Cross-talks between two paralogous cascades contribute to the regulation of the General Stress Response in *Sphingopyxis granuli* TFA.

Rubén de Dios, Eduardo Santero*, Francisca Reyes-Ramírez  
Departamento de Biología Molecular e Ingeniería Bioquímica, Centro Andaluz de Biología del Desarrollo (CABD/CSIC), Universidad Pablo de Olavide, 41013 Seville, Spain  
*Corresponding author, e-mail: esansan@upo.es

**Thematic Areas:** Microbial Biotechnology.

**Abstract:** In natural habitats, bacteria are forced to overcome many hostile conditions. In order to survive, they constantly adapt their expression profiles accordingly to the variations in the environment. Among other responses, bacteria can trigger the so-called General Stress Response (GSR), which responds non specifically to a wide variety of stresses, making them ready to thrive in conditions that otherwise would prevent their growth. In particular, α-proteobacteria have a unique signalling cascade to activate the GSR, involving three regulatory elements: an EcfG alternative σ factor, its cognate NepR anti-σ factor and a PhyR response regulator (1). Under permissive conditions, EcfG is bound to NepR, preventing the expression of the GSR regulon. Once some stress appears in the environment, Phyr receives a phosphoryl group and changes its conformation, showing a σ-like domain. This domain is able to sequester NepR, thus releasing EcfG to active the GSR. *Sphingopyxis granuli* TFA is an α-proteobacterium able to grow with the organic solvent tetralin as carbon and energy source, and the first representative of its genus described as facultative anaerobic (2). After the analysis of its genome, two paralogous genes were identified for each GSR regulatory element, having EcfG1, EcfG2, NepR1, NepR2, PhyR1 and PhyR2. Once single and double mutants (when possible) were constructed at the three levels, their capability to activate the GSR and resist to different stresses were tested, allowing us to distribute the six elements in two cascades: the main pathway PhyR1-NepR1-EcfG2 and the accessory pathway PhyR2-NepR2-EcfG1. After this, combinatory double mutants in both cascades were constructed and tested again regarding their capability to activate the GSR to resist different stresses. Compared to the literature and to the experiments with the single mutants, we expected that mutations in the main cascade prevented the activation of the GSR. Surprisingly, these mutants did not behave as if both cascades were independent, thus indicating that there might be cross-interactions from PhyR to NepR and from NepR to EcfG between the two pathways and they might contribute to the tight regulation of the GSR in TFA.

**Acknowledgements:** Work in the authors' laboratory was financed by FEDER/ Spanish Ministry of Science, Innovation and Universities- State Research Agency, grant BIO2014-57545-R. R. de D. holds a contract from the Spanish Ministry of Education (FPU Program).

**References**