Regulation of Virulence in *Streptococcus suis*

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**Abstract**

*Streptococcus suis* (*S. suis*) comprising thirty-five different serotypes, constitute a group of complex bacterial species that not only is swine pathogen, but also cause opportunistic infections in humans. A collection of virulence determinants have been largely elucidated that contribute to better understanding of pathogenesis underlying severe infections by *S. suis*. Here, we concentrated on control of *S. suis* virulence by a rainbow coalition of regulators, and discussed future perspectives in this field. It might provide a glimpse of the complex network of virulence regulation in *S. suis*.

**Editorial**

*Streptococcus suis* (*S. suis*) is a major swine pathogen that annually results in great economic loss worldwide [1,2]. Thirty-five serotypes (1-34, 1/2) have been classified, according to the differentiated capsule antigens of these heterogeneous *S. suis* species [3,4]. Of them, *S. suis* serotype 2 (SS2) is a previously-neglected but recently-emerging zoonotic agent that can lead to opportunistic infections in humans with close contact with swine and/or pork products [5-7]. Totally, more than 850 cases of human SS2 infections have been recorded, which are involved in over 30 countries and regions, esp. Southeastern Asia like Vietnam, Thailand, China, etc [1,3]. Given that strong invasiveness and high virulence is manifested by this bug, world-wide extensive studies have been conducted (esp. after a big outbreak of human SS2 endemic in China, in 2005 [7]), which led to discovery of a collection of new bacterial virulence determinants underlying SS2 pathogenicity [1,8]. In terms of recent development in this field, we presented a brief view on the regulation network of SS2 virulence from bellowed three aspects: transcription factor, two-component signal transduction system (TCSTS), plus orphan response regulator.

First, no less than five transcription factors, some of which can sense environmental signals, have been implicated into the complex regulatory network of *S. suis* pathogenicity (Table 1). AdcR is a regulator controlling zinc transport in *S. suis*, was observed to be correlated with bacterial virulence in mouse model [9]. In contrast, we failed to note that Zur, the other zinc uptake regulator from 05ZYH33 strain of *S. suis* [9,10], was observed to be correlated in SS2 virulence (Table 1). AdcR is a regulator controlling zinc transport in *S. suis* [9]. Willenborg et al. [11] had addressed the effect of the sugar metabolism regulator carbohydrate control protein A (CcpA) on *S. suis* pathogenesis. As anticipated, expression level of several virulence factors (such as ArcB, Sao, Enolase, etc.) were altered in the AdcR mutant. Moreover, the deletion of ccpA led to significant reduction of both capsule thickness and resistance to killing with porcine neutrophils. Unfortunately, its pathological role in bacterial virulence has not yet been verified with experiments of animal infections (Table 1). ArgR, a member of ArgR/AhrC arginine repressor family, was recently proved to regulate expression of *arcABC* operon encoding an arginine deiminase system that is recognized as a putative virulence factor [12,13]. Therefore, it is of much interest to test a role of argR in *S. suis* virulence (Table 1). Similar to what has been observed with Rgg regulators of other Gram-positive pathogen, we defined an rgg-like ortholog of *S. suis* 05ZYH33. Multiple roles of this regulator in bacterial metabolism were observed. Particularly, it was confirmed as a virulence determinant in the experimental model of piglets [14]. Very recently, Zhang and coworkers supplemented a Fur-like family of transcription factor, PerR, to the increasing list of virulence factors of *S. suis* [15]. This regulator is controlled by both H2O2 and metal ions, and directly modulates expression of two target genes (one is *dpr* encoding Dps-like peroxide resistance protein and the other is metQIN encoding a methionine transporter) [15].

Among the 15 putative TCSTS of the Chinese virulent SS2 strain (e.g., 05ZYH33) [16,17], four have been found to be involved in control of *S. suis* virulence (Table 1). In 2008, we reported a unique *salK-salR* TCSTS system within the 89K pathogenicity island [18]. The deletion of this two component system resulted in significant down-regulation of 26 genes’ expression level, and increased its susceptibility to polymorphonuclear leukocyte (PMN)-mediated killing. Consequently, the virulence of the *SalK-R* mutant was seriously attenuated [18]. Subsequently, *ciaR-ciaH* was determined as the second TCS system that is essential for pathogenicity of SS2 in the infection models of CD1 mice and piglets both [19]. A homolog of the *Clostridium perfringens* VirR-VirS regulatory system was also observed in 05ZYH33 strain of SS2, and the isogenic knockout mutant (*ΔvirRBS*) was found to exhibit marked attenuation of virulence observed with the infection model of mice [20]. More recently, Han et al. [21] employed bacterial genetics combined with comparative proteomics to unveil that the *ihk-irr* TCSTS is necessary for SS2 pathogenicity via modulating bacterial central metabolism.

Additionally, only two orphan response regulators have been verified to be involved in SS2 pathogenesis thus far (one is RevSC21 [22], and the other is CovR [23], Table 1). In 2009, Wu et al. [22] reported that RevSC21 regulator positively regulates expression levels of virulence factors (such as *mrp, sly, cps*, etc.), and is required for bacterial
virulence. In contrast, we had ever observed another orphan response regulator CovR with an opposite effect on S. suis pathogenicity [23]. The covR-defective (ΔcovR) mutant displayed thicker capsules and increased hemolytic activity. Furthermore, adherence of this mutant to epithelial cells was greatly increased, as well as its resistance to phagocytosis and killing by neutrophils and monocytes. Eventually, the removal of covR gene was found to be correlated with increased lethality of piglets [23].

It still remains elusive whether some connection/linking are present among the complex regulatory networks constituted by above transcription factors and TSCTs in modulating bacterial virulence. It is reasonable that presence of other transcription factors and/or regulatory systems that are associated with control of bacterial virulence in SS2. Unfortunately, nothing is known on post-transcriptional control of virulence by small non-coding RNA (sRNA) in S. suis, although it has been addressed in its closely-related organism, S. pneumoniae [24]. Thus we believed that genome-wide systematic identification and functional assignment of small non-coding RNAs might contribute to better understanding control of SS2 virulence. In similar, it is also of great interest to elucidate the potential relevance and/or linking of machineries for post-translational modifications [25] (such as acetylation [26]) to S. suis pathogenesis.

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Table 1: Transcription factors and regulatory systems required for S. suis virulence.

| Genes | Functional annotation | SS2 Strains | Animal models | Origins |
|-------|-----------------------|-------------|---------------|---------|
| perR  | PerR, a Fur-like protein | Strain SC-19 (China) | Balb/c mice | (25) |
| addR  | AdcR, a pleiotropic regulator | Strain P1/7 (Netherlands) | Balb/c mice | (1) |
| ccpp | Catabolite control protein A | Strain 10 (Netherlands) | No | (23) |
| argR | An ADS-associated repressor of the ArgR/AlrC arginine family | Strain 10 (Netherlands) | No | (9) |
| rgg  | Rgg transcription factor | 05ZMH3 (China) | Piglets | (26) |

Two component signal transduction system (4)

| InhR-inhR | A homolog of the Streptococcus pyogenes InhR/Inr TCS | Strain 05ZMH3 (China) | CD-1 mice | (13) |
| virR-virS | A homolog of the VirR-VirS regulatory system of Clostridium perfringens | Strain 05ZMH3 (China) | Balb/c mice | (20) |
| saik-salR | Two-component system in the 89K PAI | Strain 05ZMH3 (China) | Piglets | (14) |
| ciaR-ciaH | A two-component system | SC19 (China) | CD-1 mice & piglets | (5) |

Orphan response regulator (2)

| coVR | Orphan response regulator (CovR) | Strain 05ZMH3 (China) | Piglets | (16) |
| revSC21 | Orphan response regulator RevSC21 | Strain SC21 (China) | CD-1 mice | (24) |
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