

Structure determination of PilZ from *Xanthomonas axonopodis* pv. *citri* and their role in Type IV biogenesis

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The genome of the citrus pathogen *Xanthomonas axonopodis* pv. *citri* (*Xac*) codes for four proteins with PilZ domains: XAC1133, XAC3402, XAC1971 and BcsA a cellulose synthase subunit. PilZ domains have been recently been shown to bind the important bacterial second messenger bis-(3',5')-cyclic diguanylic acid (c-diGMP) and control a number complex behaviors gram negative bacteria, including quorum sensing, biofilm formation, motility and the production of exopolysaccharides and virulence factors.

The PilZ family is highly divergent and bacterial genomes normally code for several proteins with a PilZ domain. Recently the structures of PilZ domains of PA4608 from *Pseudomonas aeruginosa* and VCA0042/PilZD from *Vibrio cholerae* structures were determined in which the c-diGMP binding site was localized. The original PilZ protein from *P. aeruginosa* (PA2960), which does not interact with c-diGMP, was shown to be involved in the production type IV pili, an extracellular surface structure required for twitching motility.

We describe the expression, purification, crystallization, and structural determination by multiwavelength anomalous dispersion of *Xac* PilZ (XAC1133), an ortholog of PA2960. The PilZ crystals diffracted up to 1.9Å resolution and belong to the P6₁ hexagonal space group in which two PilZ molecules are present per asymmetric unit.

Analysis of the PilZ structure has provided leads through which may be gained information regarding its failure to bind c-diGMP and important features about its function in type IV pilus biogenesis. Functional experiments results will be presented.

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