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EDITORS
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EVALUATION OF VIRULENCE AND THE OXALIC ACID PRODUCTION ON *Cryphonectria parasitica* VIRULENT AND CONVERTED STRAINS BY CHV1 HYPOVIRUS

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ABSTRACT

Chestnut Blight, caused by *Cryphonectria parasitica* (Murrill) Bar, is a major disease in *Castanea sativa* Mill. on the European continent. Biological control by hypovirulence is a sustainable and efficient method to control the disease. The presence of *Cryphonectria hypovirus* 1 (CHV1) in *C. parasitica* reduces the fungus virulence that promote canker healing and tree recovery. Hypovirus infection results in phenotypic and metabolic changes, including the reduction of ligninolytic enzymes activity, and decreased oxalic acid production. The aim of this work was to evaluate the oxalic acid production in both virulent and converted strains on PDB (Potato Dextrose Broth, 24g/L) and to access the virulence of these strains on chestnut stems. Six isolates were converted with two characterized hypovirulent *C. parasitica* isolates (RBB111, SR44.2) and the presence of CHV1 was detected by molecular methods. Oxalic acid production was evaluated by spectrophotometry after the growth of the strains on 100 ml of PDB supplemented with 2mM $MnSO_4$ in an orbital incubator during five days. To evaluate the virulence of the isolates, chestnut stems were inoculated with the virulent isolates, their converted ones and the hypovirulent isolates. The characterized hypovirulent isolates used in this work has complete ability to convert virulent isolates with effective hypovirus transmission and PCR detection of CHV1 was obtained in all *C. parasitica* converted strains. The obtained results by spectrophotometric analysis have revealed that virulent strains always produced more

oxalic acid than converted strains. The infection area on chestnut stems caused by virulent strains was significantly higher ($P<0.05$) than the infection area caused by converted strains. The converted strains Cast26/RBB111 and Cast26/SR44.2 showed 50% and 88.6% reduction in the content of oxalic acid present in supernatant, respectively. This suggests that the reduction in enzymatic activities caused by hypovirulent strains is variable with the hypovirulent donor used in conversion.

Keywords: European chestnut, pathogenesis, oxalate