Poster

The structure of *Vibrio cholerae* transcription complexes reveal how one-component signal transduction factors ToxR and TcpP activate virulence genes

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Cholera is an acute life-threatening diarrheal infection caused by the bacterium *Vibrio cholerae*. Having brought about seven world pandemics, cholera is endemic in several countries. There has been an acute upsurge of the 7th cholera pandemic since 2021. In this regard, cholera has re-emerged in at least 30 countries, in areas that have been free of cholera for decades, with high mortality rates, prompting the WHO to classify it as a highest grade 3 emergency.

When *V. cholerae* reaches the small intestine, two integral membrane proteins, ToxR and TcpP, activate the *toxT* promoter. This triggers a regulatory cascade resulting in the secretion of cholera toxin and the expression of the toxin-coregulated pilus. ToxR alone also downregulates the expression of membrane porin OmpT and upregulates that of OmpU, leading to protection against bile salts in the human intestinal epithelium.

ToxR is a one-component signal transduction regulator with a periplasmic receiver domain and a DNA-binding effector domain. These domains are connected by a single-pass transmembrane region. Although one-component systems are common in prokaryotes, they are poorly characterized in comparison with phosphorylation-based two-component systems.

Here we have studied the activation mechanism of *toxT* and *ompU* virulence genes by ToxR and TcpP. Using X-ray crystallography, we have solved a number of structures of ToxR complexed with different DNA sequences of the *toxT* and *ompU* promoters [1]. In addition, 5 structures of transcription activation complexes, including the RNA polymerase holoenzyme, DNA promoter sequences and ToxR and TcpP transcription factors, have been solved by cryo-EM (Fig.1).

Based on the structures, we propose a model for RNA polymerase recruitment to the *toxT* and *ompU* promoters and the transcription activation of genes conferring virulence to *V. cholerae*.

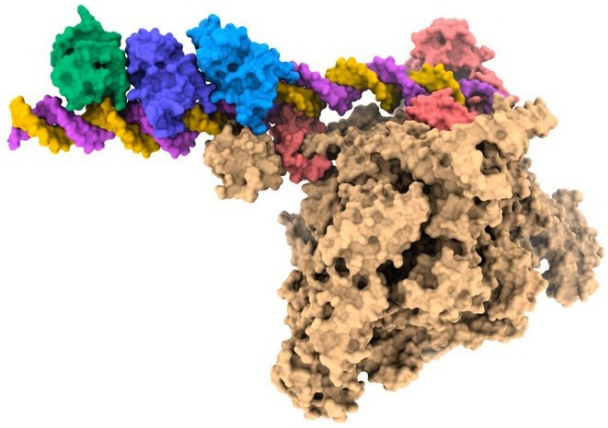

Figure 1. Cryo-EM structure of *Vibrio cholerae* transcription activation complex.

[1] Canals, A., Pieretti, S., Muriel-Masanes, M., El Yaman, N., Plecha, S.C., Joshua J. Thomson, J.J., Fàbrega-Ferrer, M., Pérez- Luque, R., Krukonis, E.S. & Coll, M. (2023). ToxR activates the *Vibrio cholerae* virulence genes by tethering DNA to the membrane through versatile binding to multiple sites. *Proc. Natl. Acad. Sci. USA* **120**, e2304378120. https://doi.org/10.1073/pnas.2304378120.