Quorum Sensing and Bacterial Communication
Talking Through Chemical Languages

Microbes, plants, animals, and human beings are intricately related to each other. In quite a few scenarios, these organisms live in close association with each other, for example, in rhizosphere, phyllosphere, etc. Epiphytes harbor microbes which prove helpful to the host. Endophytic fungi prevent diseases and provide drought tolerance to Poaceae (grasses), and mycorrhizal fungi form symbiosis with vascular plants and protect them from diseases and enable them to sequester phosphate from the soil. Among all the symbiotic relationships, nitrogen fixation by rhizobia-legume interaction can be listed as the most beneficial outcomes. Human beings are conscientiously accomplished to exploit others for their personal benefits. Associations of microbes and human beings are not viewed in the positive light. Most microbes with an ability to infect human beings are perceived to lead to an unhealthy situation. However, each organism has been bestowed with a mechanism to protect itself from attack. All organisms have distinct genetic makeup that they are able to maintain their identity and integrity with high precision. Incidentally, recent developments in microbial sciences have revealed that microbes stay on and inside our body. It has been realized that we harbor ten times more bacterial cells than our own (10 trillion). Skin microbiota is dominated by members of *Bacteroidetes, Corynebacteria*, *Propionibacterium*, *Proteobacteria*, *Staphylococcus*, etc. A second major group of microbes (around 160 species) is present in the human gut, intensive analysis of gut microbes has provided very interesting insights for human health. In fact, our well-being is dependent upon the composition of this community.

Quorum sensing (QS) is a fascinating phenomenon that occurs in many bacteria, as well as some fungi and algae. It is a sophisticated communication system that allows microorganisms to regulate their gene expression in response to changes in population density. In other words, bacteria can "sense" the presence and density of nearby bacterial cells and alter their behavior accordingly. Quorum sensing enables bacteria to coordinate their activities, such as biofilm formation, virulence factor production, and other communal behaviors.

While quorum sensing is predominantly known for its role in bacterial communication, it's important to note that similar mechanisms have been identified in animals and insects, although they are not exactly the same as bacterial quorum sensing. In these organisms,

these mechanisms are often referred to as animal quorum sensing or socio-sensory behavior.

Insects, such as ants, bees, and termites, exhibit a form of quorum sensing in their social behaviors. For example, ants use chemical signals called pheromones to communicate with each other. When foragers find a food source, they release pheromones on their way back to the nest. The intensity of the pheromone trail increases with the number of ants using it. As more ants use the trail, the stronger the signal becomes, leading to a positive feedback loop where more ants are attracted to the food source, reinforcing the trail.

While vertebrates do not have a classical quorum sensing system, they do exhibit social behaviors that involve signaling based on the density of their population. For instance, fish, birds, and mammals often engage in collective behaviors such as flocking, or herding. The decision to join or leave these groups can be influenced by the presence and behavior of others. These behaviors are often mediated through visual and auditory cues, as well as hormonal and neural signals.

Even humans exhibit behaviors that could be related to quorum sensing, although the concept is not directly applicable. Social phenomena like crowd behavior, collective decision-making, and the spread of opinions or trends can be compared to quorum sensing principles. In these cases, the collective behavior of a group is influenced by the actions and decisions of individuals, creating a social feedback loop.

Quorum sensing is a mode of bacterial signaling that is based on the production of small diffusible molecules during the bacterial growth phase known as autoinducers (AIs). QS process involves generation and accumulation of a signal molecule in the medium to particular threshold concentration, recognition of the signal molecule by a receptor and forming a signal-receptor complex which consequently activates the expression of an extensive array of genes.

Cell communication and signaling are essential for the proper growth and development of all living multicellular organisms. Because of its universal importance, it is not surprising that many fundamental aspects of cell communication have been evolutionarily conserved between plants, animals, and unicellular eukaryotes, even though these kingdoms diverged more than 1 billion years ago.

Cell communication in bacteria and in some eukaryotic microorganisms occurs in a population-density dependent manner and is based on the production of and response to small pheromone-like biochemical molecules called autoinducers.

Differential gene regulation in response to intercellular signaling provides microbes with a means to express particular behaviors only while growing in social communities. This

process has been termed quorum sensing to reflect the need for a sufficient population of microbes (and concentration of signal) to activate the system.

The Significance of Quorum Sensing

Quorum sensing is crucial for various biological processes, particularly in the context of bacterial infections and environmental adaptations. It plays a pivotal role in the virulence of many pathogens, allowing them to express virulence factors only when their population reaches a certain threshold within a host. Additionally, quorum sensing is involved in symbiotic relationships between bacteria and their hosts, such as in the case of nitrogen-fixing bacteria in plant roots. Furthermore, it influences biofilm formation, a microbial community structure that enhances bacterial survival and resistance to antibiotics.

History of Quorum Sensing

Roughly 40years ago, two papers were published reporting the same breaking news: bacteria are social organisms that communicate each other using a chemical language to coordinate group activities. In 1965 Nature published an article by Tomasz (1965) reporting the first example of a regulatory mechanism in *Pneumococcus* that uses a chemical factor, named competence factor, while only a few years later, Nealson et al. (1970) reported "The discovery of autoinducer activity in *Vibrio fischeri"* in the Journal of Bacteriology. Since then, it took almost 30years to accept the idea that bacteria are social organisms able to communicate with each other using a cell-signaling system. Finally, Fuqua et al. (1994) introduced the term quorum sensing to describe a population-density-responsive gene regulation system able to sense population density and to react only when a quorum of cells is reached. In which way does this system act? Signaling molecules, called autoinducers, provide the means of communication. They are produced and accumulated in the environments. When the quorum (i.e., the right concentration of chemical signals) is reached, these molecules bind the receptor protein and activate changes in the gene expression that result in the activation of the cascade responsible for the production of a number of factors, including virulence and biofilm formation (Fig. 1). Since then, a number of publications have reported on this signaling system in different bacterial species; it is now generally described as quorum sensing. Nowadays, QS is considered a general feature of bacteria, either Gram negative either Gram positive, differing in the architecture of their peculiar chemical signals, that allows bacteria to coordinate their collective behaviors in a way that often mimics that of multicellular organisms. This concept of cell-to-cell communication as social activity among bacteria

has been termed social microbiology. Parsek and Greenberg introduced this term in 2005 to describe this social phenomenon that includes biofilm formation and control of virulence factors.

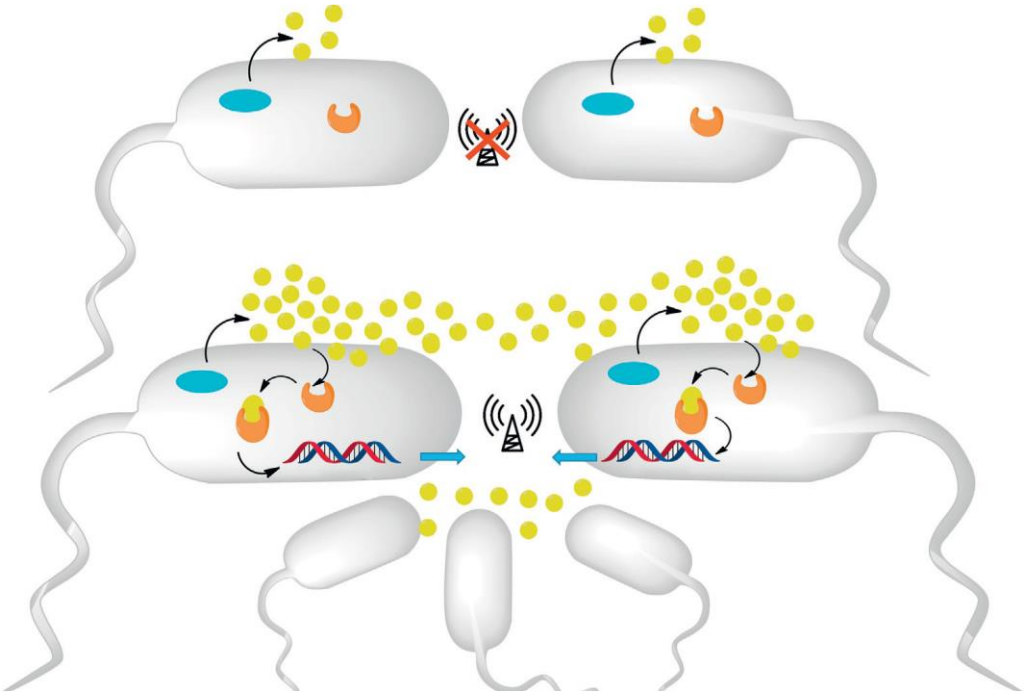

Fig. 1 Quorum sensing system in Gram-negative bacteria. QS relies on the synthesis of autoinducing signals which are produced in a population density dependent manner; when a threshold concentration (quorum) is reached, these molecules interact with a transcriptional regulator, allowing the expression of specific genes at a high cell density.

Quorum sensing in *Vibrio fischeri* is a well-studied example of how bacteria use this communication system to coordinate bioluminescence in response to changes in population density. Quorum sensing in *Vibrio fischeri* was first observed and characterized by Nealson and Hastings in the 1970s and early 1980s. They were studying the bioluminescent properties of these marine bacteria, which emit light as a result of their quorum sensing system. Early research focused on understanding how *Vibrio fischeri* regulates the production of the enzyme luciferase, which is responsible for bioluminescence. The researchers noticed that the luminescence was dependent on cell density, suggesting that it was a form of population-dependent behavior.

By the mid-1980s, researchers had identified the signaling molecule responsible for quorum sensing in *Vibrio fischeri.* This molecule was named "autoinducer" and is referred to as N-(3-oxohexanoyl) homoserine lactone (3-oxo-C6-HSL).

Further research in the 1990s focused on understanding the genetic and molecular mechanisms of quorum sensing in *Vibrio fischeri*.

Scientists discovered that the LuxI protein synthesizes the autoinducer, and the LuxR protein acts as a receptor to detect it. When the autoinducer reaches a certain concentration (quorum), it binds to LuxR, which then activates the expression of genes responsible for bioluminescence. Subsequent studies continued to explore the role of quorum sensing in *Vibrio fischeri* and its ecological significance.

Researchers have investigated how the bioluminescence of these bacteria benefits them, including how it helps them form symbiotic relationships with certain marine animals like squid. The *Vibrio fischeri* system has since become a model for studying quorum sensing in bacteria, and it has contributed significantly to our understanding of how bacterial communities communicate and coordinate behaviors. This research has implications not only in the field of microbiology but also in biotechnology, medicine, and ecological studies.

The Mechanism of Quorum Sensing

Quorum sensing relies on signaling molecules called autoinducers. Bacteria produce these molecules and release them into the environment. When the bacterial population density increases, the concentration of these autoinducers also rises. Once the autoinducer concentration reaches a specific threshold, it binds to receptor proteins in the bacterial cells, triggering a series of gene expression changes. This coordinated response allows bacteria to act collectively and adapt to their environment more effectively.

Autoinducers

Microbially derived signalling molecules act as auto inducers in bacterial quorum sensing. The Gram-negative bacteria use fatty acid derivatives called Homoserine Lactones HSLs whose synthesis is dependent on LuxI homolog or LuxR homolog encoding a transcriptional activator protein responsible for detection of the cognate HSL and the resulting gene expression which results in phenotypic changes. The Gram-positive bacteria use amino acids and short peptide derivatives for quorum sensing.

The autoinducers were first described in the marine luminescent bacterium *Vibrio fischeri*, where they control the mechanism of light production. This marine species lives in symbiotic associations with several different animal hosts and can colonise the light organ of the host. The enzyme luciferase which produced by *V. fischeri* is responsible for light production and encoded by the *luxCDABEGH* genes. The *lux* operon transcription and subsequent light production of *V. fischeri* occur only at high densities and at low densities

are suppressed. A protein called LuxI is an AI responsible for the production of a signalling molecule that diffuses through the membrane into the extracellular surroundings or back into the cytoplasm, where it binds LuxR protein. At the high bacterial density, the concentration of AI increases and the transcription factor LuxR-AI complex activated, which thereby induces transcription of the *luxCDABEGH* genes. Whereas at low bacterial densities, the concentration of AI molecules is below the threshold required to activate transcription, and the *lux* genes encoding luciferase are repressed. It was believed for years that this phenomenon was limited to a few marine organisms, but it is now broadly recognised that numerous bacterial species use quorum sensing as part of their regulatory mechanisms.

Classification of Bacterial Autoinducers

It has been more than 5 decades since the discovery of QS which is considered the most intensively studied interspecies, intraspecies, and interkingdom communication mechanism. Small chemical-signals known as autoinducers mediate this phenomenon which enables the bacterial community to exist like multicellular-organisms. Similar to humans' language, various AIs are employed by bacterial species. Over years a variety of QS signaling-molecules have been uncovered and more are likely to be discovered. However, there are few critical characteristics that need to be present in order to classify a particular molecule as QS signal-molecule such as the manufacture of the signal-molecule in regards to a specific environmental alteration or during particular growth stages, the signal-molecule should be recognized by a specific receptor when accumulated extracellularly, a collective response should be generated following accumulation of the signal-molecules to the critical thresholds, finally cellular-response must expand beyond physiological-alterations needed to detoxify or metabolize the molecule. Based on the type of AIs used we can generally classify QS-systems into the following types:

A) AHL Dependent QS System

AHLs are considered the most prevalent QS signal-molecules utilized by gram-negative bacteria for intraspecies communication. Homologs of LuxR and LuxI system of *V. fischeri* are employed by numerous gram-negative bacteria. Over 100 gram-negative bacteria species are thought to apply this paradigm for chemical-communication. LuxI function as the AI-synthase that synthesizes AHLs by using acyl-carrier protein (ACP) which is an intermediate in the biosynthesis pathway of fatty-acids along with S-adenosylmethionine (SAM) as substrates. LuxR proteins function as transcription-regulators and they consist of an amino (N) terminal domain that binds to the signals and

a carboxyl (C) terminal region that binds to the DNA. The AHLs bind to their cognate LuxR-receptors at high cell densities by doing so the latter will stabilize, fold and bind to the DNA sequence known as the lux box leading to the induction of the transcription of target genes. RhlR/RhlI and LasI/LasR of *P. aeruginosa* along with abaI/abaR of *A. baumannii* are typical examples of LuxR/LuxI paradigm.

B) Autoinducer Peptide (AIP) Dependent QS-System

Oligopeptides referred to as AIPs represent the signal-molecule employed by the majority of the gram-positive bacteria for intraspecies QS. These QS-peptides are produced as precursors (pro AIPs) by ribosomes located in the cytosol of gram-positive bacteria, then depending on the bacterial species they undergo modification either extracellularly or intracellularly to become functional and stable. Due to their inability to passively diffuse through the membrane, an active-transporter usually ATP-binding-cassette (ABC) is used for their extracellular transport. The AIPs will be sensed and bind to the transmembrane-receptors histidine-kinases once their extracellular concentrations reach the QS threshold value. This binding will activate the sensor-kinase which in turn auto-phosphorylates and passes the phosphoryl-group to a cognate-receptor in the cytoplasm that functions to regulate expression of QS-genes, including kinase-receptor genes, transporter-system genes, AIP-genes, and response-regulator genes by binding to the target promoter region. Such QS-mechanism can be observed in bacteria such as *Staphylococcus aureus*. Interestingly, some gram-positive bacteria such as *Bacillus cereus* employ another type of AIP QS-system where the secretory-systems release the pre-AIP into the extracellular environment where they are modified by the extracellular enzymes and then they are transported back inside the cell where they bind to the cytoplasmic response-regulators and alter gene-expressions.

C) Autoinducer-2 Dependent QS-System

Initially detected in *V. harveyi*, autoinducer-2 (AI-2) is a family of diffusible interconvertible molecules chemically recognized as furan borate diester utilized by both gram-positive and negative bacteria for inter and intra-species communication. AI-2s are synthesized from SAM in a series of enzymatic reactions. Different AI-2 forms are detected by bacterial species for example *Escherichia coli* uses non-borated AI-2, whereas, *V. harveyi* use boron-containing AI-2. Periplasmic-receptors recognize and bind to AI-2-molecules accumulated in the extracellular environment, thus leading to the activation of gene-transcription in the respected bacteria. To date, two types of receptors

have been discovered including LuxP which binds to boronated AI2s, and LsrB which detects non-boronated AI2s.

D) Other Signal-Molecules Dependent QS-System

` Bacterial chemical-vocabulary continues to expand as novel AIs are discovered. For instance, ***Pseudomonas* quinolones (PQS)** and a more recently detected QS signal molecules known as **IQS** are encoded by *pqsABCDH* and *ambBCDE* in the *Pseudomonas aeruginosa* and function to control the expression of biofilm and various virulence factor related genes. Initially described in the enterohemorrhagic *E. coli*, **AI-3s** are believed to exist in other enteropathogens as well where they are involved in the activation of the pathogenicity genes. Although the structure of these molecules is not yet characterized finely, studies indicate that host hormones **norepinephrine and epinephrine** can also activate the AI-3-QS pathway. Furthermore, **diffusible-extracellular-factor (DF**) along with **diffusible-signal-factors (DSF)** are fatty-acid derivates utilized as QS AIs by the plant pathogen *Xanthomonas campestris*, *Helicobacter pylori*, and *Burkholderia cepacia* Recently, a number of studies suggest that **indole** which is a bacterial-byproduct synthesized from the hydrolysis of tryptophan by the enzyme tryptophanase may function as a QS signal-molecule and can affect both indole-producing and non-indole-producing bacteria where they regulate a number of bacterial processes including motility, biofilm, and drug-resistance.

Quorum Sensing in Gram-positive and Gram-negative Bacteria

Gram-positive and Gram-negative bacteria use different types of QS systems as shown in figure (2); Gram-positive bacteria communicate using peptides, called autoinducing peptides (AIPs), as signalling molecules. These AIPs are produced in the cell, processed, and secreted. At HCD in which the extracellular concentration of the AIPs is high, the AIPs bind to a cognate membrane-bound two-component histidine kinase receptor. This binding usually activates the receptor's kinase activity; it autophosphorylates and passes phosphate to a cognate cytoplasmic response regulator. The phosphorylated response regulator activates transcription of the genes in the QS regulon.

Gram-negative bacteria use small molecules as autoinducers to communicate, which are either acyl-homoserine lactones or molecules whose formation depends on S-adenosylmethionine (SAM) as a substrate. The AIs are produced in the cell and freely diffuse through the inner and outer membranes. At HCD, when the concentration of AIs

is sufficiently high, the AIs bind to transcription factors which are cytoplasmic receptors. The AI-bound receptors control the expression of the genes in the quorum system regulon.

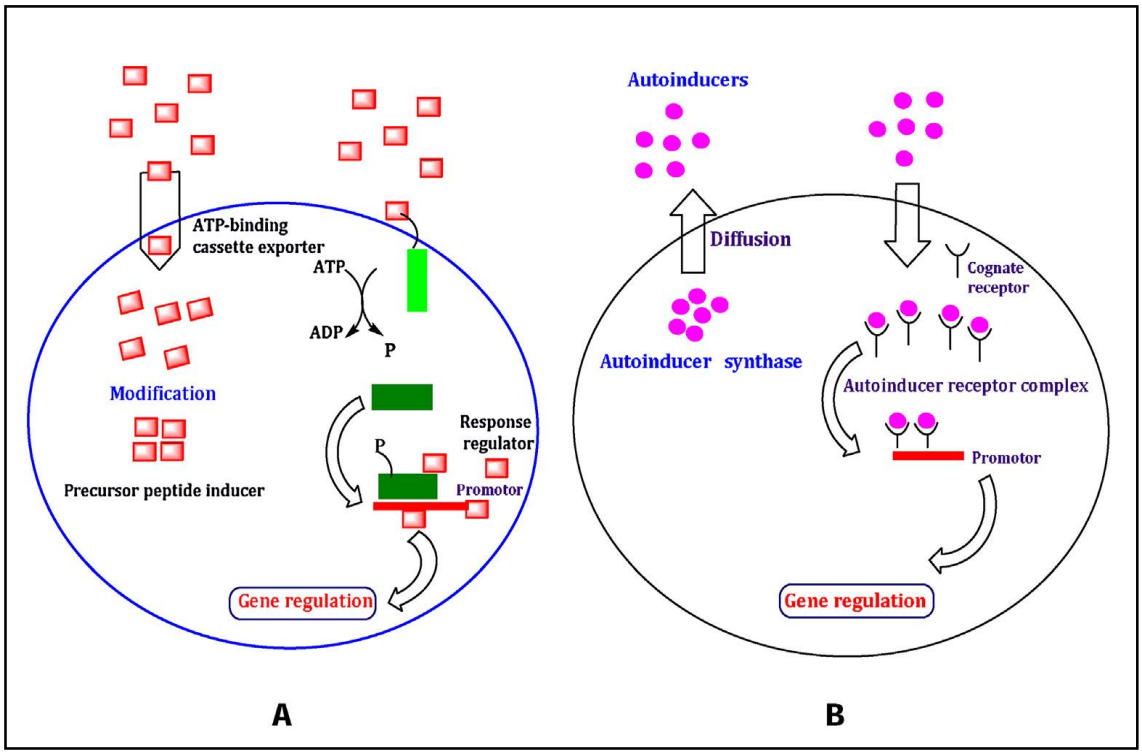

Fig. 2 Graphical presentation of QS molecular signaling network of (A) Gram-positive and (B) Gram-negative bacteria. ADP: adenosine diphosphate; ATP:adenosine triphosphate.

P. aeruginosa Quorum Sensing

P. aeruginosa is a ubiquitous Gram-negative bacterium that can cause both acute and chronic infections in humans. Typically, *P. aeruginosa* infections are found in the lungs of people with CF, which leads to declined pulmonary function and increased mortality. It also causes acute infections in people with compromised epithelial barriers; a common trait among patients with severe burns or tracheal intubation and mechanical ventilation. In both situations, *P. aeruginosa* uses QS systems to communally produce a suite of virulence factors that contribute to its ability to cause disease.

There are three QS systems in *P. aeruginosa* (Figure 3): two LuxI/LuxR-type QS circuits that work in sequence to control the expression of virulence factors as well as a third, the *Pseudomonas* quinolone signal (PQS) system which is non-LuxI/LuxR-type system. In the first circuit, 3-oxo-C12-homoserine lactone (3OC12HSL) is synthesised by the LuxI

homolog LasI. At HCD, this AHL is detected by the cytoplasmic LuxR homolog LasR. The LasR–3OC12HSL complex activates transcription of several target genes, including those encoding virulence factors such as proteases, elastase, and exotoxin A. Furthermore, the LasR–3OC12HSL regulates the lasI, which establishes an autoinducing feed-forward loop.

Another target of regulation by LasR–3OC12HSL is a second luxI homolog called rhlI. RhlI synthesis a second AHL, butanoyl homoserine lactone (C4HSL). At HCD, this autoinducer binds to a second LuxR homolog RhlR. RhlR–C4HSL activates target genes, including those encoding proteases, elastase, siderophores, and pyocyanin, it also regulates rhlI, leading to autoinduction of this second QS circuit.

P. aeruginosa uses an additional QS system to control virulence factor gene expression, which is non-LuxI/LuxR QS system called *Pseudomonas* quinolone signal (PQS). In the PQS system, 2-heptyl-3-hydroxy-4-quinolone is produced by PqsA, PqsB, PqsC, PqsD, and PqsH and is detected by the regulator PqsR. Expression of pqsH and pqsR is activated by LasR–3OC12HSL, whereas RhlR–C4HSL represses pqsABCD and pqsR. PqsR–PQS autoinduces PQS synthesis and further activates rhlI and rhlR. Accordingly, the PQS circuit is closely tied to the LasI/LasR and RhlI/RhlR QS systems and, therefore, also impacts virulence factor production. *P. aeruginosa* QS-activated virulence factors include proteases, elastase, pyocyanin, swarming motility, rhamnolipids, lectin, toxins, and biofim formation.

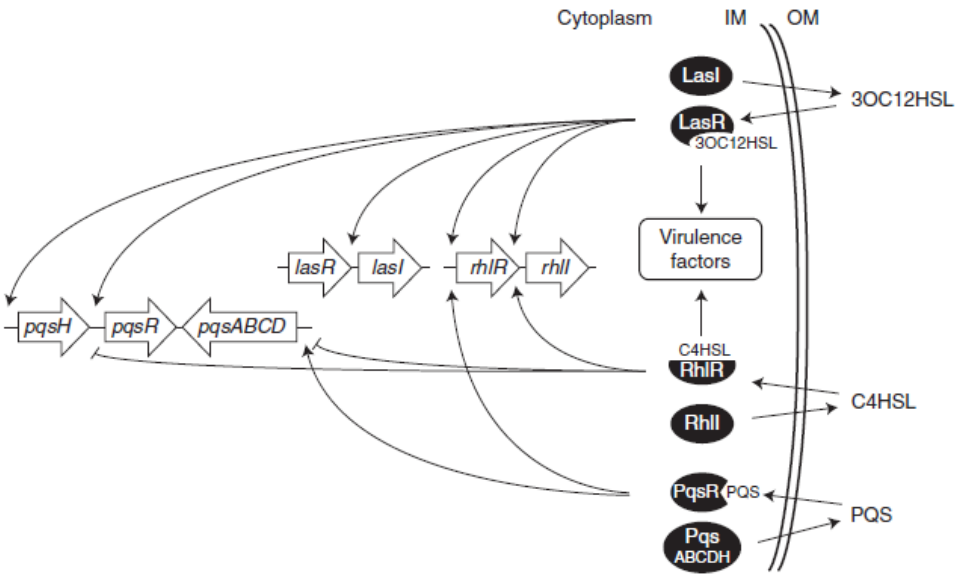

Fig 3. *P. aeruginosa* QS circuits.

Staphylococcus aureus Quorum Sensing

Staphylococcus aureus is found among the normal human skin flora. If the epithelial barrier is compromised, *S. aureus* can cause minor skin infections. These infections can lead to pneumonia, bacteremia, and sepsis. Its ability to cause disease depends on expression of an array of adhesion molecules, toxins, and compounds that affect the immune system. QS regulates expression of genes encoding these virulence factors. *S. aureus* uses a canonical Gram-positive two-component QS system encoded by the agr locus (Figure 4.). The P2 promoter drives expression of a transcript (RNAII), which encodes the four components of the QS system. agrD encodes the pro-AIP, which is processed to the final AIP and secreted by the transmembrane transporter protein AgrB. When the AIP accumulates, it binds the membrane-bound histidine kinase AgrC, which autophosphorylates at a conserved histidine and transfers the phosphate group to an aspartate on the response regulator AgrA. Phosphorylated AgrA binds upstream of theP2 promoter to autoinduce the agr operon. In addition to activating the P2 promoter, phosphorylated AgrA activates the divergently encoded P3 promoter. The P3 promoter controls expression of RNAIII.

The net result of this QS regulatory cascade is downregulation of surface virulence factors (such as protein A), and up-regulation of secreted virulence factors (such as a-toxin). Most of the effects of QS on regulation of virulence in *S. aureus* are mediated through direct and indirect regulation by RNAIII.

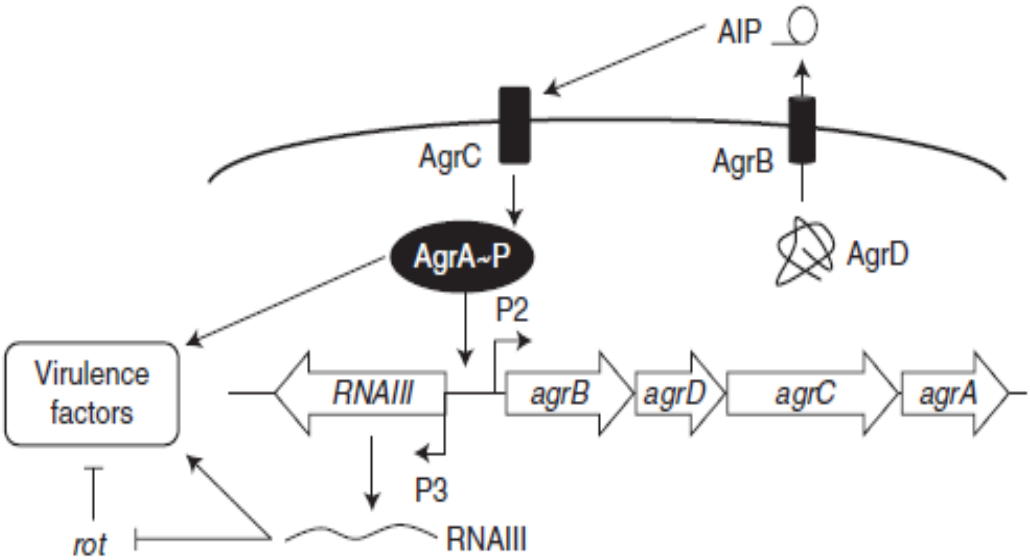

Figure 4. *S. aureus* Agr QS circuit.

Quorum Quenching

Interference of AI-dependent QS is commonly known as quorum quenching (QQ); it has been considered as the novel way to control bacterial infections. QQ can be attained in several ways. First, inhibition of AIs biosynthesis which can be achieved by inhibiting the enzymes involved in their biosynthesis. Second, the destruction of the QS signaling molecules will prevent them from accumulating. Third, inhibition of QS receptors, consequently attenuates QS-dependent virulence.

Applications of Quorum Sensing

Understanding quorum sensing has significant implications in various fields. In medicine, researchers are exploring ways to disrupt quorum sensing in pathogenic bacteria as a novel strategy to combat bacterial infections. In agriculture, it is being used to enhance plant growth by promoting beneficial symbiotic relationships between plants and bacteria. Additionally, in biotechnology, quorum sensing systems are being engineered to control the production of valuable compounds in microbial cultures.

Quorum Sensing as Targeting Therapeutic Pathway to Combat Bacterial Pathogenicity

The beginning of the 20th century brought relief to human beings from a substantial number of life-threatening illnesses through the invention of antibiotics. As the 21st century commences, excessive and indistinctive use of antibiotics has led to the emanation of multi-drug resistant (MDR) bacterial strains. Today, the universal concern is of the diminished capability to fight microbes while we are entering a post-antibiotic epoch. The fact that 16 million people die every year due to the infectious diseases caused by MDR strains led to the need for alternative strategies to combat them. One strategy to treat antibiotic-resistant strains which is less likely to assess a selection pressure for development of resistance is by the development of new mechanisms of antipathogenic treatments that act to weaken the virulence expression. New anti-QS molecules are now being considered as a valuable alternative to overcome the vast challenge posed by drug-resistant pathogens. Several strategies have been considered to interrupt and or disrupt the bacterial QS system. They include inhibition of autoinducers synthesis, inhibition of signal diffusion, inhibition of signal reception, sequestration of autoinducers using, for example, antibodies against autoinducers, degradation of autoinducers, inhibition of

autoinducers secretion/transport, and antibodies that "cover" and therefore block autoinducers receptors.

Screening for Quorum Sensing Inhibitors

To develop biological screens that will assist in the isolation and identification of QS inhibitors (QSI), it has to be realised which properties are preferentially required for an effective and applicable compound. In essence, the ideal QSI is a low-molecular-mass molecule, the activity of which causes a significant reduction in the expression of QS-controlled genes. Second, but equally important, the inhibitor exhibits a high degree of specificity for the QS regulator (i.e., the LuxR homologue) with no toxic side effects on either the bacteria or an eventual eukaryotic host. For example, if the QSI does not interfere with basal life processes such as general RNA and protein synthesis of the bacteria, and hence growth, it is possible that the selective pressure for development of resistance is decreased. In addition, the QSI should be chemically stable and resistant to metabolism and disposal by the higher host organism. *In vitro* screening of many QSIs necessitates an easily detectable reporter. The first screen for QSIs was based on the bacterium *Chromobacterium violaceum*, a Gram-negative bacterium that in the shape of a signal molecule mutant produces the purple pigment violacein in response to the exogenously added AHLs, 3-oxo-N-hexanoyl homoserine lactone (3-oxo-C6-HSL) and N-hexanoyl homoserine lactone (C6-HSL). If *C. violaceum* is plated onto an agar plate, the colonies will turn purple, and in the presence of QSI, the colonies will appear white. Another equally effective biosensor for long-chain AHL inhibitor screening is *Agrobacterium tumefaciens* NT1. On the other hand, *V. harveyi* is used most commonly as a biosensor for AI-2 type autoinducers.

The use of bacterial biosensors provide simple, rapid, and sensitive means to detect QS inhibitors without requiring any sophisticated instrumentation, but they do not allow quantitation.

Quorum Sensing Inhibitors

Although each QS system used by bacteria is unique and specific, all QS circuits share a common mechanism involved in AI production, accumulation, and detection. At least each of these steps has been targeted by QS inhibitors. Consequently, there have been described quorum-sensing inhibitors for signal inactivation and degradation, and for inhibition of QS signal biosynthesis, and detection. To select a QSI effectively, it has been proposed that it should have at least the following characteristics: first, a small molecule able to reduce the expression of QS-regulated genes efficiently, second, be highly specific

for a given QS regulator and has no adverse effect(s) on each of bacteria and the host, and third, be stable chemically and unaffected by various host metabolic systems. As a consequence of these properties of a QSI, resistance to such drugs is excluded which generally exerts selection pressure during treatment of infections. The followings are the most relevant strategies of attenuating bacterial virulence by using QS modulators and QQ:

1-Natural Quorum Sensing Modulators

Naturally originated compounds are always preferred in the biomedical field, since they are biodegradable and usually very efficient, being perfect candidates for ecological anti-infectious strategies. Studies propose the usage of natural eukaryotic- derived vegetal and animal compounds, or microbial-derived compounds for attenuating bacterial virulence and modulating QS.

2-Microbial Quorum Sensing Modulators

The human microbiota involves more than 1000 species, and it is estimated that gastrointestinal tract harbour 10 times as many bacterial cells as there are human cells within our bodies. The normal microbiota plays critical roles in mammals physiology, immunity, development, nutrition, and behaviour, such that distracting the structure and balance of this communal leads to dysbiosis and disease. The proposed competition between resident microbes and pathogens is affected by the virulence factors expressed by pathogens and by the nutritional requirements of both inhabitants. One of the most significant features in a quorum-sensing bacterial population is density-dependent suitability benefits, and bacterial density became essential in infections. These dynamics can direct the colonisation, survival, and clearance of pathogens in the gut.

3-Algae-derived Quorum Sensing Modulators

Many photosynthetic organisms ranging from algae to superior plant species have been evidenced to interfere with quorum sensing, virulence, and pathogenesis. The unicellular soil-freshwater algae *Chlamydomonas reinhardtii* and *Chlorella* spp. were shown to secrete compounds that affect the activity of the N-acyl-L-homoserine lactone, signal molecules that regulate the expression of quorum sensing genes in many bacteria Furanones and their derivatives produced by *Delisea pulchra* are the well-investigated algal QS inhibitors. Among the first recognised small-molecule inhibitors of quorum sensing were brominated furanones. Even though their toxicity prevents their commercial or therapeutic use, furanones provided suitable molecular probes in understanding signaling and the consequences of its inhibition.

4-Mammalian-derived Quorum Sensing Modulators

To find molecules protecting human and animal hosts from infections, researchers have explored mammalian signaling compounds in finding molecules able to attenuate cell-to-cell communication. It has been evidenced that bacterial QS molecules are inactivated or cleaved by mammalian enzymes after entering the host cells. QS system inactivation appears to be molecule specific, Paraoxonases are highly conserved antioxidant enzymes in human are also able to hydrolyze lactones.

Furthermore, these paraxoanases can interfere with QS, degrade AHLs in *P.aeruginosa* block the communication and reduce the extent of infection.

5-Synthetic Quorum Sensing Modulators

Since novel insights into discovering the intimate molecular support of bacterial QS signaling molecules have brought by technological progress, many efforts made to develop targeted synthetic QS modulators. In *S. aureus* Agr system controls QS signaling and the production of Agr peptides, it has been shown by, that synthetic Agr thiolactone-containing autoinducing peptides interfere with *S. aureus* virulence both *in vitro* and *in vivo*.

6-Antibody-based Quorum Sensing Inhibitors

Due to their low molecular weight and non-proteinaceous nature, AHLs and relative signal molecules were not expected to elicit an antibody-based immune response. The most effective among all antibodies screened for the ability to degrade 3OC12HSL of *P. aeruginosa* was antibody XYD-11G2.

7-Plant-derived Quorum Sensing Modulators

Plants have been used since ancient times for their antimicrobial and healing properties. Even though the action of plants on bacterial development at the molecular level is mostly unknown, they are employed in modulating of bacterial virulence, adherence and biofilm formation and also for attenuating QS communication. Studies have revealed that geranium, rose, peppermint, lavender, cinnamon, clove and rosemary oils are very potent QS inhibitors mediating QS-dependent phenotypes in Gram-negative species.