhibitors. *C. violaceum* 026, another QS bio-sensor strain used in this investigation, was a two-fold smaller than usual Tn5 mutant derived from *C. violaceum* ATCC 31532, Kan^R, Hg^R, cvil::Tn5xylE, and a spontaneous Str^R. It is unable to synthesize AHLs, but it can respond to exogenous AHLs. The CviR receptor of *C. violaceum* 026 recognizes C6-AHL as the related gene and is touchy to short- and medium-chain-length AHLs (McClean et al. 1997). *C. violaceum* 31532 is a C6-AHL overly made and is used as a positive control for *C. violaceum* 026. All *C. violaceum* strains were benevolently provided by Professor Robert J.C. McLean, Texas State University, USA. The strains were developed on Luria-Bertani (LB) agar, with or without anti-infection agents, at 30 °C, and were kept up on LB agar plates at room temperature for a short stockpiling and for long haul stockpiling in LB broth containing glycerol at − 70 °C.

Bio-sensor bioassay for anti-QS activity

Anti-QS activity was resolved at focuses lower than the MIC esteems known as sub-inhibitory fixations. Minimal inhibitory concentration (MIC) values were determined by the broth micro-dilution technique (Metzler et al. 2004). The MIC value was determined after 24 h incubation by observation of the presence or absence of visible growth. The anti-QS potential of FSE was done by bio-sensor bioassay with the help of *C. violaceum* 12472 as a base strain (Taganna and Rivera 2008). Ten milliliters of delicate liquid LB agar (0.5% *w/v*) was inoculated with 100 μL of *C. violaceum* 12472 left overnight for growth in LB broth. The agar culture solution was immediately poured over the surface of the LB agar plates. Subsequently, 2 mm wells in diameter were punched through the agar and filled with 50 μL FSE at sub-MIC. The plates were incubated for 24 h at 30 °C and examined for violacein pigment production. The violacein inhibition property was evaluated by measuring the distance across of the yellowish obscure radiance display (showing bacterial development) without the purple violacein pigmentation of the bacterial lawn (demonstrating QS hindrance) encompassing the well. Sterilized water was utilized as a method of control. For quantitative QS inhibition assay, experiments were carried out according to a previously described method and evaluated spectrophotometrically at optical density of 580 nm (UV-1800; Shimadzu, Japan) (Blosser and Gray 2000).

Biofilm assays

Biofilm was evaluated by crystal violet binding assays, as previously described (Agarwala et al. 2014). Briefly, *C. violaceum* 12472 cells were incubated statically for 24 h at 30 °C in a test tube with the presence or absence of *Forsythia suspense* extract. After incubation, the mixture was removed and the tube was rinsed three times with phosphate buffer saline, fixed with formaldehyde (10%) for 10 min, and stained with 1 ml of 0.1% in ethanol crystal violet for 15 min. Stained cells were washed with deionized water to remove unbound crystal violet, and bounded crystal violet was eluted in 100% ethanol. The absorbance was measured at 650 nm. All tests were performed in three replicates.

Effect of FSE on modulation of AHL activity

This assay was performed as depicted by Chan et al. (2011) with some changes. Briefly, aliquots of C6-HSL (10 μg/μL) (Sigma-Aldrich, St. Louis, Missouri, USA) in ethanol were administered into a sterile tube, and the dissolvable dissipated to dryness under sterile condition. The FSE was added to the

tube to rehydrate the C6-HSL. The mixture was incubated at 37 °C for 4 h with delicate shaking in a hybridization stove. Then, 50 μL mixture was inoculated into the well on agar plate seeded with the biosensor strain *C. violaceum* 026 and incubated at 30 °C. Debasement of C6-HSL is clear by the loss of purple pigmentation appearing by *C. violaceum* 026. This test was repeated in three independent triplicates.

Effect of FSE on modulation of AHL synthesis

The impact of FSE on AHL synthesis was resolved utilizing a C6-HSL over-producing strain, *C. violaceum* 31532 and its mutant *C. violaceum* 026 (McLean et al. 2004). *C. violaceum* 31532 was cultured in the presence of FSE at sub-MIC for 24 h. AHL was removed from the cell-free supernatant (8 mL) utilizing dichloromethane (3:1 *v*/*v*) and vanished under a thin stream of nitrogen gas. For deciding the AHL action, the dried AHL portions were re-suspended in 70% methanol (20 μL) and added to crisp 10 mL LB medium immunized with biosensor strain *C. violaceum* 026 which reacted to exogenous AHL by delivering violacein. Violacein by the AHL portions in *C. violaceum* 026 was measured spectrophotometrically after hatching at 30 °C for 24 h as depicted before.

Effect of FSE on AHL receptor

C. violaceum 026 overnight culture grown in LB broth was balanced to an OD 600 of 0.01. A total of 100 μL of dilution was transferred into 20 mL sterile tube with 5 mL LB followed by addition of FSE solution to different concentrations at sub-MIC. Sterilized water was included as means of negative control. The mixture was incubated for 4 h at 30 °C with gentle shaking. A total of 20 μL of the mixture was then inoculated into the well on an agar plate with 10 μg/mL of C6-HSL, and inoculation continued for 16 h at 30 °C to check for violacein production (Maurer et al. 2015).

Docking

Molecular operating environment (MOE, 2014.0901, Chemical Computing Group, Montreal, Canada) computational software was applied for testing of 10 active components from *F. suspense* with them binding to the signal receptor CviR protein in *C. violaceum* activity. These 10 active components from *F. suspense* were selected as the references by Luo and Zhang (2013) and Zhang et al. (2017). All the compounds' structures used in the study were obtained from Chem spider. The protein CviR 3D structure data file was acquired from Protein Data Bank (PDB ID code 3QP1, resolution 1.55 Å). The structural topology of CviR consisted of two domains, a DNA-binding domain (DBD) combined to a ligand-binding domain (LBD) (Chen et al. 2011). Firstly, the local ligands and water molecules were evacuated using MOE. Hydrogen was added to all ligands and the receptor. Then, the ligands were additionally limited under the force field, and the coupling pocket was auto-locked as the coupling site of the agonist ligands. Lastly, docking of compounds and protein was performed. The docking pocket of CviR protein is site 2, 1: (ILE57 VAL59 MET72 VAL75 TYR80 TRP84 LEU85 TYR88 ASP97 ILE99 MET100 TRP111 PHE115 PHE126 ALA130 MET135 THR140 ILE153 SER155). Binding energy calculations and further analysis were needed. The termination gradient is 0.01 kcal/mol, and the maximal iteration is 1000 (Aliyu et al. 2016).

Confirmation of anti-QS activity of selected compounds

Anti-QS assay based on AHL-based analysis was performed using the bio-sensor bacterial strain CV12472. All of the selected ten compounds (pinoresinol, quercetin, dimethylmatairesinol, kaempferol, homovanillyl alcohol, forsythoside B1, forsythoside A, forsythoside D, rutin, and astragalin) were purchased from Shanghai Regal Biology Technology Co. Ltd. (Shanghai, China).

Statistical analysis

All assays were performed in triplicate, and one-way analysis of variance (ANOVA) was utilized to dissect the contrast between the treatments. $p < 0.01$ was considered as huge unless determined.

Results

Quorum sensing inhibition activity detected by qualitative agar diffusion assay

The MIC of FSE to *C. violaceum* 12472 is 0.5 mg/mL. A yellowish bacterial lawn was observed around the well containing FSE, indicating violacein production was inhibited and there was no AHL existing around the well when *C. violaceum* qualitative agar dissemination was examined with the sub-MIC FSE at 0.25 mg/mL (Fig. 1). There was no clear halo around the well, indicating there was no bactericidal effect of FSE at sub-MIC.

Violacein production quantification

The FSE demonstrated consequences for violacein creation for strain *C. violaceum* 12472. Figure 2a demonstrates the inhibitory impact of different concentrations of FSE on violacein generation. FSE showed a higher inhibitory effect with 70.12% at an increase of 0.25 mg/mL than 22.48% at 0.015625 mg/mL. Bacterial cell check performed on LB agar

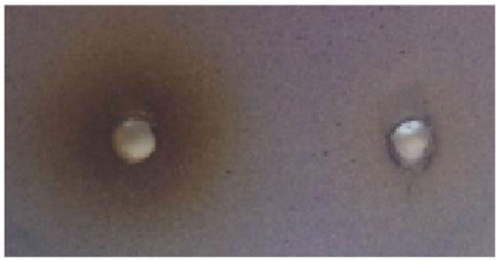


Fig. 1 Anti-QS properties of *F. suspense* extract. (**a**) 0.125 mg/ml *F. suspense* extract; (**b**) water

plates at 24 h brooding demonstrated no critical distinction in the quantity of colony-forming units (CFUs) at the FSE sub-MIC fixations tried (Fig. 2b).

Biofilm quantification

The effect of FSE on *C. violaceum* 12472 biofilm formation was evaluated (Fig. 3). Decreased biofilm formation was observed in FSE-treated *C. violaceum* 12472 (85.31% inhibition at a concentration of 0.25 mg/mL).

Degradation of C6-HSL by FSE

This test was done to affirm that the counter QS property of FSE was not because of the debasement of the C6-HSL. After being co-incubated with FSE, the C6-HSL was added to the wells on the plate inoculated with *C. violaceum* 026 to see the violacein production. FSE at sub-MIC did not influence the pigment production; this demonstrated that the tried fixations did not debase C6-HSL (Fig. 4).

Effect of FSE on AHL synthesis

The impacts of FSE on AHL synthesis were resolved utilizing a C6-HSL over-producing wild-type strain, *C. violaceum* 31532 and its mutant *C. violaceum* 026, which reacted to exogenous AHLs (McClean et al. 1997; McLean et al. 2004). Results demonstrated that the AHLs removed from *C. violaceum* 31532 culture supernatants could initiate violacein generation in the mutant *C. violaceum* 026. This showed that AHL synthesis was not influenced by the FSE.

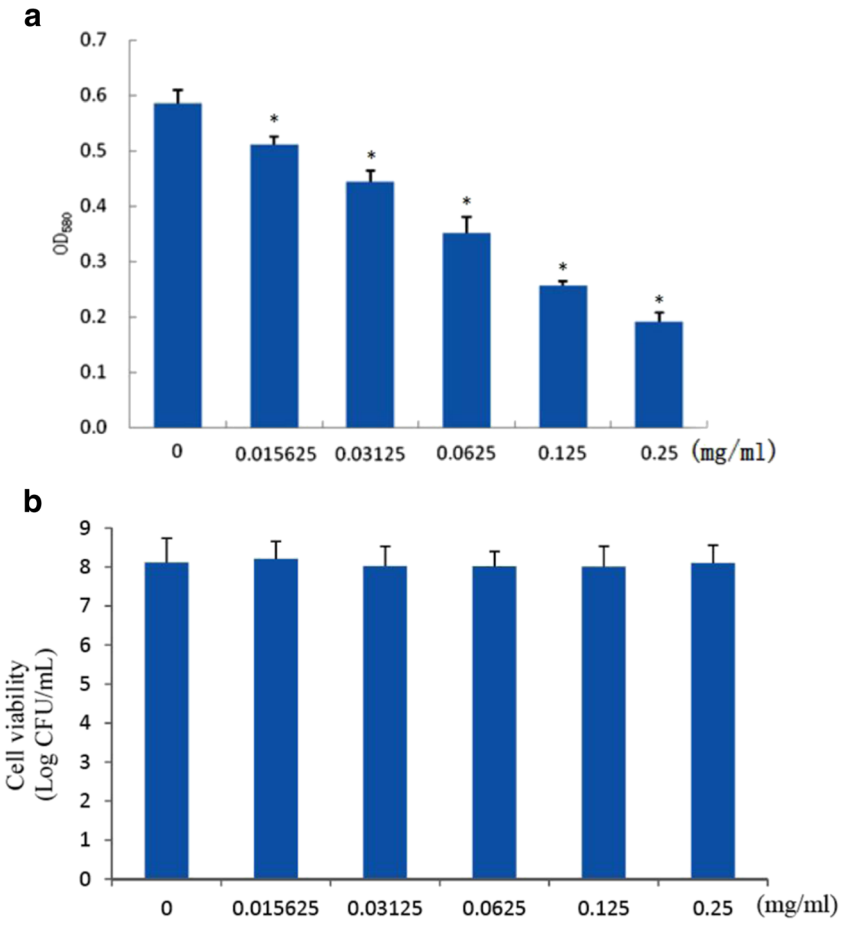


Fig. 2 (**a**) FSE inhibits *C. violaceum* 12472 violacein production in a concentration-dependent manner. The violacein production was measured spectrophotometrically and quantified by reading the OD values of the solution at 580 nm. Values are presented as mean ± SD, $n = 3$. (**b**) Cell viability of *C. violaceum* 12472 after incubation in LB broth with sub-MIC FSE treatments

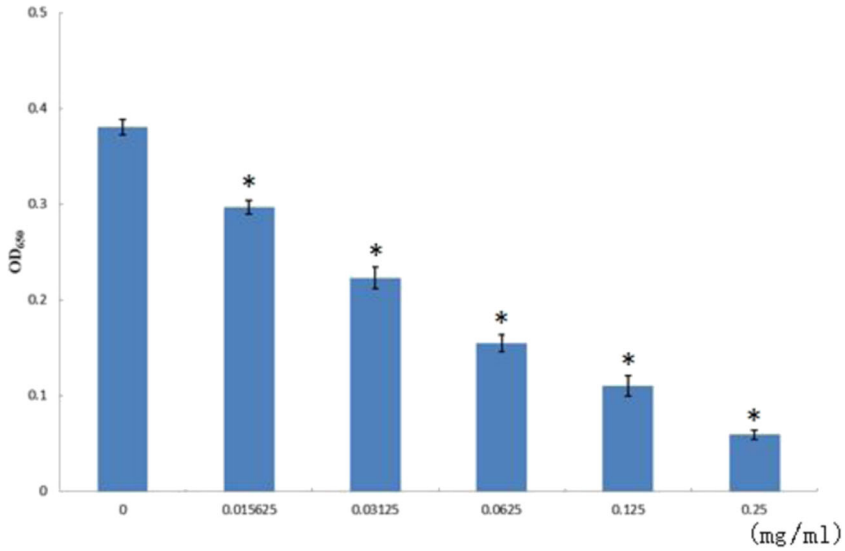


Fig. 3 Biofilm inhibition was performed with *C. violaceum* 12472 in triplicate with *F. suspense*. Error bars represent SD of three replicates

Effect of FSE on AHL receptor

Figure 5 showed that with the FSE concentration increased, violacein production decreased, which indicated FSE interference with the C6-HSL receptor.

Docking calculations and anti-QS activity of selected compounds

The analysis using MOE program recommended that three molecules among 10 competitors got higher scores than C6-HSL. The three molecules might play a major role in inhibiting QS systems. Ten active components from *F. suspense* and the energy score of interaction between the compounds and CviR were listed in Table 1. The energy score of pinoresinol (− 26.02 kcal/mol) is higher than that of C6-HSL (− 16.09 kcal/mol) and the other compounds. The crossed-domain conformation of pinoresinol:CviR and C6-HSL:CviR can be seen in Fig. 6a. A model of the pinoresinol:CviR complex suggested that pinoresinol mimics the lactone of AHLs and probably forms the canonical H-bond with Trp-111 and Ser-89 and so on (Fig. 6a). By comparison, the similarities between the two compounds' structures (Fig. 6b), the high-energy score recorded by pinoresinol may be due to the phenyl group's high complementarity with the binding site of CviR protein.

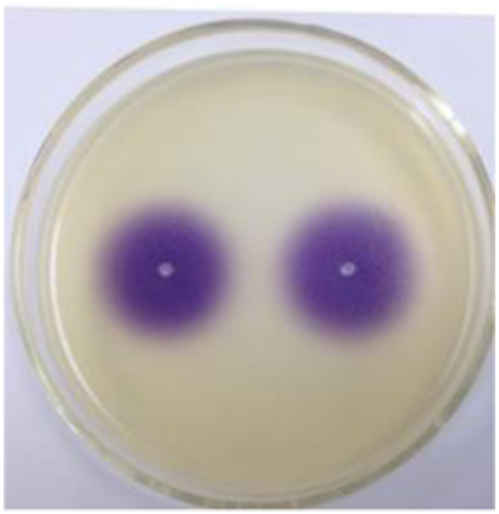


Fig. 4 *Forsythia suspense* extract did not degrade C6-HSL. (**a**) C6-HSL treated with *Forsythia suspense* extract at 0.125 mg/ml; (**b**) C6-HSL treated with sterilized water

Furthermore, in deciding the relationship between the assessed restricting scores and the real hindrance exercises, the QS restraint capacity of the selected mixes from *F. suspense* was assessed by testing the hindrance of violacein creation by *C. violaceum* ATCC 12472. All the compounds exhibited QS inhibitory activity at 0.25 mg/mL, and yellowish bacterial lawn was observed around the wells of all selected compounds after incubation at 28 °C for 24 h (Fig. 7).

Discussion

As pointed out in the "Introduction" section, the rapidly growing population of antibiotic-resistant bacteria has rendered antibiotics ineffective; therefore, the need for the discovery of a new way to fight bacteria is of much interest. QS directs an extensive variety of physiological procedures, including bioluminescence, biofilm development, level quality exchange, and destructiveness factors in bacterial pathogens (Ng and Bassler 2009). Therefore, QS has pulled in extensive enthusiasm as another objective for antimicrobial treatment and another option alternative to traditional antibiotics or disinfectants. In addition to potential medical applications, QS disruption may also be used in the agriculture and food industry. Most QS-disrupting agents known to date are effective at sub-MIC and

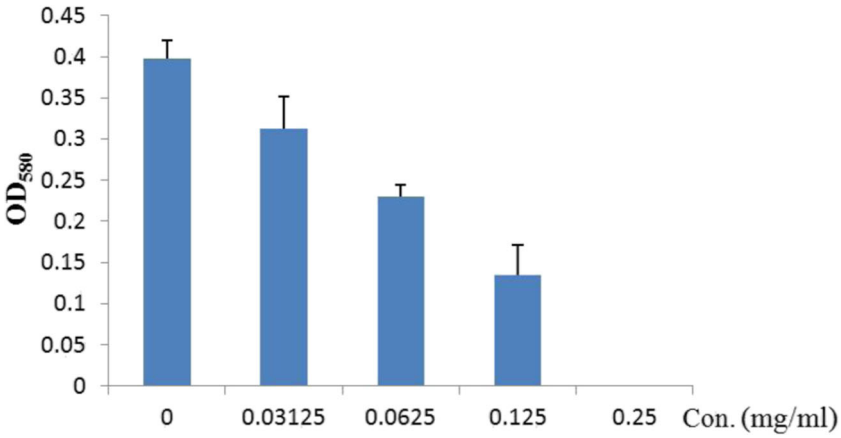


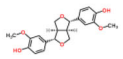
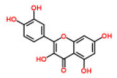
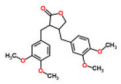
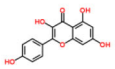
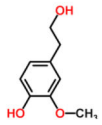
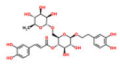
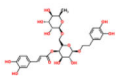
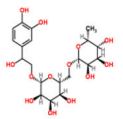
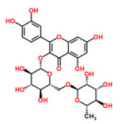
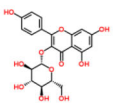
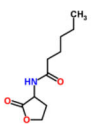
Fig. 5 Violacein production by *C. violaceum* 026 induced by AHL extracted from the culture supernatants of *C. violaceum* 31532 grown in the presence FSE different concentration at sub-MIC. Values are presented as mean ± SD, $n = 3$

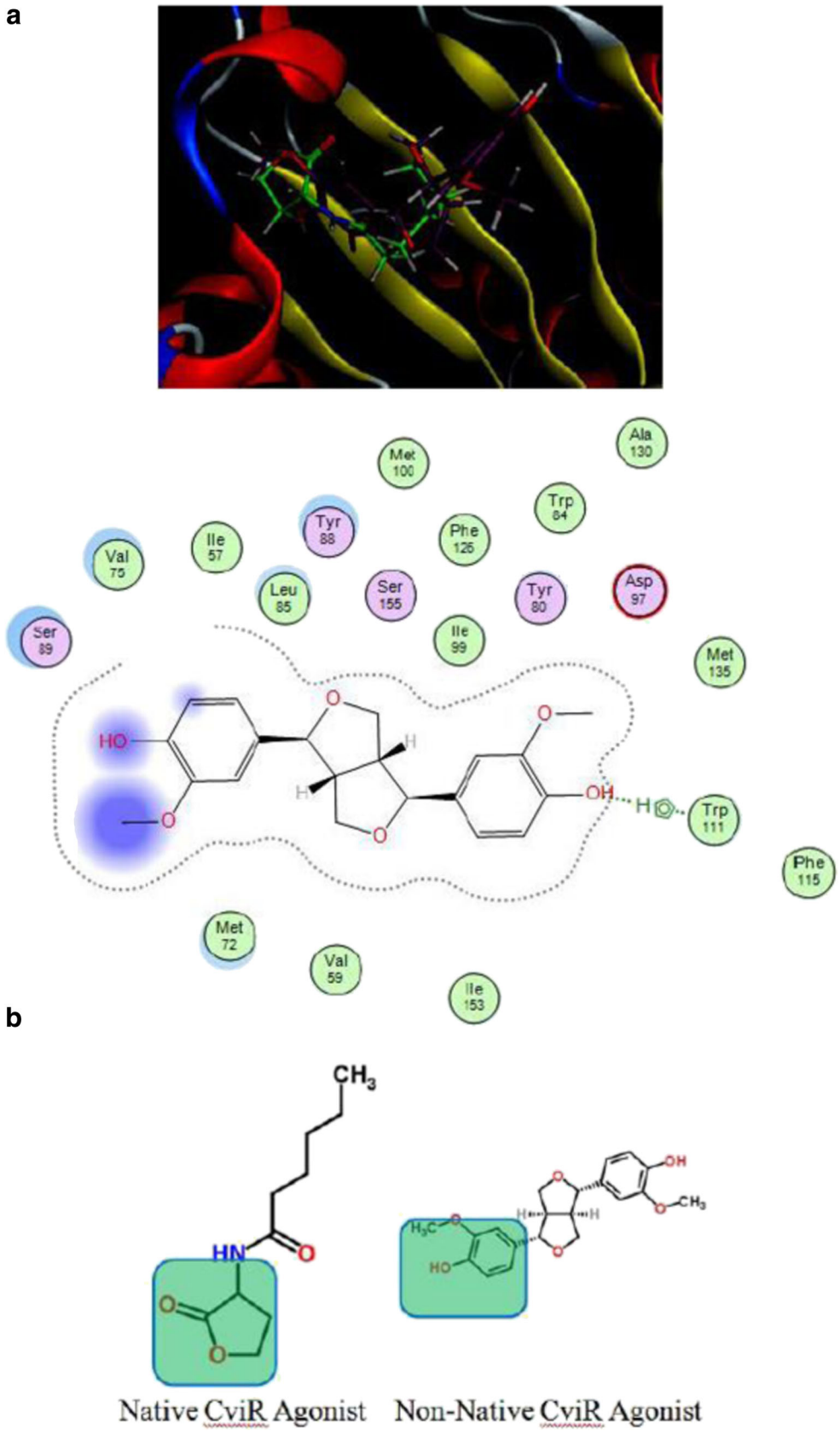
therefore do not disrupt growth or viability but interfere with bacterial pathogenicity. This approach has minimal effect on bacterial growth and therefore reduces the growth rate-induced selective pressure for the development of QS inhibitor resistance (García-Contreras et al. 2015). There are a few QS hindrance techniques accessible through which the procedure of majority detecting can be interfered with, which are as follows: (a) inhibition of the signal molecule biosynthesis or mimicking the signal molecules fundamentally by utilizing manufactured mixes as analogs of flag particles; (b) enzymatic destruction of signal molecules that will prevent them from gathering; and (c) interference with flag receptors or blockage of arrangement of signal molecule complex.

The extracts of the dried fruit from *Forsythia suspense* Vahl (Oleaceae) have been widely used for a long time as traditional Chinese medicines to treat infections, such as acute nephritis, erysipelas, and ulcers (Nishibe et al. 1982; Ozaki et al. 1997). It has been reported that the *F. suspense* extract exhibited potential antibacterial, antiviral, and anti-inflammatory effects (Zhang 2000). In this study, aqueous extract of *Forsythia suspense* was evaluated for its QS inhibitory activity. The results showed that FSE can inhibit QS-regulated violacein production and biofilm formation in *C. violaceum* without meddling with its development. We, therefore, deduced that the reduction in violacein creation was not caused by the restraint of bacterial development but instead by disturbance of the QS frameworks. Many natural compounds of plant origin are well-known for QS inhibitory ability (Koh et al. 2013). Examples include halogenated furanones extracted from *Delisea pulchra* (Martinelli et al. 2004), extracts of *Terminalia catappa* (Taganna et al. 2011), epigallocatechin gallate (Taganna and Rivera 2008), plant volatiles (Ahmad et al. 2015), and ellagic acid derivatives from *Terminalia chebula*; aqueous extracts from *Conocarpus erectus*, *Callistemon viminalis*, and *Bucida buceras* also had reported inhibitory effects on quorum sensing of *Pseudomonas aeruginosa* (Adonizio et al. 2008; Sarabhai et al. 2013). Zhao et al. found that an aqueous extract from the famous Chinese medicine, Yunnan Baiyao, inhibited the QS-related virulence, pyocyanin, protease of *Pseudomonas aeruginosa* (Zhao et al. 2013). Mihalik et al. made *Camellia sinensis* (green tea, GT) extracts suspend in distilled water, and results showed that GT extract can inhibit the outflow of harmfulness factors in *P. aeruginosa* which were managed by QS (Mihalik et al. 2008).

The present outcome in both subjective and quantitative examinations has interestingly exhibited that aqueous concentrates from *Forsythia suspense* cannot degrade AHL molecules and cannot affect QS by modulating AHL synthesis; the FSE can only interfere with AHL receptor as suggested by the results presented here. Apparently, these inhibitory exercises could most likely hinder the acyl homoserine lactone (AHL) practices by restricting intensely the AHL receptor protein (LuxR homolog) as a key quorum-sensing receptor widespread in *C. violaceum*. The LuxR-type protein CviR detected N-hexanoyl homoserine lactone (C6-HSL) autoinducer produced in *C. violaceum* at high cell density. CviR:C6-HSL complex functioned as a transcriptional activator of genes controlled by quorum sensing (Bucio-Cano et al. 2015). Dissecting the partiality of the ligand with the receptor can be enormously encouraged by the utilization of computer-aided drug design (CADD). The structure of the compound was similar to C6-HSL, which indicated that they may compete against AHLs for binding to the *N*-hexanoyl homoserine lactone receptor, CviR. Using CADD platform for predicting protein-ligand docking, the joint capability effect could be determined by the binding energy. The platform was also applied to screen for QSIs (Zeng et al. 2008). Various synthetic constituents with assorted structures have been reported from *F. suspense* [(Thunb.) Vahl (Oleaceae)]. Kang's research found that five compounds—ursolic acid, phillygenin, (+)-pinoresinol, rutin, and quercetin—were confirmed to exist in species of this genus (Kang and Wang 2010). Phillyrin (Chen et al. 2015), forsythoside (Sheng et al. 2011), pinoresinol (Su et al. 2014), linoleic acid, oleic acid, and palmitic acid (Jiao et al. 2013) also exist in *F. suspense*. Wang et al. confirmed the other six compositions—forsythoside A, phenethyl alcohol *β*-

Table 1 Binding free energy components of protein–ligand complexes (in units of kcal/mol)

Name	Molecular formula	Structural formula	Docking score (kcal/mol)
Pinoresinol	$C_{20}H_{22}O_6$		-26.02
Quercetin	$C_{15}H_{10}O_7$		-25.81
Dimethylmatairesinol	$C_{22}H_{26}O_6$		-21.74
Kaempferol	$C_{15}H_{10}O_6$		-15.38
Homovanillyl alcohol	$C_9H_{12}O_3$		-14.91
Forsythoside B1	$C_{29}H_{36}O_{15}$		-13.77
Forsythoside A	$C_{29}H_{36}O_{15}$		-9.61
Forsythoside D	$C_{20}H_{30}O_{13}$		-4.50
Rutin	$C_{27}H_{30}O_{16}$		-3.98
Astragalin	$C_{21}H_{20}O_{11}$		-3.50
C6-HSL	$C_{10}H_{17}$		-16.09


a
Ala 130
Met 100
Trp 84
Tyr 88
Phe 126
Ile 57
Val 75
Leu 85
Ser 155
Tyr 80
Asp 97
Ser 89
Ile 99
Met 135
HO
O
H
OH
Trp 111
Phe 115
Met 72
Val 59
Ile 153
b
CH3
HN
O
Native CviR Agonist
Non-Native CviR Agonist

◀ **Fig. 6** (**a**) The crossed-domain conformation of pinoresinol:CviR and C6-HSL:CviR. Pinoresinol is purple, and C6-HSL is green. Amino acid involved in binding of the native ligands to the receptors. (**b**) Structures of the native CviR agonist C6-HSL and non-native CviR agonist pinoresinol. Shaded is the similarities between the two compounds' structure

D-xylopyranosyl-(1-6)-β-D-glucopyranoside, forsythoside F, 2-(3,4-dihydroxyphenyl) ethyl-β-D-glucopyranoside, calceolarioside B, and forsythoside E—in the fruits of *F. suspense* (Thunb.) Vahl, determined by spectroscopic and chemical techniques (Wang et al. 2009). Of all the ten selected compounds, they can bind to the signal receptor CviR protein in *C. violaceum*; the most stable of these closed conformations is the pinoresinol:CviR complex, in which score of affinity is the most astounding. Studies with ligands and receptor demonstrate that they stabilize a closed conformation; the pinoresinol binding to CviR transcription factor is antagonized by the native autoinducer C6-HSL. By comparing the structure of pinoresinol and C6-HSL, we found some similarities. This may raise the possibility of pinoresinol being an antagonist. We are as of now chipping away at active compounds' recognizable proof and seclusion in FSE utilizing chromatographic techniques. In order to confirm the anti-QS activity of selected compounds, *C. violaceum* ATCC 12472 was used as the report strain, and the results have shown that all the selected compounds can inhibit violacein production, which means that QS activity has been inhibited.

The scan for such active compounds may add to the disclosure of another methodology for safe hostile to bacterial medications from home-grown sources that have bring down toxicities and do not have the danger of antimicrobial resistance.

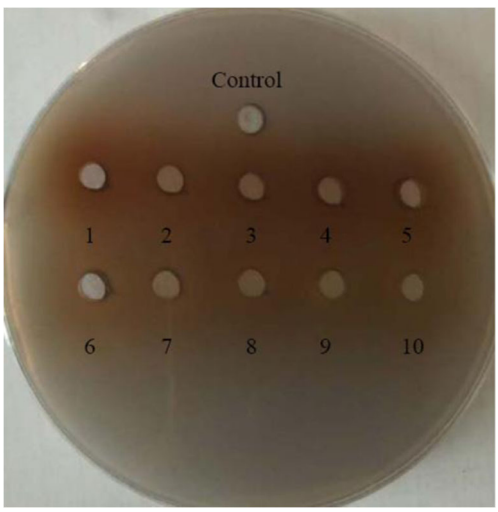


Fig. 7 QS inhibitory activity of selected compounds from *F. suspense*. 1, Pinoresinol; 2, quercetin; 3, dimethylmatairesinol; 4, kaempferol; 5, homovanillyl alcohol; 6, forsythoside B1; 7, forsythoside A; 8, forsythoside D; 9, rutin; 10, astragalin; control, water

Conclusions

The counter majority detecting action capability of *Forsythia suspense* extract was evaluated utilizing *C. violaceum* biosensor frameworks. FSE was shown to have huge focus subordinate inhibitory consequences for QS-intervened violacein generation and biofilm arrangement. The docking of ten mixes of *Forsythia suspense* into the coupling destinations of CviR recommended their differential restricting affinities for the objective proteins. It might be proposed that the *Forsythia suspense* extract can be novel threatening to hostile to bacterial specialists, with the capacity to diminish harmfulness and pathogenicity of microscopic organisms. Our results indicated that FSE maybe used as a tool in the development of new feed additives instead of antibiotics for livestock and aquaculture industry to control bacterial infection.

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Compliance with Ethical Standards

Conflict of Interests The authors declare that there are no conflicts of interest regarding the publication of this paper.

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