Microbiol. Biotechnol. Lett. (2022), 50(1), 1–14
http://dx.doi.org/10.48022/mbl.2111.11012
pISSN 1598-642X eISSN 2234-7305

MBL
Microbiology and Biotechnology Letters

Review

Acyl Homoserine Lactone in Interspecies Bacterial Signaling

Poonam Kanojiya, Rajashri Banerji, and Sunil D. Saroj*

Symbiosis School of Biological Sciences, Symbiosis International (Deemed University), Pune-412115, Maharashtra, India

Received: November 30, 2021 / Revised: December 30, 2021 / Accepted: December 31, 2021

Bacteria communicate with each other through an intricate communication mechanism known as quorum sensing (QS). QS regulates different behavioral aspects in bacteria, such as biofilm formation, sporulation, virulence gene expression, antibiotic production, and bioluminescence. Several different chemical signals and signal detection systems play vital roles in promoting highly efficient intra- and interspecies communication. Gram-negative bacteria coordinate gene regulation through the production of acyl homoserine lactones (AHLs). Gram-positive bacteria do not code for AHL production, while some gram-negative bacteria have an incomplete AHL-QS system. Despite this fact, these microbes can detect AHLs owing to the presence of LuxR solo receptors. Various studies have reported the role of AHLs in interspecies signaling. Moreover, as bacteria live in a polymicrobial community, the production of extracellular compounds to compete for resources is imperative. Thus, AHL-mediated signaling and inhibition are considered to affect virulence in bacteria. In the current review, we focus on the synthesis and regulation mechanisms of AHLs and highlight their role in interspecies bacterial signaling. Exploring interspecies bacterial signaling will further help us understand host-pathogen interactions, thereby contributing to the development of therapeutic strategies intended to target chronic polymicrobial infections.

Keywords: **Quorum sensing, interspecies signaling, LuxR solo, virulence, bacterial communication, pathogenesis**

Introduction

Communication between bacteria of different species and genus is facilitated by signaling molecules synthesized by them and receptors present on bacteria. Microbial interactions have been widely studied to gain insight into growth-inhibitory interactions. However, interspecies signaling may result in other events like antibiosis, motility, competence, and symbiosis. In recent years, various studies on microbial interactions gave us a better awareness of the association of microbes to the components of their habitat and other microbes [1]. There has been a significant rise in research over the last few years documenting a wide variety of interspecies signaling molecules and interactions [2, 3]. The identification of any transition in the growth of an organism living in a polymicrobial community may imply the existence of unnoticed compounds utilized to interact with one another. Exploring microbial relationships can thus direct identification of molecules involved in interspecies interaction. Signaling may lead to a modified use of known molecules [4]. Bacteria can regulate their gene expression as a response to fluctuations in cell density. This process of regulating the gene expression is called quorum sensing (QS). The functioning of QS occurs in a sequential manner starting from the synthesis of signal molecules. Bacteria synthesize signal molecules called autoinducers, and their concentration increases with an

***Corresponding author**
Tel.: +91-20-28116310, Fax: +91-7710005366
E-mail: sunil.saroj@ssbs.edu.in

increase in the cell population. The detection of autoinducers is critical for bacteria. This is done by receptors present on bacteria. Only after achieving a threshold concentration of autoinducers, alteration in gene expression is observed. Gram-positive and gram-negative bacteria respond to chemically distinct signaling molecules. For communication, gram-positive and gram-negative bacteria make use of oligopeptides and acylated homoserine lactone (AHL) as autoinducers, respectively [5]. Most gram-negative bacteria use AHL as intercellular signaling molecules to regulate the QS in response to cell density. These chemical signals are synthesized by the LuxI family of proteins [6]. After exceeding the threshold concentration of AHL, these signaling molecules bind to the LuxR homologs, i.e., the intracellular receptors. Binding of the LuxR to AHL molecules results in a change in gene transcription. It has become increasingly evident over the past few years that this is a very simplistic view of AHL-dependent QS, and that there is, in fact, considerable variation in the way LuxI-R homologs function [5]. To understand infections and design strategies to evade the infections, understanding the communication between microbes in mixed infection is of concern. Taking into consideration that AHLs play a significant role in interspecies communication, the current review summarizes the synthesis of AHLs, regulation of AHL QS, and focuses on findings of AHLs in interspecies bacterial signaling which will provide a better understanding of communication amongst microbes and in turn can help in developing intervention strategies.

Role of Metabolites in Interspecies Communication

N-Acyl-L-Homoserine Lactones

AHLs are the most thoroughly studied signal molecules in gram-negative bacteria. AHL synthase of the LuxI family synthesizes the signal molecule which is detected with the help of AHL receptors of the LuxR family of transcriptional regulators. The AHL autoinducers only bind to the LuxR-type cognate proteins when they cross a critical concentration. Autoinducer binding regulates the activity of the LuxR protein [7]. A wide range of functions like bioluminescence, biofilm formation, and synthesis of virulence factors are regulated by AHL dependent QS circuits [7].

Autoinducer-2 (AI-2)

AI-2 is a common signal molecule utilized by both gram-positive and gram-negative bacteria. AI-2, which is different from homoserine lactones, requires LuxS for its synthesis [8]. LuxP is a periplasmic protein that acts as a primary receptor of AI-2. LuxP on interaction with AI-2 modulates LuxQ. LuxPQ participates in signaling required for regulation of bioluminescence in *V. harveyi* [9]. Studies have revealed that human epithelia synthesize AI-2 mimic as a response to bacteria. This AI-2 mimic is capable of inducing QS in bacteria; therefore, provides evidence of cross-kingdom communication [9].

AI-3/Epinephrine/Norepinephrine

AI-3 is a bacterial signal molecule synthesized by commensals residing in the intestine of the humans. They play a role in virulence as they induce transcription of virulence genes. The detection of epinephrine and norepinephrine by prokaryotes indicates that AI-3 might play a role in communication. A two-component system homologous to PmrAB of *Salmonella* Typhimurium known as QseBC is present in *Escherichia coli*. QseC specifically binds to AI-3, epinephrine, and norepinephrine [10]. In the presence of AI-3, epinephrine and norepinephrine motility genes are expressed in *E. coli*. Host pathogen interactions are aided by motility. The regulation of adhesins is another significant role played by QseBC. The fact that QseC can interact with signal molecules from bacteria as well as eukaryotic host is novel and draws attention towards its role in interkingdom signaling [10, 11].

Indole

Many of the human and plant pathogens are capable of synthesizing indole and its derivatives [12, 13]. Indole synthesis requires tryptophan as a substrate. Two genes involved in the degradation and transport of tryptophan are *tnaA* and *tnaB* encoding tryptophanase and tryptophan permease respectively. A tryptophanase mutant shows decreased activation of *gabT* and *astD*. The conversion of gamma-aminobutyrate to succinic semialdehyde and glutamate is catalyzed by *gabT*. The conversion of succinylglutamate semialdehyde to succinate is catalyzed by *astD*. Studies show that the presence of

Table 1. Representative metabolite and respective gene involved in synthesis.

S. No.	Name of metabolite	Gene
1.	N-Acyl-L-homoserine lactones	AHL synthase
2.	AI-2	*luxS*
3.	AI-3/epinephrine/nor-epinephrine	*luxS*
4.	Indole	*tnaA* & *tnaB*
5.	Antibiotics	*ampC*

indole inhibits biofilm formation in *E. coli*. On the contrary, it favors biofilm formation in *P. aeruginosa* [14, 15].

Antibiotics

Antibiotics are organic compounds synthesized by bacteria to kill or inhibit other microbes. They bind to a specific site on the target cell to act on them. Microbes synthesize antibiotics as secondary metabolites which gives them a competitive survival advantage. Several studies revealed the property of antibiotics to act as a signaling molecule at sub-inhibitory concentrations [16–20]. Many other molecules act as signaling molecules in bacteria. Autoinducing peptides, diffusible signal factors, and diketopiperazines are few other classes of molecules involved in interspecies signaling [3]. Here we focus on AHLs as a signal molecule for interspecies interaction to gain insights on its role in communication (Table 1).

Structure and Synthesis of AHLs

Autoinducer molecules are synthesized by a wide range of organisms like *V. fischeri, P. aeruginosa, S. aureus,* and *S. liquiefaciens*. AHLs are assigned to function as signaling molecules that are only released to trigger QS. The structural elucidation of the autoinducer molecule synthesized by *Photobacterium fischeri* led to the identification of the first AHL molecule as N-(3-oxohexanoyl)-3-aminodihydro-2(3H)-furanone. Subsequently, various AHLs formed by different bacterial species were identified [21]. The constant part of the AHL molecular structure is the homoserine lactone ring. The side chain attached to the central ring can differ in the length of the chain ranging from 4 to 14 carbons in length. The amphipathic nature of AHLs helps them to access the phospholipid layer of the cell membrane in addition to aqueous intracellular and extracellular surroundings. Many bacteria like *V. fischeri, P. aeruginosa* show the presence of multiple QS systems, activated by different types of AHLs. Some of the AHLs synthesized by bacteria are 3-oxo-hexanoyl-homoserine lactone (3OC6-HSL), para-coumaroyl-homoserine lactone, butyryl-HSL, and 3-oxo-dodecanoyl- homoserine lactone (3-oxo-dodecanoyl-HSL). Taken into account the structural differences and specificity of receptors, AHLs are likely to limit the interaction of two systems present in a bacterium [22, 23]. *Rhizobium sphaeroides* and *Rhizobium leguminosarum* synthesize 7,8-cis-N-(tetradecenoyl) homoserine lactone and N-(3R-hydroxy-7-cis-tetradecanoyl)-L-homoserine lactone respectively containing the longest side chains [24]. The synthesis of the side chain of AHLs utilizes the intermediate of fatty acid synthesis or degradation as a substrate which can result in the formation of side chains longer than 14 carbons. Studies have shown that the expression of LuxI/TraI/LasI proteins in bacteria is critical and adequate to confer the capacity to produce analogous AHLs [25, 26]. Therefore, emphasizing the substrate specificity of receptors towards AHLs. An AHL molecule is synthesized in two stages. To synthesize a 3OC6-HSL molecule, the side chain part, i.e., 3-oxo-hexanoyl is produced using substrate from fatty acid metabolism, and the homoserine ring is produced using methionine [25]. Later, it was confirmed that S-adenosylmethionine (SAM) participates in the synthesis of the homoserine lactone ring portion of the AHL molecule [25, 27]. SAM is a cell metabolite used in many essential processes in a cell other than AHL synthesis.

Membrane Trafficking of AHLs

An autoinducer molecule like AHL can simply diffuse in a cell and bind reversibly to its specific receptor. Bacteria try to equilibrate the intracellular and extracellular concentration of AHLs; this was confirmed by a study that reported that the diffusion of 3OC6-HSL was strongly influenced by its concentration gradient over the bacterial envelope [28]. AHL molecule does not have to be synthesized by the same bacteria to cross the bacterial envelope. Signal molecules synthesized by one bacterium can travel through the envelope of another by

diffusion. Similarly, LuxR-type proteins are not solely activated by endogenous AHLs and can be induced by an external supply of AHLs. The findings support the fact that transport of AHLs is dependent on passive diffusion. The hydrophobicity of the AHLs increases with the number of carbon atoms in the side chain. AHLs having side chains with more than eight carbon atoms thus accumulate in the membrane. Other than the length of the side chain, the degree of substitution also affects the movement of AHL across membrane bilayer. Efflux pumps help in the extrusion of long-chain AHLs as they are not freely permeable. BpeAB-OprB and MexAB-OprD efflux systems are involved in the transport of AHLs. BpeAB-OprB is present in the pathogen *Burkholderia pseudomallei*, and MexAB-OprD is present in *P. aeruginosa* [29]. *B. pseudomallei* synthesizes six different types of AHLs all of which are transported using BpeAB-OprB efflux system as they have long side chains(more than ten carbons) [30]. In *P. aeruginosa* long-chain AHLs are transported using the MexAB-OprD efflux system and short-chain AHLs diffuse freely [28].

Overview of AHL QS

Synthesis of AHLs occurs by the activity of a solo enzyme that belongs to the LuxI family of AHL synthases. SAM and intermediates of fatty acid metabolism act as a precursor molecule for AHL synthesis [27, 31]. AHL synthase acts on substrate molecules and activates a cascade of reactions to synthesize AHLs. In *P. aeruginosa* production of butanoyl-homoserine lactone (C4-HSL) and 3-oxo-dodecanoyl-homoserine lactone (3OC12-HSL) is catalyzed by Rh1I and LasI respectively. This denotes that AHL synthases direct the synthesis of similar still chemically distinct molecules. Receptors for AHLs belong to the LuxR family of transcriptional regulators. *V. fischeri* is the marine organism used as a paradigm to study AHL mediated QS. Regulation of bioluminescence by 3OC6-HSL utilizing the LuxI-LuxR system is well researched. 3OC6-HSL can passively diffuse in the environment, and hence the concentration of the signal molecule in the surrounding can provide information about the cell density. LuxR on interacting with 3OC6-HSL induces QS regulated LuxI; after induction AHL concentration increases much swiftly, resulting in luminescence [32].

Interspecies Interactions

The concentration of AHLs in the extracellular pool is determined by the presence of the producers (bacteria documented to synthesize and secrete AHLs) and the degraders (bacteria documented to produce AHL

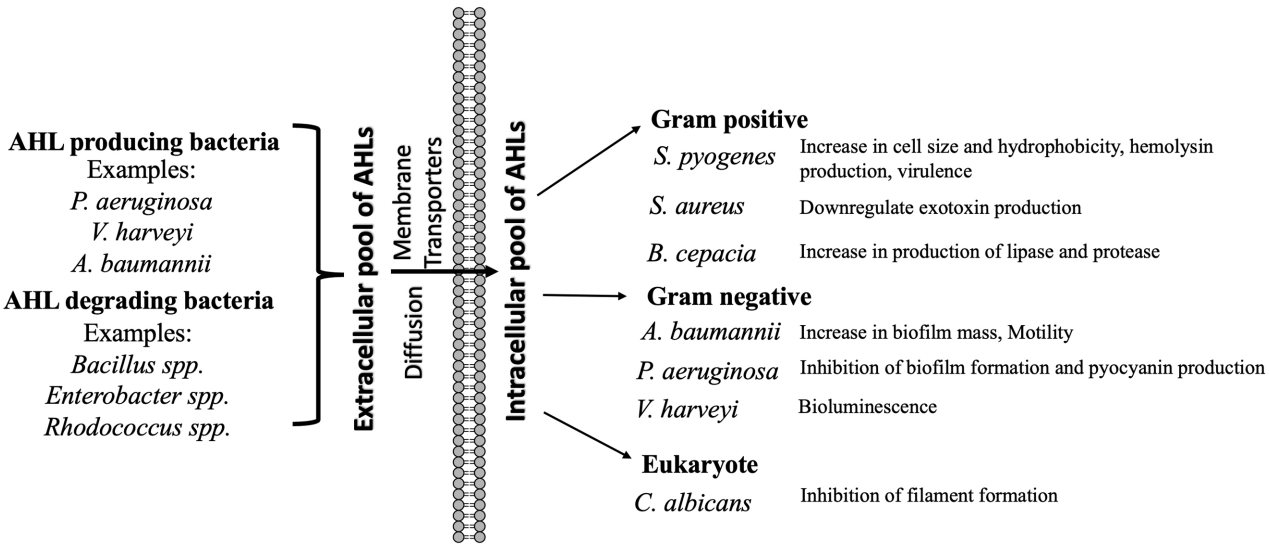

Fig. 1. Schematic representation of effect of AHLs on virulence. Presence of AHLs shows varying response on virulence of microorganisms. Effect of AHLs on various phenotypes contributing to virulence of bacteria and candida is shown.

http://dx.doi.org/10.48022/mbl.2111.11012

degrading enzymes). The AHLs from the extracellular pool can be taken by the producers as well as the non-producers which can interact with the receptors to generate a response (Fig. 1).

Vibrio harveyi

V. harveyi is a gram-negative, free-living, marine bacterium. In *V. harveyi* multiple QS systems function independently to control a group of genes. Two parallel systems control the genes regulating bioluminescence in *V. harveyi* [33, 34]. *V. harveyi* synthesizes two structurally different autoinducer molecules referred to as AI-1 (3-hydroxybutanoylhomoserine lactone) and AI-2 (3a-methyl-5,6-dihydrofuro-[2,3-d][1,3,2]dioxaborole-2,2,6,6a-tetraol). LuxLM and LuxS are autoinducer synthases that catalyze the synthesis of AI-1 and AI-2 respectively [35–37]. The two-component sensor kinase system detects the presence of autoinducers in the periplasm. Binding of AI-1 and AI-2 to LuxN and LuxP respectively activates signaling from another sensor kinase LuxQ [34, 38]. Both LuxN and LuxP contain a periplasmic and a histidine kinase domain. Next in the signal cascade is LuxU, which transmits the signal from LuxPQ and LuxN to shared integrator protein LuxO via phosphorylation. LuxO regulates the transcription of several genes and represses the promoter of *luxCDABE*. Signal transmission inactivates the repression by LuxO and leads to bioluminescence (Fig. 2) [39, 40] Inactivation of either *luxS* or *luxP* in *V. harveyi* leads to the elimination of the fatal impact on brine shrimp *Artemia franciscana in vivo*. However, a mutation in AI-1 mediated components does not show any effect on virulence [41]. This might be due to the instability of AHLs at alkaline pH.

Pseudomonas aeruginosa

P. aeruginosa is an opportunistic pathogen that has come up as a model organism for QS studies. This gram-negative bacterium inhabits a variety of different environments in nature [42] and infects plants, animals, and humans. Extensive studies on *P. aeruginosa* have already revealed detailed information on its QS network. LasR-LasI and RhlR-RhlI both are the AHL circuits present in *P. aeruginosa* which constitute of LuxR-type receptor and a LuxI-type synthase [43, 44]. Other than these a third system is known as QscR is present in *P. aeruginosa* which lacks cognate AHL synthase. These circuits regulate about 6% of the genome. LasI and Rh1I, respectively produce C4-HSL and 3OC12-HSL. The hierarchically positioned LasRI circuit regulates RhlRI circuit at the transcriptional level [45, 46].

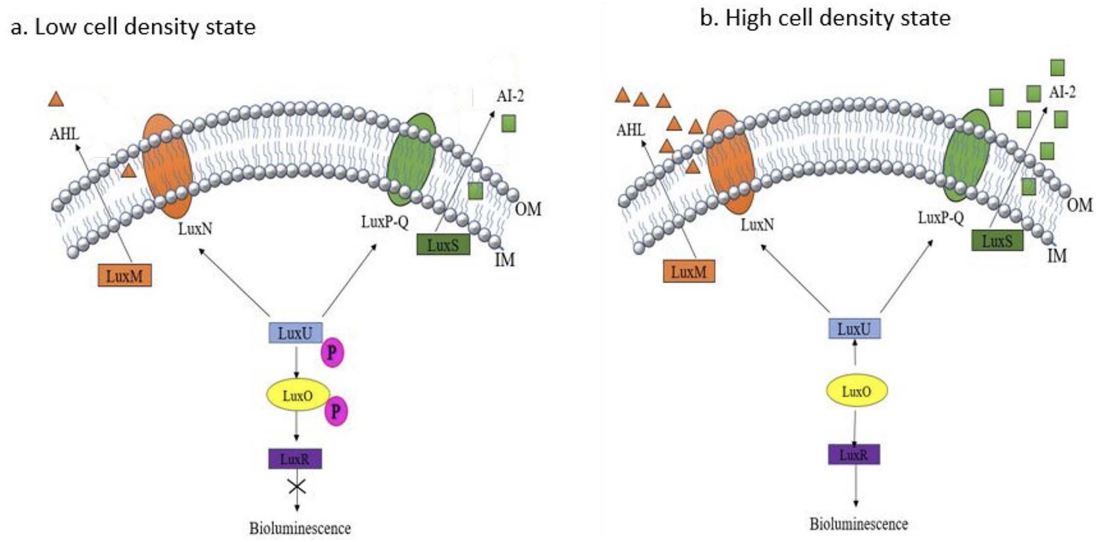

Fig. 2. Schematic representation of *V. harveyi* quorum sensing system. (a) The low cell density state - Phosphate (P) flows towards LuxO at low cell density and act in a negative manner to repress LuxR resulting in inhibition of bioluminescence. IM and OM represents inner and outer membrane respectively. (b) Schematic representation of *V. harveyi* quorum sensing system. The high cell density state - AHLs and AI-2 act in a positive manner to activate quorum-dependent genes. The phosphate flow is reversed and LuxO gets un-phosphorylated leading to binding of LuxR to *luxCDABE* resulting in bioluminescence.

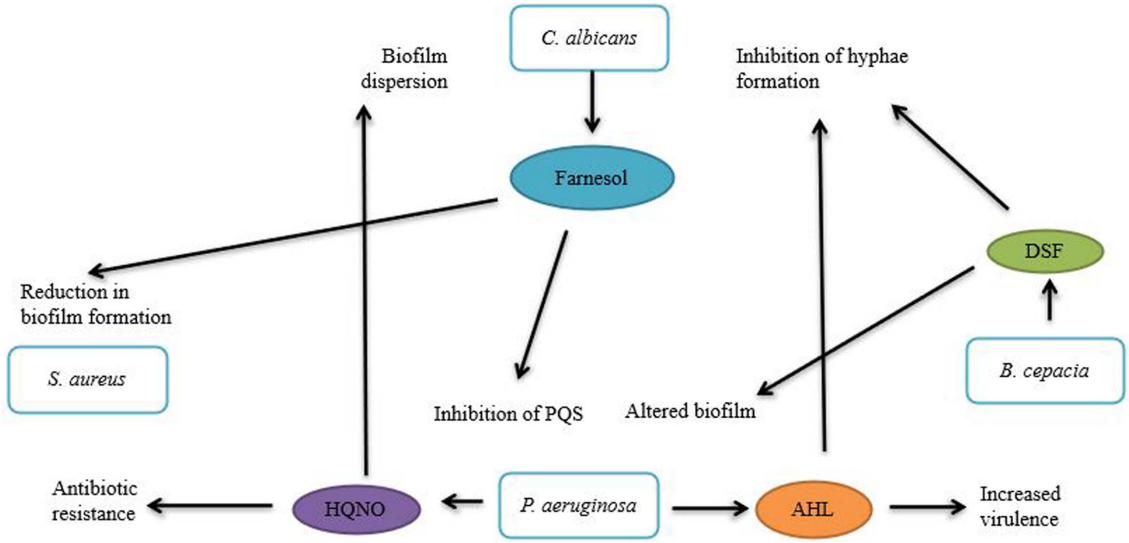

Fig. 3. Schematic representation of interaction amongst pathogens co-colonising with *P. aeruginosa* in human host. AHLs and DSF synthesized by *P. aeruginosa* and *B. cepacia* respectively can inhibit filamentation in *C. albicans*. Farnesol synthesized by *C. albicans* results in decreased virulence of *P. aeruginosa* by inhibiting PQS and reduction of biofilm formation in *S. aureus*. HQNO assists in developing antibiotic resistance in *S. aureus*.

AHL mediated QS helps in the expression of many virulence factors leading to the occurrence of chronic infections. Another system, known as *Pseudomonas* quinolone signaling (PQS) having QscR binds specifically to 3OC12-HSL [43]. Patients with burn wounds and cystic fibrosis (CF) belong to the high-risk group and can be infected more easily by *P. aeruginosa*. An increase in the pulmonary deficit is one of the major causes of high mortality in patients with CF. Patients with CF become progressively susceptible to frequent infections with pathogens like *S. aureus*, *P. aeruginosa*, and *C. albicans*. Mixed species biofilms of *C. albicans* and *S. aureus* are frequently found on catheter implants [47, 48]. The close association of *C. albicans* and *S. aureus* indicates that a similar environment triggers both the species for causing infection. The interactions between a few human-associated pathogens and *P. aeruginosa* are highlighted in this section (Fig. 3). Chronic infections due to *P. aeruginosa* may lead to premature deaths as a consequence of cardiac failure [49]. The transition from yeast form to filamentous growth is considered to be a significant factor affecting the virulence of *C. albicans* [50]. This morphological shift is dependent on several factors like pH, temperature, and presence of other extracellular molecules [51]. The presence of *C. albicans* in the lungs contributes to the prevalence of *P. aeruginosa* infections [52]. 3OC12HSL synthesized by *P. aeruginosa* inhibits the formation of filaments in *C. albicans;* however, it does not impair the growth rate [50]. Also, farnesol is a sesquiterpene molecule produced by *C. albicans* which results in reduced production of pyocyanin by altering the PQS synthesis [53]. Consequently, it leads to a decrease in the virulence of *P. aeruginosa*. 4-hydroxy-2-alkylquinolines (HAQs) are synthesized by *P. aeruginosa* participates in signaling and show antimicrobial activity. Although 4-hydroxy-2-heptylquinoline N-oxide (HQNO) can inhibit the growth of various gram-positive organisms, it infers a benefit to *S. aureus* on long term exposure. Aminoglycosides resistance is developed in *S. aureus* in the presence of HQNO as it hampers electron transport [54]. Thus it can be stated that *S. aureus* develops the ability to survive the antibiotic exposure as a consequence of HQNO induced changes. *S. aureus* also shows a unique phenotypic trait on exposure to HQNO, which is known as small colony variants [55].

Acinetobacter baumannii

A. baumannii is an aerobic, gram-negative pleomorphic pathogen that is challenging to control and cure.

Bacteremia, secondary meningitis, and infections in burn patients are caused by *A. baumannii* [56, 57]. *A. baumannii* is capable of forming a biofilm that allows it to survive in unfavorable conditions and avoid the influence of antimicrobial substances effectively [58, 59]. *A. baumannii* is found to be present in close association with other pathogens infecting patients undergone surgery [60]. Co-infection with other bacteria mostly *S. aureus* or *P. aeruginosa* has been reported in a case study on multidrug-resistant *A. baumannii* isolated from patients. As a consequence of co-infection severity of infection is increased. Reports reveal that *A. baumannii* is resistant to the effect of pyocyanin produced by *P. aeruginosa* PAO1; thus, *A. baumannii* can coexist with *P. aeruginosa* PAO1. This elevates the probability of synthesis of polymicrobial biofilms with raised antibiotic resistance, which may be hard to govern [61]. Considering the close association of *A. baumannii* and *P. aeruginosa* as both of them are related to respiratory tract infections, a comparative genomic analysis was done. The study suggests that *A. baumannii* shows similarities with the opportunistic human pathogen *P. aeruginosa;* however, the presence of only one QS system is reported in *A. baumannii* [62]. N-(3-hydroxydodecanoyl)-L-homoserine lactone (3-OH-C12 HSL) is the AHL molecule synthesized by *abaI* which forms a complex with AbaR transcriptional activator. The synthesis of AHL in *A. baumannii* is regulated by positive feedback using 3-OH-C12 HSL. Reportedly, AbaR and LasR receptor proteins can generate a response to heterologous AHLs. LasR can interact with 3-OH-C12 HSL signal molecule synthesized by *A. baumannii* [63]. Similarly, AbaR can respond to various AHL molecules. A study confirmed that biofilm mass of *A. baumannii* and *P. aeruginosa* rises in the presence of heterologous AHLs. This emphasizes the AHL based communication between them. An increase in biofilm mass occurs only in the presence of long-chain AHLs [61]. Long-chain AHLs also regulates proteolytic activity. Due to the mutual site of infection, AHL based signaling and communication influences the establishment and severity of the infection.

Burkholderia cepacia

B. cepacia is a gram-negative bacterium that is considered as an important pathogen associated with CF [64, 65]. Reports suggest the synergistic effect of extracellular factors secreted by *P. aeruginosa* assists in the attachment of *B. cepacia* to the lungs by exposing receptors of the epithelial cell surface of lungs and enabling attachment [66]. In chronic cases, the infection can lead to 'Cepacia syndrome' or fatal pneumonia [67]. The QS system in *B. cepacia* constitutes of two components, i.e., CepI and CepR. N-octanoyl- L-homoserine lactone (C8-HSL) is the signal molecule synthesized by CepI. *B. cepacia* AHL QS system regulates the expression of extracellular factors by modulating CepI and CepR, i.e., AHL synthase and transcriptional regulator respectively [68, 69]. Cep system is reported to have a different effect on various cellular processes. A study reported that *cepI* mutants show an increased synthesis of ornibactin, which helps in scavenging iron molecules in iron-deficient conditions. A decrease in proteolytic activity was also observed in the absence of AHLs. CF patients are highly susceptible to bacterial infections. Infection with *P. aeruginosa* and *B. cepacia* leads to several other complications in CF patients. As both the bacteria use AHLs to regulate the development of biofilms and virulence factors, it seems possible that besides being competent in interacting with each other such dialogue may also increase the pathogenicity. To understand the signaling mechanism between *P. aeruginosa* and *B. cepacia*; *B. cepacia* was grown in the presence of culture supernatant of *P. aeruginosa.* This resulted in an increase in the production of protease and lipase by *B. cepacia* due to the presence of AHLs in growth media [70]. *cepI* mutants of *B. cepacia* restore the ability to synthesize protease on the external addition of C8 HSL. On the other hand, when *lasI rhlI* double mutant of *P. aeruginosa* is grown in the presence of culture supernatant of AHL synthesizing strain of *B. cepacia,* the protease synthesizing ability is not restored indicating a unidirectional communication [71]. This unidirectional communication was further confirmed by using the mice model [71].

Streptococcus pyogenes

S. pyogenes is a gram-positive pathogen residing in the respiratory tract and skin of the human host. *S. pyogenes* causes a broad spectrum of diseases ranging from mild pharyngitis to life-threatening diseases such as toxic shock syndrome [72]. Autoinducing peptides (AIP) and

Table 2. Summary of organism and respective metabolites used in interspecies signaling.

No.	Name of organism	Metabolite used
1	*Vibrio harveyi*	AI-1 (3-hydroxybutanoylhomoserine lactone) and AI-2 (3a-methyl-5,6-dihydrofuro-[2,3-d][1,3,2]dioxaborole-2,2,6,6a-tetraol)
2	*Pseudomonas aeruginosa*	C4-HSL and 3OC12-HSL
3	*Acinetobacter baumannii*	3-OH-C12 HSL
4	*Burkholderia cepacia*	C8-HSL
5	*Streptococcus pyogenes*	Oxo-C12-AHL

AI-2 are the QS molecules synthesized by the gram-positive bacteria. These pheromones are synthesized and transported out of the bacterial cell. During the export event, the autoinducers undergo post-translation modification to form functional peptides. At high cell density, the ligands bind to the membrane-bound two-component system sensor kinase and further activate the signaling cascade [1]. Currently, four families of the QS system have been identified in *S. pyogenes*. These include the Rgg (regulator gene of glucosyltransferase) system, LuxS/ AI-2, lantibiotic regulatory systems, and Sil (streptococcal invasion locus) system [73]. Although *S. pyogenes* do not code for the AHL synthesis machinery, it has been observed that the pathogen can recognize the AHLs produced by the gram-negative bacteria, thereby modulating its virulence mechanism. The ferrichrome transporter FtsABCD was reported to be involved in the transportation of AHLs across the bacterial membrane in *S. pyogenes* [74]. Moreover, it was demonstrated that *Lactobacillus* released an effector molecule which was recognized by *S. pyogenes*, further leading to inhibition in the production of SLS (Streptolysin S) by the pathogen [75]. Oxo C12 is also reported to promote virulence in *S. pyogenes* [76]. Furthermore, Oxo C12 aids survival of *S. pyogenes* in macrophages [77]. Thus, *S. pyogenes* codes for several membrane-bound receptors that are unidentified which are involved in recognizing the molecules secreted by other bacteria in the vicinity. Such interspecies communication studies need further exploration to understand the relationship and behavioral aspects of the pathogens to design and develop antimicrobial strategies (Table 2).

LuxR Solo

Various studies on virulence and QS have highlighted the need to address the role of AHL QS in bacterial communication [6]. Recent studies have revealed that the genomes of different proteobacteria have sequences of QS-related LuxR AHL sensors/regulators. Numerous proteobacteria lack a cognate LuxI AHL synthase [78]. Many bacteria like *Brucella melitensis*, *S.* Typhimurium, and *E. coli* lack complete QS circuits [79]. The structure shows the presence of a HTH DNA binding domain and an AHL binding domain at C-terminus and N-terminus, respectively [80]. The presence of at least one LuxI and LuxR homolog is considered a complete QS system. Studies revealed that 13% of the bacterial genome out of 265 genomes shows the presence of a complete QS system [79]. The presence of more LuxR than LuxI homologs was reported highlighting that LuxR solos are present apparently. Moreover, the absence of an AHL synthase gene in genomes revealed the existence of LuxR solos. Therefore, LuxR solos are extensively present in bacteria that have an entire AHL QS system and in bacteria that do not presumably synthesize AHLs. The endogenously synthesized AHLs could interact with LuxR solos existing in AHL-producing bacteria and thus expand the AHL QS regulon to various gene targets. Conversely, AHLs produced by different bacterial species may have a more flexible or broad specificity. The non-AHL producing bacteria can either show synergy or can develop competitive behavior towards other bacteria by responding to AHLs produced by other bacteria with the help of LuxR solos. LuxR solo might help to eavesdrop on bacteria in surrounding to compete and survive. Furthermore, there are some LuxR solos lacking the conserved amino acid sequences. These slightly defective LuxR solos may bind to molecules other than AHLs. As a consequence, they may impart information on the location of bacteria. LuxR solos of various AHL producing bacteria like SdiA in *S.* Typhimurium, CviR of *Chromobacterium violaceum*, QscR of *P. aeruginosa*, and BisR of *Rhizobium leguminosarum* are well studied [78, 81]. The presence of multiple QS systems and the synthesis of several types of AHL molecules is reported in *P. aeruginosa.* Further insights about the role of LuxR solos will give an understanding of the purpose of synthesis of a range of AHLs by bacte-

ria occupying the same environment.

LuxR Solos in AHL-Producing Bacteria

Bacteria can detect and respond to the changes occurring in the environment. QS is one of the mechanisms by which bacteria respond to changes in population density. The presence of multiple QS systems in *P. aeruginosa* is well studied. QscR is a homolog of LasR and Rh1R with no cognate signal generator. Although the LasI/R and RhlI/R systems co-ordinate in a hierarchy, and they regulate multiple virulence factors, QS mutants are not avirulent [82]. QscR is flexible in specificity. QscR can interact with the endogenous 3-oxo-C12-HSL produced by LasI and with exogenous AHLs synthesized by other bacteria contributing to increased effective responsiveness to cell density. Premature expression of *phz1 and phz2, hcnAB, lasB, rhlI,* and *lasI* genes occurs in *qscR* mutants, which indicates repression of these genes by QscR [83].

LuxR Solos in Non-AHL-Producing Bacteria

The presence of LuxR solos in non-AHL producing bacteria indicates a possibility that AHL producers and non-AHL producers can sense and respond to the signal molecules synthesized by other microbes. In *E. coli* and *Salmonella enterica* a system similar to *Vibrio* spp. QS system is present. SdiA is the LuxR solo present in the human pathogens *Salmonella, Escherichia*, and *Klebsiella* species. Other than human pathogens plant pathogens also show the presence of LuxR solo. LuxR solo synthesized by plant pathogen *Xanthomonas* spp. is involved in the modification of virulence on binding with low molecular mass compounds produced by plants which may not be AHLs. SdiA is the only receptor that can interact exclusively with signal molecules synthesized by other bacteria in the surroundings. Unlike LuxR, SdiA can also bind with N-(3-Oxo-acyl)-homocysteine thiolactones showing flexibility and can interact with a wide range of AHLs [84]. The binding efficiency is less for 3-oxo-C6-HSL, 3-oxo-C4-HSL as compared to3-oxo-C8-HSL [85, 86]. The *rck* operon comprises six genes (pefI, srgD, srgA, srgB, rck and sgrC), which plays a role in adhesion. The expression of *rck* operon and *srgE* is dependent on *sdiA* [82]. Studies using animal models have proved that the *Salmonella sdiA* mutants are virulent and hence SdiA presumably controls accessory virulence factors. In *E. coli sdiA* is responsible for conferring resistance to antibiotics and quinolones [87]. The ability of SdiA to interact with multiple signaling molecules, i.e., AHLs and indole highlight its importance in cellular functions.

AHL Inhibition

The wide variety of bacteria that controls the expression of virulence and colonization-relevant traits through QS systems indicates that capable quorum sensor inhibitors have significant potential [88]. To reduce the expression of AHL dependent virulence factors, inhibition of AHL production is of concern. Analogs of SAM act as inhibitors as they block the signal reception. Various substrate analogs like L-S-adenosylcysteine, butyryl-SAM, holo-ACP, sinefungin, and D/L-S-adenosylhomocysteine are evaluated for their inhibitory action on AHL production [6]. *fabI* encoding enoyl-acyl carrier protein (ACP) reductase provides a precursor for AHL synthesis. The activity of ACP reductase is NADH dependent. Synthesis of C4-HSL is reduced in *fabI* mutants and inhibited by the antimicrobial chemical triclosan. Inhibition of the precursor production using triclosan (5-chloro-2-(2,4-dichlorophenoxy) phenol) results in the reduction of AHL synthesis [89]. Immucillin A derivatives are transition state inhibitors reported to have an inhibitory effect on 5-MAT/S-adenosyl-homocysteine nucleosidase that takes part in AHL synthesis [90, 91]. These molecules are potential inhibitors of AHL synthesis and may affect cellular metabolism as they hinder polyamine synthesis, quorum sensing and other cell processes. Thus, it is critical to do design inhibitors that explicitly target AHL synthesis.

QS often favors conduct that is harmful to other nearby organisms. One of the well-studied examples of this is the activation of virulence genes using AHL mediated QS in *P. aeruginosa* [92]. In the plant pathogen, *E. caratavora* antibiotic synthesis is dependent on the activation of LuxR homolog [93]. Furthermore, AHL mediated QS also regulates swarming motility in *Serratia liquefaciens,* which contributes to its virulence [94]. In addition, different species of bacteria compete and communicate with one another in mixed-species environments by interacting with heterologous AHLs. As a strategy to survive, few sensitive bacteria have devised

defense mechanisms to interrupt QS. Compounds having structural similarities to AHL molecules are capable of inhibiting AHL regulated processes. Secondary metabolites synthesized by macroalga *Delisea pulchra* hinder AHL regulated processes in prokaryotes. Various halogenated furanones which resemble AHLs are synthesized by *D. pulchra* [95]. These metabolites inhibit bacterial colonization over its surface. Eberl *et al.* reported swarming motility as a phenotype regulated by AHL mediated QS. Motility in *S. liquefaciens* is hindered by inhibiting AHL synthesis using halogenated furanones leading to interruption in colonization on the surface of *D. pulchra*.

Another approach to inhibit AHLs is the use of enzymes that alter AHLs in such a way that they are not functional anymore. Oxidized halogens react only with AHLs containing a 3-oxo group. AHLs from *Chromobacterium violaceum* dropped their activity on exposure to oxidized halogens. AHLs are also reported to restrict violacein synthesis in *C. violeaceum* [96]. Moreover, bacteria produce proteins that obstruct AHL signaling. Production of such enzymes is a benefit as it helps for survival in polymicrobial habitats. Nine genes in *Bacillus thuringiensis* are capable of producing AHL lactonase which is an AHL inactivating enzyme. This might generate potential in *B. thuringiensis* to compete with the neighboring bacteria having AHL producing ability [97]. QS signals are potential targets for disease control, and such enzymes will help in widening the approach to prevent infections.

Swarming is a type of surface translocation used by many bacteria like *Proteus mirabilis, Vibrio parahaemolyticus,* and *S. liquefaciens.* The viscosity of the medium affects the motility. In *S. liquefaciens* AHL mediated QS and *flhDC* regulates swarming motility. At high cell density QS system is activated, leading to surfactant synthesis. Furthermore, cell differentiation occurs depending upon the viscosity of media (0.4–1.2% agar), leading to extensive flagellation. As a consequence of both the events swarming motility occurs [98]. Inhibition of either of events could lead to defect in the swarming motility. A *swrI* mutant cannot synthesize the surfactant W2 and is incapable of swarming motility [99]. The external addition of N-3-oxohexanoyl-L-HSL to *swrI* mutant leads to the restoration of swarming motility. Inhibition of AHL mediated QS leads to reduced motility. This indicates AHL mediated QS plays a role in swarming motility.

The approaches mentioned above focuses on inhibiting AHL synthesis. But another interesting aspect is the inhibitory effect of AHLs on bacterial growth and virulence. The inhibitory effect of AHLs against *S. pyogenes* virulence has been investigated. This study on *S. pyogenes* M6 S165 revealed the negative regulation of expression of *sag* operon by AHLs with fatty acid side chain ≥12 carbon atoms inhibit streptolysin (SLS) mediated hemolytic activity [74]. SLS encoded by *S. pyogenes* has the ability to lyse mammalian erythrocytes and impairs the membrane of lysosomes, neutrophils, platelets, mitochondria [100, 101]. The primary hemolysin SLS is detected at the late exponential phase suggestive of hemolysis to be a cell density-dependent phenomenon.

Discussion

It is now widely accepted that bacteria live in polymicrobial communities and have specific intra and interspecies communication mechanisms. Many signaling mechanisms are established, but it seems certain that several more are still to be identified. The concept of interspecies communication is supported by various studies [5]. AHL-based communication systems have been identified in a number of pathogenic gram-negative bacteria which cause diverse problems, including food poisoning and human lung infections. Monitoring cell to cell communication enables us to explore the efficacy of molecules that restrict communication in biofilm systems as well as other complex instances. As AHLs play a role in interspecies communication in *P. aeruginosa,* they are having an impact on the virulence of microbes participating in the communication. It may be concluded that the inter bacterial communication deficiency leads to a decrease in pathogenicity. This applies to the opportunistic human pathogen *P. aeruginosa*, which infects the lungs of patients with CF.

AHLs serve to control the expression of the bacterial virulence genes via cell-cell contact, but can also be a virulence determinant by virtue of immunomodulatory properties. AHLs are reported to down-regulate host innate immunity [102]. Because AHL mediated QS is essential for the virulence of several bacteria, the signaling molecules and mechanism is the focus for designing

of new types of therapies, antipathogenic agents, compounds that cannot kill bacterial pathogens but impair the ability to trigger infections. The challenge here is to identify and test the efficacy of QS inhibitors in the treatment of infections, particularly persistent biofilm infections.

Bacteria exist in polymicrobial community in nature. As a survival strategy bacterium has adapted several characters like sensing nearby bacteria and responding to the metabolites in the vicinity, bacteria are hardly reported in seclusion, and evolution seems to have established a mechanism for sensing changes in the environment, for determining population density of self and other bacteria in mixed-species populations, and for responding to these details by regulating gene expression. AHL mediated QS helps in survival by regulating genes in response to extracellular factors, and, most significantly, compelling evidence indicates that many pathogens depend on AHL QS to facilitate infection. Detailed awareness of intra- and inter-species signaling processes will undoubtedly improve our understanding of host-pathogen relationships. It will also give us insights about the processes underlying infectious diseases and will help develop and implement new therapeutic strategies. Inhibiting the AHL mediated interspecies signaling could be one of the potential targets for development of novel antimicrobials.

Acknowledgments

PK and RB are supported by the junior research fellowship program of the Symbiosis International (Deemed University).

The work was supported by the Ramalingaswami fellowship program of Department of Biotechnology, India under grant BT/RLF/Re-entry/41/2015.

Conflict of Interest

The authors have no financial conflicts of interest to declare.

References

1. Rutherford ST, Bassler BL. 2012. Bacterial quorum sensing: its role in virulence and possibilities for its control. *Cold Spring Harb. Perspect. Med.* **2**: a012427.
2. Waters CM, Bassler BL. 2005. Quorum sensing: cell-to-cell communication in bacteria. *Annu. Rev. Cell Dev. Biol.* **21**: 319-346.
3. Ryan RP, Dow JM. 2008. Diffusible signals and interspecies communication in bacteria. *Microbiology* **154**: 1845-1858.
4. Fajardo A, Martínez JL. 2008. Antibiotics as signals that trigger specific bacterial responses. *Curr. Opin. Microbiol.* **11**: 161-167.
5. Miller MB, Bassler BL. 2001. Quorum sensing in bacteria. *Annu. Rev. Microbiol.* **55**: 165-199.
6. Parsek MR, Val DL, Hanzelka BL, Cronan Jr JE, Greenberg EP. 1999. Acyl homoserine-lactone quorum-sensing signal generation. *Proc. Natl. Acad. Sci. USA* **96**: 4360-4365.
7. Eberl L. 1999. N-acyl homoserinelactone-mediated gene regulation in gram-negative bacteria. *Syst. Appl. Microbiol.* **22**: 493-506.
8. Schauder S, Bassler BL. 2001. The languages of bacteria. *Genes Dev.* **15**: 1468-1480.
9. Ismail AS, Valastyan JS, Bassler BL. 2016. A host-produced autoinducer-2 mimic activates bacterial quorum sensing. *Cell Host Microbe* **19**: 470-480.
10. Hughes DT, Sperandio V. 2008. Inter-kingdom signaling communication between bacteria and their hosts. *Nat. Rev. Microbiol.* **6**: 111-120.
11. Sircili MP, Walters M, Trabulsi LR, Sperandio V. 2004. Modulation of enteropathogenic *Escherichia coli* virulence by quorum sensing. *Infect. Immun.* **72**: 2329-2337.
12. Verstrepen KJ, Reynolds TB, Fink GR. 2004. Origins of variation in the fungal cell surface. *Nat. Rev. Microbiol.* **2**: 533-540.
13. Lee J, Jayaraman A, Wood TK. 2007. Indole is an inter-species biofilm signal mediated by SdiA. *BMC Microbiol.* **7**: 42.
14. Kuczyńska-Wiśnik D, Matuszewska E, Furmanek-Blaszk B, Leszczyńska D, Grudowska A, Szczepaniak P, *et al.* 2010. Antibiotics promoting oxidative stress inhibit formation of *Escherichia coli* biofilm via indole signaling. *Res. Microbiol.* **161**: 847-853.
15. Lee JH, Lee J. 2010. Indole as an intercellular signal in microbial communities. *FEMS Microbiol. Rev.* **34**: 426-444.
16. Davies J. 1990. What are antibiotics? Archaic functions for modern activities. *Mol. Microbiol.* **4**: 1227-1232.
17. Davies J. 2007. Microbes have the last word: A drastic re-evaluation of antimicrobial treatment is needed to overcome the threat of antibiotic-resistant bacteria. *EMBO Rep.* **8**: 616-621.
18. Seshasayee AS, Bertone P, Fraser GM, Luscombe NM. 2006. Transcriptional regulatory networks in bacteria: from input signals to output responses. *Curr. Opin. Microbiol.* **9**: 511-519.
19. Yim G, Wang HH, Davies J. 2006. The truth about antibiotics. *Int. J. Med. Microbiol.* **296**: 163-170.
20. Yim G, Huimi Wang H, Davies Frs J. 2007. Antibiotics as signaling molecules. *Philos. Trans. R. Soc. Lond., B, Biol. Sci.* **362**: 1195-1200.
21. Eberhard A, Burlingame AL, Eberhard C, Kenyon GL, Nealson KH, Oppenheimer NJ. 1981. Structural identification of autoinducer of *Photobacterium fischeri* luciferase. *Biochemistry* **20**: 2444-2449.
22. Pearson JP, Gray KM, Passador L, Tucker KD, Eberhard A, Lglewski BH, *et al.* 1994. Structure of the autoinducer required for expression of *Pseudomonas aeruginosa* virulence genes. *Proc. Natl. Acad. Sci. USA* **91**: 197-201.

23. Pearson JP, Passador L, Iglewski BH, Greenberg EP. 1995. A second N-acylhomoserine lactone signal produced by *Pseudomonas aeruginosa*. *Proc. Natl. Acad. Sci. USA* **92**: 1490-1494.
24. Gray KM, Pearson JP, Downie JA, Boboye BE, Freenberg EP. 1996. Cell-to-cell signaling in the symbiotic nitrogen-fixing bacterium *Rhizobium leguminosarum*: autoinduction of a stationary phase and rhizosphere-expressed genes. *J. Bacteriol*. **178**: 372-376.
25. Eberhard A, Longin T, Widrig CA, Stranick SJ. 1991. Synthesis of the lux gene autoinducer in *Vibrio fischeri* is positively autoregulated. *Arch. Microbiol*. **155**: 294-297.
26. Gilson L, Kuo A, Dunlap PV. 1995. AinS and a new family of autoinducer synthesis proteins. *J. Bacteriol*. **177**: 6946-6951.
27. Schaefer AL, Val DL, Hanzelka BL, Cronan JE, Jr, Greenberg EP. 1996. Generation of cell-to-cell signals in quorum sensing: acyl homoserine lactone synthase activity of a purified *Vibrio fischeri* LuxI protein. *Proc. Natl. Acad. Sci. USA* **93**: 9505-9509.
28. Pearson JP, Van Delden C, Iglewski BH. 1999. Active efflux and diffusion are involved in transport of *Pseudomonas aeruginosa* cell-to-cell signals. *J. Bacteriol*. **181**: 1203-1210.
29. Evans K, Passador L, Srikumar R, Tsang E, Nezezon J, Poole K. 1998. Influence of the MexAB-OprM multidrug efflux system on quorum sensing in *Pseudomonas aeruginosa*. *J. Bacteriol*. **180**: 5443-5437.
30. Alcalde-Rico M, Hernando-Amado S, Blanco P, Martinez JL. 2016. Multidrug efflux pumps at the crossroad between antibiotic resistance and bacterial virulence. *Front. Microbiol*. **7**: 1483.
31. Hanzelka BL, Parsek MR, Val DL, Dunlap PV, Cronan Jr JE, Greenberg EP. 1999. Acylhomoserine lactone synthase activity of the *Vibrio fischeri* AinS protein. *J. Bacteriol*. **181**: 5766-5770.
32. Parsek MR, Greenberg EP. 2000. Acyl-homoserine lactone quorum sensing in gram-negative bacteria: a signaling mechanism involved in associations with higher organisms. *Proc. Natl. Acad. Sci. USA* **97**: 8789-8793.
33. Mok KC, Wingreen NS, Bassler BL. 2003. *Vibrio harveyi* quorum sensing: a coincidence detector for two autoinducers controls gene expression. *EMBO J*. **22**: 870-881.
34. Henke JM, Bassler BL. 2004. Three parallel quorum-sensing systems regulate gene expression in *Vibrio harveyi*. *J. Bacteriol*. **186**: 6902-6914.
35. Cao JG, Meighen EA. 1989. Purification and structural identification of an autoinducer for the luminescence system of *Vibrio harveyi*. *J. Biol. Chem*. **264**: 21670-21676.
36. Surette MG, Miller MB, Bassler BL. 1999. Quorum sensing in *Escherichia coli*, *Salmonella* Typhimurium, and *Vibrio harveyi*: a new family of genes responsible for autoinducer production. *Proc. Natl. Acad. Sci. USA* **96**: 1639-1644.
37. Chen X, Schauder S, Potier N, Van Dorsselaer A, Pelczer I, Bassler BL, *et al*. 2002. Structural identification of a bacterial quorum-sensing signal containing boron. *Nature* **415**: 545-549.
38. Bassler BL, Wright M, Silverman MR. 1994. Multiple signaling systems controlling expression of luminescence in *Vibrio harveyi*: sequence and function of genes encoding a second sensory pathway. *Mol. Microbiol*. **13**: 273–286.
39. Freeman JA, Bassler BL. 1999. A genetic analysis of the function of LuxO, a two-component response regulator involved in quorum sensing in *Vibrio harveyi*. *Mol. Microbiol.* **31**: 665-677.
40. Freeman JA, Bassler BL. 1999. Sequence and function of LuxU: a Two-component phosphorelay protein that regulates quorum sensing in *Vibrio harveyi*. *J. Bacteriol*. **181**: 899-906.
41. Defoirdt T, Bossier P, Sorgeloos P, Verstraete W. 2005. The impact of mutations in the quorum sensing systems of *Aeromonas hydrophila*, *Vibrio anguillarum* and *Vibrio harveyi* on their virulence towards gnotobiotically cultured *Artemia franciscana*. *Environ. Microbiol*. **7**: 1239-1247.
42. Silby MW, Winstanley C, Godfrey SA, Levy SB, Jackson RW. 2011. *Pseudomonas* genomes: diverse and adaptable. *FEMS Microbiol. Rev*. **35**: 652-680.
43. Schuster M, Greenberg EP. 2006. A network of networks: quorum-sensing gene regulation in *Pseudomonas aeruginosa*. *Int. J. Med. Microbiol*. **296**: 73-81.
44. Williams P, Cámara M. 2009. Quorum sensing and environmental adaptation in *Pseudomonas aeruginos*a: a tale of regulatory networks and multifunctional signal molecules. *Curr. Opin. Microbiol.* **12**: 182-191.
45. Schuster M, Lostroh CP, Ogi T, Greenberg EP. 2003. Identification, timing, and signal specificity of *Pseudomonas aeruginosa* quorum-controlled genes: a transcriptome analysis. *J. Bacteriol*. **185**: 2066-2079.
46. Wagner VE, Bushnell D, Passador L, Brooks AI, Lglewski BH. 2003. Microarray analysis of *Pseudomonas aeruginosa* quorum-sensing regulons: effects of growth phase and environment. *J. Bacteriol.* **185**: 2080-2095.
47. Crump JA, Collignon PJ. 2000. Intravascular catheter-associated infections. *Eur. J. Clin. Microbiol. Infect. Dis*. **19**: 1-8.
48. Wargo MJ, Hogan DA. 2006. Fungal-bacterial interactions: a mixed bag of mingling microbes. *Curr. Opin. Microbiol.* **9**: 359-364.
49. Duan K, Dammel C, Stein J, Rabin H, Surette MG. 2003. Modulation of *Pseudomonas aeruginosa* gene expression by host microflora through interspecies communication. *Mol. Microbiol*. **50**: 1477-1491.
50. Hogan DA, Vik Å, Kolter R. 2004. A *Pseudomonas aeruginosa* quorum-sensing molecule influences *Candida albicans* morphology. *Mol. Microbiol*. **54**: 1212-1223.
51. Trejo-Hernández A, Andrade-Domínguez A, Hernández M, Encarnación S. 2014. Interspecies competition triggers virulence and mutability in *Candida albicans–Pseudomonas aeruginosa* mixed biofilms. *ISME J*. **8**: 1974-1988.
52. Roux D, Gaudry S, Dreyfuss D, El-Benna J, de Prost N, Denamur E, *et al*. 2009. *Candida albicans* impairs macrophage function and facilitates *Pseudomonas aeruginosa* pneumonia in rat. *Crit. Care Med*. **37**: 1062-1067.
53. Cugini C, Calfee MW, Farrow III JM, Morales DK, Pesci EC, Hogan DA. 2007. Farnesol, a common sesquiterpene, inhibits PQS production in *Pseudomonas aeruginosa*. *Mol. Microbiol*. **65**: 896-906.

http://dx.doi.org/10.48022/mbl.2111.11012

54. Tashiro Y, Yawata Y, Toyofuku M, Uchiyama H, Nomura N. 2013. Interspecies interaction between *Pseudomonas aeruginosa* and other microorganisms. *Microbes Environ*. **28**: 13-24.
55. Hoffman LR, Déziel E, d'Argenio DA, Lépine F, Emerson J, McNamara S, *et al*. 2006. Selection for *Staphylococcus aureus* small-colony variants due to growth in the presence of *Pseudomonas aeruginosa*. *Proc. Natl. Acad. Sci. USA* **103**: 19890-19895.
56. Fishbain J, Peleg AY. 2010. Treatment of *Acinetobacter* infections. *Clin. Infect. Dis*. **51**: 79-84.
57. Weinstein RA, Gaynes R, Edwards JR. 2005. Overview of nosocomial infections caused by gram-negative bacilli. *Clin. Infect. Dis*. **41**: 848-854.
58. Nucleo E, Steffanoni L, Fugazza G, Migliavacca R, Giacobone E, Navarra A, *et al.* 2009. Growth in glucose-based medium and exposure to subinhibitory concentrations of imipenem induce biofilm formation in a multidrug-resistant clinical isolate of *Acinetobacter baumannii*. *BMC Microbiol*. **9**: 270.
59. Tomaras AP, Dorsey CW, Edelmann RE, Actis L. 2003. Attachment to and biofilm formation on abiotic surfaces by *Acinetobacter baumannii*: involvement of a novel chaperone-usher pili assembly system. *Microbiology* **149**: 3473-3484.
60. Dent LL, Marshall DR, Pratap S, Hulette RB. 2010. Multidrug resistant *Acinetobacter baumannii*: a descriptive study in a city hospital. *BMC Infect. Dis*. **10**: 196.
61. Bhargava N, Sharma P, Capalash N. 2012. N-acyl homoserine lactone mediated interspecies interactions between *A. baumannii* and *P. aeruginosa*. *Biofouling* **28**: 813-822.
62. Niu C, Clemmer KM, Bonomo RA, Rather PN. 2008. Isolation and characterization of an autoinducer synthase from *Acinetobacter baumannii*. *J. Bacteriol*. **190**: 3386-3392.
62. Savka MA, Le PT, Burr TJ. 2011. LasR receptor for detection of long-chain quorum-sensing signals: identification of N-acyl-homoserine lactones encoded by the avsI locus of *Agrobacterium vitis*. *Curr. Microbiol.* **62**: 101-110.
64. Govan JR, Deretic V. 1996. Microbial pathogenesis in cystic fibrosis: mucoid *Pseudomonas aeruginosa* and *Burkholderia cepacia*. *Microbiol. Rev.* **60**: 539-574.
65. Tümmler B, Kiewitz C. 1999. Cystic fibrosis: an inherited susceptibility to bacterial respiratory infections. *Mol. Med. Today* **5**: 351-358.
66. Saiman LI, Cacalano GR, Prince AL. 1990. *Pseudomonas cepacia* adherence to respiratory epithelial cells is enhanced by *Pseudomonas aeruginosa*. *Infect. Immun*. **58**: 2578-2584.
67. Isles A, Maclusky I, Corey M, Gold R, Prober C, Fleming P, *et al.* 1984. *Pseudomonas cepacia* infection in cystic fibrosis: an emerging problem. *J. Pediatr*. **104**: 206-210.
68. Lewenza S, Conway B, Greenberg EP, Sokol PA. 1999. Quorum sensing in *Burkholderia cepacia*: identification of the LuxRI homologs CepRI. *J. Bacteriol*. **181**: 748-756.
69. Gotschlich A, Huber B, Geisenberger O, Tögl A, Steidle A, Riedel K, *et al.* 2001. Synthesis of multiple N-acylhomoserine lactones is wide-spread among the members of the *Burkholderia cepacia* complex. *Syst. Appl. Microbiol.* **24**: 1-4.
70. McKenney D, Brown KE, Allison DG. 1995. Influence of *Pseudomonas aeruginosa* exoproducts on virulence factor production in *Burkholderia cepacia*: evidence of interspecies communication. *J. Bacteriol*. **177**: 6989-6992.
71. Riedel K, Hentzer M, Geisenberger O, Huber B, Steidle A, Wu H, *et al.* 2001. N-acylhomoserine-lactone-mediated communication between *Pseudomonas aeruginosa* and *Burkholderia cepacia* in mixed biofilms. *Microbiology* **147**: 3249-3262.
72. Carapetis JR, Steer AC, Mulholland EK, Weber M. 2005. The global burden of group A streptococcal diseases. *Lancet Infect. Dis*. **5**: 685-694.
73. Jimenez JC, Federle MJ. 2014. Quorum sensing in group A *Streptococcus*. *Front. Cell. Infect. Microbiol*. **4**: 127.
74. Saroj SD, Holmer L, Berengueras JM, Jonsson AB. 2017. Inhibitory role of acyl homoserine lactones in hemolytic activity and viability of *Streptococcus pyogenes* M6 S165. *Sci. Rep*. **7**: 44902.
75. Saroj SD, Maudsdotter L, Tavares R, Jonsson AB. 2016. Lactobacilli interfere with *Streptococcus pyogenes* hemolytic activity and adherence to host epithelial cells. *Front. Microbiol.* **7**: 1176.
76. Banerji R, Saroj SD. 2021. Interspecies signalling affects virulence related morphological characteristics of *Streptococcus pyogenes* M3. *FEMS Microbiol. Lett.* **368**: fnab079.
77. Banerji R, Saroj SD. 2021. Exposure to acyl homoserine lactone enhances survival of *Streptococcus pyogenes* in murine macrophages. *Microb. Ecol.* doi: 10.1007/s00248-021-01926-1. Online ahead of print.
78. Subramoni S, Venturi V. 2009. LuxR-family 'solos': bachelor sensors/regulators of signaling molecules. *Microbiology* **155**: 1377-1385.
79. Case RJ, Labbate M, Kjelleberg S. 2008. AHL-driven quorum-sensing circuits: their frequency and function among the proteobacteria. *ISME J.* **2**: 345-349.
80. Fuqua C, Greenberg EP. 2002. Listening in on bacteria: acyl-homoserine lactone signaling. *Nat. Rev. Mol. Cell Biol*. **3**: 685-695.
81. Lintz MJ, Oinuma KI, Wysoczynski CL, Greenberg EP, Churchill MEA. 2011. Crystal structure of QscR, a *Pseudomonas aeruginosa* quorum sensing signal receptor. *Proc. Natl. Acad. Sci. USA* **108**: 15763-15768.
82. Ahmer BM. 2004. Cell-to-cell signaling in *Escherichia coli* and *Salmonella enterica*. *Mol. Microbiol.* **52**: 933-945.
83. Chugani SA, Whiteley M, Lee KM, D'Argenio D, Manoil C, Greenberg EP. 2001. QscR, a modulator of quorum-sensing signal synthesis and virulence in *Pseudomonas aeruginosa*. *Proc. Natl. Acad. Sci. USA* **98**: 2752-2757.
84. Janssens JC, Metzger K, Daniels R, Ptacek D, Verhoeven T, Habel LW, *et al.* 2007. Synthesis of N-acyl homoserine lactone analogues reveals strong activators of SdiA, the *Salmonella enterica* serovar Typhimurium LuxR homologue. *Appl. Environ. Microbiol.* **73**: 535-544.
85. Michael B, Smith JN, Swift S, Heffron F, Ahmer BM. 2001. SdiA of *Salmonella enterica* is a LuxR homolog that detects mixed microbial communities. *J. Bacteriol.* **183**: 5733-5742.
86. Yao Y, Martinez-Yamout MA, Dickerson TJ, Brogan AP, Wright PE,

Dyson HJ. 2006. Structure of the *Escherichia coli* quorum sensing protein SdiA: activation of the folding switch by acyl homoserine lactones. *J. Mol. Biol*. **355**: 262-273.

87. Rahmati S, Yang S, Davidson AL, Zechiedrich EL. 2002. Control of the AcrAB multidrug efflux pump by quorum-sensing regulator SdiA. *Mol. Microbiol.* **43**: 677-685.
88. Chan KG, Liu YC, Chang CY. 2015. Inhibiting N-acyl-homoserine lactone synthesis and quenching *Pseudomonas quinolone* quorum sensing to attenuate virulence. *Front. Microbiol.* **6**: 1173.
89. Hoang TT, Schweizer HP. 1999. Characterization of *Pseudomonas aeruginosa* enoyl-acyl carrier protein reductase (FabI): a target for the antimicrobial triclosan and its role in acylated homoserine lactone synthesis. *J. Bacteriol*. **181**: 5489-5497.
90. Singh V, Evans GB, Lenz DH, Mason JM, Clinch K, Mee S, *et al*. 2005. Femtomolar transition state analogue inhibitors of 5'-methylthioadenosine/S-adenosylhomocysteine nucleosidase from *Escherichia coli*. *J. Biol. Chem*. **280**: 18265-18273.
91. Singh V, Shi W, Almo SC, Evans GB, Furneaux RH, Tyler PC, *et al*. 2006. Structure and inhibition of a quorum sensing target from *Streptococcus pneumoniae*. *Biochemistry* **45**: 12929-12941.
92. Taga ME, Bassler BL. 2003. Chemical communication among bacteria. *Proc. Natl. Acad. Sci. USA* **100(suppl 2)**: 14549-14554.
93. Eberl L, Winson MK, Sternberg C, Stewart GS, Christiansen G, Chhabra SR, *et al*. 1996. Involvement of N-acyl-l-homoserine lactone autoinducers in controlling the multicellular behaviour of *Serratia liquefaciens*. *Mol. Microbio*l. **20**: 127-136.
94. Lindum PW, Anthoni U, Christophersen C, Eberl L, Molin S, Givskov M. 1998. N-Acyl-L-homoserine lactone autoinducers control production of an extracellular lipopeptide biosurfactant required for swarming motility of *Serratia liquefaciens* MG1. *J. Bacteriol*. **180**: 6384-6388.
95. Manefield M, de Nys R, Naresh K, Roger R, Givskov M, Peter S, *et al.* 1999. Evidence that halogenated furanones from *Delisea pulchra* inhibit acylated homoserine lactone (AHL)-mediated gene expression by displacing the AHL signal from its receptor protein. *Microbiology* **145**: 283-291.
96. Borchardt SA, Allain EJ, Michels JJ, Stearns GW, Kelly RF, McCoy WF. 2001. Reaction of acylated homoserine lactone bacterial signaling molecules with oxidized halogen antimicrobials. *Appl. Environ. Microbiol.* **67**: 3174-3179.
97. Lee SJ, Park SY, Lee JJ, Yum DY, Koo BT, Lee JK. 2002. Genes encoding the N-acyl homoserine lactone-degrading enzyme are widespread in many subspecies of *Bacillus thuringiensis*. *Appl. Environ. Microbiol.* **68**: 3919-3924.
98. Givskov M, Östling J, Eberl L, Lindum PW, Christensen AB, Christiansen G, *et al*. 1998. Two separate regulatory systems participate in control of swarming motility of *Serratia liquefaciens* MG1. *J. Bacteriol.* **180**: 742-745.
99. Eberl L, Molin S, Givskov M. 1999. Surface motility of *Serratia liquefaciens* MG1. *J. Bacteriol.* **181**: 1703-1712.
100. Bernheimer AW, Schwartz LL. 1964. Lysosomal disruption by bacterial toxins. *J. Bacteriol.* **87**: 1100-1104.
101. Keiser H, Weissmann G, Bernheimer AW. 1964. STUDIES ON LYSOSOMES: IV. Solubilization of enzymes during mitochondrial swelling and disruption of lysosomes by streptolysin S and other hemolytic agents. *J. Cell Biol*. **22**: 101-113.
102. Lowery CA, Dickerson TJ, Janda KD. 2008. Interspecies and interkingdom communication mediated by bacterial quorum sensing. *Chem. Soc. Rev*. **37**: 1337-1346.

http://dx.doi.org/10.48022/mbl.2111.11012