

August. Our findings are the first evidence of SBFS species-specific patterns for timing of inoculum deposition and infection on apple fruit.

Phylogenetic analyses to assess the evolutionary origins of sooty blotch and flyspeck fungi on apple

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The sooty blotch and flyspeck (SBFS) complex of ascomycetes causes economically important blemishes of apple fruit worldwide. About 90% of SBFS species are in the order Capnodiales. However, evolutionary relationships of SBFS fungi with close relatives that do not cause SBFS remain unclear. We attempted to reconstruct the evolutionary history of major SBFS lineages by using ancestral state reconstruction, utilizing the 28S nuclear large subunit region (LSU) of rDNA and RPB2, which encodes the largest subunit of RNA polymerase II. The analyzed taxa encompass numerous genera of SBFS and non-SBFS fungi from seven families within the Capnodiales. The non-SBFS taxa were selected based on their distinct ecological niches, including plant parasites, animal parasites, and saprobes. Results of phylogenetic analysis of LSU sequences suggest that most SBFS species are closely related to plant parasitic fungi. A preliminary ancestral state reconstruction based on LSU data suggests that plant parasitism represents an ancestral state for most SBFS lineages. Further ancestral state reconstruction of ecological niche of SBFS fungi and their closest relatives are underway using a Bayesian approach and RPB2 sequences. Knowledge gained from this study may help us to better understand the ecology and evolution of epiphytic plant-inhabiting fungi on apple fruit.

Characterization of plant growth-promoting and disease suppressing abilities of certain actinomycetes isolated from groundnut rhizosphere

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Actinomycetes are widely used bacterial biocontrol agents in plant disease management. The present study focused on isolation and characterization of certain actinomycetes from groundnut rhizosphere for growth-promotion and pathogen suppressiveness. Dual culture studies were conducted to assess the antifungal abilities against aflatoxin producing fungi (*Aspergillus flavus*) and stem rot pathogen (*Sclerotium rolfsii*). Sixteen isolates showing high antagonism were further characterized for biocontrol and growth-promotion in groundnut. *In-vitro* antifungal assays indicated that 50% inhibition was exhibited by 10 isolates against *S. rolfsii* and more than 40% inhibition was shown by five isolates against *A. flavus*. Two superior isolates (RP1A-12 and RP1A-15) were selected based on fungal antagonism and crop growth promotion. Culture filtrates of RP1A-12 and RP1A-15 exhibited good antagonism against test pathogens, with crude extract of RP1A-15 showing complete inhibition of the mycelia at 1.5%. The HPLC chromatograms of RP1A-15 and RP1A-12 crude extracts showed eight and three peaks respectively. Separation of eight compounds in RP1A-15 was obtained with silica gel flash column chromatography. Bio-efficacy studies were conducted on the obtained fractions and results suggested one fraction significantly delayed the germination of sclerotia of *S. rolfsii*. Identification of these two isolates using 16S rRNA sequencing and bio-active fraction using LC-MS, NMR are in progress.

Microscopic interactions between butternut (*Juglans cinerea*) trees and the butternut canker fungus (*Ophiognomonia clavignenti-juglandacearum*)

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Butternut canker is a lethal disease of butternut (*Juglans cinerea*) trees caused by the fungus *Ophiognomonia clavignenti-juglandacearum* (Oc-j). The disease has decimated populations of the species in many areas and is one of the main impediments to maintaining *J. cinerea* as a component of hardwood forests throughout its native range. Small elliptical cankers form at wound sites and natural openings on all woody tissues. Coalescence of multiple cankers kills branches and trees of all ages. In addition to complications from disease, *J. cinerea* readily hybridizes with non-native Japanese walnut (*J. ailantifolia*). Recently, Oc-j was found to cause leaf lesions on butternut and its hybrid. Prior research on the histological interactions between the fungus and host focused solely on canker progression within stem tissue. Little is

known about surface interactions between the primary inoculum in the pathosystem (conidia) and leaf and stem tissues. In this study, detached leaflets and stem sections in moisture chambers were inoculated with deionized water suspensions of Oc-j conidia and interactions were analyzed by scanning electron microscopy. We report evidence that spore germination on plant surfaces readily occurs. Subsequent hyphal growth is apparently haphazard and no active penetration structures were observed. We further report the likely mode of infection through the surface of non-wounded leaflets, as well as infection development within foliar tissue.

***Ralstonia solanacearum* requires PopS, an ancient virulence effector, to suppress SA-mediated defenses during tomato wilt**

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Ralstonia solanacearum requires a Type III (T3) secretion system for bacterial wilt pathogenesis, but the biological functions of individual effectors remain unknown. During tomato wilt, *R. solanacearum* expresses *popS*, which encodes an AvrE-family T3 effector. *popS* homologs were present in all 17 sequenced *R. solanacearum* strains, and the phylogeny of *popS* parallels that of the *R. solanacearum* species complex, suggesting that PopS is an ancient effector needed for association with plants. We determined that *popS* is required for full virulence on multiple *Solanum* crop hosts (susceptible potato and susceptible and quantitatively resistant tomato), but not for virulence on a related epidemiologically relevant weed, *S. dulcamara*. The *popS* mutant was also significantly delayed in tomato stem colonization following direct inoculation through cut petioles. AvrE-type effectors in other plant pathogenic bacteria suppress plant defenses triggered by the plant signaling molecule salicylic acid (SA). The *popS* mutant induced higher expression of SA-responsive tomato *PR* genes than its wild-type parent. Further, pretreating plant roots with SA exacerbated the *popS* virulence defect. Finally, PopS was dispensable for bacterial colonization of SA-deficient NahG transgenic tomato plants. These results indicate that this conserved T3 effector suppresses SA-mediated defenses in tomato roots and stems, which are the natural infection courts of this soilborne vascular pathogen.

Temporal dynamic of *Aspergillus flavus* community structure in soils of fields treated with the atoxigenic biocontrol *A. flavus* AF36 in Arizona

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Aflatoxins, toxic and carcinogenic metabolites produced by *Aspergillus* Section *Flavi*, frequently contaminate crops. Atoxigenic members of *A. flavus* competitively displace aflatoxin producers and consequently reduce aflatoxin contamination. The atoxigenic *A. flavus* AF36 is a natural inhabitant of soils in Arizona and has been successfully used commercially to reduce contamination in cottonseed and other crops. After successful applications, the population structure of *Aspergillus* in the soil is modified benefiting subsequent crops. Populations in treated fields reach equilibrium with those in non-treated fields in aflatoxin-producing potential without additional applications. The time and factors influencing equilibrium have not been examined. The current study sought to describe the process of equilibrium after application of AF36. Population structures of *A. flavus* in soils of twelve fields with cotton crops treated with AF36 were examined several times during four years in two areas. Results indicate that over time, AF36 decreased, while the highly toxigenic strain S increased to reach equilibrium with populations across the areas, but at higher AF36 incidences than before applications. However, time required to reach equilibrium (strain S > 75% and AF36 < 10%) was differential between the Yuma Valley (2 years) and the Mohawk Valley (3 years). In both areas, soils of treated fields had *A. flavus* populations containing over 50% AF36 one year after application.

Transformation of *Liberibacter crescens* using two wide host range shuttle vectors

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Huanglongbing (HLB) is caused by '*Ca. Liberibacter asiaticus*' (Las), '*Ca. L. americanus*' (Lam) and '*Ca. L. africanus*' (Laf), a group of alpha proteobacteria that have not been cultured. At least three other *Liberibacter* species have been described, including '*Ca. L. solanacearum*' (Lso, affecting potato, tomato, and carrots), '*Ca. L. europaeus*' (Lep, a nonpathogenic endophyte of