

GENOMIC SEQUENCE OF THE BIOLOGICAL CONTROL AGENT *PSEUDOMONAS FLUORESCENS* PF-5

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For the first time, the complete genomic sequence is known for a biological control agent that suppresses plant disease. In July of 2005, the genomic sequence of *Pseudomonas fluorescens* Pf-5 was published (Paulsen et al. 2005). Strain Pf-5 was selected as a model biocontrol strain for a genomic sequencing project because of its success as a biocontrol agent, its use as a standard biocontrol strain by many researchers, and the body of knowledge that exists on the mechanisms by which Pf-5 suppresses soilborne plant disease. The genomic sequence provides a wealth of information about this biological control agent as well as future directions for enhancement of biological control.

Background

Pseudomonas fluorescens Pf-5 was first described for its capacity to suppress soilborne diseases of cotton caused by *Rhizoctonia solani* (Howell and Stipanovic 1979) and *Pythium ultimum* (Howell and Stipanovic 1980). Since it was first described, Pf-5 has been shown to suppress these pathogens on plant hosts including cucumber, pea, and maize. Pf-5 also suppresses a number of other soilborne or residue-borne pathogens such as *Pyrenophora tritici-repentis*, *Sclerotinia homoeocarpa* and *Dreschlera poae*, *Fusarium oxysporum* f. sp. *radicis-lycopersici* and *Erwinia carotovora*.

Pf-5 produces a suite of antibiotics including pyrrolnitrin (Howell and Stipanovic 1979), pyoluteorin (Howell and Stipanovic 1980; Nowak-Thompson et al. 1999), and 2,4-diacetylphloroglucinol (Nowak-Thompson et al. 1994). Each of these compounds has a different spectrum of activity against plant pathogens, and their roles in biological control have been established in various biological control organisms.

Highlights from the genomic sequence data

The size of the genome of Pf-5 is 7.1 Megabases, which is 0.5 to 1.0 Megabases larger than genomes of other *Pseudomonas* spp. that have been sequenced to date. As expected from the spectrum of antibiotics produced by Pf-5, many biosynthetic gene clusters for secondary metabolite production are present in the genome. 4% of the genome is devoted to the biosynthesis of secondary metabolites, which explains (at least in part) why the Pf-5 genome is larger than those of other *Pseudomonas* spp. that don't produce many secondary metabolites. Genes involved in the biosynthesis of each antibiotic are clustered together, but the clusters for the different antibiotics are distributed throughout the genome.

There is no evidence for the presence of genes involved in pathogenicity (ie. phytotoxins, Type III secretion systems, effectors, etc.) in the genome of Pf-5; such genes are found in the genomes of phytopathogenic *P. syringae*. Therefore, there are significant differences in the genomes of phytopathogenic and biocontrol strains of *Pseudomonas* spp.

Future Directions

Three new gene clusters for secondary metabolite biosynthesis were found in the genome of Pf-5, in addition to the clusters for production of the antibiotics known to be produced by this bacterium. We are currently using a combination of approaches from molecular biology and natural products chemistry to identify the three unknown metabolites produced by Pf-5. Once identified, the toxicity of these metabolites towards soilborne fungi and their potential for suppressing plant diseases will be determined.

For more information

Visit the Pf-5 genome website:

<http://www.ars-grin.gov/hcrl/Pf5genome/index.htm>

References Cited

Howell, C. R. and R. D. Stipanovic . 1979. Control of *Rhizoctonia solani* in cotton seedlings with *Pseudomonas fluorescens* and with an antibiotic produced by the bacterium. *Phytopathology* 69:480-482.

Howell, C. R. and R. D. Stipanovic. 1980. Suppression of *Pythium ultimum* induced damping-off of cotton seedlings by *Pseudomonas fluorescens* and its antibiotic pyoluteorin. *Phytopathology* 70:712-715.

Nowak-Thompson, B., N. Chaney, J. S. Wing, S. J. Gould, and J. E. Loper. 1999. Characterization of the pyoluteorin biosynthetic gene cluster of *Pseudomonas fluorescens* Pf-5. *J. Bacteriol.* 181:2166-2174.

Nowak-Thompson, B., S. J. Gould, J. Kraus, and J. E. Loper. 1994. Production of 2,4-diacetylphloroglucinol by the biocontrol agent *Pseudomonas fluorescens* Pf-5. *Can. J. Microbiol.* 40:1064-1066.

Paulsen, I. T. et al. 2005. Complete genome sequence of the plant commensal *Pseudomonas fluorescens* Pf-5. *Nature Biotechnology* 23: 873-878.

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