Quorum quenching – an alternative antimicrobial therapeutics

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New millennium is witnessing an alarming increase in multi-resistant microbes due to the indiscriminate application of compounds to kill/inhibit growth of microorganisms. Consequently infections that were once treatable have become non-treatable. Microbial resistance to traditional antimicrobials and the sparse availability of novel antimicrobial agents necessitate exploration of alternative therapies. Quorum Sensing (QS) signaling systems represent highly attractive targets for the development of novel therapeutics that may prevent, suppress, and/or treat infectious diseases. The present chapter deals with the mechanisms and key components of quorum sensing. It represents an overview of quorum quenching strategies. Attempt to cite substantial studies on small quorum sensing inhibitors, their probable sources, and mechanism as novel antimicrobial agents against different pathogens has also been made.

Key words: Quorum sensing; microbial resistance; quorum quenching; antimicrobial therapeutics

1. Introduction

Invasion and colonization of one organism in another for multiplication is a natural process of evolution. It is a conventional process employed by microbes for their sustenance and survival. This forms the basis of infection in multicellular organisms. Not all invading microorganisms are infectious in nature. Some are found to lead a symbiotic association with the host cells. These may even be protective in nature. Microbes which infect and subsequently lead to fatal diseases are of major concern for human beings. Such infecting microbes include bacteria, fungi, mycoplasma, viruses, protozoa etc.

Though, several treatment strategies have been employed for their eradication the discovery of antibiotics has been a landmark in the history of medicine. With the introduction of antibiotics, a new era of therapeutics had started which promised to be a potential tool for abolition of infectious diseases caused by bacteria. Consequences of the use of antibiotics turned out to be partially fruitful. Indiscriminate use of antibiotics leads the way for the evolution of superior kind of microbes which are no longer sensitive to antibiotics i.e. antibiotic resistant microbes. Emergence of resistant microbes forced human beings to search for better and effective therapeutic alternatives that are more secure with respect to long term effect.

Intensive research in microbial systems, their organization, and mutual interaction with each other during colonization and subsequent infection, showed the presence of a kind of communication predominant in multicellular systems. This communication which is known as quorum sensing (QS) involves secretion of certain molecules and their detection. Presently the research activities are oriented towards treating diseases caused by microbial infection by disconnecting this communication- quorum quenching (QQ). The present chapter focuses on the various aspects of quorum quenching and its potential role in treatment of infectious diseases, along with the challenges faced in such approach.

1.1Basics of Microbial Infection

Infection and disease relationship of a particular microbe in a host is governed by various factors. Study of some of the underlying factors is essential to understand the mechanism of infection leading to disease. Infection involves successful multiplication of microbes in a host by utilizing host origin molecules as resources. But all infections don’t lead to diseases, even in case of a pathogenic microbe. For manifestation of disease, couple of tens of thousands to millions of microbes (bacteria) are needed i.e. a significantly high number, which makes their easy and rapid elimination by the host defense mechanism difficult. Along with it host immune status is also an essential factor which determines the extent of infection. Certain specific biological features of microbes like, presence of capsule, flagella etc. play an important role in the pathogenicity of a microbe and even determine its virulence. Sequence of steps eventually leads to infection.

Microbial infection follows the given pathway-

- **Adhesion**- This most crucial part of infection involves interaction between pathogenic microbe and host cell or tissue. It involves attachment using adhesins, fimbriae, pili etc. besides some exceptions.
- **Penetration**- In most cases adhesion is followed by penetration of host cell and/or tissue by the bacteria as found in *E.coli, Salmonella* etc.
• Multiplication- Post penetration the bacteria, take control over the host system and utilize its molecules for their reproduction, leading to host tissue damage and disease followed by their spread to newer locations and/or hosts.
• Evasion of host immune system- In order to survive in the host, the pathogenic bacteria escape the host cell immune system by employing various mechanisms like release of toxins, molecular mimicry, antibody degradation, presence of capsule etc. [1-2].

This pathogenic progress as already has been mentioned needs certain number of microbes to be present. It takes place in form of a population, not as an individual microbe. The ultimate cause for this well orchestrated activity culminating to microbial pathogenicity is a question that may be addressed by QS studies.

1.1.1 QS and Infection
Bacterial communication i.e. QS is found to be monitored by release of certain signal molecules i.e. autoinducers as a density dependent variable. The autoinducers are released by the bacterial cells into the surrounding environment. As their concentration reaches a threshold level, it leads to development of a well coordinated response. Thus, for the signal to be significant enough to generate a response a particular cell density is essential to create threshold level signal. These signals lead to activation or suppression of certain genes leading to different changes in metabolic activity, morphology, mobility, aggregation and association with other cells of same species or different species.

1.1.2 Mechanism of QS
QS is found in both Gram positive and Gram negative bacteria. It can be species specific or non-specific i.e. operate between bacterial cells of different species. The signal molecules involved in QS are found to be diverse in nature. Mostly the molecules employed are small (< 1000 Da) organic molecules or peptides with 5-20 amino acids. In case of Gram negative bacteria the signaling molecules generally used are N-acylhomoserine lactones (AHLs), 2-alkyl-4-quinolones (AQs), long chain fatty acids and fatty acid methyl esters. Gram positive bacteria employ linear, modified or cyclic peptides such as the auto inducing peptides (AIPs). Both Gram negative and Gram positive bacteria are found to use auto inducer-2 for QS which is a group of interconvertible furanones derived from dihydroxypentanedione (DPD) [3].

1.1.3 Major types of QS Mechanisms
Different bacterial species are found to communicate via mainly two types of QS mechanisms - AHL mediated and AIP mediated.

• **AHL- mediated QS** is found in Gram negative organisms, first described in *Vibrio fischeri*. It involves two proteins LuxI which synthesizes the autoinducer AHL and LuxR which is cytoplasmic autoinducer receptor/DNA-binding transcriptional activator. Initially AHL is produced in small amount which diffuses out of the cell. Once, its concentration increases and reaches threshold level with the rise in cell density the AHL binds to LuxR. This complex activates transcription of operon associated with luciferase production. LuxR-AHL complex also acts as positive feed back activator, leading to high expression of luxI gene and production of LuxI protein.

AHL mediated QS is found in several other Gram negative bacteria that are pathogenic in nature- *Pseudomonas aeruginosa, Burkholderia cepacia, Aeromonas hydrophila* etc. *P. aeruginosa* is a causative agent of nosocomial infections and found to infect patients suffering from Cystic fibrosis and immuno-compromised patients resulting in high mortality. Its pathogenicity is found to be regulated by QS mechanisms including synthesis and release of exoproteases, siderophores, exotoxins, secondary metabolites and biofilm formation. Interspecies QS has been expressed between *P. aeruginosa* and *B. cepacia* employing AHL-based mechanism.

• **AIP- mediated system:** This is generally found in Gram positive bacteria, best example being *Staphylococcus aureus*. The microbe though a normal microflora of human can be converted into deadly pathogen upon invasion into host tissue. It has unique feature of expressing different traits at varying cell densities. Factors which promote adhesion and colonization are expressed at low cell density levels and at high cell density secretion of toxins and proteases takes place. This is mediated by QS system which involves AIP encoded by agrD gene and a two component sensor kinase-response regulator pair, AgrC and AgrA, respectively. AIP is exported out of the cell by ArgB exporter protein and is modified by the same protein by addition of thiolactone ring. When threshold level is achieved AIP binds to ArgC and leads to phosphorylation of ArgA protein. Activated ArgA induces expression of RNAIII, that leads to repression of cell adhesion factors and expression of secreted factors. Similar to AHL system it also forms a positive feed back loop causing increase in AIP generation.
Thus, QS plays a key role in pathogenesis by bacteria with its influence at the genetic level leading to regulation of various factors involved in the process. Similar kind of observations has been detected in many other human opportunistic pathogens like *Serratia liquefaciens* and *Chromobacterium violaceum* and even in several plant pathogens. So, the treatment of bacterial diseases can be developed based on this pathway rather than the conventional method of targeting the vital survival mechanisms of pathogen [4-6].

1.2 Conventional Antimicrobial Therapy

Conventional treatment follows use of antibiotics, which function either by inhibiting the growth of bacteria or by killing the bacteria. They are derived from moulds or fungi, bacteria and/or are synthetic or semi-synthetic compounds. These antibiotics are mostly used topically or internally. Depending upon their host range, the antibiotics can be divided into broad spectrum or narrow spectrum. Penicillin is a narrow spectrum antibiotic mainly used for Gram negative bacteria, whereas tetracycline is broad spectrum antibiotic with effect on both Gram positive and Gram negative bacteria.

Antibiotics may also be categorized according to their major mode of action, which are:

- Interference with cell wall synthesis.
- Protein synthesis inhibition.
- Interference with nucleic acid synthesis.
- Inhibition of a metabolic pathway.

The mode of action of different representative antibiotics is given in Table 1[7-8].

<table>
<thead>
<tr>
<th>Mode of Action</th>
<th>Antibiotics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Interference with cell wall synthesis</td>
<td>β-Lactams and Glycopeptides</td>
</tr>
<tr>
<td>Inhibition of protein synthesis:</td>
<td></td>
</tr>
<tr>
<td>Bind to 50S ribosomal subunit</td>
<td>Macrolides, Chloramphenicol etc.</td>
</tr>
<tr>
<td>Bind to 30S ribosomal subunit</td>
<td>Aminoglycosides, Tetracyclines</td>
</tr>
<tr>
<td>Bind to bacterial isoleucyl-tRNA synthetase</td>
<td>Mupirocin</td>
</tr>
<tr>
<td>Interference with nucleic acid synthesis:</td>
<td></td>
</tr>
<tr>
<td>Inhibition of DNA synthesis</td>
<td>Fluoroquinolones</td>
</tr>
<tr>
<td>Inhibition of RNA synthesis</td>
<td>Rifampin</td>
</tr>
<tr>
<td>Inhibition of metabolic pathway</td>
<td>Sulfonamides, folic acid analogues</td>
</tr>
</tbody>
</table>

1.3 Challenges of Antibiotic Treatment

The main challenge in antibiotic treatment is development of resistance. This refers to the situation where the prescribed dose of drug that usually destroys the bacteria fails to do so, making the disease clinically untreatable. This has resulted in re-emergence of once considered eradicated diseases and infections, with greater virulence which has raised the mortality and morbidity. One of the major causes of resistance is overuse and misuse of antibiotics.

Resistance can be classified into three types- intrinsic, acquired and genetic resistance. Intrinsic resistance involves the mechanisms which are present in certain bacteria naturally. For instance *Streptomyces* naturally carries some genes which give protection against its own antibiotic. Acquired resistance is observed in some bacterial species in presence of antibiotics and is found to be unstable in nature without any change in the genotype. Genetic resistance involves changes in genes due to mutations or acquisition of mobile genetic materials, like plasmids, transposons with antibiotic resistance genes. This can also be transmitted by phages during transduction.

Different mechanisms of antibiotic resistance are:

- Reduced permeability - in this case bacteria prevent the antibiotic from entering and accessing the target molecule. The membrane proteins which are responsible for transport of antibiotics acquire mutations and resist their passage. Example, in case of *P. aeruginosa* resistance to imipenem occurs by lack of specific D2 porin. Similarly, *Neisseria gonorrhoea* porin can acquire mutations which leads to resistance to penicillin and tetracycline.
Increased efflux- increase in efflux of antibiotics from interior of bacterial cells has been found in enterobacteriaceae showing resistance against tetracycline.

Enzymatic inactivation- best example is β-lactamase enzyme which can cleave β-lactam ring present in certain antibiotics rendering the bacteria resistant. These enzymes are found to be effective against penicillin and cephalosporin. Another example is aminoglycoside-inactivating enzymes which inactivate antibiotics by addition of acetyl, adenyl and phosphoryl groups.

Alteration in target- in this case the antibiotic gains access to the target but fails to act on it due to structural changes in the target. Example of this is shown by vancomycin resistance in Gram positive bacteria. Vancomycin prevents cross-linking of peptidoglycan by binding to D-Ala-D-Ala dipeptide of muramyl peptide. But in case of resistance D-Ala-D-Ala gets changed to D-Ala-D-lactone which does not bind vancomycin.

“Bypass” pathway- in this mechanism bacterium produces an alternate target molecule that is resistant to inhibition by antibiotic and simultaneously it continues to produce the original target molecule. Methicillin resistant S. aureus (MRSA) produces alternative penicillin binding protein (PBP2a) in addition to normal protein. This PBP2a is not inhibited by antibiotics like fluclaxacin and the cell continues to produce normal peptitoglycan. Thus, healthy cell wall synthesis takes place even in presence of antibiotics [8-10].

It is clearly indicated that under stressed conditions, selected bacteria develop varied strategies for survival that ultimately leads to development of resistant strains. Certainly the requirement of present medical therapeutics and research is to look beyond antibiotics, and search for alternatives which can regulate the virulence rather than killing or growth inhibition of bacteria. Opposed to conventional antibiotics, anti-pathogenic drugs would prove to be much safer, where inhibition of QS promise to play an active role.

2. QQ an alternative approach

QS Signalling systems of microbial pathogens are central regulators for expression of virulence factors. They represent highly attractive targets for development of novel therapeutics to prevent microbial infections. The use of QS based drugs to attenuate/combat bacterial pathogenicity rather than bacterial growth is particularly enticing with the emergence of increased antibiotic resistant bacteria. The obvious strategy is to prevent the signal molecules involved in QS i.e. signal generator molecule and signal receptor.

2.1 Discovery of QQ

Antibiotics kill/stop the microbial growth by interference with the essential metabolic process eventually leading to antibiotic resistance in microbial pathogens. This finding about conventional antibiotics has intrigued the scientists even more. Tapping QS based naturally occurring well evolved mechanisms has aroused significant fundamental and biotechnological research interests required to target activities of deleterious bacteria. In the quest for QS inhibitors, studies have demonstrated that eukaryotes mainly plants and bacteria themselves are anti QS substances and show significant promise. Hence an in depth activity in this area to unravel the identity and potential of similar analogues is the prime focus.

2.2 Prospective and demonstrated QQ strategies

QQ strategies mainly employ disruption of signal molecules i.e. generation, the molecule itself and interference at the reception level. For an improved understanding the demonstrated QQ strategies may be enlisted as given below:

- QQ by small quorum-sensing inhibitors (QSI).
- QQ by AHL-lactonase.
- QQ by AHL-acylase.
- QQ by paraoxonase enzymes.

2.2.1 Small QSIs

The inhibitors of QS can be roughly grouped into two categories according to their structures and functions. One group consists of molecules which structurally mimic QS signals, like halogenated furanones and synthetic AIPs that are similar to AHL and AIP signals, respectively. These inhibitors interfere with the binding of corresponding signal to the receptor or decrease the receptor concentration.

The other group of small chemicals include enzyme inhibitors. For example, triclosan a potent inhibitor of the enoyl-ACP reductase that is involved in the synthesis of acyl-ACP, one of the essential intermediates in AHL biosynthesis, reduces AHL production and closantel is a potent inhibitor of histidine kinase sensor of the two-component system.
2.2.2 AHL-lactonase mediated QQ

AHL-lactonases, hydrolysing the homoserine lactone ring of AHL signals, have been identified from a range of bacterial species. Crystal structure of AHL-lactonase shows that the enzyme contains two zinc ions in the active site. It is a metalloprotein. Residues directly involved in metal coordination are completely conserved in all AHL-lactonases. A catalytic mechanism of AHL-lactonase involves the following:

Step 1: Attack of the substrate’s carbonyl carbon by a nucleophilic water/hydroxide bridging the two Zn$^{2+}$ ions.
Step 2: Formation of negatively charged intermediate stabilized primarily by interactions with Zn ion.
Step 3: Formation of open ring product by breaking of C-O bond of lactone ring of AHL.

Compared to AHL-acylase and PON enzymes with variable substrate spectra, AHL-lactonase is by far the most specific AHL degradation enzyme. It hydrolyses both short- and long-chain AHL signals with similar efficiency, but has little or no residual activity to other chemicals, including non-acyl lactones and aromatic carboxylic acid esters.

2.2.3 QQ by AHL-acylase

Several bacterial species, (*Variovorax paradoxus*, a *Ralstonia* isolate, *P. aeruginosa* PAO1 etc.) have been reported to encode AHL-acylase. These degrade AHL signals by hydrolysing the amide bond of AHLS and by producing corresponding fatty acids and homoserine lactone.

AHL-acylases obtained from varied microbial sources exhibit varying efficiency of long chain AHL and short chain AHL degradability. Also, substrate specificity for them is different. Some degrade penicillin G and ampicillin effectively while others fail even with structural similarity. Mutagenesis and crystal structure analysis of these enzymes would be critical for predicting the molecular mechanisms involved in substrate specificity.

2.2.4 QQ by paraoxonase enzymes

First observation of strong AHL inactivation activity was done in human epithelial cells. Later, it was found to be widely conserved in the sera of six mammalian species—human, rabbit, mouse, horse, sheep and bovine. The characteristics of these AHL inactivation enzymes depend on Ca$^{2+}$ ion and lactonase-like activity. These paraoxonase enzymes (PONs), like PON1, PON2 and PON3, exhibit a wide range of physiologically important hydrolytic activities, including drug metabolism and organophosphate detoxification. Some displayed strong AHL degradation activity. These PON enzymes seem to be most active with long-chain AHL signals but less efficient with short chain AHL signals. As is the case with AHL-lactonase these enzymes also hydrolyse the homoserine lactone ring of lactone signals.

Interestingly, PON enzymes, particularly PON1, are known to catalyse the hydrolysis of many synthetic chemicals including organophosphate insecticides, nerve agents, aromatic carboxylic acid esters, cyclic carbonate esters, aromatic lactones and alkyl lactones. The physiological substrates for these proteins have not been identified. As the host pathogen interactions are ubiquitous, the AHL quorum sensing signals are cited as the first index of natural substrates of these fascinating PON enzymes.

The catalytic reaction involves:

Step 1: Deprotonation of a water molecule to generate a hydroxide ion.
Step 2: Nucleophilic attack at the ester carbonyl centre of the substrates that results in production of an oxy anionic intermediate. The negative charge of the resulting intermediates is probably stabilised by the catalytic calcium.
Step 3: The C-O bond of the ester intermediate breaks down.

Some of these enzymes are able to hydrolyse a wide range of substrates and show at least three types of enzyme activity, i.e. organophosphatase, arylesterase and lactonase. The chemical structures of the substrates are different. Hence, it is intriguing as to whether the enzyme uses the same mechanism for catalysis of the varied substrates or does it differ. These enzymes not only share overlapping substrates, but also display distinct substrate specificities [11-12].

2.3 Potential QQ/QSIs as therapeutic targets

The potency of molecules interfering with QS signals for use as anti-pathogens a probable novel therapeutic mode necessitates search for increased number of molecules for use. Requisite traits in molecules for use as QSI have been dealt with, which include:

- Low molecular weight molecule.
- Activity causing a significant reduction in expression of QS controlled genes.
- Inhibitor activity exhibiting a high degree of specificity for QS regulators without toxic side effects on either the bacteria or the eukaryotic host.
- Chemical stability and resistance to metabolism and disposal by higher host organisms.
2.3.1 Screening strategies for QSI

Several QSI screening strategies have been adopted/employed from time to time such as, use of AHL biosensor, QSI selector based screening, pigment based qualitative and quantitative QSI assay, anti-swarming based screening tests etc. Undermentioned is a list of extracts tested for QSI activity and their potential applications in Table 2.

<table>
<thead>
<tr>
<th>Plant Species (Family)</th>
<th>Part</th>
<th>Medicinal use</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polygonatum odoratum</td>
<td>Underground stem</td>
<td>Treatment of fever, common cold, sore throat, rheumatism etc.</td>
</tr>
<tr>
<td>Prunus armeniaca (Rosaceae)</td>
<td>Kernel of seed</td>
<td>Treatment of coughs, bronchial asthma, rheumatism, constipation etc.</td>
</tr>
<tr>
<td>Prunella vulgaris (Labiatae)</td>
<td>Stem, leaves, flower and fruit</td>
<td>Treatment of fever, headache, inflammation of eyes, dizziness etc.</td>
</tr>
<tr>
<td>Nelumbo nucifera (Nymphaeaceae)</td>
<td>Leaves</td>
<td>Treatment of sunstroke, diarrhea, dysentery, fever, dizziness etc.</td>
</tr>
<tr>
<td>Lycium chinense (Solanaceae)</td>
<td>Fruit</td>
<td>Treatment of anemia, back ache, vision problems, chronic cough, dizziness etc.</td>
</tr>
<tr>
<td>Punica granatum (Punicaceae)</td>
<td>Bark</td>
<td>Treatment of sore throat, vaginal discharge, bad breath etc.</td>
</tr>
<tr>
<td>Areca catechu (Palmae)</td>
<td>Seed</td>
<td>Treatment of diarrhea, indigestion, expels tapeworms and roundworm</td>
</tr>
<tr>
<td>Imperata cylindrical (Gramineae)</td>
<td>Underground stem</td>
<td>Stops bleeding, acute inflammation of kidney, cough with phlegm etc.</td>
</tr>
<tr>
<td>Terminalia catappa</td>
<td>Leaf</td>
<td>Treatment of cough, fever etc.</td>
</tr>
</tbody>
</table>

2.3.2 QSI compounds against microbial infections:

Quorum sensing inhibitors i.e.QSI compounds maybe: i) Natural QSI or ii) Synthetic QSI.

**Natural QSI:** QSI compounds can be isolated from natural sources such as plants and fungi. Some of them are expected to produce QSI compounds since, both plants and fungi have co-existed with QS bacteria for millions of years. Penicillium species produce secondary metabolites with QSI activity. Two of the compounds with QSI activity have been identified as penicillic acid (PA) and patulin. These two compounds target the Las R and Rh1R QS regulators. Brominated furanones from the alga *Delisea pulchra* are the group of compounds that exhibit QSI effect. Their QSI effect may be attributed to the binding of blockers to the receptors which induces conformational changes eventually leading to destabilization of the receptor. They prevent bacterial colonization, and thereby macrofouling, by interfering with QS-controlled motility. One example of this is the ability of halogenated furanones to inhibit the QS-controlled swarming phenotype of *Serratia liquefaciens* MG1.

Plants like carrot, soybean, water lily, tomato, pea seedlings, habanero (chilli) and garlic have been found to produce compounds capable of interfering with bacterial QS. Garlic extract contains minimum of three different QS inhibitors. One of it has been identified to be a cyclic disulphur compound, which exerts a strong antagonistic effect on *LuxR*-based QS but, interestingly, has no effect against *P. aeruginosa* QS.

**Synthetic QSI:** There are basically three ways to block QS by developing on the AHL scaffold:
- Introduction of substitutions in the acyl side chain without any change in the lactone ring.
- Introduction of substitutions and alterations in the lactone ring with unchanged acyl side chain.
- Thirdly, extensive modifications in both the acyl side chain and the lactone ring.

Groups of agonistic AHL analogues carry an acyclic or cyclic alkyl substituent on the outmost carbon atom of the side chain. Replacement of the C-3 atom with sulphur in the acyl side chain generates analogues which block expression in both LuxR and LasR-controlled QS reporters.

Other successful QSI compounds are obtained by placing aryl substituents at the end of the side chain – at least in the 3-oxo-C6 HSL/LuxR-controlled system. Antagonist effect of the compound is dependent on the size of the substituent. Replacement of C-1 carbonyl group of the side chain with a sulphonyl group in aryl AHLs have shown to further enhance the QSI effect. Exchanging the hexanone ring for a phenolic ring, result in the generation of a potent QSI compound.

QSI have also been identified by screening of random compound libraries. Several QSI compounds with structures unrelated to the signal molecules, including 4-nitro-pyridine-N-oxide (4-NPO), indole, p-benzoquinone, 2,4,5-
tribromoimidazole, indole and 3-nitrobenzene sulphone amide, have also been found to exert similar QSI effects. Undermentioned is the list of QQ molecules against microbial infections enlisted in the Table 3.

<table>
<thead>
<tr>
<th>QQ Molecules</th>
<th>Effect on QS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macrolides</td>
<td>Reduction in autoinducers (LasI and RhlI) in <em>P. aeruginosa</em></td>
</tr>
<tr>
<td>RNAIII-related compounds</td>
<td>Inhibition of action of RNAIII-activation protein in <em>staphylococci</em></td>
</tr>
<tr>
<td>Halogenated furanones</td>
<td>Inhibition of LuxR-dependent gene expression in Gram negative bacteria</td>
</tr>
<tr>
<td>Homoserine lactonases</td>
<td>Degradation of HSL signal molecules</td>
</tr>
<tr>
<td>HSL analogue</td>
<td>Antagonism of HSL activity</td>
</tr>
<tr>
<td>HSL vaccine</td>
<td>Reduced pulmonary TNF-α aggregation in pneumonia</td>
</tr>
</tbody>
</table>

### 2.3.3 Additional beneficial effects of QSI

A known fact is that bacteria living in biofilm mode of growth are often more tolerant to antibiotics, biocides and heavy metals. Concentration of antibiotics to eradicate them is too high thus precluding efficient antibiotic treatment. QSI have been reported to be beneficial in therapeutic treatments as they have been found to render the biofilms susceptible to antimicrobial therapy. Biofilms treated with furanones were eradicated by tobramycin and dispersed by detergents. Garlic extract, patulin, PA have also been found to make *P. aeruginosa* more susceptible to tobramycin.

### 2.4 Status of QQ based therapy

QQ molecules have proved to be valuable tools in addressing both basic and conceptional questions. QS is evident in many common human pathogens. *P. aeruginosa* and *S. aureus* represent two important pathogens associated with healthcare acquired infections. Subsequent production of toxins of both these strains elicits a significant response. Table 4 given below represents examples of QQ molecules against microbial infections [12-16].

<table>
<thead>
<tr>
<th>QQ Molecules</th>
<th>Host</th>
<th>Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>AHL-lactonase</td>
<td><em>P. aeruginosa</em></td>
<td>Decreases production of elastase, hydrogen cyanide and pyocyanin, and inhibits bacterial swarming</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>Attenuates pathogenicity of <em>E. carotovora</em></td>
</tr>
<tr>
<td></td>
<td><em>Burkholderia thailandensis</em></td>
<td>Reduces bacterial swarming and twitching motility, prevents β-haemolysis of sheep erythrocytes</td>
</tr>
<tr>
<td>Paraoxonase</td>
<td><em>P. aeruginosa</em></td>
<td>Serum containing it prevents biofilm formation</td>
</tr>
<tr>
<td>AHL-acylase</td>
<td><em>P. aeruginosa</em></td>
<td>Decreases swarming ability, elastase and pyocyanin production, and attenuates nematode paralysis</td>
</tr>
<tr>
<td>Synthetic AIP-II</td>
<td>Mouse</td>
<td>Treated mice show resistance to <em>S. aureus</em> infection</td>
</tr>
<tr>
<td>3-oxo-C12-(2-aminocyclohexanone) furanone</td>
<td><em>P. aeruginosa</em></td>
<td>Reduces production of virulence factors and biofilm formation</td>
</tr>
<tr>
<td></td>
<td>Mouse</td>
<td>Attenuates the virulence of <em>P. aeruginosa</em> in mouse models</td>
</tr>
</tbody>
</table>
2.5 Conclusion:

Whether QQ based therapies offers a new hope in the persistent fight against multiantibiotic resistant bacteria is not apparent but initial search results appear enticing. Interference of QS not only down regulates expression of many virulence factors but impart additional advantages in many cases. On the other hand QSIs are not miracle bullets and a complete eradication of disease, based solely on QQ based therapy may not prove a success. However, combinatorial chemotherapeutic treatment of antibiotics with anti-pathogens seems to open up new vistas of treatment for chronic infections caused by QS regulated infections. Future activities will reveal whether QSI alone is a better anti-pathogenic drug treatment or complete bacterial eradication may be envisaged with synergistic action of multiple bioactive compounds.

References


