Poster Presentation 2

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Optimization of protoplast generation and polyethylene glycol-mediated transformation of the pepper anthracnose pathogen *Colletotrichum scovillei*

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Colletotrichum acutatum is a species complex causing anthracnose disease in a wide range of crops. We isolated a *Colletotrichum* species from an infected pepper in Gangwon Province of South Korea. The isolate was identified as *C. scovillei* using combined sequence analyses of the nuclear ribosomal internal transcribed (ITS) region, partial sequences of the chitin synthesis 1 (CHS-1), β-tubulin (TUB2), actin (ACT), , histone 3 (HIS3), and an intron sequence of the glyceraldehyde-3-phosphate dehydrogenase (GAPDH). *C. scovillei* is a member of the *C. acutatum* species complex and recently shown to cause anthracnose disease on pepper in South Korea. In this study, we optimized the transformation system of *C. scovillei*. The maximum release of protoplasts was produced from young hyphae of *C. scovillei* in an enzymatic digestion of 2% lysing enzyme and 0.8% driselase in 1M NH_4Cl for 3 h incubation. The optimal concentration of hygromycin B as a selection marker was 200 μg/mL in regeneration media. Next, we tested PEG-mediated transformation of *C. scovillei* protoplats using 19 different loci. The average number of candidates for gene deletion mutants was 17.1% in the PCR screening. By southern blot analysis, we confirmed that at least one transformant among 2-5 PCR-screened positive transformants per locus had single copy integration.