## USING GENETICS TO DEFINE THE INTERACTION SURFACES BETWEEN TWO Bacillus Subtilis CELL DIVISION PROTEINS

Blasios-Jr, V.<sup>1</sup>, Bisson-Filho, A.W.<sup>1</sup>, Varvakis, T. A.<sup>1</sup>, Gueiros-Filho, F.J.<sup>1</sup>

<sup>1</sup>Departamento de Bioquímica, IQ, USP

The initiation of bacterial division is the formation of a ring of the tubulin-like protein FtsZ in the middle portion of the cell. The Z ring is a cytoskeletal structure that works as a scaffold for the recruitment of several proteins involved in the formation of the division septum. A crucial aspect of bacterial division is its precision, which is assured by proteins that interact with FtsZ and regulate its polymerization. One of the best known regulators of FtsZ is the Min system, which inhibits Z ring formation in the polar regions of the cell and, thus, favors septum formation in the middle of the cell. Here, we have adopted a genetic approach to try to define how FtsZ and MinC interact at the atomic level. Based on the principle that overexpression of Min is lethal to cell, we sought FtsZ mutants capable of surviving Min overexpression, expecting that they would bear mutations in aminoacids important for the interaction with Min. To do so, an FtsZ mutant library was created through error prone PCR and transformed into a modified Bacillus subtilis strain that overexpresses Min. 70000 transformants were screened and 49 mutants that survived Min overexpression were found. We found five substitutions after the sequencing of 11 selected mutants. Mapping of these on the FtsZ structure revealed that bur of the substituted amino acids were clustered together in helix HC3. This suggests that the HC3 helix contains the possible binding pocket for Min. We are currently sequencing the 38 remaining mutants and will soon characterize the biochemical properties displayed by these Min resistant mutants.

Supported by CAPES, CNPq and FAPESP