

Quorum Sensing in Plant Pathogenic Bacteria and Its Entanglements in Plant Disease Development

N. Olivia Devi and Bharati Lap

Ph.D Scholar, School of Crop Protection, College of Post Graduate Studies in Agricultural Sciences, CAU, Umiam, Meghalaya

SUMMARY

Quorum sensing is a chemical communication that regulates the behaviour of bacterial population in a coordinated manner by detecting signal molecules that are produced and released by bacteria. The majority of the bacteria that use quorum-sensing systems have been linked to plants or animals in some way. Most phytopathogenic bacteria employ this regulatory mechanism for virulence and pathogenicity by controlling the expression of virulence factor encoding genes. They also use QS to survive in diverse environmental conditions by controlling the expression of such genes. Several mechanisms of QS have been used by phytopathogenic bacteria such as signal molecules N-acyl-homoserine lactone (AHL), diffusible signal factor (DSF), and other virulence factors.

INTRODUCTION

Quorum sensing (QS) is a kind of cell to cell chemical communication mechanism employed by the bacteria in order to regulate collective behaviour like gene expression in a coordinated way (Baltenneck et al. 2021). It depends on the production, recognition and response to diffusible signal molecules called the autoinducers in gene density-dependent manner. Bacteria use such behaviour in order to compete, survive in nature and colonize a host. It also helps in the expression of genes in bacteria that are involved in several functions like toxin production, extracellular enzyme production, biofilm formation, plasmid conjugation and motility etc. Such regulatory mechanism of genes through quorum sensing is commonly used by bacteria in order to control biological function like expression of gene encoding for virulence factor which helps the bacteria to cause infection and survive in diverse environmental conditions. Gram negative bacteria uses N-acylhomoserine lactones (AHLs) as a signaling molecules in QS and gram positive bacteria use oligopeptides, while autoinducer 2 (AI2) are found in most bacteria. Autoinduction of QS was first reported for the regulation of gram negative bioluminescence bacteria *Vibrio fischeri* in 1970s. Later it has been reported in many plant pathogenic bacteria like *Pseudomonas aeruginosa*, *Pectobacterium carotovorum*, *Agrobacterium tumefaciens* and *Agrobacterium tumefaciens*. In recent case due to better understanding of virulence functions controlled by QS, there are reports of tactics to control phytopathogenic bacteria by development of QS disrupting approaches.

Mechanism of quorum sensing in bacteria

The bioluminescence phenomenon of *Vibrio fischeri* is the paradigm of QS in gram negative bacteria. The LuxI and LuxR proteins control the expression of the luciferase operon (*luxLCDABE*), which is required for *V. fischeri* to produce light (Gour, 2018). The most frequent autoinducer for gram-negative bacteria is AHLs (N-acyl-homoserine lactones). AHLs molecules are permeable to membrane so they diffuse freely to the exterior of the cell. When the signal reaches a certain concentration threshold it is bound to the LuxR proteins and also induces the expression of LuxI since LuxR-AHL complex is encoded in luciferase operon. The LuxI/LuxR protein complexes have a feedback regulatory impact on the synthesis of AHLs signal molecules and their receptor proteins at the same time. This regulatory setup sends a lot of signal into the environment. This sets off a positive feedback loop, causing the entire population to go into quorum sensing mode. In case of gram-positive bacteria autoinducing peptides are used to confer exchange of intraspecies communication among cells. Because peptide signals cannot freely diffuse or traverse cell membranes, they must be released with the help of ATP-binding cassette transporter or protein membranes (Singh and Ray, 2014). When peptide signals released by the gram positive bacteria in the surrounding environment reaches a certain level it binds to the cell surface receptors that triggers the two component phospho-kinase system consisting of response regulatory protein AgrA and transmembrane sensor kinase AgrC. Later, the peptides interact with cognate regulators either transcriptional or phosphatases regulators to modulate target genes expression and also starts transduction and transcription of

genes. These regulators have been found in bacilli, enterococci, and streptococci and assist in activities like virulence, persistence, conjugation, and competence.

Quorum sensing in different phytopathogenic bacteria

Ralstonia solanacearum

Bacterial wilt caused by strains of *Ralstonia solanacearum* causes wilt on a wide range of plant species. Quorum sensing contributes to the virulence of *R. solanacearum* by controlling the production of secondary metabolites, extracellular polysaccharide (EPS), cellular motility and biofilm formation. OE1-1 strain of *R. solanacearum* produces a QS signal molecule methyl 3-hydroxymyristate (3-OH MAME) synthesized by PhcB encoded S-adenosyl methionine-dependent methyltransferase and this are sensed by PhcS/PhcRQ two component regulatory system (Yoshihara et al. 2020). Through QS, PhcA a LysR- type transcriptional regulators regulate virulence through *ralA* encoding for furanone synthase and its activity is controlled by 3-OH MAME signal molecules.

Pseudomonas syringae

The AHL-mediated QS systems have been investigated in various *P. syringae* pathovars in recent years. It was first reported in *Pseudomonas syringae* strain B728a. AHL synthetase of B728a strains is named AhII, N-(3-oxo-hexanoyl)-L-homoserine lactone (3-oxo-C₆-AHL) which interacts with the regulator AhIR. AHL system positively regulates a range of qualities in *P. syringae* such as assisting in cell-aggregation, survival on leaf surface, regulates swarming motility and epiphytic fitness to cause plant diseases. *P. syringae* pv. *actinidiae* lacks the AHL synthase based QS system instead has three signal receptors viz. PsaR1, PsaR2, and PsaR3 which is homologous to LuxR.

Erwinia amylovora

AHL production by *Erwinia amylovora* in both in planta and in vivo was first reported in Italy isolates. In *E. amylovora*, QS regulates the synthesis of extracellular polysaccharides (amylovoran and levan) as well as tolerance to hydrogen peroxide. Qs system in *E. amylovora* have two component signal transduction systems consisting of bis-(3'-5')-cyclic di-GMP (c-di-GMP) and QS (Piqué et al. 2015). They also produce two signal molecules named as OHHL and HHL, while EamRI is homologues to LuxRI.

Agrobacterium tumefaciens

Virulence part of Ti plasmid and virulence protein in *A. tumefaciens* causes tumour in plants (Gohlke and Deeken 2014). QS signal molecules belonging to LuxR/LuxI class and AHLs helps in the transfer of oncogenic Ti plasmid to plasmidless agrobacteria. QS is regulated by TraR gene, a LuxR type AHL receptor which is found homologous to LuxR of *Vibrio fischeri* and responding to N-3-oxo-octanoyl-HL (3-oxoC₈HL) for its activity. In *A. tumefaciens* the opines and QS signal pathways are linked, with opines serving as the primary regulator, as a result, TraA-3-oxoC₈HSL signalling requires the presence of adequate opines. While TraR gene is regulated by an OccR (octapine responsive activator) in octapine type plasmids.

Xanthomonas spp.

The diffusible signal factor (DSF) molecule facilitates the QS regulatory systems of *Xanthomonas*. *X. campestris* pv. *campestris* (*Xcc*) commonly use this DSF mediated QS while *X. oryzae* pv. *oryzae* (*Xoo*) employs DSF and Ax21 and type I secretion proteins to express pathogenicity and virulence. It was found that the DSF regulated QS is associated with the *rfp* (regulation of pathogenicity factors) gene cluster which is needed for the synthesis and perception of DSF signal (Yu et al. 2015). Such gene cluster includes *rpfB*, *rpfF* and *rpfGHC* genes. In several *Xanthomonas* species, the DSF/Rpf regulation mechanism has been shown to contribute to virulence. In *X. oryzae* QS, StoS (stress tolerance-related oxygen sensor) and SreKRS (salt response kinase, regulator) helps in swarming, EPS production along with virulence factors production and pathogenicity (Hrp) proteins.

Xylella fastidiosa

Similar to *Xanthomonas* pathogen, *Xylella fastidiosa* also utilizes diffusible signalling factor (DSFs) to regulate its behaviour in a cell density-dependent manner in QS system. RpfF synthesis DSFs and such RpfF signal help to regulate biofilm formation, virulence and motility in *X. fastidiosa*. QS regulates switching between the plant host and insect vectors, by rpfF and rpfC mutants in *X. fastidiosa*. DSF assists in cell stickiness, which helps the bacteria cling to the xylem wall during high cell density, allowing the insect to acquire the bacterium.

CONCLUSION

Among several phytopathogenic bacteria, quorum sensing which synchronises bacterial responses within a population is widespread. Quorum sensing ability to coordinate bacterial behaviour in cell density manner helps the pathogenic bacterial in several ways like in infection process by regulating the virulence determinants and also helps evading host defence. Since QS plays a considerable role in bacterial pathogenesis it can be targeted one or other way to produce an effective strategy to manage plant diseases. It can also be employed to manipulate plant microbe interaction for improving the production of crops.

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