

Insights into *Klebsiella pneumoniae* type VI secretion system regulation

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Klebsiella pneumoniae is a Gram-negative bacterium responsible for many acute infections, mainly in the urinary and respiratory tracts. These infections represent a big challenge for public health as there are multiple antibiotic resistant strains circulating around the world, including in Brazil. Thus, the understanding of *K. pneumoniae* virulence mechanisms is still required and may, in the future, lead to novel approaches for interfering at the infection process, such as the development of more efficient and specific drugs. Recently, it was suggested that the type VI secretion system (T6SS) of *K. pneumoniae* is important to its pathogenicity. Indeed, T6SS provides competitive and adaptative advantages for bacteria that possess it. As T6SS is present in a wide variety of proteobacteria, it is not surprising that the T6SS shows different regulatory mechanisms, such as quorum sensing, biofilm formation, iron limitation, oxidative stress, changes in osmolarity and temperature as well as the independent regulation for the expression structural components and of the T6SS effectors. Up to now at least 24 transcriptional regulators of T6SS are known in species of *Vibrio*, *Pseudomonas*, *Burkholderia* and *Escherichia* genus. However, there are no studies concerning T6SS regulation in *K. pneumoniae*. Our group has previously observed that *K. pneumoniae* strain Kp52.145 expresses T6SS genes *in vivo*, however the signal triggering such mechanism has not been identified. In this work we aimed to get insights into such regulation. In order to achieve this aim, we used a computational approach to identify possible transcriptional regulators of T6SS in *K. pneumoniae*. In order to infer promoter regions, we predicted potential transcription start sites in the three T6SS loci encoded in the genome of this strain, using BPPROM algorithm. Then, we further analyzed the 13 promoter regions predicted in order to identify putative transcriptional regulators binding sites. For that, the -500 bp sequences were analyzed with Virtual Footprint software against ProDom database consisting of 59 protein binding site patterns. In overall, 523 putative binding sites of 27 different regulators were predicted. After manual curation, eight regulators and their binding sites are highlighted. They are: Fnr | *Escherichia coli*, Fur(8mer) | *Escherichia coli*, Fur | *Pseudomonas aeruginosa*, OmpR | *Escherichia coli*, OxyR | *Escherichia coli*, OxyR (SELEX) | *Escherichia coli*, RcsAB | *Escherichia coli*. Most of those regulators are part of two-component systems regulated by known environmental stimuli. We are currently performing validation experiments in order to verify computational predictions.