



Biological control of *Phytophthora capsici* root rot of pepper (*Capsicum annuum*) using *Burkholderia cepacia* and *Trichoderma harzianum*

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ABSTRACT

Objective: To control *P. capsici* using a combination of two independently effective and compatible microorganisms, *Trichoderma harzianum* and *Burkholderia cepacia*.

Methodology and results: A combination of two compatible microorganisms, *Trichoderma harzianum* and *Burkholderia cepacia*, both antagonistic to the pathogen *Phytophthora capsici*, was used to control root rot in pepper. The population of the pathogen in soil was

reduced by 71% as a result. Vegetative growth of the mycelium of *P. capsici* was inhibited in vitro on the second day after *P. capsici* and *T. harzianum* were placed on the opposite sides of the same Petri plate. *T. harzianum* was capable of not only arresting the spread of the pathogen from a distance but also, after invading the whole surface of the pathogen colony, sporulating over it. Scanning electron microscopy showed the hyphae of *P. capsici* surrounded by those of *T. harzianum*, their subsequent disintegration, and the eventual suppression of the pathogen's growth. *Burkholderia cepacia* produced a zone of inhibition, from which was obtained a compound with anti-oomycete property secreted by the bacteria. When purified by high-pressure liquid chromatography, this compound was identified as pyrrolnitrin which seems to be one of the principal compounds involved in the antagonism. Finally, a strategy was tested for increasing the level of the hydrolytic enzyme β -1,3-glucanase and chitinase in the antagonism test of *P. capsici* versus *T. harzianum* on PDA medium enriched with laminarin: glucose (3:1, v/v). Chitinolytic activity was evaluated using culture filtrates from isolates grown on chitin and the *Agaricus bisporus* mushroom as the release of p-nitrophenol from p-nitrophenyl N, N'-diacetylchitobiose and as the formation of clearing zones on chitin agar. A formulation was prepared that maintained the compound's capacity to inhibit growth of the pathogen for up to two years when stored at room temperature in the laboratory on a mixture of plantation soil and vermiculite. The two antagonists, added as a compound formulation, were effective at pH from 3.5 to 5.6 at 23 to 30 °C. The optimal dose of the antagonists in the compound formulation was 3.5×10^8 spores/ml of *T. harzianum* and 10^9 cfu/ml of *Burkholderia cepacia*.

Conclusion and application: This is the first report of a compound biocontrol formulation of these two antagonists with a potential to control root rot caused by *P. capsici*.

Key words: Biocontrol, *Capsicum annuum*, *Phytophthora capsici*, *Burkholderia cepacia*, *Trichoderma harzianum*