beta, and SFBB-gamma) have been isolated of which the SFBB-alpha genes were identified as the best candidates for pollen S-genes. To investigate sequence characteristics of the possible pollen S-gene and elucidate possible GSI mechanism of Chinese sand pear (*P. pyrifolia*), SFBB-alpha homologies in Chinese sand pear cultivars were cloned and characterized by degenerate primers, PSFBA-F and PSFBA-R, that were designed based on Japanese pear and apple SFBB-alpha sequence information. Results showed that the PSFBA-F and PSFBA-R primers successfully amplified a fragment of approximately 1,300 bp from 'Maogong' ($S_{12}S_{13}$), 'Hongsucui' ($S_4S_{12}$), 'Tianchengzi' ($S_7S_{12}$), and 'Mantianhong' ($S_4S_{12}$), corresponding to pear SFBB-alpha gene. A total of two distinct sequences derived from the 1,300 bp product were identified that were named PpSFBB12-alpha (*P. pyrifolia* SFBB12-alpha) and PpSFBB13-alpha, respectively. RT-PCR revealed that both PpSFBB-alpha genes were expressed specifically in the pollen grains. The coding region of PpSFBB12-alpha was 1,194 bp in length encoding 397 amino acids with predicted molecular weight of 45.8 kDa and isoeletric point of 5.03. The coding region of PpSFBB13-alpha was 1,179 bp in length encoding 392 amino acids with predicted molecular weight of 45.4 kDa and isoeletric point of 4.92. Both genes displayed normal structural characteristics of SFB/SLF genes, i.e. an F-box motif and four variable regions. At the deduced amino acid level, they shared 18.6% to 97.7% similarities with other SFB/SLFs of rosaceous plants. These characteristics of the two PpSFBB-alpha genes fully demonstrated that they are good candidates of pollen S-gene. Phylogenetic analysis revealed that 34 rosaceous SFB/SLFs were dived into two subfamily groups, but did not further form species subgroup. The evolutionary pattern of SFB/SLFs concurred with that of rosaceous S-RNases, suggesting that SFB/SLFs occurred after divergence of subfamily, but before the divergence of species as S-RNases in Rosaceae. The two PpSFBB-alpha sequence data should be useful for studying the interaction between SFBs and S-RNases and clarifying the mechanism of GSI at the molecular level in Chinese sand pear.

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5:45–6:00 pm

### Multiple Approaches for Genetic Engineering of Citrus for Disease and Pest Resistance

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Citrus in Florida is threatened by non-indigenous but now endemic bacterial pathogens causing the diseases Huanglongbing (HLB, caused by *Candidatus* Liberibacter asiaticus) and citrus canker (caused by *Xanthomonas citri* ssp. citri). These diseases have the potential to wipe out the entire industry due to unavailability of resistance in commercially cultivated Florida cultivars. HLB is vectored by the Asian citrus psyllid (*Diaphorina citri* Kuwayama), while canker is dispersed by windblown rain, contaminated equipment or human activity. In efforts to control the diseases, several transgenic strategies have been adopted. Citrus scion and rootstock cultivars were transformed with gene(s) encoding antimicrobial peptides, systemic acquired resistance (SAR) proteins or insecticidal proteins. Each gene was under control of an enhanced CaMV 35S promoter. Several genes were also under control of a phloem specific *Arabidopsis* SUC2 (*AtSUC2*) promoter as *C*. Liberibacter asiaticus is a phloem limited bacterium and targeting gene expression in the phloem potentially resolves issues of presence of transprotein in the fruit. Molecular analysis confirmed the presence of the gene(s) in the citrus genome. We employed a rapid micrografting technique on tender rootstocks to quickly propagate our transgenic lines. Results from preliminary disease resistance tests suggested that some of the antimicrobial peptides could provide varying levels of resistance against both HLB and canker. Several studies are currently underway to evaluate resistance to either of the pathogens and also to the insect vector.

## Oral Session 4: Jefferson A
## Horticultural Crops Culture and Management: Pest Management
## Saturday, 25 July 2009, 4:00–6:00 pm

**Moderator: Beiquan Mou, beiquan.mou@ars.usda.gov**

4:00–4:15 pm

### Effectiveness of Host Resistance Inducers and Humic Acid for Fire Blight Control and Shoot Growth on Apple Cultivars with M9 Rootstock

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Fire blight, caused by *Erwinia amylovora*, is the most devastating bacterial disease of apples with M9 rootstocks and often results in the death of the tree. The resistance-inducing compounds prohexadione-Ca (PC), harpin protein (Hp) and benzothiadiazole (acibenzolar-S-methyl) (BTH), the fertilizer humic acid (HA), the bactericides streptomycin and copper, and combinations of copper with chemicals were evaluated for their ability to control fire blight and shoot growth on apple cultivars, Gala, Red Elstar, Jonagold and Pinova, with M9 rootstock in two years. PC was applied at a rate of 125 $mg \cdot L^{-1}$ at two shoot lengths (6–12 cm and 15–20 cm), while BTH (135 $mg \cdot L^{-1}$) and harpin (50 $mg \cdot L^{-1}$) were applied when the shoots measured between 15–20 cm, and again at 30–35 cm. HA (200 $mg \cdot L^{-1}$), copper and streptomycin were applied at three phase of shoot lengths. In bioassay tests, bacterial populations were determined in plant tissues at 5th, 10th and 15th days. Disease severity and effectiveness of the applications were calculated using the following formulas: Disease severity (%) = (a/b) × 100; where a is the length of the blighted part of the shoot (cm), and b is the whole length of the shoot (cm) and Efficacy (%) = [(DIck – DItr) / DIck] × 100 where DIck is mean disease severity in water sprayed control plants and DItr is mean disease severity in treated plants. When the apple varieties and years were taken into consideration, PC alone (44% to 70%) and in combination with copper compound (48% to 63%) were the most effective treatment during both years on disease control, following by the streptomycin (76% to 95%) ($P$ □ 0.05). PC reduced both shoot length and shoot blight on the apple cultivars. The cultivar Red Elstar showed the lowest disease severity than the other cultivars and followed Jonagold, Pinova and Gala, respectively. HA applications were unsuccessful in controlling fire blight on all of the apple cultivars. Streptomycin treatment yielded the lowest rate of bacterial population, and it was followed by PC, Hp, BTH, copper and HA respectively. The use of resistance-inducing substances during the early phase of shoot growth may offer a means of managing the shoot blight phase of fire blight disease on apple cultivars with M9 rootstock.

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