

Title Potential biocontrol activity of a strain of *Pichia guilliermondii* against grey mold of apples and its possible modes of action

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Abstract

The efficacy of *Pichia guilliermondii* strain M8 against *Botrytis cinerea* on apples was evaluated under storage conditions, and its possible modes of action were investigated both *in vitro* and *in vivo* experiments. After storage at 1 °C for 120 days, M8 reduced grey mold incidence from 45.3% (control) to 20.0%. In apple juice medium (AJM) and in wound-inoculated apples, M8 at 10^9 and 10^8 cells ml⁻¹ inhibited the spore germination of *B. cinerea* and the grey mold development. When co-culturing *B. cinerea in vitro* or *in vivo* in the presence of the yeast, neither inactivated cells nor culture filtrate of the yeast had any effect on spore germination or germ tube elongation. In AJM, the spore germination was significantly recovered by the addition of 1% glucose, sucrose and fructose, or 0.5% and 1% of (NH₄)₂SO₄, phenylalanine and asparagine. When the pathogen and the yeast were co-incubated in apple wounds with addition of the same nutrients, the inhibition of rots was significantly reduced by the supplemental nutrients. Light microscopy revealed that the yeast strongly adhered to the hyphae and spores of *B. cinerea*. M8 produced hydrolytic enzymes, including β-1,3-glucanase and chitinases in minimal salt media with different carbon sources. Pretreatment with M8 at 10^8 cells ml⁻¹ followed by washing, significantly reduced grey mold lesions, suggesting an induction of defense responses. Direct attachment, competition for nitrogen and carbon sources, secretion of hydrolytic enzymes and induction of host resistance play a role in the biocontrol mechanism of *P. guilliermondii* M8 against *B. cinerea*.