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Improved *Metarhizium brunneum* Endophytism for Biological Protection of Potato Plants

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Abstract

Conventional insect pest control faces a growing number of problems, such as the decreasing efficacy of many commercial insecticides, the need for repetitive application as well as ecotoxicological issues. Biocontrol with endophytic entomopathogenic fungi is a promising alternative to chemical pesticides to protect crop plants systemically from insect herbivores, paving the way for a novel plant protection measure. Yet, current applications are limited especially by low fungal plant colonisation, a requirement for endophytic biocontrol.

Inspired by penetration mechanisms of plant pathogenic fungi, we aimed at enhancing potato plant penetration and colonisation by the endophytic entomopathogenic *Metarhizium brunneum* strain Cb15 through supplementation of plant cell-wall degrading enzymes pectinase and cellulase or corresponding substrates into beads containing mycelial biomass.

We found that after bead application to potato tubers and incubation at 18–23 °C with a LD cycle of 16:8 for 21 days, *M. brunneum* was re-isolated from surface-sterilized roots, tubers, and shoots in 75.0 ± 9.4 %, 33.3 ± 9.8 %, and 29.2 ± 15.1 % of samples, respectively. Additional verification of re-isolated *M. brunneum* was conducted with qPCR. Pectinolytic and cellulolytic enzymes were successfully induced by addition of corresponding substrates, but activity levels were low and no correlation between enzymatic activity and fungal penetration was found. However, incorporation of cellulase into beads led to a substantial increase in plant penetration by 25.0 % in roots, 54.2 % in tubers, and 16.6 % in shoots. This was accompanied by a 3.0-fold enhanced spore formation on the surface of beads to $1.91 \times 10^8 \pm 0.26 \times 10^8$ per bead. Finally, a stronger root development of treated plants was observed indicating a fertilising effect mediated by the formulation.

Our study provides first evidence that refined formulations of endophytic entomopathogenic fungi could contribute to a more effective use of these fungi in strategies priming plants against biotic and abiotic stress.

Keywords: Biological crop protection, endophytes, formulation, potato plants