Distribution of fungal endophyte genotypes in doubly infected host grasses

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Abstract

Fungal endophytes of the genus *Epichloë* live intercellulary in above ground plant parts of many poold grasses of the temperate regions. The associations are characterized by single genotype entities since a given host individual normally contains a single endophyte genotype. They can persist over the life span of the hosts. This study examines whether two fungal genotypes can coexist within a host plant, and how fungal genotypes are distributed within a host in the case of double infections. We selected four *Epichloë bromicola* strains that we identified as unique genotypes through RAPD analysis. Young Bromus erectus plants, derived from callus cultures, were artificially inoculated with all possible doublestrain mixtures of these fungal genotypes. For identification of fungal genotypes in planta, we designed genotype specific primer pairs that flanked size-variable loci in the fungal genomes. Diagnostic PCR revealed that only one fungal genotype was present in most inoculated plants, but double infections were also observed with a frequency of 8% of all infected plants. Subsequent analyses of individual tillers of doubly infected plants revealed that in a given tiller, both the leaf-blade and the leaf-sheath were colonized with only one endophyte genotype. Tillers without any detectable fungal DNA were observed as well. Thus, coexistence of multiple endophyte genotypes within a single host plant is governed by mutual exclusion at the tiller level.