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Role and Regulation of Bacterial LuxR-Like Regulators

Jing Chen and Jianping Xie*

Institute of Modern Biopharmaceuticals, Key Laboratory of Ministry of Education Eco-Environment of the Three Gorges Reservoir Region, School of Life Sciences, Southwest University, Chongqing, China

ABSTRACT

LuxR transcriptional regulator is a key player in Quorum Sensing (QS), coordinates the expression of a variety of genes, including those encoding virulence factors and antibiotics biosynthesis, motility, nodulation, plasmid transfer, bioluminescence, and biofilm formation. The characteristics and roles of this family, especially those of *Mycobacterium*, are summarized in this paper to give clues for drug target discovery. J. Cell. Biochem. 112: 2694–2702, 2011. © 2011 Wiley-Liss, Inc.

KEY WORDS: LUXR; QUORUM SENSING; REGULATION; *MYCOBACTERIUM*

LuxR family proteins were firstly described by Engebrecht in 1983 when they analyzed the Quorum Sensing-related genes [Engebrecht et al., 1983], which function as pheromone receptors and transcriptional regulators [Fuqua and Greenberg, 2002]. LuxR protein family is under intensive study [Yang et al., 2009] largely due to their important roles in quorum sensing (QS).

Quorum sensing (QS) is a way for cell–cell signaling [Rui et al., 2008]. Bacteria can secrete signal molecule called autoinducer (AI), and reconcile themselves with the ambient AI concentration [Hardman et al., 1998; Rui et al., 2008; Patankar and Gonzalez, 2009]. At high cell density, AI accretes with the bacterial cell population. Multiple target genes remain to be regulated until the AI concentration reaches a threshold [Patankar and Gonzalez, 2009], such as extracellular protease, extracellular polysaccharide (EPS), bioluminescence, swarming, antibiotic biosynthesis, biofilm formation, mobility, plasmid conjugal transfer, and virulence production [Jobling and Holmes, 1997; Hardman et al., 1998; McCarter, 1998; Shao and Hor, 2001; Kirke et al., 2004; Case et al., 2008]. These events are essential for bacteria survival and propagation.

LuxI/R signal circuits regulate most gram-negative bacterial QS. LuxI is an AI synthase, while LuxR is a regulatory protein. At low cell density, although LuxI synthesis AI, the concentration of AI between environment and intracellular tend to reach a balance via diffusion or active transport, thereby repressing the expression of target genes. At high cell density, AI concentration reaches a threshold and forms an active complex with LuxR homolog; this complex binds to specific promoter sequences called *lux-boxes* controlling the transcriptional activities of the promoter [Fuqua and Greenberg, 2002], which allows bacteria to coordinate their behaviors that are conducive to survival and propagation in hostile environment. Meanwhile, LuxR-AI complex initiates the LuxI, promoting the AI synthesis, comprising a feedback regulation (Fig. 1). LuxR family proteins affect both the survival and propagation of bacteria, and the virulence and biofilm formation [Alonso-Hearn et al., 2010]. Understanding the mechanism of action of LuxR regulators underlies further efforts for better drug targets against pathogens, such as *Mycobacterium tuberculosis*.

AI can bind to LuxR family regulators. Many bacterial AI structures are available (Table I). A typical feature can be found from these structures. An analog competitively binding to LuxR to neutralize QS strategy based on AI structure was reported recently and might herald new avenue to fight pathogens [Chen et al., 2011].

THE STRUCTURE OF LuxR-TYPE PROTEINS

LuxR-type proteins have a characteristic structure, a carboxyl-terminal module to bind specific DNA sites near target promoters and an amino-terminal module to bind AIs [Fuqua et al., 1996; Patankar and Gonzalez, 2009]. The carboxy-terminal domain has a helix-turn-helix responsible for multipolarity and binding promoters [Fuqua, 2006; Patankar and Gonzalez, 2009]. The amino acids stretch between 193 and 220 at C terminal is important for DNA binding, while other amino acids play a key role in the activation

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*Correspondence to: Jianping Xie, Institute of Modern Biopharmaceuticals, Key Laboratory of Ministry of Education Eco-Environment of the Three Gorges Reservoir Region, School of Life Sciences, Southwest University, Chongqing 400715, China. E-mail: jianpingxiefudan@gmail.com

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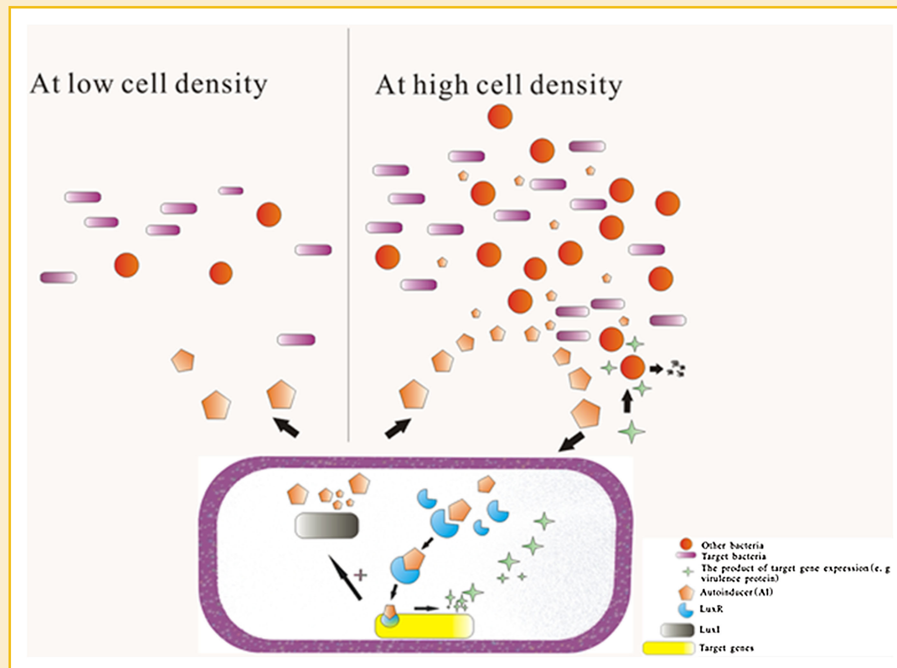

Fig. 1. The scheme of quorum sensing. Left, at low cell density, the bacterial autoinducer synthase (LuxI) produces signal molecules, the target genes do not transcribe. Right, when at high cell density, the AI concentration reaches threshold, it interacts with the LuxR homolog, which then regulates transcription of the target genes.

of *lux* operon. These domains are conservative in the LuxR family (Fig. 2).

Many sequenced proteobacterial genomes have QS-related sensors which lack homologous LuxI AI synthase [Case et al., 2008]; the unpaired LuxR-family proteins are designated as orphan LuxR homologs [Fuqua, 2006]. However, there is some controversy as to the appellation of these LuxR family members. Some hold that "solo" was more appropriate to describe this novel burgeoning family of LuxR proteins [Subramoni and Venturi, 2009]. Unlike synthase-associated LuxR proteins, LuxR solo cannot regulate the synthesis of signal molecules directly, but can interact with them to expand the existing bacterial regulatory network [Patankar and Gonzalez, 2009] to better sharing or competing resources with other bacteria.

LuxR PROTEINS

LuxR-type proteins can be grouped into three classes biochemically [Yang et al., 2009]. Class I requires AI for folding, and once folded,

TABLE I. The Structures of AI

Autoinducer	Structure	Organism	References
N-(3-oxohexanoyl) homoserine lactone		*V. fischeri*	Eberhard et al. [1981]
N-(3-oxooctanoyl) homoserine la-ctone,		*A. tumefaciens*	Zhang et al. [1993]
N-3-oxododecanoyl homoserine lactone		*Pseudomonas aeruginosa*	Pearson et al. [1994]
N-butyryl-L-homoserine lactone		*Pseudomonas aeruginosa*	Pearson et al. [1994]
N-(3-hydroxybutanoyl) homoserine lactone		*Vibrio harveyi*	Cao and Meighen [1989]

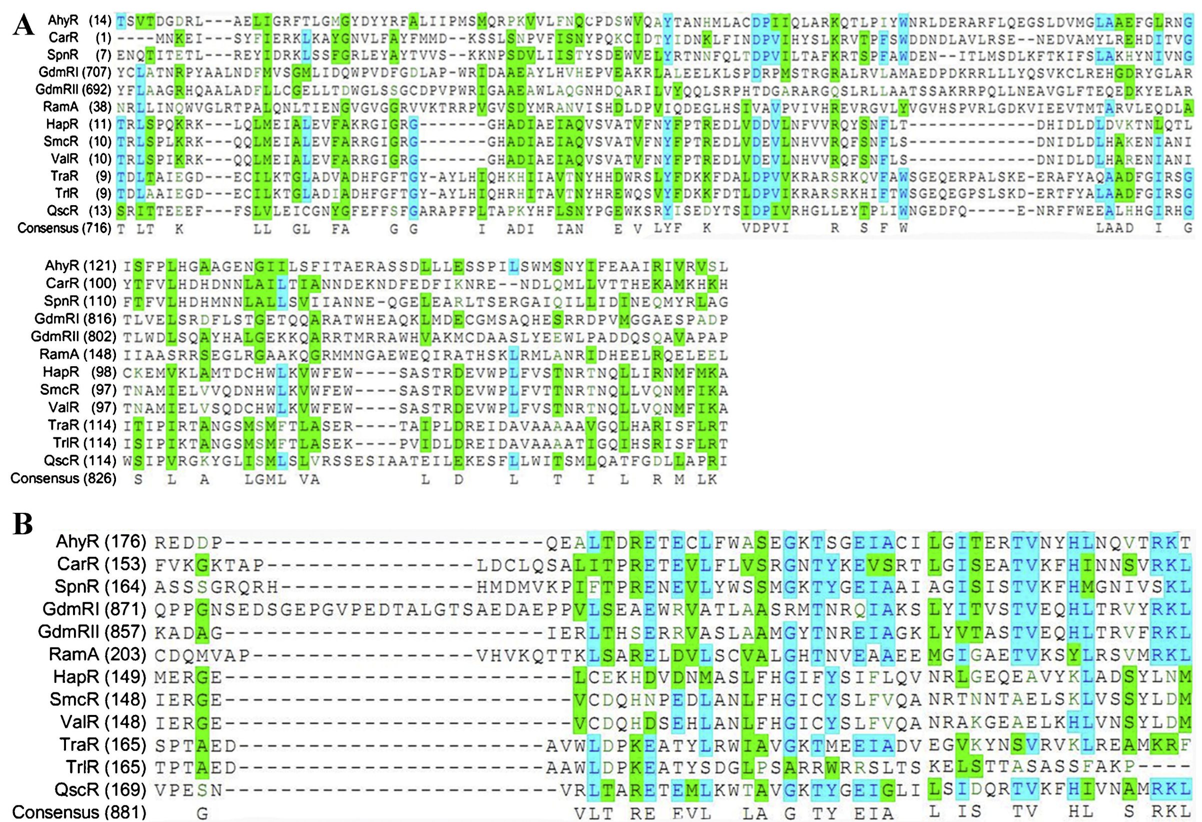

Fig. 2. The Amino acid sequence alignment of LuxR family regulator. A: The autoinducer-binding domain. (B) The HTH DNA-binding motifs at the C terminal of LuxR family regulator.

can bind to AI tightly, such as LasR and TraR of *Pseudomonas aeruginosa* [Schuster et al., 2004]. Class II requires AI for folding, binding to AI reversibly, such as *V. fischeri* LuxR [Urbanowski et al., 2004] and *P. aeruginosa* QscR [Lee et al., 2006]. Class III binds to AI reversibly, and AI is dispensable for folding, such as *Erwinia chrysanthemi* ExpR [Castang et al., 2006] and *Pantoea stewartii* EsaR [Minogue et al., 2005]; most class III LuxR family proteins function as inhibitor.

LuxR-regulated target genes can be divided into three chasses based on their binding affinity, namely the concentration of the regulators [Pompeani et al., 2008]. Class I promoters have the lowest affinity for LuxR; therefore, the LuxR concentration requirement is the highest. Class II genes need intermediate level of LuxR concentration. Class III genes require the lowest concentration of LuxR due to their promoters' highest affinity for LuxR [Waters and Bassler, 2006].

The activity of LuxR regulators is induced by bacterial signal molecules. Most LuxR regulators require homologous signal molecules for activities, but some can fold, dimerize, and bind to DNA even without signal molecules. Several LuxR regulators are reported to be suppressed by signal molecules [Tsai and Winans, 2010]. In *Pantoea stewartii*, OHHL is the AI of transcriptional regulator EsaR; although it has no effect on binding affinity, it can prevent EsaR from binding to its binding sites [Minogue et al., 2002]. In addition, OHHL alters the intrinsic fluorescence of EsaR tryptophan residues and increases EsaR thermostability [Minogue et al., 2002]. In *Pectobacterium carotovorum* and *Erwinia chrysanthemi*, OHHL blocks activation of ExpR–DNA complexes in vivo and disrupts it in vitro [Cui et al., 2005]. However, the mechanism remains to be determined.

THE REGULATION OF LuxR PROTEINS

THE REGULATION OF LuxR PROTEINS IN QS

The mechanisms that LuxR proteins regulate the expression of genes are rather complex. LuxR can be a node of more sophisticated network via interacting with other transcription regulators. Interesting reader can refer to two excellent reviews [Patankar and Gonzalez, 2009; Subramoni and Venturi, 2009] for diverse well-characterized orphan LuxR regulators in various Gram-negative bacteria such as TrlR and TraR of *Agrobacterium tumefaciens*, QscR of *Pseudomonas aeruginosa*, VirR/ExpR2 of *Erwinia* species, CarR of *Erwinia* and *Serratia* species, BisR of *Rhizobium leguminosarum*, SdiA of *Salmonella enterica* and *Escherichia coli*, XccR of *Xanthomonas campestris* and OryR of *Xanthomonas oryzae*, Orphan LuxR homologs of *Sinorhizobium meliloti*, AviR and AvhR of *Agrobacterium vitis*, VjbR and BlxR of *Brucella melitensis*, and Orphan LuxR homologs of *Burkholderia* species. The distribution

and regulation of Other LuxR members in *Mycobacteria* are elaborated in this paper (Table II).

ValR of Vibrio alginolyticus. *Vibrio alginolyticus* is a fish pathogen. Antibiotics were used to prevent the *V. alginolyticus* infection. However, antibiotics resistance and the potential damage of the residual antibiotics to human beings via food chains are great concerns. *V. alginolyticus* ZJ-51 can undergo colony phase variation between translucent/smooth (Tr) and opaque/rugose (Op). The quorum-sensing master regulator ValR is a homolog to *V. harveyi* LuxR mediating the transition. To investigate the function of ValR, an in-frame deletion of *valR* was constructed in both Op and Tr backgrounds. The mutants in both backgrounds had an intermediate colony morphotype, namely not fully translucent/smooth nor less opaque/rugose. The motility also displayed an intermediate level. However, the biofilm formation of both mutants was seriously attenuated and polar flagella disappeared. Most genes involved in polysaccharide and flagellum biosynthesis were downregulated in the mutant of Tr background but upregulated in the mutant of Op background. This suggests that ValR might regulate the flagellum biosynthesis, thereby controlling *V. alginolyticus* biofilm formation. ValR also affect the expression of genes involved in colony phase variation [Chang et al., 2010].

RamA of Corynebacterium glutamicum. *Corynebactericum glutamicum* is a high-GC content Gram-positive soil bacterium and is an industrial producer of amino acids for flavor enhancers and animal feed additives, such as L-glutamate and L-lysine [Eikmanns et al., 1993; Stephan et al., 2001]. Oxygen depletion can hinder the *C. glutamicum* growth but without apparent effect on the secretion and production of organic acid and ethanol [Okino et al., 2005].

D-Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) is a key enzyme for carbohydrate metabolism. It can catalyze the oxidation of D-glyceraldehyde-3-phosphate into 1,3-diphosphoglycerate [Toyoda et al., 2009]. *C. glutamicum gapA* and *gapB* encode GAPDH. GapA is essential for glycolysis, and GapB is important for gluconeogenesis and dispensable for glycolysis [Toyoda et al., 2009]. In carbohydrate phosphotransferase system, SugR is a transcriptional repressor [Engels and Wendisch, 2007; Tanaka et al., 2008] that can repress the expression of *gapA* in the absence of sugar [Toyoda et al., 2008]. *gapA* expression of *sugR* deletion mutant remains to be induced in the presence of glucose, as that of the wild-type strain, suggesting other transcriptional regulators might be involved [Toyoda et al., 2008]. SugR, RamA, GntR1, and GlxR can bind to the *gapA* promoter regions [Toyoda et al., 2009]. To confirm the roles of these transcriptional regulators, deletion mutants were constructed and the expression levels of *gapA* were compared by qRT-PCR. Total RNAs from the mid-exponential phase and the beginning of the stationary phase aliquots, which grows on glucose or acetate respectively, were extracted. At the beginning of the stationary phase, the activity of GAPDH in the *ramA* mutant was reduced in the presence of glucose, while in the presence of acetate, it was comparable to that of the wild strain, suggesting that, at the onset of the stationary phase, RamA upregulated expression of *gapA* in the presence of glucose [Toyoda et al., 2009]. EMSAs and DNase I footprinting assays showed that RamA specifically bound to *gapA* promoter at three sites with different affinity. Furthermore, RamA can directly activate *sugR* expression. This complex regulatory mechanism might benefit the *C. glutamicum* to reconcile the *gapA* expression level with environmental cues [Toyoda et al., 2009].

VanT of Vibrio anguillarum. *Vibrio anguillarum* is a gram-negative pathogen which can lead to a terminal hemorrhagic septicemia [Milton et al., 1997] and contains multiple quorum-sensing systems. VanT is a homolog of LuxR protein; its amino acid sequence has 81% identity with LuxR of *V. harveyi*, 74% identity with HapR of *V. cholerae*, 82% identity with SmcR of *Vibrio vulnificus*, and 82% identity with OpaR of *V. parahaemolyticus*. An in-frame *vanT* deletion was constructed to investigate its function. Gene fusion found that VanT transcriptionally regulated *empA*, *vps73*, *serA*, and *hpdA*. These four genes were transcriptionally fused with β-galactosidase encoding gene and the protease activity measured was almost the same in mutant and the wild type. Northern analysis showed that the amount of RNA from the *vanT* mutant was significantly decreased in comparison with the wild type, suggesting that VanT regulated the expression of the metalloprotease *empA* gene. TLC analysis and the LD50s showed that VanT is dispensable

TABLE II. The Regulation of Bacterial LuxR

Organism	LuxR homolog	Function	The amino acid identity with *Mycobacterium tuberculosis H37Rv*	References
Vibrio alginolyticus	ValR	ValR may control the formation of biofilm by regulating the biosynthesis of flagellum.	22% TetR family transcriptional regulator	Chang et al. [2010]
Corynebacterium glutamicum	RamA	RamA positively regulates the expression of D-Glyceraldehyde-3-phosphate dehydrogenase and sugR.	39% nitrate/nitrite response transcriptional regulatory protein NarL	Toyoda et al. [2009]
Vibrio anguillarum	VanT	VanT upregulates biofilm formation, pigment production, the biosynthesis of serine, glycine, metalloprotease gene empA expression.	23% TetR family transcriptional regulator	Croxatto et al. [2002]
Serratia marcescens	SpnR	SpnR downregulates the production of pigment, exonucleases, and biosurfactant; it has positive autoregulation	36% nitrate/nitrite response transcriptional regulatory protein NarL	Horng et al. [2002]
Aeromonas hydrophila	AhyR	AhyR regulates the formation of biofilm, the production of ectoenzyme and AhyI.	32% two component transcriptional regulatory protein DevR	Kirke et al. [2004]
Vibrio cholerae	HapR	HapR represses biofilm formation and aphR expression, in addition, it has negative autoregulation	44% TetR family transcriptional regulator	Hammer and Bassler, 2003; Lin et al. [2005]
Streptomyces hygroscopicus 17997	GdmR I and GdmR II	GdmR I and GdmR II positively regulate geldanamycin biosynthesis.	34% transcriptional regulatory protein and 38% transcriptional regulatory protein	He et al. [2008]

for 3-oxo-C10-HSL production and virulence. Biofilm formation, pigment production, biosynthesis of serine, and glycine were upregulated by VanT [Croxatto et al., 2002].

SpnR of Serratia marcescens. *Serratia marcescens* is a Gram-negative, rod-shaped bacterium. They can be used to detect the quality of bacterial filters because of their small size and the ability to produce bright red pigment called prodigiosin [Dauenhauer et al., 1984]. *S. marcescens* can cause nosocomial infections, urinary tract infections, and wound infections [Dauenhauer et al., 1984].

S. marcescens SpnR is a member of LuxR protein family. Raised levels of SpnR can reduce the production of the red pigment and the colonies became white. Deletion mutants produced more pigment, suggesting that SpnR might be a repressor of prodigiosin production [Horng et al., 2002].

Microplate format DNA degradation assay showed that the exonuclease activity was reduced by about 50% with multicopy *spnR*, whereas that of the deletion mutants increased fourfolds. This hints that SpnR was a repressor of exonuclease, and this was achieved via modulating the transcription of *nucA* [Horng et al., 2002]. In addition, SpnR regulates the sliding motility by repressing the production of biosurfactant. In *S. marcescens* SS-1ΔR, the onset time for sliding prolonged from 1.5 h to about 5 h [Horng et al., 2002]. SpnR had no effect on AHL production. The structural analysis revealed that the upstream region of *spnR* had a *lux box* consensus sequence partially overlapped with the predicted −35 promoter region, suggesting that *spnR* might be autoregulated. The promoter activity of *spnR* was monitored by a recombinant plasmid-constructed pSC503. The light-emission patterns of *S. marcescens* SS-1(pSC503) and *S. marcescens* SS-1ΔR (pSC503) showed that PspnR activity increased 2.4-fold in the presence of *spnR*, suggesting that SpnR had positive autoregulation effect which is enhanced by 3-oxo-C6-HSL. Overexpression of SpnR can repress many phenotype, while deletion of *spnR* can enhance these traits, suggesting that SpnR is a negative regulator [Horng et al., 2002].

AhyR of Aeromonas hydrophila. *Aeromonas* species are Gram-negative bacterium. Although *Aeromonads* were discovered more than 100 years ago, their roles in multiple human diseases were only studied in recent 30 years [Janda and Abbott, 1998]. They are putative culprits of gastroenteritis ranging from mild enteritis to diarrhea. However, their roles in enteric diseases remain elusive due to the lack of human volunteers and an appropriate animal model, as well as no accurate diagnostic procedures [Thornley et al., 1997]. *Aeromonas* species is also implicated in infections such as gastroenteritis, hemolytic uremic syndrome(HUS), septicemia, ocular infections, meningitis, peritonitis, wounds, respiratory tract disease, and new bacteremic syndromes [Janda and Abbott, 1998]. Furthermore, *Aeromonads* are pathogens for fish, amphibians, and reptiles [Austin, 1997].

The virulence of *Aeromonas* species is the synergy of multiple factors including extracellular enterotoxins, capsule formation, adhesions, endotoxins, fimbriae, haemolysins, S-layer, proteases, and cytotoxins [Cahill, 1990]. In *A. hydrophila*, biofilm formation and extracellular protease production were controlled by AhyR/C4-HSL-dependent QS system; the activities of metalloprotease and serine in mutation of either *ahyI* or *ahyR* were lost [Swift et al., 1999]. In mutation of *ahyR*, the biofilm had a larger available surface area than the wild-type [Lynch et al., 2002]. Western blotting showed that transcription of *ahyI* was not required for AhyR, but AhyR controls the turnover of AhyI in stationary phase and the timing of AhyI expression [Kirke et al., 2004].

HapR of Vibrio cholerae. Cholera is a common lethal diarrhea disease which can be caused by food or water contaminated with the *Vibrio cholerae*. *V. cholerae* has two primary virulence factors, one is toxin coregulated pilus (TCP) which is a critical colonization factor [Taylor et al., 1987], the other is cholera toxin (CT), which causes copious diarrhea that can lead to severe dehydration and death [Lin et al., 2005]. The expression of these genes which encode the virulence factors depend upon multiple activator and repressor proteins; these proteins were regulated by regulators AphA and AphB. AphA is a repressor that regulates the expression of genes participated in the detoxification of phenolic acids; the deletion of *aphA* prevented the production of TCP [Skorupski and Taylor, 1999]. In *Vibrio cholerae*, the phosphorylation of LuxO (LuxO∼P) activates the transcription of five genes, which encode homologous small noncoding RNAs (sRNAs) [Lenz et al., 2004] called the Qrr sRNAs [Rutherford et al., 2011]. LuxO, Qrr sRNAs, LuxR, and AphA constitute a complex regulatory network. First, LuxR activates the expression of *qrr*, which subsequently increases the production of Qrr sRNAs [Svenningsen et al., 2008]. The Qrr sRNAs repress the translation of *luxR* mRNA, which reduces LuxR production. In turn, the production of Qrr sRNAs is reduced [Rutherford et al., 2011]. Second, the Qrr sRNAs suppress *luxO* translation, which reduces the levels of Qrr due to the expression of *qrr* requires LuxO∼P [Svenningsen et al., 2008; Tu et al., 2010]. Finally, the Qrr sRNAs inhibit the expression of genes which encode the AI synthases and cognate receptors. In addition to *qrr*, AphA also inhibits *luxR* [Rutherford et al., 2011]. In *V. cholerae*, a LuxR homolog, HapR binds to a specific site of *aphA* promoter at high cell density, which reduces the expression of virulence gene. Hence, AphA and HapR repress mutually [Rutherford et al., 2011]. HapR can inhibit the biofilm formation. In Δ*hapR-lacZ* transcriptional fusion strains, it was confirmed that the ability of HapR reduces its own expression at high cell density, suggesting that HapR negatively autoregulated [Lin et al., 2005].

GdmR I GdmR II of Streptomyces hygroscopicus 17997. *Streptomyces* are gram-positive, high GC-content bacterium and can produce various secondary metabolisms including antibiotics. *Streptomyces* produces secondary metabolisms under starvation, concomitant with the slow growth rate of the colony in the transition phase and the related morphological changes. *Streptomyces hygroscopicus* 17997 is a producer of geldanamycin [Tao et al., 1997], which is active against cancer and virus.

Streptomyces have a complicated secondary metabolisms regulatory network in which regulators interacts with each other to regulate the production of antibiotic and other secondary metabolites. Several large ATP-binding regulators of the LuxR family (LAL) originally found in *Escherichia coli* [Schrijver and Mot, 1999] have also been found in *Streptomyces* antibiotic gene clusters. Geldanamycin can bind to the ATP/ADP domain of heat shock protein 90, which can downregulate target proteins, such as steroid receptors, tyrosine kinases, and transcriptional factors [Prodromou et al., 1997]. The downstream of the geldanamycin biosynthetic

gene cluster has two regulatory genes, *gdmR I* and *gdmR II*, which encode two LuxR -family transcriptional regulators. The result of HPLC assays showed that the deletion mutant strains Δ*gdmR I* and Δ*gdmR II* could not produce geldanamycin. When the intact *gdmR I* and *gdmR II* were complemented into each mutant, the production of geldanamycin was restored to 30% of wild type, indicating that GdmR I and GdmR II could positively regulate geldanamycin biosynthesis. But the growth rate and morphological characteristics are same to wild strain, suggesting that the GdmR I and GdmR II are not crucial for bacterial growth and differentiation [He et al., 2008].

THE LuxR TRANSCRIPTIONAL REGULATORS OF MYCOBACTERIA

Tuberculosis has been a leading global killer because of the 8.9 million new cases and 1.7 million deaths annually. The regime largely consisting of rifampicin, isoniazide, and streptomycin once was potent to stem the TB. However, this transient success was shattered by the emergence of multiple drug resistant *Mycobacterium tuberculosis* and HIV coinfection. One drawback is the lack of the understanding of *M. tuberculosis* physiology. More studies on *M. tuberculosis* LuxR regulators are promising to bridge this gap.

Seven genes encoding LuxR family proteins, Rv0386, Rv0195, Rv0491, Rv0890c, Rv0894, Rv2488c, and Rv3133c, were found by our scrutiny of the *M. tuberculosis* H37Rv genome blastP 16 *Mycobacterium* genomes (http://www.ncbi.nlm.nih.gov/sutils/genom_table.cgi). With their amino acid sequences found Rv3133c and Rv0491 are ubiquitous among 16 *Mycobacteria*. Rv4091 identity among 16 *Mycobacteria* is above 90%, suggesting the two genes may play a key role in *Mycobacteria*. The identities of seven genes are above 99% between *Mycobacterium bovis* and *M. tuberculosis* (Fig. 3).

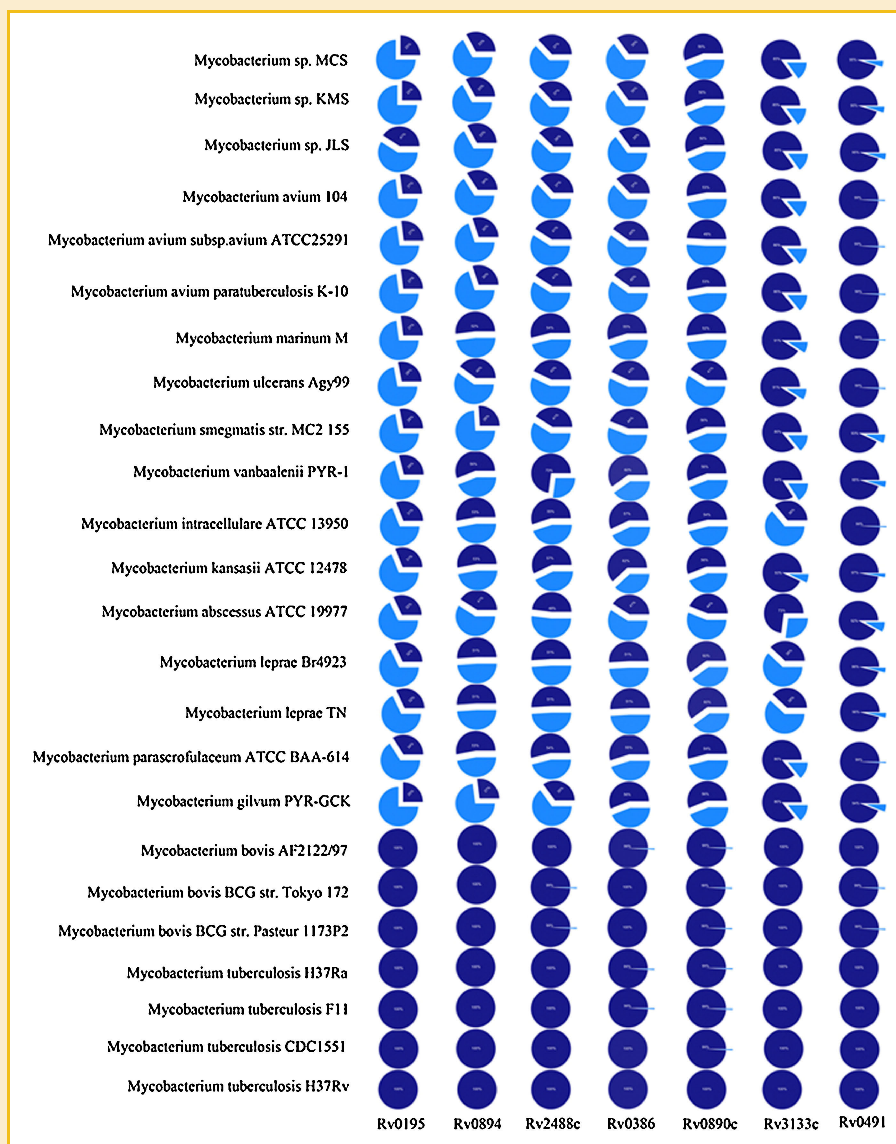

Fig. 3. The identities of the LuxR transcription regulators of Mycobacterium tuberculosis and other Mycobacteria. Blue represents identity, abscissa represents seven regulators of *M. tuberculosis*, ordinate represents various Mycobacteria.

J774 macrophage-like cells and THP-1 macrophages infected by *M. tuberculosis* undergoes intracellular cAMP burst and initiate the phosphorylation of cAMP response-element-binding protein (CREB) in a dose-dependent manner [Agarwal et al., 2009]. However, this phosphorylation is absent in macrophages infected with heat-killed bacteria. Specific pharmacological inhibitors of these signal transduction pathways leading to CREB phosphorylation were used to treat the *M.tuberculosis*-infected macrophages. Immunoblots showed that there might be another alternative source of cAMP [Agarwal et al., 2009]. On the basis of the observation that *M. avium*-induced CREB phosphorylation leads to tumor necrosis factor-α (TNF-α) secretion [Roach et al., 2005], a recombinant *M. tuberculosis* strain overexpressing endogenous phosphodiesterase (PDE) Rv0805 (Mtb-PDE) was constructed and infected macrophages. RT–PCR indicated that overexpression of PDE reduced intramacrophage cAMP levels and PKA-dependent events affected by cAMP [Agarwal et al., 2009]. Transposon mutated *M. tuberculosis* in eight adenylate cyclase genes were used to infect mice. Relative abundance of each mutant was tested via quantitative PCR. Impaired survival was observed in the Rv0386 mutant strain because the loss of Rv0386 decreased the entrance of bacterial cAMP into macrophage cytoplasm. However, the exact role of LuxR family proteins in *M. tuberculosis* remains to be elucidated.

In *Mycobacterium avium* subsp. *Paratuberculosis* (MAP), MAP0482 gene is a homolog of M. avium LuxR and has 34% identity with a conserved hypothetical protein of *M. tuberculosis*. MAP0482- overexpressing strain has more effective infectivity than the wild-type bacteria [Alonso-Hearn et al., 2010]. Transcriptional profiles show that many genes involved in lipid biosynthesis and transport are repressed in mutant, suggesting that LuxR regulate cell envelope and virulence via effecting lipid biosynthesis [Alonso-Hearn et al., 2010].

CONCLUDING REMARKS

QS can be found among various pathogenic bacteria and plays a key role in survival, virulence. So far, we have found several pathways which can disturb bacterial QS system. These methods are roughly divided into three classes: (1) Synthesizing degradation enzyme to inactivate signal molecules, such as AHL-lactonase [Hentzer et al., 2002]; (2) Using QS signal molecules to induce resistance; (3) Synthesizing signal molecules analogs competitively bind LuxR regulators, which block bacterial QS system.

In Australia, *Delisea pulchra* produces a series of halogenated furanones which can competitively combine LuxR proteins with signal molecules and consequently prevent bacteria colonizing its surface [Hoang et al., 2004]. Diketopiperazines (DKPs) were found in the supernant of bacterial culture solution, including *Pseudomonas aeruginosa*, *Citrobacter freundii*, *Enterobacter agglomerans*. This compound is a kind of AI as well; at high cell density, it has fairly pharmacodynamics and biological roles for higher organisms [Shiner et al., 2005]. Athough the mechanism of action remain obscure, DKPs are putative QS inhibitors capable of specifically binding to LuxR proteins and regulating LuxR-based QS system, which influence the bacterial survival and propagation.

With their promiscuous and significant role in diverse (patho)-physiological events, LuxR family transcriptional factors hold huge potential as new drug target to develop agents against tuberculosis and other pathogen.

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