"EXPRESSION AND CRYSTALLIZATION OF PTHA EFFECTOR PROTEIN FROM *XANTHOMONAS AXONOPODIS* PV *CITRI*"

Perez NT [1], Benedetti CE [1]

[1]Brazilian Synchrotron Light Laboratory, Campinas-SP, Brazil.

Studies have shown that A*vr* proteins delivered to the host plant cell by the type III secretion system (TTSS) play major roles during bacterial-plant interactions. PthA is the best known *avr* protein from *Xanthomonas axonopodis* pv *citri* (Xac), which causes the citrus canker disease. PthA is a multi-domain protein composed of an N-terminal, thought to be required for TTSS transfer, a central region consisting of many repeats of a 34 residue peptide, which confers host selectivity, and a C-terminal with DNA-binding characteristics. PthA isoforms differ by the number of repeats in the central domain and the way by which these repeats are folded may dictate how the protein dimerizes and is transported to the nucleus, where it may act as a transcriptional factor. As the three-dimensional structure of PthA is unknown we are attempting at crystallizing PthA for protein structure resolution. PthA2 isoform was cloned in pET vectors to express de entire protein or the C-terminal domain with a 6xHis tag at the N-terminal (6xHis-Ptha2 and 6xHis-CPtha2, respectively). In addition, a construct was made to express a truncated version carrying only 5,5 internal repeats and the C-terminal (6xHis-?PthA). All proteins were purified from the inclusion bodies. After refolding, they were analyzed by Circular Dichroism, Dynamic Light Scattering and subjected to crystallization. We are currently subcloning the constructs for expression of the proteins in yeast cells.