Sociedad Argentina de Microbiología General

XIX CONGRESO DE LA SOCIEDAD ARGENTINA DE MICROBIOLOGÍA GENERAL

22 al 25 de octubre del 2024
Centro cultural y Pabellón Argentina de la Universidad Nacional de Córdoba, Córdoba, ARGENTINA.

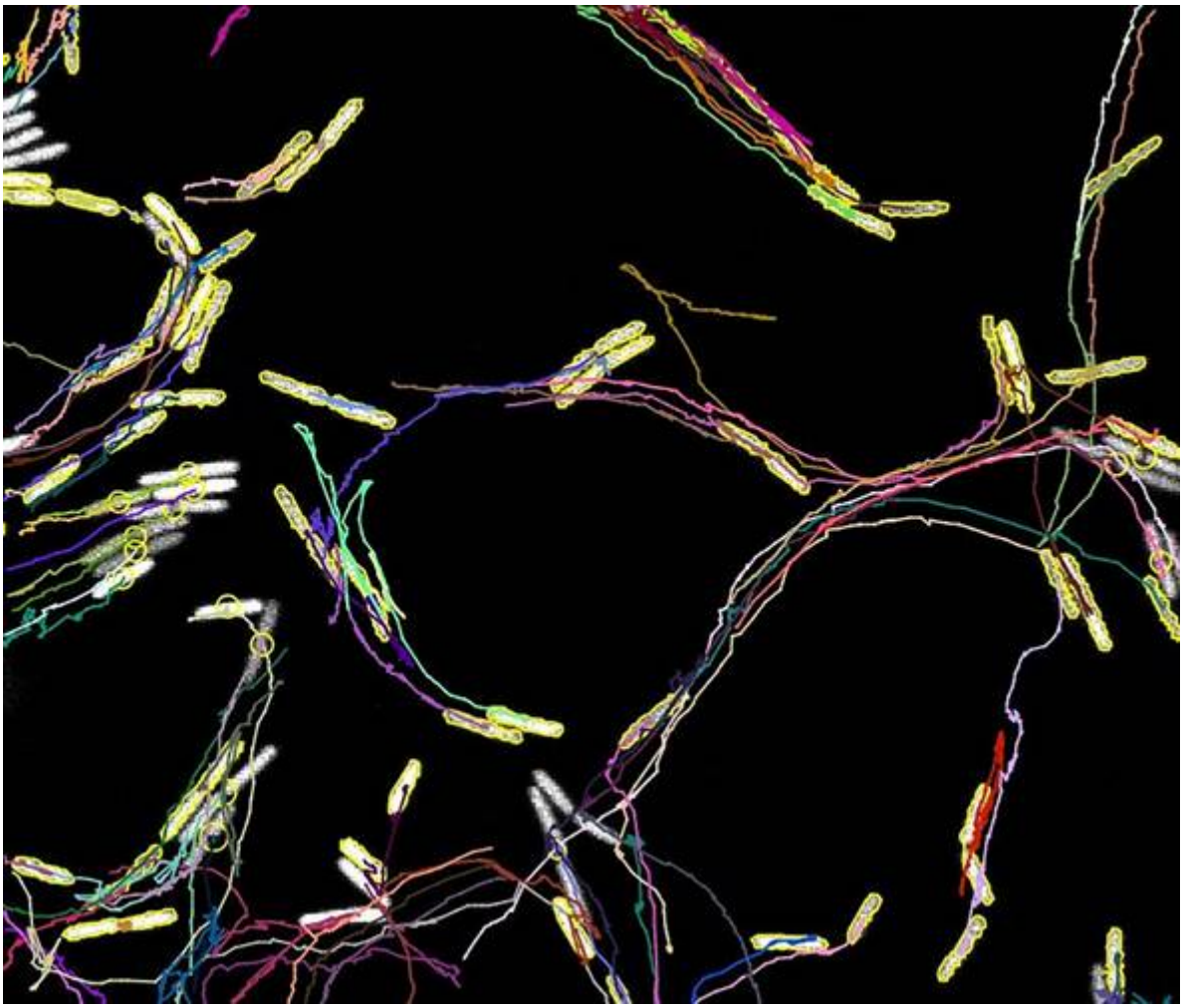

Foto: Se hace camino al andar. Celeste Dea. 1er puesto. Concurso fotográfico SAMIGE 20 años.

Evolution, Structure and Dynamics in the ArsR Family of Transcriptional Regulators

Antelo, Giuliano Tomás[1] - Pis Diez, Cristian Manuel[1] - Rondon, Johnma J[1] - Guitierrez González, Giovanni[2] - Wu, Hongwei[2] - Giedroc, David Peter[2] - Capdevila, Daiana Andrea[1]

1) Fundación Instituto Leloir, IIBBA-CONICET - C.A.B.A - Buenos Aires- Argentina.
2) Chemistry, Indiana University - Bloomington- IN - United States of America.
Contacto: antelo.gt@gmail.com

To sustain life, organisms must detect and respond to environmental stress while maintaining homeostasis. In bacterial organisms, where the ability to physically escape harmful conditions is limited, changes in gene expression are the key survival mechanism. This is primarily achieved by transcriptional regulators that sense stressful molecules and modulate the expression of response genes to alleviate stress. One prominent example is the ArsR family of transcriptional repressors. These proteins are widely distributed in nature, including in many pathogens, where they play critical roles as virulence factors, enabling pathogens to counter host immune responses or even resist antibiotics. The ArsR family is characterized by a highly conserved three-dimensional fold despite low sequence identity among its members. Each ArsR protein is capable of specifically detecting a particular stressor molecule, which can range from metal ions to reactive oxygen or sulfur species. Collectively, this family of transcriptional repressors is one of the most diverse in terms of the range of stressors sensed. Understanding the evolution of such diversity, as well as the molecular determinants of stressor specificity and its impact on gene expression, is essential to uncover how new resistance mechanisms emerge. Our previous work used Sequence Similarity Network (SSN) analysis to create iso-functional subgroups and characterize sensors of unknown inducers, like Vibrio cholerae's HlyU. Additionally, structural and dynamical studies on a zinc sensor from Staphylococcus aureus showed that changes in internal dynamics upon inducer or DNA binding are crucial for explaining allosteric communication between the sites. However, the generality of this mechanism across the family remains unclear. In this work, we extended our SSN analysis by obtaining consensus sequences for each subgroup. Assuming these reduce phylogenetic noise from low sequence identity, we used artificial intelligence tools (AlphaFold) to identify the key positions responsible for inducer specificity, DNA-operator specificity, or allosteric communication. To test these observations, we focused on SqrR, a persulfide sensor from Rhodobacter capsulatus, with known structures of its reduced (DNA-compatible) and oxidized (DNA-incompatible) forms. We solved the DNA-bound structure, providing detailed insights into each protein state and enabling functional assignments for the conserved positions from our

bioinformatic analysis. Finally, we addressed the role of internal dynamics in allosteric communication by obtaining NMR spectra of oxidized, reduced, and DNA-bound SqrR. The high-quality spectra allowed us to quantify the changes in conformational entropy between states through estimation of the average methyl order parameter. Using these advanced NMR approaches and bioinformatic analysis, we obtained an integrated view of how function may be diversified in a family of proteins.

Palabras clave: Transcriptional regulators – allostery – molecular evolution- ArsR – NMR