

THE SHORT ORF XAC3629 FROM *Xanthomonas axonopodis* pv. citri REGULATES THE OPERON *copAB* EXPRESSION

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Xanthomonas axonopodis pv. citri (*Xac*) is a phytopathogenic bacterium that causes citrus canker. After sequencing its genome, genes encoding proteins related to copper resistance were identified (*copA* and *copB*). Copper compounds have been used to control plant bacterial disease and the effectiveness of the process has been reduced by the appearance of copper resistant strains. The genes *copA* and *copB* are organized in an operon whose transcription is induced and specific to copper, as we previously described. The ORF XAC3629 encoding 152 amino acids hypothetical protein was identified upstream *copAB*. The main objective of this work is to investigate the role of the ORF on the operon expression. Previous results in our laboratory demonstrated that the inactivation of the ORF sequence by the transposon insertion led to a complete loss of the copper resistance. The mutant strains were unable to grow in culture medium containing copper, even at the lowest CuSO₄ concentration tested (0.25 mM), whereas the wild type strain was able to grow in the presence of up to 1.0 mM copper concentration. In this work we started to construct a mutant strain having the ORF XAC3629 deleted by homologous recombination. For that, a DNA fragment missing 309 bp was constructed by reverse PCR leading to plasmid pMOS-?XAC3629. The mutated ORF encodes a polypeptide sequence corresponding to the 28 N-terminal amino acids followed by the 21 C-terminal amino acids. The insert was subcloned into a suicide vector giving the construction pNPTS-?XAC3629 (*sacB* gene as selection marker) and used to transform *Xac* cells. Mutants were selected and are being evaluated in relation to copper resistance.

Keywords *Xanthomonas*, :copper resistance, operon *copAB*

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