Genetic and biochemistry analyses of the natural resistance of the fungicide fenhexamid in the phytopathogenic fungus Botrytis pseudocinerea

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Genetic and biochemistry analyses of the natural resistance of the fungicide fenhexamid in the phytopathogenic fungus *Botrytis pseudocinerea*

The *Botrytis* species complex responsible for grey mould disease on grapevine is composed of two species: *Botrytis cinerea* the major one (about 90%) and *Botrytis pseudocinerea*. Despite their genetic polymorphism, these species cannot be morphologically distinguished. However, they do differ in their response to several fungicides, especially to the sterol biosynthesis inhibitor fenhexamid. While *B. cinerea* is sensitive to this hydroxyanilide, *B. pseudocinerea* is naturally resistant. Enzyme assays showed that in *B. pseudocinerea* the fenhexamid target enzyme, the sterol 3-ketoreductase was less sensitive to fenhexamid. In addition, a synergic effect between fenhexamid and sterol 14A-demethylation inhibitors (DMIs) known to inhibit Cyp51, a cytochrome P450 monooxygenase was observed in *B. pseudocinerea*. These results could suggest detoxification of fenhexamid by cytochromes P450. The cyp684 gene showing the strongest similarity to cyp51 among all *B. cinerea* cytochrome P450 genes was found strongly overexpressed in the presence of fenhexamid in *B. pseudocinerea*. In this work, we studied separately the effect of *B. pseudocinerea* erg27 polymorphism, erg27 encoding 3-ketoreductase, and of the recently identified cytochrome P450 gene, cyp684, on resistance to fenhexamid. The objective is to determine their respective implication in resistance. Experiments were conducted by exchange between erg27*B.cinerea* and erg27*B.pseudocinerea* in *B. cinerea* and by cyp684 deletion in *B. pseudocinerea*. In parallel, metabolization studies are conducted to identify metabolites and test their activity on *Botrytis spp*.