AGENDA
Diaprepes Task Force/Citrus Growers Meeting
November 16, 2006

United States Department of Agriculture
2001 South Rock Road
Ft. Pierce, FL 34945

Conveners: Steve Lapointe and Larry Duncan

9:00	**Welcome, Introductions, Announcements**	Garvie Hall, *Chair* Constance Riherd, *Vice Chair*
	Research Updates	
	Behavior: Attractants, baits and monitoring	
9:15	Update on the search for a Diaprepes root weevil attractant	S. L. Lapointe[2], D. G. Hall,[2] and J. C. Dickens[8]
9:30	Research on monitoring adult Diaprepes root weevil using traps baited with an attractant	D. G. Hall[2] and S. L. Lapointe[2]
9:45	Concerns of citrus growers in California and Texas about Diaprepes and their interest in detection methods	R. Mankin[7]
10:00	**Discussion**	
10:20	**Break**	
	Host Plant Resistance	
10:35	Screening rootstock genotypes for tolerance to *Phytophthora-Diaprepes* under field conditions	J. H. Graham,[1] K. D. Bowman,[2] D. B. Bright,[1] and R. C. Adair[3]
10:50	Search, assessment and development of a Bt-gene based strategy for resistance to *Diaprepes*	R. Shatters, Jr.,[2] S. Lapointe,[2] A. Weathersbee,[2] and D. Hall[2]
11:05	Progress in the development of new rootstocks tolerant of the *Diaprepes/Phytophthora* complex	J.W. Grosser, [1] J.H. Graham, [1] D. Bright, [1] A. Hoyte, [1] and H.M. Rubio[1]
11:20	Potential control of *Diaprepes abbreviatus* with TMOF and Cathepsin L insect specific protein inhibitor	D. Borovsky,[5] C. Powell,[6] and R. Shatters, Jr.,[2]
11:35	**Discussion**	
12:00	**Lunch**	
	Biological control	
1:00	The effect of climate on distribution of *Diaprepes* root weevil and associated egg parasitoids	S. L. Lapointe,[2] D. G. Hall,[2] and D. M. Borchert[9]
1:15	Update on classical biological control of the *Diaprepes* root weevil	J. E. Pena,[4] D. Carrillo,[4] R. Duncan,[4] B. Ulmer,[4] C. McCoy,[1] and D. Hall[2]

1:30	Is there a role for conservation biological control with entomopathogenic nematodes?	L.W. Duncan,[1] R.J. Stuart,[1] J.H. Graham,[1] J. Zellers,[1] D. Bright,[1] D. Dunn,[1] and F.E. El-Borai[1]
1:45	**Discussion**	
	IPM	
2:05	Update of *Diaprepes* root weevil on ornamental plants	C. M. Mannion[4]
2:20	Managing *Diaprepes* through rootstock tolerance and strategic pesticide applications: the long-term field study at Southport	R. Stuart,[1] M. Rogers,[1] and W. Castle[1]
2:35	**Discussion**	
	Other	
3:00	Characterization and evolution of delta 9 desaturase from *Diaprepes abbreviatus* (L.): The first report of a desaturase gene from the Coleoptera	L.M. Boykin,[2] K.S. Katsar, [2] S.L. Lapointe, [2] and W. Hunter[2]
3:15	Regulating the growth and development of *Diaprepes abbreviatus* on artificial diets	R.P. Niedz,[2] T. J. Evens, [2] and S.L. Lapointe[2]
3:30	**Discussion**	

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UPDATE ON THE SEARCH FOR A DIAPREPES ROOT WEEVIL ATTRACTANT. *S. L. Lapointe,[1] D. G. Hall[1] and J. C. Dickens,[2] [1] U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS); [2] U. S. Department of Agriculture/Beltsville Agricultural Research Service/Chemicals Affecting Insect Behavior Laboratory, Beltsville, Md. (USDA/ARS/CAIBL)*

An aggregation pheromone for the Diaprepes root weevil (DRW) has been sought since shortly after its introduction into the United States in the 1960s. We have re-initiated the search for a chemical attractant. We have demonstrated the presence of at least two DRW-produced volatiles that elicit consistent responses from DRW antennae by GC/EAD. One compound elicits high antennal response at extremely low concentrations. The compound appears to be produced by males and females suggesting a role in aggregation.

RESEARCH ON MONITORING ADULT DIAPREPES ROOT WEEVIL USING TRAPS BAITED WITH AN ATTRACTANT *D. G. Hall and S. L. Lapointe, U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

A field study in young citrus trees was conducted to evaluate and compare boll weevil eradication traps and universal moth traps for their potential as tools for screening candidate attractants for adults of the citrus root weevil *Diaprepes abbreviatus* (L.). Extracts of adult weevil frass were used as the weevil attractant in the study. The boll weevil eradication traps (BWET) were modified by enlarging the crawl space through the base of the trap into the trap's confinement cage. Universal moth traps (UMT) had not been investigated as a trap for adult *D. abbreviatus*. Vaportape® II (DDVP) kill strips (2.5 x 10 cm, 0.229 g ai/cm^2, 5.9 g ai) are commonly used to subdue and kill insects that enter UMT. Prior to the field test, the following were shown to provide 93 to 100% mortality of adult weevils during a 24 h exposure period in UMT: whole strips (2.5 x 10 cm, 5.9 g ai), half strips (2.5 x 5 cm, 2.95 g ai) and quarter strips (2.5 x 2.5 cm, 1.475 g ai). There were no significant differences among the three kill-strip rates with respect to percentage mortality. Low percentages ($\leq$ 5.0%) of weevils escaped from UMT before being subdued/killed. No significant reduction in activity was found among kill strips left in the field for up to 10 weeks based on percentage mortality of adults exposed to the strips. For the field study, BWET were deployed at the top of the tree canopy and UMT were deployed just above the middle of the tree canopy. Over an 11 day trapping period in a grove heavily infested by adult weevils, a total of 30 and 7 weevils were collected at BWET baited and not baited, respectively, with weevil frass extract. Totals of 10 and 4 weevils were collected at UMT that were baited and not baited, respectively, with frass extract. Significantly greater numbers of adult male and female weevils were captured at BWET baited with frass extract than at the other traps. No significant differences in numbers of adults collected were observed in the absence of frass extract between BWET and UMT. Adding frass extract to UMT tended to increase captures of adults but not significantly. Significantly greater numbers of males were collected than females at traps baited with frass extract, and the ratio of males to females at these traps was 2:1. The results of this study indicated that BWET positioned in the upper canopy of citrus trees may currently be the best choice for testing candidate attractants for the weevil under field conditions, and that traps with frass extracts could be included for comparison purposes. However, based on the large population of weevils at the study site, BWET baited with frass extracts were relatively inefficient for attracting and capturing adult *D. abbreviatus*.

CONCERNS OF CITRUS GROWERS IN CALIFORNIA AND TEXAS ABOUT DIAPREPES AND THEIR INTEREST IN DETECTION METHODS *R. Mankin, U. S. Department of Agriculture/Agricultural Research Service/Center for Medical, Agricultural, and Veterinary Entomology, Gainesville, Fla. (USDA/ARS/CMAVE)*

Diaprepes was first detected in two orchards near McAllen, TX in November 2000. The area was quarantined immediately and it has been monitored using Tedder's traps since then. Diaprepes was discovered in ornamentals in urban areas of Orange County and Los Angeles County in 2005, and then in urban areas and lemon groves in San Diego County in 2006. The affected areas have been quarantined, and a public awareness campaign is under way, asking homeowners to alert inspectors of any sightings. Inspectors, researchers, and growers in both states have expressed interest in identifying and implementing technologies that have potential for detecting small infestations, but until now, visual inspection for adults has been the primary detection method.

SCREENING ROOTSTOCK GENOTYPES FOR TOLERANCE TO PHYTOPHTHORA-DIAPREPES UNDER FIELD CONDITIONS *J. H. Graham[1], K. D. Bowman[2], D. B. Bright[1] and R. C. Adair,[3] [1] University of Florida/Institute of Food of Agricultural Sciences, Citrus Research and Education Center, Lake Alfred, Fla. (UF/IFAS/CREC); [2] U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS); [3] Florida Research Center for Agricultural Sustainability, Vero Beach, Fla. (FLARES)*

Rootstock germplasm from USHRL breeding program was evaluated in each of three growing seasons at the Florida Research Center in Vero Beach. The screening site was located on Winder and Manatee fine sand soil series and was naturally infested with *Diaprepes abbreviatus* and *Phytophthora nicotianae* and *P. palmivora*. Several studies showed this site to be conducive from development of Phytophthora-Diaprepes (PD) complex and for distinguishing tolerance of rootstocks based on tree performance. Seedlings (6 mo-old?) of mandarins, sour oranges, sweet oranges, pummelos and trifoliates and sexual hybrids thereof were grown in conetainers. A mixture of rhizosphere soil with fibrous roots was harvested from the 0-10 cm depth of the soil profile beneath Sunburst trees on Swingle rootstock that supported moderate populations of both *Phytophthora* spp. To ensure that all seedlings were exposed to the two *Phytophthora* pathogens, 200 cm^3 of the root/soil mixture was placed in the bottom of each planting hole before the seedling was set. Diaprepes exposure was due to egg laying adults immigrating from older trees adjacent to the test block. Plantings were established in May of 2002 and 2003 and in January of 2005. Seedlings were fertilized and irrigated as needed with a micro-jet irrigation system to sustain Phytophthora infection.

Seedlings were harvested in January 2003, March of 2004, March of 2006 at 6, 7 and 10 months after planting in the 2002, 2003 and the 2005 season, respectively. Trees were carefully excavated with a shovel to keep the fibrous root system intact. A handful of soil was removed from below the root zone during the excavation process. Soil samples from each tree were dilution plated onto semi-selective PARPH medium for enumeration and identification of *Phytophthora spp*. Whole root systems and structural roots were visually rated for root rot by the fungi and feeding damage by the weevil on a scale from 1-5 (1 = no damage, 5 = no undamaged roots). Tissues were dried and shoot, structural root and taproot weights were measured.

When 2002 and 2004 data were combined there was a significant positive correlation between whole root system damage and total Phytophthora populations. Among the genotypes, mandarins and pummelo hybrids showed greater tolerance to PD complex than trifoliate and some of its hybrids. Tolerance was judged by whether the genotypes supported less than 20 propagules of total Phytopthora per cm^3 of soil. In 2005, screening focused on hybrids of pummelo and sour orange for which the relationship of root damage with populations was weak and the majority of the genotypes had less than 20 propagules. Overall, the tolerance of genotypes in the third year was greater than for the populations of genotypes tested in the first two years of screening.

These findings: 1) validate use of field screening of rootstock seedlings for early assessment of genotype tolerance to PD complex, and 2) confirm the promise of certain pummelo and mandarins as parents for hybrids with requisite Phytophthora resistance to develop rootstocks tolerant to the PD complex.

SEARCH, ASSESSMENT AND DEVELOPMENT OF A BT GENE-BASED STRATEGY FOR RESISTANCE TO DIAPREPES *R. G. Shatters, Jr., S. L. Lapointe, D. G. Hall, and A. A. Weathersbee III. U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

The Diaprepes root weevil (DRW) has been an important agricultural pest of citrus in Florida since its apparent introduction from the Caribbean over 40 years ago. Despite significant research on different control strategies during this 40-year period the weevil has continued to expand its range throughout the citrus production regions of Florida. More recently, it has been detected in Texas and California and threatens continued expansion in these areas as well. Meanwhile, management programs for the weevil remain expensive and less effective than needed. One DRW management approach that deserves investigation in citrus is the use of a genetically engineered rootstock with resistance/deterrence to DRW larval feeding. A number of successes in the use of genetically engineered crops for insect control have been achieved in other crops. There are a number of known that produce proteins active against insects. As part of a research project on a resistant rootstalk, we screened numerous isolates of a bacterium known to produce insect toxins, *Bacillus thuringiensis,* for activity against DRW larvae. This screening process included analysis of new Bt isolates identified from field-collected DRW cadavers and Bt strains in national germplasm collections known to have activity against beetles. Of 19 screened isolates, the one producing the lowest LC_{50} value (50.7 μg/ml) in DRW larvae was used for further study. From this isolate, the gene encoding the major toxin gene was obtained and modified for optimal expression in plants. The modified gene was inserted into the genome of both alfalfa and tobacco plants, both of which are suitable host plants for DRW larvae. These transformed plants are currently being grown for characterization of Bt toxin expression and will be tested against DRW larvae. If plants expressing the Bt toxin show resistance to DRW, experiments will be initiated to develop a citrus rootstock expressing this toxin. Successful incorporation of a DRW resistant rootstock into citrus production groves would provide a low-cost, sustainable management strategy for DRW in citrus.

POTENTIAL CONTROL OF *DIAPREPES ABBREVIATUS* WITH TMOF AND CATHEPSIN L INSECT SPECIFIC PROTEIN INHIBITOR *D. Borovsky[1], C. Powell[2] and R. Shatters, Jr.[3], [1]University of Florida/Institute of Food of Agricultural Sciences, Florida Medical Entomology Laboratory, Vero Beach, Fla. (UF/IFAS/FME); [2] University of Florida/Institute of Food of Agricultural Sciences, Indian River Research and Education Center, Ft. Pierce, Fla. (UF/IFAS/IRREC); [3]U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

Chemical control of the citrus root weevil, *Diaprepes abbreviatus*, is difficult because the larvae are subterranean, feed on citrus plant roots and are difficult to control by conventional insecticides. Thus, development of citrus rootstocks that produce inhibitors against citrus weevil digestive enzymes has been identified as a potential control strategy. Because larval *D. abbreviatus* use serine and cysteine like proteases (trypsin and cathepsin L, respectively) to digest their food, we studied the biosynthesis and the control of these enzymes using a molecular biology approach. TMOF, a mosquito decapeptide-hormone that affects trypsin biosynthesis in mosquitoes and in *D. abbreviatus* was cloned and expressed in alfalfa. Alfalfa can be used as a model plant because it is readily eaten by larval *D. abbreviatus*. Since TMOF affects only serine proteases and larval *D. abbreviatus* synthesize cysteine proteases as well as serine proteases, we searched in adult *D. abbreviatus* cDNA libraries for potential Cathepsin L inhibitors. Two polypeptides that exhibited high homology to the pre-pro region of the Cathepsin L sequence were identified. Full-length cDNAs from larval RNA were isolated, expressed, characterized and the polypeptides were identified as larval *D. abbreviatus* native Cathepsin L inhibitors. One of the inhibitors has a signal peptide suggesting that it is secreted into the gut from the epithelial cells that line the gut lumen and its physiological role is to control cathepsin L inside the larval gut. The second inhibitor lacks a signal peptide, and thus, has a possible role in controlling cathepsin L activity inside the insect's cells. The physiological role(s) and the potential bioengineering of Cathepsin L inhibitor(s) and TMOF into plants in future control of *D. abbreviatus* will be discussed.

PROGRESS IN THE DEVELOPMENT OF NEW ROOTSTOCKS TOLERANT OF THE *DIAPREPES/PHYTOPHTHORA* COMPLEX *J. W. Grosser, J. H. Graham, D. Bright, A. Hoyte, and H. M. Rubio, University of Florida/Institute of Food of Agricultural Sciences, Citrus Research and Education Center, Lake Alfred, Fla. (UF/IFAS/CREC)*

Our primary strategy for dealing with the Diaprepes/Phytophthora problem has been to develop complex rootstock hybrids that have the capacity to tolerate mechanical damage caused by weevil feeding and then recovery by exhibiting vigorous root growth in challenging soils inoculated with both *Phytophthora nicotianae* and *P. palmivora*. We have continued with annual crosses of superior allotetraploid somatic hybrid rootstocks and screening of resulting seed in high pH calcareous 'Winder' soil inoculated with both *Phytophthora spp*. in greenhouse flats. Seed from 2006 crosses was recently planted. Vigorous healthy "tetrazyg" seedlings are selected and propagated by grafting to vigorous rootstocks and subsequently rooted cuttings. Replicated *Diaprepes* force-feeding assays are conducted in conetainers®, and hybrids selected for reduced mechanical damage are replanted in a 'Winder'/*Phytophthora* mix to assess recovery potential. During the past 6 months, 3 large sets of new hybrids from 2004 crosses were screened, and several promising hybrids were identified that show excellent capacity for complete root system recovery in this greenhouse test. These are now being propagated for more extensive field evaluation. Cuttings have been produced from 2005 hybrids for screening during 2007. Data on seed production from the first group of hybrids screened will also be presented (from top-worked seed trees). Information on the first field test of greenhouse selected tetrazygs planted in the Kelly block (via R. Stuart/C.W. McCoy, spring of 2005; scion: Hamlin sweet orange) will also be presented. Citrus rootstock breeding and selection at the tetraploid level maximizes genetic diversity and selection efficiency, and shows great promise for generating new rootstocks that can tolerate the *Diaprepes/Phytophthora* complex.

THE EFFECT OF CLIMATE ON DISTRIBUTION OF DIAPREPES ROOT WEEVIL AND ASSOCIATED EGG PARASITOIDS *S. L. Lapointe[1], D. G. Hall[1] and D. M. Borchert,[2] [1] U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS); [2] USDA-APHIS, Plant Protection and Quarantine/Center for Plant Health Science and Technology/Plant Epidemiology and Risk Analysis Laboratory, Raleigh, NC*

The Diaprepes root weevil (DRW) has been a pest of citrus and ornamental plants since its introduction into Lake County, Fla. in 1964. Since then, DRW has colonized the Florida peninsula to the south of its point of introduction, but has not expanded its range to the north. A lower threshold for oviposition was estimated as 14.9°C. Eggs were highly susceptible to cold with 95% mortality ($LTime_{95}$) occurring in 4.2 d at 12°C. Relative susceptibility of life stages to cold was eggs>pupae>larvae>adults. Archived weather data from Florida was examined to guide a mapping exercise using the lower developmental threshold for larvae (12°C) and the lower threshold for oviposition (15°C) as critical temperatures for mapping DRW distribution and the potential for establishment of egg parasitoids. Probability maps using the last 10 years of weather data examined the frequency of at least 10, 15, 20, 25, or 30 d per winter when soil temperature was ≤12°C. The geographic area that experienced between 15 and 20 d per winter with mean daily soil temperature ≤12°C closely approximated the northern limit of *D. abbreviatus* in Florida. Homologous maps of Arizona, California, and Texas predict the areas where soil temperatures favor DRW establishment. Successful establishment of egg parasitoids in Florida appears to be limited to southern Florida where mean daily air temperatures fall below 15°C fewer than 25 d/yr. By this measure, we predict that egg parasitoids will not establish in Arizona, California or Texas.

UPDATE ON CLASSICAL BIOLOGICAL CONTROL OF THE DIAPREPES ROOT WEEVIL. *J. E. Peña,[1] D. Carrillo,[1] R. Duncan,[1] B. Ulmer,[1] C. McCoy[2] and D. Hall.[3] [1]University of Florida/Institute of Food of Agricultural Sciences, Tropical Research and Education Center, Homestead, Fla. (UF/IFAS/TREC); [2] University of Florida/Institute of Food of Agricultural Sciences, Citrus Research and Education Center, Lake Alfred, Fla. (UF/IFAS/CREC); [3] U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

Two new parasitoid species, *Haeckeliania sperata* (Hymneoptera:Trichogrammatidae) and *Fidiobia dominica* (Hymenoptera: Platygastridae) were released during the summer and early fall of 2006 for control of the Diaprepes root weevil eggs. Because of problems with the release protocol, possible predation of parasitized egg masses and reduction of the number of individuals in the parasitoid colonies, releases have been upheld until 2007. The biology, development of both parasitoids at different temperatures and the effect of host plants on successful parasitism of both species were studied in the laboratory. The upper development threshold (UDT) of *F. dominica* was 30.0ºC, its maximal development rate (MDR) occurred at 27.6ºC, its lower development threshold (LDT) was 9.6ºC and its thermal constant (K) for development from egg to adult of 293.11 DD. For *H. sperata*, UDT was 35.0º C, MDR occurred at 31.0ºC , LDT was 11.3ºC and K was 279.72 DD. Based on these results, both species would be able to complete 17 to 18 generations annually in South Florida. However host availability during critical periods could severely impair the ability of these egg parasitoids to establish and successfully control *D. abbreviatus* in areas where winter temperatures fluctuate around 12ºC, the LDT for this pest. Preliminary results suggest that leaf characteristics (thickness and pubescence) might alter successful egg deposition by both the parasitoids.

IS THERE A ROLE FOR CONSERVATION BIOLOGICAL CONTROL WITH ENTOMOPATHOGENIC NEMATODES? *L. W. Duncan, R. J. Stuart, J. H. Graham, J. Zellers, D. Bright, D. Dunn, and F. E. El-Borai. University of Florida/Institute of Food of Agricultural Sciences, Citrus Research and Education Center, Lake Alfred, Fla. (UF/IFAS/CREC)*

We are testing the hypothesis that endemic entomopathogenic nematodes (EPN) regulate the prevalence of *Diaprepes abbreviatus* in Florida. In surveys, both within groves and across regions, the species diversity and abundance of entomopathogenic nematodes was positively related to the size and amount of sand particles in the soil, whereas the abundance of *D. abbreviatus* was inversely related to sand content and EPN prevalence. We are also investigating the role of nematophagous fungi (NF) in regulating the spatial patterns of EPN. The temporal and spatial patterns of the EPN *Steinernema diaprepesi,* but not *Heterorhabditis zealandica* or *H. indica,* were inversely related to some species of NF in a central ridge citrus grove on deep, coarse sand. Similarly, in an east coast grove on finer texture soil, the prevalence of the sole EPN species *H. indica* was not related to that of any species of NF. Four EPN species isolated from a grove on the central ridge were introduced into a grove on the east coast in which *H. indica* was the only endemic EPN. The 3-year-old trees in the east coast grove had been planted into holes (3 m radius x 3 m deep) filled with coarse sand from the central ridge intended to improve drainage. Five months after the introduction of EPN, mortality of sentinel *D. abbreviatus* larvae was significantly greater in plots treated with *S. diaprepesi* (54% week^{-1}) or *H. indica* (50%) from the ridge than in plots containing only the endemic *H. indica* (26%). During 2 years in a grove of mature trees on the central ridge, the prevalence of NF was reduced and that of EPN was increased by mulching the soil with composted animal manure. Mortality of sentinel *D. abbreviatus* was also higher in the plots amended with manure mulch. These experiments support the feasibility of modifying the soil habitat to conserve the endemic and introduced EPN species that provide biological control of *D. abbreviatus*. In a separate line of research, significantly fewer *D. abbreviatus* and *Artipus floridanus* were recovered from ground traps adjacent to young trees growing in soil covered by woven landscape fabric compared to trees growing in bare soil. After 4 years of growth, the mean cross sectional trunk area of trees growing in fabric-covered soil was 36% greater ($P < 0.02$) than that of trees growing in bare soil. The trees are growing on the central ridge at a site with low prevalence of *D. abbreviatus* and the effects of herbivory and other factors such as soil temperature on tree growth are unknown. A second trial at a flatwoods site with high prevalence of *D. abbreviatus* is ongoing.

DIAPREPES ROOT WEEVIL ON ORNAMENTALS – AN UPDATE FROM SOUTHERN FLORIDA; *Catharine Mannion and H. Glenn, University of Florida/Institute of Food of Agricultural Sciences, Tropical Research and Education Center, Homestead, Fla. (UF/IFAS/TREC)*

The Diaprepes root weevil, *Diaprepes abbreviatus* (L.) (Coleoptera: Curculionidae) is a serious pest problem facing citrus growers in Florida and is also a severe problem for ornamental plant growers. There are numerous concerns about this pest which include detection and monitoring of soil-inhabiting stages, the destructive habits of larvae and adults, and the impact of damage on the ornamental plants, but one of the most important aspects of management in ornamental production is control of the immature stages in order to reduce the risk of spreading the pest with infested plant material.

Currently, infestations of Diaprepes root weevil have been found in three counties in southern California. Research, extension and regulatory officials are working towards potential eradication and also pursuing regulatory treatments that may be necessary for their ornamental growers. One area of interest includes the combination of soil insecticides and entomopathogenic nematodes. A review of soil-applied insecticides alone an in combination with entomopathogenic nematodes will be reviewed with an emphasis on tests conducted at the Tropical Research and Education Center. Previous research has demonstrated that bifenthrin (Talstar) is efficacious against neonates and young larvae and that some entomopathogenic nematodes are efficacious against various stages of larvae. Bifenthrin is currently recommended as a drench or incorporated into the potting media at a rate of 25 ppm based on the bulk density of the media. Tests were conducted to evaluate bifenthrin and entomopathogenic nematodes, alone and in combination, for control of older larvae (> fifth instar) in container ornamentals. In all cases, the combination treatment of bifenthrin and the entomopathogenic nematodes provided the best control suggesting a synergy or additive effect between treatments. The results of a field test did not mirror the results from the tests with containerized plants, however, there was a trend of increased control in the combination treatments. Additional testing is ongoing to evaluate additional soil insecticides in combination with nematodes.

Another area of concern is the impact of Diaprepes root weevil on select ornamental plants. Data will be reported from studies that have been conducted to examine the effects of root feeding of Diaprepes root weevil and flooding and the interaction of these two stresses on leaf gas exchange and growth. Some of the plants that have been tested include buttonwood (*Conocarpus erectus* L.), mahogany (*Swietenia mahogani* Jacq.), surinam cherry (*Eugenia uniflora* L.), and pond apple (*Annona glabra* L).

MANAGING *DIAPREPES* THROUGH ROOTSTOCK TOLERANCE AND STRATEGIC PESTICIDE APPLICATIONS: THE LONG-TERM FIELD STUDY AT *SOUTHPORT*

R. J. Stuart, W. S. Castle, J. H. Graham, M. E. Rogers and C. W. McCoy; University of Florida/Institute of Food of Agricultural Sciences, Citrus Research and Education Center, Lake Alfred, Fla. (UF/IFAS/CREC)

In a long-term field study near Southport, Osceola County, we are comparing the growth and survival of 'Hamlin' orange trees on five rootstocks: C22, C32, and C35 citranges, Cleopatra mandarin and Swingle citrumelo. The trees were planted in September 2001 within a declining mature citrus grove heavily infested with the root weevil, *Diaprepes abbreviatus*. We used a split plot experimental design and all young trees received chemical applications for pest control during the first 20 months of growth. Subsequently, half the trees received no pesticide treatments whereas the others received treatments based on weevil abundance and focusing primarily on the weevil's spring emergence peak. To date, treated trees have had higher growth rates (except Cleopatra mandarin), larger tree canopies, less tree decline, and higher yields than untreated trees; and trees on C32, C35, and Swingle citrumelo have generally outperformed those on C22 and Cleopatra mandarin. Roots damaged by weevil larvae are often infected by the plant pathogen *Phytophthora nicotianae*, a major factor in tree decline. Hurricanes in 2004 killed 18.6% of the young trees and caused major damage to an additional 27.2%. Differential storm damage among treatments disrupted the field study with larger healthier trees suffering more broken branches and fatal trunk injury whereas smaller weaker trees had less damage overall but were more frequently uprooted. Nonetheless, differences in canopy volumes among treatments showed similar patterns before and after the storms. This study demonstrates the importance of rootstock selection and effective pesticide use for maintaining citrus groves under stress from *Diaprepes* and *Phytophthora*. The Southport grove is also a venue for research on additional new rootstocks, weevil phenology, movement, and longevity, and the role of ants and entomopathogenic nematodes in the biological control of *Diaprepes*.

CHARACTERIZATION AND EVOLUTION OF DELTA 9 DESATURASE FROM *DIAPREPES ABBREVIATUS* (L.): THE FIRST REPORT OF A DESATURASE GENE FROM THE COLEOPTERA. *Laura M. Boykin, Catherine Sue Katsar, Stephen Lapointe, and Wayne Hunter; U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

A delta 9 desaturase gene was identified from an EST library constructed from teneral female *Diaprepes abbreviatus*. Desaturases are involved in the production of essential fatty acids. In insect orders such as Lepidoptera, they are commonly associated with pheromone production. In order to characterize the DRW delta 9 desaturase-like gene we conducted sequence comparisons, which included a Bayesian phylogenetic approach. Phylogenetic relationships of 50 delta 9 desaturase genes from various insects will be discussed.

REGULATING THE GROWTH AND DEVELOPMENT OF *DIAPREPES ABBREVIATUS* ON ARTIFICIAL DIETS *Randall P. Niedz, T. J. Evens, and Stephen L. Lapointe; U. S. Department of Agriculture/Agricultural Research Service, Ft. Pierce, Fla. (USDA/ARS)*

Mass rearing of Diaprepes root weevil (DRW) provides all life stages for basic and applied research into plant-insect interactions, biological control, insect physiology, novel toxin discovery (plant, fungal, bacterial), behavior, and pheromone discovery and elucidation. The standard artificial diet was modified from the boll weevil diet and is sufficient for the insect to complete development from neonate to adult. However, the individual and interactive effects of diet components on growth and development are unknown. We present preliminary data from experiments designed to quantify the main and interaction effects of nine standard diet components on larval survival and growth, time until adult emergence, sex ratio, and adult weight. Data analysis identified components that significantly affected each measured response. Quantifying component effects is useful in three aspects: 1) the significant effects can be used to predict and generate specific responses and thus provide a sufficiently characterized biological system to identify key modulators of growth and development; 2) new diet formulations that improve mass rearing efficiency are a natural byproduct of such quantification and; 3) relationships between multiple responses can be established thus providing a higher order understanding of the complete life cycle of DRW compared to single component/factor approaches.