Quorum Sensing Related Signalling Pathways

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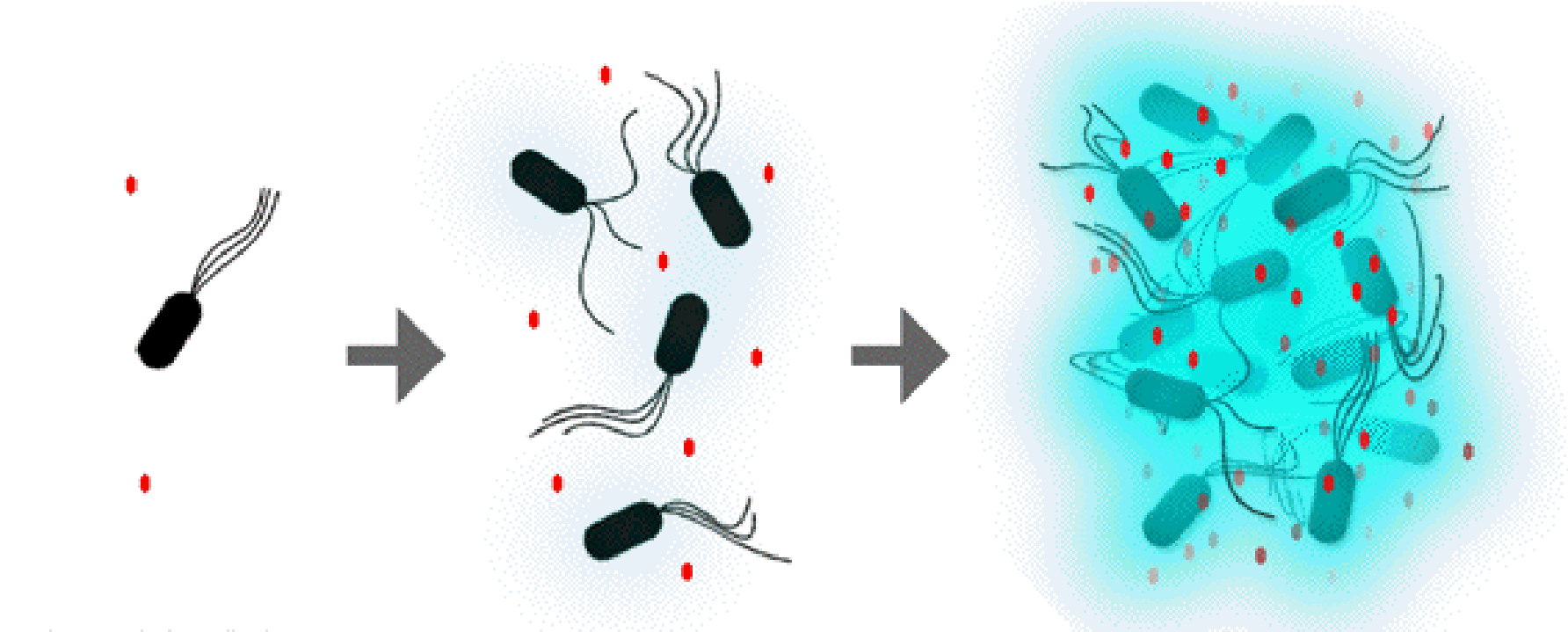
Quorum Sensing

- Bacterial cells within a population can act socially by coordinating their activities through cell-to-cell communication or ‘quorum sensing’.
- Quorum sensing (QS) regulation allows bacteria to control their gene expression in response to their population density.
- This collective behavior depends on the production and extracellular release of small signal molecules known as autoinducers.
- As the bacterial population density increases, so does the concentration of signal molecule such that the expression of quorum sensing-dependent target genes is activated (or repressed) once a critical threshold concentration of signal has been achieved (Figure 1).
- Usually processes that are regulated by QS are beneficial when a group of bacteria acts together.
- Quorum sensing permits a bacterial population to mount a cooperative response that improves access to nutrients, promotes defence against competitors, and survival in adverse environmental conditions.
- Many species of bacteria regulate several aspects of life using QS, including biofilm formation, bioluminescence, virulence, DNA exchange, etc.

Figure 1. Quorum Sensing & Bioluminescence



At low cell density the autoinducers (red dots) that are produced diffuse through the cell membrane into the growth media.

As the cell growth continues, the level of autoinducers in the media will start to accumulate in a confined environment. A very low intensity of light emission can be detected.

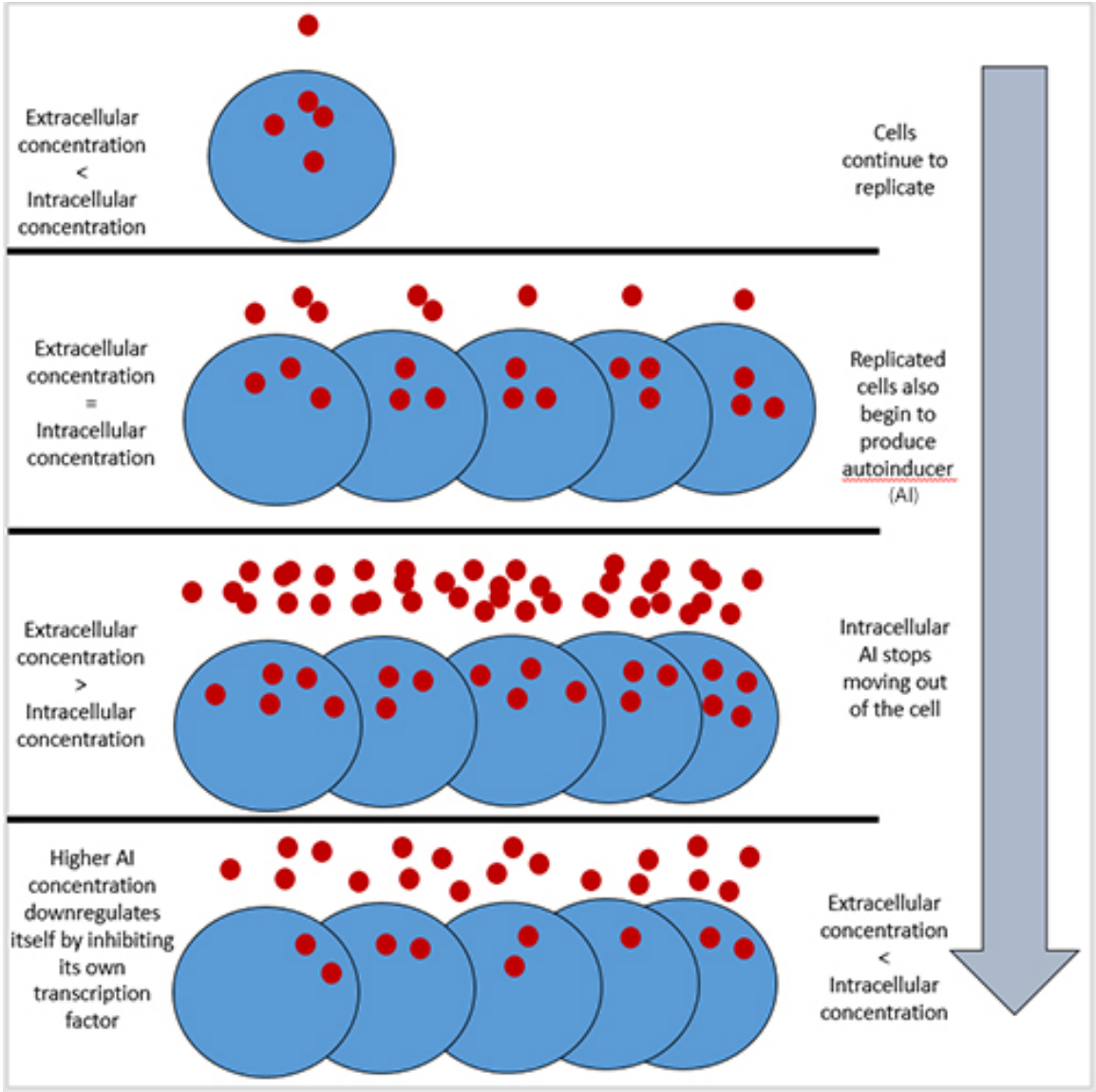
High levels of autoinducers activate the luminescent systems of the luminous bacteria in the media. The regulatory response to the autoinducer leading to induction of expression of the *lux*CDABE genes is termed "QUORUM SENSING."

Mechanism of Quorum Sensing

- During their reproductive cycle, individual bacterium synthesize autoinducers.
- Gram-negative bacteria produce acyl-homoserine lactone autoinducers that can passively diffuse through their thin cell wall.
- In contrast, Gram-positive bacterial autoinducers are made of peptide and must be actively transported through their peptidoglycan cell wall using the ATP- bind ing cassette (ABC) transporter system.
- In both cases, autoinducers move out of individual cells as they are produced.
- Since the bacteria are reproducing, there are progressively more individual cells producing autoinducers and the extracellular concentration of the autoinducers increases, eventually hitting a “critical mass.”
- That threshold makes it energetically unfavorable for intracellular autoinducers to continue to leave the cell (whether through diffusion or transport), resulting in an increase in their intracellular concentration.
- Once extracellular and intracellular concentration increases, autoinducers bind to their receptors, triggering signaling cascades that alter transcription factor activity and therefore, gene expression.
- For many bacteria, the change in gene expression includes down regulation of autoinducer synthesis in a negative feedback loop (Figure 2).

Figure 2. Overview of how quorum sensing works in bacteria

Source: W. Jon Windsor



Categories of extracellular signal molecules

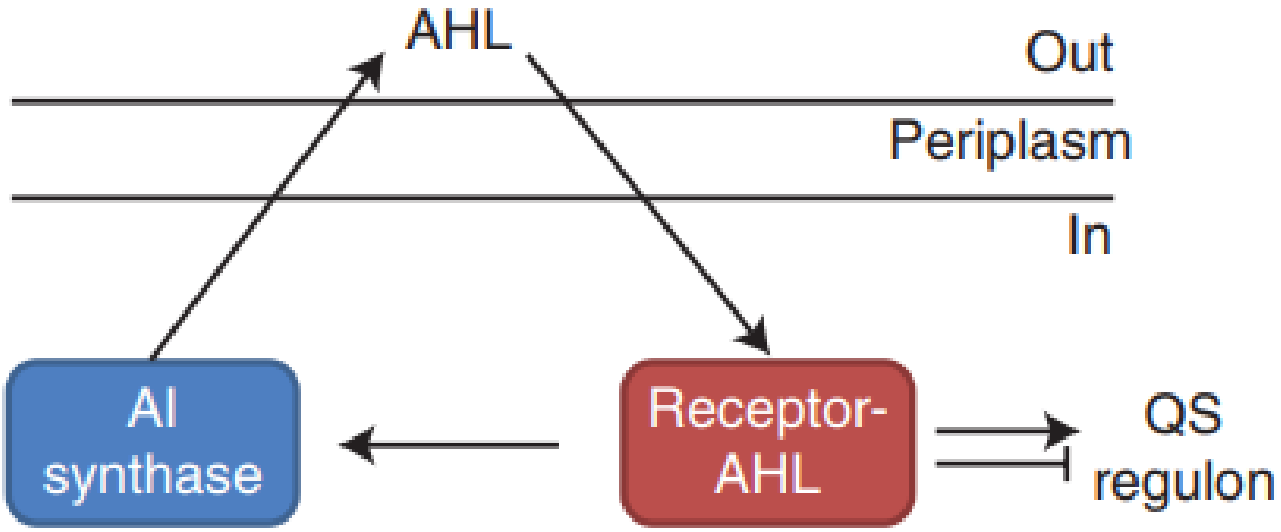
- Till today, many categories of extracellular signal molecule have been reported in bacteria .
- Most QS signals are either small organic molecules of ~ 1000 Da or peptides of 5-20 amino acids.
- Some common signal molecules utilized by Gram negative bacteria are:
  - N-acylhomoserine lactones (AHLs),
  - 2-alkyl-4(1H) quinolones (AQs),
  - long-chain fatty acids
  - fatty acid methyl esters
  - autoinducer-2 (AI-2), a collective term for a group of inter-convertible furanones derived from dihydroxypentanedione (DPD).
- AI-2 is also produced by some Gram-positive bacteria; however, usually these microorganisms prefer linear, modified or cyclic peptides such as the autoinducing peptides (AIPs)

N-Acyl-L-homoserine lactones

- AHLs is made of a conserved homoserine lactone (HSL) ring with an amide (N)-linked acyl side chain range from C4 - C18 in length.
- The variation in structure of different AHLs can be seen in acyl side chain in form of 3-hydroxy or a 3-oxo group.
- The type of AHL produced by a particular species is often strain-dependent.
- First identified in marine *Vibrio* species, *lux* type quorum sensing is based on the production of and responses to AHLs.
- In general, lux-type systems consist of 2 components, an autoinducer synthase (e.g., LuxI), which synthesizes AHLs from S-adenyosyl methionine, and a transcriptional regulator (e.g., LuxR) (figure 3).
- Because of its small size and lipophilic character, AHL freely diffuses across cell membranes.
- As the population density increases, intracellular AHL binds the functionally linked (cognate) LuxR-like receptor at a sufficient concentration within the cytoplasm to induce differential gene expression.

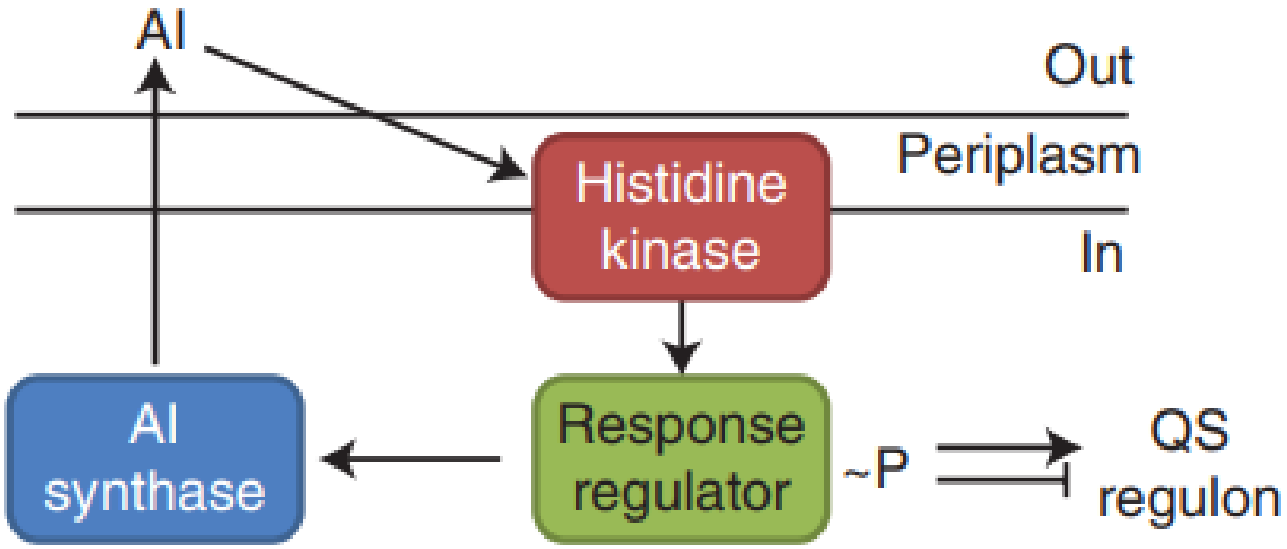
Figure 3. A LuxI/LuxR-type system

- Typically, AHL-bound LuxR-type proteins also activate luxI expression, forming a feed-forward autoinduction loop that floods the vicinity with AI.
- Numerous Gram-negative pathogens control virulence factor production using LuxI/LuxR type QS circuits.
- Some examples are LasI/LasR and RhlI/RhlR in *P. aeruginosa*, SmaI/SmaR in *Serratia marcescen*.



Autoinducers of Gram-negative Bacteria Can Also Function Through Two Component System

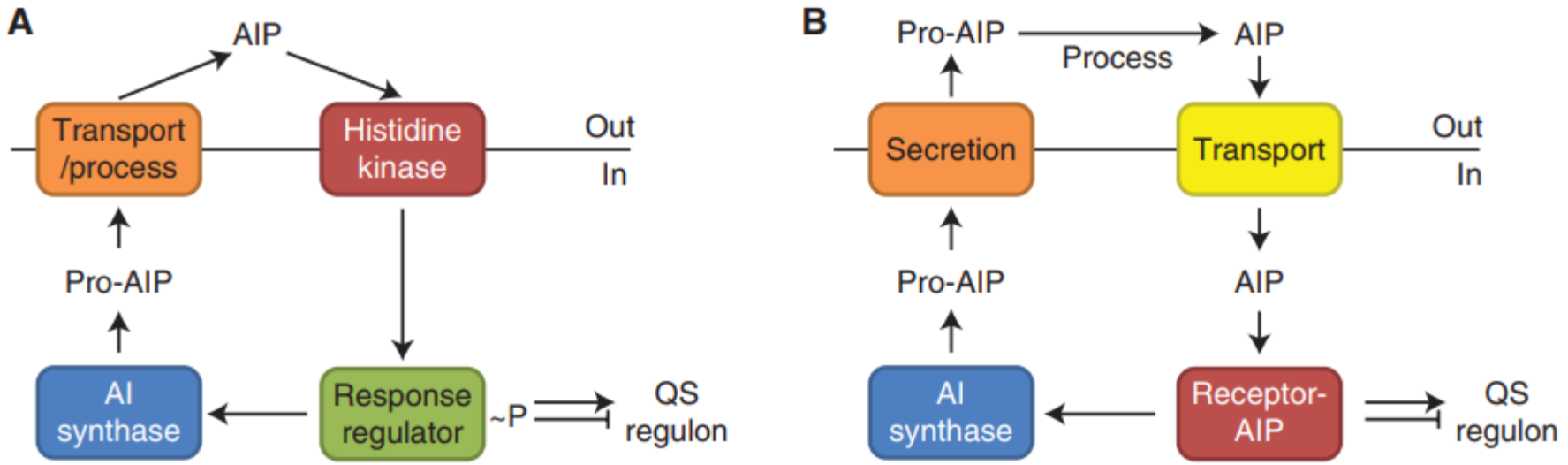
- In some cases of Gram-negative bacterial QS, autoinducers are detected by two-component histidine kinase receptors that function analogously to the process found in the Gram positive bacteria (Figure 4.).



Autoinducing peptides (AIPs)

- Gram-positive bacteria use peptides, called autoinducing peptides (AIPs), as signaling molecules.
- Once produced in the cell, release of AIP from the cell takes place through oligopeptide transporters.
- The AIP transporters also process the pro-AIPs. The final processed AIPs range in size from 5 to 17 amino acids, can be posttranslationally modified, and can be linear or cyclized.
- When the extracellular concentration of the AIP is high, which occurs at high cell density, it binds to a cognate membrane-bound two-component histidine kinase receptor.
- Usually, binding 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 (Figure 5A).
- In some cases of Gram-positive bacterial QS, AIPs are transported back into the cell cytoplasm where they interact with transcription factors to modulate the transcription factor's activity and, in turn, modulate gene expression changes (Figure 5B).

Figure 5. Autoinducing peptide (AIP) QS in Gram-positive bacteria by (A) two-component signaling, or (B) an AIP-binding transcription factor.



… Autoinducing peptides (AIPs)

- In these Gram-positive QS circuits, the pro-AIP, transporter, histidine kinase receptor, and response regulator are typically encoded in an operon.
- Expression of this operon is activated by the phosphorylated response regulator, resulting in an autoinducing feed-forward loop that synchronizes the QS response.
- Some examples of Gram-positive QS behaviors are competence in *Streptococcus pneumonia* and *Bacillus subtilis* and sporulation in *B. subtilis*.
- QS controls virulence factor production in Gram-positive human pathogens including *S. aureus, Listeria monocytogenes, Enterococcus faecalis*, and *Clostridium perfringens.*

Questions

- What is quorum sensing? Explain the major QS pathways of Gram-negative and Gram-positive bacteria.
- What is quorum sensing? Describe how it occurs and briefly discuss its importance to microorganisms.
- Write short note on:
  - Mechanism of quorum sensing
  - Autoinducers
  - N-Acyl-L-homoserine lactones signalling pathway
  - Autoinducing peptides (AIPs) signalling pathway
  - Main QS pathway of Gram-negative bacteria
  - Main QS pathway of Gram-positive bacteria.