The pivotal functionality of the amyloid protein TasA in *Bacillus* physiology and fitness

Cámara-Almirón, J¹, Navarro, Y¹, Magno-Perez-Bryan, M. C¹, Molina C¹, de Vicente, A², Pérez-Garcia, A², and Romero, D¹.

¹Departamento de Microbiología, Centro de Supercomputación y Bioinnovación, Universidad de Málaga, Calle Severo Ochoa 34, Parque Tecnológico de Andalucía, 29590 Málaga, España.

²Departamento de Microbiología, Facultad de Ciencias, Universidad de Málaga, Campus de Teatinos, 29071, Málaga, España.

Biofilms are complex bacterial communities formed on any virtual surface and composed of cells embedded in an extracellular matrix. Studies on *Bacillus subtilis* have demonstrated this tissue-like structure comprised of diverse exopolymeric substances (eps) including exopolysaccharides, the protein BslA, and TasA and TapA the two main components of the amyloid fibers that confer robustness to the architecture of the biofilm. It has been demonstrated that the genetic pathways involved in formation of biofilms are active in the interaction of *B. subtilis* with plant surfaces. Indeed, we previously showed that surfactin acts as a self-trigger of biofilm in the plant phylloplane, which connected with the suppressive activity of *B. subtilis* against phytopathogenic fungi. These findings led us to hypothesize a major contribution of the extracellular matrix in the ecology of *B. subtilis* in the poorly explore plant phylloplane. In this work, we show that the amyloid protein TasA has a meaningful role in adhesion and biofilm formation over the plant phylloplane, however, despite the inability of the *tasA* mutant to form a biofilm, it still retained a similar antagonistic activity compared to the wild type strain. An in-depth transcriptomic analysis of the mutant led us to find unexpected variations in the expression levels of over 300 genes, including the overexpression of: i) production of acetoin ii) secondary metabolites and non-ribosomal peptides iii) eps and other biofilm-related components and iv) general stress, among others. These findings suggested that besides the structural role, TasA might have a regulatory function on the physiological stage of the cells. Indeed, an allele of TasA unable to restore biofilm formation allowed us to separate both functions, supporting the importance of this functional amyloid in regulating bacterial physiology and fitness.

This project is funded by the European Union (European Research Council - Starting Grant BacBio ERC637971)