Revisiting the Staphylococcus aureus SarA regulon by high-throughput screening

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Staphylococcus aureus is a commensal bacterium and an opportunist pathogen causing a variety of human and animal infections from benign to harsh. It is also the major cause of community-associated and nosocomial infections. Its pathogenicity involves virulence and metabolic factors whose expression can be regulated by regulatory RNAs (sRNAs) or transcriptional factors (TF). Among *S. aureus* TF there is SarA, a major virulence factor involved in biofilms formation and antibiotic resistance. Our laboratory has recently shown that SarA represses the expression of two RNAs, *srn_3610_sprC* and *srn_9340*. My project is to expand the SarA regulon especially to regulatory RNAs. Using RNA-Seq, we compared the transcriptome of the isogenic Δ*sarA* mutant to the wild-type strain and we found that, in absence of SarA, the transcription of 299 genes was repressed while the transcription of 142 genes was actived. To discriminate the direct and indirect targets, ChIP-Seq experiment was realized to localize the SarA binding sites in the whole genome. Around 340 SarA binding sites were identified with a p-value under 1.10^{-250}. Combining RNA-Seq and ChIP-Seq results, we highlighted one hundred genes for which the level of transcription appears to be directly correlated to the SarA binding on their promoter region. We chose to validate 14 potential targets by Northern Blot and gel shift experiments. Among the sRNA targets validated in vitro, we can cite the $SprA2_{AS}$ antitoxin sRNA which is part of the type I toxin-antitoxin (TA) system SprA2/$SprA2_{AS}$ and the SprG2 sRNA encoding the toxin of the SprG2/SprF2 type I TA system. In the type I TA system, a drop of sRNA antitoxin level induces toxic peptide translation that results to bacterial cell death or bacteriostasis. Our results reveal a new aspect of SarA which appears, for the first time, as a novel regulator of type I TA systems strengthening its involvement in *S. aureus* virulence.

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