# Abstract

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# **Editors' Comments**

Henneberry, T. J., R. M. Faust, W. A. Jones and T. M. Perring (eds.) 2002 Sweetpotato (Strain B, Silverleaf) Whitefly Five-Year National Research and Action Plans; Fourth Annual Review of the Second Plan and Final Report for 1992 to 2002. The meeting was held in San Diego, CA, February 10-12, 2002.

This publication contains research, extension-education, industry and action agency reports of progress contributing to our knowledge of the whitefly complex and to the development of ecologically acceptable whitefly management systems. The multi-agency cooperative effort has, since 1992, provided a forum for information exchange, complementary, coordinated and cooperative research programs, avoidance of duplication of effort, and optimum return for expended research dollars. The result of the joint partnerships has been solutions and technology transfer to the stakeholders in the agricultural communities. These accomplishments have been achieved within a rapid timeframe, in large part, due to openness of communications, sharing of expertise and focus on common goals. The editors sincerely thank all those who participated in the annual reviews of the research and action plans. This year's publication supplement contains a final report for Whitefly National Research and Action Plans from 1992 to 2002. Although this year's activities terminate the formal functioning of the plan, communication guidelines and identification of other whitefly research and information exchange coordinating groups worldwide are provided in Appendix A.

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# Preface

The Sweetpotato Whitefly, Bemisia tabaci (Gennadius) Strain B (= Silverleaf Whitefly, *Bemisia argentifolii* Bellows and Perring), National Research and Action Plans (1992-1997 and 1997-2002) were developed by USDA agencies (ARS, APHIS, and CSREES), state agencies, state agricultural experiment al stations, and representatives of the cotton, vegetable, ornamental, nursery crop and chemical industries. The objectives were to establish research priorities, avoid duplication of effort, and maximize the use of existing resources. Research needs, goals and objectives, and technology transfer to clientele (scientific community, legislators, regulators, the agricultural industry, and the public) were reviewed on an annual basis. The plan was flexible allowing responsiveness to changing needs and priorities with appropriate adjustments to terminate, redirect, or add priorities based on funding, current knowledge, and program needs. The results were the development of environmentally and socially acceptable areawide, community-based whitefly management methods.

The USDA Sweetpotato Whitefly Research, Education, and Implementation Coordinating Group, throughout the duration of the work, facilitated USDA interagency and partner state agricultural experiment stations activities. The Working Group was composed of members of participating agencies and met annually to maintain communication with the USDA Coordinating Group and the Sweetpotato Strain B (Silverleaf) Whitefly Program Planning and Review Committee.

Intensive studies outlined in the research and action plans result in development of short-term relief from the devastating impact of explosive *Bemisia* populations using chemical methods. Selected insecticides applied alone or in mixtures based on developed action thresholds and Insecticide Resistance Management (IRM) methods have been highly effective on a short-term basis, but continuing long-term insecticide based management efficacy cannot be relied on. The research conducted within the guidelines of the plan has also resulted in a large base of information on Bemisia biology, ecology, behavior, physiology, systematics and other information that has improved our understanding of the essentials necessary to develop long-term strategies for SPW management. Thus, building blocks are provided for continuing efforts to develop areawide, community-based Bemisia management systems that incorporate cultural, biological, and nonchemical methods into chemical control-IRM-based control methods.

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### **Executive Summary**

The Whitefly National Research and Action Plan Reviews from 1993 to 2002 provided annual updates of the federal, state, and industry cooperative efforts to develop efficient management of methods for *Bemisia* populations. The work was initiated in 1991 when federal and state agency, agricultural experiment stations, industry and commodity group representatives, identified priority research areas for development into a national research and action plan for control of sweetpotato whiteflies. Unacceptable losses in cotton and vegetable field crop production were being experienced in California, Arizona, Texas and Florida, as well as ornamental and vegetable losses in glasshouse production.

During the years 1992 to 1997, extensive research achievements were made that provided interim solutions and a better understanding of the whitefly problem. Some of the research accomplished as part of the plan has been implemented by growers in their management efforts. A complete published review of the program showed extensive progress in all priority research areas. Over 70 examples of technology transfer to growers and the scientific community were documented.

The second 5-year plan, "The Silverleaf Whitefly, (*Bemisia argentifolii* Bellows and Perring) Research, Action and Technology Transfer Plan" was finalized at the annual review meeting at San Diego, CA on January 28-30, 1997.

The SPW has been referred to in the literature as the cassava whitefly, tobacco whitefly, poinsettia whitefly and cotton whitefly, as well as *B. tabaci* and a number of other scientific names (synonyms). The taxonomic relationships and the synonymy of the species have been reviewed on several occasions and remain unresolved. Race and/or biotype designations have also often been recorded in past literature in relation to host affinities and virus transmission vector interactions. The possibility of biotype occurrence has received continuing interest with the increasing importance of the sweetpotato whitefly in crop production on a worldwide basis. The detection of differences in electrophoretic isozyme patterns, biology and extended host range have provided evidence for the existence of different biotypes as compared to the type

previously encountered in the desert Southwestern United States crop production areas. This biotype has been referred to in the literature as the sweetpotato whitefly strain B. Based on the lack of nomenclatural resolution, *B. tabaci* strain B and *B. argentifolii* names used interchangeably in these reports are considered the same species.

### Introduction

The SPW National Research and Action Plans were developed to serve as guidelines for multi-agency, federal, state, private industry and consumer programs to reduce excessive SPW-induced losses occurring in the cotton, vegetable, nursery and glasshouse crop production industries in the United States. The extremely diverse SPW host range that included cotton, field, vegetable, ornamental and nursery crops in commercial or home garden and recreational and landscape settings resulted in implementation of the accomplishments and technology transfer not only to commercial production, but an extension of the benefits to private homeowners, urban communities and the non-agriculturally involved public sector in general.

Coordinated, multi-partner projects have a number of advantages: 1) adoption of standardized data collection protocols that are appropriate for comparison over diverse areas; 2) periodic reviews to exchange information and reprioritize research areas; 3) increased efficiency by sharing knowledge, resources and expertise; 4) multiple commodity input; and 5) increased returns per invested research dollar as opposed to multiple efforts in an uncoordinated manner.

The development of the action plans stimulated by the economic, environmental and social impact of the SPW species complex worldwide resulted in development of extensive information on SPW biology, ecology, physiology, viruses, virus-vector interactions and systematics. Implementation of this information has resulted in highly acceptable management systems with improved cost-benefit ratios. An important contributing factor to the success of the plans has been participation of all members of agricultural communities, growers, industry, scientists, bankers, and others sharing and implementing information provided by researchers. extension, and educational systems to develop large-scale multidisciplinary approaches to SPW management. Additionally, the success of the plans benefited, far beyond our ability to measure, from the experiences and research inputs from scientists of more than 14 foreign countries that participated in cooperative research and the annual plan reviews.

In this final review, we recap the research highlights and accomplishments. It would be impossible to capture the specifics of every item of investigation since over 1,000 reports of progress were published during the reviews (Table 1), but all of the work and participants contributed to the success of the plans.

# I. Sweetpotato Whitefly Strain B (SPW): Accomplishments of the National Research and Action Plans 1993 to 2002 and Future Goals.

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# Early History of the Sweetpotato Whitefly in the United States

*Bemisia tabaci* (Gennadius) was first reported in the United States in Florida in 1894 (Russell 1957, Russell 1975). It was subsequently collected on various crop and weed hosts in Washington D.C., Arizona, Southern California, Texas and Georgia. Little information on the biology of the SPW was reported that could serve to compare the cause of outbreaks occurring in the mid 1950s and in the late 1970s with those that occurred in late the 1980s and early 1990s.

Before the 1980s, SPW was primary a minor pest, except that it was known to vector certain plant diseases. Cotton Leaf Crumple (CLC) disease was observed in California in 1948 (Dickson et al. 1954). CLC was subsequently determined to be transmitted exclusively by SPW (Laird and Dickson 1959). Substantial cotton losses occurred in California and Arizona (Erwin and Meyer 1961, Allen et al. 1960, van Schaik et al. 1962) establishing SPW as an economic pest in the United States. Stub (perennial) cotton was suspected as the virus carryover source and when culture of perennial cotton was eliminated CLC incidence declined (van Schaik et al. 1962).

SPW during the 1960s seldom reached economic levels in Southern California cotton except when insecticides were used excessively (Gerling 1967). In the Imperial Valley from 1964 through 1981, no SPW were collected in 1964, 1965 and 1966; only low numbers occurred in 1967; and none were collected from 1968 to 1974 (Johnson et al. 1982). Numbers began to increase in the Imperial Valley in 1975, were variable through 1980, and increased dramatically in 1981 (Johnson et al. 1982). In 1981, SPW was reported as a rapidly escalating cotton problem in Arizona and Southern California (Butler and Henneberry 1984, 1985).

Subsequently, several virus-like diseases were found affecting a number of important commercial crops, and the diseases apparently were coincident with the high SPW populations (Duffus and Flock 1982, Duffus et al. 1986). Losses in 1981 were reported to exceed 100 million dollars (Duffus and Flock 1982). The SPW remained a problem of varying intensity in Arizona and California through the mid and late 1980s (Butler and Henneberry 1984, 1985, Butler et al. 1986). A major epidemic of CLC occurred in Arizona in 1981 (Brown and Nelson 1984) and was a problem annually thereafter through 1985 (Brown and Nelson 1986).

In 1986, SPW outbreaks in Florida resulted in losses in the tomato industry alone that were estimated to be over 140 million dollars (Costa et al. 1993). In 1991 losses for cotton and vegetables in Texas were estimated to be 24.0 and 29.0 millions of dollars, respectively (Riley and Sparks 1993). In southern California, crop losses of over 100 million dollars a year and a reduction in 3,000 agricultural jobs annually have been reported since 1992 (Henneberry et al. 1993, 1994, 1995, 1996, 1997, 1998, 1999, 2000). SPW losses for ornamentals in 1991 were reported to exceed 23 million dollars (Barr and Drees 1992). SPW has been considered a major pest in most greenhouse cultures in the Southeastern United States (Oetting and Buntin 1996), but reliable loss figures are not available. In addition several previously undescribed diseases were observed, including irregular ripening in tomato, silverleaf of curcurbits, and blanching of brocolli, that were coincident with infestations of SPW; while whitefly populations and virus diseases declined in lettuce (Bharathan et al. 1990, Byrne et al. 1990, Yokomi et al. 1990).

The serious nature of the escalating SPW problem in many areas of the United States and the economic impact on the agricultural community highlighted the need for a coordinated national effort to provide short and long-term solutions to the problem. During the same period it was noted that certain populations of *B. tabaci* differed in reproductive potential. Strains with the lower and higher fecundity were termed A and B, respectively (Bethke and Paine 1991).

# Sweetpotato Whitefly: 5-Year Plans for Development of Management and Control Methodology

# The First Five-Year Plan 1992-1996

By the end of the 1991 growing season, it was apparent that unacceptable losses in the field grown cotton and vegetable production were being experienced in California, Arizona, Texas, and Florida, as well as losses to ornamental and vegetable crops in glasshouse cultures at various locations in the South and Northeastern United States. The need for immediate management recommendations was urgent. Working from a draft plan (Anonymous 1992), representatives from federal and state agencies, agricultural experiment stations, industry and commodity groups identified six priority areas for development into a SPW national research and action plan (USDA 1992). They also identified the need for an immediate plan of research action for the 1992 growing season. Standardized experimental procedures, data collection protocols, report preparation formats, and a system for exchanging results were established for a national testing program of promising chemicals, natural products, microbial insecticides and improved application technology for SPW control on all major crops (Akey

1992). Additionally, the framework for an insecticideresistance management program (IRM) was established. Scientists in California, Texas, and Florida initiated laboratory studies monitoring insecticide responses of field populations to develop baseline information as a cornerstone for long-term SPW-IRM. Over 30 federal and state experiment station scientists were involved in the overall program in eight states. The results of the coordinated, national chemical control efficacy trials, with annual modification, improvement, and testing of additional materials have provided the basis for highly effective SPW chemical control and ongoing SPW-IRM.

The plan was designated "The Five-Year National Research and Action Plan for Development of Management and Control Methodology for the Sweetpotato Whitefly" (USDA 1992). The high priority research areas identified were: (1) Ecology, Population Dynamics, and Dispersal; (2) Fundamental Research -Behavior, Biochemistry, Biotypes, Morphology, Physiology, Systematics, Virus Diseases, and Virus Vector Interactions; (3) Chemical Control, Biorationals, and Pesticide Application Technology; (4) Biological Control; (5) Crop Management Systems and Host Plant Resistance; and (6) Integrated Techniques, Approaches, and Philosophies.

Annual workshops were held to review progress and to adjust the plan as needed (Henneberry et al. 1993, 1994, 1995, 1996, 1997, 1998, 1999, 2000). After the annual review meeting in Orlando, FL in 1994 the titles of the reports of progress for the 5-year plans were changed to recognize the revised description the SPW strain B as a new species, *B. argentifolii* Bellows and Perring (Bellow et al. 1994). Because of the continuing nomenclatural controversy, the reluctance of international researchers to accept the new disintegration, and to facilitate continuity in the literature, in this publication, we consider SPW Strain B and *B. argentifolii* Bellows and Perring as synonymous

During the years 1992 to 1997, many research achievements were made that provided interim solutions and a better understanding of the SPW problem. Some of the research results that have been accomplished through coordination with the plan have been implemented as practices by growers. A complete published review of the program showed extensive progress in all priority research areas (Henneberry et al. 1997). Over 70 examples of technology transfer to growers and the scientific community were documented. A complete management system for SPW is a primary goal for the future. The decision to develop a second 5-year plan was adopted when 85% of the individuals attending the 1996 annual review responded to a question and indicated that a sequel to the existing plan should be prepared to maintain continuity, and a continuing high level of coordinated research and technology transfer.

The Second Five-Year Plan 1997-2001 The second 5-year plan, "The Silverleaf Whitefly, *(Bemisia argentifolii* Bellows and Perring) Research, Action, and Technology Transfer Plan" (Henneberry et al. 1997, 1998, 1999, 2000), was finalized at the annual review meeting at San Diego, CA on January 28-30, 1997. The high priority research areas are: (A) biology, ecology, and population dynamics; (B) viruses, epidemiology, and virus-vector interactions; (C) chemical control, biopesticides, resistance management, and application methods; (D) natural enemy ecology and biological control; (E) host plant resistance, physiological disorders, and host-plant interactions; and (F) integrated and areawide pest management approaches, and crop management systems.

Over 1,000 abstracts of research progress were presented at reviews conducted from 1993 to 2002. Tables 2 and 3 are summaries of the research highlights for the research priority approaches for the 1992 to 1997 and the 1997 to 2002 national plans, respectively. Research credits can be identified in the original abstract sections for each for each year.

For the long-term, the extensive base of information on biology, ecology, physiology, systematics, virus, virusvector interactions and other fundamental information (Tables 2 and 3) developed and reported by the two fiveyears plans will provide the basis for new technically driven, ecologically oriented and economically acceptable SPW management programs. The practical applications and incorporation of much of this information is directly or indirectly included in the following documented management approaches. Much additional implementation can be accomplished and will improve the efficacy, the environmental compatibility, and social acceptance of insect management.

# Research Based Accomplishments in Sweetpotato Whitefly Management

*Chemical control, action thresholds and resistance* management. In the low desert agricultural areas of the Southwest U.S. the SPW was a year-round pest, successively infesting green-leafy winter vegetables, spring melons, and cotton. Outbreaks in cotton were directly related to rapid population increases in spring melons, and availability of the winter vegetable crops to serve as a haven for the pest during the winter season. Epidemic SPW populations in California and Arizona that characterized the late 1980s through the mid -1990s have not occurred in the growing seasons of 1996 to 2001 as a result of implementing programs based on our increased knowledge of SPW and consequently our ability to develop improved control methods. The early search for stop-gap control methods identified certain chlorinated hydrocarbon, (e.g. endosulfan), organophosphate (e.g. Lorsban and others), pyrethroid (e.g. Capture, Danitol, Asana) and pyrethroid-organophosphate combinations

(e.g. Danitol-Orthene) that were effective. In 1993 imidicloprid, a systemic neonicotinoid insecticide became available for management of SPW in winter vegetables, and in 1994 for melons, as well. Beginning in 1993, programs of conventional insecticides were used to manage SPW, until in 1995, resistance to certain key insecticides, e.g. the pyrethroid plus organophosphate combinations used for whitefly management in cotton occurred (Dennehy et al. 1996a and b, Naranjo et al. 1998).

New systems for SPW were urgently needed for the 1996 season. Adult and immature SPW sampling methods and action thresholds were developed and validated for cotton and melons in the field, and for greenhouse crops. The action thresholds are an integral part of the overall management program and were particularly helpful in reducing superfluous insecticide use, reducing costs, and delaying insecticide resistance development (Naranjo and Flint, 1994, 1995, Naranjo et al. 1995, 1996, Tonhasca et al. 1994a and b). In Arizona, California and Texas cotton, the action thresholds were used with two previously unavailable insect growth regulators (buprofezin and pyriproxyfen). In combination with improvements in cultural control, a program of integrated use of insect growth regulators and insecticides in rotation has effectively managed SPW populations since 1996 (Ellsworth 1999). Because the propensity of SPW to rapidly develop resistance to control measures was recognized, the two newly-available insect growth regulators were introduced in the same year, and the use of each was restricted to one application per year to conserve their efficacy.

Chemical control efficacy has been protected by the implementation of integrated resistance management (IRM) programs (Prabhaker et al. 1996, Castle et al. 1996b, Dennehy et al. 1996). Yellow sticky card, vial and leaf dip techniques have been used in resistance monitoring (Dennehy et al. 1996, Prabhaker et al. 1996). Recommendations for SPW management in Arizona, and more generally in the Southwest, have been reviewed by entomologists from USDA, the cooperating universities, and representatives of the cotton and agrichemical industries at annual meetings since 1996 (Dennehy, unpublished, and Nichols, unpublished, respectively). SPW susceptibility levels to the various modes of action that have been measured differed among geographic locations and in some instances between years at the same location. Some level of tolerance to organophosphates, pyrethroid, carbamate, and chlorinated hydrocarbon insecticides may occur at various locations within the United States (Dennehy and Williams, 1997). It has been rare, however, to attribute control failures to insecticide resistance (Li et al. 2001).

The advances in improved SPW control were not attained without increasing control costs and agricultural community involvement (Henneberry et al. 2000). The insect growth regulator treatments are costly (about 35 dollars per acre). However, they have been cost effective for cotton with resulting increased yield and quality, and they have replaced far greater numbers of insecticide treatment that were used, at times, ineffectively before their introduction. In Imperial Valley, California, cotton production was reduced from 15,000 acres in 1990-1991 to 4 to 5,000 acres in the early 1990s and has increased to about 8,000 acres in 1998 and 1999 because of the new technology. Similarly, imidicloprid has consistently provided excellent SPW control on melons and vegetables, but costs are \$65 dollars per acre. In California, Arizona and Texas almost all melon acres, of necessity, are treated with imidicloprid to protect them from SPW infestations. Recently buprofezin has been registered for SPW control on melons, thus cross commodity cooperation has been developed to avoid SPW exposure to excessive buprofezin selection pressure in the overlapping melon and cotton growing seasons. Costs associated with administrative coordination, resistance monitoring, and development and dissemination of information on resistance management programs also can be extensive.

In the Imperial Valley, as a result of whitefly populations, spring melon acres were severely reduced in the early 1990s, and continue to be below the average of 25 thousand acres that were grown in the pre-1990s. Melon acres in the Imperial Valley were about 14 thousand acres in 1999. Similarly, fall melons were grown on 10 thousand acres in 1990 and on only 2 thousand acres in 1998-1999. Indirect economic effects of SPW infestations have also occurred through reduced acres in other crops that may not be as obvious. Although new insecticidal chemistries have played a major role in providing SPW control in many of our major cropping systems, high costs may limit use of effective insecticides under some circumstances. For example, because of high prices of certain insecticides, growers of some specialty crops may not be able to take advantage of the new technologies. In more extreme cases, certain crops grown only on a relatively few acres may not have insecticides available to them because the expense of insecticide registration outweighs the anticipated returns. Often in these cases the crop is simply no longer grown commercially in the area where SPW is a problem. Extensive documentation of such cases does not exist, but termination of fall squash production in the Imperial Valley as related to SPW-transmitted squash leaf curl virus was a case in point (Agricultural Commissioner's Office, Imperial Valley, CA, Personal Communication).

In addition, to the development of new insecticide-based management systems, a number of biorational materials that include oils, soaps, and plant products have been shown effective for some crops and may be useful in insecticide rotations to avoid resistance (Stansly et al. 1995).

### **Goals for Integrated Areawide Management**

As described above, chemical control strategies, use of action thresholds, IRM programs, natural enemy conservation, with consideration for spatial and temporal crop sequencing, early harvest, and crop residue destruction have been highly effective within individual crops, with some benefit to adjacent and sequential crops due to SPW population reduction. Efforts are being made, and further efforts are needed, to expand SPW management systems to incorporate cultural, and biological methods into existing chemically based programs that in the future should include areawide, community efforts.

Cultural control. The crops grown, their temporal and spatial relationships, and their respective production inputs and practices are important considerations in SPW population dynamics. Extensive numbers of weed, ornamental, nursery stock, and cultivated crop-host-plants provide nutritional and reproductive host continuity to support SPW population growth. Inter- and intra-host SPW movement occurs during crop growing seasons (Blackmer and Byrne 1995, Byrne and Blackmer 1996). At present strategies for utilizing SPW dispersal in management are limited to grower's awareness of the importance of crop sequencing, planting times and wind direction on SPW dispersal. The proximity of other host crops, when establishing new plantings, is being considered more frequently than in the past because of increased awareness of host-pest relationships and movement. For example, spring melon to summer cotton to fall melon to winter cole crop planting cycles were modified in some areas of the West to eliminate fall melon plantings in order to break the SPW host cycle. An alternative may be to grow fall melons in areas that are distant from existing late-season cotton. Important in respect to crop sequencing is the strict adherence to early harvests of all host crops and destruction and plowdown of crop residues as soon as possible. In areas, where applicable, growers and urban community residents have participated in destruction of weed hosts to reduce another source of migrating SPW. Although, these efforts have not been quantified in relation to effects on SPW populations, they represent good farm practice, and growers have been encouraged to implement them.

Avoidance - Water management and exclusion techniques. Higher SPW populations occur in waterstressed compared to well-watered cotton (Flint et al. 1996). Irrigation schedules that effectively avoid waterstress have been recommended and are largely implemented in Arizona cotton. Also, overhead sprinkler irrigation in melon plantings appears to adversely affect SPW population development. However, overhead irrigation is not, at present, used commercially (Castle et al. 1996a). Row covers to protect plants from SPW, and other insect exclusion and reflective materials for repelling SPW have been adopted and are partially effective in some cropping systems (Chu and Henneberry 1994).

*Natural enemies*. Biological control is expected to play important roles in long-term management of SPW populations. High levels of indigenous natural enemy activity have been identified in cotton, vegetable, and peanut ecosystems, suggesting that natural enemy augmentation and conservation approaches may be important avenues for exploitation in SPW management in the future (Hoelmer 1995, Nordlund and Legaspi 1996). Improvements in conservation have been achieved through use of selective chemistries in cotton. (Naranjo 2001). Part of the efficacy of insect growth regulator use in cotton is attributable to conservation of SPW predators (Ellsworth and Martinez-Carrillo 2001). Foreign explorations have resulted in collection of numerous exotic parasite and predator species that are being considered as potential biocontrol agents (Kirk et al. 2001). Release of some exotic parasitoid and predator species have been accomplished with variable results (Hoelmer 1995). Monitoring is continuing to identify and verify establishment and impact on SPW populations. For predators, a monoclonal antibody has been developed to quantify the role of predation on SPW population regulation as well as to determine candidate predator potential (Hagler and Naranjo 1994). Initial results show positive predation for several indigenous predator species. Natural enemy efficacy, ecosystem compatibility, habitat adaptability, and other factors are being studied to assure optimum utilization of indigenous and introduced parasites and predators. Progress in developing the potential of microbial control is also progressing with the identification of numerous indigenous and exotic pathogenic fungi attacking SLW (Lacey et al. 1996). Strains of Paecilomyces fumosoroseus and Beauveria *bassiana* have been identified as promising biological control agents (Akey and Henneberry 1994).

*Host Plant Resistance Factors.* Plant resistance to several SPW-vectored-virus diseases has been identified with resistance to tomato mottle (TMV) a prominent example. Germplasm resistance to SPW has been identified in alfalfa, peanut, melon, cotton, broccoli, collard and tomato. The hirsute leaf character has been recognized to support higher whitefly populations than smooth-leaf types (Mound 1965) and verified by many scientists. Host plant preferences for melons, cotton, broccoli and lettuce appear related to the amount of vascular tissue per unit of leaf area and the proximity of vascular bundles to leaf surfaces (Chu et al. 1995). Also, leaf surface morphology has been shown to play an important role in SPW nymph establishment (Cohen et al. 1996). Studies on plant-insect interactions, physiological disorders and their mechanisms of resistance and the modes of action of whitefly vectored and of disease resistance provide leads for scientists to identify resistant germplasm for incorporation into agronomic types.

### Viruses and Virus Vectors

New virus-induced plant diseases transmitted by Bemisia, have been observed to be on the increase. In Southern California a new SPW-transmitted geminivirus affecting cucurbits was found in 1998 and also recently detected in Arizona (E. Natwick, Univ. CA Farm Advisor, Holtville, CA; Personal Communication). The long-term impact is unknown, but past experience suggests that the situation must be monitored closely. SPW populations in Texas have also declined from epidemic levels that were experienced in the early to mid 1990s. However, cantaloupe and honeydew melons in the lower Rio Grande Valley have been identified as having SPWtransmitted Cucurbit Yellow Stunting Disorder Virus (CYSDV) disease (Liu et al. 2000). The occurrence of the closterovirus is of much concern to melon growers and its spread, persistence and economic impact will be followed closely. Economic losses have already been experienced. In Florida, the SPW virus vector activity is also being experienced (Polston 1999). Symptoms characteristic of Tomato Yellow Leaf Curl Virus (TYLCV) were first observed on a few tomato plants in the field and nurseries in 1997. The virus probably entered the U.S. in 1996 or 1997 and was rapidly distributed via retail garden centers around the state. Regulatory procedures, that are costly, as well as field management practices have been implemented to minimize the movement of the virus. Even so, TYLCV has spread throughout the state since its first identification. Yield losses have been experienced as well as increased production costs. Pesticide applications to minimize SPW populations have increased. New regulations have been imposed on transplant producers of known TYLCV host plants, which include lisianthus (Eustoma grandiflorum), tobacco (Nicotiana tabacum) and tomato, to minimize the occurrence of TYLCV in certified transplants. TYLCV has been demonstrated to have a devastating effect on tomato yield particularly when plants are infected in early stages of development. The long-term impact on tomato production in Florida remains unknown.

Extension and education. The role of information delivery systems, training and technology transfer cannot be overestimated in implementation of SPW management programs and their success (Kopp et al. 1995). Integration of risk assessment information, spatial analysis, geographic information systems, communications networking, ecological modeling, and extension programs are continually improving and could provide timely information to producers for implementation of improved practices with increasing efficiency. An invaluable service to the plans was the development of an extensive *Bemisia* spp. bibliography first published in the 1995 Supplement to the 5-Year National Research and Action Plan (Butler et al. 1995). The bibliography was updated in supplements thereafter (Naranjo et al. 1996, 1997, 1998, 1999, 2000, and 2002 [this issue]).

*Summary*. The goal of integrating chemical control with resistance management, crop sequencing and host-free periods, crop residue and weed destruction, SPW population and plant disease monitoring, and other cultural controls and management options is considered a high priority for future research. Additional tools such as descriptive models and geographical information systems are envisioned as components of large areawide programs. In most areas with SPW problems, steps are being taken to organize these approaches into coordinated community-action programs. Such programs have proven effective where they have been developed (Ellsworth 1998, 1999, Ellsworth and Naranio 1999, Ellsworth et al. 1996). SPW and disease resistant crop plants, natural products and microbial insecticides, natural enemy introduction and augmentation have potential, and in some instances have been incorporated into SPW management systems when they become available.

Currently, effective SPW management has been accomplished with (1) selection of non-SPW preferred cultivars, (2) selected spatial and temporal modification of sequential crop systems, (3) intensive sampling and monitoring of whitefly populations, (4) chemical control focused on natural enemy conservation, established action thresholds, alternating chemical modes of action, and resistance monitoring, (5) optimum crop yield goals, allowing for early harvests and destruction of crop residues, and (6) active education and extension outreach to provide timely communication of new developments, SPW population dynamics, and other pertinent information to growers. Not all of the management components are applicable or used in all areas or for all crops, but are general principles that provide the agricultural community options for consideration in SPW management. Additionally, the systems remain openended and receptive to other compatible SPW management components.

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		Re	search Priorit				
Agency <sup>c</sup> /State	А	В	С	D	E	F	Total
National Research and	Action Plan for	the Develo	pment of Ma	nagement an	d Control M	ethodology f	or the
Sweetpotato Whitefly,	1993-1997 <sup>a</sup>						
		1993 1	Review, Tem	pe, AZ			
APHIS	-	-	1	1	-	1	3
ARS	7	11	16	13	6	-	53
AZ	2	4	3	1	-	2	12
CA	4	3	4	2	3	-	16
FL	2	3	2	2	2	-	11
GA	-	-	5	-	2	-	7
NY	1	-	1	1	-	-	3
OH	-	-	1	1	-	-	2
TX	1	1	2	-	2	3	9
OTHERS	-	-	1	-	1	-	2
TOTAL	17	22	36	21	16	6	118
		1994 F	Review, Orla	ndo, FL			
ADA	-	-	1	-	-	-	1
APHIS	-	-	-	3	-	-	3
ARS	7	14	13	10	5	1	50
AZ	7	4	5	4	2	3	25
CA	4	5	13	6	3	1	32
CDFA	-	-	-	2	-	-	2
FL	-	3	5	3	2	2	15
GA	-	-	1	-	_	_	1
HI	1	1	_	-	-	_	2
SC	-	1	_	_	-	_	1
TX	1	-	1	2	1	_	5
WI	-	2	-	-	-	_	2
OTHERS	1	-	4	2	_	-	- 7
TOTAL	21	30	43	32	13	7	146
TOTAL	21				15	,	140
		1995 Re	eview, San D	iego, CA			
APHIS	1	1	-	6	-	-	8
ARS	4	4	19	12	4	-	43
AZ, ASU	8	5	5	1	-	2	21
CA	3	7	7	7	2	4	30
CDFA	-	-	-	2	-	-	2
FL	1	2	1	4	1	-	9
GA	-	-	1	-	-	-	1
HI	-	1	-	1	-	-	2
KY	-	-	-	-	1	-	1
NM	1	-	-	-	-	-	1
TX	1	-	2	1	1	1	6
WI	-	1	1	-	-	-	2
OTHERS	-	3	6	7	2	2	20
TOTAL	19	24	42	41	11	9	146

# Table 1. Numbers of Research Abstracts published in 1993 to 2002 Sweetpotato Whitefly Strain B National Research and Action Plans Annual Progress Reviews.

# 1996 Review, San Antonio, TX

APHIS	2	-	-	12	-	-	14
ARS	3	19	16	19	7	1	66
AZ, ASU	4	6	10	1	-	4	25
CA	3	2	12	1	-	-	18
CDFA	-	-	-	3	-	3	6
FL	-	1	2	5	5	1	14
GA	-	-	1	-	-	-	1
TX	-	3	-	1	2	-	6
OTHERS	-	-	6	8	1	1	16
TOTAL	12	31	47	50	15	10	166

Silverleaf Whitefly, National Research, Action and Technology Transfer Plan, 1997-2001<sup>b</sup>

# 1997 Review, San Diego, CA

APHIS ARS AZ CA CDFA FL GA TX OTHERS TOTAL	- 7 2 3 - 2 - 1 15	- 9 8 7 - - 1 25	- 7 2 7 - 3 - 1 4 24	12 10 7 2 4 5 - 1 2 43	2 5 - 1 - 1 1 - 1 1 1	- 2 2 1 - 1 - 1 7	14 40 21 21 4 12 1 3 9 125
		1998 Re	view, Charles	ton, SC <sup>b</sup>			
APHIS ARS AZ CA FL GA TX OTHERS TOTAL	12 - 1 1 - 3 17	- - 1 1 - - 2	- 6 1 7 - - - 14	11 13 - 5 - 2 1 32	- 6 1 1 - 1 15	- 1 2 3 - - 1 9	13 38 3 23 3 1 2 6 89
		1999 Re	eview, Albuqu	ierque, NM			
APHIS ARS AZ CA FL TX OTHERS TOTAL	- 16 1 - 1 5 24	- 3 - 1 - 4 1 5	- 6 - 5 1 5 1 17	3 4 3 5 - 3 23	- - - - 3 9	6 - 4 - - 2 12	9 29 8 18 1 10 15 90

# 2000 Review, San Diego, CA

APHIS	-	-	-	2	-	-	2
ARS	15	1	6	7	2	2	33
CA	2	1	5	5	7	1	21
FL	-	1	-	-	-	-	1
TX	-	1	1	1	1	1	5
OTHERS	3	2	-	9	-	3	17
TOTAL	20	6	12	24	10	7	79
		2001/2002	Review, San	Diego, CA			
APHIS	-	-	-	3	-	-	3
ARS	14	3	3	5	3	3	31
AZ	1	2	-	-	-	-	3
CA	-	1	8	2	7	-	18
FL	-	1	4	-	2	1	8
TX	-	-	4	-	1	-	5
OTHERS	11	3	-	2	1	1	18
TOTAL	26	10	19	12	14	5	86

<sup>a</sup> For 1993-1997, A = Ecology, Population Dynamics and Dispersal; B = Fundamental Research, Behavior, Biochemistry, Biotype, Morphology, Physiology, Systematics, Virus Diseases and Vector Interactions; C = Chemical Control, Biorationals, and Pesticide Application; D = Biocontrol; E = Crop Management Systems and Host Plant Resistance, and F = Integrated Techniques, Approaches, and Philosophies.
 <sup>b</sup> For 1997-2002, A = Biology, Ecology, and Population Dynamics; B = Viruses, Epidemiology and Virus-Vector

<sup>b</sup> For 1997-2002, A = Biology, Ecology, and Population Dynamics; B = Viruses, Epidemiology and Virus-Vector Interactions; C = Chemical Control, Biopesticides, Resistance Management, and Application Methods; D = Natural Enemy Ecology, and Biological Control; E = Host-Plant Resistance, Physiological Disorders, and Host Plant Interactions; F = Integrated and Areawide Pest Management Approaches, and Crop Management Systems.

<sup>c</sup> APHIS = USDA, Animal and Plant Health Inspection Service; ARS = USDA, Agricultural Research Service.

# Table 2. Summary of Section A: Ecology, Population Dynamics and Dispersal Research Progress for the 5-Year National Research and Action Plan on Sweetpotato Whitefly Strain B (SPW) (1993-1997).

Research Approaches	Research Result Highlights
Define biology, phenology, and demography of SPW on greenhouse, field crop and wild host plants.	An extensive SPW host range of weeds, landscape vegetation and cultivated crops was identified in multistate surveys. Low winter temperatures reduced SPW survival and population development. Dispersal from cucurbits to cotton in the spring to alfalfa in the fall occurred in California and Arizona. In Texas, dispersal from cabbage to melons to cotton was common. SPW movement between sequentially planted crop hosts and naturally occurring weed hosts was evident. SPW population dynamics differed in different localities. Biology differed on different crops and also varied from year-to-year among and between geographical areas. Moisture deficit conditions increased SPW cotton densities in Arizona. Predators and severe wind/rain were important sources of mortality in life table studies. Significant correlations occurred between <i>Orius tristicolor</i> and <i>Chrysoperla carnea</i> and SPW population density. Parasitism was found to be extensive in some ecosystems in surveys in Arizona, California, Texas and Florida, but was not a limiting factor in population development.
Develop efficient SPW sampling plans for research and decision making.	Vertical population distribution of SPW adults and nymphs within host plants revealed that sampling in the top 20% of the plant provided good population estimates. Leaf disks from the 5th main stem nodes on cotton was the most efficient sample unit for eggs and nymphs. Based on this data, sampling plans for adults (5 to 10 adults/leaf turn) and nymphs ( $0.3 \pm 0.05$ nymphs cm <sup>2</sup> of leaf area) were developed and evaluated for cotton and further refined for cantaloupes. The cotton sampling plan was validated in over 8000 grower acres and both plans were adopted by agriculturalists. The intraplant distribution of SPW eggs, nymphs, and pupae were also studied on tomato. Field sampling methods for honeydew and cotton stickiness showed that lint stickiness was uniformly to randomly distributed within cotton fields compared with the highly clumped distribution of SPW nymphs.
Develop economic thresholds for SPW in relation to feeding damage, honeydew production and virus transmission.	Sticky cotton as a result of honeydew contamination is a major concern in the cotton textile industry. Identification of the sugars in honeydew and subsequent determination of their relationships to whitefly population and stickiness determined be thermodetector analyses were major accomplishments. SPW densities were correlated to reduced cotton lint quality. The action threshold for minimizing honeydew contamination of lint and yield protection were found to be between 5 and 10 SPW adults per leaf. In melons, SPW population relationships to melon harvest weights, % sugars, and other crop performance characteristics were established. Action thresholds of 0.5 large nymph per 7.6 cm <sup>2</sup> of leaf area were suggested. Action thresholds for adult whitefly have been incorporated in binomial sampling plans for cotton and melons. Rainfall and sampling variations for lint stickiness confound sampling method efforts.
Develop and test population models to describe and predict SPW dynamics.	Life process oriented models and more general spatial models were partially developed to identify influences of abiotic and biotic factors on site-specific and regional population dynamics. They can be valuable tools to identify research needs, assess priorities, and aid development of management systems. Model simulation results compared reasonably with trends observed in the sampling data, and were highly sensitive to natural enemy parameters. LandSat data were obtained and areawide models were developed and tested for the Imperial Valley. SPW population data for various crops were linked to cropping data and the effects of various cultural control strategies evaluated with the computer model.

Determine factors influencing	SPW flight was shown to be sustained at temperatures up to 47° C and for 2 h in a small fraction of the population (males
SPW dispersal.	longer than females). Diet, sex and age (longest flights 3 to 6 days after adult eclosion) were shown to affect flight
	performance. Dispersal was influenced by plant phenology, plant amino acid content, abiotic factors (diurnal cycles,
	temperature, light, wind) and physiological and morphological conditions. Marked and released individuals moved as far as
	4.8 km from the release site. Adult mark and recapture studies provided quantitative estimates of dispersal from melons and
	observational studies showed declining densities of SPW in cotton at progressively greater distances from source melons.
	Adult trapping studies in Texas indicated overall increases and declines in capture were correlated with increase and decline
	of the cotton crop.

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<sup>&</sup>lt;sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 2 (Continued): Summary of Research Progress for Section B: Fundamental Research - Behavior, Biochemistry, Biotypes, Morphology, Physiology,Systematics, Virus Diseases, and Virus Vector Interactions of the 5-Year National Research and Action Plan on Sweetpotato Whitefly Strain B (SPW) (1993-1997).

<b>199</b> 7).	
Research Approaches	Research Result Highlights
Studies of feeding	Extensive and detailed information was developed on SPW feeding, digestion, cuticular characterization and morphology. SPW
behavior: sensory	honeydew from cotton contained at least 30 sugars created from sucrose in phloem sap. Trehalulose and sucrose were the stickiest
receptors,	lint contaminants. Treatment of sticky cotton lint with enzymes at harvest removed the stickiness more quickly and efficiently than
ultrastructure,	water treatments alone. A and B biotype SPW wax glands were similar, surface cuticular lipids were characterized. Abundance of
morphology, digestive	vascular bundles in leaf tissues of some crops appeared to affect SPW feeding preference. Microscopic examination of the SPW
physiology; intra- and	digestive system indicates that it serves as an organ of disease transmission and inferences can be made concerning food
interspecific competition.	movement within the system, how water balance is regulated and in which tissues viral particles may accumulate. SPW egg stalks had walls of thick multiple layers of fibrous material; cytoplasm or yolk was not seen in the egg stalk. Observations on adults and nymphs indicate modes of stylet penetration, sheath formation, and feeding. Histological study of SPW stylet penetration has correlated host plant microstructure with success of whitefly colonization. Serine proteinase activities were undetectable in whole extracts of whiteflies. Results may expedite breeding of new cultivars of some crop plants. Nymphs target only minor vascular bundles, and stylets of surviving nymphs always reach bundles and phloem cells. Lower surfaces of the youngest leaves of most plant species tested were preferred.
Studies of biochemistry, physiology, nutrition, development and reproduction.	Rates of water loss (10%), and oxygen consumption for A and B SPW biotypes were similar. Thermal maximum thresholds were about 52°C. SPW survivorship was best on zucchini, intermediate on cabbage and lowest on sugar beet. Development times were longer on cabbage (30 d) than on zucchini (23.1 d). SPW females appear to prefer to oviposit on plants that enhance nymphal development. Host selection studies and host effects on tritrophic interactions have shown definite correlations between host plant and whitefly population levels. Artificial feeding systems were established to evaluate effects of chemical, toxins, and antibiotics on whitefly fecundity, development, and honeydew production, and to study feeding behavior, relative to intake of phloem sap and geminiviruses in phloem sap. One feeding chamber experiment tested semi-synthetic diet presented through a Parafilm membrane and demonstrated an ability to rear whiteflies successfully from egg to adult. Egg hatching rates were 2-50%, and specific dietary constituents were critical to development past third instar. Feeding behavior of immatures was studied using ingestion of fluorescent markers, and feeding structures active during ingestion and processing were video-recorded. Constituents of phloem sap ingested by whiteflies and composition of honeydew were studied; certain amino acids in phloem sap were in reduced forms, suggesting host amino acids are metabolized in whiteflies. Radiolabeled plant proteins were found to be taken up and converted by whiteflies, and radiolabel was found in new proteins and secreted into plants. Toxins from microorganisms were assayed against adult whiteflies using a new assay.

Studies to discover and analyze diagnostic characteristics of SPW, including component taxa, and to determine biological and genetic basis for development of biotypes, host races, and species, genetics and genetic diversity. Develop dsRNA and cDNA probe. DNA analyses showed differences between species and A and B SPW biotypes. Esterase analyses of SPW showed the B biotype is widely distributed worldwide. ACHE enzyme systems in A and B are different. rRNA analysis of A and B types suggest they are conspecific. Isoelectric focusing techniques used to analyze SPW from various locations in US, Egypt, Mexico and Spain identified A and B type. A, B and Hybrids have been reported. Unique dsRna was associated with the B-biotype." RAPDs analyses of DNA from a collection of *B. tabaci* indicates genetic variability between populations and that RAPDs were not useful markers for assigning phylogenetic associations. Direct genetic analysis of ribosomal (nuclear) and mitochondrial DNA of the *B. tabaci* complex was undertaken. The B biotype was proposed as a new species, *Bemisia argentifolii*, but key morphological characters to distinguish the A and B biotypes, or other biotypes are still lacking. Morphological plasticity and data from molecular studies indicate the most likely classification for the group is as a *B. tabaci* species complex. Some morphological plasticity stems from stimuli received by crawlers based in plant geography, hairiness, and crowded conditions on the leaf. Three whitefly complexes in the genus *Bemisia* have been defined based on classical taxonomic criteria and tentatively corroborated with molecular sequencing data. Molecular markers are under investigation for inference of phylogenies that can be compared to those predicted by morphometric and other methods. Successful diagnostics for natural enemy (especially parasitoid wasp) identification were identified. Many species were differentiated within the genera, *Encarsia* and *Eretmocerus*, using genetic fingerprinting by RAPD-PCR.

# Develop systematic analysis of the genus *Bemisia* utilizing various methods.

Ultrastructural studies of SPW eggs showed that mycetomes and their symbionts form a prominent mass at the distal end of each egg as in aphids and other homopterans. The surface lipids and wax particles of SPW (biotypes A & B) were characterized and no differences were found. The major wax esters were C46 and the major acid and alcohol moieties in the esters were C20 and C26, respectively. Major components of the wax particles were a C34 aldehyde and a C34 alcohol plus small amounts of C32 aldehyde and alcohol. Morphological studies for taxonomic considerations of the A and B biotypes focus on the pupal vasiform orifice, and mapping of setae and pore arrangements. Widely scattered loci of the two strains could not be separated. The submarginal setae, ASMS4, present in the A biotype, in type material from Greece, and in type material from poinsettia is absent from the B biotype 95% of the time. Because the character is not present or absent 100% of the time, it was not useful as a taxonomic character for differentiating the A and B biotypes or sibling species. A 550 bp sequence of whitefly 16S ribosomal mt RNA gene showed promise as an informative molecular marker, based upon phylogenies inferred from several trial datasets. The B biotype, or silverleaf whitefly, clusters with other Old World whitefly sequences in a consensus tree derived from 100 bootstrap samples of rDNA sequences, suggesting the silverleaf whitefly is of Old World origin.

# Identify and define SPW toxicogenic effects.

Plant growth regulator-induced silvering in cucurbits similar to SPW-induced silvering, but rootmass, chlorophyll and intercellular fluid expression was different. Gibberellic acid overcame PGR-induced symptoms and SPW-induced silvering, suggesting GA modification from SPW feeding. Feeding by biotype B nymphs causes squash silverleaf which is in part the result of the plant responses including autolysis of mesophyll cells at the feeding site. Other responses to B biotype feeding include the appearance of proteins at Mr 31,000 and 70,000 and the suppression of a Mr 60,000 protein in intercellular fluids of sweet sugar pumpkin. Other proteins that were suppressed were detected by 2-dimensional electrophoresis. While precise mechanisms of direct (toxicogenic) effects of whiteflies (especially B biotype) on host plants have not been discovered, some of the phenomena have now been histologically defined and nymphs identified as the source. New approaches to the molecular bases of plant syndromes such as tomato irregular ripening and squash silverleaf were considered. One such method that showed promise was the differential display of messenger RNA. Duration of feeding and distance from fruiting structures affected the severity of tomato irregular ripening. (á-Tocopherol, squalene, and linolenic acid increased in squash (pumpkin) plants prior to expression of squash silverleaf. Direct toxicogenic effects of the B biotype were expressed as uneven ripening in tomato, silverleaf in squash, stem streaking and thickening in *Brassica*, and paleness of leaves in lettuce and other composites. Pathogenesis -related proteins are typically sequestered in vacuoles or transported out of cells into the intercellular fluid (IF). The IF from silvered leaves of B biotype-infested cucurbits contained induced proteins of 31K and 70K Mr, whereas a characteristically prominent constitutively expressed protein (60K) was shown to be absent.

# Characterize SPW endosymbiont (SPWe) influence on metabolism, host range, and biotype formation.

Endosymbionts from A- and B-biotype SPW as well as several other whitefly species have been grouped according to ribosomal RNA sequences. Rod-shaped endosymbionts purified from adult SPW were morphologically similar to prokaryotes localized ultrastructurally in the mycetomes of third instar nymphs. Mycetomes in immature B. tabaci contain microorganisms which appear to be symbiotic to whiteflies and necessary for survival. Antibiotic treatment reduced mycetome size in offspring produced from treated adults and also reduced the immatures' ability to induce phytotoxic SSL. Ultrastructural studies by TEM indicated at least two morphologically different types of microorganisms. Microscopic analysis of endosymbionts has begun to define specific forms of symbiotic bacteria associated with the mycetomes of *Bemisia*, and to define differences among genetically defined populations. Artificial feeding systems were developed to rear whiteflies from egg to adult for the first time. Fundamental biological studies have also been conducted to evaluate the effects of insecticidal compounds on whiteflies and the transmission pathway of geminiviruses in adult whiteflies in relation to feeding behavior. Feeding chambers have also been utilized in the identification of sugars that contribute to stickiness due to whitefly honeydew. Mycetome-like organs containing prokaryotic-like and pleomorphic microorganisms were observed in immature instars, and at the distal tip of each egg. Transfer of endosymbionts in mycetocytes from the female to developing ova is a process that occurs continuously, prior to oviposition. Mycetocyte-containing bacteria were observed in oocytes inside a common membrane in the region that becomes the pedicel end of the egg. In the final stages of ovum development, the plasma of the oocyte completely surrounds the mycetocyte and the chorion thickens. Ooctyes with mycetocytes were present in adult females 16 hr after emergence, and none were observed in individuals that were less than 2 hrs old. Antibiotics known to have a negative impact on prokaryotic processes and metabolism were detrimental to whiteflies, whereas whiteflies appeared to be unaffected by those specifically inhibiting prokaryotic cell membrane synthesis or affecting cell walls. Negative effects were attributed to mortality of the primary endosymbiont in whiteflies.

Investigate etiology of diseases; biological and molecular characterization of causal agents; develop understanding of relationship; molecular probes for viral diseases; diagnostics and resistance; virus-vector specificity and interactions Several SPW-transmitted geminiviruses in Florida have been identified in bean, cabbage, tomato and weed species and compared at the nucleotide level at two sites in the genome. B. tabaci mouthparts and associated organs are similar in structure to those found in aphids. The salivary glands and the digestive system have been observed in detailed morphological studies. Diseases of vegetable and ornamental crops due to the whitefly-transmitted geminiviruses were also observed in Hawaii and Yucatan, Mexico. A PCR-based assay was developed to specifically and universally detect subgroup III geminiviruses. PCR primers target the core or central fragment of the coat protein (CP) gene and yield a 550 bp diagnostic product. The products are cloned and sequenced, and incorporated into the geminivirus CP gene sequence database. The approach has facilitated geminivirus identification. Whiteflytransmitted closteroviruses in lettuce was found that was vectored by the B biotype of B. tabaci. Artificial feeding chambers were used to study steps in the transmission process in which whiteflies ingest, acquire, and transmit geminiviruses. Geminivirus bNA was detected by PCR in saliva of the whitefly vector for the first time, whereas virus DNA was detectable in whole body extracts and honeydew of both vector and non-vector whiteflies. In Arizona, molecular cloning and characterization were the focus for isolates of cotton leaf crumple, squash leaf curl, and an uncharacterized geminivirus of Solanaceae. Studies were initiated to identify geminiviruses of cotton, kenaf, pepper, squash, tomato, tomatillo, watermelon, and weed species in Texas, and to determine the epidemiology of squash leaf curl in cucurbits. Studies in Florida concentrated on determining experimental and natural host ranges and identification of sources of inoculum for tomato mottle virus. TMoV infects species within the Solanaceae and Leguminosae, and that the most important source of TMoV inoculum is infected tomato fields. TMoV has been cloned and sequenced and has been shown to be related to other New World bipartite WFT geminiviruses. Breeding for virus resistance/tolerance in tomato was initiated with Lycopersicon chilense as a potential source of resistance gene(s). Engineered pathogen-derived resistance approaches have made promising strides using expression of *mutant CP*, BCJ, and Rep. An epidemic in bean caused by Macroptilium mosaic geminivirus were reported in Florida for the first time. a recently introduced strain of bean golden mosaic virus from the Caribbean Basin and is distinct from the BGMV -FL strain. A strain of TYLCV, nearly identical to TYLCV-Israel, was found for the first time in The Dominican Republic, Jamaica, and Cuba. A new WFT closterovirus, tomato infectious chlorosis (TICV) was discovered in coastal California tomato greenhouses and is transmitted by the greenhouse whitefly, but not B. tabaci. The virus was also identified in tomato greenhouses in Italy.

Study epidemiological parameters: vector population dynamics; disease thresholds; identify sources of inoculum, distribution, severity, and prevalence of pathogens. Correlate efficiency of transmission with biotypes, diversity and parameters of cropping systems. Squash lines screened show variable degrees of susceptibility to SPW silvering. Hybridization profiles and partial host ranges have been generated for over 12 new viruses in tomato and pepper in the U.S. PCR-based methods are being developed to determine virus prevalence in existing cropping systems and to determine virus vector relations. New biotype specific viruses are being characterized (ToMoV, putative new geminivirus in crucifers) and vector efficiency is being evaluated with respect to previously characterized viruses (LIYV). Biotype A transmitted lettuce infectious yellows (LIYV) more effectively than B biotype. Result of reduced transmission of LIYV has been larger yields in certain crops because A biotype populations have been reduced. Whitefly-mediated transmission studies were undertaken to standardize experimental transmission methods for broad-scale use. A PCR technique was developed for universal detection of WFT geminiviruses. PCR primers target the core region (nts 494-1048) of the capsid protein gene, the most highly conserved gene among subgroup 111 viruses. PCR products of diagnostic 550bp size indicate positive infection. Fragments are cloned and DNA sequences obtained to verify identity with sequences from well characterized viruses. A collection of core coat protein gene sequences has been established in a database. This database will be useful for geminivirus identification, molecular epidemiological studies, and for making predictions about phylogenetic relationships between geminivirus isolates. Among isolates collected and analyzed from New World sites, all but one virus, now known to be introduced into the Caribbean Basin from the Eastern Hemisphere, cluster with New World viruses. This suggests that the majority of viruses problematic in the US, Mexico, and Caribbean Basin originated in the New World, and are not introduced viruses. Further, these data extends the hypothesis that recent epidemics in the New World are primarily the result of indigenous viruses that rarely reached epidemic status in the past due to mobilization by new whitefly biotypes and altered disease pressures. Cloning and sequence of a second key region of the genome, the common region, are underway for the same library of geminiviruses for identification and to provide a second molecular maker in a key region of viral genomes. Comparative analysis of core coat protein and common region sequences permitted delineation of (putative) genetically related subclusters within the WFT subgroup, and indicates a wider range of variability within the subgroup than previously thought. Ability to define WFT geminiviruses as constituents of particular genetic subclusters by key viral sequences will permit a better understanding of evolutionary relationships, permit early recognition of new and emerging viruses, and suggests a possible strategy for selecting a range of prototype isolates for classical breeding or engineered resistance efforts.

# Study mating and oviposition behavior.

A and B biotype mating studies suggest reproductive isolation, but identified hybrid populations have occurred. Attempts to identify a SPW sex pheromone have been unsuccessful. Phosphorus deficiency cotton seedlings resulted in reduced oviposition on young leaves in greenhouse and growth chambers. Host acceptance was associated with low leaf sucrose concentrations (greenhouse), low total sugars and high leaf water (growth chambers) suggesting that factors affecting plant osmotic potential impact cotton susceptibility to SPW. Different whitefly species were shown to have different mating behaviors, and if extended to studies of B. tabaci populations has relevance to the (putative) reproductive isolation observed between biotypes. Studies of mating behavior and effects of mating on fecundity, viability, and population structure of whitefly on cotton indicate distinct behaviors for the B biotype and other whitefly genera. Viability of eggs can be much lower from an unfertilized female. Oviposition rate during the first days of adult life is not influenced by mating history. Mating behaviors for three whitefly species were captured on film using a video recorder attached to a binocular microscope. Leaf age and position on the plant influenced oviposition rates. Differences in behavioral patterns and temporal modulation of activities were documented during courtship and copulation. The basis for apparent reproductive isolation between certain populations of *B. tabaci* is not known, and has been another confounding factor in resolving the taxonomic status of *B. tabaci*.

# Determine factors influencing host plant selection and host acceptance.

Sequential feeding periods on different hosts or exposure to different hosts did not affect SPW fecundity or survival, suggesting no relation to preference. B biotype immatures developed faster than A's on the same host leaves, different host leaves or different cucurbit plants. Host preference was positively correlated to the abundance/length of vascular bundles per unit leaf volume. The preferred habitat is the under side of the leaf and this characteristic may be related to the shorter distance to vascular bundles from the lower compared to the upper leaf surface. Although leaf surface pubescence is an important factor in ovipositional behavior which resulted in females choosing the lower leaves which have fewer leaf hairs for deposition of 90-95% of the eggs laid, negative geotropism was the overriding factor influencing oviposition on lower vs. upper leaves. Acceptability of host plant feeding sites, based on oviposition rates, was greatest on youngest leaves compared to older leaves or cotyledons. High osmotic potential of leaves was positively correlated with adult whitefly acceptability of the host plant. Surface and internal structures of leaves from different host plant species and cultivars were shown to affect immature whitefly morphology and ability to colonize (density of immatures) and feed. The latter is due to variation in density and proximity of available vascular bundles within leaves. SPW is strongly attracted to surfaces reflecting in yellow-green wavelengths at certain times and to blue/UV reflecting surfaces at others. Host acceptance or rejection seems to be largely due to taste response following short leaf probing in the mesophyll. Influence of host and host choice on whitefly fecundity was examined, and at times, eggs were deposited on plants suitable for adult feeding that were not conducive to progeny survival. AC feeding monitors were used to correlate ovipositional activity with adult feeding patterns, feeding patterns of females were apparently more indicative of ovipositional site preferences than adult nutrition.

**Identify plant nutritional** A minimum of eight different proteins (four different Mr's) were isolated from plants colonized by the 'B' biotype. At least six other proteins of three different Mr's appear to be suppressed by SPW B biotype feeding on sweet sugar pumpkin. Measurements of and defensive responses to SPW and their effects chitinase activities show an inverse relationship with the length of SPW B biotype feeding time, i.e., the longer the plant is a host on SPW and natural for SPW, and the more progressive the silverleaf symptom, the lower the amount of chitinase activity. SPW were shown to be unsuitable prey for some predators, due to nutritional deficiencies. In cucurbits, several natural products (alpha-tocopherol, enemies squalene, and linolenic acid) were analyzed in developing leaves after whiteflies fed on lower leaves. Quantitative changes in these lipids may have nutritional consequences for the whitefly and/-physiological implications for the host plant. Linolenic acid and alpha-tocopherol (vitamin A) may induce host defensive responses and serve as vitamins for whiteflies, while squalene is a precursor of sterols which serve as critical structural components of plant cells and are also required by insects. Studies of protein expression in tomato indicated that several proteins were induced by adult and larval feeding. Though total leaf activities of chitinase, peroxidase, beta-I,3-glucanase, and chitosanase were greater than for controls; two proteins induced within 24 h were identified as chitinase and glucanase

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<sup>&</sup>lt;sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 2 (Continued). Summary of Research Progress for Section C: Chemical Control, Biorationals and Pesticide Application Technology of the 5-year National Research and Action Plan for Sweetpotato Whitefly Strain B (SPW) (1993-1997).

Research Approaches	Research Result Highlights
Identify new chemicals and	Greenhouse and field studies in Arizona, Georgia, California, Texas, Florida, Ohio, Maryland, New York and Mississippi
biopesticides for registration, that selectively control SLWF with	identified conventional chemicals alone or in combination that had potential for SPW control on cotton, vegetables and ornamentals. Section 18's were obtained that allowed the emergency use of Admire, Capture, and Danitol in several
minimal effects on beneficial organisms	states. The insect growth regulators, pyriproxyfen and buprofezin were highly effective for control of SPW as a foliar spray on cotton and melons. Their use in Arizona and California resulted in less damage and reduced tolerance to pyrethroids. Chemical control to maintain SPW populations below economic thresholds and avoid economic losses was demonstrated in many commercial crops. Yield responses to insecticidal control were demonstrated in virtually all host crops. Examination of plant derived oils, <i>Nicotiana</i> spp. extracts, neems, a "mycoinsecticide" <i>Beauvaria bassiana</i> , and several soaps appeared as useful alternatives in combination with insecticides, in chemical rotational systems, and for special use purposes.
Economic thresholds: Relate schedules and methods of application to economic thresholds.	Tentative economic and action thresholds based on yield vs. SPW density relationships in cotton, melons, cole crops and lettuce were established. Relationships between SPW and sticky cotton were investigated in California, Arizona and Texas. Similarly, significant relationships between SPW adult and immature densities were established on cantaloupe yield and quality in Arizona and Texas. An action threshold between 5 and 10 adults per cotton leaf generate the best economic return. A 190-acre trial in AZ validated the SPW per leaf threshold.
Insecticide resistance: Elucidate mechanisms, monitor whitefly populations and develop management systems	Baseline SPW insecticide susceptibility data was obtained using several techniques in California, Arizona, Texas and Florida; and Sonoran and Baja California, Mexico. Arizona and California bioassays showed a trend toward increasing pyrethroid resistance. PCR-based molecular diagnostics were used to investigate cyclodiene resistance in SPW biotypes. Synergists were used to implicate esterase detoxification and oxidative metabolism in resistance to chlorpyrifo and bifenthrin. Resistance management experiments and/or programs in cotton, based on rotation of insecticide types have reduced or eliminated resistance.
Application technology: Develop improved methods for application or delivery of materials to enhance control and conserve natural	Aerial application investigations involving air speed, above crop height, and nozzle type suggest improved delivery of sprays to target leaf areas. Electrostatic, air assist and hydraulic, ground operated spray systems have not shown clear advantage for increased underleaf coverage, but optimizing pump pressure, spray volume and nozzle configuration of conventional hydraulic systems improved underleaf coverage.

enemies..

Integration with other control tactics: Integrate chemical control with biological and cultural control. Florida, Texas and California studies indicated that parasitoids and predators responded differently to biorationals, IGRs and conventional insecticides. Although progress was made in studying the impact of chemicals and biorationals on natural enemies, no significant progress has been made on evaluating application methodologies on the SLW and natural enemy complexes.

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<sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 2. Summary of Research Progress for Section D - Biological Control of the 5-year National Research and Action Plan for Sweetpotato Whitefly Strain B (SPW) (1993-1997).

Research Approaches	Research Result Highlights
Determine effects of indigenous natural enemies on regulating SPW populations.	High levels of SPW parasitism occur in urban weed host habitats, but not cotton fields. Drapetis spp. flies were identified as a SPW adult predator. Predaceous coccinellids and chrysopids were identified as potential biocontrol agents. Natural enemy activity exists in most ecosystems and is responsible for some degree of population suppression. Surveys of parasites revealed at least one <i>Eretmocerus</i> sp. in Arizona, California, Texas and South Carolina. Up to 6 <i>Encarsia</i> spp. were found in southeastern U.S., 5 <i>Encarsia</i> spp. in Texas and 2 species in the southwestern desert regions. Parasitism rates vary seasonally and on different host plants from 0 to over 70% depending on crop or weed hosts. In the San Joaquin Valley of CA, the overall percentage parasitism was 1.5% during 1993-1994. Seven parasitoid species and five predator species were collected from the Lower Rio Grande Valley of TX during 1994; yet, this assemblage of 12 species was unable to control <i>Bemisia</i> populations below economic injury levels. An egg specific monoclonal antibody technology was developed and results suggest that <i>Orius tristicolor, Lygus hesperus, Chrysoperla rufilabris</i> and <i>Georcoris punctipes</i> were dominant predators of whitefly eggs in the West.
Develop methods for enhancing habitats with refuge plantings to conserve natural enemies.	Moderate to high SPW parasitism in urban and weed host areas suggest they may be potential refuges. Refuge plantings of sunflower, kale, collard and kenaf were established in Texas in association with unsprayed cotton plots. Rates of parasitization in the Texas refuge plantings indicates 3-4 parasite species are common seasonally. A native plant species, <i>Heterotheca grandifolia</i> , native to the desert southwest, and kenaf, a crop grown as a potential substitute for paper production harbor large numbers of whitefly parasitoids, ( <i>Eretmocerus</i> sp.). In the Imperial Valley, CA, refuge plantings have had limited impact on adjacent plantings of cantaloupe and cole crops due to the poor performance of the indigenous parasitoids in these crops. Annual plants as refuges are not suitable because of their poor sustainability and high costs, but perennials may be satisfactory. A new banker plant method using vegetable transplants is being tested in TX.
Identify new natural enemies in areas of SPW origin; foreign exploration, importation and release.	SPW natural enemies (parasites, predator, fungi) were collected in Egypt, Greece, India, Nepal, Pakistan and Spain and held in quarantine facilities. Availability of extensive exotic biological material was encouraging. At least 19 species of natural enemies including <i>Encarsia</i> and <i>Eretmocerus</i> parasite species, predators and several isolates of fungi were cultured in California, Florida, Texas, and France. Additional natural enemy collections from Argentina, Brazil, Cyprus, Italy, Israel, Malaysia, the Philippines, Taiwan and Thailand yielded three fungal pathogens ( <i>Paecilomyces fumosoroseus, Fusarium coccophilum</i> and <i>Beauvaria bassiana</i> ), at least 6 species of <i>Eretmocerus</i> and <i>Encarsia</i> , and 1 predator ( <i>Illeis koebele</i> ). Quarantine procedures including identification of adaptation and hyperparasitism for the collected species were accomplished and permits issued for their release. Extensive release programs occurred in California, Arizona, and Texas with many different parasitoid strains released. Exotic parasitoid and predator species were reported in CA, AZ, TX and FL indicating establishment and impact on SPW populations in selected systems.
Determine natural enemy host selection processes and mechanisms.	Host-plant, host-whitefly, and natural enemy interactions associated with plant hairiness, glandular exudates, and plant nutritional quality were demonstrated to influence the efficiency of natural enemies. Peanut and soybean parasitoids had either different parasitization efficiencies or different microhabitat preferences that were based on foliar pubescence of the host plant. Behavioral studies exa mining the tritophic relationships of SPW and <i>Delphastus pusillus</i> suggest that foraging efficiency was influenced by the phenotypes of both the whitefly and its host plant.

Inoculate/augment parasite and predator populations through propagation and release.	<i>Encarsia formosa</i> gave effective SPW control in greenhouses in Ohio but not in New York. <i>Eretmocerus californicus</i> releases were promising in field cage cotton studies. Various strains of <i>Encarsia formosa</i> failed to provide SPW control on eggplant in Hawaii and failed to establish within field cages in the Imperial Valley, CA. However, releases of 2 species of <i>Eretmocerus</i> into cotton in the San Joaquin Valley, CA provided significant reductions of whitefly densities. Artificial diets are being investigated to improve cost-effectiveness of mass-produced predators. Large scale field trials employing augmentation of parasitoids showed promising results in CA and TX. Augmentative biological control using the native <i>Eretmocerus</i> (AZ) is effective against whiteflies in the greenhouse.
Determine effects of pathogens on regulating SPW populations	Over 50 fungal pathogens were collected for SPW pathogencity screening. A staining method was developed to identify spore viability. <i>Beauveria bassiana</i> results were promising for SPW control on cotton. A large number of <i>Beauveria bassiana</i> and <i>Paecilomyces fumosoroseus</i> isolates from diverse geographic regions of North America and Asia were found highly effective against immature whiteflies in the laboratory. Applications of oil-formulated <i>B. bassiana</i> and <i>P. fumosoroseus</i> produced highly variable levels of control in field and greenhouse trials. Trials with a commercial strain of <i>Beauveria bassiana</i> for control of SPW infesting greenhouse tomatoes and ornamentals, and for cotton in the southern US provided control similar to the best insecticide-based programs. Separate trials with 4 isolates of <i>Beauveria bassiana</i> provided consistently high levels of whitefly control in experimental plots of cantaloupe and cucumber during both the spring and fall seasons. Trials conducted in cotton during the summer and in spring-planted tomatoes yielded poor results, yet trials in fall-planted tomatoes produced > 90% reduction in whitefly populations.
Evaluate compatibility of pesticides with SPW natural enemies.	Residues of seven insecticides on cotton leaves were tested for effects on four species of adult parasitoids. Buprofezin was non-toxic to all parasitoids; amitraz showed intermediate effects. <i>Eretmocerus mundus</i> from Spain exhibited some tolerance to the four insecticides.

Systematics of predators, parasites	Extensive biological material has been curated and identified. A protocol was established and agreed to by ARS, APHIS,
and pathogens	and SAES scientists for vouchering of specimens from importations and a centralized database for all importations
	established for Encarsia and Eretmocerus of world. Molecular technique classification of Eretmocerus and Encarsia has
	been accomplished and an identification key of the North American Eretmocerus spp. published. Publications with keys
	to identify the various other parasitoid species are in preparation.

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<sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

 Table 2 (Continued). Summary of Section E: Crop Management Systems and Host Plant Resistance Research Progress of the 5-Year National Research and

 Action Plan on Sweetpotato Whitefly Strain B (SPW) (1993-1997).

Action Plan on Sweetpotato Whitefly Research Approaches	Research Result Highlights
Determine effect of traditional crop	Frequent cotton irrigations reduced SPW populations verifying that water stress facilitated population development.
production inputs on SPW	Weekly irrigated plants also required fewer insecticide application to maintain adults below treatment thresholds.
population development.	Research conducted on evaluation of the effect of water stress on populations has been accepted and implemented by cotton growers. Integrating the use of reduced water stress by weekly irrigation and conventional insecticides has provided the best control. Water stressed plants had more SPW eggs than non-stressed plants. Sprinkler irrigation were found to reduce SPW populations compared to furrow irrigation in cotton. Whitefly flight behavior was reduced in irrigated plants as compared with water stressed plants. Populations of native parasitoids persisted in foliar applied imidacloprid plots, although their abundance was reduced compared with plots not receiving insecticide.
Determine temporal and spatial effects of host plants on SPW populations and dispersion.	SPW population differences within and between major cultivated crops were identified were apparently due to mechanism of resistance. The selection, crossing and genetic analysis to incorporate resistance characters into acceptable agronomic types has progressed with some favorable results. For example, smooth leaf cottons have shown less SPW numbers than hairy leaf varieties. The timing of SPW infestation occurrence on different crops in relation to spatial arrangements have allowed cultural practices such as planting times, harvest dates, crop residue destruction, etc., to be manipulated to reduce damage in some production areas. Large scale cropping patterns in SPW using LandSat data to map crops for modeling the spatiotemporal dynamics and SPW migrations have been integrated in areawide management systems. Areawide approaches focus on wind patterns, distance adults migrate, distribution of migratory SPW from point sources, and the ability of whiteflies to reproduce in relation to migration distances.
Determine effect of colored mulches, trap crops, intercropping, row covers, and other innovative cultural practices as potential SPW control methods.	The mode of action, disturbance, physical, mortality, etc. under sprinkler irrigation systems appears to be for reduced populations. Row covers were effective in limiting SPW cantaloupe infestations through the early season. Plastic mulches (white and silver reflective materials) repel SPW adults but have not been effective enough to justify their expense. Melons as a trap crop reduced the SLW infestations in cauliflower and interplanting melons in the system appear to increase efficacy. Fine-mesh screens exclude migrating SPW adults from greenhouses. Forced positive air flow through SPW-proof filters reduced the influx of SPW and incidence of virus transmission. Aluminum plastic mulch, from mesh screen row covers and living soil covers delayed the incidence of tomato mottle geminivirus on tomato by reducing early infestations in Florida.

# Develop reproducible evaluation techniques to isolate resistant germplasm

Experimental and mesophyll leaf tissue morphological differences appear to be associated with SPW numbers on different cotton lines. Trichome exudate production in tomatoes was inversely related to SPW feeding and oviposition. Cotton trichome density was related to high SPW densities on cottons. Methods to assess host plant resistance were developed in: alfalfa, broccoli, collard, eggplant, lettuce, melon, pepper, soybean, and tomato. More SLW oviposition occurred on pubescent and hirsute near-isoline soybeans versus a glabrous genotype. In melon, there was a positive correlation between long-leaf trichomes and number of SPW adults and immatures. Type IV and VI trichomes of Lycopersicon hirsutum accessions were evaluated for their role in resistance against Bemisia. Differences were mainly associated with day length and accessions. Plants under 8 hour day length had the most dense type IV trichomes and received the least oviposition versus 12 and 16 hour day plants. Trichome density did not explain all behavioral differences by SPW among accessions. A simulation model for evaluating plant germplasm was developed based on antibiotic factors. A testing procedure was established to provide a repeatable protocol for evaluation of lettuce germplasm for SPW feeding. Also, procedures were established for evaluation of SPW feeding, honeydew contamination and sooty mold contamination in alfalfa. Leaf thickness was highly correlated with the depth of vascular bundles in cotton. Work on other resistance mechanisms in cotton includes reduced trichome numbers, okra-leaf shape, and red plant color. Two collard hybrids had fewer whiteflies than open-pollinated cultivars, but three glossy genotypes were the most resistant. Pubescence was a factor in host oviposition selection and vertical distribution of oviposition on soybean, but survival and development were effected. Leaf thickness was found to be highly correlated with the depth of vascular bundles in cotton and may be developed as a resistance mechanism.

Identify resistant germplasm to SPW and associated viruses and plant disorders. SPW population differences among *Brassica oleracea* groups suggest plant size and color affected preference. Oviposition preference appears to be a factor in sweetpotato breeding line evaluations. Significant differences also occurred between peanut cultivars, melons, soybeans, cotton and tomatoes. High levels of resistance to SPW were not found in any crop species or cultivars tested. Evaluation of 15 Cotton cultivars for resistance to SPW resulted in no significant differences among cultivars. Differences were detected among 12 melon cultivars in ovipositional rate, nymphal numbers, adult survivorship in field trials, and greenhouse lifetable studies. Significant differences were detected among 12 broccoli cultivars for SPW colonization preference. Two zucchini lines (Sunseed 3 and A21-7) had resistance to SPW symptoms, but not to whitefly infestations.

Conduct plant breeding studies to	Tomato plant selection and crossing for tomato mottle resistance identified several sources of resistant germplasm.
select SPW resistant plant	Breeding lines were investigated for both melons and tomatoes. Snakemelon was identified as a parental material that had
germplasm.	low SPW counts/unit area. Several crosses with this material produced lines that had fewer SPW than snakemelon.
	Tomato lines were evaluated for stickiness and horticultural traits resulting in selection of 52 for low tomato mottle virus
	symptom expression, high stickiness and fruit-set. Breeding lines, half-sib "families" of alfalfa from a genetically diverse
	germ plasm pool, resulted in several crosses that had lint and well as fewer SPW immatures and less stickiness from
	honeydew. Alfalfa germplasm with whitefly resistance and other good qualities should soon (ca. 1998) be available for
	seed increases by industry. A glabrous type cotton with SPW resistance, good yield and other good qualities, was
	released and is commercially marketed as Texas 121. SPW on broccoli lines are being back crossed with commercial
	cultivars to improve agronomic characteristics.

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- <sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 2 (Continued). Summary of Research Progress for Section F Integrating Techniques, Approaches and Philosophies Research Progress of the 5-Year National Research and Action Plan on Sweetpotato Whitefly Strain B (SPW) (1993-1997).

Research Approaches	Research Result Highlights
Risk Assessment.	In Texas, large SPW nymph numbers provided the best index for relationships to yield and quality measurements. One large nymph/6.7 cm <sup>2</sup> of leaf at the 6th node or three adults/leaf at the 3rd node were initial thresholds. SPW population survey results were incorporated into GIS systems. GIS systems for predicting virus spread were used for tomato growers in northern Mexico. A broader understanding of virus-whitefly plant interaction, increased understanding of the role of different host plants in population development, increased understanding of SPW host plant resistance, and sampling techniques for monitoring population development are leading information for management development. There is an increased understanding of insecticide resistance management in whitefly management programs. Economic losses from California, Texas, and the Mexicali area of Mexico, showed a general downward trend to SPW infestations with new management approaches that included cultural practices such as destruction of old host crops and close attention to planting dates. Selected insecticides for vegetable crops grown prior to cotton production has reduced SPW in cotton early in early season. Fluctuation in populations occurred from season to season, but predictions are not possible. A more complete understanding of the role of different host plants in populations are essential for areawide, community wide Integrated Management Basis or Systems.
Spatial Analysis and GIS.	Crop sequencing and spatial arrangement studies are on an area-wide basis in AZ, CA, and TX have been displayed on GIS systems to SPW areawide management is now generally accepted. An informal network of scientists interested in the remote sensing applications to SPW dynamics has formed. Satellite crop map information is being coupled with pest management issues related to spatial distribution. Methods for mapping crop systems from satellite (Landsat) image data were developed and applied to the San Joaquin, Imperial and Lower Rio Grande Valleys. Methods of interfacing satellite based crop maps with insect simulation models have been developed and used in simulations.
Ecosystem modeling.	Survey data from CA, TX, and FL have been used to develop simple models for SPW growth and movement. Work in the HERMES environment to model interactions between SPW and two of its natural enemies had begun. GIS technology has been developed and is bring applied to models to evaluate and predict SPW population development and dispersal within certain Agroecosystems. Field Sampling is being used to validate prediction capabilities of whitefly population models. New methods of modeling and visualizing spatial dynamics have been developed which use the fast Fourier transform for solving the spatial dispersal/reproduction process in a spatial model of whitefly dynamics making it possible to use of PCs for simulation of spatially extended systems. A satellite image analysis system (TNTmips) has now been installed at Kearney Agricultural Center in the San Joaquin Valley. A full Landsat scene (IOOxI15mi) of the Valley (July 3, [994) has been provided thus demonstrating the potential of this technology to producers. Field Sampling in Texas, Arizona, and California is being used to validate prediction capabilities of whitefly population models.

Networks.	The NBCI and SPW management committee bulletin board in on line and accessible through Internet. It has a SPW forum and is available for the dissemination of information and for communication. Pest alert database is available via gopher at the University of Florida and includes whitefly and virus information. A series of homepages and 1-800 hotlines for consumers on whiteflies have been put in place to date in Florida, California, and Arizona. A broad network of whitefly information is linked on the WWW with homepages in Arizona, California, Florida, and Texas.
Integrated Extension Programs.	Area wide management programs have been initiated in Arizona, California and Texas. Publications and resource materials produced by scientists in Arizona, California, Florida and Texas are shared. Local leadership and team building, whitefly management committees and task forces have formed to address area wide management approaches. Training programs were developed and implemented in California and Arizona. Satellite crop map information is being coupled with pest management issues related to spatial distribution. This information is being targeted to IPM implementation programs as they relate to crop sequencing in areas wide management approaches. Whiteflies are an important component in cotton and vegetable IPM programs which have been implemented in Arizona, Mexico, California, and Texas. Growth and acceptance is visible in each program as they expand in geographic area and crops covered. A "train the trainer" program was developed and implemented in California and Arizona, which did much to strengthen the ties and cooperation between growers, extension, and research scientists. Satellite crop map information is being coupled with and related to pest population data, climatic data, whitefly population migration data, spatial distribution, and pest management data. This information is being targeted to IPM implementation programs as they relate to crop sequencing for community wide management approaches. A state regulatory agency (Arizona Dept. of Agriculture and a commodity group (Arizona Cotton Growers Association) took responsibility for stewarding a progressive Section 18 process which enabled uses of selective <i>Bemisia</i> growth regulators, namely Applaud and Knack. Cooperative Extension through industry collaboration developed the section 18 and implemented a proactive educational program.

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<sup>&</sup>lt;sup>1</sup> Summaries of research progress abstracts were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 3. Summary of Research Progress for Section A Biology, Ecology, and Population Dynamics of the 5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly Strain B (SPW) (1997-2002).

Research Approaches	Research Result Highlights
Determine life cycle vulnerabilities (life tables), population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.	<i>Bemisia</i> life table analyses on cotton in Arizona showed that natural factors, including predation and physical dislodgment were major mortality factors. High winter mortality occurs. Parasitism was a minor source of mortality. Survivorship from egg to adult ranged from 0-18.2% over 6 generations in sprayed and unsprayed fields. Studies on wild hosts in Israel indicate that parasitoids may contribute to maintaining low levels of SPW on lantana.
Develop sampling methodology, action and economic thresholds for all major crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Relationships between whitefly density and the occurrence of tomato irregular-ripening as well as preliminary sampling plans for SPW on tomato were developed.
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day-degree sub-models for estimating phenology and temporal patterns of whitefly, natural enemies and host crops	Partial development of large-scale temporal and spatial models and temperature-dependent, site-specific population dynamics models was accomplished. Additional biological and ecological detail, as well as information on various aspects of pest management needs to be integrated into the models to make them useful as exploratory research tools. A temperature-dependent, site-specific population dynamics model in cotton and cantaloupe was completed. Additional refinements, enhancements and field validation are needed for predicting population dynamics under various management regimes and environmental conditions.
Develop sampling methods for quality of cotton lint, vegetables and other commodities.	Cotton lint stickiness was shown to be randomly distributed in cotton fields. Preliminary sampling plans were developed for estimating pre-harvest cotton lint stickiness. Comparative evaluations of manual and high speed cotton stickiness thermodetectors revealed differences in performance that have important implications for the development of measurement scales for stickiness and the number of samples that would need to be collected for the precise estimation of stickiness. Thermodetector sticky cotton measurements in relation to amounts of honeydew and SPW populations were highly correlated. Plant stress can affect honeydew production.
Quantify whitefly and natural enemy dispersals and contribution to population dynamics.	Aerial distribution of SPW dispersing from cantaloupe fields to cotton and other hosts were documented. The SPW, <i>Eretmocerus eremicus</i> average 12 min. flight durations, respond to plant cues shortly after take off.
Define mating behavior, reproductive isolation, species, biotypes.	Surveys worldwide document the spread of SPW Strain B. Electorphoretic analyses demonstrated its presence in Australia and Brazil. Heterozygotes between <i>B. argentifolii</i> and the Australian <i>B. tabaci</i> corroborated previous laboratory results and support the existence taxonomic problems within the <i>Bemisia</i> species complex. Two SPW biotypes, cassava and sweetpotato, were identified in India.

Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Comparative morphological analyses of <i>Bemisia</i> pupae from around the world show highly variable characters suggesting that pupal morphology should not be the sole criteria for classifying individuals within the <i>Bemisia</i> species complex.
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Several antibiotics that interfere with bacterial protein synthesis affected SPW nymph growth and development. None affected adult oviposition rates or sex ratios. <i>Wolbachia</i> endosymbiotic bacteria was discovered in SPW suggesting significant implications for development of control strategies manipulating reproductive biology.
Characterize nutrient uptake and metabolism	Sorbitol was suggested to function as a thermoprotectant in SPW that enables them to thrive in desert environments. The unique pathway of sorbitol synthesis and degradation in SPW may offer an opportunity to develop transgenic plants that disrupt sorbitol synthesis and compromise SPW ability to deal with heat stress. Nymphs and adult feeding, stylet lengths and feeding mechanism descriptions were adequately described – nymph stylets 110-200 µm long, adult up to 217µm long.
Develop whitefly artificial diets and natural enemy mass-rearing.	An artificial diet and feeding system for rearing SPW immatures was been developed. Rates of instar development were comparable to those reported on various host plants. Adult <i>Encarsia</i> , parasitoid wasps, have been successfully produced using the diet and feeding system for SPW hosts.
**Pursue specific genetic and biological basis for variability in whitefly biotypes, strains, and species; determine impact of different genetypes/phenotypes on whitefly-mediated transmission and on the epidemiology of virus diseases.	A cell line from the silverleaf whitefly, BtB-2.97-Hunter & Polston was tested for its ability to support replication of the insect iridescent virus 6 (IIV-6). Several lines of evidence indicated that a productive infection was achieved. The cells displayed cytopathic effects, virus particles accumulated in defined areas of the cytoplasm, the cell-associated virus titer was detected at three orders of magnitude higher than that released into the media, and western blot analysis indicated CIV structural proteins were being expressed. Virus was also detected in nymphs by PCR and electron microscopy, but the infections were not highly pathogenic. Infection by whiteflies by IIV-6 suggests that more pathogenic viral isolates may be found for this insect and that the virus has potential to be used as an intracellular probe.

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<sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

Research Approaches	Research Result Highlights
Identification and characterization of new or emerging whitefly- transmitted viruses and strains.	Tomato yellow leaf curl virus (TYLCV) is a monopartite virus within the 17 geminiviruses infecting tomato alone. The group is considered the most economically important. Various techniques (antibodies, blots, PCR, DNA sequencing) have been developed for virus identification. TYLCV introduction into Florida may have significant economic impact on Florida tomato production. A new cucurbit-infecting geminivirus was identified in Imperial Valley, CA. Cucurbit yellow sticky disorder virus (CYSDV) was identified in the Rio Grande Valley, TX. Characterization of several new geminiviruser is being accomplished. New technologies are developing with expanding needs.
Molecular epidemiology: identification of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses.	PCR and DNA sequencing confirmed TYLCV in Florida. A number of symptomless weed hosts were found to have TYLCV. Host free periods effectively manage TYLCV. New systemic insecticides have been promising in some virus-disease problems and cultural practices such as time of planting are additional useful approaches to management.
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission.	Differences in feeding behavior of aphids and SPW adults relating to each of cell punctures early stylet probing was observed. Evidence for transovarial transmission of TYLCV in SPW and sexual transmission between males and females has been found.
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Whitefly host (tomato, beans, melons and peppers) free periods of 3 months have been shown to reduce TYLCV incidence. Living covers (coriander and perennial peanuts) and plastic mulch were found to reduce TYLCV.

# Table 3 (Continued). Summary of Research Progress for Section B Epidemiology and Virus Vector Interactions of the 5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly Strain B(SPW) (1997-2002).

Control of virus diseases: development of virus resistant germplasm through conventional and engineered/ molecular approaches. Define prospective strategies for selecting candidate viruses, identifying specific virus diseases to target, and prioritize specific crops and cultivars for protection approaches.

Gene marking of viruses has been useful in studying virus movement and infection activity. Potentially resistant CLCrV have been identified. TYLCV resistant tomatoes have been made commercially available.

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<sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

Table 3 (Continued).       Summary of Research Progress for Section C Chemical Control, Biopesticides, Resistance Management, and Application Methods of the	
5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly Strain B (SPW) (1997-2002).	

Research Approaches	Research Results
Improve insecticide efficacy: Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Numerous new chemistries, biorational plant extracts, potential biologicals and the like have been evaluated to expand conventional chemistry control options. Neonicitinoid systemics are effective for SPW control on numerous crops.
Develop improved methods of application including formulation and delivery of materials to improve control.	Higher volumes, increased pressure result in increased underleaf coverage. Conventional hydraulic spray systems hav been as effective as air-assist, electrostatic systems.
Conserve insecticide efficacy: Relate action thresholds to insecticide usage patterns.	Action threshold implementation were demonstrated to reduce pesticide use. Also, they are important in resistance management and natural enemy conservation.
Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.	Modes of action, genetic and biochemical basis for resistance need further emphasis. Monitoring of resistance has no revealed resistance to new chemistries (IGRs, neonicotinoids, etc.).
Improve insecticide efficacy: Improve techniques for monitoring resistance.	Resistance management monitoring and implementation of action thresholds have been implemented in most cropping systems and their value in pest management demonstrated repeatedly. Uptake of systemics and relationships to mortalities appear to be a promising approach to improve efficacy and efficiency.
Develop, evaluate and refine resistance management systems.	Rotations, tank mixes, and new chemistries integrated into SPW management systems have resulted in conservation of natural enemies in large area programs and prevents delay and/or forego resistance development. Cost commodity approaches to conserving use patterns for multicrop IRM is essential.

Integrate chemical control with other<br/>tactics.Increasing awareness of potentials for combining control strategies has stimulated implementation of control systems<br/>using new chemistry selections (IGR, neonicitinoids), biorationals, smooth leaf cottons and other resistant crops, and<br/>resistance management techniques. Conservation of natural enemies has become a reality with some of the new<br/>chemistries.

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<sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

Research Approaches	Research Results
Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	New insect growth regulators tested well under field conditions, and reduced loss of natural enemies. A Life Table analysis was conducted on natural enemies in cotton. Role of predators in cotton identified; importance of narrow spectrum insecticides highlighted. Life history tables have been constructed comparing mortality factors of natural enemies in conventional vs IGR treated cotton. Functional response data available for several parasitoid species. Imidicloprid and IGR's are more compatible with natural enemies. Pathogenic fungi may be promising. Annual refugia plantings are of limited value, perennials are promising.
Evaluate potential of alternate plants to act as in-field refuges or insectaries for natural enemies.	Combinations of annuals and some perennials show promise as within field natural enemy refugia. They are attractive to parasites but support low numbers of whiteflies. Annuals served as outdoor insectaries when releasing exotic parasitoid Perennial plants capable of growing in Imperial Valley identified, selected for a pilot project at a commercial organic farm. Research in the Imperial Valley has shown that perennial refuges support large numbers of whitefly and parasitoids that migrate to adjacent systems.
Augmentation of natural enemies: Develop natural enemy mass-rearing systems.	Foreign exploration exhaustive. Evidence for establishment following releases. Diets are being developed for generalist predators. Improvements have been made in rearing parasitoids, increasing rearing efficiency. Field studies have identified promising candidates for augmentative releases. Whitefly, parasitized by <i>Encarsia</i> , were grown on an artificia diet long enough for parasitoids to emerge as adults. First such report. Potential for research and commercial rearing. Mass rearing methods on SLWF has been accomplished. Artificial diets are still being researched, with economics undetermined. Parasite release monitoring suggest establishment in some areas. Several fungi appear promising for control.
Develop release technologies to maximize the effectiveness of mass- reared natural enemies in the field.	A novel "Banker Plant" field release strategy shows promise for increasing efficacy of releases. Releases of <i>Eretmoceru</i> into greenhouses controlled <i>Bemisia</i> attacking poinsettias when done at low pest densities. First year results show banker plants may prove more efficacious than releases of parasitoids by hand. Two species of coccinellids evaluated, compared for greenhouse use. Several release technologies have or are being tested. Banker plant technology appears to be very effective. Capsule delivery methods being tested. Cold storage of parasitoid pupae also being tested.
Evaluate augmentative parasitoid, predator, or pathogen releases.	Augmentative releases of parasitoids controlled <i>Bemisia</i> in large demonstration fields. These releases can be integrated with conventional pest management practices. Impact of <i>Beauveria bassiana</i> on generalist predators determined. Parasitoid dispersal was determined using new protein marking technique. Parasitoid release rates have been determined for major crops. Strategies for releasing/integrating parasitoid and predator in greenhouse crops have been determined. Significant information currently available on application of fungal pathogens in various crops.

Table 3 (Continued). Summary of Research Progress for Section D Natural Enemy Ecology and Biocontrol of the 5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly (SPW) Strain B (1997-2002).

Importation biological control: Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Combinations of annual plants that make excellent insectaries and can be farmed under local climatic conditions have been identified. Homeowners are being recruited to care for plants used for making releases. Several new exotics have persisted over several years and are multiplying and spreading in Texas and California. Whitefly suppression by exotic parasitoids determined by multiple researchers. Preliminary data suggests significant spread of established exotic parasitoids in some regions. Dispersal rates of natural enemies still under study. Book being prepared for publication.
Systematics, ecology, and population of	lynamics of natural enemies:
Clarify systematics of predators, parasitoids and pathogens.	Taxonomic studies have been completed on the exotic <i>Eretmocerus</i> and a key to their identification is in press. PCR techniques have been developed to identify the purity of cultures and aid in identification of recovered parasites. Key on exotic <i>Eretmocerus</i> published. Program developed for curating, cataloging recovered parasitoids. Several taxonomic keys developed for imported parasitoid species. RAPD-PCR techniques proven as quick identification method. Preliminary Satellite DNA techniques proven, however, still under development.
Determine <i>Bemisia</i> - natural enemy- host plant (Tritrophic) interactions.	Controlled laboratory studies showed that <i>Bemisia</i> and aphelind oviposition rates varied depending on host plant. Parasitoid foraging, oviposition varied in response to different plants (crops) and host whitefly. Plants varied in color, architecture, and semiochemicals. Tri-trophic interactions determined for <i>B. bassiana</i> / SLWF / tomato. Some research completed on parasitoid / host / plant interactions.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	The role of autoparasitism in native populations of <i>Encarsia</i> and its impact on native <i>Eretmocerus</i> has been evaluated. Results from one study show no adverse affect of <i>Encarsia</i> on overall parasitism of SLWF. No interference competition measured, with respect to whitefly control, when mixing primary parasitoids and autoparasitoids. Some life history data collected on parasitoid and predator populations in cotton. BioControl-Parasite simulation model available for testing/ validation. Some Laboratory data available for testing theoretical predictions of field level performance.

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<sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 3 (Continued). Summary of Research Progress for Section E Host-Plant Resistance, Physiological Disorders, and Host-Plant Interactions of the 5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly (SPW) Strain B (1997-2002).

Research Approaches	Research Results
Characterize resistance mechanisms and identify chemical/ morphological components, and study effects of insect adaptation.	Potential sources of germplasm for SPW resistance in alfalfa, cotton, melon, cole crops, and cucurbits; and resistance to virus symptoms and silverleaf disorder in cotton and cucurbits have been identified. Selection for SPW resistant alfalfa is close to completion. Partially SPW-resistant crops including cotton, collard, and melons have been identified. Varieties of cotton and tomato with resistance or partial resistance to transmitted viruses also have been identified. In collards, the glossy leaf trait, and in cotton, the okra-leaf trait have been suggested as mechanisms of whitefly resistance. Increased levels of phenolics and peroxidase in response to plant stress have been associated with decreased whitefly performance in tomato. In <i>Datura wrightii</i> , glandular trichomes were demonstrated to be a very effective resistance mechanism to whiteflies. SPW fecundity on alfalfa was higher for alfalfa-reared whiteflies than for cotton-reared whiteflies. In cotton, glabrous-leaf trait has been demonstrated as a mechanisms of partial SPW resistance against whiteflies in a tomato variety carrying the <i>Mi</i> gene has been shown to be due to factors encountered by whiteflies before they penetrate sieve elements rather than factors in the phloem sap. During initial contact with the plant on good host to poor hosts detected little difference in occurring SPW behavior and oviposition occurred on poor hosts. SPW probably will oviposit on resistant crops and mechanisms of plant resistance will be continuously challenged.
Develop molecular level techniques to produce resistant germplasm.	Characterization of plant genomone was demonstrated in tomato and squash. Pathogenesis related mRNAs accumulated in response to SPW feeding on tomato leaves. SPW probing behavior indicates that host evaluation phase is dominated by probing. The natural plant products, neem seed extract, azadiractin, and extract of bitterwood, were shown to be effective insecticides against silverleaf whitefly. The natural plant products, azadiractin, was shown to be an effective insecticide against silverleaf whitefly. Tomato transformation successful for ToMoV, appeared resistant to TYLCV
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition.	Morphological plant traits such as okra-leaf and large distance from leaf surface to vascular bundles in cotton, and glandular trichomes in <i>Datura wrightii</i> have been suggested as resistant characteristics. Fluctuations in amino acid concentrations over the lifespan of melon leaves appeared correlated with whitefly performance. Also in melons, group feeding by whiteflies was shown to create a nutrient sink in the plant, and thus provide the whiteflies with improved amino acid nutrition. Senescence in poinsettia reduces host plant quality for SPW. In cotton, decreased nitrogen fertilization decreases populations. In tomato, plant stress caused by fertilizer and/or water deficiency reduced host plant quality. In cotton, the okra-leaf trait and glabrous-leaf trait have been confirmed as a mechanisms of partial resistance. Factors encountered by whiteflies during their stylet penetration to vascular bundles has been shown to confer partial resistance in a tomato variety with the <i>Mi</i> gene. Phloem sap factors do not appear to play a role in this resistance. The known host plant range of SPW has been expanded to include some medicinal plants and weed species.

Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>c</sup>	SPW host evaluation was shown to be dominated by probing. Time spent in a particular behavior was affected by imidacloprid when the whitefly came into contact with the chemical in its diet rather than on the leaf surface. Intercropping of resistant within susceptible cole crops did not lessen the abundance of whiteflies. Improvements have been made in a system for rearing whiteflies on an artificial liquid medium. This will allow direct experimentation on the role of specific plant nutrients and allelochemicals on whitefly feeding and performance. Stylet contact with minor vascular bundles is essential for successful whitefly feeding on cotton. The fine structure of whitefly eggs and their attachment to host leaves has been studied with electron microscopy, and the distal end of the egg petiole that is inserted into the host leaves possesses morphological structures that suggest a role in water uptake from the host leaf which is a very important process for egg survival. Morphological studies on whitefly stylets indicate that they are sufficiently long to reach minor vascular bundles (the major feeding site) from virtually any place on the abaxial leaf surface of cotton. This makes variation in vascular bundle depth an unlikely mechanism of resistance to whiteflies in cotton. Variation in nitrogen fertilization has been shown to decrease amino acid concentrations in phloem sap and thus affects nutrition available to whiteflies. Whitefly feeding differentially induces pathogenesis -related (PR) proteins in two cucurbit species, cantaloupe and watermelon, and apparently is not affected by treatment with plant growth-promoting rhizobacteria (PGPR).
Study whitefly toxicogenic plant reactions.	Research on tomato identified a gene that is specifically induced by whitefly feeding. Four classes of genes were identified in inducing squash leaf silvering. These genes were further characterized by hybridization, sequence analysis and complementation studies. Genes specifically induced by whitefly feeding have been identified in tomato and in squash. These genes may play a role in the plant's defensive response to the whitefly and/or the plant's toxicogenic reaction such as irregular ripening in tomato and silverleaf symptom in squash. Two genes, one of which appears to be a general plant defense, have been shown to be differentially induced in squash by silverleaf and sweetpotato whiteflies. This may be related to the different toxicogenic effects of these two whitefly species on squash. The activation of these genes is systemic. In tomatoes, feeding by both silverleaf whitefly and greenhouse whitefly induced pathogenesis related genes, but not genes regulated by the octadecanoid pathway. These studies indicate that tomato plants perceive phloem-feeding silverleaf and greenhouse whiteflies in a manner distinct from that of chewing insects.

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<sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

# Table 3 (Continued). Summary of Research Progress for Section F Integrated and Areawide Pest Management Approaches, and Crop Management Systems. of the 5-Year National Research, Action and Technology Transfer Plan on Sweetpotato Whitefly (SPW) Strain B (1997-2002).

Research Approaches	Research Results
Development: Study whitefly-crop interactions as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/termination/harvest dates, other farm practices, intercrop relationships.	The roles of soil fertility (nitrogen), water-stress & some other agronomic factors on <i>Bemisia</i> population dynamics have been identified. Cross-commodity integration of pesticides used across multi-crop ecosystems and potential impact on whitefly management were developed. Progress was made with studies in Texas on seasonal dynamics of <i>Bemisia</i> on spring collards, and the impact of cotton defoliants on <i>Bemisia</i> and parasitoid populations. Economic study in Arizona showed > 80% reduction in pink bollworm-SPW IPM systems. Crop-free was demonstrated to break the tomato yellows leaf curl virus cycle.
Develop behavioral barriers to whitefly colonization and population development, e.g., mulches, trap crops, inter-cropping, row cove rs, etc.	Row covers and screens as physical barriers, mulches and oils as behavioral barriers, living mulches as behavioral barriers have proven useful in some situations. Investigations on intercropping took place in both desert and tropical environments. Approaches have also been identified as having potential.
Integration: Develop Integrated Pest Management systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	1) Insect Growth Regulators & biological control in cotton (conservation), 2) imidacloprid & other chemical control taction & various forms of biological control, especially in vegetables, 3) studies of direct & indirect effects of chemical control on bio-control agents. A number of field studies employed multiple tactics directed against whitefly populations. Biorational insecticides were examined in combination with IGRs and other biopesticidal agents such as <i>Beauvaria bassiana</i> for control efficacy of silverleaf whitefly. There was an indication of inhibitory action by <i>B. bassiana</i> when used in combination with imidacloprid as well as deleterious effects to predators contacted by <i>B. bassiana</i> treatments. Neem products were used to reduce whitefly populations and incidence of yellow mosaic virus in India. A melon trap crop was integrated with chemical control to focus potentially disrupting treatments into a limited area while preserving natural mortality factors in cotton as the principle crop. Much progress with integrating control tactics: 1) Life table evaluation of both natural and insecticide-based mortalities, 2) Compatibility of IGR's for whitefly control in greenhouses 3) IPM development in cotton for <i>Bemisia</i> and other cotton pests, 4) Augmentative biocontrol using crop transplants inoculated with parasitoids in Admire <sup>®</sup> -treated fields.
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision- making, biological control, etc.	1) <i>Bemisia</i> distributions have been examined in tomato, 2) new binomial sampling system for large nymphs in cotton, & integration with thresholds for IGR decisions, 3) sampling & IGR re-treatment decisions tested in cotton. Analysis of types and patterns of chemical treatments made on a large number of cotton fields in central Arizona over a 4 year period revealed extraordinary differences in the number of treatments and amount of time that whiteflies exceeded threshold levels prior to and following the advent of the IGRs buprofezin and pyriproxyfen. The proactive initiative taken by Arizona growers to pursue chemical use harmonization across commodities required consideration of all aspects of pest and crop management. A similar whole system appraisal was made in the San Joaquin Valley with an emphasis on integrating multiple practices with diverse insecticide classes as part of an insecticide resistance management program. Fourth consecutive year of monitoring <i>Bemisia</i> populations in the Imperial Valley using the CC trap. Sampling-based refinement of action thresholds for IGR's in Arizona cotton show better conservation of predators with high threshold (1 vs. 5).

Delivery and Implementation: Elevate single field/farm practices to areawide community-based contexts; develop methodology for installing and evaluating areawide control technologies and their impact.	Areas dominated by cotton production were identified in AZ & CA for implementation of cooperative programs. Areas of melon and vegetable production were identified in TX for potential area-wide programs. Area-wide sampling, & decision-making was the main focus of most programs; however, coordinated natural enemy releases were also conducted. Large areas in the San Joaquin Valley observed specific guidelines for IPM and IRM in cotton with evaluations continuing on the benefits attained over areas that did not observe these guidelines. Community wide evaluations were made on quality of whitefly management according to chemical control practices. The successful IPM and IRM programs practiced in Arizona cotton continued for a third consecutive year. Further cross-commodity development of these programs is under way. Economic analysis of the use of IGR's in Arizona cotton.
Implement and deliver Integrated Pest Management and Integrated Crop Management systems or system components to clientele.	IPM recommendations were distributed in AZ, CA, Mexico & FL. Bilateral discussions between Brazil & U.S. took place. A 'best agricultural practices' demonstration project was conducted on 50.5 acres (Univ. of AZ Maricopa Agricultural Center) with inputs from agronomy, entomology, irrigation management, weed sciences and plant pathology. SPW management was fully integrated with management of other insect pests and required only a single application of pyriproxyfen. Lint yields of 2.81 bales/acre were higher than the historical as well as the 1998 farm-wide average. An integrated areawide management program involving the cooperation of growers, PCAs, ginners and state and university researchers was expanded during a second year in the San Joaquin Valley. Sticky cotton bulletin published by University of Arizona and Cotton Incorporated. International development of IPM for managing whiteflies and geminiviruses. Development of <i>Bemisia</i> -resistant alfalfa cultivars. Crop and pest management demonstration project on cotton in Arizona.

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- <sup>1</sup> Summaries of research progress abstracts or tables as listed were prepared by the indicated authors. See individual abstracts for research credits and more detail.

## II. Plenary Session Keynote Address Summaries

#### Section A: Plenary Session Summary

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#### Whitefly Metamorphosis—Timing, Regulation and Influences on the Development of its Parasitoid, *Encarsia formosa*

A staging system based on body depth (thickness) and maturation of the adult eye was used to track development and identify physiologically synchronous 4<sup>th</sup> instar/pharate adult silverleaf (SLWF) and greenhouse (GHWF) whiteflies. The median depth of Stages 1, 2, 3 and 4 is 0.1, 0.15, 0.2 and 0.25 mm, respectively, and the depth of Stage 5 is  $\ge 0.27$  mm. As the whitefly progresses through Stages 6, 7, 8, and 9, the eye changes from a small, intense red dot (Stages 1 - 5) to a diffuse dot (Stage 6), to a light red adult eye (Stage 7), to a medium bipartite red (Stage 8) and finally dark red or redblack bipartite adult eye (Stage 9). An examination of histological sections of last instar SLWFs revealed that adult eye and wing development were initiated in Stage 6, the stage in which external examination showed that the eye had just begun to undergo pigment diffusion. In contrast, adult eye and wing development of GHWFs was first observed in Stages 4 or 5, a time prior to the initiation of eye pigment diffusion. Wings had become deeply folded by Stages 6 and 7 in GHWF and SLWF, respectively. Although adult development was initiated later in the SLWF than in the GHWF, metamorphosis occurred at a very rapid rate in both species of whitefly. Within approximately 24 h, the simple bi-layered wing bud developed into a wing of nearly adult proportions, and within an additional 12-24 h, the nymphal eye and wing bud had been replaced by the well-differentiated eye and wing of the whitefly adult. Ecdysteroid titers, as measured by a very sensitive ELISA (range = 500 -40,000 fgs), peaked at Stages 4 - early 6 in the SLWF and at Stages 4 - 5 in the GHWF, a time just prior to or at the onset of adult development. It is noteworthy that at their peak, ecdysteroid titers were three to four times greater in the SLWF than in the GHWF, although for both species, titers were 100 to 1000 times less than those reported for species from other orders of insects. The identity of the host plant did not significantly affect the magnitude or timing of the ecdysteroid peak in SLWF last instars. However, leaf pubescence did impact the length, width and depth dimensions of 4<sup>th</sup> instar/pharate adults. When reared on the pubescent leaves of green beans and tomatoes the period of whitefly development prior to becoming pharate adults could be divided into five stages with depth reaching a maximum of 0.25 - 0.3 mm during

stages 4-5. In contrast, when reared on the glabrous leaves of cotton, collard or sweet potato, whitefly body thickness rarely surpassed 0.17 mm, a thickness equivalent to Stage 2. However, whitefly nymphs on glabrous plants were significantly longer and wider than those developing on pubescent leaves. Thus, it appears that Stages 3-5 of nymphs that are reared on pubescent leaves are developmentally similar to Stage-2 nymphs that are grown on glabrous leaves.

The critical feeding period (CFP), the time after which 50% of whiteflies can be removed from the leaf and complete development, i.e., emerge, was also determined. Stages 1 through 9 were removed from the leaf, maintained under high humidity and monitored for percent adult emergence. It was surprising that although adult development was initiated earlier in GHWFs than in SLWFs, the CFP occurred at a later stage in the GHWF. At 25°C and L : D 16 :8, more than 50 % of test GHWFs and SLWFs successfully completed development if removal from the leaf was performed on or after whiteflies reached Stages 4 and 3, respectively. When removed at Stage 2, adult emergence was observed in 40% of SLWFs but in no GHWFs.

It was confirmed that the developmental rates of *Encarsia formosa* differed significantly based upon the whitefly instar parasitized. Development was the most rapid and synchronous when 3<sup>rd</sup> and young 4<sup>th</sup> instar whiteflies were parasitized. In addition, adult longevity was greatest and emergence was most synchronous when these older instars were selected for oviposition. It is significant that our results indicate that no matter which GHWF instar until the host has reached Stage 4/5 of its last instar. It appears, then, that a condition(s) associated with host adult formation may be required for the parasitoid's final molt.

#### Section B: Plenary Session Summary

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### Diversity of Begomoviruses and the Whitefly Vector in the US Sunbelt States: An Overview

Recent upsurges in populations of the whitefly *Bemisia tabaci* (Genn.) have drawn much attention to its worldwide importance as an insect pest and as the vector of emergent begomoviruses (Family: *Geminiviridae;* Genus: *Begomovirus*). Several begomoviruses that are considered 'new' and others previously regarded as minor pathogens have been linked to recent epidemics. Recent studies have revealed much variation in begomoviruses, despite the view that DNA-containing viruses do not rapidly accumulate mutations. Also, certain *B. tabaci* 'variants' are known that more effectively or selectively transmit certain begomoviruses and exhibit biotic differences that may influence their spread. Patterns of distribution and dissemination of begomoviruses transmitted by *B. tabaci* are poorly understood because standardized molecular-based tracking methods have not been available. Understanding virus/whitefly vector/host plant interrelationships in the context of emerging problems can be achieved only by linking predicted evolutionary histories with epidemiology using molecular phylogenetic approaches. To address etiology and investigate the molecular epidemiology and ecology of begomoviruses, we have employed a molecular marker approach targeting key viral sequences that enable provisional virus identification. Polymerase chain reaction (PCR) and degenerate primers have been designed to amplify the 'core' region of the viral coat protein gene (core CP) of virtually all begomoviruses (Genus: Begomovirus; Family: Geminiviridae) (Wyatt and Brown, 1996). Subsequent 'positives' are subjected to additional PCR and sequence analysis using a suite of degenerate primers to target additional informative sequences. We applied this approach to identify and map the global distribution of begomoviruses. Analysis of the begomoviral CP sequence reveals an Old and New World clade. Old World begomoviruses are further separated based on major geographical barriers. In contrast, New World begomoviruses cluster as a single, large polytomy containing certain distinctive lineages that are somewhat representative of geographical boundaries in the region; however, definitive barriers are less apparent than for Old World exemplars. Withincluster divergence for New World viruses is somewhat less than for Old World viruses, suggesting that extant begomoviruses have evolved from fewer founder events than occurred for the Old World viruses, or that only a few lineages survived despite multiple founder events. All begomoviruses are transmitted by a single species of whitefly, Bemisia tabaci (Genn.) (Aleyrodidae: Hemiptera). A species complex has been proposed to encompass whiteflies identifiable as *B. tabaci*, which is rich in biological variants that lack distinguishing morphological characteristics. We have previously shown that the mitochondria cytochrome oxidase I (mtCOI) gene is useful for studying genetic variation within this group. A fragment (~800 bp) of the mtCOI gene was amplified using polymerase chain reaction and degenerate primers and sequence analysis reveal numerous clusters (or clades) with a basis in geographical origin. New World B. tabaci cluster as two groups: (1) North/Central America & the Caribbean region and (2) South America, which exhibited <2-6% divergence, while eight or more Old World clades are well-supported at ~8-12+% divergence from other Old clusters and from the two New World clades as well. These results provide no

overwhelming rationale for erecting the B biotype as a distinct *Bemisia* species, in light of the extensive, but as yet, poorly understood basis for the diversity evident in this complex. Also, begomoviral CP and whitefly vector mt COI trees are generally concordant with respect to extant geographical origin, suggesting that markers are also reliable predictors of present-day distributions of viral or whitefly genotypes. Interestingly, virus-vector relationships corroborate a tight evolutionary relationship between all *B. tabaci* and the Genus: Begomovirus that is thought to have a basis in virusvector specificity. Indeed, the CP gene is the most conserved among begomoviral ORFs. This observation, together with the demonstrated congruence between vector and virus trees, reflects a co-evolved relationship between the whitefly vector and viral capsid protein and confers properties necessary for vector-mediated transmission.

> Robert L. Gilbertson University of California, Davis

# Tomato yellow leaf curl virus in the Dominican Republic-A case study

Tomato yellow leaf curl disease is one of the most devastating diseases of tomato (Lycopersicon *esculentum*). Infected plants show stunted and erect or upright growth and leaves with upward curling, crumpling and interveinal chlorosis. Yield losses of 100% are common, particularly if plants are infected at an early stage in development. The disease is caused by the whitefly-transmitted geminivirus (Genus Begomovirus, Family *Geminiviridae*), *Tomato yellow leaf curl virus* (TYLCV). TYLCV is transmitted by Bemisia tabaci and *B. argentifolii* (*B. tabaci* biotype B) in a persistent manner. It is a phloem-limited virus that is not transmitted via seed or by sap-inoculation. TYLCV was first described in Israel in 1931 and has been reported from many countries around the Mediterranean Basin and Africa. It is a limiting factor for tomato production in countries such as Spain, Cyprus, Jordan, Israel, Egypt, Senegal, Mali, Thailand, and India. Moreover, it has been established that tomato yellow leaf curl disease is caused by more than one begomovirus species, depending on the geographical location. The name TYLCV now refers to the monopartite begomovirus that causes TYLCV in Israel.

TYLCV had not been reported from the Western Hemisphere but, in the early 1990s, tomatoes in the Caribbean island, the Dominican Republic (DO), developed disease symptoms that were similar to those caused by TYLCV. Using the polymerase chain reaction (PCR) and a pair of TYLCV-specific primers designed to direct the amplification of a full-length TYYLCV DNA fragment, the expected size DNA fragment (~2.8 kb) was amplified from tomato tissues showing TYLCV symptoms from the DO. This DNA was cloned and sequenced. DNA sequence analyses revealed that the cloned DNA was nearly identical (97%) to TYLCV and that the genome organization of the Dominican Republic TYLCV (TYLCV-DO) was identical to that of TYLCV. Using squash blot hybridization analysis and the cloned TYLCV-DO as a probe, it was established that TYLCV had spread throughout the major tomato growing areas of the Dominican Republic and that the primary host was tomato. Thus, TYLCV had been inadvertently introduced into the DO and it had spread rapidly, presumably via already existing *B. tabaci/B. argentifolii*. TYLCV was not detected in weeds with or without symptoms, with the exception of *Datura stramonium* plants that showed leaf curl and interveinal yellowing.

As part of a TYLCV management strategy, a whitefly host-free period was instituted in the two major processing tomato growing areas (one in the North and one in the South) in the DR. To assess the efficacy of this approach, a PCR-based method for assessing the incidence of TYLCV in whiteflies was used to monitor TYLCV incidence over time. By the end of the tomato growing season, TYLCV was detected in nearly all of the whiteflies collected from tomato fields in the North and the South. However, during the host-free period, the incidence of TYLCV decreased dramatically in whiteflies (now collected from weeds around the fields). Thus, the whitefly host free period resulted in a considerable decrease in the incidence of TYLCV in the DO. More importantly, the host free period provided a lag period of 6-8 weeks before newly planted tomatoes developed TYLCV symptoms.

Thus, the whitefly host-free period resulted in a dramatic decrease in the incidence of the virus, but it was able to persist during the host free period. Using squash blot hybridization analysis of numerous crop and weed plants in and around TYLCV-infected tomatoes, very few potential TYLCV host were identified. Interestingly, while pepper plants commonly showed symptoms of virus infection in the DO, few were positive for TYLCV, even in cases where pepper fields were adjacent to heavily infected tomato fields. An exception to this was a pepper field in the North in January 2000 that had high whitefly populations and numerous plants with stunted growth and upcurled and yellowed leaves; TYLCV was detected in these plants by PCR. Thus, TYLCV can infect pepper in the DO; the low frequency may relate to host preference of the whitefly vector. TYLCV infection of common bean in the DR was demonstrated by planting seed of cv. Topcrop near a tomato field with a high incidence of TYLCV infection. Many bean plants showed stunted growth, curled and crumpled leaves and chlorosis; these plants were infected with TYLCV based on PCR analysis. Interestingly, it appears that the smaller seeded beans (e.g., pinto bean, cv. Othello) may develop symptomless infections. Thus, beans are also a host for TYLCV in the DR. These results demonstrate that it is important to

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include these crops as well as tomato in the host-free period.

Squash blot hybridization failed to show TYLCV infection in most weeds. However, TYLCV was detected in weeds using the more sensitive PCR approach. The virus was detected in a number of common weeds, including *Croton lobatus*, *Cleome viscosa* and *Solanum nigrum*. Thus, TYLCV may persist in weeds as a symptomless infections; the inefficient nature of these infections may explain why it takes weeks before newly planted tomatoes develop TYLCV infections via inoculum from weeds.

Management of TYLCV in the DR involves an integrated approach using the whitefly host-free period, whitefly control (e.g., imidicloprid), and the use of early maturing hybrids and resistant varieties. Using this approach, levels of tomato production in the DR have returned to pre-TYLCV levels.

# Section C: Plenary Session Summary

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# Insecticide resistance in *Bemisia tabaci*: Past expectations and present reality

Pesticide resistance is considered to be one of the limiting problems in agriculture and human health. It is therefore critical that we understand the nature of resistance and the steps that can be taken to avoid its occurrence. This is especially true for a handful of recidivist organisms that repeatedly develop resistance to novel chemistries in agricultural systems and environments all over the world. Bemisia tabaci is one such organism that has a welldocumented legacy of resistance to all major classes of insecticides. Moreover, populations of *B. tabaci* in some parts of the world have already developed high levels of resistance to novel compounds that have proven so effective in the southwestern USA. Because insecticides remain a primary control option against *B. tabaci* in the USA and much of the world, there remains a high potential for further cases of resistance development. However, the rate at which resistance develops will depend upon genetic, biological/ecological and operational factors. The importance of some of these factors will be considered with reference to the case of whiteflies in California and other regions of the world. The current status of insecticide resistance in *B. tabaci* to various insecticides in various regions of the world will be discussed. More importantly, the differences among regions will be compared and contrasted to ascertain the principle factors that determine whether resistance development will be a significant or trivial occurrence.

Resistance to organophosphates and pyrethroids in B. tabaci was reported during the early 1980s in Sudan and California. Additional examples of resistance to OPs and pyrethroids were identified in the following years at a time when choice of chemistry was much more limited than at the present. Less effective, more broad-spectrum chemistry used repeatedly as the only recourse to increasing whitefly populations would certainly represent an untenable situation from a resistance management standpoint. It is little surprise that the resistance examples of the 1980s were all related to cotton production, a longseason crop that relies heavily on insecticides. In the California example, the Type A strain of B. tabaci was principally a cotton pest in that its population growth cycle occurred in cotton and not on a series of crops prior to and in addition to cotton, as was to become the case for the Type B strain. Consequently, refuge areas for preserving susceptible genes were not part of the set of ecological factors that would become so important for the Type B strain during the 1990s in California.

The most significant change that took place in the 1990s was the invasion of North America by the Type B stain of B. tabaci. Although this had been initiated as early as 1985 in Florida, it wasn't until the late 1980s that signs of the Type B strain were apparent in Arizona and California. After simmering for some period of months and perhaps years in the desert agricultural valleys, B. tabaci Type B erupted in 1991 in the Imperial Valley in a sensational display of biotic force. What had been mostly a late season cotton pest and vector of viruses to fall vegetables in the form of Type A was now seemingly transmutated to a year round pest of cotton, fall and winter vegetables, and spring melons in the form of Type B. This meant heavier use of insecticides on multiple crops, and from what people assumed based on unmanageable numbers of whiteflies, high levels of insecticide resistance.

A resistance-monitoring program was initiated in the Imperial Valley in 1992 to determine relative susceptibilities of whiteflies to the various insecticides that were being used on crops year round. Additionally, cross-resistance patterns between chemistries and the influence of management strategies to maintain long-term viability of products were also studied in the laboratory and the field. Results of four years of whitefly monitoring data to bifenthrin, endosulfan, chlorpyrifos and methomyl showed no progression towards higher resistance in spite of heavy dependence on these compounds. Instead, whiteflies were less sensitive in 1993 compared to the following 2-3 years as indicated by lower LC<sub>50</sub>s, thereby demonstrating reversion rather than progression of resistance. This was not the case for all whitefly treatments used in the Imperial Valley. A comparison of whitefly responses to a mixture of danitol<sup>®</sup>+orthene<sup>®</sup> from 1994 to 1999 showed some loss of susceptibility between 1994 and 1997, but stabilized through 1999. However, the modest shift towards

reduced susceptibility observed in the Imperial Valley contrasts sharply with the high resistance levels to danitol+orthene observed in central Arizona. A year-byyear comparison between 1995 and 1998 consistently showed a flatter response to increasing concentrations of danitol+orthene for Maricopa whiteflies compared to whiteflies collected in the Imperial Valley, even though whiteflies from both regions were collected from cotton that received a similar number of danitol+orthene treatments. The fact that whiteflies from different regions, but same crop, responded so differently to similar exposures of insecticides argued strongly for the importance of the different ecological conditions under which each population was exposed. Insecticide selection pressure for Maricopa whiteflies in terms of the proportion of the regional population exposed to repeated treatments of danitol+orthene was almost certainly much higher than that proportion of the Imperial Valley regional population exposed to danitol+orthene. This is due to the proportionally low acreage of cotton grown in the Impeial Valley relative to other crops such as alfalfa as well as ornamentals grown at homes and businesses in the more densely populated Imperial Valley. In contrast, cotton is the highest acreage summer crop grown in central Arizona, and this region is sparsely populated by humans. Thus, whiteflies would be concentrated in cotton and would therefore receive a greater exposure quota of insecticide treatments as a function of the total regional population. It is somewhat analogous to the situation in the Imperial Valley in the 1980s where the concentration of the A type in cotton led to greater proportional exposure of the whole population with little refuge or conservation of susceptible genotypes.

The neonicotinoid insecticides have become an indispensable group of compounds for combating whiteflies on crops grown worldwide. Monitoring data of 3 neonicotinoids, acetamiprid, imidacloprid and thiamethoxam in 1999 and 2000 showed variations from field to field, however, the  $LC_{50}$ s were extremely low (<10 ppm) and gave no indication of reduced sensitivity. An interesting trend was evident in our study which showed highest survivorship to imidacloprid occurring with whiteflies collected from fall melons. The highest susceptibility to imidacloprid was during early fall when insects were collected from cotton compared to whiteflies collected later in the fall on brocolli plants. These results confirm the interesting phenomenon of shifting levels of susceptibility depending on crop, type of treatments applied, and other ecological events that may interact to maintain an overall high level of susceptibility.

Comparative bioassay results with imidacloprid on whiteflies from Guatemala collected on melon crops compared to the Imperial valley insects indicated a plateauing of mortality at the higher concentrations with <80% mortality at 100 ppm suggesting that they may be resistant to this compound. Similarly insects from Almeria appear to be resistant to imidacloprid in addition to some other chemistries. A comparison from the literature on whiteflies from India also report resistance to imidacloprid and pyrethroids. A number of factors that may have influenced the resistance pattern to insecticides as evident by the differences in responses of whiteflies from various parts of the world are compared and discussed with emphasis on agro-ecosystem, crop rotation and insecticide use patterns as well as the refugia concept.

An analysis of chemical control has shown the importance of research and development of resistance management programs' for continuing use of insecticides for whitefly control during the last decade. An evaluation of the benefit of experience gained over the last decade with this species has proven that whitefly specialists are better able to assess the resistance risks based on the ecological conditions of the agro-environment with help of new chemistries with novel modes of action. Now we have proven formulas for managing resistance based on examples from Arizona and Israel.

## Section D: Plenary Session Summary

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## Application of Genetic Diagnostics to Biological Control of Silverleaf Whitefly

Genetic technology has had a significant impact on the history and delivery of the biological control program against silverleaf whitefly. Early challenges in identification of the imported natural enemies were met with a specific genetic diagnostic, RAPD-PCR. Later taxonomic studies corroborated the genetic data. Genetic patterns assigned to natural enemy collections from all over the world set a precedent for facilitating mass rearing, quality control, ecological studies, and field evaluation of natural enemies. MPPC imported and cultured over 56 populations of Encarsia spp. and Eretmocerus spp., several of which were new species. Parasitoids in quarantine were categorized with RAPD-PCR and morphologically based systematics. Integration of the two techniques proved to be useful in capturing the maximum amount of species diversity with a minimum amount of duplication in cultures. Rearing personnel needs were reduced by 11 service years over a five-year period and insect quarantine space was used effectively. In field evaluation efforts, the two methods were integrated for identifying indigenous and imported parasitoids. Cooperators have initiated an additional technique to identify specific satellite DNA probes for development into a squash blot kit that could alleviate the cost and time constraints of RAPD assays.

*Eretmocerus* and *Encarsia* were separated into distinct groups using the morphology of the pupae and adult females. Individuals from each unique accession were

immediately characterized at the MPPC Genetics Diagnostic Laboratory using RAPD-PCR with primers C04 and A10 from Operon Technologies. Cohorts of the original parental material were sent to cooperating systematists. Information from the collaborating systematists and geneticists allowed for characterization of quarantine material while the original parental cohort was still alive. Typically, material was characterized using both methods within two to three days after acceptance into quarantine. Based on RAPD patterns, unique parasitoid accessions (possible cryptic species) were set up in pure cultures and reared on the local SLWF. The integration of the morphology-based systematics and the molecular techniques has optimized the efficient characterization of natural enemies in quarantine. The integration has allowed each new cryptic species to be fully evaluated and maintained as a pure culture while cooperating systematists continue to access the genetically unique accessions. Apart from differences in RAPD patterns, new species were only distinguishable from each other by minute differences in the first funicular antennal segment of the female. Screening with RAPD assays was really the only method available for distinguishing such cryptic species at the time of culture initiation and efficacy studies. Without RAPD assays, multiple cultures of widely distributed species, such as E. mundus would have used most of the quarantine resources.

The techniques were also equally valuable in field-testing. It was possible to test multiple species simultaneously in the field and sort out species in the recovery samples by their DNA profile. Combining the use of RAPD-PCR and classical systematics allowed for an increase in the numbers of individuals that could be characterized to species in the evaluation effort. Ultimately this led to the best possible determination of which species showed the greatest efficacy in the field. The molecular methods were only valuable when combined with classical systematics. Properly identified and curated specimens will provide the best permanent record of the species released and established. Representatives of all the accessions were cryogenically stored at MPPC and vouchered at the Texas A&M University, Department of Entomology Collection, College Station, Texas and the USDA-ARS, Systematic Entomology Laboratory, Washington, D.C.

#### Section E: Session Summary

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#### Overview of Whitefly Feeding Behavior: What We Know and What We Need to Know

#### **Mechanical Movement of the Stylets**

The mouthparts of whiteflies are typical of the order Homoptera. The mandibles and maxillae are modified as very thin and elongate stylets that pierce into the plant tissue. The paired maxillary stylets are tightly joined together along their length with an interlocking tongueand-groove coupling. The tongue-and-groove design provides a very tight coupling while allowing each maxillary stylet to move forward and back independent of one another. The paired maxillary stylets enclose a food canal and salivary canal, and their primary function is ingestion and salivation. The mandibular stylets fit on either side of the joined maxillary stylets, and enclose the maxillary stylets on their lateral aspect. The mandibular stylets are serrated at their tip, and their main function is to penetrate through the plant tissue. The maxillary stylets, with their food and salivary canals, penetrate the plant tissue along with the mandibular stylets, but it is primarily the mandibular stylets that do the mechanical work cutting and penetrating through the plant tissue.

Each stylet is operated by a pair of antagonistic muscles in the head: a protractor muscle that pushes the stylet deeper into the plant tissue, and a retractor muscle that pulls the stylet back. The four stylets can be moved independently of one another by their individual protractor and retractor muscles. The protractor and retractor muscles are short, and thus each muscle contraction can advance or retract their respective stylet only a short distance. Consequently, stylet advancement is incremental and consists of a repeating series of short distance advancements of the stylets. First, the mandibular stylets are advanced a short distance, followed by the maxillary stylets. Stylets also can be withdrawn from the plant tissue in an incremental manner similar to stylet advancement, or they can be withdrawn more rapidly by a mechanism described later. The mandibular stylets are naturally curved inward (medially) which facilitates turning of the stylets in the plant tissue. Because of their medial curvature, the left mandibular stylet tends to turn to the right in the plant tissue while the right mandibular stylet tends to turn left. If the left and right mandibular stylets advance simultaneously, their tendency to curve in opposite directions counteracts each other, and the stylets advance straight into the plant tissue. If the left and right mandibular stylets do not advance simultaneously, the stylets will turn in the plant tissue. For example, if the left mandibular stylet advances by itself, it will turn right in the plant tissue; then the right mandibular stylet advances, and is forced to turn to the right by following the pathway previously cut by the left

mandibular stylet. A turn to the left is accomplished by a similar, but vice versa manner. The stylets are capable of turning only left and right. To allow 360° of movement of the stylets, the whitefly has to pivot its body around the point of stylet insertion. This rotates the stylets in the plant tissue so that the left-right plane of the stylets is rotated into any position, thus allowing the stylets to turn in any direction. The stylets are remarkable flexible, and microscopic examination of stylets in plant tissue reveals that they twist and turn in virtually every direction, making the stylet pathway usually very tortuous.

In adult whiteflies, the labium is elongate, foursegmented, and has a longitudinal groove that encloses the stylets when the insect is not feeding. When not feeding, the stylets extend the entire length of the labial groove. During stylet penetration, the apex of the labium is pressed against the plant surface and the protractor muscles begin to advance the stylets into the plant tissue. As the stylets are advanced, the head is lowered closer to the plant surface and the labium shortens, allowing the stylets to extend past the tip of the labium and into the plant tissue. The labium does not penetrate the plant tissue. The length of the stylets in the plant tissue equals the length that the labium shortens, so the length of stylet penetration can be indirectly measured by measuring the length of labial shortening. Note that since the pathway of the stylets in the plant tissue is usually very tortuous rather than linear, the length of stylet penetration is greater than the depth of stylet penetration (depth = distance perpendicular from the plant surface to the apex of the stylets).

A long stout apophysis, called the crumena, extends internally from the apical labial segment into the head capsule. Powerful crumena protractor muscles attach to the base of the crumena in the head capsule. This allows very rapid withdrawal of the stylets from the plant tissue as follows. When the stylets are inserted into the plant tissue, the labium shortens, as described above. When the crumena protractor muscle contracts, it pushes the apical labial segment away from the head, rapidly extending the labium to its full length. The labium lengthening forces the stylets to pull out of the plant tissue and back into the labial groove.

#### Sucking Pump and Precibarial Valve

The food canal at the base of the maxillary stylets opens to a narrow canal, called the precibarium, that is enclosed anteriorly by the epipharynx and posteriorly by the hypopharynx. The precibarium widens proximally into the cibarium which also is enclosed by the epipharynx and hypopharynx. There is a valve, the precibarial valve, between the precibarium and cibarium. The pro ximal end of the cibarium opens to the anterior-most segment of the alimentary canal, the pharynx. The cibarium functions as the sucking pump. A series of muscles extends from the midline of the inner clypeal wall to the midline of the epipharyngeal wall of the cibarium and to the epipharyngeal wall of the precibarial valve which is immediately distal to the cibarium. When the muscles are at rest, the epipharyngeal wall presses tightly against the hypopharyngeal wall due to its natural elasticity. This closes the valve. Also, when the muscles are at rest, the epipharyngeal wall of the cibarium rests close to the hypopharyngeal wall (again due to natural elasticity), and the volume of the cibarium is at a minimum. When these muscles contract, the epipharyngeal wall of both the precibarial valve and the cibarium is pulled away from the hypopharyngeal wall. This has two effects. First, it opens the precibarial valve, allowing fluid to flow from the maxillary food canal into the cibarium; and second, it dilates the cibarium, creating the suction needed to pull fluid from the maxillary food canal into the cibarium. When the muscles relax, the epipharyngeal wall of the cibarium returns to its resting position, constricting the volume of the cibarium, and forcing fluid out of the cibarium. At the same time, relaxation of the muscles cause the precibarial valve to close, preventing backflow of fluid from the cibarium into the maxillary food canal. Since the precibarial valve is closed when the cibarium constricts, there is only one direction the fluid can leave the cibarium: into the pharynx.

It should be noted that the primary food source, phloem sap, is under very high turgor pressure. Consequently, suction may not be always necessary to ingest phloem sap. The high turgor pressure of the phloem sap will force sap up through the food canal as soon as the maxillary stylets penetrate a sieve element, in a manner analogous to an artesian well. In fact, if the base of the stylets are severed above the plant surface when the stylet tips are in a phloem sieve element, the turgor pressure of the phloem sap forces sap up through the severed stylets where it can be collected for chemical analysis. Consequently, ingestion rate of phloem sap may be regulated more by the precibarial valve than by the cibarial pump. The pump would play a more important role when the whiteflies occasionally ingest xylem sap (which is under negative pressure) or when the stylets get clogged with one of two sealing mechanisms in phloem sieve elements: coagulation of P-protein or deposition of callose.

#### Salivary Glands and Salivary Pump

The salivary glands and salivary pump has been described only in the adult stage of whiteflies. Adult whiteflies have two pairs of salivary gland, the primary glands and accessory glands. The ducts from each gland eventually join to form a common salivary duct that enters the salivary pump. The salivary pump functions as a syringe and connects the common salivary duct to the salivary canal enclosed by the maxillary stylets. Muscles attaching to the dorsal wall of the pump pull the dorsal wall away from the ventral wall, creating suction that draws fluid into the pump. When the muscles relax, the dorsal wall snaps back against the ventral wall (due to natural elasticity), expelling fluid from the pump. Opening and closing of valves on the afferent and efferent ends of the pump in synchrony with contraction and relaxation of the syringe muscles ensure a unidirectional flow of saliva from the salivary glands to the maxillary salivary canal.

#### Saliva

Whiteflies, as well as all other homopterans that have been studied, produce two kinds of salivary secretions: sheath saliva which gels upon secretion to form the salivary sheath surrounding the stylets in the plant tissue, and watery saliva which contains salivary enzymes and metabolites. At least in aphids, the pH of sheath saliva is slightly acidic while the pH of watery saliva is alkaline with values up to 9.

Sheath saliva. As the stylets penetrate into plant tissue, saliva is secreted intermittently from the stylet tips, and this saliva gels soon after secretion, forming the "salivary sheath" or "stylet sheath" which characteristically encases the stylets in a continuous sheath from the initial point of penetration on the plant surface to the tips of the stylets in the plant tissue. The advancing stylets produce the salivary sheath in a sequence of repeating events in synchrony with the incremental advancement of the stylets that was described previously. First, a small volume of sheath saliva, as it is called, is secreted from the stylet tips. It is initially liquid, but gels soon after secretion, encasing the stylet tips in a semi-solid matrix. The stylets then advance a few microns until the tips just break through the recently gelled sheath saliva. Then another small increment of sheath saliva is secreted from the stylet tips, encasing them again in a solid matrix. The stylets then advance a few more microns, breaking through the recently gelled sheath saliva, and the process is repeated over and over as the stylets advance deeper into the plant tissue. Each increment of sheath saliva adheres to the increment secreted previously, so the increments form a continuous sheath encasing the entire stylet length in the plant.

The incremental production of sheath saliva as the stylets advance can be detected by electrical penetration graphs (EPGs) as alternating periods of low electrical resistance, when the sheath saliva is secreted, and high electrical resistance, in between the incremental secretions. Since the rate of increments of salivary sheath production is a function of the rate of stylets advancement, we can detect different rates of stylet advancement through plant tissue by measuring the rate of resistance fluctuations in the EPGs. EPG studies have thus revealed that when silverleaf whiteflies are feeding on non-host plants or are having a first encounter with a novel host, the whitefly spends a much larger proportion of its time with the stylets stationary in the plant tissue without advancing in comparison to when it is feeding on a familiar host plant species.

The production of sheath saliva during stylet penetration appears to be characteristic of the entire order Homoptera and many Hemiptera. Studies on aphids, pentatomids, and lygaeids indicate that the salivary sheath is mainly protein and contains about 10% phospholipids and probably some conjugated carbohydrate. It is mainly hydrogen-bonded but is stabilized with disulfide linkages. The precursors of the sheath material are secreted by specialized lobes of the salivary gland. If the contents of these lobes are mixed together in vitro, the sheath material does not solidify as long as oxygen is excluded. Within the glands, the sheath precursors are maintained in solution by reducing conditions. The precursors also are mixed with amino acids that are presumed to prevent the formation of hydrogen bonds by maintaining a high dielectric constant. Once the precursors are secreted into the plant, the amino acids diffuse out, allowing formation of hydrogen bonds. Reducing components also diffuse out, allowing exposure to oxygen in the plant tissue to initiate disulfide bonding.

While the production of salivary sheaths by homopterans has been known for a very long time, the function of the salivary sheaths is uncertain. Several hypothetical functions proposed in the literature include: (1) the salivary sheath may assist in keeping the stylet bundle (the two maxillary stylets and 2 mandibular stylets) cohesive, preventing them from separating; (2) the salivary sheath may provide a relatively frictionless sheath in which the stylets can easily advance, retract, or rotate; (3) the salivary sheath may seal punctured sieve elements (the primary feeding site - see below) to prevent loss of sap which may trigger a plant defensive response that shuts down the flow of sap to damaged sieve elements; (4) the salivary sheath may provide a relatively inert barrier between the stylets and the plant tissue to minimize the plant wound response to piercing.

Stylet sheaths remain in the plant even after the stylets are withdrawn, and they stain very readily so they can be easily distinguished from the surrounding plant tissue. Consequently, for research purposes, they conveniently provide a detectable record of all places in the plant tissue where the whitefly has inserted its stylets.

*Watery saliva*. In contrast to sheath saliva, watery saliva does not gel upon secretion. Watery saliva contains salivary enzymes and metabolites. Enzymes detected in whitefly salivary glands and/or salivary secretions that presumably are from watery saliva include: amylase, invertase, and alkaline phosphatase. The role of these enzymes in the feeding biology is unknown. At one time, the presence of amylase had been the basis of a hypothesis that whiteflies sometimes feed on mesophyll cells, but more recent evidence indicates that whiteflies do not obtain significant nutrition from mesophyll cells.

#### Stylet Pathway in the Plant

The target site of stylet penetration for whiteflies is the sap conductive cells of the phloem, the sieve elements. The pathway of the stylets from the plant surface to the sieve element is primarily intercellular, and the stylets weave their way between and around plant cells prior to puncturing a sieve element. Their great flexibility and ability to twist and turn facilitate their tortuous pathway between and around cells. There is conflicting evidence of whether the pathway of the stylets through the epidermis is directly through the epidermal cells or between adjacent cells; but once past the epidermis, evidence from different laboratories is consistent that the stylet pathway is primarily intercellular through the mesophyll.

The nature of the physical contact between the stylets and the punctured sieve element has not been reported for whiteflies. However, the physical contact between the stylets and the punctured sieve elements has been examined with transmission electron microscopy for aphids, and until proven otherwise, the most parsimonious assumption is that whiteflies behave similarly. In aphids, the mandibular stylets stop at the cell wall of the sieve element, and only the maxillary stylets penetrate a short distance into the sieve element. Sheath saliva also stops at the cell wall and does not extend into the sieve element.

Occasionally, during the primarily intercellular stylet penetration, the stylets penetrate into cells, a phenomenon that can be readily detected in EPG recordings. These intracellular penetrations are usually very brief (< 30 s), and the whitefly then withdraws its stylets from the cell (or pushes them all the way through) back into intercellular space. Aphids also have a primarily intercellular stylet pathway and make brief intracellular punctures along the way from the plant surface to the sieve elements. However, there are some interesting quantitative and qualitative differences between the brief intracellular punctures of aphids and whiteflies. Most aphids that have been studied produce many brief intercellular punctures along the way to the sieve elements, sometimes up to 50 or more between the plant surface and the sieve element. Whiteflies produce far fewer; on average only 3-6 between the plant surface and the sieve element. Most aphids also begin making brief intracellular punctures almost as soon as stylet penetration begins, frequently within the first 30 s of stylet penetration. Whiteflies, on the other hand, do not make intercellular punctures until much later in the probe, generally 8 - 30 min after initiation of stylet penetration. Also, EPG recordings strongly suggest that during the brief intracellular punctures, the behavior of the insect differs between aphids and whiteflies. These differences between whiteflies and aphids likely explain why nonpersistent viruses are transmitted primarily by aphids and very few are transmitted by whiteflies (of 211 known non-persistent viruses, 208 are transmitted by aphids and only 3 are transmitted by whiteflies). Non-persistent viruses are transmitted during brief intracellular punctures, especially intracellular punctures produced early in the probe; thus the production of relatively few intracellular punctures by whiteflies, and the virtual absence of intracellular punctures early during probes, are not conducive to transmission of non-persistent viruses by whiteflies. Furthermore, EPGs detect specific behaviors

during brief intracellular punctures by aphids that are associated with inoculation and acquisition of nonpersistent viruses, and these behaviors are not detectable in EPGs of whiteflies.

The differences between brief intracellular punctures of aphids versus whiteflies suggests different functions for intracellular punctures in these two families of phloemfeeders. As noted previously, aphids begin making intracellular punctures very early in a probe (usually within the first 30 s) while whiteflies make brief intracellular punctures only late during probes. If the function of the brief intracellular punctures is to sample cell contents to discriminate host from nonhost plants, it would be logical that the intracellular punctures should occur early during probing rather than later after considerable time and energy has been expended penetrating the plant tissue. Thus, I hypothesize that one of the functions of intracellular punctures for aphids but not for whiteflies is host discrimination. Whitefly intracellular punctures generally occur shortly before a phloem sieve element is penetrated and phloem sap ingestion occurs. Thus, I further hypothesize that in whiteflies, brief intracellular punctures represent trialand-error attempts to locate sieve elements once the stylets are deep within the plant tissue. This may or may not be an additional function of brief intracellular punctures in aphids. These hypotheses need testing.

#### Sensory Organs Involved in Feeding

Whiteflies have several olfactory receptors on their antennae, but their role in host selection and feeding is unknown. There are only a few experiments reported in the literature to determine if whiteflies are attracted to host plants by their odor. There is no strong evidence for olfactory attraction to host plants. Two papers report some degree of attraction to host plant odor, but the methodology and analysis were so poorly described, that the reader has no way of evaluating their validity. The fact that both male and female whitefly antennae are well equipped with olfactory organs suggest an important role of odor detection in their biology. While there is evidence that greenhouse whitefly females produce a pheromone that attracts males, the presence of welldeveloped olfactory organs on both sexes, as well as in parthenogenic species like Parabemisia myricae, make it tempting to deduce that the olfactory organs also may be used to detect host plant odor. This is a hypothesis that needs future investigation.

As do many homopterans, whiteflies generally rub the tip of their labium on the plant surface prior to inserting their stylets. Whiteflies have seven pairs of sensory organs on the tip of their labium, four of which are mechanoreceptors and three of which are compound mechano/chemoreceptors. In aphids, which have only mechanoreceptors at the tip of their labia, the mechanoreceptors are thought to assist the aphids in locating intercellular grooves on the plant surface to

facilitate stylet penetration between adjacent epidermal cells. Whiteflies also tend to initiate probes in intercellular grooves, so the function of the mechanoreceptors at the tip of the labium of whiteflies may be the same as that postulated for aphids. The role of the chemoreceptors at the tip of the whitefly labium is unknown. Several possible functions are postulated. They may detect relevant chemicals on the plant surface simply by rubbing the tip of the labium over the plant surface or by salivating on the plant surface and pressing the tip of the labium into the saliva where the chemoreceptors could detect plant surface chemical dissolved in the saliva. Alternatively, it is possible that the apical labial chemoreceptors detect chemicals *inside* the plant tissue. While the labium never penetrates the plant surface, it is still at least theoretically possible that the apical chemore ceptors can detect internal fluids in the plant. As discussed previously, whiteflies produce a salivary sheath that extends into the plant as the insect advances its stylets. The salivary sheath actual begins on the plant surface, and during stylet penetration, the tip of the labium with its chemoreceptors is pressed into the salivary sheath on the plant surface. Since the sheath is continuous from the surface to the internal tissues of the plant, it is possible that the apical labial chemoreceptors could detect internal plant chemicals that diffuse up the salivary sheath and come in contact with the chemoreceptors pressed into the surface part of the salivary sheath. Research is needed to test these hypotheses and determine the role of the apical labial chemoreceptors in feeding and host selection.

Despite the known chemoreceptors on the tip of the labium and the well documented behavior of rubbing the tip of the labium on the plant surface prior to inserting the stylets, it is not known if whiteflies taste the surface of the plant and whether or not plant surface chemistry plays a role in host selection by whiteflies. There is some circumstantial evidence that plant surface cues are used by whiteflies: *Parabemisia myricae* discriminates between suitable and unsuitable leaves prior to probing. While plant surface chemistry is an obvious candidate as an explanation for this pre-probing discrimination, other explanations can be postulated. Research is needed to determine what, if any, role plant surface chemistry plays in whitefly host selection and feeding behavior.

The mandibular stylets of whiteflies are hollow through most of their length and each mandibular stylet is traversed by a pair of dendrites. These are believed to be mechanosensory and provide the whitefly with sensory feedback about the movement of the stylets. The maxillary stylets are not associated with any dendrites.

The precibarium, formed by the opposing hypopharynx and the epipharyngeal wall of the labrum provides the pathway of fluid from the maxillary food canal to the cibarial pump. The precibarial valve, as described previously, regulates the flow of fluid through the precibarium. Close to the precibarial valve are 10 chemoreceptors distal to the valve and 8 chemoreceptors proximal to the valve. These sensillae presumably taste the ingested fluid passing through the precibarium. The role of these sensillae in feeding and host selection are not known, but their location and the lack of any other known chemoreceptors in the stylets or anterior alimentary canal appear to make them the primary taste organs for ingested fluid. Thus, they probably are used to taste plant sap as it is being ingested. At least in some hemipterans, they apparently are also used to taste the plant surface as follows: the insect secretes a small amount of saliva on the plant surface which dissolves surface chemicals, and then ingests the saliva where the precibarial sensillae can detect the dissolved plant surface chemicals. Whether or not this occurs in whiteflies is unknown.

#### Sieve Element Contact and Feeding

The primary feeding site of whitefly adults and nymphs is the phloem sieve element. However, fewer than 10% of probes by whiteflies succeed in reaching a phloem sieve element. This suggests that locating a sieve element during a probe is not an easy task. How whiteflies and other phloem feeders locate the sieve elements has intrigued researchers for nearly a century, but there is still no conclusive data indicating how they accomplish this. Perhaps it may be simply a trial and error process, randomly piercing cells after the stylets are deep in the plant tissue, as hypothesized previously. The time from the beginning of a successful probe to initial penetration of a sieve element has been examined for several combinations of whitefly species and plant species. The average times for these different combinations range from 16 - 66 min.

Once a sieve element is pierced, whether this is the result of trial and error or result of a more direct method, sieve elements have several physiochemical characteristics that could provide the insect with the information that a sieve element rather than a non-sieve element has been penetrated. These characteristics include very high turgor pressure, very high sugar concentration, and slightly alkaline pH. Which, if any, of theses cues is used by whiteflies or other phloem feeders to identify sieve elements is not known for certain. However, the phagostimulatory effect of sucrose in artificial diets fed to whiteflies and other phloem-feeders suggests that the high sugar concentration of sieve elements may be at least one cue used by the insects to identify them.

When an adult whitefly first penetrates a sieve element, it begins salivating into the sieve element. The salivary phase lasts several minutes, and it is during the salivary phase that persistent viruses, such as geminiviruses, are inoculated. After the salivary phase, the whitefly enters an ingestion phase where phloem sap in ingested. The salivary phase may appear again interspersed with periods of ingestion. The function of the salivary phase is unknown. After a variable period of "phloem phase" (the combined term for salivary + ingestion phase), which for *Bemisia* adults generally lasts several minutes to several hours, the whitefly withdraws its stylets from the sieve element, terminating phloem phase, and may continue probing and eventually initiate another phloem phase, or it may terminate the probe altogether. Whitefly nymphs, on the other hand, tend to stay in the same sieve element for much longer periods of time (measured in days) and have very regular alternating periods of salivation phase is unknown but it may be involved in conditioning the sieve elements for continued ingestion, and very likely is when the whitefly nymph introduces factors responsible for the systemic physiological disorders inflicted on some host plants by silverleaf whitefly nymphs.

#### Ingestion from Non-Phloem Tissues

As noted previously, the presence of amylase had been the basis of a hypothesis that whiteflies sometimes feed on mesophyll cells, but this hypothesis has recently been rejected. EPG studies suggest that whiteflies penetrate cells other than sieve elements, but these intracellular penetrations are too brief (usually < 30 s) to be a significant source of ingested nutrients. It is likely that the intracellular punctures serve to sample plant sap with their precibarial sensillae either to aid in host discrimination or to locate sieve elements.

Whiteflies (at least adult whiteflies) occasionally ingest xylem sap, but this is much, much less frequent than phloem sap ingestion. Xylem sap is extremely dilute with very low concentrations of nutrients. This, combined with the infrequency of xylem sap ingestion, indicates that xylem sap is not a significant source of nutrition for whiteflies. Then why ingest xylem sap? Evidence in aphids, which also are primarily phloem feeders that occasionally ingest xylem sap, indicates that xylem sap ingestion is the equivalent of drinking water, and occurs much more frequently if the aphid is dehydrated. Phloem sap, with its very high osmolarity, would not be ideal for addressing a water deficit in the insect.

#### **Critical Questions Needing Answers**

In addition to the questions posed in the above text, the following are some important questions. Do whiteflies get gustatory information from the plant surface? Do whiteflies get gustatory information from the apoplast (intercellular space and fluid) during intercellular penetration? What behaviors occur during brief intercellular punctures? How is host acceptance/rejection decided? What is the composition of whitefly saliva? Does the composition of saliva differ between nymphs and adults? Does the composition of saliva differ during different phases of feeding? What are the roles of the different salivary components? How do whiteflies circumvent phloem sealing responses?

#### Section F: Plenary Session Summary

Steve Castle USDA-ARS, Western Cotton Research Laboratory, Phoenix, AZ

## Outbreak Occurrences: Factors that Contribute and Tactics that Suppress

The status of *Bemisia tabaci* as a major pest species of agriculture throughout the world has intensified in recent decades. With its expansion into previously uninfested production systems and increased activity in regions where it has long been established, significant losses have occurred on a global scale irrespective of pest management capabilities. An unprecedented interest in *B. tabaci* biology and management has arisen in recent years in response to the serious challenges posed to agriculture.

Explanations for the upsurge in *B. tabaci* outbreaks have often centered on failures of management. In part icular, insecticides have been extensively incriminated for causing large-scale disruptions in control. For example, in the Sudan Gezira during the late 1970s, destructive outbreaks of *B. tabaci* were attributed either to the direct effects of insecticide use, i.e. resistance development and fertility-stimulating effects of hormoligosis, or the indirect effects of insecticide use, i.e. depredations of natural enemy populations. Thereafter, anywhere in the world that a *B. tabaci* outbreak occurred, an insecticidebased explanation would generally be a central element of the post-outbreak head scratching.

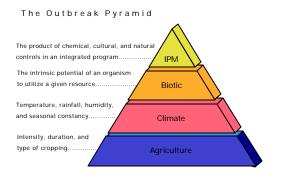
While many regions in the world have been impacted by severe infestations of *B. tabaci*, these represent only the latest in a series of *B. tabaci* outbreaks world-wide dating back to the 1920s. Accounts of historical *B. tabaci* infestations provide an essential perspective on present day outbreaks by demonstrating serious pest potential under a range of agricultural and management conditions. The set of observations, historical and modern, allows for a broad consideration of factors that determine the size of *B. tabaci* populations in agricultural systems. When contrasted to agricultural regions with only endemic populations of *B. tabaci*, an even more complete picture of manifold factors becomes possible.

Two approaches have been taken for comparing *B. tabaci* infestations in various systems. One has been the comparison of modern outbreaks with historical outbreaks by examining published accounts. Historical documentation of *B. tabaci* outbreaks prior to 1940 establish their capacity as serious agricultural pests prior to the advent of synthetic organic insecticides. The second approach has been to examine outbreak vs. non-outbreak regions in present times to deduce principle factors contributing to outbreaks. *B. tabaci* occurs in agricultural zones around the world, but does not reach outbreak status in all. It is instructive to examine various

regions where infestations of varying magnitude occur and contrast their physical and cultural features for clues concerning the central factors governing infestation levels. The diversity of California agriculture including cropping variations and climatic differences has served well for deducing important factors contributing to B. *tabaci* populations. In particular, comparisons among Imperial, Coachella, and San Joaquin Valleys have helped to illustrate the role of temperature and cropping system in the buildup, or lack thereof, of *B. tabaci* populations. Although other elements of *B. tabaci* population dynamics are clearly recognized, there has been no synthesis of the fundamental factors that contribute to *B*. *tabaci* outbreaks. The Outbreak Pyramid represents a conceptual synthesis of 4 general factors that determine the size of *B. tabaci* infestations. The foundation of the Outbreak Pyramid is represented by the Agriculture layer, which forms the resource base subject to exploitation by whiteflies and other pest populations. The quality of the Agriculture layer from a resource standpoint depends on the types of crops grown, the area over which they are grown, or intensity, as well as the length of season, or duration, that they are available for exploitation. The next layer of the pyramid is represented by Climate that stands as the most unalterable component. The Climate layer will dictate to some degree the composition of the Agriculture layer, but only in defining the limits of what can be grown. The particular character of agriculture in any specific region will be more a product of market forces, historical precedent, and physico-environmental limitations such as water availability and soil characteristics. But whatever the composition of the Agriculture layer, climate will exert a major influence on the dynamics of insect populations. Climate factors such as temperature, rainfall, and humidity impinge on many facets of the ecology of *B. tabaci* including growth rates, survival or demise on a host plant during intense weather, and dispersal between crops. In particular, temperature is a driving force in generation time and how fast *B. tabaci* populations grow, whereas low rainfall favors greater survivorship. The Biotic layer represents the intrinsic capacity of *B. tabaci* to exploit the resources available as determined by the agricultural makeup and within the constraints of the physical environment as defined by the climate. At the pinnacle of the pyramid is the management or IPM layer, representing the product of chemical, cultural and natural controls, and is the layer subject to the most manipulation by humans.

The essence of the Outbreak Pyramid is that in circumstances where the basal 2 layers of the pyramid are "broad" with agro-ecological potential, the biotic characteristics of *B. tabaci* permit exploitation of the resource base in excess of the various types of control, both natural and manipulated. Conceptually, the volume of the IPM layer at the top of the pyramid is dwarfed in comparison to the combined volume of the three layers that form the pyramid base. In practice, management efforts that are targeted at individual fields, even if they

provide superior results, have been overwhelmed by region-wide population increases of *B. tabaci* on crop and non-crop vegetation. It is not a failure of management per se to control regional populations of *B. tabaci*, but rather a function of the combined potential contained within the climate, agriculture and biotic levels of the pyramid that eventually exceed the capacity of local crop protection.



Regions with year-round crop production such as the desert southwest USA have experienced incremental buildups of B. tabaci populations on a succession of crops. Management of whitefly infestations within crops grown early in the annual cycle has often proved adequate through harvest time. But with each crop transition, and with an increasingly favorable environment as summer temperatures arrive, whiteflies break away from local control and eventually saturate an entire region. In some instances, as crops near harvest, local field controls are relaxed because of the access required for hand picking. After the harvest, there is often considerable delay in plowing the field that serves only to generate large numbers of whiteflies ready to disperse to neighboring crops. These are examples of vulnerability in crop production that whiteflies rapidly exploit. The tools of management cannot always be used fight back whitefly onslaughts because of regulatory issues, e.g. pre-harvest intervals, as well as economic issues such as the cost of insecticide applications on low value crops. The broad polyphagy of *B. tabaci* allows for field, vegetable, and tree crops to all serve as hosts. Moreover, some regions harbor a large biomass of non-crop hosts in the form of both wild and ornamental hosts, and these often contribute significantly to whitefly numbers.

In regions where *B. tabaci* outbreaks have occurred chronically, modifications targeted at 1 or more levels of the Outbreak Pyramid have been pivotal in tempering further outbreaks. Changes can only be made at the Agriculture level or at the IPM level as the macro climate and the intrinsic biotic characteristics of the organism are unmanipulable. The IPM level is the most flexible level in terms of modifying control practices or instituting new information-gathering techniques for improving the decision-making process. It is the level that must be expanded in volume in order to be able to suppress the explosive potential represented by the expansion of the lower 3 levels. Expanded IPM happens through the development of sampling programs, determination of action thresholds, conservation of natural enemies, and the integration of chemical, cultural and natural controls that becomes possible through research and information gathering. When true integration of control tactics occurs as opposed to haphazard and uncoordinated implementation, the result can be spectacularly successful as it has been in Arizona from 1996 to the present. Expanded IPM in Arizona and California has also occurred with the arrival of superior insecticides that target multiple stages of *B. tabaci*, are residually active, and have relatively low impact on natural enemy populations. Modification of the Agriculture layer has played an important role in curbing *B. tabaci* outbreaks in California's Imperial Valley. Relatively few acres of fall melons are grown now compared to pre-1991 levels, and spring melon and fall vegetable acreages have also dropped. The Agriculture layer of the Outbreak Pyramid has contracted in the Imperial Valley, and in combination with the expansion of the IPM layer, B. tabaci outbreaks have been brought under control.

### III. Reports of Research Progress – 2001 & 2002

#### **Reports of Research Progress**

#### Section A: Biology, Ecology, and Population Dynamics Co-Chairs: Jackie Blackmer and David Byrne

Investigator's Name(s): Ma. del Rosario Avila, P. Cano, U. Nava and E. López

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

Dates Covered by Report: 2000

#### Identification of the Whitefly Species and their Plant Hosts in the Comarca Lagunera, Mexico, 2000

In the Comarca Lagunera area from 1994 to 1996, three species of whiteflies were identified, which were: sweet potato whitefly (SPWF), *Bemisia tabaci* Gennadius, the silverleaf whitefly (SLWF), *Bemisia argentifolii* Bellows & Perring and the greenhouse whitefly, *Trialeurodes vaporariorum* Westwood. During 1997 four additional whitefly species were identified: the banded wing whitefly, *Trialeurodes abutilonea* Haldeman, the wooly whitefly, *Aleurothrixus floccosus* Maskell, the acacia whitefly, *Tetraleurodes acaciae* Quaintance and the citrus blackfly, *Aleurocanthus woglumi* Ashby.

At the beginning of the sampling on the cultivated fields during 1994 the whitefly species identified were: the SPWF *B*. *tabaci* and the SLWF *B*. *argentifolii*. However, the more recent (1996-97) whitefly samples indicated that dominant species in the cultivated fields was *B*. *argentifolii* and that the *B*. *tabaci* species has been practically displaced.

Additionally, 108 plant species were identified as SLWF, *B. argentifolii* plant hosts. The main crop hosts were: *Brassica oleracea* L. vars. *botrytis* and *capitata*, *Cucumis melo* L., *Cucumis sativus* L., *Cucurbita pepo* L., *Citrullus lanatus* (Thunb.) Mansf. and *Gossypium hirsutum* L. It was found that ten weed had high SLWF infestation, but, because of their higher distribution in the crop fields the most important ones were: *Convolvulus arvensi* L., *Flaveria trinervia* (Spreng.) Mohr, *Ipomoea spp*, *Solanum elaeagnifolium* Cav. and *Xanthium strumarium* L. In the urban areas 38.7% of the plant hosts had from medium to high SLWF infestation. The plant species with the highest SLWF infestation were: *Acacia berlandieri* Benth., *Acalypha wilkesiana* Muell., *Althaea rosea* Cav., *Bahuinia divaricata* L., *Camellia japonica* L., *Sedum spp*, *Ipomoea spp*. *Jasminum spp*, *Lantana camara* L. and *Oenothera spp*.

Investigator's Name(s): Jacquelyn L. Blackmer<sup>1</sup> and Dale Cross<sup>2</sup>

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

#### Dates Covered by the Report: 2001

## Response of *Eretmocerus eremicus* (Hymenoptera: Aphelinidae) to Skylight and Plant Cues in a Vertical Flight Chamber

In the southwestern United States, Eretmocerus eremicus (Hymenoptera: Aphelinidae) is a native parasitoid of the Bemisia complex (Homoptera: Aleyrodidae). However, little information currently exists on its potential as a biological control agent of whiteflies or on the factors that influence its tendency to disperse. Information on this parasitoid's dispersal propensity might alter the release strategy employed. Low dispersal rates could mean that multiple release sites might be necessary for optimal distributions relative to its host. Our study examined the host-habitat/host location phase(s) of host-finding behavior for E. eremicus. In this study, we examined the flight behavior of male and female E. eremicus in response to skylight (here simulated by a mercury-vapor lamp) and plant cues (a 550-nm filter) in a vertical flight chamber. The visual plant cue was presented to the parasitoid in the absence of whitefly stimuli. Approximately 90% of the parasitoids took off in response to the skylight cue. Both sexes were capable of sustained flights in excess of 60 min; however, average flight durations were  $15.6 \pm 3.8$  min for males and  $7.8 \pm 2.6$  min for females. When a plant cue was presented during the parasitoid's phototactic flight, four relatively distinct responses were observed. Fifty-one percent of the individuals responded to the plant cue throughout their flight by flying toward or by landing on the cue. The majority of these parasitoids were females. Approximately 12% of the wasps exhibited an intermittent, positive response to the plant cue. Twenty percent exhibited a 'migratory' response. These parasitoids, which were predominantly males, failed to respond to the plant cue until they had flown for a considerable period. Finally, 17% failed to respond to the target during their flight. Approximately 37% of the individuals that showed a positive response to the plant cue actually landed on it and the majority of these were female. However, in the absence of additional whitefly stimuli, tenure time on the visual plant cue was relatively short. The differential response to the plant cue by male and female parasitoids could be, in part, because females are driven to locate hosts in which to oviposit, and males are driven to find mates. Additional studies need to determine how biotic and abiotic factors influence the dispersal tendencies of *E. eremicus*, the role of visual and olfactory information in the host-finding process, and whether the vertical flight chamber might be a useful tool for screening the flight propensity of potential biological control agents.

## Investigator's Name(s): James S. Buckner and Marcia M. Hagen

Affiliation & Location: USDA-ARS, Insect Genetics and Biochemistry Research Unit, Red River Valley Agricultural Research Center, Fargo, ND

Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

### Dates Covered by the Report: 2001

## The Triacylglycerol Composition of Silverleaf Whitefly Adults

*Bemisia argentifolii* Bellows and Perring (Homoptera: Aleyrodidae) adults were collected from a whitefly colony maintained on hibiscus plants, weighed and stored at -80°C. Groups of adults (30-50 mg) were placed into a 20-ml glass vial containing 8-10 ml of CHCl<sub>3</sub>/methanol (2:1) and covered with a Teflon-lined cap. To homogenize (sonicate) the tissues, the vial was placed in the water bath of a Fisher Scientific, Model FS 60, ultrasonic cleaner for 15-30 min. The homogenate was filtered through glass wool with CHCl<sub>3</sub> and the CHCl<sub>3</sub> washed with water. The lipid extract in CHCl<sub>3</sub> was spotted on 250-µm silica gel TLC plates and the triacylglycerol (TAG) fraction was separated from other lipid fractions by developing the TLC plates in hexane/ethyl ether/formic acid (80:20:1). The purified TAG fraction was visualized with iodine vapors, scraped from the TLC plates and eluted from the silica gel with CHCl<sub>3</sub>.

High performance liquid chromatography (HPLC) was used to separate the TLC-purified mixture of TAG using a ChromSpher 5 Lipid column (Chrompack, 5 µm, 4.6 X 250 mm). Mass detection of TAG constituents was performed using a Sedex (Model 55) evaporative light scattering detector (ELSD). Peak areas from the ELSD were used to determine quantities of resolved TAG components and structural identifications were made by HPLC-MS. Individual TAG components were identified by comparing their column retention times to those of authentic TAG standards and by comparative mass spectral analysis using the Waters Model 2690 Separations Module linked to a Waters Thermabeam Mass Detector.

For the mixture of triacylglycerols from *B. argentifolii* adults, the major fatty acid constituents were identified as oleic acid (18:1), palmitic acid (16:0), stearic acid (18:0) and linoleic acid (18:2), in descending order of abundance. Lesser amounts of palmitoleic acid (16:1) and arachidonic acid (20:0) were detected, as well as trace amounts of myristic acid (14:0). The major intact TAG in decreasing order of abundance, as analyzed by HPLC-MS, were 1-palmitoyl-2,3-dioleolylglycerol (POO), 1,2-dipalmitoyl-3-oleoylglycerol (PPO), 1-palmitoyl-2-stearoyl-3-oleoylglycerol (PSO), 1,2,3-trioleoylglycerol (OOO), 1-stearoyl-2,3-dioleoylglycerol (SOO), 1-palmitoyl-2-oleoyl-3-linoleoylglycerol (POL) and 1-palmitoyl-2-linoleoyl-3-stearoylglycerol (PLS). The fatty acids of the TAG from *B. argentifolii* adults were also analyzed as their methyl ester derivatives following hydrolysis with 5% KOH in methanol and subsequent reaction of the free fatty acids with 10% HCl in methanol to form methyl esters. The distribution of fatty acids was similar to that indicated above for intact TAG: approximately 58% as 18:1, 29% as 16:0, and lesser amounts of 18:0, 18:2, 18:3 and 16:1. These methods developed for characterization of TAG from adults will be used to determine the TAG composition of *B. argentifolii* nymphs and to provide useful information in regard to the lipid nutrient reward for whitefly predators and parasitoids.

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Affiliation & Location: USDA-ARS, Insect Genetics and Biochemistry Research Unit, Red River Valley Agricultural Research Center, Fargo, ND

Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

### Dates Covered by the Report: 2001

#### The Fatty Acid Composition of the Internal Lipids from Silverleaf Whitefly Nymphs and Adults

The cuticular lipids of *Bemisia argentifolii* Bellows and Perring (Homoptera: Aleyrodidae) nymphs (1) and adults (2, 3) have been characterized, but the identification and quantification of the internal lipids of whiteflies have not been determined. The assessment of the internal lipids as a source of nutrient reward is especially relevant to specific parasitoids of whiteflies since they consume their host from within. The identification and composition of the fatty acids associated with the major lipid classes (triacylglycerols, free fatty acids and phospholipids) within *B. argentifolii* nymphs were determined. Comparisons were made to the fatty acids present in the internal lipids of adult *B. argentifolii*.

Feeding silverleaf whitefly nymphs were raised on young cantaloupe plants and adult whiteflies were maintained on hibiscus. Groups of 200-300 third and fourth instar nymphs were carefully removed from leaves, placed in glass vials, and their surface lipids were removed and discarded using hexane. The surface lipids from groups of pre-chilled adults were also removed with hexane. The internal lipids were then extracted by sonication for 45 min in 7-8 ml of chloroform/MeOH (2:1). The chloroform/MeOH homogenate was filtered through glass wool and partitioned twice between chloroform and water. The internal lipids of whiteflies were separated into neutral lipids, free fatty acids and phospholipids using a column of 80-180 mg of Porasil Prep silica (Waters Corp., Milford, MA). The separation was achieved using a modification of a novel solvent system containing tertiary butylmethylether (t-BME) (4). Lipid samples were applied to the column in hexane and lipid fractions eluted using the following solvents: hydrocarbons, wax esters, aldehydes (99.5:0.5 hexane/t-BME); triacylglycerols (TAG), alcohols (96:4 hexane/t-BME); free fatty acids (FFA) (100:2 hexane/acetic acid); phosphatidylethanolamine (PE) (20:4:1 t-BME/MeOH/0.001M ammonium acetate, pH 8.6) and phosphatidylcholine (PC) (5:4:1 t-BME/MeOH/0.001M ammonium acetate, pH 8.6). The purity of lipid fractions was verified by silica HPTLC. Fatty acid methyl esters (FAME) were formed by alkaline hydrolysis of the triacylglycerol and phospholipid fractions followed by treatment of the free fatty acids with methanolic HCl. The FAME derivatives of fatty acids were separated by capillary gas chromatography (CGC) and analyzed using splitless injection at 200°C, a Supelco SP-2380 capillary column (30m x 0.25mm id.) in a Hewlett-Packard Model 5890 GC. The quantities of FAME were determined by converting integrated peak area data from the FID response using nonlinear regression slope data for increasing amounts (0.78-200 ng) of the authentic methyl esters of 14:0, 16:0, 16:1, 18:0, 18:1, 18:2, 18:3 and 20:0 fatty acids.

All lipid classes contained variable distributions of 8 fatty acids: the saturated fatty acids, myristic acid (14:0), palmitic acid (16:0), stearic acid (18:0), arachidonic acid (20:0); the monounsaturated fatty acids, palmitoleic acid (16:1), oleic acid (18:1); the polyunsaturated fatty acids, linoleic acid (18:2), linolenic acid (18:3). Fourth instar nymphs had 5-10 times the quantities of fatty acids as compared to third instar nymphs and 1-3 times the quantities from adults. The quantity differences of fatty acids between fourth and third instar nymphs related to their size and weight differences: fresh weights of 19-28 mg/nymph and 5-9 mg/nymph, respectively. For nymphs and adults, TAG lipids were the major source of fatty acids, with oleic (18:1) and palmitic (16:0) acids as major components. For nymphs and adults, the majority of the polyunsaturated fatty acids, linoleic (18:2) and linolenic (18:3) were present in the phospholipid fractions (PE + PC).

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

Dates Covered by the Report: November 2000-December 2001

## Seasonal Ecology of *Bemisia tabaci* in Arizona: Low Temperature and Host Plant Effects on Field Populations and Associated Mortality Factors

The current ongoing study has examined seasonality and mortality patterns of *B. tabaci* on different hosts during the year. Plots of six representative host plants (broccoli, cantaloupe, cotton, alfalfa, Lantana and various weeds) were established at the Yuma, Maricopa and Marana Agricultural Centers in Arizona. These sites represent the range of geographic and climatic areas of the state for cotton production. Here we report findings from the first year of the study. Two host plants, cantaloupe and Lantana, were observed to provide a good environment for the buildup of B. tabaci populations during the fall. B. tabaci populations declined steadily with decreasing temperatures during the winter. Low temperatures affected the host plants, with Lantana plants losing all foliage due to freezing temperatures. However, a significant number of Lantana plants produced new foliage during the spring and summer. After the decline during the winter, populations survived in broccoli and Malva parviflora (cheeseweed), and built up in broccoli, cantaloupe and Lantana. Populations of B. tabaci were extremely high in Lantana and cantaloupe, which allowed for significant densities in cotton once this crop was established. In addition, during the middle of the summer, ground cherry plants hosted large densities of *B. tabaci*. The prevalence of different weed species through the year also allowed for increased populations. From the three different regions, Yuma showed earlier increases in B. tabaci populations, followed by Maricopa and Marana. Yuma populations were in general smaller than those of Maricopa and Marana. B. tabaci populations in alfalfa were low at all sites. Life table analyses demonstrated differences in survivorship patterns on the different hosts. In general, the highest survivorship was observed in cantaloupe at all sites. The highest B. tabaci survival at Marana occurred in cantaloupe (about 47%) followed by Lantana (15%), weeds (15%) cotton (4%), alfalfa (3%) and broccoli (2%). Yuma had the highest survival in cantaloupe (45%) followed by broccoli (31%), Lantana (19%), weeds (17%) and cotton (7%). Maricopa had the highest survival in cantaloupe (45%) followed by broccoli (45%), Lantana (33%), alfalfa (28%), weeds (15%) and cotton (4%). From the different mortality factors observed, both predation and dislodgment accounted for a significant portion of the mortality at all locations. Dislodgment and desiccation were important in some host plants and probably were influenced by low temperatures. For instance, broccoli cohorts suffered high mortality due to desiccation and dislodgment. Rates of predation were relatively high in all host plants but more so in cotton across the different locations. Parasitism by Encarsia and Eretmocerus varied from host to host and from location to location. Parasitism was highest in Lantana, especially at Maricopa (32%). Despite high levels of B. tabaci mortality at the three locations, it was insufficient to prevent population outbreaks in most plant hosts. The examination of the biodemographic characteristics of *B. tabaci* during the year, especially during the winter months will provide important information relative to the cold hardiness and survival of this species. Such information will aid the prediction, and possibly the prevention, of outbreak populations in the late spring and summer

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## Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

### Dates Covered by the Report: 2000-2001

#### Differential Host Acceptance of Cotton and Melon by Bemisia tabaci

A series of greenhouse experiments were conducted to evaluate host acceptance of melons relative to cotton by *Bemisia tabaci*. The objectives were to quantify the differences between melons and cotton in terms of the number of adult whiteflies that settled on either plant, then determine the levels of oviposition that occurred over the course of the experiment. Two types of choice assays were used to examine adult settlement and oviposition. For the first, acetate cylinders 0.6 m high by 0.28 m diameter were fashioned with 3 equidistant slots cut into the cylinder wall at a height of 0.46 m, large enough for fully expanded melon and cotton leaves to be inserted into the interior space of each cylinder. The tops of the cylinder interiors remained attached to their plants. A foam-rubber collar was sandwiched around each leaf petiole and then snugly fit into the rectangular slots in the cylinder walls so that no whiteflies would be able to escape from the interiors. Once all cylinders were collected into an aspiration tube, then transferred through a sleeve opening to be released inside each cylinder cage. As many as 18 cylinders were set up at one time and infested with adult whiteflies. A total of 10 cylinder cage experiments were conducted.

For the  $2^{nd}$  type of assays, experiments were conducted in 2 small greenhouses with 16 plants of each type, melon and cotton, arranged into a randomized complete block design with 4 blocks consisting of 8 plants each. The 4 blocks were arranged on a bench on one side of each greenhouse. Whiteflies were collected separately from cotton and melon cultures. Approximately 1200 adults were released into each of the 2 greenhouses. Leaf counts of adults were made over the next 2.5 days, morning and late afternoon. Following the 5<sup>th</sup> and final count, all leaves were collected from each test plant for egg counts. Whitefly adult and egg counts were expressed as the mean number (±SEM) of whiteflies per leaf at each leaf node.

In both the cylinder cage and greenhouse experiments, *Bemisia tabaci* consistently demonstrated a much higher affinity for melons compared to cotton. Adult settling and oviposition rates were greater on melon in the greenhouse experiments that used whole plants and for which whiteflies were free to move from plant to plant. A similar pattern was observed in the cylinder cage tests, but the differential in adult settling and oviposition between melons and cotton, although highly significant, was not as great as the greenhouse test. Overall, a ratio of 2:1 or greater was observed for mean numbers of adults on the melon leaf relative to either one of the two cotton leaves. In some cases, i.e. experiment 1, the differential was approximately 5:1 in favor of melon leaves. The relative number of whitefly eggs on either host tended to follow the same pattern observed for adults.

The differential between cotton and melon plants was more pronounced in the 2 greenhouse experiments in terms of mean whitefly adult and egg densities. The mean numbers of whitefly adults on cotton leaves compared to melon leaves ranged between 8-31-fold greater on melon leaves. Egg densities on melon leaves exceeded those on cotton leaves between 8-56-fold. There was little difference in results with respect to the origin of the whiteflies used in the greenhouse experiments. However, there was a marked tendency for more eggs overall to be deposited on leaves if the whiteflies originated from the melon colony.

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

Dates Covered by the Report: 1998-2000

# Modification of CC whitefly traps

Modifications of CC whitefly traps are in progress to improve their potential for adult whitefly control in greenhouses. Adult catches in the modified CC traps have been increased by 50% by coating trap tops with Tanglefo t and removing the deflector plates. In laboratory studies, installation of a lime green LED light in the trap top resulted in catches of 281 adults/trap/24-h compared with catches of 18 and 12 adults/trap/24-h for traps with white LED light and no light, respectively. Studies are in progress to test the effects of the modified CC traps on catches of whitefly parasitic wasps.

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

# Dates Covered by the Report: 2000

# Preliminary Study of the Morphological Development of Silverleaf Whitefly Stages in the Field

Leaf samples were taken from field grown cotton and cantaloupe in Phoenix, AZ in 2000. The morphological development of silverleaf whitefly stages were determined at Fargo, ND. Nymphs on cotton leaves were longer and wider compared with nymphs on cantaloupe leaves. From cotton, the length of the four nymph stages were 257, 355, 589 and 724  $\mu$ m, respectively, and widths 140, 217, 395 and 498  $\mu$ m, respectively. For cantaloupe nymphs, the lengths were 244, 350, 456, and 601  $\mu$ m and widths were 129, 192, 267 and 411  $\mu$ m, respectively.

With nymphs placed on their sides, dorso-ventral measurements for nymphs on cantaloupe leaves were greater except for the third instars compared with dorso-ventral measurements for nymphs on cotton leaves. The values for nymphs from cotton and cantaloupe leaves were 63 vs.  $66 \mu m$ , 78 vs. 98  $\mu m$ , 139 vs. 124  $\mu m$  and 214 vs. 259  $\mu m$  for the first, second, third and fourth instars, respectively. As nymphs developed from first to fourth instars, the ventral half of the body increased more compared with the dorsal half of the body. The ratios of ventral and dorsal halves of bodies were 0.53, 0.57, 0.63 and 0.70 for first, second, third and fourth instars, respectively. On the average, adult body length from body lengths from head to tail were 953 and 1127  $\mu m$  and body weights were 17 and 39  $\mu g$  for males and females, respectively. Egg we ight was 0.8  $\mu g$  per egg. Exuvia weighed 0.8 and 5.4  $\mu g$  for non-parasitized and parasitized from fourth instar nymph's exuvia, respectively. The significances of the differences for each stage of development on the two plant species are being analyzed.

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

# Dates Covered by the Report: 2001-2002

## Preliminary Study of the Morphological Development of Sweetpotato Whitefly Stages in the Fields

Leaf samples were taken from field grown cotton and cantaloupe in Phoenix, AZ in 2000 and 2001. Measurements from eggs to nymphs and exuvia for the morphological development of whitefly stages were made at Fargo, ND. The 4<sup>th</sup> instars on cotton leaves were longer and wider compared with that on cantaloupe leaves. From cotton, the length of the four nymph stages were 250, 355, 434 and 724 mm, respectively, and widths 135, 214, 270 and 498  $\mu$ m, respectively. For cantaloupe nymphs, the lengths were 244, 350, 456, and 603  $\mu$ m and widths were 129, 192, 267 and 411  $\mu$ m, respectively. With nymphs placed on their sides, dorso-ventral measurements for nymphs on cantaloupe leaves were greater compared with nymphs on cotton leaves. The values for nymphs from cotton and cantaloupe leaves were 57 vs. 66  $\mu$ m, 76 vs. 98  $\mu$ m, 71 vs. 123  $\mu$ m and 214 vs. 258  $\mu$ m for the first, second, third and fourth instars, respectively. As nymphs developed from first to fourth instars, the ventral half of the body increased more compared with the dorsal half of the body. The ratios of the ventral and dorsal halves of the bodies were 0.53, 0.57, 0.61 and 0.70 for first, second, third and fourth instars, respectively. On average, adult body length from head to tail were 807 and 884  $\mu$ m and body weights were 17 and 39  $\mu$ g for males and females, respectively. Eggs weighed 0.8  $\mu$ g per egg. Exuvia weighed 1.2 and 3.6  $\mu$ g for non-parasitized and parasitized exuvia, respectively. The significances of the differences for each stage of development on the two plant species are being analyzed.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by the Report: 2000

# The Mechanics of Stylet Penetration by Bemisia argentifolii.

Most adult silverleaf whitefly probe sites occurred along the margins of the abaxial epidermal cells. Even though the penetration sites were in the area of the cell margins, we found that the stylets passed directly through the cytoplasm of the epidermal cell and not through the common radial wall between epidermal cells. Adult stylets averaged 217  $\mu$ m long. During non-feeding periods the stylet is completely enclosed within the labium, which consists of four distinct segments. The stylet bundle enters between the first and second labial segment and extends to the tip of the labium. After the tip of the labium is anchored to the epidermal surface the adult whitefly lowers its head pushing the stylet bundle down the labial groove into the host plant. The entire length of the stylet can be inserted into the leaf in this manner. The depth of penetration can be determined by examining the position of the labrum along the labial groove.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

#### Dates Covered by the Report:: 2000-2001

### What I Know About Silverleaf Whitefly Nymph Feeding and What I Would Like to Know

Nymphs feed during all four stages of development. Once they begin to feed, they settle down and become flattened against the leaf surface. The rostrum of the nymph is short and has sensilia on the tip similar to those found on the labium of the adult. The rostrum also has a groove through which the stylet bundle passes similar to the labial groove of adults. The tip of the mandibular stylets are ridged or serrated and used to penetrate the host leaf tissue. Stylets range from 110  $\mu$ m in first instar to over 200  $\mu$ m in fourth instar. The stylets can be at least partially withdrawn and reinserted during the feeding process. The nymphs increase in length and width with each molt and the newly formed stylets are also longer with each molt. However, the stylets do not increase in length proportional to the increase in size of the nymphs.

What are the mechanisms involved in the insertion and withdrawal of the nymphal stylet? Do the stylets of subsequent molts use the salivary sheaths developed by an earlier instar? What is the function of the sensillia on the tip of the rostrum in sedentary nymphs? How does the nymph stylet find the phloem tissue in host leaves? Does a salivary sheath form within the sieve tube element and do the stylets move from one sieve tube element to another?

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

Dates Covered by the Report: 2001

## Silverleaf Whitefly Nymph Stylets and Feeding Characteristics

After emerging from the egg, silverleaf whitefly (*Bemisia argentifolii* Bellows & Perring) nymphs move across the surface of the leaf in search of a feeding site. The crawler is the only nymphal stage that is capable of locomotion. The crawler must begin feeding quickly if it is to survive. Prior to the onset of feeding the crawlers stylet is folded within the body of the nymph. When the crawler begins to feed it becomes flattened against the leaf surface. Like the adult whitefly, the nymph produces a flange like material that appears to cement the tip of the rostrum to the surface of the host leaf. Staining techniques can be used to determine both probe and feeding sites. The nymph stylet bundle consists of a pair of mandibular stylets serrated at the tip and a pair of maxillary stylets locked together to form the food canal. The ultrastructure of the epidermal layer of the leaf the stylet moves through the apoplast of mesophyll all the way to the phloem tissue. The stylet path can be studied by examining the salivary sheath which remains in the leaf even after the stylet has been removed. The salivary sheaths are often highly branched, demonstrating that the stylet can be at least partially withdrawn and then extended in a different direction. Prior to each molt, the stylets are withdrawn into the body of the nymph and left behind in the cast exuviae. With each successive molt the stylet becomes longer and in the fourth instar may reach lengths greater than 200  $\mu$ m.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by the Report:: 1999-2000

## Replication of an Iridovirus In Whitefly Cells and Whitefly Nymphs

A cell line from the silverleaf whitefly, BtB-2.97-Hunter & Polston, was tested for its ability to support replication of the insect iridescent virus 6 (IIV-6). Several lines of evidence indicated that a productive infection was achieved. The cells displayed cytopathic effects (CPE) that included a change in cell shape and the appearance of cytoplasmic inclusions. Virus particles accumulated in defined areas of the cytoplasm. The cell-associated virus titer was detected at three orders of magnitude higher than that released into the media  $(1.3 \times 10^8 \text{ vs} \cdot 1.1 \times 10^5)$ . Fluorescent antibody staining was used to visualize the virogenic stroma that initially was localized to areas adjacent to the nucleus in the cytoplasm of infected cells. The accumulation of viral DNA and proteins in the virogenic stroma led to the formation of the cytoplasmic inclusions that were observed as CPE. In addition, western blot analysis indicated CIV structural proteins were being expressed. Each of these lines of evidence indicated that whitefly cells could support a productive CIV infection. Nymphs were infected by feeding the virus in diet on an artificial rearing system. Virus was detected in nymphs by PCR and electron microscopy, but the infections were not highly pathogenic. Infection of whiteflies by IIV-6 suggests that more pathogenic viral isolates may be found for this insect and that the virus has potential to be used as an intracellular probe.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics.

Dates Covered by the Report:: Sept 2000 - July 2001

## Physiological Considerations in the Penetration of Eretmocerus mundus into Bemisia Nymphs

*Eretmocerus mundus* oviposits under its host, *Bemisia tabaci*. Egg hatch occurs within ca. 3 days of oviposition. The first instar larva penetrates the host and development proceeds within a capsule of epidermal origin. Recent studies have shown that no matter under which host instar *E. mundus* eggs are laid, penetration into the host proceeds only during the host's fourth instar, typically sometime prior to the transition to adult characteristics. Thus, it is understandable that while *E. mundus* will oviposit under any of its host's four instars, the parasitoid prefers to lay an egg under either a second or third instar. Since approximately three days are required for parasitoid egg development, oviposition under a second or third instar whitefly will result in hatch occurring under a third or young fourth instar host; the period of time during which first instar parasitoid will be forced to remain outside its host will be minimal as will be its chances to miss the window during which the whitefly is vulnerable to penetration.

Epidermal capsule formation accompanies parasitoid penetration. The capsule surrounds/engulfs the parasitoid and must contribute to parasitoid development. Although the function(s) of the very large glands that are apparent in the first instar parasitoid larva is not known, it is possible that they influence the production of the capsule and that they may be essential for facilitating *E. mundus* penetration and/or development. The question as to whether host ecdysteroid levels are altered upon parasitization also was examined. Results showed that the level of ecdysteroids was always lower in a parasitized whitefly into which a first instar parasitoid had just begun to penetrate than in a healthy whitefly of the same developmental stage. Therefore, it does not appear that a parasitoid-induced increase in host ecdysteroid levels is involved in the stimulation of capsule formation. Rather, other regulatory molecules such as growth factors may be involved.

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Research & Implementation Area: Section A: Biology, Ecology, and Population Dynamics

## Dates Covered by the Report: 2000

## Sweetpotato Whiteflies, Cotton Aphids, and Sticky Cotton

We compared the sugars extracted and cotton lint exposed to SPW and CA under laboratory conditions; also their effects on cotton lint stickiness as measured by thermodetector counts. Sweetpotato whitefly (SPW), *Bemisia tabaci* (Gennadius) Strain B (=*B. argentifolii*) and cotton aphid (CA), *Aphis gossypii* Glover, are the two most common honeydew producing insect species that occur on cotton. Honeydew contaminated lint is a serious problem in lint processing at the textile mill. It can also be difficult to harvest and gin. The major sugar components of the honeydew of both insect species are glucose, fructose, sucrose, trehalulose, and melezitose. Trehalulose and melezitose are insect-produced sugars. SPWs produce more trehalulose in relation to melezitose and the opposite is true for CAs. We exposed clean cotton lint to SPW or CA in the laboratory. The total sugar contents of water extracts of honeydew-contaminated lint after exposure to the insects were significantly correlated to increasing thermodetector counts (a measure of stickiness) that occurred as a result of increasing durations (days) of exposure. Higher concentrations of total sugars measured in these extracts occurred following exposures to SPW compared with CA. However, numbers of SPW and CA were different and the results are therefore not directly comparable. Research is continuing to further define differences and similarities in cotton lint stickiness as a result of honeydew lint contamination by SPW or CA.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by the Report: 2000

## Open Cotton Boll Exposure and Development of Sticky Cotton in Whitefly Infested Fields

We used thermodetector analysis to determine the effect on lint stickiness of different numbers of days of exposure of open cotton bolls to whitefly, *Bemisia tabaci* (Gennadius) Strain B (=*Bemisia argentifolii*) populations. Thermodetector lint stickiness measurement is accomplished by manipulating a 2.5 g sample of cotton lint into a fine mat, which is placed between two sheets of aluminum foil and heated under pressure. The foil sheets are separated and the number of sticky spots counted. Thermodetector counts do not distinguish between the contributions of the different sugars in honeydew or plant physiological sugars. However, the counts are an overall assessment of cotton lint stickiness. We also studied the effect of rainfall on sticky cotton.

Trehalulose and melezitose produced by *Bemisia* and thermodetector counts in cotton lint increased with increasing numbers of days of exposure of open cotton bolls in infested cotton plots. Thermodetector counts were significantly correlated to amounts of trehalulose and melezitose. Rainfall of 0.5 inch reduced trehalulose and melezitose in cotton lint within 5 h following the rain. The results suggest dissolution of the sugars followed by runoff as opposed to microbial degradation.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by Report: 2000-2001

## Cotton Water Stress Effects on Bemisia tabaci Strain B (Homoptera: Aleyrodidae) on Honeydew Production

Comparisons of honeydew production by sweetpotato whitefly (SPW), *Bemisia tabaci* (Gennadius) Strain B while feeding on water-stressed or non-water-stressed cotton showed that more honeydew sugars were produced on non water-stressed leaves of cotton plants (four days after irrigation) compared to those feeding on leaves of cotton plants 7 or 13 days after irrigation (water stress). Leaf water potentials, as a measure of water stress, decreased with increasing numbers of days following irrigations. Leaf water potentials of furrow irrigated cotton and furrow irrigated plus supplementary drip irrigated cotton (1 h per day) showed the same decreasing leaf water potential patterns. The decreases, however, were less in furrow irrigated plus supplementary drip irrigation compared with furrow irrigation alone. SPW feeding on plants in the field with furrow plus supplementary drip irrigation and in the greenhouse on non-stressed cotton plants produced more micrograms of honeydew sugars per gram of honeydew compared to SPW feeding on plants with furrow irrigation alone in the field and on water-stressed plants in the greenhouse.

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Research & Implementation area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by the Report: 1999

## Distribution of Immature, Adult and Immature Parasitoids of *Bemisia tabaci* Genn. (Homoptera: Aleyrodidae) in Cotton, Cukurova, Turkey

Cotton is one of the important cultivated plants in Cukurova, Turkey. The climate in Cukurova region is subtropical, with hot dry summers and mild winters. The cotton growing season lasts from the end of March to the beginning of October. Many pests are attacked cotton in this period. *Bemisia tabaci* Genn has been one of the most important pest of cotton under irrigated conditions. Although its populations vary from year to year, it is still the key pest in Cukurova, causing economic damage and yield reduction. However, there is no efficient sampling plan to estimate or classify populations of whitefly for management purposes. The first step in developing a sampling plan is to define the sampling unit and random variable of spatial distribution.

Studies were conducted to determine the sampling unit and spatial distribution of immature and adult *B. tabaci* and immature parasites. For this purpose 20 plants were examined at five different dates for immature *B. tabaci* and parasites. Adults were counted by leaf turn method on a randomly selected leaf from top, middle and bottom part of the cotton plants at nine different dates. Also adults counted on individual mainstem leaf from node 2 through 8 (terminal:node 1) at four different date. All studies were conducted in 1998-1999, but only data from 1999 was used to determine the distribution pattern.

We looked at 3 parameters to describe the distribution of whitefly and parasitoid life-stages among various leaf positions as follows: (1) the leaf position with the highest percentage of a particular stage (2) leaf position where insect counts were best correlated with counts on the total plant (3) the coefficient of variation. The pattern of aggregation for immature, adult *B. tabaci* and immature parasites was measured by Taylor's power law. All 3 distribution parameters changed throughout the season. Across the entire season, the 3 distribution parameters for whitefly eggs were associated with leaf positions 2, 3 and 4; for nymphs with leaf positions 5, 6 and 7; with pupae (red-eye) leaf positions 6, 7, 8, 9 and 10; and for immature parasitoids with leaf positions 8, 9, 10 and 11. Based on consideration of Taylor's power law and precision, it was most efficient to sample leaf 3 for eggs, leaf 5 for nymphs, leaf 10 for pupae and immature parasitoids.

Adult were consistently more abundant on leaves from the top than leaves from the middle and bottom strata. According to the 3 parameters, it was most efficient to sample mainstem leaf 4 for whitefly adult stage.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered by the Report: January 2000-December 2001

#### Development of Sampling Methodology for Estimating Cotton Lint Stickiness due to Infestations of *Bemisia tabaci*

Sticky cotton continues to be a worldwide problem that may presently be considered by the cotton industry as the most serious factor affecting cotton quality. The nature and causes of lint stickiness are complex. Plant-produced sugars have been implicated in the sticky cotton problem; however, a high percentage of the documented lint stickiness problems have been associated with the occurrence of honeydew-producing insects such as aphids and whiteflies. Honeydew induced stickiness is problematic at many post-harvest phases of lint processing including ginning, carding, and particularly spinning. The sticky cotton problem in the U.S. has increased with recent population outbreaks of the whitefly *Bemisia tabaci* and research is currently addressing an array of issues from characterization of sugars to management of plant and insect sources.

The accurate determination of cotton lint stickiness in the field could greatly aid management and research efforts. Field sampling of stickiness could augment pest monitoring techniques and improve overall decision-making for pest suppression. Field sampling could also identify unacceptable levels of stickiness at harvest that may require some remedial action such as the use of enzyme treatment systems that are currently being investigated. Our research targets the development of standard methodologies for collecting lint samples from fields to ensure statistically precise estimates of stickiness. Two assay systems were investigated, the manual thermodetector (SCT) and the high speed (automated) sticky cotton thermodetector (H2SD). The development of sampling plans involves four inter-related components: 1) description of the sampling distribution, 2) selection of the optimal sample unit, 3) partitioning of variance components, and 4) determination of the number and allocation of sample units.

The newest H2SD system consistently detects more thermodetector spots than the SCT and the relationship between the platforms appears to be nonlinear, at least for field-collected samples encompassing a very wide range of stickiness levels. The distribution of sticky lint in the field is random regardless of which platform is used to assay the lint. Time and cost considerations are critical to any sampling procedure. Examination of many different sizes of field sample units suggest that smaller sample units (e.g. all lint from 1 plant or lint from 20 bolls collected at random) are more cost-efficient than larger sample units. Further analyses of variance components suggests that only a single assay for each sample unit should be conducted with the SCT, but that 3 assays should be conducted for each sample unit on the H2SD. This analysis takes into account the relative costs of sample collection in the field and the time required to complete a single assay with each platform.

The assay system (SCT or H2SD) used is critical to the development of a final field sampling plan; however, there is considerable flux in the industry relative to a standard platform for stickiness testing. Given this uncertainty, we have developed sampling plans for both platforms. Reasonably precise estimates of stickiness can be achieved with a sample size of about 15 on either platform using the 1-plant sample unit. Even fewer samples would be required using a 20-boll unit. Sample sizes as high as 41 or 82 would be required for estimates with very high precision on the H2SD or SCT platform, respectively. Overall sampling costs are much lower for the H2SD because of the speed with which samples can be assayed. Sampling efficiency could be further improved with sequential sampling strategies or with sample plans focused on categorizing (non-sticky vs. sticky) rather than estimating actual levels of stickiness. These sampling applications await further research to define critical levels of stickiness and/or the implementation of wide-scale testing where any time savings would be at a premium.

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## Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

### Dates Covered by the Report: January 2000-December 2001

#### Movement and Mortality of Bemisia tabaci Crawlers on Cotton

Greenhouse studies were conducted to examine movement and estimate rates of mortality of crawler stage *Bemisia tabaci* on cotton. Five female whiteflies were allowed to oviposit for 24 h in clip cages on the lower surface of a single leaf (generally the 2<sup>nd</sup> to 3<sup>rd</sup> leaf from the terminal) of cotton plants that had 6-8 expanded leaves. After removal of adults, plants were then placed in organdy cages to prevent further oviposition. One to two weeks after oviposition plants were destructively sampled, and the location (leaf node, upper or lower leaf surface) and number of settled nymphs were recorded. The petiole lengths of all leaves and internode distances were measured and recorded. The number of hatched and unhatched eggs on the oviposition leaf also were recorded. The experiment was replicated 20 times in still air and 20 times with plants exposed to a table-top fan blowing air at a velocity of approximately 2.2 meters/second. This latter condition was used to simulate the effects of a moderate breeze on crawler movement and mortality.

The survival of crawlers did not differ statistically between still-air and wind treatments and averaged 85.5% overall. Survival ranged from 50 to 100% on individual plants. Over 99% of all surviving crawlers settled on the lower surface of leaves and 99.9% settled on the leaf where eggs were oviposited. Over both treatments only 3 individuals (out of 2700 surviving nymphs) moved from the leaf of origin and they all moved to leaves lower on the plant. Assuming that these individuals crawled rather than fell, the mean d istance moved was 200 mm. These results suggest that crawlers move very little and that survival is high on young cotton plants in the absence of other natural factors such as natural enemies. Further study is needed in the field over a wider range of conditions to evaluate natural levels of mortality for life table analyses.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

#### Dates Covered by the Report: 2000 and 2001

### Comparison of the Feeding Process of Nymph and Adult Whiteflies

With scanning electron microscopy we have examined the feeding processes of nymph and adult whiteflies. The *Bemisia argentifolii* adult feeding apparatus consists of a four-segmented labium in which is contained a slender stylet bundle (1.5-2 micrometers in diameter), consisting of two mandibles and two maxillary stylets. The stylet bundle of the adult is completely contained within the groove in the labium when not feeding. To penetrate the leaf, the adult places the tip of the labium against the leaf and then lowers its head in order to slide the stylet bundle down the labial groove and push the tip of the stylet bundle into the leaf. The length of the stylet bundle, averaging 217 micrometers, is sufficient to allow the adult to reach phloem tissue from any position on the leaf, from either abaxial or adaxial surfaces.

The total length of the adult stylet bundle is determined by measuring the distance from the junction of labial segments 1 and 2 to the tip of the labium of non-feeding adults. The length that the stylet bundle has penetrated the leaf can be determined by measuring the distance from the junction of labial segments 1 and 2 to the position of the head (where the stylet bundle enters the labial groove) along the labial groove. This distance varied from 43 to 151 micrometers indicating that the adult uses less than 70% of the length of its stylet to reach a phloem bundle in cotton. The distance from the abaxial epidermal surface to the phloem in the cotton leaves we exa mined ranged from 53 to 127 micrometers.

The stylet bundle of the immature whitefly, *Bemisia argentifolii*, appears to be looped or coiled within the body. The end of the bundle enters a groove in the rostrum and extends beyond the rostrum tip. In order to penetrate the leaf, the nymph extends the stylet bundle beyond the tip of the rostrum in an unknown manner. The length of the stylet bundle ranges from a minimum of 110 micrometers in the crawler to a maximum of 200 micrometers in the 4th instar. The length of the stylet bundle only increased 39% while the length of the nymph increased 168%. The length of the nymph stylet bundle appears to be of sufficient length to reach phloem from any position on the leaf surface.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

Dates Covered By The Report: January 2000 - December 2000

#### Life Table Studies of Bemisia tabaci Genn. on Cassava

An investigation was undertaken to study the age specific mortality of individuals in the whitefly population on cassava. Cassava plants (cv. M4 & H226) were raised in mud pots (21' diameter) placed in five groups of ten plants each. When cohorts of settled first instar nymphs were established from natural population; three leaves were selected from each plant and their location was marked with a non-toxic indelible marker pen. The fate of the each individual was then tracked by visual observation using a pocket microscope (10-60x magnification), every third day to estimate mortality due to predation, parasitisation and missing/unknown factors.

During the first observational period (18 January - 20 February, 2000) first instar nymphs selected were 17.2/leaf, and pupae and adults emerged were 2.0 and 2.5/leaf, respectively. Predation was the prime source of mortality (43.6%) followed by missing/unknown (28.3%) and parasitisation (15.2%). During the second observational period (2 February - 22 February 2000), first instar nymphs mean of 17.4/leaf was observed and emergence of pupae and adults averaged 2.9 and 0.9/leaf, respectively. In this period, 50.4% of mortality was due to missing/unknown factors and 26.8% and 22.4% due to predation and parasitisation, respectively. During the third (9 - 31 March), fourth (4 -30 April) and fifth observational period (9 - 26 May), more adults emerged. Out of 11.4, 17.6 and 13.8 nymphs/leaf during March, April and May, respectively; 5.2 pupae and 4.4 adults during March, and 9.7 pupae and 8.2 adults during April, and 8.0 pupae and 7.1 adults emerged during May. This may be probable due to the favourable climatic/weather factors. In the last three observational periods, missing / unknown factors contributed to maximum mortality (36.8% / leaf, March, 22.9%, April and 19.7% May). While predator induced mortality was 15.9, 11.6 and 19.3 % during March, April and May, respectively. During April, parasitisation (13.8%) outnumbered predation (11.6%).

Natural enemies observation were aphelinid parasitoids, predatory coccinellids, dolichopodids, mites and spiders. Adult and larval stage of the coccinellids were found to feed on all the stages of the host. Usually adult predatory mites sucked on the body fluid of the host. Missing/unknown factors were a significant source of mortality in most of the experiments, followed by predation and parasitism. However, predation and parasitism contributed to high mortality to immature stages.

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Research & Implementation: Section A: Biology, Ecology and Population Dynamics

Dates Covered by the Report: January -December 2000

### Presence of Biotypes in Bemisia tabaci Genn. in India

Studies were undertaken to investigate the host associated variation in *B. tabaci*. Host transfer studies using clip cages and host preference studies using host plants, cassava, sweetpotato, egg plant, tobacco and cotton showed two host associated strains of *B. tabaci* in India. The two strains were identified as cassava biotype and sweet potato biotype. The whitefly population from sweet potato did not colonize on cassava, similarly whiteflies from cassava showed limited colonization on cotton but failed to complete its development on sweet potato. Egg plant and tobacco proved to be the common host for both. The cassava biotype showed 96.7% adult emergence on cassava compared to 70.0% on egg plant followed negligible on cotton and nil on sweet potato. Whereas with sweet potato biotype showed 90.7% adult emergence on sweet potato followed by 78.8% on cotton, 55.2% on egg plant and no adult emergence on cassava.

The duration or the life cycle of sweet potato whitefly was maximum on brinjal (20.2 days). The cassava whitefly showed the highest total developmental duration on cassava (19.2 days). The longevity of cassava whitefly on sweet potato and cotton and that of sweet potato whitefly on cassava, and cotton were also studied. The longevity of the cassava whitefly ranged from 4-10 on cotton with a mean of 7.8 days and 4-6 on sweet potato with mean of 4.7 days. The longevity of the sweet potato whitefly ranged from 2-7 days on cassava and 14-29 days on cotton, with their means 4.2 and 17.7 days respectively.

The studies on esterase profile corroborated the presence of two different biotypes. There was clear difference in the esterase banding pattern between the two biotypes.

Cassava mosaic virus transmission studies conducted using seedlings of cassava and cassava whitefly biotype could make successful transmission of CMD from infected to healthy seedlings. Symptoms developed within 9-30 days and were confirmed by ELISA.

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Research & Implementation: Section A: Biology, Ecology and Population Dynamics

Dates Covered by the Report: January 1999- December 2001

## Life Table Studies of Bemisia Tabaci Genn. on Cassava and Laboratory Evaluation of Potentiality of Predators

An investigation was undertaken to study the age specific mortality of individuals in the whitefly population on cassava. Cassava plants (cv. *M4 & H226*) were raised in mud pots (21' diameter) placed in five groups of ten plants each. When cohorts of settled first instar nymphs were established from natural population; three leaves were selected from each plant and their location was marked with a non-toxic indelible marker pen. The fate of the each individual was then tracked by visual observation using a pocket microscope (10-60x magnification), every third day to estimate mortality due to predation, parasitisation and missing/unknown factors.

The observations were grouped into four observational periods. During the first observational period adult survivorship (11.3%) was very low due to increased predation followed by unknown factors. The predator mortality was highest in the first instar to pupal stage. During the second observational period, adult survivorship (4.72%) was lowest among the four observational periods. Mortality from parasitisation was maximum in the pupal to adult stage. Mortality for missing/unknown factors was highest during the third observational period. The percentage of adult survival was 46.6%. Adult survivor (55%) was highest due to decreased predation and parasitisation.

The overall survival from the first instar to the adult ranged from 4.72-55% during these periods. Parasitisation was the dominant mortality factor from pupae to adult emergence.

Natural enemies noticed during observations included aphelinid parasitoids, predatory coccinellids, mites and spiders. Adult and larval stages of the coccinellids were found to feed on all stages of the host. Predatory mites were found to suck the body fluid of the host, and this was the most dominant mode of predation. Missing/unknown factors were a significant source of mortality in most of the experiments, followed by predation and parasitism. However, predation and parasitism together contributed to high mortality of immature stages.

The predation efficiency of five coccinellid predators and predatory mite were studied in the laboratory. *Serangium paracestosum* (91.45  $\pm$  5.32) was found to be dominant over the other five predators. *Anegleis cardoni* was less effective than *S. paracesetosum* but superior to the other four species. The predation efficiency of the four other predators was at par and showed no significant differences.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

Dates Covered by the Report: January 1999 - December 2001

## Isozyme, Vector Capability and Cross Breeding Studies to Confirm the B biotypes in Bemisia tabaci (Genn.) in India

Host associated variation studies revealed the presence of two strains of *Bemisia tabaci*- Cassava whitefly and Sweet potato whitefly, which showed clear-cut host preferences. Additional confirmations were carried out by analysing the variations in the isozyme patterns, vectoring capabilities and by doing cross-breeding studies.

Two enzyme systems studied under isozyme analysis includes Esterase and Malate dehydrogenase (MDH). The esterasebanding pattern in sweet potato whitefly differed from the cassava whitefly in having an additional fast moving esterase band (Rf value 0.293). The migration distances of the two slow moving bands, common to both were also different (0.150 and 0.204 in CWF and 0.108 and 0.165 in SPWF). Banding patterns using the MDH system (Rf values 0.104 and 0.162) was identical in both biotypes.

Investigations on the role cassava biotype as a vector of Indian Cassava Mosaic Virus (ICMV) was confirmed by ELISA using monoclonal antibodies raised (MAbs) to ICMV isolates. Cassava biotypes could successfully transmit ICMV, whereas sweet potato biotype failed to transmit. A single whitefly could transmit ICMV with 10h AAFP & IAFP. Maximum percentage of transmission was obtained when AAFP & IAFP were 48h. This fact corroborated the indication of biotypes of *B. tabaci* in this region.

The biological consequences of mating interaction between the two biotypes of *B. tabaci* were studied by cross breeding female SPWF and male CWF, and female CWF and male SPWF. This showed an increased proportion of male progeny indicating that cassava and sweet potato are two biotypes and not separate species.

Thus biological and electrophoretic studies and vectoring efficiency have very clearly revealed the presence of two distinct biotypes of *B. tabaci* – a cassava biotype and a sweet potato biotype in India. This is the first report on a scientific basis.

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Research & Implementation Area: Section A: Biology, Ecology and Population Dynamics

## Dates Covered By The Report: Summer, 2001

#### Engineering-Based Computer Simulation for Modeling Greenhouse Whitefly Population

Development of a mathematical model is a suitable technique for understanding insect population dynamics. It offers a possibility to bring together the results of the extensive research conducted and recognizes possible gaps in our knowledge. Once the model has been verified and validated, it can be used to obtain qualitative and quantitative information on the importance of certain relations or parameters and it may serve to predict the reaction of a system on a change of the values of parameters, variables etc.

It was decided to restrict the model to one host-plant species, from which the most experimental data were available, so tomato was used as a host-plant in this research. A simulation model was constructed since dynamic model was desired. SLAM II is used as an engineering-based computer simulation language. This simulator was originally developed for modeling industrial manufacturing processes. This software was used to simulate the population growth of the greenhouse whitefly. Temperature was used as one of the abiotic parameters. Whitefly females were then given a value for daily fecundity equal to 5. At this point, the female was checked and if its lifetime was terminated it was routed to a death or termination node. If it was not terminated, it was allowed to lay its complement of eggs and then routed back to the fecundity node. It continued in this cycle until death. The eggs were each assigned the current simulation time as their "birth day". Next 6.1% of eggs were routed to termination node corresponding to egg mortality. The remaining eggs were sent to another assignment node that was assigned egg development time. The model continued in this manner until all eggs developed to pupae. The total population was obtained by summing these five activity counts. The program of the whitefly model was ran (with TRACE option).

To validate the part of the model that described the developmental stages, there were two data sets available for comparison. The data came from an experiment that was carried out in the greenhouse. In this experiment the numbers of developmental stages of two sets of 106 and 115 whitefly eggs on tomato leaves were followed and recorded daily at average 18 C° and %65±5 RH. The average developmental period from egg to adult took 52 days, while it lasted 56 days in the simulation. The average developmental period for the second data set took 52 days, while it lasted 58 days in the simulation. The mortality was lower than simulated particularly during egg and second larval stages. The average of mortality in greenhouse was 21.2% and in model was 24%.

To validate the part of model that described the population growth, there were two data sets available for comparison. The data came from an experiment carried out in greenhouse conditions. In this experiment, the numbers of empty pupae of two sets of 106 whitefly eggs on tomato leaves were followed and recorded weekly at  $18\pm2$  C° and %65 RH in autumn and 100 whitefly eggs in average 24 C° and 65%±5 RH in spring.

The greenhouse data was above the curve of the model. This may be related to temperature conditions in the model and greenhouse. The development of immature stages of greenhouse whitefly can be predicted with this model. But sensitive analysis shows the developmental duration has the most effect on population growth.

#### Section A Research Summary

Compiled by D. N. Byrne and J. L. Blackmer

Recent greenhouse studies examined dispersal by *B. tabaci* crawlers on cotton. Crawlers were found to move as far as 200 mm. Ongoing studies examined seasonality and mortality patterns of *B*. tabaci on a variety of crop plants and weeds. Owing to their polyphagy, a large number of plant species support whitefly populations at various times of the year. Predation and dislodgment were found to account for a significant portion of Bemisia mortality. Age specific mortality was investigated on cassava plants. Adult survivorship depended on time of year. On cassava, parasitism was the dominant mortality factor from pupal to adult emergence. In laboratory studies, Serangium *paracestosum* was the most important predator. Studies were conducted to determine the most appropriate sampling unit as well as spatial distribution of immature and adult Bemisia. Similar factors were examined for immatures and parasitoids. Units and distribution were stadiadependent. As a result of two related studies, improvements have been made in the CC trap. Two assay systems for detecting sticky cotton were investigated, the manual thermodetector and the high speed (automated) sticky cotton thermodetector. Sampling costs are much lower for the automated system because samples can be assayed more quickly. Field and laboratory studies in the Southwest indicated that Eretmocerus eremicus may have a more limited ability to disperse than Bemisia. This may limit their use as biological control agents in southwestern agriculture. In India, studies revealed the presence of two strains of Bemisia *tabaci* - the cassava whitefly and the sweet potato whitefly. The ability of the cassava biotype to vector Indian Cassava Mosaic Virus was confirmed. A system based on body depth and maturation of the adult eye was used to track development and identify physiological synchronous 4<sup>th</sup> instar/pharate adult *Bemisia*. An examination of histological sections of the last instar revealed that adult eve and wing development were initiated in Stage 6. Wings were deeply folded by Stages 6 and 7. Ecdysteroid titers peaked at Stages 4-early 6, just before the onset of adult development. In Mexico, numerous whitefly species have been identified including Bemisia, Trialeurodes vaporariorum, T. abutilonea, Aleurothirixus floccosus (Maskell), Tetraleurodes acaciae and Tetraleurodes acaciae. The identification and composition of the fatty acids associated with the major lipid classes within Bemisia nymphs were determined. All lipid

classes contained variable distributions of eight fatty acids: myristic acid, palmitic acid, stearic acid, arachidonic acid, palmitoleic acid, oleic acid, linoleic acid, and linolenic acid. The triacylglycerol composition of *Bemisia* adults was characterized, and the major fatty acid constituents were identified as oleic acid, palmitic acid, stearic acid, and linoleic acid. Several different types of bacteria were cultured from surface-sterilized *Bemisia* adults and nymphs, including *Bacillus* spp. Enterobacter cloacae, was found within the gut cells of adults. The rostrum of the Bemisia nymphs was found to be short and with sensilla on the tip. Stylets range from 110 um in the first instar to over 200 µm in the fourth instar. Prior to the onset of feeding, the crawlers stylet is folded within the body of the nymph. A newly developed artificial rearing system was used to determine how biotic and abiotic factors influenced egg hatch, crawler establishment and development of Bemisia. Egg age, day length, light intensity and density of eggs placed on the artificial membrane influenced hatch rates, establishment of crawlers, and to a certain extent subsequent development of nymphs. Recent studies indicated that no matter on which host instar E. mundus eggs are laid, penetration into the host proceeds only during the host's fourth instar. A cell line from the Bemisia. BtB-2 tested for its ability to support replication of the insect iridescent virus 6 (IIV-6).

Research Approaches	Year 1 Goals Statement	Progress A Yes	Achieved No	Significance
Determine life cycle vulnerabilities (life tables) <sup>a</sup> , population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.	Whitefly and natural enemy sampling in cultivated crops, urban planting and weed hosts.	X		Partial life table analyses have been completed for <i>B. argentifolii</i> on cotton in Arizona. Natural forces, including predation and dislodgment are major mortality factors; parasitism was a minor source of mortality. Survivorship from egg to adult ranged from 0-8.5% over 4 generations in sprayed and unsprayed fields. Studies on wild host crops in Israel indicate that parasitoids may contribute to low levels of whitefly on lantana. Whitefly and natural enemy populations were monitored in cropping systems in the Imperial and San Joaquin Valleys of California, Maricopa, Arizona and the Rio Grande Valley of Texas. The spread of <i>B. argentifolii</i> is being documented in Brazil. Life table studies provide valuable quantitative information on sources of whitefly mortality; surveys define the temporal and spatial dynamics of pest and natural enemy populations. This information is critical in developing and refining more biologically-based management systems.
Develop sampling methodology, action and <sup>b,c</sup> economic thresholds for all major crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Initiate whitefly to identify spatial and temporal distributions in major cultivated crops.	Х		Relationships between whitefly density and the occurrence of tomato irregular-ripening as well as preliminary sampling plans for whitefly on tomato have been developed. Evaluations of a reusable trap for surveying adult whiteflies in various crops are continuing. Studies of the effects of various insecticides on whitefly natural enemies are ongoing. Sampling plans and action thresholds are still needed for a number of affected crops.

## Table A. Biology, Ecology, and Population Dynamics.

		Progress A	chieved	
Research Approaches	Year 1 Goals Statement	Yes	No	Significance
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day- degree sub-models for estimating phenology and temporal patterns of whitefly, natural enemies and host crops.	Summarize whitefly biology, ecology and plant phenology to identify whitefly host plant interfaces.	Х		Development of large-scale temporal and spatial models and temperature-dependent, site-specific population dynamics models continues. Such models have the potential to encapsulate our current knowledge and provide a framework for developing more efficient management systems. However, considerable biological and ecological detail, as well as information on various aspects of pest management is available and needs to be integrated into these models to make them most useful as exploratory tools.
Develop sampling methods for quality of cotton lint, vegetables and other commodities.	Initiate sampling of seed cotton in the field during the season, at harvest, after picking, moduling and ginning.	Χ		Research has characterized the temporal distribution of honeydew deposition by <i>B. argentifolii</i> in cotton, improved our understanding of the relationship between lint stickiness and whitefly abundance and compared the production of trehalulose and melezitose between nymphs and adults. Studies reveal that cotton lint stickiness is randomly distributed in cotton fields. Preliminary sampling plans have been developed for estimating pre- harvest cotton lint stickiness. Stickiness constitutes one of the most important problems currently facing the cotton industry.
Quantify whitefly and natural enemy dispersals and contribution to population dynamics.	Review and analyze existing knowledge of whitefly dispersal.	Х		Studies have characterized the aerial distribution of whiteflies dispersing from cantaloupe fields and have examined the trade-offs between oogenesis and flight activity. Studies on whitefly parasitoid dispersal are ongoing. Understanding and predicting the timing and extent of the movement of whiteflies and their natural enemies is an important component in developing areawide management systems.

Research Approaches	Year 1 Goals Statement	Progress Achie Yes No	
Define mating behavior, reproductive isolation, species, biotypes.	Initiate studies on mating, oviposition and other behavior.	X	Surveys worldwide continue to document the spread of <i>B. argentifolii</i> . Electorphoretic analyses demonstrate the presence and extent of this pest in throughout Australia and Brazil. <i>B. argentifolii</i> appears to be displacing <i>B. tabaci</i> Biotype A in Brazil and is having a large impact on agricultural production through direct feeding and geminivirus transmission. Reports of heterozygotes between <i>B.</i> <i>argentifolii</i> and the extant Australian type <i>of B.</i> <i>tabaci</i> corroborates previous laboratory and highlight the taxonomic challenges within the <i>Bemisia</i> species complex.
Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Continue examination of <i>Bemisia</i> sp. for distinct morphological character differences.	Х	Comparative morphological analyses have been completed on <i>Bemisia</i> pupae from around the world. Several of these characters are highly variable among populations suggesting that pupal morphology should not represent the sole criteria for classifying individuals within the <i>Bemisia</i> species complex.
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Identify endosymbionts in whitefly.	Х	The effects of antibiotics on the biology of <i>B.</i> <i>argentifolii</i> have been examined. Several antibiotics that interfere with bacterial protein synthesis affected growth and development of immatures, but none affected oviposition rates or sex ratio. Results have important implications for the use of antibiotics to disrupt the function of whitefly endosymbionts and other associated microbes as potential control methods.
Characterize nutrient uptake and metabolism	Determine the process of uptake and metabolism of carbohydrates, amino acids and other nutrients.	Х	High levels of a polyol, sorbitol, were associated with elevated ambient temperatures. Sorbitol may function as a thermoprotectant in whiteflies that enables them to thrive in desert environments. The pathway of sorbitol synthesis and degradation in <i>B.</i> <i>argentifolii</i> is unique and may offer and avenue to develop transgenic plants which could disrupt sorbitol synthesis and compromise the whiteflies ability to deal with heat stress.

Research Approaches	Year 1 Goals Statement	Progress A Yes	Achieved No	Significance
Research Approaches		103	110	Significance
Develop whitefly artificial diets and natural enemy mass-rearing.	Identify whitefly nutritional components in plant tissue.	Х		An artificial diet and feeding system for rearing immatures of <i>B. argentifolii</i> has been developed. Rates of development of individual instars were comparable to those estimated on various host plants. The feeding system has proven to be a useful bioassays for examining diet components and for studies of primary metabolism based on defined diets, and has the potential to provide a means of mass rearing whitefly parasitoids.

<sup>a</sup> Natural enemy research complements from Section D, see Table D.
 <sup>b</sup> Action and economic thresholds also apply in Section C, see Table C.
 <sup>c</sup> Sampling technology applicable to all other sections, see Tables B to F.

Research Approaches	Year 2 Goals Statement	Progress A Yes	Achieved No	Significance
Determine life cycle vulnerabilities (life tables) <sup>a</sup> , population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.		X		Life table studies continued to characterize and quantify mortality factors for immatures of <i>B.</i> <i>argentifolii</i> on cotton. Predation and dislodgment accounted for much of the mortality in untreated fields and survivorship from egg to adult ranged from 0-18.2% over 6 generations. Several perennial plants species show potential to serve as refugia for exotic and native parasitoids. Life history and reproductive potential has been studied on various crop and weed hosts in the US and Italy. Whitefly population dynamics and virus incidence has been examined in cropping systems in Costa Rica, India and Guadaloupe. These ecological and biological studies form the foundation of effective pest management strategies.
Develop sampling methodology, action and <sup>b,c</sup> economic thresholds for all major crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Analysis and identification of needed additional sampling research to develop appropriate sampling protocol.	Х		A multistate study determined action thresholds for cotton in Arizona and California. Evaluations of a reusable trap for surveying adult whiteflies in various crops are continuing. Studies of the effects of various insecticides on whitefly natural enemies are ongoing. Sampling plans and action thresholds are still needed for a number of affected crops.
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day- degree sub-models for estimating phenology and temporal patterns of whitefly, natural enemies and host crops.		Х		The first version of a temperature-dependent, site- specific population dynamics model of <i>B</i> . <i>argentifolii</i> in cotton and cantaloupe was completed. Additional refinements, enhancements and field validation are needed to improve the utility of the model for predicting whitefly population dynamics under various management regimes and environmental conditions. In general, considerable biological and ecological data are available and need to be integrated into these models to make them most useful as exploratory tools.

## Table A. Biology, Ecology, and Population Dynamics.

		Progress Achie	eved
Research Approaches	Year 2 Goals Statement	Yes No	- 8
Develop sampling methods for quality of cotton lint, vegetables and other commodities.	Based on year 1 results, expand and repeat sampling protocols as described.	Х	Comparative evaluations of manual and high speed cotton stickiness thermodetector revealed differences in performance that have important implications for the development of measurement scales for stickiness and the number of samples that would need to be collected for the precise estimation of stickiness. Research on quality- related problems in other affected crops is needed.
Quantify whitefly and natural enemy dispersals and contribution to population dynamics.	Validate times of whitefly dispersal, environmental factors and identify modifying factors.	Х	Studies on whitefly and parasitoid dispersal are ongoing in the desert southwest. Understanding and predicting the timing and extent of the movement of whiteflies and their natural enemies is an important component in developing areawide management systems.
Define mating behavior, reproductive isolation, species, biotypes.	Define interspecies interbiotype mating interactions.	Х	Research continues on the role of reproductive isolation in the formation of species and biotypes, using insects from around the globe. There has been little detailed study of mating behavior <i>per se</i> , and its relevance for mating incompatability.
Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Develop genetic molecular level and acceptable species level separation.	х	Molecular characterization of the global whitefly complex is ongoing to clarify the taxonomic relationships between <i>Bemisia</i> whitefly populations. The whitefly karyotype has been determined and is an important development in our understanding of whitefly reproduction.
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Determine role of endosymbionts in whitefly biological functioning.	Х	The discovery of <i>Wolbachia</i> endosymbiotic bacteria in whiteflies is a new development that has significant implications for development of control strategies targeting the reproductive biology of whiteflies.

		Progress A	Achieved		
Research Approaches	Year 2 Goals Statement	Yes	No	Significance	
Characterize nutrient uptake and metabolism	Determine the biochemical pathways for metabolism of compounds essential for whitefly development.	Х		Fundamental questions about the nutritional physiology of whiteflies are being answered with the aid of artificial diets. Biochemical pathways for carbohydrate metabolism and polyol synthesis hav been determined. Metabolism of plant toxins is being studied to assess the ability of <i>Bemisia</i> to detoxify plant deterrent compounds. The role of nitrogen fertilization in whitefly-cotton interactions was determined in field trials.	
Develop whitefly artificial diets and natural enemy mass-rearing.				Development of an artificial feeder for whiteflies that will support development from egg to adults has been successful, and improvements continue to increase the proportion of <i>Bemisia</i> adults produced. This system has been tested for its effectiveness at supporting parasitoid wasp development, and adult <i>Encarsia</i> have been successfully produced in this system. Further research is needed to optimize the system for both whitefly and parasitoid development.	

<sup>a</sup> Natural enemy research complements from Section D, see Table D.
 <sup>b</sup> Action and economic thresholds also apply in Section C, see Table C.
 <sup>c</sup> Sampling technology applicable to all other sections, see Tables B to F.

## Table A. Biology, Ecology, and Population Dynamics.

Research Approaches	Year 3 Goals Statement	Progress A Yes	Achieved No	Significance
Determine life cycle vulnerabilities (life tables) <sup>a</sup> , population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.	Identify potential low population manipulation on vital host links for survival	Y		Low survivorship was documented both on cotton during peak season and on citrus during overwintering. Mortality factors were identified and shown to have overlapping influence. Seasonal distribution on various host species was determined.
Develop sampling methodology, action and <sup>b,c</sup> economic thresholds for all major crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Validate and refine sampling methods.		Х	
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day- degree sub-models for estimating phenology and temporal patterns of whitefly, natural enemies and host crops.	Provide model simulation of whitefly populations and multiple cropping systems.		х	
Develop sampling methods for quality of cotton lint, vegetables and other commodities.Develop sampling protocol for and harvest and processing sam and determine interrelationship		Х		Correlations established between thermodetector measurements of cotton stickiness and amounts of honeydew on the fiber.
Quantify whitefly and natural enemy dispersals and contribution to population dynamics.	Determine proportion of whitefly population that are migratory and their reproductive potential.	Х		Trapping studies showed low dispersal for October to early June and evidence for sudden dispersal activity
Define mating behavior, reproductive isolation, species, biotypes.	Define factors involved in mating, cues, feedback mechanisms, etc.		Х	
Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Discuss results, plan additional research, arrive at a consensus decision.	Х		Difference in ecdysteroid titers and waxy particle length, width and morphology documented between <i>Bemisia</i> and other whiteflies.

Research Approaches	Year 3 Goals Statement	Progress A Yes	Achieved No	Significance
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Determine potential for manipulating, interfering with or inhibiting endosymbiont function.		Х	
Characterize nutrient uptake and metabolism	Determine the physical and biochemical processes involved in uptake of carbohydrates, amino acids and other essential nutrients.	Х		Length of stylet determined and correlated with vascular bundle depth. Basic metabolic rate and minimum carbohydrate content to support this rate were determined. Preliminary analysis of amino acid metabolism was conducted.
Develop whitefly artificial diets and natural enemy mass-rearing.	Conduct addition, deletion studies to identify essential nutritional needs.	Х		Optimal conditions of pH and egg load were established. Hatching and survivorship was evaluated on various artificial diets.
<ul> <li>**Pursue specific genetic and biological basis for variability in whitefly biotypes, strains, and species; determine impact of different genetypes/phenotypes on whitefly- mediated transmission and on the epidemiology of virus diseases.</li> <li>Continue with work from prev years. Study impact of biotyp strains, and species difference disease spread, crop damage, a specific control measures to re whitefly vector populations. L with biological and chemical c sections.</li> </ul>			Х	This research approach transferred to section A

<sup>a</sup> Natural enemy research complements from Section D, see Table D.
 <sup>b</sup> Action and economic thresholds also apply in Section C, see Table C.
 <sup>c</sup> Sampling technology applicable to all other sections, see Tables B to F.
 \*\* Transferred from Table B 3/31/2000.

Research Approaches	Year 4/5 Goals Statement	Yes	Achieved No	Significance
Determine life cycle vulnerabilities (life tables) <sup>a</sup> , population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.	Initiate studies to manipulate host sequences to determine potential influence on whitefly population. Continue 4 and finalize analysis of the potentia l of habitat modification as a management tool.	x		Greenhouse studies examined movement and mortality rates of <i>B.</i> <i>tabaci</i> crawlers on cotton. Airflow did not influence crawler survival, which averaged 86%. Crawlers moved as far as 200 mm, and over 99.9% settled on the lower surface of the same leaf where eggs were oviposited. Ongoing studies examined seasonality and mortality patterns of <i>B. tabaci</i> on broccoli, cantaloupe, cotton, alfalfa, <i>Lantana</i> and various weeds in Yuma, Maricopa and Marana, AZ. These sites represented a range of geographic and climatic areas in the state. Cantaloupe and <i>Lantana</i> served as good hosts for <i>B. tabaci</i> population buildups in the fall. In the winter, populations steadily declined. Whiteflies survived the winter best in broccoli and cheeseweed, and shortly thereafter built up in broccoli, cantaloupe and <i>Lantana</i> . Large populations in cantaloupe and <i>Lantana</i> were probably responsible for the initial buildups in cotton. During the summer, ground cherry and other weeds also played a role. Yuma showed the earliest increase in whitefly numbers in the spring, but had lower numbers throughout the season when compared to Maricopa and Marana. Whiteflies survived best on cantaloupe at all sites. Predation and dislodgment accounted for a significant portion of the mortality at all locations. Parasitism was highest in Lantana and at the Maricopa site. In India, age specific mortality factors were investigated for whiteflies reared on potted cassava plants. Adult survivorship depended on time of year. Predation, parasitism and unknown factors accounted for variation in survivorship. Overall survivorship from first instar to adult ranged from 5-55%. Parasitism was the dominant mortality factor from pupa to adult emergence. Natural enemies included aphelinid parasitoids, predatory coccinellids, mites and spiders. Missing and unknown factors were the dominant mortality factor in most experiments, followed by predation and parasitism. In laboratory studies, <i>Serangiu</i> <i>paracestosum</i> was the most important predator.

# Table A. Biology, Ecology, and Population Dynamics, 2001-2002. Pre

		Progress A	Achieved	
<b>Research Approaches</b>	Year 4/5 Goals Statement	Yes	No	Significance
Develop sampling methodology, action and <sup>b,c</sup> economic thresholds for all major	Implement sampling protocols through cooperative extension outlets and other technology transfer methods	Х		In Cukurova, Turkey, studies were conducted to determine the most appropriate sampling unit and spatial distribution of immature and adult <i>B. tabaci</i> , and immature parasitoids. Whitefly eggs were most often associated with cotton leaf positions 2-4; nymphs with positions
crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Finalization, implementation and use in IPM systems.	Х		5-7; pupae with positions 6-10; and immature parasitoids with positions 8-11. Based on Taylor's power law it was most efficient to sample leaf 3 for eggs, leaf 5 for nymphs, leaf 10 for pupae and immature parasitoids, and leaf 4 for adults. The CC trap was modified by coating the trap tops with Tanglefoot. This resulted in a 50% increase in trap catches. The addition of a lime green LED (light- emitting diode) to the trap resulted in a 15-23 fold increase in whiteflies trapped relative to traps with a white LED or no light.
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day-degree sub- models for estimating phenology and temporal patterns of whitefly, natural enemies and host	Identify weak points and needed information to improve model simulations. Validate and expand effort to provide predictive models capabilities for whitefly population development and crop interfaces	X X		In Iran, an engineer-based computer simulation model was used to predict greenhouse whitefly development. Developmental time from egg to adult was longer and mortality was higher particularly during the egg and second nymphal stages with the computer simulation relative to greenhouse data. Incorporation of additional biotic and abiotic factors into the model may improve predictability.

crops.

		Progress Ach	ieved
<b>Research Approaches</b>	Year 4/5 Goals Statement	Yes N	No Significance
Develop sampling methods for quality of cotton lint, vegetables and other commodities.	Extend sampling protocols to textile mill and verify field findings in relation to mill problems. Modify, refine and complete sticky cotton sampling protocols from the field to the mill.	X	Two assay systems for detecting sticky cotton were investigated, the manual thermodetector (SCT) and the high speed (automated) sticky cotton thermodetector (H2SD). The newest H2SD system consistently detected more thermodetector spots than the SCT and the relationship between the platforms appeared to be nonlinear. Examination of many different sizes of field sample units suggested that smaller sample units were more cost-efficient than larger sample units. Further analyses of variance components suggested that only a single assay for each sample unit should be conducted with the SCT, but that 3 assays should be conducted for each sample unit. Even fewer samples ize of about 15 on either platform using the 1-plant sample unit. Even fewer samples would be required using a 20-boll unit. Samples sizes as high as 41 or 82 would be required for estimates with very high precision on the H2SD or SCT platform, respectively. Sampling costs are much lower for the H2SD because samples can be assayed faster. Sweet potato whitefly feeding on plants in the field with furrow plus supplementary drip irrigation and in the greenhouse on non-stressed cotton plants produced more micrograms of honeydew sugars per gram of honeydew compared to whiteflies feeding on plants with furrow irrigation alone in the field and on water-stressed plants in the greenhouse. Thermodetector naalysis was used to determine the effect of days of exposure to <i>B. tabaci</i> and lint stickiness. The effect of rainfall on lint stickiness was also examined. Trehalulose and melezitose produced by <i>Bemissia</i> and thermodetector counts were significantly correlated to amounts of trehalulose and melezitose. Rainfall of 0.5 inches reduced trehalulose and melezitose. SPW produced more trehalulose relatives in cotton lint within 5 h. Sweet potato whitefly and cotton aphid honeydew sugars were compared, and their effect on cotton lint stickiness was addite to the modetector counts, and increasing duration of exposure to both insects.

 Table A. Biology, Ecology, and Population Dynamics, 2001-2002. (Continued)

 Progress Achieved

		Progress	Achieved	
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance
Quantify whitefly and natural enemy dispersals and contribution to	Quantify the role of dispersal in population dynamics on different crop systems.	Х		The flight behavior of male and female <i>Eretmocerus eremicus</i> in response to skylight and plant cues was examined in a vertical flight chamber. Both male and female parasitoids were capable of flying in
population dynamics.	Formulate theory for manipulating and/or using dispersal as a tool in IPM.	X excess of 60 min, but a were presented during t wasps responded to the these wasps were femal consistent with migratio they had flown for an e A separate study, also extended period in the the longest flights. Thi was found that female f	excess of 60 min, but averaged 12 min in duration. When plant cues were presented during the parasitoid's phototactic flight, 63% of the wasps responded to the plant cue shortly after takeoff, and most of these wasps were females. Another 37% exhibited a response consistent with migration. They failed to respond to the plant cue until they had flown for an extended time. Most of these wasps were males. A separate study, also found that <i>E. eremicus</i> could fly for an extended period in the vertical flight chamber. Virgin females made the longest flights. This work was validated in field studies where it was found that female flight was wind-aided and directional. Males flew for shorter distances, mostly within the boundary layer	
Define mating behavior, reproductive isolation, species, biotypes.	Develop potential methods of utilizing behavioral information in management strategies	Х		In India, host-associated variation studies revealed the presence of two strains of <i>Bemisia tabaci</i> – the cassava whitefly and the sweet potato whitefly. Two enzyme systems were used to study variations in the
	Review, summarize and propose additional needed research.	Х		<ul> <li>isozyme patterns: Esterase and Malate dehydrogenase. The esterase-banding pattern in sweet potato whitefly differed from the cassava whitefly in having an additional fast moving esterase band (Rf value 0.293). The migration distances of the two slow moving bands, common to both biotypes were also different (0.15 and 0.204 in CWF and 0.108 and 0.165 in SPWF). Banding patterns using the Malate dehydrogenase system was identical in both biotypes. The ability of the cassava biotype to vector Indian Cassava Mosaic Virus was confirmed by ELISA. The sweet potato biotype was unable to transmit ICMV. In crossbreeding studies an increased proportion of male progeny indicated that cassava and sweet potato are two biotypes and are not separate species.</li> <li>Host-associated variation of <i>B. tabaci</i> in terms of development and survival also were examined. Cassava, sweet potato, egg plant, tobacco, and cotton were used for these studies. Whitefly populations from sweet potato could not colonize cassava and whiteflies from cassava could not colonize sweet potatoes.</li> </ul>

Research Approaches	Year 4/5 Goals Statemer	nt	Progress Achieved Yes No Significance
Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Publish verification of new species or other appropriate taxa	X	YesNoSignificanceA staging system based on body depth (thickness) and maturation of the adult eye was used to track development and identify physiological synchronous 4 <sup>th</sup> instar/pharate adult silverleaf and greenhouse whiteflies. An examination of histological sections of last instar SLW. revealed that adult eye and wing development were initiated in Stage 6 Wings were deeply folded by Stages 6 and 7. Ecdysteroid titers peake at Stages 4-early 6, just before the onset of adult development. Leaf samples were taken from field-grown cotton and cantaloupe, an developmental differences in whitefly length and width were determined. Fourth instar nymphs on cotton were longer and wider than nymphs on cantaloupe leaves. In Mexico, numerous whitefly species have been identified including SPWF, SLWF, the greenhouse whitefly, banded-wing whitefly, woolly whitefly, acacia whitefly and citrus whitefly. Between 1994 surveys and 1996-97 surveys, SPWF has nearly been displaced by SLWF. More than 100 plant species hav been identified as hosts for SLWF. The identification and 
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Determine associated enzymes and/or other endosymbionts and whitefly relationships Summarize and implement findings with suggestion for additional research.	Х	Several different types of bacteria were cultured from surface-sterilized <i>Bemisia argentifolii</i> adults and nymphs, including <i>Bacillus</i> spp. <i>Enterobacter cloacae</i> , was found within the gut cells of adult whiteflied and was mildly pathogenic.

		0	s Achieved	
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance
Characterize nutrient uptake and metabolism	Determine the potential for blocking key steps in nutrient uptake and/or metabolism Implement findings by developing inhibitors of nutrient uptake and/or	Х	Х	It was found that whitefly nymphs feed during all four stages of development. The rostrum of the nymphs is short and has sensilla on the tip, which are similar to those found on the labium of the adult. Stylets range from 110 $\mu$ m in first instar to over 200 $\mu$ m in the fourth instar. Prio to the onset of feeding the crawlers stylet is folded within the body of the nymph. Like the adult whitefly, the nymph produces a flange like materia
	metabolism.			that appears to cement the tip of the rostrum to the surface of the host leaf Prior to each molt, the stylets are withdrawn into the body of the nymph and left behind in the cast exuviae. Most adult whitefly probe sites occurred along the margins of the abaxia epidermal cells. Stylets passed directly through the cytoplasm of the epidermal cell and not through the common radial wall between epiderma cells. Stylets averaged 217 $\mu$ m long. The adult whitefly uses less than 70
				of the length of its stylet to reach a phloem bundle in cotton.
Develop whitefly artificial diets and natural enemy mass-	Evaluate developed diets on whitefly fecundity/longevity biology, behavioral characteristics.	Х		A newly developed artificial rearing system to determine how biotic and abiotic factors influenced egg hatch, crawler establishment and development of SLWF. Egg age significantly influenced hatch rates, and
rearing.	Develop whitefly rearing system and adapt for production of natural enemies.	Х		a lesser extent survival and development of nymphs reared on the artificia diet. There were negative associations between the number of eggs placed on the membranes and both hatch rate and establishment of crawlers. Egg oviposited on and then subsequently removed from plants held under long day conditions (14:10 L:D) or high light intensity had higher hatch rates than eggs oviposited under short-day conditions or low light intensity. Long-day conditions during oviposition also significantly enhanced survival of nymphs through Day 20 and developmental rate for Day 6 counts. Light intensity, at least for the range tested, did not significantly affect development or survival of whitefly nymphs. Recent studies indicated that no matter which host instar <i>E. mundus</i> eggs are laid, penetration into the host proceeds only during the host's fourth instar, typically sometime prior to the transition to adult characteristics. Since approximately three days are required for parasitoid egg development,
				oviposition under a second or third instar whitefly will result in hatch occurring under a third or young fourth instar host. Epidermal capsule formation accompanied parasitoid penetration. The capsule surrounds/engulfs the parasitoid and must contribute to parasitoid development. The question as to whether host ecdysteroid levels are altern upon parasitized whitefly into which a first instar parasitoid had just begu to penetrate than in a healthy whitefly of the same developmental stage.

		Progress .		
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance
**Pursue specific genetic and biological basis for variability in whitefly biotypes, strains, and species; determine impact of different genetypes/phenotypes on whitefly- mediated transmission and on the epidemiology of virus diseases.	Identify potential factors related to specific genetic and biological variability that may be manipulated to reduce disease spread. Develop molecular approaches to track biotypes, strains, and species re lative to disease spread, based on differential molecular markers. Summarize results, identify new research needs and make recommendations for implementation or expansion of research	X		A cell line from the silverleaf whitefly, BtB-2.97- Hunter & Polston was tested for its ability to support replication of the insect iridescent virus 6 (IIV-6). Several lines of evidence indicated that a productive infection was achieved. The cells displayed cytopathic effects, virus particles accumulated in defined areas of the cytoplasm, the cell-associated virus titer was detected at three orders of magnitude higher than that released into the media, and western blot analysis indicated CIV structural proteins were being expressed. Virus was also detected in nymphs by PCR and electron microscopy, but the infections were not highly pathogenic. Infection by whiteflies by IIV-6 suggests that more pathogenic viral isolates may be found for this insect and that the virus has potential to be used as an intracellular probe.

<sup>a</sup> Natural enemy research complements from Section D, see Table D.
 <sup>b</sup> Action and economic thresholds also apply in Section C, see Table C.
 <sup>c</sup> Sampling technology applicable to all other sections, see Tables B to F.
 \*\* Transferred from Table B 3/31/2000

#### **Reports of Research Progress**

#### Section B: Viruses, Epidemiology, and Virus-Vector Interactions Co-Chairs: Bob Gilbertson and Judy Brown

**Investigator's Name(s):** J.K. Brown<sup>1</sup>, A.M. Idris<sup>1</sup>, and J. Bird<sup>2</sup>

**Affiliation & Location**: <sup>1</sup>Univ of Arizona, Plant Sciences Dept; Tucson AZ 85721/<sup>2</sup>Univ of PuertoRico, Rio Piedras, PR 00928

Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions.

## Dates Covered by the Report: 2001

#### Cloning and Molecular Characterization of the A and B Components for Macroptilium Mosaic Virus from Puerto Rico.

Macroptilium mosaic virus (MaMV) is a begomovirus that infects common bean and *Macroptilium lathyroides* in Puerto Rico. Leaf samples were collected from *M. lathyroides* plants exhibiting typical bright yellow mosaic symptoms. Total nucleic acids were extracted and aliquots were incubated with selected restriction enzymes. Digestion products were analyzed by Southern hybridization using PCR-amplified of MaMV A and B component-specific probes. As Cla I digestion linearized dsDNA viral forms, it was used to clone A and B viral components. Recombinant plasmids containing full-length viral inserts were selected and their identity was confirmed by Cla I digestion. DNA sequence comparisons revealed that MaMV shared 66.3 and 59.8 % nt sequence identity with the A and B components, respectively, of its closest relative, *Bean golden mosaic virus PR*. MaMV is a new bean-infecting begomoviral species, and only the second begomovirus identified in the BGMV-Caribbean cluster to date.

Investigator's Name(s): J.K. Brown and A.M. Idris

Affiliation & Location: Univ of Arizona, Plant Sciences Dept; Tucson AZ 85721

Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered by the Report: 2001

### Cloning and Sequencing of Cotton Leaf Crumple Virus -a Begomovirus Infecting Cotton in the Sonoran Desert

Cotton leaf crumple virus (CLCrV) is a bipartite, whitefly-transmitted geminivirus from the southwestern US and Sonora, Mexico that has been known to infect cotton since the 1950's. The CLCrV DNA A and DNA B components for isolates from Arizona and Sonora were cloned and the nucleotide sequence was determined. Sequence comparisons indicated that the DNA A component (GB Accession AF480940) shared the highest nucleotide sequence identities with members of the SLCV group, while the closest relatives for the CLCrV DNA-B component (GB Accession AF480941) were begomoviruses from the Caribbean, Central America, and Mexico. Parsimony and maximum likelihood analyses indicated that CLCrV is the first member of a previously undiscovered begomovirus group from the New World. The Rep binding element within the common region of CLCrV, GGAGT-CT-GGAGT, is 100% conserved for both DNA-A and DNA-B components, indicating they are cognate for the same virus. Lack of phylogenetic congruence between CLCrV DNA A and DNA B indicate that they have evolved along different paths, and/or that recombination may have occurred. Investigator's Name(s): C.L. McKenzie, Robert G. Shatters, Jr., and R.T. Mayer.

Affiliation & Location: USDA-ARS, Horticultural Research Lab, Ft. Pierce, FL

Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered by the Report: 2000

## Differential Plant Response and Whitefly Performance in Comparisons of Whitefly and Whitefly-Tomato Mottle Virus (ToMoV) Complex Challenged Tomato.

Whitefly challenges of tomato are known to induce the accumulation of a class of proteins termed pathogenesis related or PR-proteins. It is also known that in general different types of plant challenges can induce different sets of PR-proteins. The present work was to determine how activity and buildup of a group of PR-proteins responded to an insect (whitefly) versus an insect-virus complex (whitefly-ToMoV) challenge.

Tomato PR-proteins (chitinase, ß-1,3-glucanase, peroxidase, P2 and P4) responses were measured over time in plants divided into three treatments: untreated controls, plants infested with healthy whiteflies, and plants infested with ToMoV carrying whiteflies. Six- to seven-leaf plants were infested with ~5 adult whitefly per leaf. Subsequent plant samples were pulled prior to whitefly infestation and at 14, 28, 42, and 56 days post-infestation for protein and enzyme analyses. By 56 days, there were 2.5- and 4.5-fold more whitefly eggs and nymphs respectively on the plants infested with viruliferous whiteflies than on the healthy whitefly infested plants. A significant increase in the enzymatic activity of all measured PR-proteins, as compared to control plants, was only seen in viruliferous whitefly infested plants. There was no significant difference observed in enzyme activities between uninfested and healthy whitefly infested plants; however, a trend for healthy whitefly induced increases in PR-protein activities was observed. Foliar protein (protein g/leaf (mg)) in tomato did not differ significantly among treatments for any of the sample dates indicating that any significant changes in enzyme levels was a result of differences in the specific measured enzymes. At 56 days post-infestation, virus infected plants exhibited severe virus symptoms and PR enzyme activities declined. The greatest differences for all PR-proteins assayed was observed 42 days after treatment initiation. Western blot analyses showed that the differences in PR-protein activities among the treatments were due to changes in specific enzyme levels within the plant and were associated with concomitant increases in levels of P2 and P4 PR-proteins; however, the level of viral induction of PRs was variable among virus infected plants. Under our experimental conditions, it is clear that the whitefly-ToMoV complex is a much stronger inducer of tomato PRproteins response than whiteflies alone. Because of the increased egg and nymph production by the viruliferous whiteflies, the cause of the increased PR-protein response when the virus is present may be the result of either increased feeding pressure of viruliferous whiteflies or direct interactions of virus and plant.

Investigator's Name(s): C.L. McKenzie

Affiliation & Location: USDA-ARS, Horticultural Research Lab, 2001 South Rock Road, Ft. Pierce, FL

Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered by the Report: 2000-2001

### Tomato Mottle Virus (ToMoV) Effects Whitefly Oviposition and Adult Survivorship on Healthy Tomato

The effect of plant viruses on the reproductive potential of the vector is key to understanding geminivirus epidemiology and developing effective control measures. The objective of this study was to determine the effect of tomato mottle virus (ToMoV) on whitefly oviposition and survival rates on healthy tomato.

Adult *B. tabaci* biotype B were obtained from laboratory colonies maintained by the U.S. Horticultural Research Laboratory, Ft. Pierce, FL. Whiteflies used in these experiments were originally obtained from Dr. Lance Osborne, University of Florida, Apopka, FL and have been maintained on dwarf cherry tomato (*Lycopersicon esculentum* cv. Florida Lanai) since 1996 by serial transfer. In 1997, a ToMoV whitefly colony was established by obtaining tomato plants infected with ToMoV from Dr. Philip Stansly, University of Florida, Immokalee, FL and infesting with whiteflies from the healthy colony. Whitefly biotyping was based on RAPD PCR analysis using primers developed by De Barro and Driver (1997). Nonviruliferous and viruliferous whitefly colonies were housed separately in screened Plexiglass cages located in separate growth chambers at 25  $\pm$  1°C and a 16:8 L:D photoperiod. Whiteflies from the viruliferous colony were confirmed to be infected with ToMoV prior to infestation by PCR analysis.

In each experiment, one male and one female whitefly of unknown age from healthy or ToMoV-infected whitefly colonies were confined in clip cages attached to the terminal leaf of the 3<sup>rd</sup> fully expanded leaflet of a healthy cv. Florida Lanai plant. Clip cages were made from clear plastic cups (PC100 30 ml cups, Jet Plastica, Harrisburg, PA) fitted with a foam seal on the bottom and an organdy window on top for ventilation. The foam bottom was backed with a thin square of balsa wood, and an aluminum hair clip was glued to the balsa wood and cup portion. Whiteflies were introduced through a small hole in the side of the cup. After a 48-hr access period, adult whiteflies were removed and eggs were counted. Treatments were maintained separately at  $25 \pm 1^{\circ}$ C and a photoperiod of 16:8 L:D. For each experiment, 20 test plants were typically used for each treatment; however, the final number of replicates (=clip cages) per treatment varied when leaves of test plants died or were severed during the experiment. A minimum of 8 replicates was used for all treatments. Experiments were repeated five times. There were no significant interactions between experiment\*treatment (F=2.0; df=4,80; P=0.10) or treatment\*clip cage (F=0.66; df=19,80; P=0.85) so results were pooled over experiments (*n* = 15) were used to include survival to adult emergence which was evaluated 30 days after egg lay to ensure that all viable whiteflies had emerged.

Whiteflies infected with ToMoV deposited significantly more eggs (F=19.51; df=1,80; P < 0.01) on healthy tomato leaves than nonviruliferous whiteflies (~40%). There was no significant difference between virus-infected and nonviruliferous whiteflies for the number of adults emerged or the proportion of those adults surviving from the egg stage. There was no significant correlation between the number of eggs deposited per female and progeny survival rates on healthy tomato for whitefly infected with or without the virus.

In our experiments, whiteflies were well adapted to the host plant, either with or without ToMoV. High survival of both healthy and ToMoV-infected whitefly reflect this host-plant adaptation. Plants from the virus treatment exhibited characteristic ToMoV symptoms 30 days after clip cages were removed. This suggests adaptation to the host plant and virus by the vector could override any adverse effect the virus had on host plant physiology. Insect adaptation to the host-plant is a critical factor that should be considered on a host-by-host basis when evaluating insect biology and vector-host-plant interactions for polyphagous insect species.

Investigator's Names: R.T. McMillan, Jr., M. J. Davis, and Z. Ying

Affiliation & Location: University of Florida, TREC, Homestead, FL

Research & Implementation Area: Section B : Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered by the Report: 1997-2001

#### Tomato yellow leaf curl geminivirus Management in South Florida

Tomato yellow leaf curl geminivirus (TLCV) was identified in the United States for the first time in late July 1997 in a field planting at a commercial breeding facility in Florida. Shortly thereafter, infected tomato plants were also found in several retail garden outlets in Florida(personal communication J. Polston). The source of these plants was traced back to two commercial nurseries near Homestead in south Florida that had shipped tomato plants to retail outlets in Florida and other states and out of the country. Subsequently, infected tomatoes from retail garden outlets have been found in Virginia (confirmed by laboratory tests) and possibly other locations in the southern United States supporting the possibility that TYLCV will become a regional problem.

Due, apparently, to the introduction and spread of the silver leaf whitefly, there has been a recent emergence of seventeen whitefly-transmitted geminiviruses in tomato in the Western Hemisphere. Of these, only tomato mottle virus was present in Florida before TYLC. We used polymerase chain reaction (PCR) and DNA sequence analyses to confirmed the recent introduction of TYLCV into Florida. Degenerate primers for geminiviruses were used to amplify a fragment of the viral genome containing part of the coat protein gene. The partial gene sequence had greater than 98% homology with that of an Israel strain of the TYLCV. These results suggest that the virus is an Eastern Mediterranean strain and possibly the same strain previously introduced into the Dominican Republic from Israel in 1991 and subsequently found in Jamaica and Cuba. We have designed non-degenerate primers for PCR detection of TYLCV based on our DNA sequence data for the virus. Detection of TYLCV in tomato plants with these primers has been 10-100 times more sensitive than with the degenerate primers, which even with the degeneracy have mismatched bases.

Tomatoes are a winter crop in south Florida, and TYLCV was detected in October, 1997, in newly established commercial plantings. Tomatoes are still being planted in south Florida at the present time. The incidence of TYLCV-infected plants initially appeared to be due mostly to primary spread into the fields. Sources of inoculum are presumed to be weeds outside of the tomato fields.

Several studies have been conducted at TREC to manage SLW in tomatoes using insecticides of various classes. Most of these studies are yet to be completed. Several generalist predators (minute pirate bug, lady bug, lacewing) were evaluated in the laboratory for control of SLW, but none of them provided satisfactory control. Compatibility of these predators with various chemical and biological insecticides were also determined. All foliar sprays significantly (90-100%) affected survival of these predators. Soil drench of imidacloprid did not cause any mortality of SLW predators. Various fungal insecticides were also evaluated in the laboratory and greenhouse situations to control SLW. A commercial formulation of *Beauveria bassiana* (Mycotrol®) was consistently more effective than *Paecilimyces fumosoroseus* (PFR-97®). The performance of *Acromonium* and *Verticillium lecanii* was inconsistent. Compatibility of these fungal insecticides with fungicides was also determined. Control of SLW with both *B. bassiana* and imidacloprid offers promising possibilities and needs to be evaluated for management of TYLCV.

Whitefly population densities have been generally low, and even less in tomatoes due to the extensive application of the systemic insecticide, imidacloprid, to transplants. However, the occurrence and incidence of TYLCV in cultivated tomatoes have steadily increased during the growing season. Secondary spread within fields has become more prevalent and whitefly population densities are on the increase. TYLCV appears to be firmly established in Florida, and will continue to be a major problem in the region in years to come.

Florida Agricultural Experiment Station, Journal Series No. N-02188.

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Research & Implementation Area: Section B: Viruses, Epidemiology and Virus-Vector Interactions

Dates Covered by the Report: January 1, 2001 - December 31, 2001

## Whitefly-borne Virus Epidemiology: Lessons Learned from the Greenhouse Whitefly and Tomato Infectious Chlorosis virus

We sampled 57 weed species and 13 crop species in Orange County, California in order to describe the host range and seasonality of greenhouse whitefly (GHWF). This survey identified 21 weed hosts and 9 crop hosts of GHWF. The agricultural areas in the survey region are farmed intensively; most land carries 2 to 3 crops per year. Strawberries are planted in September and October and harvested from the following winter into summer. Zucchini squash and cucumbers are grown year-round; young plants are grown under plastic tunnels in late winter, while old plantings are harvested into early and mid- winter as long as the plants survive. Beans generally are present except in midwinter. Tomatoes are planted from winter, in tunnels until summer, and are harvested from spring into December. Thus crop plants are available throughout the year as GHWF hosts.

The most consistent aspect of GHWF infestation of these crops was that most fields surveyed had very few whiteflies. Very few fields that had low numbers of GHWF were found to be associated with a GHWF source, while fields with a maximum GHWF count of greater than 100 were, in almost every case, near a source of GHWF. Closer examination of fields typically revealed that there was a sharp decrease in GHWF numbers with increased distance from the whitefly source. GHWF outbreaks over the past several years were not regional but local in nature. Growers can minimize their own GHWF infestations by distancing new plantings from sources of infestation, by interrupting serial plantings which perpetuate infestations, and by eliminating infested crop residues as soon as possible after harvest. In our survey we found that some fields separated by as little as 0.1 mile from GHWF sources escaped even moderate infestation.

The ability to control GHWF levels is most important for tomato growers, because those numbers affect tomato infectious virus (TICV) infection. In epidemiological studies, we found that disease levels were low and seldom occurred before the onset of harvest. Tomato fields categorized according to their maximum GHWF levels revealed that few lightly infested fields became infected with TICV, while over half of the fields with high GHWF numbers were found to have virus. Additional data from individual sites supported the connection between GHWF numbers and rates of TICV infection, particularly when the whitefly source was tomato. The presence and proximity of GHWF sources strongly influenced subsequent infestations and local TICV epidemics.

Investigator's Name(s): Yash Pal S. Rathi

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Research & Implementation Area : Section B: Viruses, Epidemiology, and Virus-Vector Interactions

Dates Covered by the Report : 1998-1999, 1999-2000

## Frenchbean Crinkle Stunt: Epidemiology and Management

Crinkle stunt, a recently identified disease has become a potential threat to the cultivation of frenchbean (*Phaseolus vulgarls L.*) particularly in the *Tarai* and Hill regions of Uttranchal and Uttar Pradesh states of India. The disease incidence was higher in Rabi as compared to spring planting season. The diseased plants remain stunted showing characteristic symptoms of leaf rolling, crinkling, rugosity and vein chlorosis with dark green colour of the leaf lamina. Diseased plants show complete sterility or bear a very few pods. Yield losses may go even up to 100 per cent in case of a susceptible genotype if infected at an early stage of crop growth. Transmission studies showed that the frenchbean crinkle stunt virus (FbCSV) was not seed, soil, sap or dodder transmissible. It is vectored only by whitefly (*Bemisia tabaci* Genn.). For host range studies, 87 varieties of 50 host species belonging to 10 fa milies, were inoculated with viruliferous whiteflies, however, the virus remained restricted only to frenchbean.

The virus (FbCSV)-vector (*Bemisia tabaci*) relationship studies were conducted in glasshouse. The minimum acquisition and inoculation feeding periods were 6 and 4 hrs, respectively. Three hrs starvation before acquisition and inoculation accesses increased the transmission efficiency of the whitefly adults. An incubation period of 12 hrs was observed in the vector. Singly whitefly adult was able to transmit the virus. However, increased number of viruliferous whitefly adults per test plant, increased percentage of transmission. The viruliferous adults were infective for a maximum period of 7 days indicating that virus is the persistent (circulative). Female whitefly adults were more efficient vectors as compared to male adults.

Delayed planting (Oct- 30), narrow plant spacing (30 x 10 cm) & increased levels of N alone or in combination with P not only significantly decreased the disease incidence but also increased the yield. Two insecticidal (Monocrotophos 0.1% + Endosulfan 0.1%) sprays at 10 day interval decreased the population of whiteflies considerably. Out of 50 germplasm lines 15 were found resistant after artificial screening in glasshouse. Since none of the individual control measure is effective, an integrated management strategy has to be developed.

## Investigator's Name(s): Y.P.S. Rathi

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Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered by the Report: Last 4 Kharif seasons

## Mungbean Yellow Mosaic Virus: Epidemiology and Management

Mungbean yellow mosaic virus (MYMV) also called as a "yellow plague" of *Kharif* pulses is vectored by whitefly (*Bemisia tabaci* Genn.) in a persistent circulative manner. Currently, it is a number one problem causing substantial yield losses in urdbean (*Vigna mungo*), mungbean (*V. radiata*), mothbean (*V. aconotifolia*) and soybean (*Glycine max*) in India. The host crop exhibits general yellowing and the diseased fields can be recognized from far distance.

Studies on epidemiology in *Tarai* region of Uttaranchal State revealed that besides the population of whiteflies, weather parameters like temperature, rainfall and humidity play important role in the development of the disease epidemic. Whitefly population increased with increase in temperature. High relative humidity, heavy showers and strong winds in rainy season were found detrimental to whitefly adults. Further studies on host range revealed that ratoon crop of pigeonpea and/or some other weeds might be serving as primary source of inoculum.

Experiments were conducted at Crop Research Centre of Pantnagar University to contain this disease through agronomic practices (manipulation in planting dates and inter- cropping with barrier (non-host) crops, insecticides (foliar and soil application, seed treatment) and host resistance. Low disease incidence was recorded in early (June) and late (August) plantings as well as in low plant spacing (5 cm). Seed dressing with carbofuran 3-G or phorate l0-G with two foliar sprays of monocrotophos (0.1%) + endosulphan (0.1%) also reduced the disease incidence. Spray of 2% mineral oil + 0.5% Neem oil + 5% detergent + 0.1% monocrotophos was equally effective. Inter-cropping of non host crops with mungbean, soybean and urdbean were not found very effective barriers. Some genotypes viz., Pant U 19, 30, 35 and PU-1 GD (urdbean), PM 1, 2, 3 & 4 (mungbean) and PK 416, PK 564, PK 1029, SL 142 (Soybean) resistant to MYMV have been identified.

None of the individual management approach was found highly effective against the vector/disease due to high vector population and wide range of host plants serving as initial foci of the whitefly vector and the virus through out the year. Moreover a single whitefly adult is sufficient to initiate the infection. The only promising way of effective management of this yellow plague (MYMV) appears to be the development of integrated management strategy having tolerant varieties in core.

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Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus-Vector Interactions

## Dates Covered By The Report: January 2000 - December 2000

## Integrated Management of Bemisia tabaci Genn. and Yellow Mosaic Virus in Arid Legumes

Mothbean, Mungbean and cowpea (*Vigna aconitifolia, V. unguiculata* and *V. radiata*) are the important legumes commonly grown in the arid region of India. But the productivity of these crops is extremely poor (125-250 kg /ha). The incidence of Whitefly, Ben*isia tabaci* Genn. and the heavy incidence of yellow mosaic virus (YMV) are the major factors affecting the yield of these important arid legumes. Field studies conducted at Central Arid Zone Research Institute, Jodhpur have revealed that both whitefly and YMV are responsible for the yield loss of 15.23 to 73.15 percent in different cultivars of these crops. Four hundred lines of these crops were screened for their susceptibility to the incidence of whitefly and YMV. Seven cultivars of mothbean viz. IPCMO-943, IPCMO-1035, T-16, T-2, Jadia, PLMO-240 and PLMO-216 and four of cowpea i.e. JC5, JC10, HFG42-1 and FS-68 and two cultivars of mungbean V7 and V8 were isolated to be least susceptible to the attack of whitefly and YMV. Recommendation of early sowing preferably in the 1<sup>st</sup> or 2<sup>nd</sup> week of July and cultivation of above listed seed material is most profitable in these arid legumes. In the early sowing of these cultivars, the loss due to whitefly and YMV is minimised (14%) as compared to the late sown local cultivars were the loss can extend up to 35 to 54 percent.

Neem oil, quality Neem seed preparations and the commercial available Neem products were tested both under laboratory and field conditions against whitefly and YMV. The quality Neem seeds selected and processed were from the candidate plus trees which contain 48.23 percent oil and 0.892 percent Azadirachtin. These plant extracts were tested either alone or as mixture with a synthetic insecticide (monocrotophos) for the efficacy in the control of whitefly and YMV. Whitefly population per plant varied significantly in different treatments. Subsequently, there was significant reduction in YMV in all the three arid legumes. Use of monocrotophos was superior to all treatments followed by the treatment of Neem oil and Neem seed extracts. The use of Neem oil mixture with synthetic insecticide gave the highest yield of 875 kg /ha and was at par with Neem oil treatment (846kg/ha) followed by NSKE with monocrotophos (823 kg/ha) in mungbean crop. The data reported also involve the utilization of different management strategies in a compatible manner and designed to suppress the pest population and disease in these arid legumes. The use of Neem products either alone or in combination with synthetic insecticide will help to minimise the use of insecticide and conserve the beneficial organisms. This will also help to maintain the ecological situations with long-term benefits.

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Research & Implementation Area: Section B: Viruses, Epidemiology, and Virus Vector Interactions

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## Complementation for Transmission by Non-vector Whiteflies among Tomato-infecting Criniviruses

*Tomato chlorosis crinivirus* (ToCV) and *Tomato infectious chlorosis crinivirus* (TICV) appear to have largely distinct geographical distributions, but have been found together in field-grown tomato. TICV is transmitted only by *Trialeurodes vaporariorum*, while ToCV is transmitted by *T. vaporariorum*, *T. abutilonea* and *Bemisia* species. Both viruses have similar genome size and organization, suggesting the potential exists for transmission by non-vector whiteflies from mixed infections. We established *Physalis wrightii* source plants, containing either TICV alone, ToCV alone, or both viruses together, confirmed by northern blot to virus specific probes. *T. vaporariorum* and *T. abutilonea* were allowed to feed separately on all virus sources, as well as virus-free plants for 24 hours, then were transferred to young host plants. Symptomatic plants were tested by northerns as before, and transmission of TICV by *T. abutilonea*, and *B. argentifolii* were allowed to feed initially on ToCV source plants, followed by TICV source plants, then transferred to test plants did not result in transmission of TICV by non-vector whiteflies. Although cross-transmission does not appear to occur frequently, this rate is substantial considering the high whitefly populations that can occur in the field. Cross-transmission also suggests that genetic compatibility between these viruses may be possible, and could lead to evolution of TICV strains with expanded vector transmissibility.

#### Section B. Summary

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#### Begomoviruses.

Cotton leaf crumple virus (CLCrV). CLCrV is a bipartite, whitefly-transmitted geminivirus from the southwestern US and Sonora, Mexico that has been known to infect cotton since the 1950's. The CLCrV DNA A and DNA B components for isolates from Arizona and Sonora were cloned and the nucleotide sequence was determined. Sequence comparisons indicated that the DNA A component (GB Accession AF480940) shared the highest nucleotide sequence identities with members of the SLCV group, while the closest relatives for the CLCrV DNA-B component (GB Accession AF480941) were begomoviruses from the Caribbean, Central America, and Mexico. Parsimony and maximum likelihood analyses indicated that CLCrV is the first member of a previously undiscovered begomovirus group from the New World. The Rep binding element within the common region of CLCrV, GGAGT-CT-GGAGT, is 100% conserved for both DNA-A and DNA-B components, indicating they are cognate for the same virus. Lack of phylogenetic congruence between CLCrV DNA A and DNA B indicate that they have evolved along different paths, and/or that recombination may have occurred.

Crinkle stunt disease (CLD). CLD is a new whiteflytransmitted viral disease of frenchbean (Phaseolus vulgarls L.) in the Tarai and Hill regions of Uttranchal and Uttar Pradesh states, India. Diseased plants are stunted and exhibit leaf rolling, crinkling, veinal chlorosis, and a dark green leaf lamina, and yields are severely reduced. Transmission studies showed that the Frenchbean crink1e stunt virus (FbCSV) is vectored by whitefly (Bemisia tabaci Genn.) and is not seedborne. Host range studies revealed that of 50 species in 10 families examined, only frenchbean is a virus host. The minimum acquisition and inoculation feeding periods were determined as 6 and 4 hrs, respectively. A12 hr latent period was observed and single whiteflies transmitted the virus. Viruliferous adults transmitted virus for 7 days. Females were more efficient than males as vectors. Delayed planting, narrow spacing (30 x 10 cm, and increased N applications, alone or in with P, decreased disease incidence and increased yields. Two insecticidal (Monocrotophos 0.1% + Endosulfan 0.1%) treatments at 10 day intervals reduced whitefly populations. Fifteen of 50 germplasm lines examined under glasshouse conditions exhibited virus resistance.

**Macroptilium mosaic virus (MaMV).** MaMV is a begomovirus that infects common bean and *Macroptilium lathyroides* in Puerto Rico. Total nucleic acids were extracted and aliquots were incubated with selected

restriction enzymes. Digestion products analyzed by Southern hybridization using PCR-amplified of MaMV A and B component-specific probes indicated Cla I digestion linearized dsDNA viral forms, thus it was used to clone A and B viral components. Recombinant plasmids containing full-length viral inserts were selected and their identity was confirmed by Cla I digestion. DNA sequence comparisons revealed that MaMV shared 66.3 and 59.8 % nt sequence identity with the A and B components, respectively, of its closest relative, *Bean golden mosaic virus PR*. MaMV is a new bean-infecting begomoviral species, and only the second begomovirus identified in the BGMV-Caribbean cluster to date.

#### Mungbean yellow mosaic virus (MYMV). MBYMV

causes significant loss of urdbean (Vigna mungo). mungbean (V. radiata), mothbean (V. aconotifolia) and soybean (*Glycine max*) crops in India. Studies on disease epidemiology in Tarai region of Uttaranchal State revealed that whitefly pressures and weather parameters (temperature, rainfall and humidity) influence epidemic development. Agronomic practices to reduce disease pressures were examined, including manipulation of planting dates, inter-cropping with barrier (non-host) crops, insecticide treatments (foliar and soil application, seed treatment), and planting of disease resistant varieties. Disease incidence was low in early (June) and late (August) plantings and in low plant spacing (5 cm). Seed dressing with carbofuran 3-G or phorate 10-G with monocrotophos (0.1%) + endosulphan (0.1%) treatment also reduced disease incidence. A treatment of 2% mineral oil + 0.5% Neem oil + 5% detergent + 0.1% monocrotophos was as effective. Non-host crops intercropped with mungbean, soybean, and urdbean were not effective virus or vector barriers. No management approach alone was highly effective against the vector/disease due to high vector population pressures and the availability of a broad range of plants that are virus and/or whitefly vector hosts, indicating that disease management requires an integrated approach that includes resistant varieties.

At the Central Arid Zone Research Institute, Jodhpur, whitefly and yellow mosaic disease (caused by MBYMV) are responsible for 15.23 to 73.15 percent losses in cowpea, mothbean, and mungbean. Four hundred lines were screened for susceptibility to whitefly and YMV. Seven cultivars of mothbean: IPCMO-943, IPCMO-1035, T-16, T-2, Jadia, PLMO-240 and PLMO-216, four of cowpea: JC5, JC10, HFG42-1 and FS-68, and two cultivars of mungbean: V7 and V8 were least susceptible whitefly and YMV pressures. Early sowing (preferably in the 1<sup>st</sup> or 2<sup>nd</sup> week of July) of tolerant germplasm is recommended, based on only14% losses, compared to the 35-54% losses when local cultivars are planted later in the season. Neem oil, quality Neem seed preparations, and commercially available Neem products were tested under laboratory and field conditions against whitefly and YMV. The quality Neem seeds selected and processed were from the candidate trees which contained 48.23 % oil and 0.892% Azadirachtin. These extracts were tested alone or as mixture with a synthetic insecticide (monocrotophos) for whitefly and YMV control. Results indicated a significant reduction in virus damage in all three legume crops. Monocrotophos was superior to all treatments, followed by Neem oil, and Neem seed extract. Neem oil was more effective when mixed with the synthetic insecticide than when applied alone, resulting in the highest yields (875 kg /ha), and was at par with Neem oil treatment (846kg/ha), followed by NSKE with monocrotophos (823 kg/ha). Uses of Neem products either alone or in combination with synthetic insecticides will also aid in protect ion of beneficial organisms.

Tomato mottle virus (ToMoV). Experiments were conducted to determine the effect of ToMoV on whitefly oviposition rates and adult survivorship. In 3 of 6 experiments, significantly more eggs were oviposited by viruliferous whiteflies compared to non-viruliferous controls. Although there was no significant difference between treatments for half of the experiments, a trend for higher oviposition by viruliferous whiteflies was observed. Data combined for all dates revealed a significant difference in whitefly oviposition. Adult survivorship was significantly greater for viruliferous whiteflies for one replicate, while no significant difference was seen in two others. When data were combined, the percent eggs developing into adults was the same (~90%), but the number of adults surviving between treatments was significant. Virus assays revealed that not all whiteflies harbored ToMoV, suggesting an explanation for somewhat inconsistent results.

The effect of ToMoV on whitefly oviposition and survival rates on healthy tomato was investigated. Whiteflies exposed to ToMoV deposited more eggs on healthy tomato than did nonviruliferous whiteflies. There was no significant difference between virus-infected and nonviruliferous whiteflies in number of adults emerged or proportion of adult survival. There was no significant correlation between the number of eggs deposited and progeny survival rates on virus-free tomato for viruliferous versus non-viruliferous whitefly. High survival of whiteflies colonizing both virus-free and ToMoV-infected plants reflected adaptation of whiteflies to their host. Virusinfected plants exhibited characteristic ToMoV symptoms 30 days after inoculation, suggesting that host adaptation by the whitefly may counter the otherwise adverse effects that virus infection may have on host physiology. Hostadaptation of the whitefly vector to the host plant is an important consideration to vector-host-plant studies.

A study was carried out to evaluate PR-proteins in plants exposed to whitefly compared to insect-virus complex (whitefly-ToMoV) challenge. Tomato PR-proteins (chitinase,  $\beta$ -1, 3-glucanase, peroxidase, P2 and P4) responses were measured over time in plants for three treatments: untreated control, whitefly-infested, whitefly exposed to ToMoV infected tomato. At 56 days after infestation, 2.5- and 4.5-fold more whitefly eggs and nymphs, respectively were found on plants infested with viruliferous whiteflies than for the other two treatments, together with a significant increase in enzymatic activity for PR-proteins. Amount of leaf protein (gm/mg)) did not differ significantly among treatments indicating that significant changes in enzyme levels were due to differences in specific enzymes. At 56 days postinfestation, virus infected plants exhibited severe disease symptoms, concomitant with a decline in PR enzyme activities. Western blot analyses indicated that differences in PR-protein activities were due to changes in levels of specific enzymes, and were associated with an increase in P2 and P4 PR-proteins. Thus, the whitefly-ToMoV complex more strongly induces PR-proteins in tomato that does whiteflies alone. PR-protein induction may be due to whitefly feeding or to interactions between virus and host plant.

Several generalist predators (minute pirate bug, ladybug, and lacewing) were evaluated in the laboratory for control of SLW to reduce ToMoV incidence, but none of them provided satisfactory control. Compatibility of these predators with various chemical and biological insecticides was examined. Foliar sprays significantly (90-100%) affected survival of predators, whereas, a soil drench of Imidacloprid did not cause predator mortality. Various fungal insecticides were also evaluated in the laboratory and greenhouse situations to control the whitefly vector. A commercial formulation of Beauveria bassiana (Mycotrol®) was more effective than Paecilimyces fumosoroseus (PFR-97®). Performance of Acromonium and Verticillium lecanii treatments was inconsistent for both. Whitefly control using *B. bassiana* plus Imidacloprid offe red promising possibilities for virus disease management by reducing vector populations.

Tomato yellow leaf curl virus (TYLCV). TYLCV was introduced into Florida in1997 and infects vegetable and nursery crops. The source of these plants has been traced to two commercial nurseries near Homestead in south Florida that shipped tomato plants to retail outlets in Florida, other locations in the US, and as exports abroad. Subsequently, infected tomatoes from retail garden outlets were documented in Virginia and possibly other locations in the southern US, suggesting that TYLCV-IS will become a regional problem. Polymerase chain reaction (PCR) using degenerate primers for a fragment of the coat protein gene (CP) and DNA sequencing of amplicons were employed to identify the specific viral strain or species. The CP sequence for the Florida isolate shared greater than 98% identity with an Israel strain of the TYLCV (TYLCV-IS) and is probably the same as that introduced into the Dominican Republic in 1991, and subsequently in Jamaica and Cuba. Non-degenerate primers for TYLCV were designed for specific detection

of TYLCV-IS by PCR and were 10-100 times more sensitive than degenerate CP primers.

#### Criniviruses.

Tomato infectious chlorosis virus (TICV). A survey of 57 weed species and 13 crop species was carried out in Orange County, California to identify the host range and seasonal occurrence of the greenhouse whitefly (GHWF). Results indicated 21 weed hosts and 9 crop hosts, including strawberries, squash, tomato, and cucumber, some of which are cultivated year-round. Most fields had very few whiteflies and outbreaks were localized, not regional. Thus, growers can minimize GHWF infestations by distancing new plantings from sources of infestation, interrupting serial plantings, and eliminating infested crop residues after harvest. Some fields separated by as little as 0.1 mile from GHWF sources escaped moderate infestation. Ability to control GHWF is most important for tomato growers, because this whitefly is a vector of TICV in tomato. Disease incidence was low and infection seldom occurred before the onset of tomato harvest. Tomato fields categorized according to their maximum GHWF levels revealed that few lightly infested fields were infected with TICV, while over half of the fields with high GHWF numbers also harbored virus. Thus, presence and proximity of GHWF sources strongly influences local TICV epidemics.

Tomato chlorosis virus (ToCV) and TICV. ToCV and TICV have largely distinct geographical distributions, but may occur together in field-grown tomato. TICV is transmitted by Trialeurodes vaporariorum, while T. vaporariorum, T. abutilonea and the B biotype of Bemisia tabaci (Genn) transmit ToCV. (syn. B. argentifolii). Both viruses infect tomato, suggesting the potential for transmission by a non-vector whitefly when viruses occur in a mixed infection. Experiments were conducted in which T. vaporariorum, T. abutilonea, and B. tabaci B biotype were allowed to feed on ToCV source plants, followed by TICV source plants, prior to transfer to test plants, revealing that TICV was not transmitted by nonvector whiteflies. However, transmission of TICV by T. abutilonea from a mixed virus infection was confirmed in two of sixty plants. Although such cross-transmission was infrequent, the frequency could be substantial under high whitefly vector pressures. Cross-transmission also suggests the possibility for genetic compatibility between these viruses that could facilitate evolution of TICV strains that are transmissible by additional vector species.

Research Approaches	Year 1 Goals Statement	Progress . Yes	Achieved No	Significance
Identification and characterization of new or emerging whitefly-transmitted viruses and strains.	Monitor crops for presence of whitefly- transmitted diseases, and determine relative disease incidence. Begin virus identification and strain differentiation.		х	Rapid techniques are available for identification and characterization of geminiviruses through sequencing of PCR-amplified viral DNA fragments. This approach was used to show 98% sequence identity between the tomato yellow leaf curl gemini virus (TYLCV) from the Dominican Republic and an Eastern Mediterranean virus strain indicating that the virus was probably introduced on tomato transplants from the Eastern Mediterranean area The use of such sequences in comparisons of viruses are important in establishing their relatedness and origin. Several other assays are available for rapid detection of geminiviruses such as dot blot and squash blot hybridization analysis.
Molecular epidemiology: identification of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses.	Monitor and identify host plants, virus reservoirs in affected areas. Linkages to diagnostic methods for virus ID and tracking.		Х	The use of squash blot analysis using a TYLCV- specific DNA probe to assess the role of weeds as hosts in the Dominican Republic showed that they were not infected with TYLCV and not significant molecular sources for the virus. TYLCV newly discovered in Florida was also 98% identical to the Dominican Republic strain. Geminiviruses, are known throughout the world and distinct viruses are known to occur in many countries. For instance, tomato mottle virus ToMoV) was first detected in Florida in 1989 and is thought to have originated from that state.

# Table B. Viruses, Epidemiology and Virus Vector Interactions.

Research Approaches	Year 1 Goals Statement	Progress A Yes	Achieved No	Significance
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission.	Initiate studies on virus-vector interactions and on basis for the specifcity of whitefly -mediated geminivirus transmission.		X	Studies on feeding duration and position has demonstrated differences in aphids and whiteflies. These differences may determine why some geminiviruses are transmitted by one group and not the other. The use of the autoflourescent GFP gene, in tracking the virus movement and replication in plants indicated that a cell to cell movement of the virus occurred and the virus was not phloem limited. Understanding the movement of the virus in terms of insect feeding behavior may play a role in developing resistant varieties.
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Develop approaches to managing cropping systems to reduce vector densities to decrease transmission frequency and inoculum sources, taking into account weed and crop reservoirs in disease incidence and distribution.		Х	Host-free practices used in the Dominican Republic for TYLCV have been successful in reducing the incidence of this disease. In Florida, management of whiteflies with insecticides, field sanitation, and clean transplants has reduced the incidence of ToMoV. In whitefly reduction studies using biological control based IPM, there was a 10% reduction in geminiviruses in squash (See Table D).
Control of virus diseases: development of virus resistant germplasm through conventional and engineered/ molecular approaches. Define prospective strategies for selecting candidate viruses, identifying specific virus diseases to target, and prioritize specific crops and cultivars for protection approaches.	Define strategies for resistance efforts. Identify target viruses. Identify germplasm with virus resistance. Initiate efforts toward defining prospective engineered resistance strategies. Identify candidate crops and recipient cultivars.			Resistance to the geminivirus, bean dwarf mosaic virus (BDMV), was found in Pinto bean variety, Othello. Using the GFP gene as a marker, virus infection in this variety was compared with that in a susceptible variety. In the resistant variety, there was a collapse of tissue at the infection site and continuing necrosis in the vascular areas indicating a hypersensitive reaction to the virus. The gene(s) involved in this response may be a source of resistance to this virus either through conventional breeding efforts or by identifying the gene(s) involved. In cotton, some resis tance to the cotton leaf crumple virus was reported (See Table F).

# Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued)

		Progress A	Achieved		
Research Approaches	Year 1 Goals Statement	Yes	No	Significance	
Pursue specific genetic and biological	Identify differences in species, strains			No reports in this area.	
basis for variability in whitefly	and biotypes with respect to				
biotypes, strains, and species;	transmission, host range, mating				
determine impact of different	compatabilities, molecular variability,				
genetypes/phenotypes on whitefly-	and map the biogeographic distribution				
mediated transmission and on the	of distinct types within the <i>B. tabaci</i>				
epidemiology of virus diseases.	species complex.				

# Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued)

Research Approaches	Year 2 Goals Statement	Progress A Yes	Achieved No	Significance
Identification and characterization of new or emerging whitefly-transmitted viruses and strains.	Virus identification and characterization. Develop methods for identifying causal agents and for tracking viruses and strains using molecular methods.	X		<ol> <li>Significant progress has been made in the detection and characterization of tomato yellow leaf curl geminivirus in Florida. A comprehensive survey of the incidence and distribution of TYLCV has been made.</li> <li>Evidence has been obtained of a synergistic interaction among three geminivirus DNA components associated with chino del tomate disease of tomato (pepper huasteco geminivirus [PHV] DNA-A and DNA-B and another distinct DNA-A component, chino-A). Here, the disease symptoms induced in three hosts (Nicotiana benthamiana, tomato, and pepper) by PHV plus the chino-A are much more severe than symptoms induced by PHV alone. These results establish that (i) chino del tomate disease may be caused by a complex of geminivirus components, (ii) that complexes of geminivirus components can dramatically influence disease symptom expression and (iii) that identification of geminiviruses based on disease symptoms alone is difficult.</li> <li>Tomato geminivirus diseases in Guadeloupe are caused, at least in part, by a strain of potato yellow mosaic geminivirus (PYMV).</li> </ol>

# Table B. Viruses, Epidemiology and Virus Vector Interactions.

Molecular epidemiology: identification of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses.       Continue field studies. Determine economic input of diseases on crop production and associated losses.       1. The spread of TYLCV in Florida has been extensively documented. The virus has been disseminated throughout the state, including some northern counties. The highest incidences of TYLCV have been correlated with high populatio of whiteflies. Extensive host range studies are being conducted with TYLCV in Florida, and TYLCV have been found to infect and cause diseas in petunia and common bean. Detection in petunia could have serious implications in terms of exporting this ornamental plant.         2. Efforts are being conducted to understand how TYLCV survives in the Dominican Republic durin the three-month whitefly host-free period. Using a	Research Approaches	Year 2 Goals Statement	Progress A Yes	Achieved No	Significance
of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses. economic input of diseases on crop production and associated losses. economic input of diseases on crop production and associated losses. economic input of diseases on crop production and associated losses. extensively documented. The virus has been disseminated throughout the state, including some northern counties. The highest incidences of TYLCV have been correlated with high populatio of whiteflies. Extensive host range studies are being conducted with TYLCV in Florida, and TYLCV has been found to infect and cause diseas in petunia and common bean. Detection in petunia could have serious implications in terms of exporting this ornamental plant. 2. Efforts are being conducted to understand how TYLCV survives in the Dominican Republic durin the three-month whitefly host-free period. Using a				110	
relative contamination of whiteflies with TYLCV, was found that by the end of the tomato-growing season, TYLCV was readily detected in whiteflies collected from all tomato fields tested. However, within one month of the host-free period, the amount of virus detected in whiteflies collected from plants surrounding tomato fields decreased tremendously. By the end of the host free period, little or no virus could be detected in whiteflies. These results suggest that whiteflies themselves ar not likely to be the primary way in which the virus survives during the host-free period. Weeds and other plants in and around fields during the host-fre period were then collected and tested for TYLCV using PCR. A number of weeds were found to be symptomless carriers of TYLCV. These results suggest that such symptomless hosts may be the	of economic viruses, host plants, and reservoirs, and determination of	economic input of diseases on crop	X		extensively documented. The virus has been disseminated throughout the state, including some northern counties. The highest incidences of TYLCV have been correlated with high populations of whiteflies. Extensive host range studies are being conducted with TYLCV in Florida, and TYLCV has been found to infect and cause disease in petunia and common bean. Detection in petunia could have serious implications in terms of exporting this ornamental plant. 2. Efforts are being conducted to understand how TYLCV survives in the Dominican Republic during the three-month whitefly host-free period. Using a polymerase chain reaction test to determine the relative contamination of whiteflies with TYLCV, it was found that by the end of the tomato-growing season, TYLCV was readily detected in whiteflies collected from all tomato fields tested. However, within one month of the host-free period, the amount of virus detected in whiteflies collected from plants surrounding tomato fields decreased tremendously. By the end of the host free period, little or no virus could be detected in whiteflies. These results suggest that whiteflies themselves are not likely to be the primary way in which the virus survives during the host-free period. Weeds and other plants in and around fields during the host-free period were then collected and tested for TYLCV using PCR. A number of weeds were found to be symptomless carriers of TYLCV. These results suggest that such symptomless hosts may be the primary way that the virus survives during the host-

# Table B. Viruses, Epidemiology and Virus Vector Interactions.

		Progress A		
Research Approaches	Year 2 Goals Statement	Yes	No	Significance
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission.	Determine specific cellular and molecular factors involved in virus transmission. Study role of endosymbionts in virus acquisition and transmission.		X	Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies and other potentially effective approaches. Continue studies of management approaches for disease abatement. Interdisciplinary studies in conjunction with whitefly control methods in Sections B and C.
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Continue studies of management approaches for disease abatement. interdisciplinary studies in conjunction with whitefly control methods in Sections B and C.	Х		In Costa Rica, experiments conducted using living covers (such as coriander and perennial peanuts) and silver plastic mulch demonstrated that these strategies reduced the incidence of geminivirus infection of tomato under moderate whitefly/ geminivirus pressure, but not under high pressure. Thus, living covers and/or silver plastic represent a promising management tool, but one that needs to be used in combination with other practices that lead to reduced inoculum pressure. In the Dominican Republic, the mandatory whitefly host- free period continues to provide an effective management tool for TYLCV. There is a lag period of approximately one-month after planting tomatoes before TYLCV appears and this lag period allows for early-planted tomatoes to provide good yields. This strategy, together with the use of insecticides and tolerant varieties for late season planting, have allowed for the almost complete recovery of the processing tomato industry in the Dominican Republic.

## Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued)

		Progress A	Achieved	
Research Approaches	Year 2 Goals Statement	Yes	No	Significance
Control of virus diseases: development of virus resistant germplasm through conventional and engineered/molecular approaches. Define prospective strategies for selecting candidate viruses, identifying specific virus diseases to target, and prioritize specific crops and cultivars for protection approaches.	Continue to define suitable strategies for determining target viruses. Isolate and characterize virus-resistant germplasm. Continue work toward engineered resistance in target crops and selected viruses.	Х		<ol> <li>Cotton varieties have been screened under field conditions in the Imperial Valley of California for resistance to cotton leaf crumple geminivirus (CLCrV). A number of lines looked promising, particularly C95-387, which showed no symptoms of infection and in which no virus was detected. Two other lines, C95483 &amp; C95383 also showed potential resistance to CLCrV.</li> <li>Efforts are underway to identify tomato germplasm that is resistant to TYLCV as well as to develop genetically engineered tomatoes with resistance to TYLCV.</li> </ol>
Pursue specific genetic and biological basis for variability in whitefly biotypes, strains, and species; determine impact of different genetypes/phenotypes on whitefly- mediated transmission and on the epidemiology of virus diseases.Continue to study differences in species/strains/biotypes with respect to transmission, host range, mating compatibilities, molecular variability. Determine molecular basis of observed variability in biological, molecular & genetic terms. Infer molecular phylogenies from molecular markers.			Х	

# Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued)

Progress AchievedResearch ApproachesYear 3 Goals StatementYesNo	Significance
Identification and characterization of new or emerging whitefly-transmitted induces and strains.       Continue etiological studies and virus characterization. Apply molecular diagnostics to virus identification and evaluation of disease incidence and virus distribution.       X	<ul> <li>1.A new cucurbit-infecting geminivirus, <i>Cucurbit leaf crumple virus</i> (CuLCrV) was identified in the Imperial Valley of California. This virus causes leaf crumpling and yellowing in watermelon, cantaloupe, and muskmelon, but no symptoms were observed in honeydew melons. Very high incidences of the virus were detected in fall melons in the Imperial Valley and the virus was detected in melons with leaf crumple symptoms from Blythe, CA and Yuma, AZ. Significant progress has been made in the characterization of this virus (e.g., much of the DNA sequence of the virus has been elucidated) and tools are in hand for the monitoring for the virus in the spring and fall melon crops in 2000.</li> <li>2.<i>Cucurbit yellow stunting disorder virus</i> (CYSDV), a closterovirus in the Genus <i>Crinivirus</i>, was identified for the first time in the United States in the Rio Grande Valley of Texas. CYSDV is transmitted by the silverleaf whitefly. Previously, CYSDV was only known to occur in Europe and the Middle East. Like most of the so-called yellowing viruses, this virus causes yellowing symptoms in older leaves of infected melons. These symptoms can resemble nutritional deficiencies. Thus, infections by these viruses can be hard to diagnose. Molecular tools are available for detection of CYSDV and other related closteroviruses, particularly a degenerate PCR primer pair designed based upon the heat shock protein-like gene that is found in all of these viruses. It will be important to carefully monitor the spread of this virus in Texas and to look for it in other melon growing areas.</li> </ul>

# Table B. Viruses, Epidemiology and Virus Vector Interactions.

Research Approaches Year 3 Goals Statement	Yes	Achieved No	Significance
Molecular epidemiology: identification of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses. Establish geographic distribution of viruses and identify sources of inoculum. Assess role of alternative host virus reservoirs on spread of diseases.	X		<ul> <li>1. The distribution of CuLCrV in the Imperial Valley of California in 1999 was extensively documented using PCR and degenerate primers for whitefly-transmitted geminiviruses and using squash blot hybridization with a general probe for these viruses. The results of these analyses revealed that the virus had spread extensively by late fall 1999 and that it was infecting muskmelons throughout the Imperial Valley. It will be important to use these tools to monitor for CuLCrV in melons in spring and fall 2000. Infectious DNA clones of CuLCrV are being generated and will be used to determine the host range of this virus.</li> <li>2. Molecular tools are now in place to study the distribution of whitefly-transmitted geminiviruses in Central America and South America. This information will be important in order to develop resistance strategies.</li> <li>3. Many criniviruses have the capacity to infect weed and other hosts. Thus, it will be important to carefully monitor areas in which CYSDV has become established to assess the potential for weed and alternate hosts to contribute to the epidemiology of this disease. Findings from this work will impact management strategies.</li> </ul>

# Table B. Viruses, Epidemiology and Virus Vector Interactions.

		Progress A	Achieved	
Research Approaches	Year 3 Goals Statement	Yes	No	Significance
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission.	Continue studies in progress to determine specific factors involved in virus transmission, and the role of endosymbionts in virus acquisition and transmission.	Х		Important advances have been made in understanding the interaction of <i>Tomato yellow leaf</i> <i>curl virus</i> and <i>B. tabaci</i> . There is now evidence for transovarial transmission (rates ranging from 0- 10%) and sexual transmission between male and female insects. How these finding impact the epidemiology of the virus and disease management remain to be determined. It was also reported that a chaperonin protein produced in whiteflies by endosymbionts called GroEL may be involved in the protection of the virus in the insect body during it's journey from the gut to the salivary glands. This may involve an interaction between the GroEL chaperonin protein and the viral capsid protein.
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Continue studies of management approaches for disease abatement. Focus on interdisciplinary studies in conjunction with whitefly control methods in Sections B and C.	Х		It was reported that living ground covers and silver plastic reflective mulches can reduce virus spread in Costa Rica and Florida, but only under conditions of low to moderate virus pressure. Of the living ground covers tested, perennial peanuts seemed to be the best. Overall, silver plastic reflective mulch was the best for slowing spread of virus.

# Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued)

Table B. Viruses, Epidemiology and Virus Vector Interactions. (Continued	able B.	Viruses,	Epidemiology	and Virus	Vector 2	Interactions.	(Continued
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				<u> </u>
			No	
Research Approaches ontrol of virus diseases: development f virus resistant germplasm through onventional and ngineered/molecular approaches. efine prospective strategies for electing candidate viruses, identifying becific virus di seases to target, and rioritize specific crops and cultivars or protection approaches.	Year 3 Goals Statement Further identification of resistant germplasm and develop new methods of incorporating resistance into crop plants. Evaluate resistance strategies with respect to broad spectrum or virus- specific protection.	Progress A Yes X	Achieved No	Significance 1.Tomato varieties resistant to TYLCV are now commercially available and some of these provide high levels of resistance. Research in Israel suggests that use of highly resistant varieties (e.g., TY172) will reduce the rate of virus transmission compared to susceptible cultivars. However, moderately resistant cultivars (e.g., Fiona) may enhance virus transmission and actually enhance epidemics of TYLCV because the provide better sources of inoculum over longer periods of time compared with susceptible cultivars, which becoms severely diseased or die thereby making them poor sources of inoculum. These findings may influence how management strategies are developed. 2.Cotton varieties continue to be screened under field conditions in the Imperial Valley of Californi for resistance <i>to Cotton leaf crumple virus</i> (CLCrVA A number of lines continue to look promising, particularly line C95-387, which showed no symptoms of infection and in which no virus was detected. Two other lines, C95 483 and C95 383 also continue to show potential resistance to CLCrV. It will be very important to screen these materials in India or Pakistan to see if they have a resistance to the devastating cotton leaf curl virus, another whitefly-transmitted geminivirus that infects cotton. 3.Progress has been made in the generation of transgenic crops that are resistant to geminivirus infection. The major strategy that has been pursued to date is that of pathogen-derived resistance in which a wild-type or mutated virus gene or sequenced is introduced into the crop plant of choice. The idea is that expression of the viral sequence/ protein will interfere with the normal lifi cycle of the virus and, thus, interfere with the viral infection process. A number of viral genes have been evaluated to date including: capsid protein,

Table B.	Viruses.	Epidemiology	and Virus	Vector	Interactions.	(Continued)

		Progress	Achieved	
Research Approaches	Year 3 Goals Statement	Yes	No	Significance
**Pursue specific genetic and	Continue with work from previous		Х	This research approach should be transferred to
biological basis for variability in	years. Study impact of biotypes,			section A
whitefly biotypes, strains, and species;	strains, and species differences in the			
determine impact of different	disease spread, crop damage, and			
genetypes/phenotypes on whitefly-	specific control measures to reduce			
mediated transmission and on the	whitefly vector populations. Linkages			
epidemiology of virus diseases.	with biological and chemical control			
	sections.			

\*\* Transfer to Table A 3/31/2000

		-	Achieved	
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance
Identification and characterization of new or emerging whitefly-transmitted viruses and strains.	Continue etiological studies and virus characterization efforts. Apply molecular diagnostics to virus identification and evaluation of disease incidence and virus distribution Summarize and review results. Determine areas of new research.	x		<ol> <li>Continued characterization of the new cucurbit- infecting geminivirus, <i>Cucurbit leaf crumple virus</i> (CuLCrV)/<i>Cucurbit leaf curl virus</i> (CuLCV), has occurred. Infectious clones of isolates from Arizon and California have been obtained and sequenced completely. Results support the concept that it is a new virus species that is closely related to <i>Squash</i> <i>leaf curl virus</i> (SLCV). 2. Two viruses responsible for causing golden mosaic symptoms in <i>Macroptilium lathryoides</i> in Florida have been characterized. One virus is <i>Macroptilium mosaic</i> <i>virus</i> (MaMV) and the other is <i>Macroptilium goldu</i> <i>mosaic virus</i> (MAYMV). Infectious clones have been generated and sequence and other analyses suggest that these are distinct begomoviruses. The are infectious in common bean plants, though their role in bean golden mosaic disease in Florida remains to be established. 3. A squash blot hybridization assay has been commercialized by AgDia Co. (Elkhart, IN) and is available as a general tool for detection of geminiviruses in plant4. A dot blot assay has been developed for detection of criniviruses in plants. <b>New areas of research:</b></li> <li>Continue characterization of new emerging whitefly-transmitted begomoviruses, criniviruses and potyviruses. 2. Continue to refine detection an identification methods, e.g., develop virus-specific detection tools. 3. Improve our understanding of virus distribution: regional and global</li> </ol>

# Table B. Viruses, Epidemiology and Virus Vector Interactions, 2001-2002.

## Table B. Viruses, Epidemiology and Virus Vector Interactions, 2001-2002 (Continued)

	Tus vector interactions, 2001-2002 (Conti	Progress .	Achieved	
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance
Molecular epidemiology: identification	Identify and characterize virus	Х		1. It was established that Tomato yellow leaf curl
of economic viruses, host plants, and	involvement in disease establishment			virus could be effectively managed in the
reservoirs, and determination of	and spread. Assess potential methods			Dominican Republic through the use of a whitefly
of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses.				
				In the case of Bean golden yellow mosaic virus, the main reservoir appears to be common bean. Thus, time of planting is very important; i.e., do not establish young plants near older infected plantings. 3. Good progress has been made on understanding the epidemiology of <i>Tomato infectious chlorosis virus</i> (TICV) in Southern California fresh market tomato fields. TICV is vectored by the greenhouse whitefly and is common in southern California coastal production areas and greenhouses. The virus occurs in a number of weed hosts as well as tomato, but tomato seems to be the key host in terms of viral epidemiology. Thus, efforts for disease management need to focus on effective whitefly management and regional planning in terms of establishing tomato plantings. <b>New areas of research</b> : 1. Identify CuLCV/CuLCrV reservoirs and distribution 2. Continue efforts to define reservoir hosts for criniviruses (e.g., TICV, ToCV and <i>Cucurbit yellow stunt disorder virus</i> )

		Progress A	Achieved		
Research Approaches	Year 4/5 Goals Statement	Yes	No	Significance	
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission.	Continue virus-vector interactions studies toward the development of approaches for disease control. Summarize findings and suggest new research needs; implementation of existing knowledge	X		<ol> <li>Progress has been made in understanding the nature of whitefly transmission of TYLCV and Tomato mottle virus in Florida. In contrast to reports on TYLCV in Israel, neither TYLCV in Florida nor ToMoV was found to be transovarially transmitted. This has important implications for disease epidemiology.</li> <li>New areas of research:         <ol> <li>More studies need to be conducted on transovarial transmission (or not) with different begomoviruses. 2. Need more information on</li> </ol> </li> </ol>	
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Evaluate strategies for crop management and impact on disease epidemiology.	Х		endosymbiont/whitefly interactions Success in managing whitefly-transmitted geminivirus diseases was reported for TYLCV in the Dominican Republic based upon implementation of a whitefly host free period. The	
	Evaluate approaches and identify areas of future research for disease control by management of cropping systems. Linkages with IPM approaches			success was based upon the implementation of a regional approach, in which the host free period was enforced in the major tomato growing regions of the island. In addition, efforts were made to plow under crop debris after harvest. The recovery of the tomato industry from devastating losses caused by TYLCV has resulted in the acceptance of this regional approach by growers and others in the Dominican Republic. Similarly, effective control of BGMV in Florida has been achieved by better planting strategies and the use of a BGMV-resistant variety. Management of TICV and <i>Tomato</i> <i>chlorosis virus</i> (ToCV) has been achieved through	

the use of crop rotation and other approaches that reduce populations of the greenhouse whitefly.

1. Continue to optimize and modify cropping systems for effective disease management.

New areas of research:

## Table B. Viruses, Epidemiology and Virus Vector Interactions, 2001-2002 (Continued)

Research Approaches	Year 4/5 Goals Statement	Progress Achieved Yes No		Significance	
Control of virus diseases: development of virus resistant germplasm through conventional and engineered/ molecular approaches. Define prospective strategies for selecting candidate	Continue development of resistant varieties. Evaluate resistance strategies with respect to broad spectrum or virus-specific protection. Define mechanisms of resistance.	X	110	Commercially available tomato varieties with TYLCV resistance are now being used in the Dominican Republic and allowing for relatively high yields even under high virus pressure. Together with the imple mentation of the whitefly host free period, the use of these varieties has helped allow the tomato industry in the Dominican	
viruses, identifying specific virus diseases to target, and prioritize specific crops and cultivars for protection approaches.	Evaluate resistant plants in greenhouse and field experimentation, and identify additional research. Molecular-based monitoring of transgenes in environment	X		Republic recover to per-TYLCV levels. Two cotton lines were identified continue to show very high levels of resistance to Cotton leaf crumple virus (CLCrV). It will be very important to screen these materials in India or Pakistan to see if they have any resistance to the devastating cotton leaf curl virus, another whitefly-transmitted geminivirus that infects cotton. Promising levels of resistance to TICV have been identified in wild tomato species. This could result in the introgression of this resistance into <i>L</i> <i>esculentum</i> germ plasm. An agroinoculation procedure has been developed that allows for the screening of cucurbit germ plasm for resistance to CLCuV/CuLCrV. Progess has been made in the development of bush beans with resistance to Bean golden mosaic virus. This has been accomplished through a traditional breeding approach. These varieties have been released for commercial use in Florida and are helping in the management of this disease. <b>New areas of</b> <b>research:</b> Continue to evaluate strategies for generating genetically modified crops with resistance to begomoviruses. <b>Technology transfer:</b> The squash blot hybridization method has now been made into a commercial kit for the detection of whitefly- transmitted geminiviruses. A crinivirus dot blot assay may also be commercialized in the near future. TYLCV-resistant tomato cultivars are now available and these allow for respectable yields in area with high incidences of TYLCV, and BGMV-resistant bean varieties also have been developed. There is a need to evaluate the CLCrV-resistant lines in Pakistan and India for resistance to <i>Cotton leaf curl</i> <i>virus</i> . An agroinoculation method has been developed for CLCuV/CuLCrV and it is being evaluated for screening cucurbits for resistance to this new begomovirus.	

# Table B. Viruses, Epidemiology and Virus Vector Interactions, 2001-2002 (Continued) Progress

#### Reports of Research Progress Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods Co-Chairs: Shirley Taylor and John Palumbo

Investigator's Name: S. J. Castle<sup>1</sup>, F. J. Byrne<sup>2</sup>, N. Prabhaker<sup>2</sup>, & N. C. Toscano<sup>2</sup>

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**Research & Implementation Area**: Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by the Report: 2000-2001

## Sources of Variation in Systemic Uptake Bioassays of Neonicotinoid Insecticides

Interpretation of insecticide bioassays is predicated on the assumption that all test subjects exposed to a dosage of insecticide actually received that dosage and not one greater or lesser. Mortality at a series of insecticide dosages can then be analyzed statistically and the pertinent statistics for different populations compared to assess relative differences in susceptibility to a particular insecticide. Most insecticides are effective as contact poisons that act either by direct exposure with a contact spray, or by indirect exposure to a residue deposited on a plant or other surface within the insect's environment. Accordingly, bioassays designed to test the susceptibility of a sample population to a contact insecticide have relied upon various devices to ensure equal exposure of test subjects either by topical application or by uniform deposition of a residue. Precise quantities of known concentrations of an insecticide could then be deposited upon a test insect or surface to assess the susceptibility of the sample subjects. Treatment replication helps to compensate for minor variability in test conditions and reduce experimental error.

Neonicotinoid insecticides are active both as contact and as ingested insecticides. However, bioassays performed worldwide on imidacloprid, the longest available and most widely used of the neonicotinoids, have relied principally upon systemic uptake of imidacloprid solutions of known concentrations by severed plants or leaves. Interpretation of bioassay results have been made on the basis of the concentration of the imidacloprid solutions only, without regard for how much solution is actually taken up by the sample leaf or small plant. In many laboratories, a disk is punched from the uptake leaf and placed on agar within a petri dish or vial, to which test subjects are added and subsequently evaluated for mortality. In addition to substantial variation in uptake both within and between treatment concentrations of imidacloprid by the severed leaves or plants, variability in the distribution of imidacloprid within leaves can also affect mortality assessment of the test insects. We have measured uptake by detached leaves over a series of concentrations of imidacloprid and thiamethoxam ranging between 1 and 1000 ppm and found that volume uptake by leaves progressively declines with increasing concentrations. This is especially true for imidacloprid, a less soluble compound than thiamethoxam, at concentrations above c. 300 ppm. Variation in volume uptake within a concentration among individual detached leaves can vary by as much as 4-fold. This degree of variability among leaves often is carried over into variable mortality among replicates within a treatment concentration. However, volume uptake by individual leaves tends to be a poor predictor of mortality, suggesting that within-leaf distribution of active ingredients is non-uniform.

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**Research & Implementation Area**: Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by Report: 2000-2001

# Effects of Applaud and Knack on Sweetpotato Whitefly (SPW) Nymph Mortality and Adult and Nymph Honeydew Production

Sweetpotato whiteflies, *Bemisia tabaci* (Gennadius) produce honeydew that results in cotton lint contamination causing reduced lint processing efficiency. The insect growth regulators, Applaud® and Knack®, provide effective control of SPW on cotton by interfering with their reproduction and development. We investigated their effect on SPW honeydew production. Amounts of the major sugar components of honeydew produced by adults and nymphs collected on day six following Applaud or Knack applications to cotton field plots were not significantly different compared to amounts produced by those collected from untreated plots. Similar results occurred with SPW adults confined for 48 h on Applaud or Knack < 1 to 48 h-old residues in the laboratory following cotton leaf dips. Also in the laboratory, mortality of adults was not affected by nebulizer applied contact spray applications and honeydew sugars were not significantly reduced. In contrast, mortality of first and second instar SPW nymphs on leaves was higher on day six following leaf dips in Applaud solutions compared with leaf dips in Knack or water solutions. Nymph mortality on day six following leaf-dips in Knack solutions was higher than mortality of nymphs following leaf-dips in water. Honeydew collected from nymphs during two to 50 h intervals after leaf dip treatment had reduced amounts of glucose, fructose and trehalulose, but not sucrose and melezitose per nymph compared with honeydew from nymphs on leaves dipped in water.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by the Report: 2000

## Effects of Defoliants Alone and in Combination with Insecticides on Silverleaf Whitefly

Silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring, is one of the most important pests of cotton, vegetables and other crops in the US. The spring-summer cotton and fall-winter-spring vegetable cropping system in south Texas is unique, this sequencing of crop production allows whiteflies to thrive year-round. Although defoliants are used in every season in south Texas, the role of these defoliants on *B. argentifolii* and their natural enemies are unknown. The objective of this study was to determine the effects of defoliants on survival, migration and dispersal of *B. argentifolii* and their parasitoids after cotton plants were defoliated. Def 6 (S,S,S-tributylphosphorotrithioate); Karate (lambda-cyhalothrin), and Guthion (azinphosmethyl), were used. Filed rates were: full rate of Def at (2 pts/ac), full rate of Dropp at (0.2 lb/ac), full rate of Karate at (0.3 lb [AI]/ac), half rate of Guthion (0.25 lb [AI]/ac), full rate of Def + full rate of Karate, full rate of Def + half rate of Guthion. Untreated plots were used as controls. Each plot was 6 rows of cotton and 45 m long. Each treatments had three replications. Cotton leaves, 4th or fifth leaf from each plant, were collected directly from the treated plants 1 day after the application of chemicals. Twenty living individuals of each developmental stage were marked with an ink pen. The immatures were checked daily for survival until they developed to the next stage or died. Yellow sticky cards were used to trap whiteflies and parasitoids in the field. Five cards were placed in the middle of the plot, about 8 m apart. The cards were collected and replaced with new ones at 1-3 d intervals after the chemical applications. All arthropods trapped on the cards were counted in the laboratory.

Def and in combination with insecticides did significantly affect the survival rates of young nymphs but not eggs, older nymphs and pupae. Significant lower survival rates (13.3-30.0%) were found in Def alone and in combination with insecticides, compared with 70% survival rate in untreated control. Generally, the cotton leaves treated with Def alone had higher survival rates in all immatures stages than those treated with combinations of Def+insecticides. Karate and Guthion alone did not have significant effects on *B. argentifolii*, although lower survival rates were found. However, we do not know if Guthion and Karate, in combination with Def, added any synergistic effects on B. argentifolii to Def. Although numbers of B. argentifolii adults caught on yellow sticky cards varied greatly from date to date among the treatments, there were no clear picture showing significant effects among the treatments. Generally, more whitefly adults were caught on yellow sticky cards in the plots treated with Karate and Guthion, and slightly fewer in the plots treated with Def and untreated control. It seems that fewer whitefly adults took off and fly high in the untreated plots because the plants were green, and abundant young and nutritious leaves were available. However, we do not know exactly why fewer whitefly adults were caught in the plots treated with Def or Def+insecticide. Numbers of parasitoids, Eretmocerus spp. and Encarsia spp., caught on the yellow sticky cards are shown in Figure 2. Generally, among the treatments, yellow sticky cards placed in Def-treated plots caught similar numbers of parasitoids to those in untreated plots, but had significantly fewer parasitoids than from other chemical-treated plots. Numbers of parasitoids caught on yellow sticky cards placed in the Karate-treated plots was the greatest, followed by those placed in Guthion-and Def+Karate-treated plots. Results from this study indicate that Karate had no significant effects on the parasitoids. Although the yellow sticky cards in Guthion-treated plots than those in Karate-treated plots caught fewer parasitoids, the effects might not be directly related to Karate and Guthion themselves. Whereas we do not know the reason why the yellow sticky cards caught similar numbers of parasitoids in untreated and Def-treated plots, possible explanations might be that in the Def-treated plots, application of Def might affect the emergence or migration of parasitoids, whereas in untreated plots, parasitoids might not likely fly high or disperse with the presence of hosts (live whitefly nymphs), and therefore, fewer were caught on the yellow sticky cards. Because Karate and Guthion had no effects on leaf defoliation, we do not know why more parasitoids were caught on the yellow sticky cards. Perhaps, the odors or other chemical cues stimulated the parasitoids to take off, fly high and be caught. Many species of insects and spiders were caught on yellow sticky cards.

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Dates Covered by the Report: 2000

## Interactions of Planting Dates & Applications of Imidacloprid for Management of Silverleaf Whitefly on Spring Melon in South Texas

The silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring, continues to threaten the melon production in south Texas. Melon was severely damaged by the whitefly if Admire or other measure is not applied. In south Texas, the silverleaf whitefly infests melons from late March, and the whitefly population gradually increase in April, and reach the peak in late April or May. Melons planted late usually are more vulnerable to whitefly attack and have more severe damage than the melons planted earlier. The objective of this study was to determine the relationships and interactions between planting dates and applications of Admire (imidacloprid) for management of the silverleaf whitefly on spring melons in south Texas. There were 16 treatments, and each will had 4 replications. Each plot was 30 ft long with two separate rows 80 in wide, and 10-20 plants each. The plots were arranged in a randomized complete block design. Melons were planted in four dates, 13 and 27 Jan., and 10 and 24 Fed., 2 weeks apart. In each planting date, there were four treatments, Admire were applied in full rate (16 oz/ac) at transplanting, full rate in the mid-season, half rate in the mid-season, and an untreated control. Whitefly adults were sampled when whitefly population increase significantly. Thereafter, plants were sampled in 7-day intervals. Adults was counted by leaf turn method. When plants have <6 leaves, adults on the oldest leaf were counted, and when plants have >6 leaves, adults on the 4<sup>th</sup> or 5<sup>th</sup> leaf from the proximal were counted. Red-eyed nymphs (pupae), empty pupal cases per 4 leaf-discs (1 cm diameter) per leaf were counted. Virus incidence, leaf area covered by sooty mold, yield and sugar contents were recorded.

Interactions of planting dates and insecticide application were significant, indicating that numbers of adults on the plants with different planting dates and insecticide application were significantly different. Results also indicated that the later the plants were planted, the more whitefly adults were found, with 5.7, 4.5, 3.1 and 2.7 adults per leaf on the plant that were planted on 24, 10 Feb., 27 and 13 Jan., respectively. On the plants planted on 13 Jan., fewest adults were found on the plants treated with Admire in full rate at transplanting, followed by the plants treated Admire in full rate in mid-season. On the plants planted on 27 Jan., numbers of whitefly adults on the plants treated with Admire whether applied at transplanting, mid-season, full rate or half rate, were significantly reduced before mid -April, and were not significantly different in the last 4-5 weeks. Numbers of adults on the plants planted on 10 and 24 Feb. were very low until mid -April., and increased rapidly in late April, and peaked in early May. Plants treated with Admire (all treatments) had fewer adults on the plants planted on 10 Feb. compared with untreated plants. However, numbers of adults on the plants planted on 24 Feb. were significantly different among the treatments, with significantly fewer adults on the plant treated with Admire applied in full rate in mid-season, followed by the plants treated Admire applied in full rate at transplanting. Few red-eyed nymphs were found on the plants that planted in all four different planting dates until late March. Numbers of red-eyed nymphs increased gradually until early April, and then increased rapidly from late April until the end of the season. Generally, numbers of whitefly nymphs on the plants planted on different dates were significantly different. The plants planted early on 10 Jan. had the fewest numbers of nymphs, followed by the plants planted on 27 Jan. and on 24 Feb., the plants planted on 10 Feb. had the most. Numbers of red-eyed nymphs on the plants were also significantly different among the plants applied Admire in different rates and on different dates. The plants treated with Admire in full rate at transplanting had the fewest nymphs, followed by the plants with Admire applied full rate in mid-season, and then the plants with Admire in half rate in mid-season. Untreated plants had the most number of nymphs. On the plants planted on 13 Jan., numbers of nymphs on the plants treated with Admire at full rates at transplanting or mid-season were significantly fewer than those on the plants treated with Admire applied in half rate and those on untreated plants. In contrast, numbers of nymphs on the plants planted on 27 Jan. were not significantly different among the three treatments with Admire applied in full rates at transplanting, in mid-season, and half rate in mid-season. On the plants planted on 10 Feb., numbers of nymphs on the plants treated Admire in full rates at transplanting or in mid-season were significantly fewer than those on other treatments, especially in the late season. Numbers of nymphs on the plants planted on 27 Feb. among the four treatments were similar to those on the plants planted in 13 Jan., with fewest nymphs on the plants treated Admire applied in full rate at transplanting, followed by Admire applied in full rate in mid-season. Number of whitefly nymphs on the plants treated with Admire applied in half rate in mid-season was not significantly different from those on untreated plants. The numbers of melons harvested from plots of different treatments differ significantly. Total numbers of melons harvested from each plots among the treatments of different planting dates and insecticide application were significantly different. Among the planting dates, more melons were harvested from the plants planted on 13 Jan. and on 24 Feb. Numbers of melons harvested from the plants among the treatments of Admire applied in different rates and on different date were not significantly different. However, the size and weights of the melons among the treatments were not significantly different.

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## Dates Covered By The Report: 2000

## Effects of Planting Dates and Applications of Imidacloprid For Management of Silverleaf Whitefly in Melons

The silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring, continues to threaten the melon production in south Texas. Melon was severely damaged by the whitefly if Admire or other measure is not applied. The objective of this study was to determine the relationships and interactions between planting dates and applications of Admire (imidacloprid) for management of the silverleaf whitefly on spring melons in south Texas. There were 16 treatments, and each will had 4 replications. Each plot was 30 ft long with two separate rows 80 in wide, and 10-20 plants each. The plots were arranged in a randomized complete block design. Melons were planted in four dates, 13 and 27 Jan., and 10 and 24 Fed., 2 weeks apart. In each planting date, there were four treatments, Admire were applied in full rate (16 oz/ac) at transplanting, full rate in the mid-season, half rate in the mid-season, and an untreated control. Whitefly adults were sampled when whitefly population increase significantly. Thereafter, plants were sampled in 7-day intervals. Adults were counted by leaf turn method. When plants have <6 leaves, adults on the oldest leaf were counted, and when plants have >6 leaves, adults on the 4<sup>th</sup> or 5<sup>th</sup> leaf from the proximal were counted. Red-eyed nymphs (pupae), empty pupal cases per 4 leaf-discs (1 cm diameter) per leaf were counted.

Interactions of planting dates and insecticide application were significant. Results also indicated that the later the plants were planted, the more whitefly adults were found, with 5.7, 4.5, 3.1 and 2.7 adults per leaf on the plant that were planted on 24, 10 Feb., 27 and 13 Jan., respectively. On the plants planted on 13 Jan., fewest adults were found on the plants treated with Admire in full rate at transplanting, followed by the plants treated Admire in full rate in mid-season. On the plants planted on 27 Jan., numbers of whitefly adults on the plants treated with Admire whether applied at transplanting, mid-season, full rate or half rate, were significantly reduced before mid -April, and were not significantly different in the last 4-5 weeks. Numbers of adults on the plants planted on 10 and 24 Feb. were very low until mid -April., and increased rapidly in late April, and peaked in early May. Plants treated with Admire (all treatments) had fewer adults on the plants planted on 10 Feb. compared with untreated plants. However, numbers of adults on the plants planted on 24 Feb. were significantly different among the treatments, with significantly fewer adults on the plant treated with Admire applied in full rate in mid-season, followed by the plants treated Admire applied in full rate at transplanting. Few red-eyed nymphs were found on the plants that planted in all four different planting dates until late March. Numbers of red-eyed nymphs increased gradually until early April, and then increased rapidly from late April until the end of the season. Generally, numbers of whitefly nymphs on the plants planted on different dates were significantly different. The plants planted early on 10 Jan. had the fewest numbers of nymphs, followed by the plants planted on 27 Jan. and on 24 Feb., the plants planted on 10 Feb. had the most. Numbers of red-eved nymphs on the plants were also significantly different among the plants applied Admire in different rates and on different dates. The plants treated with Admire in full rate at transplanting had the fewest nymphs, followed by the plants with Admire applied full rate in mid-season, and then the plants with Admire in half rate in mid-season. Untreated plants had the most number of nymphs. On the plants planted on 13 Jan., numbers of nymphs on the plants treated with Admire at full rates at transplanting or mid-season were significantly fewer than those on the plants treated with Admire applied in half rate and those on untreated plants. In contrast, numbers of nymphs on the plants planted on 27 Jan. were not significantly different among the three treatments with Admire applied in full rates at transplanting, in mid-season, and half rate in mid-season. On the plants planted on 10 Feb., numbers of nymphs on the plants treated Admire in full rates at transplanting or in mid-season were significantly fewer than those on other treatments, especially in the late season. Numbers of nymphs on the plants planted on 27 Feb. among the four treatments were similar to those on the plants planted in 13 Jan., with fewest nymphs on the plants treated Admire applied in full rate at transplanting, followed by Admire applied in full rate in mid-season. Number of whitefly nymphs on the plants treated with Admire applied in half rate in mid-season was not significantly different from those on untreated plants.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by the Report: 2001

## Effects of Defoliants Alone and in Combination with Insecticides on Silverleaf Whitefly and Parasitoids, 2001

Effects of Def and Dropp alone and in combination with two insecticides, Karate (a pyrethroid) and Guthion (an organophosphate) on silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring were determined in 2001. Two defoliants, Def 6 (S,S,S-tributylphosphorotrithioate, Bayer, Kansas City, MO); Dropp (50% thidiazuron, Aventis [AgrEvo], Wilmington, DE), and two insecticides, a pyrethroid, Karate 2.08CS (lambda-cyhalothrin, Zeneca, Wilmington, DE), and an organophosphate, Guthion 2L (azinphosmethyl, Bayer, Kansas City, MO), were used in this study. There were eight treatments with different combinations of defoliants and insecticides at different rates: 1. Def (2 pts/ac)+Dropp (0.21b/ac) + Guthion (0.25 lb/ac); 2. Def (1 pt/ac) + Dropp (0.1 lb/ac); 3. Dropp (0.2 lb/ac) + Guthion (0.5 lb/ac); 4. Def (2 pt/ac) + Guthion (0.25 lb/ac); 5. Def (2 pt/ac) + Karate (0.03 lb AI/ac); 6. Guthion (0.5 lb AI/ac); 7. Karate (0.03 lb AI/ac), and 8. untreated control. To test the effect of applied chemicals on adult emergence for *B. argentifolii*, the third, and fifth and seventh leaf from the terminal was collected after chemical applications. The leaves were placed in paper bags and held in the laboratory for 3-4 weeks. Numbers of whitefly and parasitoid adults emerged from each bag were examined.

Numbers of whiteflies were significantly reduced on the leaves sampled on 26 July treated with defoliants and their combination with Karate and Guthion except for the cotton leaves treated Karate and Def + Dropp at 0.5X rate on which number of whiteflies was nor significantly different from those on untreated leaves. Defoliants and their combinations with insecticides also significantly reduced the number of silverleaf whitefly parasitized. Again, untreated leaves had the most parasitized whiteflies, followed by Karate-treated leaves, and then other treated leaves. Cotton leaves that treated with Def + Dropp + Guthion al 0.5X rate and Def + Guthion at 1.0X rate had the least number of parasitized whiteflies. Few whiteflies and parasitoids were found on the cotton leaves sampled on 16 August when leaves were almost defoliated. Therefore, there were no significant differences in numbers of whiteflies and parasitoids on sampled leaves. Numbers of *B. argentifolii* adults emerged from treated leaves after treatment. Cotton leaves treated with defoliants alone or in combination with Karate and Guthion reduced number of whitefly adults emerged in the samples on 26 July, although numbers of adults emerged in the treatments of Guthion and Karate alone and Def + Guthion at 1.0X rate were not significantly different from that in untreated control. There were no significant differences for both whitefly adult and parasitoids emerged for the samples in 16 August.

In conclusion, defoliants, Def and Dropp, and their combinations with Guthion and Karate significantly affected the infection and survival of both silverleaf whiteflies and their parasitoids, *Encarsia* spp. and *Eretmocerus* spp, although the effects varied greatly among the treatments. Karate had no significant effects on silverleaf whitefly and its parasitoids. Combining defoliants and insecticides increased the effectiveness on whiteflies and parasitoids on cotton.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by the Report: 2000-2001

## Efficacy of Sucrose Octanoate to Whitefly Using a Plant-based Bioassay

The objectives of this study were to determine the insecticidal activity of a synthetic analogue of natural sugar esters (sucrose octanoate (SO)) found in leaf trichomes of wild tobacco to egg, nymph and adult sweetpotato whitefly, *Bemisia tabaci* biotype B, (=silverleaf whitefly, *Bemisia argentifolii*), vector of Begomoviruses.

A plant-based petri dish bioassay system was developed to hold single tomato leaves infested with whitefly for their entire life cycle (egg to adult). A hole was cut in the lid of a Polystyrene petri dish (100x20 mm) to facilitate ventilation and ultrafine screen mesh was used to cover the hole to prevent whitefly escapes. Polystyrene conical tubes (15 ml, 17x120 mm) were filled with a plant tissue culture water agar and fertilizer (9-45-15) mixture. A hole was cut at the base of the petri dish to hold the conical tubes so that it fit tightly. Tomato leaves were cut so that the terminal leaf of the leaflet would fit in the petri dish. The remaining leaves of the leaflet were cut off and the stem was placed as far into the agar as possible with the abaxial leaf surface facing the screen to facilitate counting. A small hole was drilled into top of the conical tube above the agar line so that  $H_20$  could be added as needed (leaves will sprout roots and last > 28 d).

The SO solutions were applied using an ultra-low volume spray device that consisted of a spray platform that holds a pressurizable spray bottle at the proper distance and angle. Measured amounts ( $200 \mu$ I) of SO solution were placed in a test tube ( $12 \times 75 \text{ mm}$ ). The spray bottle siphon tube was placed in the test tube so that the test tube fit into the nozzle -pump body. Parafilm was wrapped around the top of the test tube to insure a tight fit and the spray bottle was pressurized to ~10 psi w/ 20 strokes of the pump mechanism to deliver a fine spray for each application. Both the abaxial and adaxial sides of the leaf were sprayed with 200 µl of SO solution.

Whiteflies were obtained from a laboratory colony maintained at USHRL, Ft. Pierce, FL. Sucrose octanoate solutions were prepared in concentrations ranging from 125 to 48,000 ppm in ddH<sub>2</sub>0 plus a ddH<sub>2</sub>0 control. Rates varied depending on the lifestage evaluated and were applied to eggs, nymphs or adults using an ultra-low volume spray device. Freshly laid whitefly eggs (24-48 hrs old) were sprayed and evaluated 7, 14, 21 and 28 DAT (until adult emergence). Whitefly nymphs were sprayed at the crawler/2nd instar stage and evaluated at 1, 3, 7 and 14 DAT (until adult emergence). Whitefly adults were sprayed and evaluated 3, 6, and 24 hours after treatment (HAT). Each concentration was replicated 5 times.

Whitefly egg and nymph mortality increased over time whereas whitefly adult mortality virtually remained the same. Freshly laid eggs sprayed with the higher concentrations (12,000 - 48,000) had significantly fewer adults emerge. Lower concentrations applied to freshly laid eggs had little effect on adult emergence. Toxicity of SO to whitefly nymphs ranged from  $LC^{90}$  values of 55,827 (20,162 - 774,817) ppm to 2,225 (1,517 - 11,255) ppm at 1 and 14 DAT (=adults emerged), respectively. Adult whiteflies were killed immediately if they were going to die and  $LC^{90}$  values calculated 1 DAT were 5,174 (4,345-6,602) ppm. Preliminary results indicate SO could be an effective tool for nymph and adult whitefly control to levels of >90% at higher concentrations of SO. Good coverage is key to efficacy.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

## Dates Covered by Report: September 2000 through December 2000

## Neonicotinoid Insecticide Efficacy for Silverleaf Whitefly Control In Cabbage, 2000

Cabbage var. Charmant was sown at UC Desert Research & Extension Center 4 October 2000. Six insecticide treatments and an non-treated control were replicated five times in a randomized complete design experiment. Insecticide treatments were as follows: Admire 2F at 0.25 lb ai/acre and Platinum 2SC at 0.141 lb ai/acre injected 3 inches below the seed-line on 15 September, and Assail 70 WP at 0.0375 lb ai/acre, Assail 70 WP at 0.05 lb ai/acre, Assail 70 WP at 0.075 lb ai/acre, and Actara 25 WG at 0.047 lb ai /acre were applied as foliar spray treatments on 24 October, 8 and 16 November. Silverleaf whitefly, *Bemisia argentifolii*, were sampled by counting adults via leaf turn of basal leaves on ten plants at random from each plot and nymphs were counted on 1.65 cm<sup>2</sup> of leaf surface from basal leaves of ten plants at random from each plot on 16, 23 & 30 October, 6, 13, 20 & 27 November, 4, 11, & 18 December.

The seasonal silverleaf whitefly adult per leaf mean for the non-treated control (4.2) was greater than the seasonal means for Admire 2F (3.0), Assail 70 WP at 0.05 lb ai/acre (3.3) and Assail 70 WP at 0.075 lb ai/acre (3.5), but was the non-treated seasonal adult mean was not greater than seasonal means for Platinum 2SC (3.8), Actara 25 WG (3.7), nor Assail 70WP at 0.0375 lb ai/acre (4.1); P#0.05. The seasonal silverleaf whitefly nymphs per cm<sup>2</sup> of leaf mean for the non-treated control (11.1) was greater than the seasonal mean for Admire 2F (4.7) but was not greater than any of the other treatments.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

Dates Covered by Report: March 2000 through July 2000

## Insecticides Efficacy against Silverleaf Whitefly In Spring Planted Cantaloupe Melons, 2000.

A stand of Cantaloupe melons, var. Topmark, was established at UC Desert Research & Extension Center 23 March 2000. Nine insecticide treatments and a non-treated control were replicated four times in a randomized complete design experiment. Insecticide treatments were as follows: Platinum 2 SC applied via drip irrigation at rates of 0.14 and 0.17 lb ai/acre and Admire 2 F was applied through the drip irrigation at 0.25 lb ai/acre, but was not followed by foliar sprays, Admire 2 F was applied through the drip irrigation at 0.25 lb ai/acre followed by foliar sprays treatments of LQ 215 (insecticidal soap) at 0.4% solution, foliar sprays of LQ 215 at 0.4%, foliar sprays of Capture 2 EC at 0.1 lb ai/acre, foliar sprays of Capture 2 EC at 0.1 lb ai/acre plus LQ 215 at 0.4% 3 EC, foliar sprays of Capture 2 EC at 0.1 lb ai/acre alternating with LQ 215 at 0.4% 3 EC, and foliar sprays of Actara 25 WG at 0.063 lb ai/acre. Drip irrigation insecticides treatments were applied 29 March. Foliar spray insecticide treatments were applied 19 & 26 April, 3 & 10 May. Silverleaf whitefly, *Bemisia argentifolii*, were sampled by counting adults on the fourth leaf from the terminal of the main stem cane from ten plants at random in each plot via the leaf turn method and whitefly nymphs were counted on 1.65 cm<sup>2</sup> leaf disks from ten crown leaves extracted from randomly selected melon plants in each plot. Adult silverleaf whitefly and nymphs were sampled on the following dates:11, 17 & 24 April, 1, 8, 15, 22 & 31 May, 2000.

Adult whitefly population levels were suppressed by both rates of Platinum 2 SC and Admire 2F for 5 weeks following drip irrigation application. Adult whitefly population levels were suppressed by all foliar insecticide spray applications except LQ 215 used alone. Silverleaf whitefly nymphal population levels were suppressed by Platinum 2 SC treatments and by Admire 2 F for 9 weeks following drip irrigation application. Silverleaf whitefly nymphal population levels were suppressed by all foliar treatments except LQ 215 used alone and LQ 215 alternating with Capture 2 EC.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Dates Covered By Report: March 2000 through December 2000

## Insecticides Efficacy Against Silverleaf Whitefly In Cotton, 2000

A stand of cotton, var. DPL 5415, was established at UC Desert Research & Extension Center 27 March 2000. Seven insecticide treatments and a non-treated control were replicated four times in a randomized complete design. Insecticide treatments were as follows: Assail 70 WP (acetamiprid) at 0.075 and 0.1 lb ai/acre, Actara 25 WP (thiamethoxam) at 0.05 and 0.06 lb ai/acre, Provado 1.6 F (imidacloprid) at 0.05 lb ai/acre, Danitol 2.4EC (fenpropathrin) + Orthene 97S (acephate) at 0.2 and 0.5 lb ai/acre, and Thiodan 3 EC (endosulfan) at 1.0 lb ai/acre. Silverleaf whitefly adults were sampled from ten plants at random in each plot via the leaf turn method using the fifth main stem leaf from the terminal. Silverleaf whitefly nymphs were counted on 1.65 cm<sup>2</sup> leaf disks from 5th position, main-stem terminal leaves extracted from ten randomly selected plants in each plot. Whitefly adults and nymphs were sampled on 6, 13, 20 & 27 June, 4, 11, 18 & 25 July, 1, 8 & 15. Seed cotton was hand picked from 0.002 acre per plot and seed cotton yield data were recorded. Seed cotton samples were ginned at the USDA-ARS Western Cotton Research Laboratory in Phoenix, AZ and lint samples were sent to the USDA/ARS Cotton Quality Research Station in Clemson, SC for stickiness and sugar analysis.

There were no differences among the treatments for silverleaf whitefly means on any of the sampling dates except 25 July when Thiodan 3 EC at 1.0 lb ai/acre, Assail 70 WP at 0.1 lb ai/acre and Danitol 2.4 EC + Orthene 97S had adult means lower than the non-treated control, P# 0.05. There were no differences among the treatments for silverleaf whitefly seasonal means. Assail 70 WP at 0.1 lb ai/acre gave the best control of silverleaf whitefly nymphs through the season followed by Assail 70 WP at 0.075 and Danitol 2.4 EC + Orthene 97S and all three of these treatments had silverleaf whitefly seasonal means lower than the non-treated control. The seasonal means of silverleaf whitefly nymphs for both rates of Actara 25 WP, for Provado 1.6 F and for Thiodan 3 EC were not different from the non-treated control.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

Dates Covered By Report: March 1997 through December 2000

## Comparison of Neonicotinoid Insecticides with Silverleaf Whitefly Insecticide Standards for Cotton

Silverleaf whitefly insecticide efficacy research trials were conducted during the cotton seasons of 1997 through 2000 at the University of California Desert Research and Extension Center in the Imperial Valley, CA to evaluate neonicotinoid insecticides and standard insecticides for control of silverleaf whitefly in cotton. Neonicotinoid insecticidal compounds, acetamiprid (Assail®), imidacloprid (Provado®), and thiamethoxam (Actara®) were compared to the standard whitefly insecticide fenpropathrin (Danitol®) in a tank mixture with an organophosphate, acephate (Orthene®) and compared to a cyclodiene compound endosulfan (Thiodan® or Phaser®) for control efficacy of whitefly adults, eggs and nymphs. Cotton stands, var. DPL 5415, were established at UC Desert Research and Extension Center in March for each year of study for the establishment of silverleaf whitefly insecticide efficacy trials. Each year the insecticides treatments and non-treated controls were replicated four times in randomized complete design experiments. Plots measured 15 m long and 8 m wide.

During four years of study, lint yields were not often different between the pyrethroid standard treatment (Danitol® 2.4 EC + Orthene® 90S) and the neonicotinoid treatments (Assail®, Actara® and Provado®). Treatments resulting in lower numbers of whitefly adults, eggs, and nymphs generally produced higher values of seed cotton pounds per acre and lint pounds per acre. In these experiments other factors that could influence yield included relative susceptibility of western flower thrips, cotton leafperforator and *Empoasca* sp. leafhoppers to the various insecticides. Insecticides in these studies vary in their spectra of activity. Danitol®, Orthene® 90S and endosulfan are active against a broad range of cotton insect pests and Assail®, Actara®, and Provado® have narrower ranges of activity.

The neonicotinoid insecticides provide silverleaf whitefly control in cotton at levels similar to the pyrethroid plus organophosphate standard Danitol® + Orthene®. The 0.06 lb ai/acre rate of Actara® and the 0.01 lb ai/acre and 0.075 lb ai/acre rates of Assail® 70 WP, and Danitol® + Orthene® maintain similar levels of silverleaf whitefly adult, egg, and nymph throughout the cotton season. The neonicotinoid insecticide, Assail®, and Danitol® plus Orthene® treatments provided the highest levels of control for silverleaf whitefly.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

Dates Covered by Report: March 2001 through July 2001

#### Insecticides Efficacy against Silverleaf Whitefly in Cantaloupe Melons, 2001

A stand of Cantaloupe melons, var. Topmark, was established at UC Desert Research & Extension Center 29 March 2001. Seven insecticide treatments and an untreated control were replicated four times in a randomized complete design experiment. The following insecticide treatments were applied as foliar sprays: Calypso 4 SC (clothianidin) at 0.094 lb ai/acre, Assail 70 WP (acetamiprid) at 0.05 lb ai/acre, Actura 25 WG (thiamethoxam) at 0.047 lb ai/acre and at 0.63 lb ai/acre, Danitol 2.4EC (fenpropathrin) at 0.2 lb ai/acre plus Thiodan 3 EC (endosulfan) at 0.5 lb ai/acre, Capture 2 EC (bifenthrin) at 0.08 lb ai/acre plus Thiodan 3 EC at 0.5 lb ai/acre, and Knack 0.86 EC (pyriproxyfen) at 0.054 lb ai/acre. Foliar insecticide spray treatments were applied 18 May, 2001. Silverleaf whitefly, *Bemisia argentifolii*, were sampled by counting adults on the fourth leaf from the terminal of the main stem cane from ten plants at random in each plot via the leaf turn method and whitefly nymphs were counted on 1.65 cm<sup>2</sup> leaf disks from ten crown leaves extracted from randomly selected melon plants in each plot. Adult silverleaf whitefly and nymphs were sampled on the following dates: 14, 21, 24, 29, 31 May, and 4 June 2001.

The adult whitefly mean for the untreated control was significantly greater than the means for all other treatment on 24 May, but the adult whitefly mean for the untreated control was not different from other treatments on all other sampling dates;  $P \le 0.05$ . The whitefly nymph means for all insecticide treatments were significantly lower than the untreated control on 21, 24 and 29 May. Seasonal means for whitefly nymphs were significantly lower for all insecticide treatments compared to the untreated control.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Dates Covered By Report: September 2001 through January 2002

#### Insecticide Efficacy for Silverleaf Whitefly Control In Cauliflower, 2001

Cauliflower var. Minuteman was transplanted into plots at UC Desert Research & Extension Center 21September 2001. Five insecticide treatments and an untreated control were replicated five times in a randomized complete design experiment. The insecticide treatments Actara 25 WG (thiamethoxa m) at 0.05 lb and 0.06 lb ai/acre, Fulfill 50 WG (pymetrozine) at 0.086 lb ai/acre, Thiodan 3 EC (endosulfan)at 0.75 lb ai/acre, and Provado 1.6 F (imidacloprid) at 0.05 lb ai/acre were applied as foliar sprays on 16 and 26 October and on 9 November 2001. Silverleaf whitefly, *Bemisia argentifolii*, were sampled by counting adults via leaf turn of basal leaves on ten plants at random from each plot on 2, 9, 16, 23, 30 October, 6, 14, 20 November 2001. Yield data as numbers of marketable cauliflower heads per 0.001 acres and weight as pounds of marketable cauliflower heads per 0.001 acres were collected 15 January 2002.

The post treatment mean for silverleaf whitefly adults for the untreated control (52.5) was significantly greater (P 0.05) than the seasonal means for all insecticide treatments except the mean for Fulfill (48.7). The post treatment adult whitefly mean for Provado 1.6 F (27.1) was significantly greater than the means for Actara 25 WG at 0.05 lb ai/acre (21.9) and 0.06 lb ai/acre (21.5). The post treatment mean for silverleaf whitefly nymphs per cm<sup>2</sup> of leaf for the untreated control (24.9) was significantly greater than the seasonal means for all insecticide treatments except the mean for Fulfill (20.7). The post treatment mean for silverleaf whitefly nymphs per cm<sup>2</sup> of leaf for Fulfill was significantly greater than all other insecticide treatments with post treatment means for whitefly nymphs ranging from 15.8 for Actara 25 WG at 0.05 lb ai/acre to 12.8 for Thiodan 3 EC at 0.75 lb ai/acre. There were no significant differences among treatment means for numbers of marketable cauliflower heads per 0.001 acre (10.5) nor for pounds marketable cauliflower heads per 0.001 acre. Investigator's Name: Eric T. Natwick

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Dates Covered By Report: September 2001 through January 2002

#### Neonicotinoid Insecticide Efficacy for Silverleaf Whitefly Control In Cauliflower, 2001

Cauliflower var. Minuteman was transplanted into plots at UC Desert Research & Extension Center 21September 2001. Seven insecticide treatments and an untreated control were replicated four times in a randomized complete design experiment. The insecticide treatments Admire 2F (imidacloprid) at 0.156 lb and 0.25 lb ai/acre, and Platinum 2SC (thiamethoxam) at 0.071 lb, 0.11 lb, 0.125 lb, 0.141 lb, and 0.172 lb ai/acre were injected 3 inches below the seed-line on 20 September 2001. Silverleaf whitefly, *Bemisia argentifolii*, were sampled by counting adults via leaf turn of basal leaves on ten plants at random from each plot and nymphs were counted on 1.65 cm<sup>2</sup> of leaf surface from basal leaves of ten plants at random from each plot on 2, 9, 16, 23, 30 October, 6, 14, 20 November 2001. The plant height (cm) was measured 13 December and yield data as numbers of marketable cauliflower heads per 0.001 acres and weight (lb) of marketable cauliflower heads per 0.001 acres were collected 15 January 2002.

The seasonal silverleaf whitefly adult mean for the untreated control (48.9) was significantly greater (P 0.05) than the seasonal means for all insecticide treatments. Admire 2F at 0.25 lb ai/acre had a seasonal mean for adult whitefly (12.2) that was significantly lower than all other insecticide treatments except Admire 2F at 0.156 lb (14.5) ai/acre and Platinum 2 SC at 0.172 lb ai/acre (13.5). The seasonal mean for silverleaf whitefly nymphs per cm<sup>2</sup> of leaf for the untreated control (11.1) was significantly greater (P 0.05) than the seasonal means for all insecticidetreatments. Admire 2F at 0.25 lb ai/acre had a seasonal mean for whitefly nymphs (1.9) that was significantly lower than all other insecticide treatments except Admire 2F at 0.156 lb (3.0) ai/acre and Platinum 2 SC at 0.172 lb ai/acre (2.0).

The untreated control plants mean height in cm (66.7) was significantly lower than the means for all insecticide treatments. The mean plant height (72.2 cm) for Platinum 2 SC at 0.071 lb ai/acre was significantly lower than the means for all other insecticide treatments, except Platinum 2 SC at 0.125 lb ai/acre with a plant height mean of 74.5 cm. The plant mean heights for all other insecticide treatment did not differ significantly and ranged from 76.6 cm for Platinum at 0.11 lb ai/acre to 78.2 cm for Admire 2F at 0.25 lb ai/acre.

The mean number of marketable cauliflower heads per 0.001 acre (10.5) was significantly lower than the mean numbers of marketable heads for Platinum 2 SC at 0.141 lb ai/acre (18.0) and for Platinum 2 SC at 0.172 lb ai/acre (19.0). The mean pounds marketable cauliflower heads per 0.001 acre (22.0) was significantly lower than the mean pounds of marketable heads for all insecticide treatments except Platinum 2 SC at 0.071 lb ai/acre (28.6) and for Platinum 2 SC at 0.125 lb ai/acre (25.8). The means for marketable cauliflower heads for all other insecticide treatment did not differ significantly and ranged from a mean of 36.6 lb per 0.001 acres for Platinum 2 SC at 0.11 lb ai/acre to 45.6 lb per 0.001 acres for Platinum 2 SC at 0.141 lb ai/acre.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

Dates Covered by Report: March 2001 through December 2001

#### Insecticides Efficacy Against Silverleaf Whitefly In Cotton, 2001

A stand of cotton, var. DPL 5415, was established at UC Desert Research & Extension Center 21 March 2001. Seven insecticide treatments and an untreated control were replicated four times in a randomized complete design. The following insecticide treatments were applied as foliar sprays: Assail 70 WP (acetamiprid) at 0.075 and 0.1 lb ai/acre, Danitol 2.4EC (fenpropathrin) at 0.2 lb ai/acre plus Orthene 97 (acephate) at 0.5 lb ai/acre, Danitol 2.4EC at 0.2 lb ai/acre plus Thiodan 3 EC (endosulfan) at 0.5 lb ai/acre, Knack 0.86 EC (pyriproxyfen) at 0.054 lb ai/acre, and Novaluron 0.83 EC (benzoylurea) at 0.013 lb ai/acre and at 0.026 lb ai/acre. Foliar insecticide spray treatments were applied 11 July, 7, 21, 30 August 2001. Silverleaf whitefly adults were sampled from ten plants at random in each plot via the leaf turn method using the fifth main stem leaf from the terminal. Silverleaf whitefly nymphs were counted on 1.65 cm<sup>2</sup> leaf disks from fifth position, main-stem terminal leaves extracted from ten randomly selected plants in each plot. Whitefly adults and nymphs were sampled on 10, 16, 20, 23, 26, 31 July, 6, 13, 20, 27 August, 4, 10, 17 September 2001. Seed cotton was hand picked from 0.002 acre per plot and seed cotton yield data were recorded. Seed cotton samples were ginned at the USDA-ARS Western Cotton Research Laboratory in Phoenix, AZ and lint samples were sent to the USDA/ARS Cotton Quality Research Station in Clemson, SC for stickiness and sugar analysis.

Seasonal means for whitefly adults for all of the insecticide treatments were significantly lower than the untreated control except the Knack 0.86 EC treatment,  $P \le 0.05$ . The Assail 70 WP treatments and the Danitol 2.4 EC + Orthene 97S treatment had seasonal means for whitefly adults that were significantly lower than the means for the Novaluron insecticide treatments. Seasonal means for whitefly nymphs for all of the insecticide treatments were significantly lower than the untreated control treatment. The Assail 70 WP treatments had seasonal means for whitefly nymphs for all of the insecticide treatments were significantly lower than the untreated control treatment. The Assail 70 WP treatments had seasonal means for whitefly nymphs that were significantly lower than the means for all other insecticide treatments. The seasonal mean for whitefly nymphs was significantly lower for the Knack 0.86 EC treatment compared to the means for all other insecticide treatments except the Assail 70 WP treatments. The Danitol 2.4EC plus Orthene 97 treatment had a seasonal mean for whitefly nymphs that was significantly lower than the means for Danitol 2.4EC plus Thiodan 3 EC and the Novaluron 0.83 treatments. There were no differences among the treatments for pounds of seed cotton per acre.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Date Covered by the Report: 2000

# Effectiveness of Neemix in Managing Silverleaf Whitefly (SLW) and Tomato Yellow Leaf Curl Virus in Tomatoes, 2000

Tomato seedlings were planted in a Rockdale fine gravely loam with pH 7.0 – 7.3 on 12 May 2000 at Homestead, Florida. Seedlings were placed 18 in. apart within rows and 6 ft. between rows and were drip irrigated. Plots were arranged in a randomized complete block design with four replications. Treatments evaluated were: 1) Admire at 8 ozs. as soil drench at planting followed by weekly foliar application of Neemix at 4 ozs./A; in this treatment plants were treated with one application of Neemix at 4 ozs./A; one week before planting the plants. 2) Admire at 8 ozs. as soil drench at planting followed by two weekly foliar application of Neemix at 4 ozs./A; 3) Admire at 8 ozs. as soil drench at planting followed by two weekly foliar application of Neemix at 4 ozs./A; 3) Admire at 8 ozs. as soil drench at planting followed by two weekly foliar application of Neemix at 8 ozs./A and then followed by weekly foliar application of Neemix at 4 ozs./A; 4) Admire 8 ozs./A as a soil drench at planting followed by Knack at 8 ozs. in rotation with Applaud at 8 ozs.; 5) Weekly foliar application of Knack at 8 ozs. followed by Applaud at 8 ozs.; 6) Admire at 16 ozs. as soil drench at planting; 7) Admire at 8 ozs. as soil drench at planting; 8) Neemix at 8 ozs./A as weekly foliar application; 9) A nontreated check.

Application of foliar treatments were made by using a backpack sprayer with two nozzles/row at 30 psi delivering 70 gpa. Weekly application of treatments were made on four dates-21, 28, May 2000, 4 and 12 June, 2000. Application of insecticides was terminated when about 80% of the plants in all treatment plots showed TYLCV symptoms. Evaluation of treatments was made 24 h after each application by counting SLW adults on one leaf/plant of randomly selected five plants per treatment plot. Finally, treatments were evaluated by harvesting all marketable fruits from all plants in a 25 ft. long middle row of each treatment plot.

Silverleaf whitefly adults were significantly fewer in all plants treated with various insecticides when compared with the nontreated control. This reduction in the numbers of SLW was consistent in all sampling dates during the present study. However Treatment 3, where Admire at the rate of 8 ozs. was drenched at planting followed by two weekly applications of Neemix at the rate of 8 ozs./A and then followed by weekly application of Neemix at 4 ozs./A, provided highest reduction of SLW. This reduction in the number of SLW was significantly different from Admire at the rate of 8 ozs./A. Marketable fruits were significantly greater in plants treated with the Admire -Knack-Applaud combination which did not differ from Admire alone and Admire -Neemix treated plants. In Admire -Neemix treated plants where Neemix at 8 ozs./A was applied two times followed by Neemix at the rate of 4 ozs./A, provided the second highest marketable yield next to Admire-Knack-Applaud treatment.

This study demonstrates that Neemix in combination with Admire provides significant reduction of SLW and its transmitted Tomato Yellow Leaf Curl Virus.

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Date Covered by the Report: 2000

#### Control of the Silverleaf Whitefly (SLW) and its Associated Bean Golden Mosaic Virus in Beans, 2000

Snap bean was planted on 15 March, 2000 into a Rockdale fine gravely loam with pH 7.0-7.3 pH at the Tropical Research and Education Center research plot. Treatment plots consisted of three 30-ft. long rows which were separated by 30-in space. Eight treatments were arranged in a randomized complete block design with four replications. A 5-ft. planted area separated the replicates. Treatments evaluated were: 1) Actara at 9 & 11 ozs./A as soil drench at planting; 2) Admire was drenched at planting at the rate of 24 ozs./A followed by four weekly applications of Knack at 8 & 10 ozs./A; 3) ) Admire was drenched at planting at the rate of 24 ozs./A followed by weekly foliar application of Knack at 8 ozs. in rotation with Applaud at 8 ozs; 4) weekly foliar application of Provado at 3.75 ozs/A; 5) weekly foliar application of Rimon at 6 ozs./A, and 6) a nontreated control. Evaluations of insecticides against the silverleaf whitefly were made by randomly collecting 5 leaves, one leaf/plant, 24 h after each application. Numbers of silverleaf whitefly adults were recorded 24 h after each foliar application by counting all adults on a leaf/plant of five randomly selected plants. Effects of insecticide treatments on the management of BGMV were determined by counting all healthy plants in the center row. All plants from 10-feet area in the center of the middle row were collected and weighed to determine effect of insecticide treatments on plant quality. Finally, 10 plants were collected from the center row of each treatment plot and number of fruits and weight of those fruits were recorded.

Silverleaf whitefly adults were significantly fewer on plants when treated with Actara at 11 ozs./A than on plants treated with other insecticide treatments. Silverleaf whitefly eggs were significantly fewer in all treatments when compared with the nontreated control. Silverleaf whitefly nymphs were significantly fewer on plants treated with all insecticides except in Rimon - treated plants. Lowest number of nymphs was observed in plants treated with Actara at 11 ozs./A.

Percentage healthy plants, free from BGMV, were significantly higher when they had been treated with Actara at 11ozs./A. Provado and Rimon did not provide any control of Bean Golden Mosaic Virus on beans. Treatments did not show pronounced effects on weights and heights of the treated plants. Numbers of marketable fruits and fruit-weights were significantly higher when plants were treated with Actara or Admire followed by Knack and Applaud. Provado and Rimon did not increase the numb ers of fruits or fruit weight. Although numbers of fruits increased on plants treated with Admire at 24 ozs. and Knack at 8 ozs., fruit weights did not increase. This inconsistency in fruit weights was caused by variable drying of fruits in the field environment before harvest.

Overall, Actara and Admire applied as soil drenches provided significant reduction of Bean Golden Mosaic Virus incidence. However, Admire in soil does not provide significant control of silverleaf whitefly for more than 4 weeks. After this critical period, Admire application should be followed by Knack to suppress both silverleaf whitefly and Bean Golden Mosaic Virus Investigator's Name(s): D. R. Seal

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### Date Covered by the Report: 1998

#### Effectiveness of Mycotrol in Controlling Silverleaf Whitefly

<sup>N</sup>Pod Squad' beans were planted in a Rockdale fine gravely loam with pH 7.0-7.3. Plant spacing was 2.5 cm in the row and 0.9 m between rows. Atrazine was broadcast in the furrow at planting at the rate of 0.5 kg/ha to control weeds in the rows, and the field was cultivated biweekly to clean weeds between the rows. Fertilizer was applied in kg/ha at 366 N, 732 P, 732 K on two dates at planting and 2 weeks after planting. The plots were irrigated by overhead sprinkler system at weekly intervals at approximately 5 cm. depth. Plots were four rows each 30 ft long. Six treatments were arranged in a randomized complete blocks with 4 replications. A nontreated 3 m wide section separated the plots. Treatments evaluated were: 1) Fipronil at 0.05 and 0.075 lb. a.i./A; 2) Provado 1.6F at 0.05 lb. a.i./A; 2) Mycotrol at 2 qts./A; 3) Provado at 0.025 lb. a.i./A in combination with 2 qts./A of Mycotrol; and 4) a nontreated control. Treatments were applied weekly using a backpack sprayer with two nozzles per row. During the spray of insecticides, the backpack sprayer was maintained at 30 psi. delivering 70 to 100 gallons/A. Treatments were evaluated by recording Bemisia argentifolii adults on randomly selected 10 leaves, one leaf/plant, per treatment plot. This was conducted by gently turning the leaves. In addition, B. argentifolii eggs and nymphs were recorded on randomly selected ten leaves per plant, one leaf/plant. These leaves were incised and placed in a plastic bag to transport them to the laboratory. Leaves were checked using a binocular microscope (10X) to record number of eggs and nymphs.

All treatments significantly reduced B. argentifolii eggs, nymphs and adults when compared with the nontreated control. Fipronil and Provado significantly reduced B. argentifolii adults and eggs after the first application. This reduction in the numbers of B. argentifolii eggs and nymphs was consistent in the subsequent weeks of study. Pupal populations did not differ among treatments after the first application. Pupal population on Fipronil and Provado treated plants decreased significantly when compared with the nontreated control. Among treatments, Mycotrol treated plants had more whiteflies than the others; although significantly different from nontreated control. Addition of Mycotrol with Provado did not improve management of silverleaf whitefly on beans

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**Research & Implementation Area:** Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods

#### **Date Covered by the Report:** 1998

#### Effectiveness of Fipronil in Controlling Silverleaf Whitefly (SLW) on Beans

'Pod Squad' beans were planted on 2 January 1998. Crop was maintained following recommended cultural practices. Plots were 2 rows by 30 feet long. Treatment plots were arranged in a randomized complete block design with four replications. Treatments evaluated in this study were: 1) Admire 2F (32 ozs./A as soil drench); 2) Provado 1.6F (3.75 ozs./A as foliar); 3) three rates of Fipronil 70WP (0.044, 0.05 & 0.075 lb. [a.i.]/A; and 4) a non-treated control. Admire was drenched in an 1 in.-deep furrow 4 in. apart from the seed row 3 weeks after planting. Provado and Fipronil were applied on foliage on 1/12, 1/19, 1/29 and 2/9/98 depending on the abundance of Bemisia argentifolii on the treated plants. Foliar application of insecticides was made by using a backpack sprayer with two nozzles per row at 30 psi. delivering 70 gpa. Prespray samples were collected once on 8 January and postspray samples were collected on 14, 20, 22, 27, 30 January and 24 Feb. 1998. Treatments were evaluated by counting numbers of *Bemisia argentifolii* adults and nymphs on randomly 5 plants, one leaf/plant, from the two center rows of each treatment plot on each sampling date. Admire, Provado and Fipronil significantly reduced SLW adults and nymphs on beans when compared with the nontreated control plants. Mean numbers of SLW nymphs and adults did not differ among rates. Use of Admire, Provado and Fipronil in rotation in a management program may provide an excellent control of SLW.

#### Section C Research Summary - Chemical Control, Biopesticides, Resistance Management and Application Methods.

#### John Palumbo

#### **Current Research Activities**

Chemical control continues to be a very important part of silverleaf whitefly management. Area -wide suppression and management of whiteflies has occurred largely due to the successful deployment of various chemical control approaches. Development and registration of new insecticides chemistries, insect growth regulators and biopesticides continues to be important and will continue to provide producers with a number of pest management options. In addition, industry, government and university scientists continue to collaboratively develop and refine integrated and resistance management programs within a number of cropping systems. Over the past 10 years, these efforts have resulted in a reduction and harmonization of chemical use, and a return to economic crop production in the United States.

Progress was made on the evaluation of systemic neonicotinoid insecticides, insect growth regulators (IGRs) and biopesticides. A significant amount of work was reported on the development of use patterns for the neonicitinoid class of chemistry. Formulations of thiamethoxam and acetamiprid applied as sprays to foliage and injected into the soil appear promising and may provide alternatives to prophylactic uses of imidacloprid in vegetables and melons crops. Use of IGRs remains an important chemical control approach as illustrated by their continued success in desert cotton crops and melons, and the development of action thresholds in tomatoes. Over the past several years, the uses of systemic insecticides and insect growth regulators have lead to a lessening of crop losses due to whitefly while opening the door to a greater role for biological control. A considerable amount of work was conducted across disciplines to examine the impacts of several IGRs on whitefly parasitoids and predators which has lead to the understanding of "bioresiudal". Workers continued to evaluate several biopesticides, azidirachtin, entomopathogenic fungi, oils, and sucrose esters for whitefly control and compatibility with natural enemies in integrated management programs. .

Because growers rely heavily on insecticides for whitefly control, monitoring and management of insecticide resistance continues to be a primary focus. Studies focusing on our basic understanding of resistance have examined the genetics and biochemistry of resistance and cross-resistance. Baselines continue to be established for insect growth regulators and imidacloprid across many growing regions. Continued work on new chemistries is important in detecting shifts early in a products use. The establishment of resistant colonies to various classes of insecticides has increased providing resistant specimens for use in resistance studies. In particular, genetic and biochemical studies have been focused on the role of MFO-based metabolism and esterases in resistance to imidacloprid. The metabolic role of synergists DEF and piperonyl butoxide to enhance the toxicity of imidacloprid was also investigated. A significant effort has been made by several laboratories to examine the cross-resistance patterns between three neonicotinoids, acetamiprid, thiamethoxam and imidacloprid. Although no definitive cross-resistance patterns were observed in a number of whitefly populations, the development of cross-resistance with extensive use is a possibility because of similarity in structure. Consequently, in order to avoid high selection pressure on any one chemical from this group, there is an urgent need for integrating the neonicotinoids into a diversified management program.

The development of resistance management programs at the grower level has been a priority in the desert growing regions of the southwestern U.S. Significant progress was made in Arizona to implement a cross commodity IRM approach for the shared use of buprofezin and neonicotinoids among commodities such as cotton, melons and vegetables. It is too early to determine the success of the Arizona program, but workers continue to collect bioassay data on insecticide susceptibility and providing this information to producers on a regional basis. Within the next few years, scientists and extension workers are confident that based on the above science. integrated programs in numerous cropping systems will be in place that optimizes neonicotinoid, IGR and biopesticide efficacy while minimizing their impacts on biological control efforts.

Research Approaches <sup>a</sup>	Year 1 Goals Statement	Progress Achieved Yes No	Significance
Improve insecticide efficacy:			
Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Develop new chemistries and natural products. Develop improved techniques for evaluating efficacy of insecticides. Support registration of desirable new products by providing information to regulatory agencies.	Х	New studies reported in this area in 1997 = 39. New biopesticides like <i>Petunia</i> extract and <i>Melia</i> extract tested. New biorationals tested or reported on included benzyl phenal urea napthaphenol and antibiotics (to act against symbiotic bacteria).
Develop improved methods of application including formulation and delivery of materials to improve control.	Develop spray systems for better underleaf coverage. Evaluate rates, timing, placement in relation to efficacy. Consider formulation, UV protectants, and other means to improve efficacy. Develop improved methods to evaluate application efficacy. Field test under commercial conditions for technology transfer.	Х	New studies = 10. Thermal fogger evaluated for greenhouse use. However, a comparison of five- sprayers in the field trials showed no significant differences between hydraulic, air-assist and electrostatic technology.
Conserve insecticide efficacy:			
Relate action thresholds to insecticide usage patterns.	Refine action thresholds based on insecticide efficacy and input from other control strategies.	Х	New studies = 8. Cost-benefit study of IPM system in cotton. Life table approach to evaluate impact of mortality factors initiated. Training effort to extend threshold information to growers in Arizona.
Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.	Establish whitefly strains resistant and susceptible to various classes of insecticide. Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide.	Х	New studies = 4. Imidacloprid binding site elucidated. Studies completed on stability of resistance in <i>Bemisia</i> including agricultural and ecological factors.

# Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods.

		Progress	Achieved	
Research Approaches <sup>a</sup>	Year 1 Goals Statement	Yes	No	Significance
Improve insecticide efficacy:				
Improve techniques for monitoring resistance.	Establish baseline data on toxogenic responses of whitefly populations to new insecticides.	Х		New studies = 9. Bioassays developed for testing sensitivity to imidacloprid. Baseline data obtained on sensitivity to imidacloprid and IGRs pyriproxyfen and buprofazin.
Develop, evaluate and refine resistance management systems.	Evaluate the effects of mixtures and rotations of new and old chemistries to mitigate selection for resistance.	Х		New studies = 14. Area-wide plans for management of resistance refined in Arizona and California. Large-scale trials of resistance management strategies conducted.
Integrate chemical control with other tactics.	Evaluate selectivity of synthetic insecticides and natural products to key whitefly natural enemies.	Х		New studies = 10, including laboratory and field studies on compatibility with whitefly natural enemies. Also a study on effects of pyrethroids on anitbiotic factors bred into crops.

### Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods. (Continued)

<sup>a</sup> See Table A for complementary research on thresholds.
 <sup>a</sup> See Table B for complementary research on virus/vector interactions.
 <sup>a</sup> See Table D for complementary research on biological control.
 <sup>b</sup> See Table E and F for complementary research on systems management.

		Progress Achiev	/ed
Research Approaches <sup>a</sup>	Year 2 Goals Statement	Yes No	Significance
Improve insecticide efficacy:			
Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Determine new modes of action of effective materials. Elucidate biochemical pathways of synthesis and degradation of natural products.	Χ	<ul> <li>(1) Section 3 registration of IGRs. (2) Section 18's supported, acetamiprid summer '99. (3) progress evaluating soil applied modes of action, sugar esters and entomopathic fungi., integration of biorationals and conventional chemistries.</li> <li>Need to evaluate future impact of FQPA.</li> <li>References 40, 69, 71, 78, 81, 103, 104, 108, 145, 165, 166, 167, 168, 180, 186, 187, 188, 212, 220, 262, 263, 273, 274, 297</li> </ul>
Develop improved methods of application including formulation and delivery of materials to improve control.	Develop spray systems for better underleaf coverage. Evaluate rates, timing, placement in relation to efficacy. Consider formulation, UV protectants, and other means to improve efficacy. Develop improved methods to evaluate application efficacy. Field test under commercial conditions for technology transfer.	Х	Akey's work with high PSI systems, increasing stability for azadiractin and utilizing digital photographs to evaluate application efficacy. References 47, 131, 139
Conserve insecticide efficacy:			
Relate action thresholds to insecticide usage patterns.	Refine action thresholds based on insecticide efficacy and input from other control strategies.	Х	Mint sampling plan/thresholds, distribution patterns validated, References 35, 125, 212, 317
Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.	Establish whitefly strains resistant and susceptible to various classes of insecticide. Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide. Evaluate the role of refuge habitats (weeds, tolerant crops, urban areas) to assure input of susceptible genes in whitefly population.	Х	Resistant colonies exist to endosulfan, chlorpyrifos, imidacloprid, bifenthrin; genetic and biochemistry studies are concentrated on acetylcholinesterase (Byrne) and nicotinyls; cross resistance being studied between nicotinyls and neonicotinyls. Impact of ecological factors such as nutrition, host plant response, local cropping patterns are being studied. Role of alfalfa as a refuge has been evaluated. References 18, 22, 46,

# Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods.

		Progress Achieved	
Research Approaches <sup>a</sup>	Year 2 Goals Statement	Yes No	Significance
Improve insecticide efficacy:			
Improve techniques for monitoring resistance.	Establish baseline data on toxogenic responses of whitefly populations to new insecticides. Expand comparative studies of resistance levels in diverse agro-ecosystems. Evaluate relationship between monitoring results and field efficacy.	Х	Baselines have and are being established for IGRs and systemic insecticides. Work in Cal. & Arizona ongoing to evaluate regional resistance management techniques which include four distinct agro- ecosystems. Work in ornamentals is increasing. Relationships between monitoring results and field failure are primarily anecdotal at this time. References 140, 227, 286, 314
Develop, evaluate and refine resistance management systems.	Evaluate the effects of mixtures and rotations of new and old chemistries to mitigate selection for resistance. Develop methods to evaluate and augment the beneficial influence of refuges as sources of susceptible genes to the population pool.	Х	Prabhaker et al. in press. Studies to increase horizontal integration of resistance management programs are addressing influence of refuges in diverse agro-ecosystems. References 20, 47, 76, 77, 235, 245, 284, 310
Integrate chemical control with other tactics.	Evaluate selectivity of synthetic insecticides and natural products to key whitefly natural enemies. Test compatibility of biological control with selective synthetic or natural product insecticides as required.	Х	Most efficacy trials include a compatibility evaluation, selectivity evaluations include systemics, entomopathic fungi; life tables are contributing to our understanding here. References 7, 9, 10, 11, 13, 24, 25, 27, 31, 32, 37, 38, 49, 70, 89, 90, 93, 96, 97, 98, 105, 110, 129, 147, 149, 153, 160, 169, 174, 194, 200, 247

# Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods. (Continued)

<sup>a</sup> See Table A for complementary research on thresholds.
 <sup>a</sup> See Table B for complementary research on virus/vector interactions.
 <sup>a</sup> See Table D for complementary research on biological control.
 <sup>b</sup> See Table E and F for complementary research on systems management.

Research Approaches <sup>a</sup> Improve insecticide efficacy:	Year 3 Goals Statement	Progress Achieved Yes No	Significance
Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Develop new chemistries and natural products. Develop improved techniques for evaluating efficacy of insecticides. Support registration of desirable new products by providing information to regulatory agencies. Determine new modes of action of effective materials. Elucidate biochemical pathways of synthesis and degradation of natural products. Same as Year 2. Evaluate the potential for transforming plants with natural product genes.	X	Section 3 registration of IGRs & Admire in additional crops, Section 18's supported, work with Rimon a new IGR, continued work with neonicotinyls as soil and foliar applications, sugar esters, azadirachtins and entomopathic fungi, toxicity of abamectin, tebufenozide, chlorfenpyr & pymetrozine evaluated, integration of biorationals and conventional chemistries, work on products that induce resistance has some promise, sampling plan in cantaloupes, life table studies, evaluated the impacts of FQPA References: 59, 60, 83, 145, 164, 167, 183, 184, 189, 192, 210, 214, 252 Tomato transformations – Florida.
Develop improved methods of application including formulation and delivery of materials to improve control.	Develop spray systems for better underleaf coverage. Evaluate rates, timing, placement in relation to efficacy. Consider formulation, UV protectants, and other means to improve efficacy. Develop improved methods to evaluate application efficacy. Field test under commercial conditions for technology transfer. Same as Year 2	Х	Additional work with swivel nozzles, several commercial studies examining drip and at planting methods of application, ultrasonic fogging devices for greenhouses, application methods to improve efficacy of entomopathic fungi, the use of DEF & PBO to enhance efficacy and overcome resistance. References: 76, 77, 244,
Conserve insecticide efficacy:			
Relate action thresholds to insecticide usage patterns.	Refine action thresholds based on insecticide efficacy and input from other control strategies Same as Year 2	Х	Continued work on sticky cotton sampling plan, development of IGR specific action thresholds in melons, Yield, sticky cotton – whitefly relationships studied. References: 18, 95, 182

## Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods.

		Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 3 Goals Statement	Yes	No	Significance
Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.	Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide. Evaluate the role of refuge habitats (weeds, tolerant crops, urban areas) to assure input of susceptible genes in whitefly population. Evaluate the influence of host plant on susceptibility to insecticides.	Х		Genetic and biochemistry studies are concentrated on the role of MFO-based metabolism and esterases in resistance to nicotinyls and bifenthrin; cross resistance being studied with nicotinyls and neonicotinyls. Impact of ecological factors such as nutrition, host plant response, local cropping patterns are being studied. Role of alfalfa as a refuge has been evaluated. Host plant influence on efficacy has been evaluated based on seasonality, external & internal uptake, changes in metabolites and several compounds screened on various host plants. References: 12, 128, 185, 220
Improve insecticide efficacy:				
Improve techniques for monitoring resistance.	Establish baseline data on toxogenic responses of whitefly populations to new insecticides. Same as Year 1. Expand comparative studies of resistance levels in diverse agro-ecosystems. Evaluate relationship between monitoring results and field efficacy. Same as Year 2. Summarize, analyze, and produce standardized comparable monitoring systems.	Х		Baselines have and are being established for IGRs and systemic insecticides. Work in Cal. & Arizona are evaluating regional resistance management techniques which include four distinct agro- ecosystems. Work in ornamentals is increasing. Relationships between monitoring results and field failure are primarily anecdotal at this time. References: 73, 74, 75

 Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods. (Continued)

× •	s, resistance management, and reprication	Progress			
Research Approaches <sup>a</sup>	Year 3 Goals Statement	Yes	No	Significance	
Develop, evaluate and refine resistance management systems.	Evaluate the effects of mixtures and rotations of new and old chemistries to mitigate selection forresistance. Same as Year 1. Develop methods to evaluate and augment the beneficial influence of refuges as sources of susceptible genes to the population pool. Same as Year 2. Develop criteria for integration of successful strategies in agricultural systems. Field test resistance management systems as long range components of successful IPM.	Х		Prabhaker et al. in press. Studies to increase horizontal integration of resistance management programs are addressing influence of refuges in diverse agro-ecosystems. Areawide resistance management programs in place in Cal. and Arizona.	
Integrate chemical control with other tactics.	Evaluate selectivity of synthetic insecticides and natural products to key whitefly natural enemies. Same as Year 1. Test compatibility of biological control with selective synthetic or natural product insecticides as required. Same as Year 2. Integrate systems with host plant resistance and cultural controls	Х		Most efficacy trials include a compatibility evaluation, selectivity evaluations include IGR's, neonicotinyls, entomo pathic fungi; life tables are contributing to our understanding here. Integration of smooth leaf varieties of cotton with chemical control being practiced.	

#### Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods. (Continued) Ashiavad

<sup>a</sup> See Table A for complementary research on thresholds.
 <sup>a</sup> See Table B for complementary research on virus/vector interactions.
 <sup>a</sup> See Table D for complementary research on biological control.
 <sup>b</sup> See Table E and F for complementary research on systems management.

Research Approaches <sup>a</sup>	Year 4/5 Goals Statement	Progress Achieved Yes No	d Significance
Improve insecticide efficacy: Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Evaluate the potential for transforming plants with natural product genes.	X	Several new registrations were granted for a number of products including: sucrose esters, neem basewed products, Applaud (Courier), Esteem and Platinum, Actara and Assail. Work to determine efficacy and chemical use patterns continued with neonicotinyls as soil and foliar applications, sugar esters, azadirachtins and entomopathic fungi, and pymetrozine. No progress was achieved in evaluating the potential for transforming plants with natural product genes
Develop improved methods of application including formulation and delivery of materials to improve control.	Develop spray systems for better underleaf coverage. Evaluate rates, timing, placement in relation to efficacy. Consider formulation, UV protectants, and other means to improve efficacy. Develop improved methods to evaluate application efficacy. Field test under commercial conditions for technology transfer	X	No new advancements in foliar spray delivery systems or assessment techniques were reported in 2002. A considerable amount of progress was made in evaluating efficacy relative to insecticide rates, timing, and placement. Progress in this area is highlighted in several abstracts.
Conserve insecticide efficacy: Relate action thresholds to insecticide usage patterns.	Refine action thresholds based on insecticide efficacy and input from other control strategies. Summarize and recommend in IPM systems. Summarize and recommend in IPM systems.	X	Very little new work on action thresholds was reported in 2002. The most significant work was done in tomatoes to develop AT for IGR use (D.J. Schuster). See section F for technology transfer.
Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.	Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide. Evaluate the role of refuge habitats (weeds, tolerant crops, urban areas) to assure input of susceptible genes in whitefly population. Evaluate the influence of host plant on susceptibility to insecticides.	X	Progress was made in characterizing cross resistance among neonicotinoid insecticides. Monitoring of whitefly susceptibility to IGR, neonicotinoids and other chemistries continued in in AZ, CA, and FL with no major shifts in susceptibility measured.

# Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods.

		Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 4/5 Goals Statement	Yes	No	Significance
Improve insecticide efficacy:				
Improve techniques for monitoring resistance.	Summarize, analyze, and produce standardized comparable monitoring systems. Develop standard systems for general use including user friendly techniques to assist growers and extension agents to evaluate susceptibility of whitefly populations to commonly used insecticides.	Х		Significant progress was made in determining the parameters of chemical uptake of imidacloprid through petioles in systemic bioassays. This information will allow for the improvement of adul bioassays for systemic compounds based on concentration taken up by leaves.
Develop, evaluate and refine resistance management systems.	Develop criteria for integration of successful strategies in agricultural systems. Field test resistance management systems as long range components of successful IPM. Technology transfer.	Х		Progress was made in AZ to expand the AZ Whitefly IRM/IPM program to implement recommendations for the shared use of buprofezin and neonicotinoids among commodities such as cotton, melons and vegetables.
Integrate chemical control with other tactics.	Test compatibility of biological control with selective synthetic or natural product insecticides as required. Integrate systems with host plant resistance and cultural controls	Х		Significant progress was made in determining the effect of IGR and other chemistries in conservation of natural enemies and additive effects to chemical control – "Bioresidual "
	Integrate systems with host plant resistance and cultural controls. Summarization and technology transfer			

# Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods. (Continued)

<sup>a</sup> See Table B for complementary research on virus/vector interactions.
 <sup>a</sup> See Table D for complementary research on biological control.
 <sup>b</sup> See Table E and F for complementary research on systems management.

#### **Reports of Research Progress**

#### Section D: Natural Enemy Ecology and Biological Control Co-Chairs: Bill Roltsch and Greg Simmons

Investigator's Name(s): Earl Andress<sup>1</sup>, Julie Gould<sup>2</sup>, Larry Heilman<sup>3</sup>, Arland Oleson<sup>3</sup>, and Mark Quinn<sup>4</sup>

Affiliation & Location: 1. USDA-APHIS, Imperial Valley Research Station, Brawley CA., 2. USDA-APHIS-PPQ, Otis Protection Center, Otis MA, 3. Department of Biochemistry, North Dakota State University, Fargo ND, 4. Department of Crop and Soil Sciences, Washington State University, Pullman WA

Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

Dates Covered by the Report: January 1, 2001 - December 31, 2001

#### Exotic Parasitoids in Four Crops in the Imperial Valley

Exotic parasitoid species introduced into the Imperial Valley of California were recovered throughout the agricultural system in 2000 and 2001. No parasitoids were released at all in 2001, and the last of the mass releases were made during the summer of 1999. Samples were collected from four crops in 2000 and 2001 and tested for parasitism using the Squash blot methods developed by (Heilman et al. ibid). Percentages of whiteflies parasitized by exotic parasitoid species were as high as 32, 28, 52, and 28 in broccoli, cantaloupe, alfalfa, and cotton respectively, and were as low as 0. The high variability in parasitism by exotics observed here may suggest that although populations of exotic introduced species of Aphelinid parasitoids are well established in the Imperial Valley, their influence on whiteflies has the potential to continue to grow in coming years. In some cases exotic parasitoids even outnumbered indigenous parasitoid. The broad range of parasitism levels observed confirms that factors affecting parasitism of whiteflies are highly variable within the Imperial Valley agricultural system. Factors that will be quantified and analyzed for their effect on both whitefly and parasitoid populations observed during the course of this study will include distance to the edge of the agriculture areas, coverage of host crops, coverage of non-host crops, proximity to urban areas, insecticide use, and predation.

Investigator's Name(s): James S. Buckner<sup>1</sup> and Walker A. Jones<sup>2</sup>

**Affiliation & Location:** <sup>1</sup>USDA-ARS, Insect Genetics and Biochemistry Research Unit, Red River Valley Agricultural Research Center, Fargo, ND; <sup>2</sup>USDA-ARS, Beneficial Insects Research Unit, Subtropical Agricultural Research Center, Weslaco, TX.

Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: 2001

#### Methyl-Branched Alkanes as Possible Marking Pheromones for Female Eretmocerus mundus, a Parasitoid of Bemisia argentifolii

Previous studies with *Bemisia argentifolii* Bellows and Perring (Homoptera: Aleyrodidae) nymphs have shown that wax esters were the major components of the cuticular lipids with lesser amounts of hydrocarbons, long-chain aldehydes and long-chain alcohols (1). The hydrocarbon fraction was identified as a homologous series of odd-carbon-number *n*-alkanes,  $C_{31}$  (40%),  $C_{33}$  (31%),  $C_{35}$  (18%) and  $C_{29}$  (11%) and trace amounts of even-carbon-number *n*-alkanes ( $C_{28}$ - $C_{34}$ ). In a more recent study, the cuticular lipids from *B. argentifolii* nymphs parasitized by the whitefly parasitoid *Eretmocerus mundus* Mercet contained measurable quantities of two additional components in their hydrocarbon fraction (2). Capillary gas chromatography (CGC) and CGC-mass spectrometry (CGC-MS) analyses and comparisons with an authentic standard indicated that the two hydrocarbons were the monomethyl-branched alkanes, 2-methyltriacontane (31 carbons) and 2-methyldotriacontane (33 carbons). In parallel experiments, no appreciable changes in lipid composition were observed for the cuticular lipids of whiteflies parasitized by another parasitoid, *Encarsia pergandiella*. In contrast to *Encarsia* spp., *Eretmocerus* spp. oviposit externally (under the nymph), yet still readily avoid superparasitism suggesting that an external discrimination cue must be present (2). Previous behavioral observations with *E. mundus* showed that, following oviposition, females often make apparent "host marking movements" that may leave a chemical that enables the searching female to distinguish parasitized from unparasitized hosts (3). Those comparative findings suggest a possible function of 2-methylalkanes as marking pheromones for ovipositing female *E. mundus*.

The following set of experiments decreased the possibility that the extra 2-methylalkanes on cuticular surfaces of *E. mundus*parasitized nymphs arose from biosynthesis and surface deposition during growth and development of the parasitoid, and supported the possibility for transfer of these compounds onto the surface of the host (nymphs) by searching/ovipositing *E. mundus* females. *B. argentifolii* nymphs were exposed to *E. mundus* females for approximately 8 hr and then removed from leaves after 48 hr. Only those nymphs with visible signs of an oviposited egg beneath each nymph were removed and pooled for subsequent analysis of their cuticular lipids. The hydrocarbon fraction of nymph samples that had been exposed to ovipositing *E. mundus* females showed the same 2-methylalkanes that were also present on those parasitized *B. argentifolii* nymphs that had been held 10 days after exposure to *E. mundus* females. In addition to 2-methylalkanes, small quantities of 31- and 33-carbon dimethyl-branched-alkanes were also identified in the hydrocarbon fraction from those nymphs exposed to *E. mundus* for 48 hr. Preliminary CGC-MS analyses of the cuticular lipids from *E. mundus* females have revealed major quantities of the same 2-methylalkanes and dimethyl-branched-alkanes that were on the cuticular surfaces of *E. mundus*exposed *B. argentifolii* nymphs. These findings clearly demonstrate that the origin of the 'extra lipids' on the surfaces of *E. mundus*exposed *B. argentifolii* nymphs is from the ovipositing female and that these lipids could be playing a role as host marking chemicals. Current studies are focused on defining the process for transfer of these methyl-branched lipids from the ovipositing parasitoid to its host.

#### References

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- 2. Buckner et al. 2000. Arch. Insect Biochem. Physiol. 44: 82-89.
- 3. Foltyn and Gering 1985. Entomol. Exp. Appl. 38: 255-260.

Investigators Name(s): Matthew A. Ciomperlik & John A. Goolsby<sup>2</sup>

**Affiliation & Location**: USDA-APHIS-PPQ Mission Plant Protection Center, P.O. Box 2140, Mission, TX 78573-2140; <sup>2</sup>USDA-ARS, Australian Biological Control Laboratory, Private Mailbag #3, Indooroopilly, Queensland, Australia, 4068

Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: Sep 1999 -Dec. 2000

#### Establishment of Introduced Parasitoids of Silverleaf Whitefly in the Lower Rio Grande Valley of Texas

Since 1993, a large number of exotic *Eretmocerus* and *Encarsia* parasitoid species/strains have been evaluated in quarantine, laboratory, and field studies to determine promising species for release in subtropical agroecosystems in the lower Rio Grande Va lley (LRGV) of Texas. Those evaluations suggested that *Eretmocerus hayati* Rose & Zolnerowich (M95012, Pakistan), *Eretmocerus* nr *emiratus* (M96076, Ethiopia), and *Eretmocerus mundus* (M92014, Spain) might become established on whitefly in crops grown in the region, because they attacked significantly greater numbers of hosts than the native parasitoid species. Prior to exotic parasitoid releases, the native parasitoid complex attacking silverleaf whitefly in the LRGV was comprised of six parasitoid species. Those species included; *Eretmocerus tejanus* Rose & Zolernowich, *Encarsia pergandiella* Howard, *E. meritoria*, *E. nr strenua*, and *E. formosa*. Of those, *Eretmocerus tejanus* and *Encarsia pergandiella* were the most abundant species collected from all crop types and weed host plants sampled (Riley & Ciomperlik, 1997). These two parasitoid species were widely distributed across the region and collected from almost all crop and weed host plant types.

Field releases of exotic parasitoid species/strains occurred from 1993 to 1998 across multiple release sites, as either direct releases of small to large numbers of parasitoid adults onto hosts within the crop, or as large scale evaluations of parasitoid release methods (Goolsby & Ciomperlik 1996). Establishment evaluations during June of 1997 showed that the native parasitoids comprised approximately 98% of all parasitoids collected. Subsequent monthly evaluations in 1998 showed increasing numbers of exotics becoming established through time (Goolsby et al. 1999). Those evaluations during 1998 showed that *Eretmocerus hayati* and *Eretmocerus mundus* were at times as abundant as the native *Eretmocerus tejanus*.

Field sampling to determine parasitoid establishment, abundance trends, and distribution were conducted in 1999 and 2000. Three sampling methods were routinely used to collect parasitoid samples from economically important crops and weed host plants within the LRGV. The three sampling methods were; 'recovery samples' where release sites were re-visited either 1 or 2 years after initial releases were made, 'random samples' where crops were sampled at multiple random locations within the same county as where the parasitoids were released, and 'triplicate sentinel' host plants infested with second instar silverleaf whitefly placed in large acreage crop production areas. In the case of the recovery and random samples, leaf samples containing whitefly pupae were returned to the laboratory, where individual parasitized pupae were isolated in <sup>1</sup>/<sub>4</sub> dram glass vials for emergence. In the case of the triplicate sentinel plant method, three seedling plants including cotton, cantaloupe, and cucumber infested with 2<sup>nd</sup> instar whitefly were placed within the crop canopy at each sample site. During the winter months, cabbage seedlings were substituted for the cucumber seedlings. After three days of exposure, the three seedling sentinel plants were returned to the laboratory and held for parasitoid development and handled as described above. All of the emerging parasitoids adults were identified to genus and species where possible using morphological characters. All suspect identifications were confirmed by RAPD-PCR analysis.

Analysis of all samples indicate that *Eretmocerus hayati* (Pakistan) is ubiquitously distributed amongst all the crop and weed host plants sampled in all counties with the exception of Starr County. This parasitoid accounted for 96.5% of the *Eretmocerus*, and 88% of all species of parasitoids collected in the establishment evaluations (n=866 parasitoids). *Eretmocerus mundus* comprised about 3%, while *E*. nr *emiratus* comprised about 0.6% of all *Eretmocerus* individuals collected. *Eretmocerus tejanus* and *Encarsia pergandiella* were infrequently collected, comprising 0.6% and 4.8% of all parasitoids collected. Attack rate patterns by the various native and exotic parasitoids are still under analysis at this time. The low numbers of recoveries of native parasitoids may suggest that exotic and native populations have not reached equilibrium densities, therefore establishment evaluations will continue in 2001.

**Investigator's Names:** Larry J. Heilmann<sup>1</sup>, Jianzhong Zhang<sup>1</sup>, Earl R. Andress<sup>2</sup>, and Arland E. Oleson<sup>1</sup>, Dennis R. Nelson<sup>3</sup>, Juli R. Gould<sup>4,6</sup>, Matthew W. Ciomperlik<sup>5</sup>, John A. Goolsby<sup>5,7</sup>, and Don C. Vacek<sup>5</sup>

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Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

Dates Covered by the Report: 1996-2001

#### DNA Based Species Identification Test for Parasitic Wasps of the Whitefly

A significant problem in using parasitic wasp species in biological control of the whitefly is identification of the individual wasp species parasitizing the nymph. This is particularly important in areas where imported foreign species are released. It is necessary to follow these releases to determine which species establish and reproduce, if they effectively parasitize local whitefly populations and if they displace native parasitoids. Current taxonomic tests for species identification are time consuming and require experienced and trained personnel.

We have developed a simple and relatively quick DNA based species identification test that can be applied to squashes of adult, pupal, and larval stage *Eretmocerus* and *Encarsia* wasps parasitizing whiteflies. The test utilizes highly repetitive but species specific DNA sequences to identify the parasitizing wasp in one to two days. We currently have cloned DNA probes for *Eretmocerus mundus (Spain), E.sp. (Ethiopia), E. hayati*, and *E. eremicus* as well as *Encarsia formosa* and *Encarsia sophia (transvena)*. The probes range from highly species-specific to only being selective for old world vs new world species. Each of the cloned probe DNAs has been sequenced and their distribution in relevant parasitoid species quantified.

In a major field test of the procedure over 26,000 whitefly larvae from the Imperial Valley of California were squashed on filters and hybridized with the probes. These larvae were from multiple locations, multiple crops and different times of the year. Approximately 20% of these larvae were parasitized by the native California species, *E. eremicus*, with individual field variations ranging from 0 to 77%. Imported species from Europe, Africa and Asia parasitized only 3-4% of the larvae. Results on this field test will be presented.

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#### Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: Summer-fall 2001

#### Parasitism of the Silverleaf Whitefly in North Florida

The objective of this study is to assess the population densities of the silverleaf whitefly (SLWF), *Bemisia argentifolii*, and the degree of parasitism (from native and exotic parasites) in various vegetables and alternate plant hosts in north Florida. Surveys for silverleaf whitefly were conducted during the summer of 2001 in vegetables, including tomatoes, squash, cabbage, watermelon, and okra and in ornamentals, such as poinsettias and hibiscus. During September to October 2001, a minimum of 10 leaves were collected from soybean (*Glycine max*), poinsettia (*Euphorbia pulcherrima*), and lantana (*Lantana camara*) in Leon and Gadsden Co. FL and returned to the laboratory for data processing. Counts of the different stages and numbers of whiteflies per leaf were made and recorded using a stereomicroscope. The leaf samples were kept in emergence cages (plastic petri dishes or plastic boxes) that were painted black with a glass vial attached to the side of the cage. Each leaf petiole was inserted into a small plastic vial filled with water to keep the leaves from drying inside the cage. The emergence cages were kept in an environmental growth chamber at 28-30 °C and 14L:10D photoperiod. Parasites that emerged were collected in the vial and placed in 70% alcohol. The parasite specimens were sent to local taxonomists for identification of species.

Surveys for silverleaf whitefly populations during the summer of 2001 indicated a very low population of SLWF. Preliminary results indicated that the mean numbers of whitefly per leaf were highest in lantana (53.3) followed by poinsettia (31.50) and soybean (14.50). Average percentage of parasitism of SLWF was highest in soybean (12.12%), followed by poinsettia (7.89%) and lantana (3.81%). The parasite:whitefly ratio was calculated as 7.25, 11.67 and 25.26 in soybean, poinsettia and lantana, respectively. The main parasites of SLWF that emerged from poinsettia were of the genus *Encarsia* sp. while those from lantana and soybean were of the genus *Eretmocerus* sp. (both Hymenoptera: Aphelinidae). Further species identification will be confirmed by local taxonomists.

Investigator's Name(s): C.L. McKenzie, W.B. Hunter, S.L. Lapointe and P. Dang

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Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: 2000-2001

#### Iridovirus: Potential Entomopathogenic Biocontrol Agent for Whitefly

Few microbial biocontrol agents and no known viral agents are commercially available for use on whiteflies. A newly described entomopathogenic virus was recently discovered in south Florida whitefly populations and was determined to be an iridovirus by DNA analysis. Invertebrate iridescent virus 6 (IIV-6) is known to be pathogenic to insects including whiteflies. Demonstrated modes of transmission for iridovirus in other insect systems have been shown to be through oral ingestion, cuticular wounding (abrasions) and occasionally sexually transmitted. Traditionally, many biological agents are applied by foliar spray application; therefore, we attempted to determine if this would be a feasible method of application of iridovirus to control whiteflies on tomatoes.

A plant-based petri dish bioassay system was developed to hold single tomato leaves infested with whitefly for their entire life cycle (egg to adult). A hole was cut in the lid of a Polystyrene petri dish (100x20 mm) to facilitate ventilation and ultrafine screen mesh was used to cover the hole to prevent whitefly escapes. Polystyrene conical tubes (15 ml, 17x120 mm) were filled with a plant tissue culture water agar and fertilizer (9-45-15) mixture. A hole was cut at the base of the petri dish to hold the conical tubes so that it fit tightly. Tomato leaves were cut so that the terminal leaf of the leaflet would fit in the petri dish. The remaining leaves of the leaflet were cut off and the stem was placed as far into the agar as possible with the abaxial leaf surface facing the screen to facilitate counting. A small hole was drilled into top of the conical tube above the agar line so that  $H_20$  could be added as needed (leaves will sprout roots and last > 28 d).

The iridovirus solutions were applied using an ultra-low volume spray device that consisted of a spray platform that holds a pressurizable spray bottle at the proper distance and angle. Measured amounts (200 or 400  $\mu$ l) of iridovirus solution were placed in a test tube (12 x 75 mm). The spray bottle siphon tube was placed in the test tube so that the test tube fit into the nozzle-pump body. Parafilm was wrapped around the top of the test tube to insure a tight fit and the spray bottle was pressurized to ~10 psi w/ 20 strokes of the pump mechanism to deliver a fine spray for each application.

Uniform infestations of whitefly were allowed to acclimate on tomato leaves in the petri dish bioassay system. Abaxial and adaxial leaf surfaces were sprayed with 200  $\mu$ l of crude virus preparation in a cold room. Controls were sprayed with PBS buffer only. Petri dishes were placed upright in a Percival Scientific incubator at  $22 \pm 1^{\circ}$  C under a L/D, 16:8 photoperiod. Whitefly were harvested at 10, 14 and 18 days post spray and were subjected to PCR analysis for presence of IIV6. The experiment was repeated using the purified virus preparation and only the adaxial leaf surfaces were sprayed with 400  $\mu$ l of iridovirus solution.

We confirmed adult whitefly infection with IIV-6 through oral ingestion via sucrose parafilm feeding chamber experiments and PCR analysis. Whiteflies were fed a 15% sucrose solution with crude or purified virus preparation for 72 hr and transferred to tomato leaves in the petri dish bioassay system for 14 days to allow the virus to replicate. Controls were fed sucrose only. Whiteflies were analyzed via PCR for presence of IIV6.

Although we could demonstrate infection in whiteflies *per os*, spray applications were inconclusive. When virus solutions were sprayed onto the insects and plant tissues no apparent virus infection was detected. This may be due low virus titer in our spray solutions; whiteflies did not ingest surface droplets when feeding, and/or the inability of the virus to remain viable on the leaf surface. Experiments are underway to try and address these concerns.

**Investigator's Names:** Steven E. Naranjo<sup>1</sup> & Nilima Prabhaker<sup>2</sup>

**Affiliation & Location:** <sup>1</sup>USDA-ARS, Western Cotton Research Laboratory, Phoenix, AZ and <sup>2</sup>Department of Entomology, University of California, Riverside, CA

Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

Dates Covered by the Report: January 2000-December 2001

#### Toxicological Studies of Two Insect Growth Regulators on Geocoris punctipes, Orius insidiosus and Collops vittatus

Laboratory experiments were continued to evaluate the direct effects of two insect growth regulators (IGR), buprofezin (chitin synthesis inhibitor) and pyriproxyfen (JH analog), on survival and reproduction of *Geocoris punctipes*, *Orius insidiosus* and *Collops vittatus*, 3 common generalist predators found in cotton. Both topical and contact residue assays were performed on adults and terminal-stage nymphs at recommended field rates of the materials. Buprofezin was found benign to all species and stages of exposure in terms of survivorship and reproduction. This is consistent with previous dose-response studies with *G. punctipes* where is was impossible to estimate lethal concentrations. Pyriproxyfen applied to adults had no effect on survival or reproduction of *G. punctipes* or *O. insidiosus*. However, egg viability, but not survival, of *C. vittatus* appeared to be affected by pyriproxyfen. Pyriproxyfen applied to terminal nymphal stages of *G. punctipes* and *O. insidiosus* caused some mortality and adult deformities compared to the control. Pyriproxyfen did not affect egg viability of treated adults or adults arising from treated nymphs of either of these species.

#### Affiliation & Location: USDA-ARS, Western Cotton Research Laboratory, Phoenix, AZ

#### Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: January 2000-December 2001

#### Comparison of Natural Enemy Fauna and Function in Transgenic Bt and Non-Bt cotton

Field studies were conducted in 1999-2001 to evaluate the combined direct and indirect effects of transgenic-Bt cotton on populations of natural enemies. Paired non-Bt and Bt fields of the same background were established and monitored weekly for foliar dwelling natural enemies. In 2001, each treatment was further split to contrast completed unsprayed plots to those receiving insecticides as needed for whitefly, *Lygus* and lepidopteran pests. Analyses are incomplete, but preliminary results indicate no consistent differences in predator populations between Bt and non-Bt cotton. Parasitism by aphelinids attacking *Bemisia tabaci* was very low and unaffected by Bt cotton. Additional studies were conducted in 2001 to examine effects on natural enemy function. Sentinel pink egg masses were placed in plots to estimate rates of predation and parasitism on pink bollworm eggs. Cohort-based life table studies were conducted to estimate rates of predation and parasitism on immature *B. tabaci*. Preliminary results indicate that rates of natural enemy -induced mortality on these two key pests were the same in Bt and non-Bt cotton.

**Investigators Name(s)**: Alberto Pantoja<sup>1</sup>, Matthew A. Ciomperlik<sup>2</sup>, Norberto Gabriel<sup>3</sup>, Pedro Vazquez<sup>3</sup>, Leyinska Wiscovitch<sup>3</sup>, and Wilfredo Robles<sup>1</sup>

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Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: 1999-2001

#### Evaluation of the Establishment and Distribution of Exotic Biological Control Agents of Silverleaf Whitefly in Puerto Rico

Silverleaf whitefly (SLWF) is a key pest of many broadleaf vegetables, field crops, and ornamentals in tropical and subtropical regions. Losses caused by plant debilitation, sooty mold growth, physiological disorders and transmission of geminiviruses by whiteflies, and control costs, have impacted vegetable production systems in Puerto Rico significantly. In efforts to combat these losses, a biological control program was cooperatively implemented by USDA/APHIS/PPQ and the University of Puerto Rico to control SLWF. The biological control program included both classical and augmentative approaches. From 1996 to 2000 the USDA/APHIS/PPQ laboratory in Mission, Texas and the local USDA/APHIS/PPQ office released 1,262,536 parasitoids in 20 locations throughout Puerto Rico. The species released include *Encarsia formosa*, *Encarsia sophia (transvena)*, *Eretmocerus mundus*, *E. hayati*, *E. emiratus*, *Eretmocerus sp. nr* emiratus and a predatory coccinellid *Serangium parcesetosum*.

Recovery studies were initiated during 1999 to determine establishment and distribution of the parasitoids released in the collaborative effort. Five hundred and thirty specimens have been recovered from twenty locations and shipped to the USDA-APHIS-PPQ Mission Biological Control Center (MBCC) in Texas for genetic identification. The specimens were subjected to RAPD-PCR analysis using the OPC-04 primer at the MPPC. *Eretmocerus mundus, E. tejanus, and E. haytii* represents 16, 0.6, and 1% respectively of the specimens recovered.

Reconciliation of data records indicating the initial release and the recovery site indicates that parasitoids can be recovered three years after the initial liberation. This suggests that the parasitoids have established on the island. The data also suggest that several native *Encarsia* species might be present. The native parasitoid fauna of Puerto Rico has not been well established, however, the predominant parasitoids collected during pre-release field sampling indicated that several *Eretmocerus* species are present but are less numerous than the native species of *Encarsia transvena* (M. Ciomperlik, Unpublished Data). From pre -release evaluations, it was clear that the native parasitoid complex was not well suited to utilize *Bemisia argentifolii* as a host. After the release of the exotic *Encarsia* and *Eretmocerus* species, growers have indicated that whitefly pressures have been less severe, that yields have increased, and the use of insecticide reduced. After two years the savings associated to the reduction of insecticide use by augmenting natural enemies is estimated to exceed \$1200 /acre (Pantoja unpublished data). The data indicates that the exotic parasitoids have become established and are regulating whitefly populations.

Investigator's Name(s): C. H. Pickett, J. Brown, G. Simmons<sup>1</sup>, J. Goolsby<sup>2</sup>, and Bill Abel<sup>3</sup>

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Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

Dates Covered by the Report: August 1997 to November 2001

#### Releases of Exotic Parasitoids for Permanent Establishment in Central California

The silverleaf whitefly, *Bemisia argentifolii*, was an increasingly important pest of cotton in the San Joaquin Valley from 1994 through 1997 when this project was initiated. Field studies suggested that citrus had become an important overwintering site for this whitefly. Cotton has the highest incidence of whitefly infestations in areas of the Valley with a matrix of citrus orchards and cotton fields. We report on large scale releases of *Eretmocerus emiratus* (M95104, U.A.E.), *Eretmocerus* nr. *emiratus* (M96076, Ethiopia), *E. mundus* (M92014, Spain), *E. hayti* (M95012, Pakistan), and trace numbers of *Encarsia sophia* (= *transvena;* M95107, Pakistan) into four citrus orchards. The study had two goals: (1) to determine if exotic parasites released into citrus during the fall will overwinter in this habitat and move into cotton the following spring; and (2) to permanently establish new populations of exotic parasites specific for the silverleaf whitefly.

Three study sites were identified initially, one each in Fresno, Tulare, and Kern counties. A fourth was added because one of the original growers stopped farming cotton (Kern Co.). Sites consisted of citrus and cotton acreage managed by the same owner. Cotton is grown directly adjacent to the citrus, and growers have had a history of silverleaf whitefly problems. We began releasing parasites in early August or September 1997, 1998, 1999, and 2000 when migrating whitefly nymphs were first recorded from citrus leaves. Typically, over 100,000 parasites were released weekly at each location and a total of 4.05 million were released in 1997, over 10 million in fall 1998, 3.2 million in 1999, and 124,000 in 2000. The dispersal of the released parasites was recorded using sticky cards with identification based on the adult males since they could be readily distinguished from native *Eretmocerus* while on the sticky cards.

Parasitism of silverleaf whitefly on citrus was generally quite low, averaging 28% overall. However this value is quite high with respect to an earlier survey in which less than 1.5% of nymphs examined on cotton from the same region were found parasitized. Whitefly densities on citrus remained very low, usually less than 0.1 nymphs per cm<sup>2</sup> leaves. During years in which exotics were being released, most of the parasitoids recovered from weed samples taken within 1 mile of citrus orchards were exotic, 81% to 95%. Two years after the last releases of exotic *Eretmocerus* spp., fall 2001, the proportion of all parasitoids sampled from weeds that were exotic dropped to 11%. Although primarily *Eret. emiratus* (M95104 + M96076) was released, *Eret. mundus* (M92014) was the dominate species recovered in fall 2001. The density of silverleaf whitefly on weeds has varied about the same from 1998 to 2001, most samples from 0.1 to less than 20 per gram dry weight.

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Research & Implementation Area: Section D: Natural Enemy Ecology and Biological Control

Dates Covered by the Report: August 1994 - Nov. 2001

#### Establishment of Introduced Parasitoids of the Silverleaf Whitefly in Imperial Valley, CA

Several species and strains of exotic *Eretmocerus* and *Encarsia* were evaluated in field cages, and released in large numbers in commercial fields, refuge nursery plots and urban yards in Imperial Valley by State, Federal and University scientists. The most promising *Eretmocerus* for this desert region include *E. emiratus* Zolnerowich & Rose, *E. sp.* nr. *emiratus* (from Ethiopia) and *E. mundus* Mercet. *Encarsia sophia* (*=Encarsia transvena*) from Maltan, Pakistan appears promising as well. Identification to species was accomplished using recently published morphological keys and by DNA analysis (RAPD-PCR) by the USDA-APHIS, Plant Protection Center, Mission, TX. This report is a summary of silverleaf whitefly (SLW), *Bemisia argentifolii* Bellows & Perring, parasitoid population development in long-term refuge nursery plots from 1994 to 2001, and a three-year, late season survey of exotic parasitoids in commercial cotton within the Valley.

From 1994 through 1997, exotic parasitoids were released into long-term refuge field plots on multiple occasions each year. Plots (1/2 to 1 acre) were located at the Imperial Valley Research Center near Brawley, CA, and at an organic farm at the south end of the county. During the warm season, the plots consisted of okra and basil. During the cool season, cole crops (esp. collard) and sunflower were present. Kenaf, roselle and eggplant were also periodically present (1994-1996) along with adjacent plantings of cotton and spring cantaloupe. Leaf samples were taken approximately 6 times during each year to determine the status of parasitoid population increase and persistence. Neither E. tejanus nor E. stauferi (i.e., Eretmocerus spp. from Texas) have been recovered following their release in 1994. During 1995, E. melanoscutus was released in large numbers but recoveries of this parasitoid were rare. Releases of E. mundus, E. hayati and E. emiratus began in April of 1996. Numbers of exotic parasitoids compared to natives were high during early summer; however, the proportion of the sample consisting of exotic species dropped markedly by late July, indicating poor performance (population increase and persistence) during this very warm summer period. During 1997, E. emiratus (origin: United Arab Emirates) and E.sp. nr. emiratus (origin: Ethiopia) were released. The relative performance of exotics was considerably better than in 1996. Several samples from May to September consisted of up to 90% exotic Eretmocerus spp. versus native Eretmocerus eremicus. The proportion of exotic *Eretmocerus* relative to native *Eretmocerus eremicus* declined once again during the fall, however, not to the same extent. During 1998, none of the long-term refuge plots was inoculated with exotic whitefly parasitoids. This made possible the assessment of populations released in previous years at these sites, in terms of their ability to overwinter and compete with native species of SLW parasitoids. Overwintering on cole crops was confirmed albeit in low numbers. During the summer of 1998, 1999, & 2000 exotic Eretmocerus spp. were dominant over native E. eremicus on okra, basil and adjacent cotton in nearly all samples taken. The order of dominance of exotic Eretmocerus species is E. sp. nr. emiratus, E. emiratus and E. mundus. Encarsia sophia reached high densities during the summer and fall of 1998 through 2000 in several of the refuge field plots as well. During 2001 SLW densities on most plant species were very low. From a considerably smaller number of samples compared to previous years, it was indicated that exotic *Eretmocerus* spp. were not nearly as dominant as in the past three years, resulting in most samples containing well over 50% native E. eremicus.

Further evidence regarding the extent of exotic parasitoid establishment was provided by surveying cotton fields from 1998 to 2000. Leaf samples were obtained from three edges of conventionally managed cotton fields in Imperial Valley during each year during September and October. Exotic *Eretmocerus* were detected in 10 of the 23 fields (i.e., 43%) sampled in the fall of 1998, 31 of 42 fields (i.e., 74%) sampled in the fall of 1999 and 23 of 24 (i.e., 96%) sampled in 2000. In those fields where exotic *Eretmocerus* were detected, 4% of the *Eretmocerus* were exotics in 1998 and 21% were exotic in 1999 and 48% were exotic in 2000. Similarly, an increase in *Encarsia sophia* was noted as well from 1998 to 2000. *Encarsia sophia* was detected in only one of 23 cotton fields (i.e., 4%) in 1998. However, *E. sophia* was detected in 27 of 42 cotton fields (64%) in 1999, and 24 of 32 (75%) fields in 2000.

In summary, up to four exotic species of silverleaf whitefly parasitoids are well established in Imperial Valley. Compared to native *Eretmocerus eremicus*, their relative yearlong abundance increased consistently from 1998 through 2000. Evidence in 2001 suggested that the level of activity by exotic parasitoid species may have declined, however, it is likely that this represents a simple year-to-year fluctuation in activity.

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#### Augmentative Biological Control of Silve rleaf Whitefly Infesting Melons with Releases of Exotic *Eretmocerus* Species: a Review of the Progress.

The primary goal of this project was to increase biological control of the silverleaf whitefly in spring melons by rearing and releasing several species of whitefly parasites. Rates of parasitism by native parasites of whitefly (Eretmocerus eremicus & Encarsia spp.) are generally low in the spring melon crop in desert production areas of California and Arizona, which is where whitefly populations first start to rapidly increase leading to high regional populations that go on to infest cotton and alfalfa after the melon harvest. If parasitoids are introduced directly into the crop, they can provide control within the crop as well as lead to lower rates of whitefly migration from melons to cotton and alfalfa, reducing whitefly population densities on these crops. Furthermore, releases into spring melons will lead to higher regional populations of parasitoids, that can then go on to attack whitefly in successive crops. Seven years of field investigations of augmentative biological control strategies of whitefly infesting spring melons have demonstrated that parasitism can be increased and whitefly populations suppressed by making open field releases of exotic *Eretmocerus* spp. in commercial melon field plots ranging from <sup>1</sup>/<sub>2</sub> to 28 ha in size. Releases of Eretmocerus mundus (ex Spain, M92014), E. emiratus (ex United Arab Emirates, M 95104) and E. nr emiratus (ex Ethiopia, M96076) into both non-treated and Admire<sup>tm</sup> (imidacloprid) treated melons have all proven to be about equally effective and increased parasitism greater than releases of the native E. eremicus. Mean rates of parasitism ranged from 25-43%. These rates of parasitism were significantly higher than the 7-11% parasitism by naturally occurring native Eretmocerus observed in no-release plots. Rates of parasitism in individual plots were as high as 85% indicating that high rates of parasitism are possible. Whitefly densities were reduced by as much as 59% in release plots and were significantly lower than whitefly densities in the no-release control plots. In another study, parasitoid release into Admire treated fields was compared to the standard control measure of pyrethroid applications that growers often used to control late season populations of whitefly that develop after the Admire application is no longer effective. The Admire + parasitoid release treatment achieved levels of whitefly control equal to the Admire + pyrethroid treatment and whitefly densities were about half that of the Admire only treatment, suggesting that parasitoid release could be substituted for pyrethroid treatment. Several techniques for releasing parasitoid pupae were tested including placing pupae in the field in paper cups, using melon transplants inoculated with whitefly nymphs and parasitoids, and releasing pupae with tractor driven release systems using gravity release with dry carriers and sprayer technology with liquid adhesive carriers. The two best release techniques were using transplants, which resulted in the highest levels of control with the fewest number of released parasitoids, followed by releases in paper cups. Parasitoid release using the tractor driven release system with the dry carrier was less effective as wasp emergence rates were reduced (range of 43-78%) relative to the transplant and the paper cup techniques (range 85-100%). It appears that the dry carrier (vermiculite) may have caused desiccation of pupae and that the mechanized gravity drop system itself did not affect emergence. Because the tractor delivery system can increase control by providing a more uniform distribution of released parasitoids within a field, lower labor costs, and increases grower acceptance of the use of augmentative biological control, it would be worthwhile to continue to work with these systems to improve emergence rates. The liquid adhesive release systems resulted in very poor emergence rates as *Eretmocerus* spp. pupae are unable to reorient within the whitefly pupa and if the whitefly pupa is stuck to the leaf surface with the dorsal side down, the wasp is unable to emerge. This results in an average reduction in emergence of 50%. The use of this kind of release system is not recommended for *Eretmocerus* spp. though should be explored for release of *Encarsia* spp. pupae as they are able to reorient within the whitefly and emerge normally. Release rate studies showed that a range of releases of 98-198 thousand parasitoid pupae/ha were the most effective in controlling whitefly. Releasing parasitoids at this rate would cost an estimated \$592 -\$1,186/ha, which is about \$100 - \$686/ha greater than melon growers spent on whitefly control during the worst years of the whitefly outbreak. It is possible that improved release techniques and lower rearing costs could further reduce release rates making augmentative parasitoid releases cheaper. In recent years, regional population levels of whitefly are lower, which means that release rates may be further reduced. Finally, in the high value spring melon crop, using augmentative releases may be helpful to growers by eliminating the need to observe cumbersome worker safety regulations and lengthy reentry intervals associated with restricted use pesticide applications. Future research needs include developing economic thresholds for the use of augmentative releases in melons, refining release methods, and the development of predictive population models to determine the best timing and frequency of parasitoid release. Some additional benefits of this program included: the release of more than 60 million exotic species of whitefly parasitoids into the Imperial valley, which helped establish several new species; the transfer of rearing technology and new more effective whitefly parasitoids to the beneficial insect industry; and cooperation with industry to improve the use of mechanized beneficial insect delivery systems.

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Research & Implementation Area: Section D. Natural Enemy Ecology and Biological Control

#### Dates Covered by the Report: Jan. 1-Dec. 31, 2000

#### Microhymenopteran Insect Species as Biocontrol Agents for Silverleaf Whitefly: Differentiation by Use of Hybridization Probes Directed at Highly Repeated Satellite DNA Sequences.

Highly repetitive satellite DNAs have been employed as hybridization targets for identification of various species of insects. Previously, we have described sequences from selected species of *Encarsia* and *Eretmocerus*, two genera that are being used in the biological control of whiteflies in greenhouse and field environments. The present communication describes efforts to extend the array of probes available for differentiation of various *Encarsia* and *Eretmocerus* species and provides additional information on the dis tribution and properties of the selected sequences.

The development of these probe sequences entails a multistep process: (1) isolation of genomic DNA from the parasitoid wasp species under study, (2) preparation of a library of cloned DNA restriction fragments in a suitable vector, (3) selection of a subset of the library representing highly repeated sequences by probing with radiolabeled genomic DNA from the insect under study, and (4) examination of the distribution of the subset of cloned repetitive sequences by probing with radiolabeled genomic DNA from the insect genomic DNA preparations from other parasitoid wasp species. With several wasp species, clones of unique repeated sequences were recovered. In contrast, some wasp species only yielded cloned sequences that were present in other species as well. Fortunately, the copy numbers of some of these shared repeated sequences vary substantially from one species to another, and many of these sequences are also of use in differentiation of parasitoid wasp species.

Our efforts have been directed at five *Eretmocerus* species (*E. hayati*, Pakistan; *E. emiratus*, United Arab Emirates; *E. mundus*, Spain; *E.* sp., Ethiopia; and *E. eremicus*, USA) and three *Encarsia* species (*E. formosa*, Egypt; *E. sophia*, Thailand; and *E. pergandiella*, USA). These organisms represent the major species that are indigenous or have been released in whitefly biocontrol efforts in the Southwestern United States. Species-specific probes were obtained for *E. eremicus*, *E. formosa*, and *E. sophia*. These sequences have repeat sizes of 143, 33, and 120 bp, respectively, and they comprise approximately 2%, 7%, and 1% of the wasp genome, respectively. A cloned 260-bp sequence from *E. hayati* comprises 1.5% of the genome in this organism; this sequence cross reacts at a very low level (3%) only with *E. emiratus*. A 172-bp sequence from *E. mundus* has applications as a non-native selective probe. No hybridization signal was displayed by *E. eremicus*, but all other *Eretmocerus* species gave strong signals. This sequence, which represents 2.5% of the *E. mundus* genome, was also the most common clone recovered from the *E. sp.* (Ethiopian) library. The latter library also yielded a clone of a 185-bp moderately repeated element (0.15% of genome) that hybridized strongly with *E. sp.* (Ethiopian) and *E. emiratus*, weakly (6% relative signal) with *E. mundus* and *E. hayati*, and not at all with *E. pergandiella*. Hybridization of insect squash blots with one or more of these probes allowed differentiation of all of the tested parasitoids.

Some of these repeated sequences have been tested for their utility as fluorescent, nonradioactive probes of squash blots of parasitoid wasps. In general, oligonucleotide sequences bearing a single fluorescent tag at the 5'-terminus did not produce sufficient signal to allow detection of the cognate wasp species, but larger polynucleotide structures containing many reporter groups appear to provide signals that permit detection of the parasitoids in insect squash blots.

#### Section D: Natural Enemy Ecology and Biological Control

#### Compiled by W. J. Roltsch and G. S. Simmons

Biological control continued to play a prominent role in the Silverleaf Whitefly National Research, Action, and Technology Transfer Plan and was a primary component in work on basic biology, population ecology, host-plant interactions, chemical control technology, and the development of integrated pest management programs. Progress on biological control of *Bemisia* over the last decade has been recently reviewed (Faria and Wraight, 2001. Crop Protection 20, 767-778; Gerling et al., 2001. Crop Protection 20, 779-799; and Naranjo, 2001. Crop Protection 20, 835-852).This research summary covers several specific topics including: classical biological control, augmentation, conservation, natural enemy biology and technology transfer.

Biological control is reliant on sound systematics and the identification of natural enemies and their whitefly host. This topic of key importance is briefly reviewed.

Foreign Exploration and Classical Biological Control.

The foreign exploration for new biological control agents was completed by 1997. Over 56 populations of Eretmocerus and Encarsia parasitoids were collected worldwide and cultured in quarantine, in addition to several predator species. Most accessions underwent laboratory and field evaluation. This was followed by the mass-rearing and release of the most promising species and strains. Mass-rearing was conducted in Texas, California, and Arizona through cooperative work plans with Federal, State, County, Universities, and private industry. Because the silverleaf whitefly cycles among numerous short-lived annual crops and urban environments in the harsh arid climates of the southwest United States, an emphasis was placed on making large inoculative releases to increase the probability of establishment of new species. Several release strategies were used including: urban releases, field releases, and refuges near commercial fields. In addition, the establishment effort was aided by extensive releases made in agriculture field plots as part of the evaluation of augmentative biological control using the most promising exotic species.

In California, the silverleaf whitefly severely affected the desert region of Imperial Valley and to a lesser extent the San Joaquin Valley. By 2000, collections from field samples showed that several exotic species were well established. *Eretmocerus* sp. nr. *emiratus* (*ex* Ethiopia) was the dominant exotic species present. A late season survey in cotton indicated that nearly half of the *Eretmocerus* in commercial cotton fields were *E*. sp. nr *emiratus* or *E. emiratus* (*ex* United Arab Emirates). Small numbers of *Eretmocerus mundus* were also collected.

These samples were composed of approximately 50% native *Eretmocerus eremicus*. The exotic species *Encarsia sophia* was also found to be present in most of the cotton fields. During 2001, sampling efforts across all cropping systems, including a multivariate valley-wide survey effort, suggested that exotic parasitoid activity was less than in 2000. It is unlikely that sufficient time has passed for exotic and native species to achieve a state of equilibrium and year-to-year fluctuations are expected. In the San Joaquin Valley, *Eretmocerus mundus* is the predominant exotic parasitoid established and has been collected from common weeds near citrus and in adjacent cotton fields.

In the lower Rio Grande Valley (LRGV) of Texas, field releases of exotic parasitoids ended in 1998. Parasitoid establishment has been monitored by collecting field samples from numerous host plant species at different releases sites, from various host plant locations and by using a sentinel plant technique. Results from 2000 indicate that *Eretmocerus hayati* has become the primary exotic parasitoid established in the LRGV representing about 95% of the parasitoid species recovered on weed species and commercial crops. *Eretmocerus mundus* and *E.* sp. nr. *emiratus* represented 3% and 0.6% respectively of the species composition of collected material.

Surveys from 2000 in Arizona showed that, *E*. sp. nr. *emiratus* is the most common exotic species. A survey of *Bemisia* parasitoids in Florida, the Caribbean, and Central and South America was conducted. Evaluation of parasitoid species composition in northern Florida has recently begun.

#### Augmentation

Continuing progress has been made on the development of pathogens for whitefly control in both greenhouse and field crops and was recently reviewed (Faria and Wraight, 2001. Crop Protection 20, 767-778). Advances in production, formulation, and application technology have improved the efficacy of fungal pathogens from strains of Verticillium lecanii, and led to the development and registration of several new products based on strains of Beauveria bassiana and Paecilomyces fumosoroseus. Efficacy of fungal pathogens has been improved by making applications early (before whitefly populations increase), targeting specific stages of *Bemisia* most susceptible to fungal infection, making applications when environmental conditions are favorable to spore survival and growth, and by identifying and avoiding the use of incompatible fungicides. Economic advantages of using conventional insecticides limits wider adaptation of these materials, but continued problems with chemical insecticide resistance, increased regulation of broad spectrum pesticides, and continuing consumer interest in pesticide-free foods should increase the use and acceptance of these materials. A recently discovered entomopathogenic virus was isolated from silverleaf

whitefly and has been identified as an iridovirus. Bioassay systems for this virus have been developed and investigations on its potential for use as an insecticide are underway.

Several studies on augmentative releases of *Eretmocerus* spp. in melon crops in California, Arizona, and Texas have shown that parasitoid releases can increase parasitism and decrease whitefly densities. Releases of Eretmocerus spp. using melon transplants inoculated with parasitoid pupae lowered release rates and achieved increases in rates of parasitism and decreases in whitefly density similar to releases by other methods requiring higher release rates. The use of tractor driven mechanized release systems with dry and liquid carriers were explored to learn if more uniform and efficacious control could be achieved by providing more uniform release of parasitoid pupae within the melon crop. Emergence rates were lower relative to other release methods. Because of the potential for better control with more even distributions of released parasitoids, lower labor costs, and increased grower acceptance, continued research is recommended to improve the use of these systems.

Further research on augmentative release strategies is needed for refinement of release techniques, to develop economic thresholds for the use of augmentative biological control in melons, and to develop predictive population models to determine the best timing and frequency of parasitoid releases.

#### Conservation

Several biorational insecticides including insect growth regulators (IGRs) and juvenile hormone analogs (JH analogs) have been integrated into the pest management of cotton production and vegetable crops. These selective materials are considered to have less impact on natural enemies (predators and parasitoids) of silverleaf whitefly and other crop pests than conventional broad-spectrum insecticides. Natural enemy data from field tests with IGRs indicate that differences in predator numbers in treated and untreated plots were low to moderate for a range of species. Reductions in treated plots typically ranged from 15-35 %. However, reduction in population densities as great as 80% of the ladybug species Hippodamia convergens was observed. From these data, it could not be determined whether reductions in ladybug densities were a direct effect of the treatment or an indirect effect of reduced host availability. Laboratory studies have shown that certain IGRs and JH analog products have negligible to low impact on survivorship and reproductive potential of several common predators (Geocoris punctipes, Orius insidiosus and Collops vittatus).

The entomopathogenic fungi *Verticillium leacanii*, *Beauveria bassiana* and *Paecilomyces* are effective against whiteflies in many greenhouse and field crops. Though non-target effects on some insect natural enemy species have been noted, the use of fungi should improve the conservation of biological control agents in whitefly affected crops (see Faria and Wraight, 2001. Crop Protection 20, 767-778).

Refuges have been used very effectively to provide a good environment for releasing and establishing exotic parasitoid species during the summer and fall period. This included the use of basil and okra (both annual plant species) that were inoculated in early summer with exotic parasitoids and grown into late fall as a parasitoid nursery site for the specific purpose of achieving regional establishment of new species. In contrast, utilizing refuge plantings consisting of annual species in a site-specific pest management mode in Imperial Valley failed. That is, unsatisfactory parasitoid to whitefly ratios occurred when planting a sequence of annual plant species (collard, sunflower, basil and okra) as a yearlong habitat to provide a continual source of parasitoids (native or exotic) to control whitefly in adjacent commercial crops. It is possible that refuge systems composed of perennial plant species could fill such a need. Silverleaf whitefly perennial host plants that have been identified as potentially valuable refuge plants, host low to moderate densities of whitefly for extended periods; whereas several annual plant species (esp. collard) commonly exhibit extreme fluctuations in silverleaf whitefly densities. This needs further investigation.

#### Natural Enemy Biology

Several studies on dispersal and flight characteristics of both native and exotic species of *Eretmocerus* were conducted in laboratory flight chambers and in open field releases in melons and cotton. In flight chamber studies, flight duration of the native *Eretmocerus eremicus* was affected by mating status with both unmated females and males flying longer than when mated. In field releases in cotton and melons, most *E. eremicus* were shown to move only a few meters from the release point, though some movements as far as 82 m were observed. Field releases of exotic E. emiratus in cotton and into surrounding okra and melon plots showed that the majority of recaptures of this species were made within a few meters of the release point. A pattern of more active flight activity in the morning hours was noted as well as sex-based differences in recapture rates, with more males recaptured than females. Information from these studies should be useful for designing more effective augmentative release strategies. Studies on long distance movement of parasitoids and predators, which may be dependent on specific conditions or time of the year, are still needed to better understand establishment processes of new species. Such information could be used to better implement classical as well as augmentative and conservation biological control efforts.

Several years of detailed life-table studies in both treated and untreated cotton has shown that mortality by conserved natural enemies can be as high as 40%. This information has led to the development of improved IPM for whitefly in cotton.

Field cage studies have provided little evidence that interspecific interactions among parasitoids can significantly disrupt the suppression of whiteflies. Silverleaf whitefly suppression was not reduced (from the highest level of suppression observed by any one parasitoid) when *Eretmocerus eremicus* and *Encarsia sophia* were tested separately or in combination. When *Eretmocerus mundus*, *Encarsia formosa* and *Encarsia pergandiella* were evaluated in all combinations, some reduction in suppression was observed in only one of the combinations that included the two *Encarsia* species. When all three were present, no reduction in suppression was observed.

Studies on parasitoid marking pheromones have been conducted for *Eretmocerus* and *Encarsia* species. This information may play a role in understanding the fundamental differences among species or species group performance at the population level. Research has also uncovered an iridovirus that affects silverleaf whitefly. This is a significant discovery as little is known about viruses of sucking insects.

The effects of leaf surface characteristics (trichomes, exudates etc.) on host plant attractiveness to parasitoids, and parasitoid searching efficiency can be considerable. *Eretmocerus* tenure on melon leaves (having a high density of trichomes) is far less than on cotton (having a considerably smoother leaf surface). Furthermore, trichome exudates of certain plants have the potential of entrapping searching parasitoids and inflicting a considerable degree of mortality. Such studies demonstrate the wide range of influences that silverleaf whitefly host plants may have on parasitoid performance.

#### **Systematics and Species Identification**

The ability to identify natural enemy species and populations is imperative to prevent costly duplication and misdirected work. Because of small size and remarkably little variation in morphology among species, the aphelinid Hymenoptera are difficult to identify. Several morphological keys were developed for native and recently introduced species of *Eretmocerus* and *Encarsia*. Because morphological identification of some species is problematic, and body structures with subtle character differences can be easily destroyed, the development of genetic diagnostic techniques has been invaluable. RAPD-PCR and DNA probe technology have been developed for identifying many of the Eretmocerus and Encarsia species. RAPD-PCR was used for identifying field specimens and testing the purity of cultures. The more recent development of satellite DNA probes allow for the identification of large numbers of field collected immature whitefly specimens.

#### **Technology Transfer**

Exotic parasitoid species and rearing technology were transferred to several commercial insectaries. Cooperative research with the beneficial insect industry to develop the use of mechanized parasitoid delivery systems for melon crops was conducted. Life-table studies along with toxicology studies on selective insecticides led to the development of more effective IPM in the cotton system in Arizona and elsewhere. Biochemical and molecular studies were developed to identify new and native species of whitefly parasitoids. An artificial diet for lacewings was developed and transferred to industry. The use of the new diet dramatically decreased costs of producing lacewings and made the use of lacewings to control whitefly in greenhouse crops more feasible. The new diet should also allow research on lacewing releases in field crops.

## Table D. Natural Enemy Ecology and Biocontrol.

Research Approaches <sup>a</sup>	Year 1 Goals Statement	Progress Achieved Yes No	Significance
Natural control and conservation:			
Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	Conduct life table analyses of indigenous and introduced natural enemies to identify key mortality factors of natural enemy populations.	х	New insect growth regulators tested well under field conditions, and reduced loss of natural enemies. A Life Table analysis was conducted on natural enemies in cotton.
Evaluate potential of alternate plants to act as in-field refuges or insectaries for natural enemies.	Identify potential plants for natural enemy population development and assess risks of these plants to foster additional pest problems.	Х	Combinations of annuals and some perennials show promise as within field natural enemy refugia. They are attractive to parasites but support low numbers of whiteflies. Annuals served as outdoor insectaries when releasing exotic parasitoids.
Assess cues used by natural enemies to locate whitefly and to identify potential methods for enhancing natural enemy activity.	Conduct laboratory tests to identify cues used by natural enemies to locate and attack whitefly.	Х	Some research has been initiated but was not reported at this meeting.
Augmentation of natural enemies:			
Develop natural enemy mass- rearing systems.	Identify natural enemies with the highest potential for controlling whitefly in key cropping systems.	Х	Diets are being developed for generalist predators. Improvements have been made in rearing parasitoids, increasing rearing efficiency. Field studies have identified promising candidates for augmentative releases
Develop release technologies to maximize the effectiveness of mass- reared natural enemies in the field.	Identify natural enemies with the highest potential for controlling whitefly in key cropping systems and that may be economically mass produced.	Х	A novel "Banker Plant" field release strategy shows promise for increasing efficacy of releases. Releases of <i>Eretmocerus</i> into greenhouses controlled <i>Bemisia</i> attacking poinsettias when done at low pest densites.

Table D. Natural Enemy Ecology and	Biocontrol. (continued)	Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 1 Goals Statement	Yes		Significance
Evaluate augmentative parasitoid, predator, or pathogen releases.	Initiate studies on natural enemy augmentation with identified high potential natural enemies.	Х		Augmentative releases of parasitoids controlled <i>Bemisia</i> in large demonstration fields. These releases can be integrated with conventional pest management practices
Importation biological control:				
Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Identify sites suitable for the release and subsequent evaluation of each candidate natural enemy. Conduct inoculative releases of natural enemies.	Х		Combinations of annual plants that make excellent insectaries and can be farmed under local climatic conditions have been identified. Homeowners are being recruited to care for plants used for making releases
Systematics, ecology, and population dynamics of natural enemies: <sup>b</sup>				
Clarify sytematics of predators, parasitoids and pathogens.	Conduct taxonomic studies of species within targeted releases sites. Verify taxonomic purity of mass-reared natural enemies. Complete taxonomic work on poorly characterized but important groups. Assist in determining most suitable natural enemies for release through biogeographical analysis.	Х		Taxonomic studies have been completed on the exotic <i>Eretmocerus</i> and a key to their identification is in press. PCR techniques have been developed to identify the purity of cultures and aid in identification of recovered parasites.
Determine <i>Bemisia</i> - natural enemy- host plant (Tritrophic) interactions.	Initiate studies to identify mechanisms involved in <i>Bemisia</i> - and natural enemy plant attraction.	Х		Controlled laboratory studies showed that <i>Bemisia</i> and aphelind oviposition rates varied depending on host plant.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	Assess the value of the <i>Bemisia</i> biological control research to evaluate key issues to the science of biological control.	Х		The role of autoparasitism in native populations of <i>Encarsia</i> and its impact on native <i>Eretmocerus</i> has been evaluated. Results from one study show no adverse affect of <i>Encarsia</i> on overall parasitism of SLWF

# Table D. Natural Enemy Ecology and Biocontrol. (continued)

<sup>a</sup> See Table C for complementary research. <sup>b</sup> See Table A for complementary research.

Research Approaches <sup>a</sup> Natural control and conservation:	Year 2 Goals Statement	Progress Achieved Yes No	Significance
Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	Evaluate predator gut contents. Conduct life table analysis.	х	Role of predators in cotton identified; importance of narrow spectrum insecticides highlighted.
Evaluate potential of alternate plants to act as in-field refuges or insectaries for natural enemies.	Determine refugia plant phenology in relation to cultivated crop phenology.	Х	Perennial plants capable of growing in Imperial Valley identified, selected for a pilot project at a commercial organic farm.
Assess cues used by natural enemies to locate whitefly and to identify potential methods for enhancing natural enemy activity.	Determine potential methods for manipulating cues as part of a whitefly management program.	Х	No work reported.
Augmentation of natural enemies:			
Develop natural enemy mass- rearing systems.	Determine nutritional, physiological, and ecological requirements for mass- rearing.	Х	Whitefly, parasitized by <i>Encarsia</i> , were grown on an artificial diet long enough for parasitoids to emerge as adults. First such report. Potential for research and commercial rearing.
Develop release technologies to maximize the effectiveness of mass- reared natural enemies in the field.	Evaluate the fate of natural enemy life stages under field conditions to identify the appropriate developmental stage to be released.	Х	First year results show banker plants may prove more efficacious than releases of parasitoids by hand. Two species of coccinellids evaluated, compared for greenhouse use.
Evaluate augmentative parasitoid, predator, or pathogen releases.	Conduct releases on selected crop systems at various rates of release.	Х	Impact of <i>Beauveria bassiana</i> on generalist predators determined. Parasitoid dispersal was determined using new protein marking technique

Research Approaches <sup>a</sup>	Year 2 Goals Statement	Progress Achieved Yes No	Significance
Importation biological control:			U
Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Evaluate establishment of exotic natural enemies within target release area. Determine if additional releases are necessary.	Х	Several new exotics have persisted over several years and are multiplying and spreading in Texas and California.
Systematics, ecology, and population dynamics of natural enemies: <sup>b</sup>			
Clarify sytematics of predators, parasitoids and pathogens.	Provide taxonomic support for importation and mass-rearing programs. Publish keys to assist in species identifications.	Х	Key on exotic <i>Eretmocerus</i> published. Program developed for curating, cataloging recovered parasitoids.
Determine <i>Bemisia</i> - natural enemy- host plant (Tritrophic) interactions.	Study plant characteristics mediating whitefly and natural enemy population densities.	Х	Parasitoid foraging, oviposition varied in response to different plants (crops) and host whitefly. Plants varied in color, architecture, and semiochemicals.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	In conjunction with field evaluations, validate predictions made by behavioral and population models important to biological control.	Х	No interference competition measured, with respect to whitefly control, when mixing primary parasitoids and autoparasitoids.

# Table D. Natural Enemy Ecology and Biological Control. (continued)

<sup>a</sup> See Table C for complementary research. <sup>b</sup> See Table A for complementary research.

		Progress A		o: :c
Research Approaches <sup>a</sup> Natural control and conservation:	Year 3 Goals Statement	Yes	No	Significance
Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	Conduct manipulative experiments to evaluate the impact of each natural enemy mortality agent on whitefly suppression.	х		Life history tables have been constructed comparing mortality factors of natural enemies in conventional vs IGR treated cotton. Functional response data available for several parasitoid species.
Evaluate potential of alternate plants to act as in-field refuges or insectaries for natural enemies.	Conduct field tests to assess whether refuges act of natural enemy and whitefly sinks or sources to adjacent cropping systems.	Х		Research in the Imperial Valley has shown that perennial refuges support large numbers of whitefly and parasitoids that migrate to adjacent systems.
Assess cues used by natural enemies to locate whitefly and to identify potential methods for enhancing natural enemy activity.	Conduct small scale trials to enhance whitefly suppression by manipulating natural enemy location and attack of whitefly.		Х	
Augmentation of natural enemies:				
Develop natural enemy mass- rearing systems.	Develop rearing systems on selected hosts and on artificial diets. Determine economic feasibility of the procedure.	Х		Mass rearing methods on SLWF has been accomplished. Artificial diets are still being researched, with economics undetermined.
Develop release technologies to maximize the effectiveness of mass- reared natural enemies in the field.	Develop necessary technology for release of the appropriate natural enemy life stage.	Х		Several release technologies have or are being tested. Banker plant technology appears to be very effective. Capsule delivery methods being tested. Cold storage of parasitoid pupae also being tested
Evaluate augmentative parasitoid, predator, or pathogen releases.	Identify optimal release strategies for key cropping systems.	Х		Parasitoid release rates have been determined for major crops. Strategies for releasing/integrating parasitoid and predator in greenhouse crops have been determined. Significant information currently available on application of fungal pathogens in

# Table D. Natural Enemy Ecology and Biological control.

various crops.

Research Approaches <sup>a</sup>	Year 3 Goals Statement	Progress Achieved Yes No	Significance
Importation biological control:			
Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Assess spread of established natural enemies and their ability to suppress whitefly populations	Х	Whitefly suppression by exotic parasitoids determined by multiple researchers. Preliminary data suggests significant spread of established exotic parasitoids in some regions. Dispersal rates of natural enemies still under study.
Systematics, ecology, and population dynamics of natural enemies: <sup>b</sup>			
Clarify sytematics of predators, parasitoids and pathogens.	Provide taxonomic support for importation and mass-rearing programs.	Х	Several taxonomic keys developed for imported parasitoid species. RAPD-PCR techniques proven as quick identification method. Preliminary Satellite DNA techniques proven, however, still under development.
Determine <i>Bemisia</i> - natural enemy- host plant (Tritrophic) interactions.	Study compatibility of characteristics of plant traits mediating whitefly populations with the abilities of natural enemies to suppress whitefly populations.	Х	Tri-trophic interactions determined for <i>B. bassiana</i> / SLWF / tomato. Some research completed on parasitoid / host / plant interactions.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	Assess deviations between theoretical predictions and field data.	Х	Some life history data collected on parasitoid and predator populations in cotton. BioControl-Parasite simulation model available for testing / validation. Some Laboratory data available for testing theoretical predictions of field level performance.

# Table D. Natural Enemy Ecology and Biological Control. (continued)

<sup>a</sup> See Table C for complementary research. <sup>b</sup> See Table A for complementary research.

		Progress A		
Research Approaches <sup>a</sup>	Year 4/5 Goals Statement	Yes	No	Significance
Natural control and conservation: Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	Conduct a feasibility study and economic assessment of altered crop management practices that may enhance the impact of indigenous natural enemies.	Х		IGR's compared to conventional insecticides reduce negative impacts on natural enemies. Pathogenic fungi have been shown to be of use. Annual refuge plantings are of limited value, however, perennial plant refuges are promising as a source of parasitoids while providing minimal source populations of SLW.
	Develop and evaluate area wide programs to facilitate full implementation.	Х		Insect growth regulators have been integrated into pest management programs .
Evaluate potential of alternate plants to act as in-field refuges or insectaries for natural enemies.	Conduct field tests to evaluate spacing of refuges necessary to achieve satisfactory whitefly suppression.	Х		Spacing studies have not been conducted, however, other field studies indirectly provide information pertaining to this topic, including several parasitoid movement studies.
	Conduct a feasibility study and economic assessment of alternate plantings in terms of an entire crop management program.		Х	
Assess cues used by natural enemies to locate whitefly and to identify potential methods for enhancing natural enemy activity.	Conduct large scale field trials and evaluate product development for commercial investment as necessary.		Х	No, however, recent work has been done on chemically-based intraspecific communication cues between parasitoid individuals.
	Transfer technology (as needed) to commercial interests for full implementation.		Х	
Augmentation of natural enemies: Develop natural enemy mass- rearing systems.	Evaluate rearing system effects on natural enemy life history characteristics, behavior, and ability to suppress whitefly populations.	х		Parasitoid development on <i>Bemisia</i> produced on artificial diet has been examined. Predator fitness when produced on artificial diets has been studied. Additional work in this area, including large scale production using plant reared <i>Bemisia</i> to rear parasitoids is in need of further study.
	Facilitate transfer of mass-rearing technology to commercial interests as necessary.	Х		Contracts were developed with private industry for using new parasitoid species. Artificial diets for several predator spp. are available.

# Table D. Natural Enemy Ecology and Biological control, 2001-2002.

		Progress A		
Research Approaches <sup>a</sup>	Year 4/5 Goals Statement	Yes	No	Significance
Develop release technologies to maximize the effectiveness of mass- reared natural enemies in the field.	Evaluate re lease technology effects on natural enemy life history characteristics, behavior, and ability to suppress whitefly populations.	Х		Tractor pulled, parasitoid pupal drop box release method needs further refinement. Technique developed using "Banker plants" with parasitoid pupae, to inoculate crops. Several container based release methods for pupae were evaluated and the best methods identified.
	Facilitate transfer of mass-rearing technology to commercial interests as necessary.	Х		Contractual agreements made to provide commercial insectaries with several promising parasitoid species for field and greenhouse augmentation.
Evaluate augmentative parasitoid, predator, or pathogen releases.	Continue evaluation of releases, determine need for additional releases. Compare results in different cropping systems and environments.	Х		Evaluations conducted in cotton, cantaloupe and greenhouse crops. Use in greenhouse environment is particularly promising.
	Analyze information and make recommendation regarding need for expansion of the approach.	Х		Studies in greenhouse crops completed and analyzed. Recommendations have been developed. Extensive evaluation of augmentative biocontrol in spring cantaloupe fields using parasitoids has occurred. Analysis nearly completed.
Importation biological control: Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Continue to assess the spread of established natural enemies and their ability to suppress whitefly populations. Evaluate program progress and determine if additional strategies are necessary	х		Extensive assessment conducted in CA., TX, AZ, & FL. Long term field plots, sentinel plants, area wide field surveys, and life table methods used. One to three parasitoid species established in CA, TX and AZ. Establishment process is ongoing, probably taking several more years. Results to date look promising in several regions.
	Complete program analysis. Publish program assessment and conduct an economic assessment.	Х		Book in preparation, K. Hoelmer and J. Gould editors.

# Table D. Natural Enemy Ecology and Biological Control, 2001-2002. (continued)

		Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 4/5 Goals Statement	Yes	No	Significance
Systematics, ecology, and population	dynamics of natural enemies: <sup>b</sup>			
Clarify systematics of predators, parasitoids and pathogens.	Provide taxonomic support for importation and mass-rearing programs.	Х		Numerous publications developed for <i>Eretmocerus</i> and <i>Encarsia</i> spp. identification. Extensive development and availability of molecular based identification methodology.
Determine <i>Bemisia</i> - natural enemy- host plant (Tritrophic) interactions.	Assess the implementability of favorable tritrophic interactions within the context of an whitefly management program.	Х		Relationships among leaf surface characteristics, SLW and parasitoid performance identified in several instances. Associations identified between plant species and parasitoid activity.
	Implement and evaluate large scale crop management programs for suppression of whitefly populations.	Х		Crop vulnerability windows identified.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	Evaluate behavioral or population level parameters that may explain observed deviations	Х		Life table analysis, laboratory and field cage studies of interspecific parasitoid interactions, field cage and in- field sex ratio studies, and studies of parasitoid indosymbionts were conducted.
	Quantify the impact of basic research on the development of feasible biological control programs for <i>Bemisia</i> and the advancement of the field as a science.	Х		Artificial diets developed for mass rearing several predators. Although an artificial diet for SLW is unlikely to support mass rearing of parasitoids, its development has been very useful for conducting detailed biological studies. Parasitoid movement studies have provided a basis for developing augmentative release approaches and interpreting exotic parasitoid establishment data.

# Table D. Natural Enemy Ecology and Biological Control, 2001-2002. (continued)

<sup>a</sup> See Table C for complementary research.
 <sup>b</sup> See Table A for complementary research.

## **Reports of Research Progress**

# Section E: Host-Plant Resistance, Physiological Disorders, and Host-Plant Interactions

Co-Chairs: Greg Walker and Cindy McKenzie

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

# Dates Covered by the Report: 2001

#### Parasitism of Bemisia tabaci on Numerous Species of Host Plants

The influence of numerous vegetable and other agronomic plant species on incidence of parasitism of the B-biotype sweetpotato whitefly, Bemisia tabaci (Gennadius), by augmentation with parasitoids was determined in field plots. Tests were conducted on 16 taxonomically diversified plant species (Beta vulgaris L., Brassica oleracea var botrytis L., Brassica oleracea var capitata L., Citrullus lanatus (Thunberg) Matsum. & Nakai ssp. lanatus., Cucumis melo L., Cucumis sativus L., Glycine max (L.) Merrill, Gossypium barbadense L., Helianthus annus L., Ipomea batatas L., Lantana camara L., Lycopersicum esculentum Miller, Phaseols vulgaris L., Solanum melongena L., Solanum tuberosum L., and Vigna sinensis L.). Parasitism in plots with feral infestations by B. tabaci was evaluated through augmentations with Eretmocerus mundus Mercet from a laboratory colony, and comparisons were made with check plots in which no parasitoid releases were made. Plots were set up at five locations in Egypt (Beihera, Beni-Suef, Kafr ElShikh, Minufiya, and Qalyubiya). Each plot (0.13 hectares) contained a single plant species. Adult *E. mundus* were released during each of 15 weeks in treatment plots, and parasitism data were collected weekly over 15 weeks. The release rate was 5-12 parasitoids per plant and the releases were done using vials of parasitoids that were attached to the plants. Parasitism was enhanced in all plots where augmentations were made. In some plots, e.g., both species of Brassica, B. vulgaris, and G. max, overall parasitism was enhanced at a relatively high rate while in other plots, e.g., C. lanatus, S. melongena, and L. esculentum, the enhancement of overall parasitism was relatively low. In both treated and untreated plots for all plant species, parasitism peaked 7-12 weeks after the first augmentation date. Results from this study describe the relative seasonal abundance and relative degree of augmentative enhancement of parasitism of Bemisia among numerous plant species of economic importance.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

# Dates Covered by the Report: Jan. - Dec. 2000

## Effect of Bemisia argentifolii and Squash Silverleaf Disorder on Zucchini Plant Growth and Yield

Squash silverleaf disorder is a serious systemic physiological disorder of *Cucurbita* spp. Previous studies have shown that large whitefly populations can reduce plant vigor, growth and yield. However, we had no idea of the Bemisia argentifolii infestation levels that would cause damage to zucchini plants. Heavy whitefly feeding and expression of squash silverleaf disorder are two factors that may cause zucchini plants to become stunted, weak and lose yield. Two closely-related zucchini breeding lines were selected to separate these two factors; ZUC61 is silverleaf susceptible, whereas ZUC76-SLR is silverleaf resistant. Experiments to test the effects of whitefly infestation levels on zucchini seedling development and mature plant fruit yield were conducted in the greenhouse and outdoor screened rooms, respectively, in the spring and fall of 2000. In the greenhouse experiments, zucchini seedlings were infested with 0, 15, 30 or 60 pairs of B. argentifolii adults at the 2leaf stage in the spring (Jan. 28 – Mar. 20), and with 0, 30, 60 or 90 pairs in the fall (Aug. 24 – Oct. 6). Whiteflies were confined on plants and allowed to feed and oviposit for 72 h. Whitefly eggs remained on plants to develop to nymphs. All whitefly-infested leaves, usually the 3-4 oldest ones, were removed immediately before whitefly adults emerged. Plant dry weight and height, and petiole length of silvered leaves were measured to evaluate zucchini plant growth 14 d after whitefly infested leaves were removed. Our results indicated that the progeny of 60 pairs of whiteflies significantly stunted ZUC61 and ZUC76-SLR zucchini seedling growth in the spring (13% dry weight reduction), whereas as few as 30 pairs and their progeny did so in the fall (9 % dry weight reduction). Plant height and the petiole length of silvered leaves of ZUC61 and ZUC76-SLR were also shortened after whitefly infestation. The effect of whitefly nymphal feeding continued at least 2 weeks after whitefly-infested leaves were removed. The silverleaf severity of ZUC61 did not differ significantly among the three treatment infestation levels in both spring and fall experiments. However, significant dry weight reduction only appeared at the highest infestation level 60 in the spring experiment. In the fall experiment, significant plant dry weight reduction appeared at all treatment infestation levels. Plant dry weight was reduced more at infestation level 90 (16%) than at infestation level 30 (9%). Although silverleaf symptoms were nonexistent or slight (grade 1) on ZUC76-SLR leaves, this breeding line experienced the same reduction in seedling growth as ZUC61.

In the screen room experiments, we used four whitefly infestation levels of 0, 30, 60 and 120 pairs of adults in both spring (Apr. 8 – June 3) and fall (Aug. 23 – Oct. 24) seasons. Whiteflies were released onto zucchini plants at the 2-leaf stage once a week for 3 weeks. Silverleaf severity of the  $4^{th}$  leaf (length > 8 cm) from the shoot apex and the leaves immediately above and below it was graded once a week until the end of the experiments. Female flowers were pollinated manually. Fruits were harvested at 16-17 cm in length in the spring, and at 96 h after pollination in the fall. The harvest was continued for 4 weeks from the beginning of fruit production in each season. Serious silverleaf symptoms of grade 4-5 were expressed on all whitefly-infested ZUC61 plants, whereas slight silverleaf symptoms of grade 1-3 were expressed on ZUC76-SLR plants. However, the silverleaf severity of each zucchini breeding line did not differ significantly among treatment infestation levels in both spring and fall experiments. Zucchini fruit number was significantly decreased at whitefly infestation level 60 for ZUC61 plants and at infestation level 120 for ZUC76-SLR plants in the spring. Fruits at infestation levels 60 and 120 developed more slowly and took approximately 1 more day to reach the same length as fruits from control plants. Fruit length from plants under these two whitefly infestation levels was shorter than that of control fruits on the 3<sup>rd</sup> d after pollination. However, fruits from all infestation levels weighed almost the same when they reached the same harvestable size. All plants at infestation levels 60 and 120 died toward the end of the experiment in the fall, but no plants died at any whitefly infestation levels in the spring. No fruit was produced on plants at infestation level 120, and only five fruits were harvested from 14 plants at infestation level 60 in the fall. Significant fruit yield reduction appeared at infestation level 30 for both ZUC61 and ZUC76-SLR breeding lines.

Based on these studies, we concluded that the stunting of zucchini seedling growth and yield reduction were mainly induced by the feeding of high whitefly populations, not by expression of silverleaf symptoms.

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Research & Implementation Area: Section E. Host Plant Resistance, Physiological Disorders and Host-plant Interaction

# Dates Covered by the Report: January 2000-2002

# Expression of SLW3 (silverleaf whitefly-induced) Gene in the Transgenic Plants

Two silverleaf white-induced genes, *SLW1* and *SLW3*, were isolated by differential RNA display. The transcripts of *SLW* genes accumulated differentially by silverleaf whitefly (SLWF) and sweetpotato whitefly (SPWF). After SLWF nymph feeding, *SLW1* and *SLW3* RNAs accumulate in the apical, non-infested leaves. In contrast, neither gene was induced by SPWF in apical leaves. Both *SLW* transcripts were accumulated in the local, infested leaves of SLWF-infested plants; however, only *SLW3* was induced in the local leaves of SPWF-infested leaves. The expression of *SLW1* was detected during flower and fruit development, while *SLW3* RNAs were not. The regulation of *SLW1* was involved in the two plant defense signals, jasmonic acid and ethylene, but the defense signals that regulate *SLW3* are still unknown. Both *SLW* genes were induced by the water deficit but not by other stresses such as wounding and pathogen infection. Database searches indicated that *SLW1* and *SLW3* encode a M20B-like peptidase and  $\beta$ -glucosidase, respectively; however their roles in the plant defense response to the SLWF infestation are still not understood. The reverse genetic approach has been used for investigating the expression and function of *SLW* genes in the plant response to SLWF feeding. The *SLW3:GUS* transgenic tomato and *Arabidopsis* have been generated and the *SLW3* expression studies under abiotic and biotic stresses and during development are ongoing. In addition, transgenic plant lines ectopically expression *SLW3* have been constructed.

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Research & Implementation Area: Section E: Host-Plant Resistance, Physiological Disorders, and Host-Plant Interactions

## Dates Covered by the Report: 2000-2001

#### Silverleaf Whitefly Populations and Trichome Densities of Upland Cottons

The study was conducted in 2000 at the University of Arizona Agricultural Research Center, at Maricopa, AZ. Cotton cultivars were five normal-leaf cotton cultivars (Deltapine [DPL] 20B, DPL 50 B, DPL 90B, NuCOTN 33B, and Stoneville [ST] 474) and four okra-leaf cultivars (E0223, E0798, E1028, and Siokra L-23). All entries were smooth leaf cultivars except for the hairy -leaf ST 474. On average, ST 474 had 71.3 stellate trichomes/cm<sup>2</sup> of leaf disk and other cultivars fewer than 1.0 stellate trichomes/cm<sup>2</sup>. ST 474 had 12.9 adults per leaf-turn count and other cultivars had fewer than 6.2 adults. ST 474 had 36.0 eggs/cm<sup>2</sup> and 13.3 nymphs/cm<sup>2</sup> leaf disk. Other cultivars had fewer than 15.4 eggs/cm<sup>2</sup> and 6.0 nymphs/cm<sup>2</sup> leaf disk. On average of all cultivars studied, leaves on the top main stem leaf node (#1) had the highest number of stellate trichomes but the lowest numbers of eggs, nymphs and adults compared with leaves on nodes #2, #3, #4, #5 or #7.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

# Dates Covered by the Report: 2000

# Field Evaluations of the Six Selected Cantaloupe Varieties for Silverleaf Whitefly Resistance

Silverleaf whitefly, Bemisia argentifolii Bellows & Perring, continues to be the most important pest insect for cucurbits, in south Texas. In the past few years, many varieties, lines, and PIs had been studied under field conditions for whitefly resistance in south Texas. It has been reported that the glabrous leafed varieties (lines) are highly resistant to whitefly. Unfortunately, the yield and quality of these smooth leafed varieties or lines are not commercially desirable. Results from the laboratory experiment for the six selected varieties indicated that among the six varieties, Tam Sun, Tam Sun x gl., and Hymark show some degree of resistance to the silverleaf whitefly, whereas other varieties did not show significant differences. The resistance of these varieties to the silverleaf whitefly under the field condition will confirm and verify the results from the laboratory study. The objective of this study was to determine the resistance of six selected varieties to silverleaf whitefly under field conditions to verify the data obtained from the laboratory study. Six varieties and lines were used in this study: Hymark, Tam Sun x gl, Explorer, Tam Sun, Primo, and Perlita. The seedlings (10-11 cm high) were transplanted in the field in middle or late January. Each plot was 30 ft (12 m) long with two separate rows 80 in (2 m) wide, and 10-20 plants each. The plots were arranged in a randomized complete block design with 4 replications. Adult sampling was conducted when whitefly population increase significantly. Thereafter, plants were sampled in 7-day intervals. Adults were counted by leaf turn method. When plants had <6 leaves, adults on the oldest leaf were counted, and when plants had >6leaves, adults on the 4<sup>th</sup> or 5<sup>th</sup> leaf from the proximal were counted. Pupae (re -eyed nymphs), empty pupal cases per 4 leafdiscs per leaf were counted. Yield, sugar content and foliage damage (by sooty molts) were also evaluated.

Whitefly adult population was relatively low on the plants in all 6 varieties before late March, and increased gradually to mid April. Numbers of adults increased rapidly, and reached the peak in late April and early May. Among the six varieties, numbers of adults per lead were significantly different. The variety that had the least numbers of whitefly adults throughout the season was TP45, a Weslaco line, followed by Tam Sun, a TAES variety. The variety that had the greatest number of whitefly adults was Primo, as many as 660 adults per leaf were found on 3 May. Number of adults on Impact and Hymark were also relatively high, with>300 adults per leaf in late April and early May. Significant differences were found for both the adults and nymphs among the six varieties. Primo had the most and TP45 had the fewest whitefly adults, and difference between the two varieties was >8 folds. Primo, Hymark and Impact had more nymphs than other varieties, and again, TP45 had the fewest number of nymphs among the six varieties. Tam Sun was the only variety that has fewer melons than other varieties; whereas the total numbers of melon harvested from other five varieties were not significantly different. Impact and TP45 had more larger-melons compared with other varieties; whereas Tam Sun x gl. and Impact had the most numbers of small melons. TP45 not only had the most numbers of melons, but also had the most large-melons (Table 4). Impact also had relatively greater weight, which, however, was not significantly different from Hymark, Tam Sun x gl., and Primo. Total weight for Tam Sun was the least, although it was not significantly different from Hymark and Tam Sun x gl. Results from this study indicated that Significantly differences in responses to silverleaf whitefly infestation were found among the six cantaloupe varieties. TP45, a Weslaco line, had the fewest whiteflies (adults and red-eyed-nymphs or pupae) compared to other five varieties. TP45 also had the highest yield (more and larger melons). However, melons of TP45 had the lowest sugar contents (3.80-3.86%) compared 7.6-9.5% in other varieties. Other varieties which had fewer silverleaf whiteflies were Tam Sun and Tam Sun x gl. Primo, Impact and Hymark had more silverleaf whitefly than others. However, those varieties had higher yield and sugar contents than others. Similar results were found in the laboratory experiment that Tam Sun, Tam Sun x gl., and Hymark show some degree of resistance to the silverleaf whitefly.

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Affiliation & Location: Dept. of Entomology & Nematology, University of Florida, Gainesville, FL Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions Dates Covered by the Report: 1995-2000

## Mechanisms and Genetics of Resistance to Squash Silverleaf Disorder in Cucurbita spp. Squash

Squash silverleaf disorder (SSL) is a serious physiological disorder of commercial *Cucurbita* species associated with the feeding of nymphal *Bemisia argentifolii*. Screening of the National Plant Germplasm System repository of *C. moschata*, *C. pepo*, and *C. maxima* yielded accessions that showed no SSL symptoms in field trials in Mayaguez, Puerto Rico and Leesburg, FL, where other accessions around them silvered heavily. Field and greenhouse studies of the mechanisms of resistance indicated that although SSL-resistant germplasm did not silver, plants received similar levels of oviposition by whiteflies and supported similar whitefly survival and rates of development as did SSL-susceptible accessions. The mechanism of resistance in two *C. pepo* SSL-resistant lines (ZUC76-SLR and ZUC33-SLR/PMR) and two *C. moschata* lines (UP9606-13-4 and UP9706-3-1) was tolerance to the effects of whitefly feeding. Controlled infestation of SSL-resistant and susceptible germplasm with similar whitefly populations resulted in no silvering in resistant lines and grade 5 SSL symptoms in susceptible lines.

Silvering of leaves begins at the apex of young leaves of *C. pepo* and proceeds basipetally to cover the whole leaf. Chloroplasts are smaller in silvered leaves even before macroscopically visible symptoms develop. Silvering results from an abnormal air space between the upper epidermis and the upper palisade layer of cells. Leaf cross-sections reveal that the development of the silvering occurs as normal air space formation is occurring. Cell separation between epidermis and palisade cells is not a consequence of differential cell division or expansion between the two layers because the number of cells per mm in each layer over time is the same in whitefly-infested plants and controls. Air space formation is more likely a result of increased cell wall degradation during normal air space formation. Leaves of SSL-resistant ZUC76-SLR showed no anatomical response to whitefly feeding but did have lighter green and shorter petioles as did susceptible plants. In grafts of the resistant plants onto susceptible plants challenged with whitefly nymphs, the resistant plants did not develop leaf silvering. Susceptible scions grafted onto resistant rootstocks did exh ibit leaf silvering. These experiments indicate that the site of SSL resistance is at the young developing leaves and not at the mature leaf where insect feeding occurs. Susceptible and resistant *C. pepo* germplasm responded systemically to application of chlormequat chloride on mature leaves or as soil drenches with shortened internodes and silverleaf-like symptoms on new foliage. Petioles and new leaves of ZUC61 and ZUC76-SLR showed similar reductions in chlorophyll content after whitefly feeding on mature leaves.

Greenhouse and outdoor screen room studies indicated that zucchini seedlings were stunted when infested with whiteflies and infested plants yielded less fruit than uninfested plants. To separate the effects of whitefly feeding versus expression of SSL symptoms on growth and yield reduction, two closely related breeding lines were infested with several levels of adult whiteflies; ZUC61 was SSL-susceptible and ZUC76-SLR was resistant (see abstract by Chen et al., section E). Both ZUC61 and ZUC76-SLR seedlings were stunted by feeding of 30-60 pairs of adult whiteflies and their progeny for 2 weeks, showing ca. 10% reduction in dry weight. ZUC61 plants showed grade 5 SSL symptoms at these infestation levels whereas ZUC76-SLR had no or mild (grade 1) symptoms. Season-long feeding by 60-120 pairs of whiteflies and their progeny caused significant yield and quality reduction in fruit for both ZUC61 and ZUC76-SLR. Seedling stunting and yield reduction appear to be more related to intensity of whitefly feeding than to susceptibility to SSL. The genetics of resistance to SSL have been investigated in C. moschata and C. pepo germplasm. In C. moschata, SSL resistance appears to be related to the presence of natural leaf mottling. The recessive gene for silverleaf resistance appears to work only in combination with the lack of natural leaf mottling. Leaf mottling is conditioned by a single partially dominant gene (M), thus only those genotypes with ssmm are resistant to SSL. In C. pepo, SSL resistance appears to be conditioned by two genes, one recessive and one incompletely dominant. In both C. moschata and C. pepo, the severity of SSL symptoms can be modified by whitefly infestation level and environmental conditions, such as light intensity and temperature. I gratefully acknowledge the important contributions of graduate students. Yasmin Cardoza and Jiang Chen, and collaborators, Drs. Judy Schmalstig. Bruce Carle, Linda Wessel-Beaver, and Susan Webb. Research was supported by USDA Special Research Topic grants FLA-ENY-03443 and -03721.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

Dates Covered by Report: March 2000 through December 2000

# **Resistance To Whitefly Transmitted Cotton Leaf Crumple Disease**

Ten cotton, *Gossypium hirsutum* L., cultivars or breeding-lines were evaluated in the field for resistance to the cotton leaf crumple (CLCr) disease caused by (Genus *Begomovirus*, Family *Geminiviridae*) *Cotton leaf crumple virus* (CLCrV) transmitted by silverleaf whitefly, *Bemisia argentifolii* Bellows and Perring. The cultivars Texas 121, AP 4103, AP 6101 and Stoneville 474 and the breeding-lines were NK 2165C, NK 2108SS, NK 2387C, NKX C429-93-2ct, NKX 2907, and NKX 2207 were in Imperial Valley, CA. Cotton entries were rated for severity of CLCr disease symptoms and the presence of CLCrV in leaves of selected plants of each cultivar/breeding-line was determined by dot blot hybridization with a CLCrV DNA probe and PCR analysis with degenerate geminivirus primers. DNA sequencing of geminivirus DNA-A and DNA-B fragments, amplified from symptomatic cotton plants, was used to confirm geminivirus infection and partial characterize CLCrV. Results showed differences in whitefly infestation levels and virus disease symptoms among cotton entries. The cultivar AP 4103 had a higher CLCr disease rating than other entries except AP 6101. The breeding-line NK 2387C, with Cedix parentage, had a lower CLCr disease rating than other entries except Stoneville 474 and NK X2207. There were visible CLCr disease symptoms in Stoneville 474 and NKX 2207, but KN 2387C did not display visible CLCr disease symptoms nor was viral DNA detected in this line.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

Dates Covered by Report: January 2000 through December 2000

# Susceptibility of Upland Cotton Cultivars to Infestation by Silverleaf Whitefly.

Sixteen upland cotton, *Gossypium hirsutum* L., cultivars and experimental breeding-lines were evaluated in the field for susceptibility to silverleaf whitefly, *Bemisia argentifolii* Bellows and Perring, sown at the UC Desert Research & Extension Center, Imperial Valley, CA, into plots of a randomized complete block design experiment replicated four times, and irrigated 24 March 2000. The normal leaf cultivars were DP 20, DP 50, DP 90, DP 5415, DP 5432, DP 5461, DP 5557, HCR 9257, HCR 9240, and Stoneville 474 and the okra-leaf cultivars and experimental breeding-lines were Siokra L23, FiberMax 832, FiberMax 819, E1028, E0798 and E0223. Individual plots measured 14 m in length with 8-beds on 1 m centers or 8m wide. Silverleaf whitefly adults were sampled from ten plants at random in each plot via the leaf tum method using the 5th main stem leaf from the terminal. Silverleaf whitefly nymphs were counted on 1.65 cm<sup>2</sup> leaf disks offrom ten 5th position leaves down from the terminal extracted from randomly selected plants in each plot. Adults and nymphs were sampled on 23 & 29 May, 5, 12, 19, & 26 June, 3, 10, 17, 24 & 31 July, 7, & 14 August, 2000. on 30 June, 7, 14, 20, 28 July, 4, 11, 18, & 25 August. Seed cotton was hand picked from 0.002 acre per plot and yield data were recorded. The okra-leaf entries as a group had fewer silverleaf whitefly adults and nymphs than the normal leaf cotton entries. The okra-leaf experimental breeding-lines had the lowest numbers of silverleaf whitefly adults and nymphs among the okra-leaf entries. Stoneville 474, a hirsute leafed cotton, had the greatest numbers of silverleaf whitefly adults and nymphs among the normal leaf cottons. ?There were no differences in seed cotton yield among the entries, p 0.05, SNK?.

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Research & Implementation Area: Section E: Host-Plant Resistance, Physiological Disorders, and Host Plant Resistance

Dates Covered by Report: May 2000 through September 2001

# High Level of Resistance to Silverleaf Whitefly in the Cotton Relative, Gossypium thurberi

In 1993, F. D. Wilson et al. from USDA's Western Cotton Research Laboratory reported a high degree of resistance to silverleaf whitefly, *Bemisia argentifolii*, in the cotton relative, *Gossypium thurberi*. *G. thurberi* has been successfully crossed with commercial cotton in the past, and therefore could serve as a source of whitefly resis tance genes that could be introduced into commercial cotton. The study presented here was initiated to verify the results of Wilson et al. (1993) and to determine the degree of resistance of *G. thurberi* to silverleaf whitefly.

# **Materials & Methods**

Two field studies (one in 2000, the other in 2001) with a randomized complete block design were conducted at the University of California Desert Research and Extension Center in Holtville, CA. The preliminary test in 2000 had two entries: *G. thurberi* and DP 5415 of Delta and Pine Seeds. The 2001 test had four entries: *G. thurberi*, DP 5415 of Delta and Pine Seeds, Siokra L23 of Cotton Seed Distributors Ltd. of Australia, and Stoneville 474 of Stoneville Pedigree Seed Company. On each sample date, whitefly adults (*Bemisia argentifolii*) were sampled using the leaf turn method. Samples of ten 5<sup>th</sup> node leaves per plot were excised from plants at random on the same dates adult whitefly counts were taken. Whitefly eggs and nymphs were counted on single leaf disks of 1.65 cm<sup>2</sup> taken from the lower left quadrant from each leaf.

In an additional test, greenhouse grown plants were used for experiments comparing nymphal survival and development time of *Bemisia argentifolii* on *G. thurberi* and DP 5415. Plants were young seedlings at the 2-4 leaf stage.

# **Results and Discussion**

The 2000 field experiment demonstrated that *G. thurberi* had a high level of resistance against silverleaf whitefly when compared to DP 5415. *G. thurberi* has okra leaf shape and DP 5415 has normal shaped leaves. Okra leaf shape has sometimes been associated with whitefly resistance; consequently the 2000 field test was unable to determine whether the resistance in *G. thurberi* was due to okra leaf shape or due to a novel factor. Both *G. thurberi* and DP 5415 have smooth leaves, a trait associated with reduced whitefly susceptibility.

The 2001 field experiment included three commercial cotton cultivars including the okra-leaf Siokra L23. *G. thurberi* exhibited a high level of resistance against silverleaf whitefly when compared to all three commercial cultivars, including the okra-leaf Siokra L23. Siokra L23 and DP 5415 also are smooth-leaf varieties while Stoneville 474 has hairy leaves. The resistance of *G. thurberi* relative to the commercial smooth-leaf and okra-leaf cultivars suggests that *G. thurberi* has a novel mechanism of resistance against silverleaf whitefly.

The greenhouse results starkly contrast the field results. Silverleaf whitefly performance, measured as nymphal survivorship and developmental time, was slightly better on *G. thurberi* than on DP 5415. We suspect that the contrasting greenhouse and field study results are due to the use of greenhouse grown seedling plants that had not hardened off. We plan to test this hypothesis in the oncoming season.

Despite the anomaly of the greenhouse experiment, the data strongly suggest that *G. thurberi* has a very high level of resistance against *Bemisia argentifolii* in the field, and that the mechanism of resistance in something not previously known.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

Dates Covered by the Report: January 1, 2001 - December 31, 2001

# Preference and Performance of Silverleaf Whitefly on Selected Poinsettia Cultivars

Studies showed that silverleaf whitefly females clearly showed a preference for ovipositing on the poinsettia cultivar Peterstar. The cultivars Pepride and Red Velvet were the least preferred of 7 cultivars tested. The development of immatures that eclosed from these eggs were consistent with the numbers of eggs laid on the cultivar (good development on Peterstar, and poor development on Pepride and Red Velvet). Females laid significantly fewer eggs on another cultivar, Petoy, than they laid than Peterstar, however the development of immatures was highly successful on Petoy. Further work with these 4 cultivars showed a very high intrinsic rate of increase (Rm=0.434) on Petoy and a low value (Rm=0.190) on Pepride for the  $F_0$  generation. This is interesting, since Petoy was not the most preferred host for oviposition. On the other hand, Pepride was among the least preferred oviposition host and this cultivar had the lowest intrinsic rate of increase. These results are not conclusive that the most preferred cultivars are those on which the whitefly will perform the best.

From a practical standpoint, the difference in Rm values for the cultivars can significantly alter the management of silverleaf whitefly on poinsettia. Small differences in Rm values result in remarkable differences in expected population growth over time. Our results indicate that the predicted density of whiteflies on the cultivar 'Petoy' would be ten times higher than on the relatively resistant cultivar 'Red Velvet' (189,094 vs. 17,676 respectively) after just 30 days. Using these empirical curves, and an arbitrary treatment threshold of 100 whiteflies, we compared the hypothetical number of treatments required across a 90 day time frame for the four varieties evaluated in chapter 3. 'Petoy' would require 7 pesticide treatments within 90 days, while 'Red Velvet' would require only 6 pesticide treatments within 90 days. Thus, the attributes of a single cultivar may result in fewer pesticide applications and possible overall savings in the costs associated with chemical control for growers. Growers should utilize this information on Pepride and incorporate this cultivar into their breeding programs.

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# Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host Plant Interaction

## Dates Covered by the Report: 2000

#### Watermelon Germplasm for Resistance to Bemisia

Like many vegetable crops, the cultivated watermelon, *Citrullus lanatus* var. *lanatus*, is plagued by *Bemisia*. *Citrullus* contains four known diploid species. However, there have not been any reports on screening *Citrullus* germplasm for resistance to whiteflies. This study was conducted to identify potential germplasm sources that could be used in the improvement of watermelon for whitefly resistance. The study was conducted on 42 *Citrullus* genotypes. Seven watermelon cultivars, a triploid line, and 16 U.S. Plant Introduction accessions (PIs) of *C. lanatus* var. *lanatus*; 10 PIs of *C. lanatus* var. *citroides*; and 8 PIs of *C. colocynthis* were evaluated for resistance to B-biotype *B. tabaci*. Bioassays were conducted on all genotypes studied, but the abundance of the insects and the survival of the nymphs varied greatly among genotypes. Most of the watermelon cultivars and *C. lanatus* PIs tested were highly susceptible to whitefly infestation, while the *C. colocynthis* PIs exhibited whitefly resistance. Trichome density on the resistant genotypes (*C. colocynthis* PIs) were elevated compared with the susceptible genotypes. However, we believe that the observed resistance was primarily related to effects of plant nutrition on the whiteflies.

This study identified useful sources of germplasm (such as PI 386015, PI 386018, and PI 386024, all of *C. colocynthis*) which can be used for the improvement of watermelon for resistance to whiteflies. Currently,  $F_2$  populations (Charleston Gray x PI 386024) are being constructed and will be used to determine the mode of inheritance of whitefly resistance in watermelon.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

# Dates Covered by the Report: 2001

# Movement of Whitefly Crawlers on Several Types of Vegetable Plants

The nymphal stage of whitefly pests is important in terms of its relationship to plant damage, plant disorders, and pest management measures. It is well known that the B-biotype sweetpotato whitefly, *Bemisia tabaci* (Gennadius), feeds on and damages numerous species of vegetable crops. The crawler, i.e., active first instar, is the only mobile form of the immature whitefly. A study was conducted to determine any influence of vegetable plant species and temperature on net distance moved (between the egg site and final resting site) by crawlers of the B-biotype *B. tabaci*. Tests were conducted in the greenhouse as well as under controlled laboratory conditions on five vegetable hosts: cantaloupe, *Cucumis melo* L.; collard, *Brassica oleracea* ssp. *acephala* de Condolle; cowpea, *Vigna unguiculata* (L.) Walpers ssp. *unguiculata*; pepper, *Capsicum annuum* L. ssp. *annuum*; and tomato, *Lycopersicon esculentum* Miller. On the different host species, the average net distance that crawlers moved ranged from 2-15 mm with the shortest distance on collard. Individuals on pepper and cowpea ceased traveling 50 and 62 mm, respectively, from the site of hatching. Observations on intact collard leaves in the laboratory indicate that the crawlers traveled an average of 21 minutes before developing to the 2<sup>nd</sup> instar. No effect of constant temperature (16-34°C) was detected on the net travel distance of the crawler. The data suggest that among the plant species in this study, collard is highly attractive for feeding and/or it offers suitable feeding sites that are easy to locate by the crawler. The results of this study help define the behavior of crawlers on several species of vegetable plants and help in ongoing research on host plant resistance.

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Research & Implementation Area: Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions

Dates Covered by the Report: January-2000-December 2000

## Silverleaf whitefly-induced (SLW) Genes: Species-Specific Induction of Novel Signaling Pathways

The squash (*Cucurbita pepo*) genes *SLW1* and *SLW3* were differentially expressed in response to silverleaf whitefly (*Bemisia argentifolii*) and sweetpotato whitefly (*B. tabaci* Type A) nymphs (van de Ven., 2000). *SLW1* and *SLW3* RNAs accumulated systemically in apical, silvered leaves after silverleaf whitefly feeding, but not sweetpotato whitefly feeding. Both RNAs accumulated, although at different levels, in leaves on which silverleaf and sweetpotato whitefly nymphs were feeding. The expression of *SLW1* and *SLW3* in response to aphids, caterpillars, nematodes and a bacterial pathogen were evaluated. Consistent with the lack of *SLW1* and *SLW3* expression after mechanical wounding, *SLW1* and *SLW3* transcripts did not accumulate in response to tobacco hornworm (*Manduca sexta*) larvae. Surprisingly, other animals that feed on the vasculature, such as the cotton aphid (*Aphis gossypii*) or root-knot nematodes (*Meloidogyne incognita*), did not cause local accumulation of *SLW1* and *SLW3* RNAs in squash leaves or roots, respectively, or systemic RNA accumulation in non-infested leaves. Infiltration of leaves with *Pseudomonas syringea* pv *syringea* did not induce *SLW1* or *SLW3*. Collectively, these data suggest that *SLW1* and *SLW3* are not typical pathogen- or pest-response genes, since they were preferentially expressed by a single whitefly species.

To investigate the signaling pathways important for *SLW* gene expression, squash plants were treated with a variety of defense and wound signals. *SLW1* (a M20B peptidase gene) was regulated by both JA (jasmonic acid) and ethylene and therefore is activated by an established JA/ethylene-dependent pathway. *SLW3* (a â-glucosidase gene) RNAs did not accumulate in response to JA, ethylene, salicylic acid, abscisic acid, nitric oxide, hydrogen peroxide or a combination of these defense signals. These data indicate that *SLW3* is regulated by a novel, defense-signaling pathway not previously recognized in plants. *SLW1* and *SLW3* RNAs were abundant after water-deficit stress suggesting that the signals generated after silverleaf whitefly feeding and water-deficit stress may overlap or *SLW* genes may respond to multiple signaling pathways. A homolog of *SLW1*, *DRIP-1*, was recently found in watermelon and was also induced after water-deficit stress (Kawasaki et al, 2000).

To understand the tissue specificity of *SLW* gene expression and to evaluate the impact of *SLW1* and *SLW3* proteins on plant defense, we are using the tools of reverse genetics. *SLW1* and *SLW3* promoters were isolated, sequenced, and fused to the â-glucuronidase (GUS) reporter gene in a *Ti*-plasmid binary vector. These chimeric constructs have been used in *Agrobacterium tumefaciens*-mediated transformation. Transgenic tomato and *Arabidopsis* plants expressing *SLW1:GUS* or *SLW3:GUS* transgenes are being regenerated and characterized. These plants will aid in unraveling the similarities and differences between the signals that induce *SLW1* and *SLW3*. Transgenic tomatoes and *Arabidopsis* over-expressing *SLW3* and *SLW1* are also being constructed and will allow us to determine the role of *SLW* genes on whitefly infestation and leaf-silvering.

#### Section E Research Summary

Compiled by C. L. McKenzie and Greg Walker

#### **Host-Plant Resistance:**

Advancements continue to be made in breeding and screening plants for resistance to whitefly and whiteflytransmitted viruses and include studies with cultivars, breeding-lines and germplasm of cantaloupe, watermelon, poinsettia, and cotton.

Six cantaloupe varieties were evaluated under field conditions for whitefly resistance in south Texas: Hymark, Impact, Tam Sun x gl, Tam Sun, Primo, and TP45. TP45, a Weslaco line, had the fewest whiteflies (adults and red-eyed-nymphs or pupae) throughout the season, followed by Tam Sun, a TAES variety. Although Tam Sun had the second lowest whitefly numbers, it also had the fewest and lowest total weight of melons. Primo had the greatest number of adult whiteflies (660 / leaf) followed by Hymark and Impact (>300 / leaf). Number of adult whiteflies was > 8-fold higher on Primo compared to TP45. TP45 and Impact had more large melons compared with other varieties whereas Tam Sun x gl. and Primo had more small melons. TP45 also had the highest yield, but unfortunately sugar content was the lowest (~3.8%) compared to other varieties (7.6 - 9.5%). Results from this study indicated that significant differences in yield responses to silverleaf whitefly infestation were found among the six cantaloupe varieties and confirm earlier laboratory findings.

In South Carolina, watermelon germplasm (42 Citrullus genotypes) was screened for resistance to whitefly infestation to identify potential germplasm sources that could be used in the improvement of cultivated watermelon varieties. Seven watermelon cultivars, a triploid line, 16 U.S. Plant Introduction accessions (PIs) of C. lanatus var. lanatus, 10 PIs of C. lanatus var. citroides, and 8 PIs of C. colocynthis were evaluated for whitefly resistance. Bioassays were conducted on whitefly non-preference, oviposition and survival. Most of the watermelon cultivars and C. lanatus PIs tested were highly susceptible to whitefly infestation, while the C. colocynthis PIs exhibited whitefly resistance. This study identified useful sources of germplasm (PI 386015, PI 386018, and PI 386024, all of C. colocynthis) which can be used for the improvement of watermelon for resistance to whiteflies.

Seven poinsettia cultivars were evaluated at U CA in Riverside for preference and performance of silverleaf whitefly. Peterstar was the most preferred cultivar while Pepride and Red Velvet were the least preferred. Development of immature stages that eclosed from these eggs were consistent with the numbers of eggs laid on the cultivar (good development on Peterstar, and poor development on Pepride and Red Velvet). Females laid significantly fewer eggs on Petoy compared to Peterstar, however the development of nymphs was highly successful on Petoy (Rm=0.434). In contrast, Pepride was among the least preferred oviposition host plus it had the lowest intrinsic rate of increase (Rm=0.190). Results are not conclusive that the most preferred cultivars are those on which the whitefly will perform the best. Growers should utilize this information on Pepride by incorporating this cultivar into their breeding programs.

Sixteen upland cotton types were evaluated in the field for susceptibility to silverleaf whitefly infestations in California's Imperial Valley and included 10 normal leaf and three okra-leaf cultivars plus three experimental okraleaf breeding-lines. The okra-leaf entries as a group had fewer whitefly adults and nymphs than the normal leaf cotton entries. The okra-leaf experimental breeding-lines had the lowest numbers of whitefly adults and nymphs among the okra-leaf entries. Stoneville 474 (hirsute leaf) had the greatest numbers of whitefly adults and nymphs among the normal leaf cottons. However, no significant differences in seed cotton yield among the entries were detected.

Ten cotton cultivars or breeding-lines were evaluated in Imperial Valley, CA for resistance to cotton leaf crumple (CLCr) disease caused by cotton leaf crumple virus (CLCrV) and transmitted by the silverleaf whitefly. Results showed differences in whitefly infestation levels and virus disease symptoms among cotton entries in terms of severity of CLCr disease symptoms and the presence of CLCrV. The cultivar AP 4103 had a higher CLCr disease rating than other entries except AP 6101. The breedingline NK 2387C, with Cedix parentage, had a lower CLCr disease rating than other entries except Stoneville 474 and NKX 2207. Visible CLCr disease symptoms were observed in Stoneville 474 and NKX 2207 entries, but NK 2387C did not display visible CLCr disease symptoms nor was viral DNA detected implicating this line as a potential source for improvement of cotton resistance to CLCrV and disease development.

# **Physiological Disorders:**

Great strides have been made in understanding the plant molecular mechanisms and genetics of squash silverleaf (SSL) disorder, a serious physiological disorder of commercial *Cucurbita* species associated with the feeding of nymphal *Bemisia argentifolii*, and how that translates into reduction in plant growth and yield observed in the field.

At the U CA (Riverside) researchers discovered that silverleaf whitefly-induced (*SLW*) genes utilize novel signaling pathways that appear to be species -specific. The squash (*Cucurbita pepo*) genes (*SLW1* and *SLW3*) were differentially expressed in response to silverleaf whitefly (SLWF) and sweetpotato whitefly (SPWF) nymphs. Both *SLW* genes accumulate systemically in apical, non-infested, silvered leaves after SLWF feeding, but not after SPWF feeding. Both *SLW* transcripts accumulate in the local, infested leaves of SLWF infested plants, however only *SLW3* was induced in the local leaves of SPWF infested leaves. The expression of *SLW1* was detected during flower and fruit development, while *SLW3* RNAs were not. In contrast, neither gene transcript accumulated in response to insects, nematodes or a bacterial pathogen. Collectively, these data suggest that *SLW1* and *SLW3* are not typical pathogen- or pestresponse genes, since they were preferentially expressed by a single whitefly species.

*SLW1* (a M20B peptidase gene) is regulated/activated by an established jasmonic acid/ethylene-dependent pathway. However the signaling pathway for SLW3 (a âglucosidase gene) appears to be regulated by a novel, defense-signaling pathway not previously recognized in plants because plant RNA transcripts did not accumulate in response to known defense and wound signals. In addition, both genes were abundant after water-deficit stress suggesting that the signals generated after SLWF feeding and water-deficit stress may overlap or *SLW* genes may respond to multiple signaling pathways.

The reverse genetic approach is being used to investigate the tissue specificity of *SLW* gene expression and function of *SLW* genes in the plant response to *SLWF* feeding and evaluate the impact of *SLW* proteins on plant defense. *SLW3: GUS* transgenic tomato and *Arabidopsis* have been generated and *SLW3* expression studies under abiotic and biotic stresses and during plant development are ongoing. In addition, transgenic plant lines ectopically expressing *SLW3* have been constructed and will aide in future studies to determine the role of *SLW* genes on whitefly infestation and leaf silvering.

Silvering results from an abnormal air space between the upper epidermis and the upper palisade layer of cells, beginning at the tip of young leaves and proceeding downward to cover the entire leaf. The number of cells in each layer over time is the same in whitefly-infested plants and controls indicating cell separation between epidermis and palisade cells is not a result of differential cell division or expansion between the two layers. Silvering develops simultaneously as normal air space formation is occurring and is more likely a result of increased cell wall degradation. No anatomical response to whitefly feeding was observed in SSL-resistant zucchini, but plants were lighter green with shorter petioles and comparable to SSL-susceptible plants. In grafts of resistant plants onto susceptible plants challenged with whitefly nymphs, the resistant plants did not develop leaf silvering. However, susceptible scions grafted onto resistant rootstocks did exhibit leaf silvering indicating that the site of SSL resistance is at the young developing leaves and not at the mature leaf where whitefly feeding occurs.

Companion field trials conducted in Mayaguez, Puerto Rico and Leesburg, FL screened *Cucurbita* spp. (squash) accessions obtained from the National Plant Germplasm System repository and identified several resistant lines to SSL disorder. Researchers found that even though SSLresistant germplasm did not silver, plants received similar levels of oviposition and supported similar rates of development and survival as did SSL-susceptible accessions indicating that the mechanism of resistance was tolerance to the effects of whitefly feeding. The genes responsible for SSL resistance appear to be related to the absence of natural leaf mottling and are conditioned by one or two genes depending on the accession. Environmental conditions and whitefly infestation level can modify the severity of SSL symptoms.

Studies to determine the effect of whitefly and squash silverleaf disorder (SSL) on zucchini plant growth and yield were conducted at the UFL, Gainesville in greenhouse and outdoor screened room experiments using two closely-related zucchini breeding-lines, ZUC61 (silverleaf susceptible) and ZUC76-SLR (silverleaf resistant), and four infestation levels. Greenhouse results indicated seedlings for both zucchini breeding-lines were stunted, had shortened plant height and petiole length of silvered leaves, and showed ~ 10% reduction in dry weight after infestation of 30-60 pairs of adult whiteflies and their progeny over 2 weeks. Susceptible plants exhibited severe (grade 5) SSL symptoms whereas ZUC76-SLR had no or mild (grade 1) symptoms, yet the same reduction in seedling growth was observed. In outdoor screened room experiments, serious silverleaf symptoms (grade 4-5) were expressed on all susceptible plants and slight silverleaf symptoms (grade 1-3) were expressed on resistant plants, but silverleaf severity of each zucchini breeding-line did not differ significantly among whitefly infestation levels (spring or fall). Significant fruit yield reduction appeared at infestation level 30 for both breeding-lines. Season-long feeding by higher infestation levels (60-120 pairs) caused significant yield and quality reduction in fruit for both breeding-lines and even resulted in plant death in fall experiments. Zucchini seedling stunting and yield reduction appears to be more related to intensity of whitefly feeding than to susceptibility to SSL and expression of silverleaf symptoms.

#### **Host-Plant Interactions:**

In Egypt, parasitism of whitefly on 16 taxonomically diverse host plants by augmentation with *Eretmocerus mundus* Mercet was determined in field plots across five locations. Vegetable and other agronomic host plants included bean, broccoli, cabbage, cantaloupe, cotton, cucumber, egg plant, lantana, potato, southern pea, soybean, sugar beet, sunflower, sweet potato, tomato, and watermelon. Parasitism was enhanced in all plots where augmentations were made, but rates differed. In some plots, e.g., cabbage, broccoli, bean, and soybean, overall parasitism was enhanced at a relatively high rate while in other plots, e.g., watermelon, egg plant, and tomato, the enhancement of overall parasitism was relatively low.

Researchers in South Carolina conducted greenhouse and laboratory experiments to determine the influence of five vegetable hosts (cantaloupe, collard, cowpea, pepper and tomato) and temperature on net distance traveled by whitefly crawlers. No effect of constant temperature (16-34°C) was detected on the net travel distance of the crawler. The average net distance that crawlers moved ranged from 2-15 mm on the different hosts, but crawlers on pepper and cowpea ceased traveling 50 and 62 mm, respectively, from the site of hatching. Crawlers moved the shortest distance on collards and traveled an average of 21 minutes before developing to the 2<sup>nd</sup> instar. The data suggest that among the plant species in this study, collard is highly attractive for feeding and/or it offers suitable feeding sites that are easy to locate by the crawler.

Nine upland cotton cultivars were evaluated for whitefly populations in relation to trichome densities in field trials over two growing seasons at the U AZ Agricultural Research Center, at Maricopa, AZ. Entries included five normal-leaf and four okra-leaf cotton cultivars with all but the hairy-leaf ST 474 being smooth-leaf. ST 474 had 71fold and > 6-fold more stellate trichomes/cm<sup>2</sup> of leaf disk compared to smooth-leaf cultivars in 2000 and 2001, respectively. ST 474 had > 2-fold more  $eggs/cm^2$  and nymphs/cm<sup>2</sup> leaf disk in 2000 and >2-fold more adults per leaf-turn count compared to smooth-leaf cultivars in both years. In 2001, ST 474 had > 4-fold more eggs/cm<sup>2</sup> and nymphs/cm<sup>2</sup> leaf disk compared to smooth-leaf cultivars. On average of all cultivars studied for both years, leaves on the top main stem leaf node (#1) had the highest number of stellate trichomes, but the lowest numbers of eggs, nymphs and adults compared with leaves lower in the canopy.

Research approaches	Year 1 Goals Statement	Progress Achieved Yes No	Significance
Characterize resistance mechanisms and identify chemical/morphological components, and study effects of insect adaptation.	Identify potential sources of germplasm for disease, plant disorders and whitefly resistance. <sup>a</sup>	Х	Research was conducted on identifying potential sources of germplasm for whitefly resistance in alfalfa, cotton, melon, cole crops, and cucurbits; and resistance to virus symptoms and silverleaf disorder in cotton and cucurbits, respectively. These studies included research on plant tolerance, antibiosis, and antixenosis. Antixenosis was found not to be responsible for resistance to squash silverleaf in two zucchini lines.
Develop molecular level techniques to produce resistant germplasm.	Identify physiological processes of whiteflies to target for inhibition.	Х	Characterization of plant genomone was demonstrated in tomato and squash. Pathogenesis related mRNAs accumulated in response to whitefly feeding on tomato leaves. Data on whitefly probing behavior indicates that host evaluation phase of <i>Bemisia</i> -host interaction is dominated by probing.
Incorporate resistance traits into commercial genotypes.	Identify and isolate genetic sources of resistance for transformation and/or breeding.	Х	From promising genetic materials, inbreeds, $F_1$ and $F_2$ progenies, and assorted cultivars were studies for whitefly resistance (in alfalfa, cotton, melon and squash), and susceptibility to diseases (in cotton) and plant disorders (in squash). Including plant geneticists and other specialists on the research team has been an asset.
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition. <sup>b</sup>	Characterize nutritional and other preference properties of various host plants.	Х	Research was studied on the acceptability of cotton and vegetable hosts on whitefly feeding behavior. Work was conducted on distance from abaxial surface to minor veins, and feeding response on abaxial and adaxial surfaces of different hosts.
Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>c</sup>	Investigate approaches for interruption of feeding, assimilation, development and reproduction.	Х	The host evaluation phase of <i>Bemisia</i> -host interactions was shown to dominate by probing, and the time spent in a particular behavior was affected by imidacloprid when the whitefly came into contact with the chemical in its diet rather than on the leaf surface. Intercropping of resistant within susceptible cole crops did not lessen the abundance of whiteflies.

		Progress Achieved	
Research approaches	Year 1 Goals Statement	Yes No	Significance
Study whitefly toxicogenic plant reactions.	Determine effects of whitefly feeding on host plant physiology, morphology and anatomy.	Х	Research on tomato identified a gene that is specifically induced by whitefly feeding. Four classes of genes were identified in inducing squash leaf silvering. These genes were further characterized by hybridization, sequence analysis and complementation studies.

<sup>a</sup> See Table B for additional plant disease resistance research.
 <sup>b</sup> See Section A.

<sup>c</sup> See Section A, approach #9.

		Progress Achieved	
Research approaches	Year 2 Goals Statement	Yes No	Significance
Characterize resistance mechanisms and identify chemical/morphological components, and study effects of insect adaptation.	Determine physiological and/or morphological basis for resistance, & effects of host-plant history and insect adaptation on plant resistance to whiteflies. Continue to identify resistant germplasm.	X	Selection for a whitefly resistant variety of alfalfa is close to completion; release of a commercial variety is expected within a few years. Whitefly-resistant or partially whitefly-resistant varieties of a number of crops have been identified, including cotton, collard, and melons. Varieties of cotton and tomato with resistance or partial resistance to whitefly-transmitted viruses also have been identified. In collards, the glossy leaf trait, and in cotton, the okra-leaf trait and large leaf surface to vascular bundle depth have been implicated as mechanisms of whitefly resistance in plants. Increased levels of phenolics and peroxidase in response to plant stress have been associated with decreased whitefly performance in tomato. In <i>Datura wrightii</i> , glandular trichomes were demonstrated to be a very effective mechanism of resistance to whiteflies.
Develop molecular level techniques to produce resistant germplasm.	Identify natural products for inhibiting processes.	Х	The natural plant products, neem seed extract, azadiractin, and extract of bitterwood, were shown to be effective insecticides against silverleaf whitefly.
Incorporate resistance traits into commercial genotypes.	Insert genes into plants <sup>b</sup> via plant transformation.	Х	Resistant commercial lines of alfalfa are close to release and commercial varieties of collard have been shown to exhibit whitefly resistance. Also, lines of cotton and melon have been identified with partial whitefly resistance. No progress has been made in the specific year 2 goal of inserting whitefly resistance genes into plants via transformation.

		Progress Achieved	
Research approaches	Year 2 Goals Statement	Yes No	Significance
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition. <sup>b</sup>	Determine the biochemical mechanism regulating adaptation to host plants.	X	Morphological plant traits such as okra-leaf and large distance from leaf surface to vascular bundles in cotton, and glandular trichomes in <i>Datura wrightii</i> have been shown to provide partial or complete whitefly resistance. Fluctuations in amino acid concentrations over the lifespan of melon leaves were correlated with whitefly performance. Also in melons, group feeding by whiteflies was shown to create a nutrient sink in the plant, and thus provide the whiteflies with improved amino acid nutrition. Senescence in poinsettia reduces host plant quality for silverleaf whitefly. In cotton, decreased nitrogen fertilization decreases whitefly populations. In tomato, plant stress caused by fertilizer and/or water deficiency reduces host plant quality for silverleaf whitefly.
Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>c</sup>	Identify physiological and morphological mechanisms regulating processes.	Χ	Improvements have been made in a system for rearing whiteflies on an artificial liquid medium. This will allow direct experimentation on the role of specific plant nutrients and allelochemicals on whitefly feeding and performance. Stylet contact with minor vascular bundles is essential for successful whitefly feeding on cotton. The fine structure of whitefly eggs and their attachment to host leaves has been studied with electron microscopy, and the distal end of the egg petiole that is inserted into the host leaves possesses morphological structures that suggest a role in water uptake from the host leaf which is a very important process for egg survival.
Study whitefly toxicogenic plant reactions.	Determine biochemical basis for physiological response of plant.	Х	Genes specifically induced by whitefly feeding have been identified in tomato and in squash. These genes may play a role in the plant's defensive response to the whitefly and/or the plant's toxicogenic reaction such as irregular ripening in tomato and silverleaf symptom in squash.

<sup>a</sup> See Table B for additional plant disease resistance research.
<sup>b</sup> See Section A.
<sup>c</sup> See Section A, approach #9.

		Progress Achiev	
Research approaches	Year 3 Goals Statement	Yes No	Significance
Research approaches Characterize resistance mechanisms and identify chemical/morphological components, and study effects of insect adaptation.	Year 3 Goals Statement Elucidate biochemical and molecular basis for resistance. Continue to identify resistant germplasm.	0	Selection for a whitefly resistant variety of alfalfa is close to completion; release of a commercial variety is expected within a year. Whitefly fecundity tests on clonal propagules of alfalfa plants chosen in the field for whitefly resistance indicate that the field- selection criteria reflect actual resistance. Fecundity of whiteflies on alfalfa was higher for alfalfa-reared whiteflies than for cotton-reared whiteflies. This suggests whitefly adaptation to a crop species. In cotton, the okra-leaf trait and glabrous-leaf trait have been again demonstrated as a mechanisms of partial resistance against whiteflies; however, closer scrutiny of an earlier report that a large leaf surface to vascular bundle depth confers whitefly resistance has been discredited. This is useful information so that resources can be focused on examining effective
			has been discredited. This is useful information so
			ovipositing female. This suggests that whiteflies will readily oviposit on resistant crops, and consequently mechanisms of plant resistance will be continuously challenged by migrating whiteflies.

Research approaches	Year 3 Goals Statement	Progress Achieved Yes No	Significance
Develop molecular level techniques to produce resistant germplasm.	Isolate the relevant biosynthetic enzymes that encode for natural products inhibiting processes.	Х	The natural plant products, azadiractin, was shown to be an effective insecticide against silverleaf whitefly. Presently, there are no attempts to insert the genes for this plant product into crop plants.
Incorporate resistance traits into commercial genotypes.	Evaluate potential of newly transformed germplasm.	Х	Whitefly-resistant commercial lines of alfalfa are close to release. Commercially available varieties of cotton, cantaloupe, and poinsettia that are resistant or partially resistant against whiteflies or whitefly- transmitted viruses have been identified. No genetically transformed germplasm has yet been evaluated for whitefly resistance.
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition. <sup>b</sup>	Determine changes in whitefly gene expression in response to host manipulation.	Χ	In cotton, the okra-leaf trait and glabrous-leaf trait have been confirmed as a mechanisms of partial resistance against whiteflies; however, closer scrutiny of an earlier report that a large leaf surface to vascular bundle depth confers whitefly resistance has been discredited. Factors encountered by whiteflies during their stylet penetration to vascular bundles has been shown to confer partial resistance in a tomato variety with the <i>Mi</i> gene. Phloem sap factors do not appear to play a role in this resistance. The known host plant range of silverleaf whitefly has been expanded to include some medicinal plants and weed species. An abundance of host plant species suitable for overwintering in California's San Joaquin Valley have been identified; thus a strategy of host- free periods for whitefly management is not very promising in the San Joaquin Valley.

		Progress A	chieved	
Research approaches	Year 3 Goals Statement	Yes	No	Significance
Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>c</sup>	Determine biochemical and molecular basis for inhibiting processes.	X		Morphological studies on whitefly stylets indicate that they are sufficiently long to reach minor vascular bundles (the major feeding site) from virtually any place on the abaxial leaf surface of cotton. This makes variation in vascular bundle depth an unlikely mechanism of resistance to whiteflies in cotton. Variation in nitrogen fertilization has been shown to decrease amino acid concentrations in phloem sap and thus affects nutrition available to whiteflies. Whitefly feeding differentially induces pathogenesis - related (PR) proteins in two cucurbit species, cantaloupe and watermelon, and apparently is not affected by treatment with plant growth-promoting rhizobacteria (PGPR).
Study whitefly toxicogenic plant reactions.	Elucidate changes in plant gene expression.	Х		Two genes, one of which appears to be a general plant defense, have been shown to be differentially induced in squash by silverleaf and sweetpotato whiteflies. This may be related to the different toxicogenic effects of these two whitefly species on squash. The activation of these genes is systemic. In tomatoes, feeding by both silverleaf whitefly and greenhouse whitefly induced pathogenesis related genes, but not genes regulated by the octadecanoid pathway. These studies indicate that tomato plants perceive phloem-feeding silverleaf and greenhouse whiteflies in a manner distinct from that of chewing insects.

<sup>a</sup> See Table B for additional plant disease resistance research.
<sup>b</sup> See Section A.
<sup>c</sup> See Section A, approach #9.

Research approaches	Year 4/5 Goals Statement	Progress Achieved Yes No	Significance
Characterize resistance mechanisms and identify chemical/morphological components, and study effects of insect adaptation.	Determine potential for transfer of resistance traits.	X	Natwick & Walker's work identified high levels of whitefly resistance in the wild cotton species, <i>Gossypium thurberi</i> , a Gossypium species that in the past had been successfully used to introduce new genes into commercial cotton. Chu and Natwick's work on Siokra cotton added to this body of work. Heather McAuslane's lab (U FL) determined that the genes responsible for SSL disorder appear to be related to the absence of natural leaf mottling and are conditioned by one or two genes depending on the accession.
	Evaluate potential for incorporating <i>Bemisia</i> , plant disorder and disease resistance into acceptable plant type. <sup>a</sup>	Χ	Larry Tueber (UC Davis) developed and released a commercial whitefly resistant alfalfa variety (UC- Impalo WF) for desert SW alfalfa growing regions. Tong-Xian Liu (TAES) evaluated cantaloupe varieties for whitefly resistance in south Texas and found significant differences in yield responses to whitefly pressure. R.T. McMillan (U FL) identified whitefly transmitted virus resistant beans and released a commercially available bean cultivar (Genuine) for resistance to Bean Golden Mosiac virus in south Florida. Natwick evaluated cotton for resistance to CLCrV and identified NK 2387C as a potential source for improvement of cotton resistance to CLCrV and disease development. McAuslane's lab identified several squash accessions from the National Plant Germplasm System repository that were resistant to SSL disorder.
Develop molecular level techniques to produce resistant germplasm.	Insert genes into plants via plant transformation. <sup>b</sup>	Х	Jane Polston (U FL) successfully transformed tomato for resistance to ToMoV and found that plants resistant to ToMoV also appear to convey resistant to TYLCV. Linda Walling's lab (UC Riverside) has generated <i>SLW3:GUS</i> transgenic tomato and <i>Arabidopsis</i> for expression studies.
	Evaluate potential of newly transformed germplasm. <sup>b</sup>	Х	Polston's lab is currently evaluating transformed tomato for field performance and resistance to TYLCV.

	Progress Achieved		
Research approaches	Year 4/5 Goals Statement	Yes No	Significance
Incorporate resistance traits into commercial genotypes.	Continue to refine resistance factors to improve resistance in newly transformed germplasm.	Х	Through conventional breeding, Teuber and Walker are working on improving whitefly resistance in alfalfa and are making progress identifying the mechanism of resistance.
	Incorporate other desirable plant characteristics for crop production	Х	Teuber's continued breeding to incorporate more desirable traits into the whitefly-resistant alfalfa that he developed.
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition. <sup>b</sup>	Relate changes in gene expression to whitefly physiology	Х	Walling's lab determined that silverleaf whitefly- induced genes utilize novel signaling pathways that appear to be species-specific. These pathways appear to overlap with signals generated by water- deficit stress or <i>SLW</i> genes may be responding to multiple signaling pathways.
	Summarize and disseminate results	X but limited	Manuscripts are in preparation.
Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>c</sup>	Determine potential for transfer of resistance traits.	Χ	Perring's lab (UC Riverside) looked at whitefly preference and the intrinsic rate of increase between poinsettia cultivars and found Pepride to be a promising cultivar for improving breeding programs. Simmons evaluated the effect of temperature and five vegetable hosts on net distance traveled by crawlers and found no effect of constant temperature (16- $34^{\circ}$ C) with crawlers traveling the shortest distance on collards < cowpea < pepper before developing into the 2 <sup>nd</sup> instar. Chu consistently found whitefly to prefer hairy-leaf cotton (ST 474) to normal-leaf and okra-leaf cultivars evaluated with > 2 to 4-fold more eggs, nymphs and adult whiteflies observed. Researchers in Egypt found parasitism to be enhanced by augmentation with <i>Eretmocerus mundus</i> Mercot in all of the 16 taxonomically diverse host plants studied, but rates differed from relatively high (cole and legume hosts) to relatively low (crucifer and solanaceous crops).
	Insert genes into plants <sup>a</sup> via plant transformation	Х	See notes above regarding Polston and Walling's work.

# Table E. Host-Plant Resistance, Physiological Disorders, and Host-Plant Interactions (Continued) Progress Achieved

Research approaches	Progress Achieved			
	Year 4/5 Goals Statement	Yes No	Significance	
Study whitefly toxicogenic plant reactions.	Identify resistance germplasm.	Х	See notes above regarding Natwick & Walker's work on <i>Gossypium thurberi</i> and Chu & Natwick's work on Siokra cotton. See R. T. McMillan, Tong-Xian Liu, McAuslane above. Also, Walker has identified several genotypes of alfalfa highly resistant to <i>Bemisia</i> . Simmons (USDA-ARS) screened watermelon germplasm and identified <i>Citrullus</i> <i>colocynthis</i> accessions and other PIs, which are being used to improve cultivated watermelon varieties for resistance to whiteflies.	
	Evaluate potential for transferring new germplasm.	Х	McMillan and Tueber have released commercially available resistant bean and alfalfa varieties, respectively. Other programs with tomato, melon, squash, and cotton are in various stages of getting resistant cultivars to the grower.	

<sup>a</sup> See Table B for additional plant disease resistance research.
 <sup>b</sup> See Section A.
 <sup>c</sup> See Section A, approach #9.

## Reports of Research Progress

## Section F: Integrated and Areawide Pest Management Approaches, and Crop Management Systems Co-Chairs: Steve Castle and Peter Ellsworth

Investigator's Name(s): C. C. Chu, P. Alexander, C. G. Jackson, and T. J. Henneberry

Affiliation & Locations: USDA-ARS, Western Cotton Research Laboratory, Phoenix, AZ

**Research & Implementation Area:** Section F: Integrated and Areawide Pest Management Approaches, and Crop Management Systems

## Dates Covered by the Report: 1998-2001

# A Light-Emitting Diode Equipped CC Trap

Trap catches of adult *Bemisia tabaci* (Gennadius), biotype-B, and their *Eretmocerus* spp. and *Encarsia* spp. parasitoids were compared in cage studies in the greenhouse. Average catches of adult *B. tabaci* in CC traps were 41% of the numbers caught on 100 cm<sup>2</sup> yellow sticky card (YC) traps. Coating the insides of CC trap tops with Tanglefoot® (TCC trap) and removing the deflector plate, increased adult *B. tabaci* catches to 65% of the number caught on YC traps. Equipping TCC traps with light-emitting diodes (LED-CC trap) increased adult *B. tabaci* catches by 614% compared with TCC traps. Few *Eretmocerus* spp. and *Encarsia* spp., were caught in LED-CC traps. The LED-CC traps may have potential in greenhouse crop production systems where YC traps are used to supplement parasite releases for *Bemisia* control.

Investigator's Name(s): C. C. Chu<sup>1</sup>, A. M. Simmons<sup>2</sup>, P. J. Aexander<sup>1</sup>, and T. J. Henneberry<sup>1</sup>

**Affiliation & Locations:** <sup>1</sup>USDA-ARS, Western Cotton Research Laboratory, Phoenix, AZ, and <sup>2</sup>USDA-ARS, U.S. Vegetable Laboratory, Charleston, SC 29414

**Research & Implementation Area:** Section F: Integrated and Areawide Pest Management Approaches, and Crop Management Systems

# Dates Covered by the Report: 2000-2001

## A Light-Emitting Diode Equipped Yellow Sticky Card Trap

Yellow sticky card (YC) traps were modified with 530 nm lime green light-emitting diodes (LED-YC) to increase the efficacy of capture of *Bemisia tabaci* (Gennadius) and other greenhouse insects as compared with catches on standard YC traps. More adult *B. tabaci* (biotype-B) and fungal gnats, *Bradysia coprophila* (Lintner), were caught on LED-YC traps compared with standard YC traps. Likewise, more *Delphastus pusillus* (LeConte) (coccinellid predators), were caught on the LED-YC traps compared with standard YC traps in a greenhouse test with collard plants, but not in a test with melon plants. However, in tests on both crops, the LED-YC traps did not catch more *Eretmocerus* spp. (parasitoids of whiteflies) than the standard YC traps. Moreover, the capture of western flower thrips, *Franklinella occidentalis* (Pergrande) was not affected by the LED-YC trap compared with the standard YC trap. These results demonstrate the potential of using LED-YC traps in greenhouses for insect detection, monitoring, and control.

Investigator's Name(s): Robert T. McMillan, Jr., M. J. Davis, Z. Ying and Dakshina Seal

Affiliation & Location: University of Florida, IFAS, TREC 18905 SW 280th Street Homestead, FL 33031

**Research & Implementation Area**: Section F. Integrated and Areawide Pest Management Approaches, and Crop Management Systems

Dates Covered by Report: 1999-2000 Crop Season

# Tomato Yellow Leaf Curl Gemini Virus Management in South Florida

Tomato Yellow Leaf Curl Geminivirus (TYLCV) was identified in the United States for the first time in late July 1997 in a field planting at a commercial breeding facility in Florida. Shortly thereafter, infected tomato plants were also found in several retail garden outlets in Florida (personal communication J. Polston). The source of these plants was traced back to two commercial nurseries near Homestead in south Florida that had shipped tomato plants to retail outlets in Florida and other states and out of the country. Subsequently, infected tomatoes from retail garden outlets have been found in Virginia (confirmed by laboratory tests) and possibly other locations in the southern United States supporting the possibility that TYLCV will become a regional problem.

Due, apparently, to the introduction and spread of the silver leaf whitefly, there has been a recent emergence of seventeen whitefly-transmitted geminiviruses in tomato in the Western Hemis phere. Of these, only tomato mottle virus was present in Florida before TYLCV. Polymerase chain reaction (PCR) and DNA sequence analyses were used to confirm the recent introduction of TYLCV into Florida. Degenerate primers for geminiviruses were used to amplify a fragment of the viral genome containing part of the coat protein gene. The partial gene sequence had greater than 98% homology with that of an Israel strain of the TYLCV. These results suggest that the virus is an Eastern Mediterranean strain and possibly the same strain previously introduced into the Dominican Republic from Israel in 1991 and subsequently found in Jamaica and Cuba. We have designed non-degenerate primers for PCR detection of TYLCV based on our DNA sequence data for the virus. Detection of TYLCV in tomato plants with these primers has been 10-100 times more sensitive than with the degenerate primers, which even with the degeneracy have mismatched bases.

Tomatoes are a winter crop in south Florida, and TYLCV was detected in October 1997, in newly established commercial plantings. Tomatoes are still being planted in south Florida at the present time. The incidence of TYLCV-infected plants initially appeared to be due mostly to primary spread into the fields. Sources of inoculumare presumed to be weeds outside of the tomato fields. Silverleaf whitefly (*Bemisia argentifolii*) population densities have been generally low, and even less in tomatoes due to the extensive application of the systemic insecticide, imidacloprid (Admire), to transplants. However, the occurrence and incidence of TYLCV in cultivated tomatoes have steadily increased during the growing season. Secondary spread of TYLCV within fields has become more prevalent and Silverleaf whitefly population densities are on the increase. TYLCV appears to be firmly established in Florida, and will likely become a major problem in the region in years to come.

Recent field studies with combinations of Admire at 8 oz. and Neemix at 4 oz. have reduced the whitefly populations, as well as the incidence of TYLCV and improved tomato yields. Also, the combination of Admire at 8 oz. plus Knack at 8 oz. plus Applaud at 8 oz., reduced whitefly populations, incidence of TYLCV and improved tomato yields.

Investigator's Names: Steven E. Naranjo<sup>1</sup>, Peter C. Ellsworth<sup>2</sup>, C. C. Chu<sup>1</sup> & Thomas J. Henneberry<sup>1</sup>

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**Research & Implementation Area:** Section F: Integrated and Areawide Pest Management Approaches, and Crop Management Systems

Dates Covered by the Report: January 2000- December 2001

## Conservation of Predatory Arthropods in Cotton: Role of Action Thresholds for Bemisia tabaci

Studies were conducted in 1994 and 1995 to measure the effects of prescriptive insecticide use for Bemisia tabaci in cotton on populations of arthropod predators in Imperial Valley, California and Maricopa, Arizona. Replicated plots were sprayed with a pyrethroid + organophosphate mixture when densities of adult *B. tabaci* exceeded 2.5, 5, 10, or 20 per leaf. Untreated plots served as controls. Application of insecticides significantly reduced population densities of spiders, *Geocoris punctipes* (Say), G. pallens (Stål), Orius tristicolor (White), Nabis alternatus Parshley, Zelus renardii Kolenati, Hippodamia convergens Guérin-Méneville. Spanogonicus albofasciatus (Reuter), Drapetis sp., and Chrysoperla carnea Stephens in one or both years and sites compared to untreated controls. Use of higher B. tabaci thresholds conserved some species and groups relative to lower thresholds. Stepwise regression analyses indicated that reductions in predator populations were generally influenced more strongly by the timing of the first insecticide application than by the total number of sprays necessary to maintain suppression of the pest below any given action threshold. A predation index, which weights the importance of predator species based on their known frequency of predation on B. tabaci and another key pest, Pectinophora gossypiella (Saunders), was developed and analyzed. General patterns were similar to results based on changes in abundance alone, but the index generally revealed less severe effects of insecticides on overall predator function. The current action threshold for conventional insecticidal control of B. tabaci in Arizona and southern California is 5 adults per leaf. Results here suggest that predator conservation may be enhanced by raising the initial threshold to delay the first application or initially using more selective materials such as insect growth regulators.

## Investigator's Name(s): Yash Pal S. Rathi

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**Research & Implementation Area**: Section F. Integrated and Areawide Pest Management Approaches, and Crop Management Systems

## Dates Covered by Report: 1999-2000

## Management Of Mungbean Yellow Mosaic Virus, A Yellow Plague Of Kharif Grain Legumes

Mungbean yellow mosaic virus (MYMV) also called as a "yellow plague" of *Kharif* grain legumes is transmitted by whitefly (*Bemisia tabaci*) in persistent circular manner. Currently, it is a number one problem causing substantial yield losses in urdbean (*Vigna mungo*), mungbean (*V. radiata*), mothbean (*V: aconotifolia*), french bean (*Phaseolus vulgaris*], soybean (*Glycine max*) and pigeonpea (*Cajanus cajan*) in India. The host crop exhibit general yellowing and the diseased fields can be recognized from far distance.

Experiments were conducted at Crop Research Centre of Pantnagar University to contain this disease through agronomic practices (manipulation in planting dates and inter-cropping with barrier (non-host) crops, insecticides (foliar and soil application, seed treatment) and host resistance. Low disease incidence was recorded in early (June} and late (August) plantings as well as in low plant spacing (5 cm). Seed dressing with Carbofuran 3G or Phorate 10G with two foliar sprays of Monocrotophos (0.1%) + Endosulphan (0.1%) also reduced the disease incidence. Use of barrier crops used as inter-cropping with mungbean, soybean and urdbean were not very effective. Some genotypes viz., Pant U19, 30 and 35, (urdbean) PM 1, 2, 3 and 4 (mungbeari) and PK 416, PK564, SL 142 (soybean) resistant to MYMV have been identified.

As none of the individual control measure was found highly effective against the disease due to high vector population and wide range of host plants serving as initial foci of the whitefly and the MYMV in this area, integrated management modules have been developed keeping the tolerant varieties as core.

#### **Section F Research Summary**

## Integrated and Areawide Pest Management Approaches And Crop Management Systems

Compiled by Steve Castle

Ten years of whitefly meetings and what do you have to show for it? *A well-managed pest*.

The story should end so simply, unfettered by dreary pontification about advances in knowledge and changes in practices that brought about quantum improvements in how we coexist with Bemisia tabaci, Bemisia argentifolii, or just plain Bemisia. But if simple statements don't suffice in a world obsessed with post facto analysis, then let it be stated that significant achievements were realized in each of the 6 sections that comprised the 5-Year Plans. The crowning achievement was that an outbreak pest has now been subdued through multi-disciplined contributions distilled and synthesized into viable control programs. Every abstract and presentation, coffee-break conversation and skeptical thought, every right idea and wrong direction contributed to the process of working out a solution to a devastating problem. But to focus only on the practical successes on the farm would be an injustice to the countless successes attained at more fundamental levels of investigation. Significant advancements were made in our understanding of *B. tabaci* as an organism with a complex genetics that affect host range, virus transmission, insecticide resistance and undoubtedly many other interactions with its biotic and abiotic environment. We gained valuable knowledge of how B. tabaci adapts physiologically to a hostile thermal environment, how developmental morphology proceeds temporally, how mouthpart anatomy influences feeding biology, how feeding biology influences virus transmission, and how virus transmission can be influenced by plant host and cultivar differences. The listing could go on at great length, but this would be redundant as all abstracts have been included in each annual edition of the 5-Year Plan Reviews. The combined set of Reviews serves as a testimonial to the breadth of work initiated in response to the outbreak crises, as well as to the power of an organized and collaborative approach to solving monumental pest management problems.

Unfortunately, some problems proved more solvable than others, and universal agreement over taxonomic identification happens to be among the others. But so far as Section F is concerned, correct taxonomy will always be more of an academic issue than a practical problem. What is done in the field to control whitefly populations will likely remain unchanged whether the target population is comprised of Type A, B, or Q, or a combination of all 3. Some might argue that the

particular biotype involved may have unique characteristics such as a particular insecticide resistance profile that should be factored into the choice of chemical treatment. Or perhaps there might be a host biotype preference among parasitoids as some considered to be a possibility in the shift from Type A to B that occurred in North America. Although differences among biotypes with respect to insecticide susceptibilities may exist, they also exist between populations within a biotype. Insecticide resistance profiles would unlikely ever become so specific and universal within a biotype as to be recognized as a characteristic and a basis of decisionmaking. On the other hand, biotype preferences could very well exist in certain parasitoids principally due to the interaction of the primary and tertiary trophic levels. Foraging parasitoids may be less likely to search and/or accept a whitefly host on one particular plant species compared to another. Therefore, variations in host plant ranges among *B. tabaci* biotypes could alter the efficiency of parasitoids that exhibit putative host plant preferences. However, biocontrol workers have a long way to go to first demonstrate that such a scenario occurs, then validate that it has any practical significance to the quality of management. Meanwhile, sound IPM practices tailored to a particular crop system that produce sterling results against 1 biotype will almost certainly produce the same results against a different biotype.

Every growing region across North America that struggled with whitefly control in the late 1980s and early 1990s benefited by the addition of far more effective chemical tools beginning in the early 1990s. While the potency, residual activity, and in many cases the specificity of these new tools are unparalleled, so too was the collective research that revealed not only their practical utility, but how and when the newer tools should be applied and incorporated into a coordinated control program. Sampling plans for whitefly in cotton and melons were developed and joined with action thresholds to produce highly effective control, especially in places where their implementation was preceded by extensive communication to, and education of, grower clientele. Nowhere else was the sampling and threshold foundation along with product label and IRM guidelines better developed than in Arizona. Industry representatives worked closely with Arizona scientists to construct product labels that limited per field use of pyriproxyfen and buprofezin to 1 application each per cotton season and mandated that purchasers of either product be certified through attendance at a training session. Nowhere else were education and training sessions in addition to cohesive guidelines, web site, and printed reference materials more skillfully delivered to clientele and subsequently implemented than in Arizona. The result of this expert blending of research and transfer has been an extraordinarily successful suppression of whitefly populations ever since 1996. That was the first year that the IGRs became available commercially and the strategy for their implementation was formalized into a pest management framework. The overall effect of this combined action has been progressive decreases in whitefly pressure each year and a concomitant drop in insecticide applications that target whiteflies.

In addition to the aforementioned factors that contributed to the success of the Arizona IPM program for cotton, there are other intangible factors that have possibly played significant roles in the decline of whitefly numbers. Certainly among these would be to what extent buprofezin and pyriproxyfen treatments act across developmental stages of whiteflies to produce their full effects on a population. Product descriptions may actually underestimate the full impact that each compound has when various stages are exposed in the field, including a number of sub-lethal effects on adults that occur beyond the well-recognized sterility effect produced in females by pyriproxyfen. Another factor that has begun to be quantified is the degree to which natural enemy populations are left intact following treatment with either of the IGRs, and what level of bio-residual activity (sensu Ellsworth and Naranjo) continues to act against whiteflies and other pest populations. Still another intangible is what role education and training sessions of growers and advisors played in leading to more effective area-wide control. More synchronous insecticide applications over wide areas may have been an outcome of the emphasis placed on timing the first IGR application according to whether or not the prescribed action threshold had been reached. The upshot of the thorough research that produced sampling plans and action thresholds, in addition to the outstanding communication and transfer of this information to the grower clientele, is that what was essentially delivered as an individualized, grower-by-grower program with the ultimate purpose of protecting each one's livelihood, soon transcended into a larger phenomenon that saw individual growers benefiting not only by their own knowledge-driven actions, but by the synchronous actions of all of their peers acting on the same knowledge. Rather than disjunct applications made in a patchwork manner over a region prior to 1996 (not having had the benefit of disciplined decision-making instilled by a coherent IPM framework), treatments against whiteflies beginning in 1996 instead became a de facto area-wide program of synchronous applications that yielded blanket-like suppression of whitefly populations.

The transformation of pest management practices that took place in Arizona beginning in 1996 makes an interesting study of how a combination of crisis and novelty can sublimate ordinary pest management into something extraordinary. Pest management as it was being practiced in Arizona prior to 1996 was anything but ordinary because sampling plans, action thresholds, and a cotton IRM had been under development for years and were all in place as a cohesive set of recommendations to growers by 1995. However, the impact these recommendations had on whitefly management in 1995 was still deficient as control of late-season generations was lost, resulting in poor cotton quality and yields. Among the repercussions of the 1995 whitefly crisis was a pervasive realization that another way had to be found to manage whiteflies in cotton. Fortunately, years of research by Arizona and Florida scientists had identified buprofezin and pyriproxyfen as highly effective compounds with unique modes of action. The volumes of data accumulated made it possible to rapidly put together a strong case for an emergency registration of both compounds. The package submitted for registration also benefited by the example in Israel where both compounds had been used effectively to help control whitefly populations. A tough resistance management component that precluded use of either compound more than once per season and mandated at least a 14-day separation between use of the IGRs was built into the package as well.

Where the novelty aspect enters into the formula that transformed whitefly pest management is in the nature of each compound's activity against whitefly populations and the special instructions required to maximize each product's potential. The unique, slow-acting properties of each product could well be deceptive to novice users otherwise accustomed to rapid knockdown by conventional insecticides. Increasing grower awareness at seminars and training sessions held at multiple locations throughout Arizona was an essential component to the successful use of the IGRs. During these sessions. emphasis was also placed on the critical timing required to produce the best possible results, and how sampling was the key to correct timing. The crisis of the previous year produced a sense of urgency to the process of adapting to novel products, and the novelty of the products demanded their deployment under the best IPM practices. Had some other pair of insecticides with more conventional activities been introduced, it is possible that growers would not have been mandated to attend training sessions in order to become certified to use the products. Their expertise would already have been in the use of conventional-acting products, and would therefore not have required the specialized training accompanied by an emphasis on using them with accurate observance of action thresholds. Or if they had, the same important message of employing the recommended sampling program and treating at the right threshold would have been given, as it had been previously in 1995, but it would have fallen too well within their comfort zone of operation so long as they had only to be concerned about new products that acted the same as all previous products. I submit that the novelty of the IGRs altered the sprayand-knockdown culture to such a degree that, in combination with the crisis mode still pervasive from the previous year's disaster, growers and PCAs became more receptive to the teachings of IPM than ever before. The product of this grower education, mandated collaboratively by company representatives for both products and by University of Arizona scientists that recognized the need for grower training through their own research experience, was a sensational turnaround in the first year that has been sustained ever since.

While other regions in North America that had previously suffered under whitefly attack also benefited by collective advances in whitefly management, none developed coordinated programs as sophisticated as the one in Arizona. Early on, whitefly outbreaks had flourished from one region to another, but there were no satisfactory management solutions because the necessary tools were as yet underdeveloped. A few more years of experience in whitefly management, some more time to develop the basic tools of IPM, and perhaps the next outbreak situation would be suppressed. That opportunity arose in 1995 in Arizona at a time when all of the components of good IPM were in place and in practice, yet still were inadequate against late-season whiteflies in cotton. It was this latter-day crisis, however, that spawned the search for a reliable solution to the one-too-many outbreaks at the propitious time that groundwork for a novel solution had already been completed. The combination of events resulted in the Arizona cotton IPM for whitefly that has few equals in the realm of IPM solutions in terms of the quality of research, education, and implementation that have contributed to its sustaining success.

## Development, Integration, and Delivery and Implementation

Progress was made in each one of the research areas that subdivided Section F (see Table F synopsis). Development in whitefly-crop interactions has continued to advance with life history studies being conducted throughout the year in various crops and ornamental hosts. The cumulative data from these studies and ones conducted previously in cotton will provide enormously valuable information on the population dynamics of B. *tabaci* as well as on the relative efficiencies of various management tactics. Development of heuristic frameworks describing the interacting strategies and components of IPM in one case, and in another the factors that contribute to whitefly outbreaks, helped synthesize existing and new information into conceptual models that advanced understanding of why outbreaks occur and how management can most effectively respond. Most impressively, many of the concepts contained in the IPM Pyramid have been validated by real world examples where implementation has brought about management successes. In addition to the case already described for Arizona cotton IPM, a coordinated program was instituted in the Dominican Republic to counter epidemics of tomato yellow leaf curl virus (TYLCV) in production tomato fields. By implementing a mandatory crop free period to reduce inoculum potential, incidence of TYLCV in tomatoes has greatly declined while production has fully recovered. This is an example of avoiding a pest or disease problem by manipulating crop plantings and seasons as spelled out in the Avoidance layer of the IPM Pyramid.

Further refinement in the implementation of pest management will continue to improve existing practices. For example, reassessment of the data that contributed to setting the action threshold at 5 adults/leaf showed that predator conservation increased with higher thresholds or if the date of first application was delayed. Improved integration of natural and chemical controls will occur as a more complete picture develops over the consequences of applying inputs to agricultural systems. In this case, slight restraint on the timing of the first application may pay dividends in the form of greater natural control and reduced pesticide use overall. On a wider scale, integration of chemical classes within and across crop seasons is a goal of cross-commodity efforts to conserve insecticide efficacies. This becomes more of a challenge as insecticides gain registrations on additional crops. The most pressing concern is that multiple uses in multiple crops will add up to too many uses on a pest that moves sequentially from one crop to another. Resistance selection pressure can be expected to increase if exposure levels increase with multiple-crop usage of a particular insecticide or insecticide class. However, much progress is being made in Arizona to harmonize insecticide use across commodities so as to limit maximum exposures to any one product or insecticide class.

		Progress Achieved	
Research Approaches <sup>a</sup>	Year 1 Goals Statement	Yes No	Significance
Development:			
Study whitefly-crop interactions <sup>b</sup> as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/termination/harvest dates, other farm practices, intercrop relationships.	Identify potential beneficial or exacerbating farm practices or inputs for testing.	X but limited	Only minor progress has been made on this approach (since last 5-yr review), & mainly in area-wide programs. This work is correlative, & little experimental work has been planned for or reported. Past work identified the potential or described the role of fertility status, water-stress & some other agronomic factors on <i>Bemisia</i> population dynamics. Conceptual discussion was presented on the role of pesticidal & non-pesticidal factors on <i>Bemisia</i> outbreaks.
Develop behavioral barriers <sup>b</sup> to whitefly colonization and population development, e.g., mulches, trap crops, inter- cropping, row covers, etc.	Review potential behavioral disrupters and evaluate as potential IPM components.	Х	<ul> <li>Progress has been made in several areas:</li> <li>row covers and screens as physical barriers,</li> <li>mulches and oils as behavioral barriers,</li> <li>living mulches as behavioral barriers.</li> </ul>
Integration:			
Develop Integrated Pest Management <sup>c</sup> systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	Identify candidate dual or multiple control tactic systems, e.g., IGRs and natural enemy conservation.	Х	<ul> <li>Significant activity on this goal has occurred:</li> <li>Insect Growth Regulators &amp; biological control in cotton (conservation)</li> <li>imidacloprid &amp; other chemical control tactics &amp; various forms of biological control, especially in vegetables</li> <li>studies of direct &amp; indirect effects of chemical control on bio-control agents.</li> </ul>
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision- making, biological control, etc.	Develop or modify sampling systems for new crops; integrate with thresholds and decision-making.	Х	<ul> <li>Limited progress has been made in this area:</li> <li><i>Bemisia</i> distributions have been examined in tomato,</li> <li>new binomial sampling system for large nymphs in cotton, &amp; integration with thresholds for IGR decisions</li> <li>sampling &amp; IGR re-treatment decisions tested in</li> </ul>

cotton.

## Table F. Integrated and Areawide Pest Management Approaches, and Crop Management Systems.

Research Approaches <sup>a</sup>	Year 1 Goals Statement	Progress Achieved Yes No	Significance
Delivery and Implementation:			
Elevate single field/farm practices to areawide community-based contexts; develop methodology for installing and evaluating areawide control technologies and their impact.	Identify agricultural communities amenable to areawide management; conduct thorough pre-implementation evaluation.	Х	<ul> <li>Significant progress was made in this area mainly in cotton:</li> <li>areas dominated by cotton were identified in AZ &amp; CA for implementation of cooperative programs.</li> <li>areas of melon and vegetable production were identified in TX for potential area-wide programs.</li> <li>area-wide samp ling, &amp; decision-making was the main focus of most programs; however, coordinated natural enemy releases were also conducted.</li> </ul>
Implement and deliver Integrated Pest Management and Integrated Crop Management systems or system components to clientele.	Develop and distribute provisional IPM & ICM recommendations.	Х	<ul> <li>Continued progress was made in this area:</li> <li>IPM recommendations were distributed AZ, CA, Mexico &amp; FL; bilateral discussions between Brazil &amp; U.S. took place.</li> <li>IPM &amp; ICM guidelines were coordinated in AZ cotton.</li> </ul>

<sup>a</sup> See Tables A to E for additional complementary research.
 <sup>b</sup> See Tables A for additional complementary research.
 <sup>c</sup> See Tables E for additional complementary research.

Research Approaches <sup>a</sup>	Year 2 Goals Statement	Progress Achieved Yes No	Significance
Development:			
Study whitefly-crop interactions <sup>b</sup> as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/termination/harvest dates, other farm practices, intercrop relationships.	Determine nature and character of relationship between interaction and whitefly population dynamics.	Х	Nitrogen fertilization at different rates in cotton and its impact on whitefly population densities and honeydew deposition was studied. Considerable development occurred on cross-commodity integration of pesticides used in multi-cropped situations and in conceptualization of the multiple levels and factors upon which whitefly management depends.
Develop behavioral barriers <sup>b</sup> to whitefly colonization and population development, e.g., mulches, trap crops, inter- cropping, row covers, etc.	Conduct field-level trials; quantify impact to crop and whitefly dynamics	Х	Investigations on intercropping took place in both desert and tropical environments. Although reductions in whitefly densities were observed in both systems, further experimentation is required to establish the effectiveness of the trap crops relative to more conventional management techniques.
Integration:			
Develop Integrated Pest Management <sup>c</sup> systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	Initiate field testing of candidate systems.	Χ	A number of field studies employed multiple tactics directed against whitefly populations. Biorational insecticides were examined in combination with IGRs and other biopesticidal agents such as <i>Beauvaria bassiana</i> for control efficacy of silverleaf whitefly. There was an indication of inhibitory action by <i>B. bassiana</i> when used in combination with imidacloprid as well as deleterious effects to predators contacted by <i>B. bassiana</i> treatments. Neem products were used to reduce whitefly populations and incidence of yellow mosaic virus in India. A melon trap crop was integrated with chemical control to focus potentially disrupting treatments into a limited area while preserving natural mortality factors in cotton as the principle crop.

Research Approaches <sup>a</sup>	Year 2 Goals Statement	Progress Achieved Yes No	Significance
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision- making, biological control, etc.	Establish practical utility of system through economic analyses; field efficiencies and costs.	X	Analysis of types and patterns of chemical treatments made on a large number of cotton fields in central Arizona over a 4 year period revealed extraordinary differences in the number of treatments and amount of time that whiteflies exceeded threshold levels prior to and following the advent of the IGRs buprofezin and pyriproxyfen. The proactive initiative taken by Arizona growers to pursue chemical use harmonization across commodities required consideration of all aspects of pest and crop management. A similar whole system appraisal was made in the San Joaquin Valley with an emphasis on integrating multiple practices with diverse insecticide classes as part of an insecticide resistance management program.
Delivery and Implementation: Elevate single field/farm practices to areawide community-based contexts; develop methodology for installing and evaluating areawide control technologies and their impact.	Install control technologies into community; develop systems for evaluation.	Х	Large areas in the San Joaquin Valley observed specific guidelines for IPM and IRM in cotton with evaluations continuing on the benefits attained over areas that did not observe these guidelines. Community wide evaluations were made on quality of whitefly management according to chemical control practices. The successful IPM and IRM programs practiced in Arizona cotton continued for a third consecutive year. Further cross-commodity develoment of these programs is under way.

		Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 2 Goals Statement	Yes	No	Significance
Research Approaches <sup>a</sup> Implement and deliver Integrated Pest Management and Integrated Crop Management systems or system components to clientele.	Year 2 Goals Statement Conduct whole farm/operation demonstrations of IPM systems.	Yes X	No	Significance A 'best agricultural practices' demonstration project was conducted on 50.5 acres at the University of Arizona Maricopa Agricultural Center that included inputs from extension specialists in agronomy, entomology, irrigation management, weed sciences and plant pathology according to university recommendations. Whitefly management was fully integrated with management of other insect pests and required only a single application of pyriproxyfen. Lint yields of 2.81 bales/acre were higher than the historical as well as the 1998 farm-wide average. An integrated areawide management program involving the cooperation of growers, PCAs, ginners and state and university researchers was expanded during a
				second year in the San Joaquin Valley.

<sup>a</sup> See Tables A to E for additional complementary research.
 <sup>b</sup> See Tables A for additional complementary research.
 <sup>c</sup> See Tables E for additional complementary research.

Research Approaches <sup>a</sup>	Year 3 Goals Statement	Progress Achieved Yes No	Significance	
Development:				
Study whitefly-crop interactions <sup>b</sup> as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/termination/harvest dates, other farm practices, intercrop relationships.	Identify mechanisms governing relationship and alter or manipulate factors that suppress whitefly dynamics.	Х	Progress was made with studies in Texas on seasonal dynamics of <i>Bemisia</i> on spring collards, and the impact of cotton defoliants on <i>Bemisia</i> and parasitoid populations. Work continued in California on the affect of various nitrogen levels in cotton with <i>Bemisia</i> population densities	
Develop behavioral barriers <sup>b</sup> to whitefly colonization and population development, e.g., mulches, trap crops, inter- cropping, row covers, etc.	Apply promising technologies to high-value crop systems; field test and evaluate	Х	<ul> <li>Further progress with research on:</li> <li>Living ground covers for managing whitefly- transmitted gemini viruses</li> <li>Behavioral disruptance by UV-blocking barriers</li> <li>Melons as a trap crop for cotton</li> </ul>	
Integration:				
Develop Integrated Pest Management <sup>c</sup> systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	Continue field testing & evaluate feasibility of large scale testing; add components as necessary.	Χ	<ul> <li>Much progress with integrating control tactics:</li> <li>Life table evaluation of both natural and insecticide-based mortalities</li> <li>Compatibility of IGR's for whitefly control in greenhouses</li> <li>IPM development in cotton for <i>Bemisia</i> and other cotton pests</li> <li>Augmentative biocontrol using crop transplants inoculated with parasitoids in Admire<sup>®</sup>-treated fields</li> </ul>	
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision- making, biological control, etc.	Integrate additional control components into sampling, threshold & decision-making systems	Х	<ul> <li>Fourth consecutive year of monitoring <i>Bemisia</i> populations in the Imperial Valley using the CC trap.</li> <li>Sampling-based refinement of action thresholds for IGR's in Arizona cotton</li> </ul>	

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Research Approaches <sup>a</sup>	Year 3 Goals Statement	Progress Achieved Yes No	Significance
Delivery and Implementation: Elevate single field/farm practices to areawide community-based contexts; dewlop methodology for installing and evaluating areawide control technologies and their impact.	Identify additional IPM/ICM compatible components. Re -assess and adapt program. Conduct areawide economic analyses.		<ul> <li>Economic analysis of the use of IGR's in Arizona cotton</li> <li>Further development of cross-commodity planning and cooperation</li> <li>Multivariate techniques used to assess parasitoid establishment in the Imperial Valley based on</li> </ul>
Implement and deliver Integrated Pest Management and Integrated Crop Management systems or system components to clientele.	Expand sites of testing with grower cooperators; conduct validation studies.		<ul> <li>crop and land use</li> <li>Sticky cotton bulletin published by University of Arizona and Cotton Incorporated</li> <li>International development of IPM for managing whiteflies and geminiviruses</li> <li>Development of <i>Bemisia</i>-resistant alfalfa cultivars</li> <li>Crop and pest management demonstration project on cotton in Arizona</li> </ul>
<ul> <li><sup>a</sup> See Tables A to E for additional con</li> <li><sup>b</sup> See Tables A for additional compler</li> <li><sup>c</sup> See Tables E for additional complen</li> </ul>	nentary research.		* *

		Progress Ach	
Research Approaches <sup>a</sup>	Year 4-5 Goals Statement	Yes No	D Significance
Development: Study whitefly-crop interactions <sup>b</sup> as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/termination/harvest dates, other farm practices, intercrop relationships.	Refine system, add other compatible components, evaluate economic impact; conduct field testing and evaluations. Conduct economic analyses and determine next level of IPM/ICM systems evaluation. Develop recommendations of best management practices.	X	Significant progress made in the realm of intercrop relationships with comparative life history studies of <i>B. tabaci</i> on multiple crop and non-crop hosts. An economic impact analysis was developed for IGR use in Arizona as well as for pesticide use in general. The IPM Pyramid was advanced as a conceptual framework for understanding the interacting components in an IPM program for whiteflies. The Outbreak Pyramid was advanced as a conceptual device for understanding causes of outbreaks. Avoidance as a major component of IPM along with conservation of natural enemies through selective chemistry were advanced as topics and supported by research. A crop-free period was pivotal in breaking epidemics of TYLCV in the Dominican Republic was elucidated, but so too the role of symptomless weeds in perpetuating the virus.
Develop behavi oral barriers <sup>b</sup> to whitefly colonization and population development, e.g., mulches, trap crops, inter- cropping, row covers, etc.	Refine system, add other components, and conduct economic feasibility analyses. Summarize and evaluate results; prepare crop systems -specific recommendations.	Х	Synthesis of field exclusion techniques published as a review article. Cost-benefit analysis of insect- exclusion screens used on greenhouses in Israel demonstrating benefit for the grower and consumer.
Integration : Develop Integrated Pest Management <sup>c</sup> systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	Initiate large-scale experiments; incorporate economic evaluation. Evaluate multiple component system as potential deliverable; prepare recommendations.	х	Large plot studies conducted in Arizona demonstrated marginal mortalities associated with different control tactics or through various natural sources. Evaluation of parasitoid releases in concert with imidacloprid treatment of spring melons. Recommendations prepared and delivered in Mexico's Yaqui Valley and in the Dominican Republic.

## Table F. Integrated and Areawide Pest Management Approaches, and Crop Management Systems, 2001-2002.

		Progress A	Achieved	
Research Approaches <sup>a</sup>	Year 5 Goals Statement	Yes	No	Significance
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision- making, biological control, etc.	Evaluate in whole field systems. Identify weaknesses; target improvements. Evaluate redesigned decision systems; continue field testing and economic analyses	X		Further analysis of sampling/action threshold database that demonstrates greater conservation of predator species when the initial treatment threshold of 10 adults/leaf is used rather than the standard of 5 adults/leaf.
Delivery and Implementation: Elevate single field/farm practices to areawide community-based contexts; develop methodology for installing and evaluating areawide control technologies and their impact.	Formulate clientele surveys; develop & begin to implement protocols for evaluating areawide technologies. Refine, reevaluate and identify weaknesses. Formulate recommendations for future areawide management systems. Conduct surveys.	Х		Pesticide use information in Arizona as a validation of improved pest management; in California, pesticide use incorporated into GIS analyses to evaluate whitefly distributions. Cross-commodity efforts to harmonize pesticide use and to identify weaknesses in cross registration of neonicotinoids and IGRs.
Implement and deliver Integrated Pest Management and Integrated Crop Management systems or system components to clientele.	Incorporate new information and economics into recommendations. Validate new components; finalize recommendations; expand to new crops.	X		Development of web sites as whitefly management resources for clientele to access and download. Crop free period in Dominican Republic and recommendations for dealing with symptomless weed hosts, beans and peppers.

<sup>a</sup> See Tables A to E for additional complementary research.
 <sup>b</sup> See Tables A for additional complementary research.
 <sup>c</sup> See Tables E for additional complementary research.

## Appendix A: Selected National and International Bemisia Research and Information Coordination

## Selected National and International Bemisia Research Projects and Information Coordination

Several national and international cooperative *Bemisia* projects have been implemented worldwide to facilitate communication, research, technology transfer, and rapid response to problem solving (Appendix IX, Table 1). Additional information on these efforts is available on the listed websites. The list is not all inclusive, and these are numerous local groups with commodity-orientation that meet informally to exchange information and experiences, and discuss current control efforts. It does, however, serve as a starting point for identifying sources of whitefly information and contacts to gain additional information.

These international, regional and national networks for *B. tabaci* and *B. tabaci*-transmitted plant viruses and other whitefly species essentially function as information exchanges. The active international and national collaborative research projects have improved operational efficiency, by optimizing returns from financial, physical and human resources. Open lines of communication and discussion identified priority research areas and focused efforts that increased returns for each invested research dollar compared to numerous small projects utilizing diverse approaches. The economic, environmental and social impact of research coordination and information exchange groups for the *B. tabaci* species complex worldwide have stimulated much new and useful research resulting in advanced knowledge of biology, ecology, physiology, and virus-vector interactions of the species complex.

	Year	
Project	Created	Accomplishments
International Bemisia	1984	Information exchange, meeting announcements, electronic conference,
Newsletter		Whitefly-L. Coordinators:
		D. Gerling <u>dangr@ccsg.tam.ac.il</u> , W. Jones <u>wjones@weslaco.ars.usda.gov</u>
Sticky Cotton Action Team, Cotton Incorporated (CI), Raleigh, NC	1990	Provided communication between the cotton industry and researchers on sticky cotton research. CI and state support committees provided \$1.9 million for research. Coordinated multi-federal, -state, -university, and -industry agencies efforts that resulted in efficient whitefly management and reduced sticky cotton problems. Financed research also contributed to implementation of insect growth regulator use patterns and resistance management. Also, stimulated improved sticky cotton lint sampling methods, demonstration of the relationships of whiteflies and aphids to sticky cotton, improved sticky cotton measurement instrumentation and enzyme approaches to eliminate honeydew sugars from contaminated lint. Coordinators: R.L. Nichols <u>BNichols@cottoninc.com</u> , W. Lalor <u>BLabr@cottoninc.com</u>
Latin America and Carribean Whitefly and Geminivirus Network	1992	Quarterly newsletter, Mosca Blanca al Dia, <u>www.catie.ac.cr/cooperacion/boletin.htm</u> Promotes researh, diagnostic activities, and IPM technology transfer in 21 countries including Spain. Annual meeting. Network host: Tropical Agricultural Research and Higher Education Center (CATIE) Costa Rica. Coordinator: L. Hilje, <u>lhilje@catie.ac.cr</u>
Integrated Management of Whitefly in Northwestern Mexico	1992	Identified soybeans as the key host and eliminated it in the cropping system to break the reproductive host continuity. Published eight scientific summaries of 262 articles. Covered 4 states, Baja California Norte, Baja California Sur, Sonora and Sinola. Participating agencies are the National Research Institute of Forestry, Agriculture and Animal Husbandry (INIFAP), Regional Centers for Agricultural Research and Mexicali Experiment Station. Coordinators: J. L. Martinez Carrillo jlmc@cirno.inifap.conacyt.mx, J. Jose Pacheco Pacheco@cirno.inifap.conacyt.mx

## Table 1. Selected national and international Bemisia tabaci research and information exchange projects.

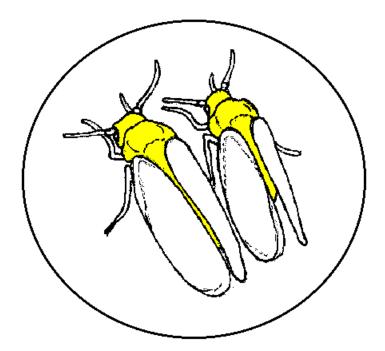
Five-Year National Research and Action Plan for Development of Management and Control Methodology for the Sweetpotato Whitefly. Revised as Five-Year Silverleaf Whitefly Research, Action and Technology Transfer Plan, in 1997.	1992-1996 1997-2001	National and international cooperative programs that led to action thresholds, effective chemical control, new chemistry and resistance management principles and agricultural community management systems. Extensive basic biology, physiology, ecology for long-term suppression systems (see text). Annual meetings. Nine annual publications with over 1000 abstracts of research progress. A bibliography of more than 4,800 entries: <u>http://www.wcrl.ars.usda.gov</u> Coordinators: T. J. Henneberry <u>thenneberry@wcrl.ars.usda.gov</u> , N. Toscano <u>ntoscano@ucrac1.ucr.edu</u> , W. Jones, T. Perring <u>perring@ucrac1.ucr.edu</u> , R. Faust <u>rmf@ars.usda.gov</u>
The Safe and Effective Use of Pesticides Project	1995	USAID-funded project carried out under the umbrella of the Middle East Peace Process. Annual meetings of IPM and public health experts in Egypt, Jordan, Israel and Palestine for information exchange. Pest monitoring, training of farm workers, blood testing, reports on safety progress. Information <u>www.mercph.org</u> Coordinating Center: National Institute of Health (NIH), Bethesda, MD. Coordinator: K. Abdo <u>abdok@niehs.nih.gov</u>
CGIAR Whitefly IPM Project	1996	Consultative Group for International Agricultural Research (CGIAR) project, under the umbrella of the Systemwide Program for Integrated Pest Management (see text). Coordinating Center: International Center for Tropical Agriculture, Cali, Colombia. Coordinator: P. Anderson <u>p.anderson@cgiar.org</u>
European Whitefly Studies Network	1997	Gathered and disseminated whitefly information. Working group meetings. Established website: <u>www.jic.bbsrc.ac.uk/hosting/eu/ewsn</u> Network host: John Innes Center, United Kingdom. Coordinator: Ian Bedford <u>ian.Bedford@bbsrc.ac.uk</u>
Brazilian Agricultural Research Enterprise (EMBRAPA) Project (1998-2000)	1998	National Whitefly Commission by EMBRAPA (3 year project). Identified priorities, coordinated efforts of the ministry, federal, state, agricultural agencies and growers. Identified infested areas, identified effective insecticides, transferred technology from other countries. Coordinator: MRV Oliveira <u>vilarin@cenargen.embrapa.br</u>
Brazilian virology project to characterize geminiviruses tomatoes	1998	Characterized 8 new tomato-infecting geminiviruses in Brazil. Developed standardized virus survey protocols, shared results, initiated varietal resistance efforts. Coordinators: F. Murilo Zerbini <u>zerbini@mail.ufv.br</u> , S.G. Ribeiro <u>simone@cenargen.embrapa.br</u>

<sup>a</sup> This table was reproduced, with the permission of the author from the publication "History, Current Status and Collaborative Research Projects for *Bemisia tabaci* by Oliveira, M.R.V., T. J. Henneberry and P. Anderson. 2001. Crop Protection 20: 709-723.

Appendix B. Bibiolography of *Bemisia tabaci* (Gennadius) & *Bemisia argentifolii* Bellows and Perring

Complete Bibliography of

# Bemisia tabaci (Gennadius) & Bemisia argentifolii Bellows and Perring



Steven E. Naranjo George D. Butler, Jr. Thomas J. Henneberry

February 2002

## Bibliography of Bemisia tabaci (Gennadius) and Bemisia argentifolii Bellows & Perring

## February 2002

Steven E. Naranjo, George D. Butler, Jr., and Thomas J. Henneberry

*Bemisia tabaci* (Gennadius) was described over 100 years ago as a tobacco pest in Greece and has since become one of the most important pests of food, fiber and ornamental crops in the world. The recent geographic expansion of this pest has been closely associated with a new and more virulent biotype, known widely as biotype B, that may represent a new species (Bemisia argentifolii Bellows and Perring). The taxonomy and systematics of this pest remains confused and controversial. Recent analyses, using a wide array of morphological and molecular tools, suggest that *B. tabaci* may represent a species complex. Regardless, this insect has had an extraordinary impact on agricultural and horticultural production systems worldwide. In addition to direct feeding damage, this insect vectors a number of devastating plant viruses, causes debilitating plant disorders of unknown etiology and, by the excretion of honeydew, reduces the quality of harvested products. We face monumental challenges in the development of economically-efficient and environmentally-sound management systems for Bemisia. The insect has a reported host-range of well over 500 plant species, a high reproductive rate, the ability to readily disperse among hosts and breed year-round, and a propensity to develop resistance to insecticides. Previous distributions of this insect were limited to regions between the 30<sup>th</sup> parallels. However, in the past two decades, the pest has invaded every continent in the world except Antarctica, and commercial trade has facilitated the regular occurrence of populations in temperate greenhouse production systems in many parts of the world.

In 1992 we began to catalog the world literature on *Bemisia tabaci* and *Bemisia argentifolii* as an aid to researchers, educators, extension personnel, agricultural producers, industry, and governmental administrators. In 1995 we published a bibliography that attempted to cover the world literature through the end of 1994 (Butler et al. 1995). Since that time, addenda to this bibliography have been published annually (Naranjo et al. 1996, 1997, 1998, 1999, 2000, 2001). Here we present the entire bibliography based on literature cataloged through the end of February 2002. This bibliography attempts to cover all of the scientific literature on *B. tabaci* and *B. argentifolii* (peer-reviewed journal articles; books and book chapters; symposia and workshop proceeding and abstracts; and governmental, university, and industry reports). It also attempts to cover much of the literature on viruses vectored by *Bemisia* and that dealing with issues related to honeydew contamination.

This bibliography has been compiled from a number of sources. The Current Awareness Literature Service of the National Agricultural Library (NAL) has been instrumental in conducting searches of abstracting databases, such as AGRICOLA (NAL), AGRIS (United Nations, Food & Agriculture Organization), Biological Abstracts (BIOSIS), Commonwealth Agricultural Bureaux Abstracts (CABI), and Zoological Records (BIOSIS). We have also searched Current Contents (Institute for Scientific Information), Dissertation Abstracts International (University Microfilms International), and our personal reprint collections. Finally, this bibliography includes citations from the two annotated bibliographies of Cock (1986, 1993), and the proceedings of various international conferences, workshops and symposia. We have noted throughout the bibliography those references which have been cited by Cock. His bibliographies provide abstracts for many of the papers and because many references may be difficult to obtain we thought researchers might find it useful to check Cock's abstracts before beginning the arduous task of locating original papers.

The current bibliography and future annual addenda can be downloaded electronically from the Western Cotton Research Laboratory web site at (<u>www.wcrl.ars.usda.gov</u>) in ProCite (2.0 for DOS; 4.0 for Windows), MS Word (2000), and ASCII text formats. The bibliography also can be searched directly on this web site. Finally, a runtime version of the ProCite software and the databases can be provided on CD (contact SEN at snaranjo@wcrl.ars.usda.gov).

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# FIFTH ANNUAL PROGRESS REVIEW FIVE-YEAR SILVERLEAF WHITEFLY RESEARCH, ACTION & TECHNOLOGY TRANSFER PLAN Feb. 10-12, 2002

## Sunday, February 10, 2002

10:00 a.m.	Registration (PACIFIC FOYER)		
	Poster set-up begins (HARBORSIDE ROOM)		
1:00 p.m.	Welcome and Announcements	Walker Jones & Thomas M. Perring	
	(PACIFIC BALLROOM A)		
1:15 p.m.	Charge to the Conference	Robert Faust	
1:30 p.m.	Section A Paper Session	Jackie Blackmer & David Byrne	
3:35 p.m.	Section A Discussion		
4:05 p.m.	Break		
4:20 p.m.	Section F Paper Session	Steve Castle & Peter Ellsworth	
6:00 p.m.	Section F Discussion		
6:30 p.m.	Mixer and Poster session (HARBORSIDE	ROOM)	

## Monday, February 11, 2002

7:00 a.m.	Continental breakfast (PACIFIC BALLROOM A)				
8:00 a.m.	Section C Paper Session	John Palumbo & Shirley Taylor			
9:50 a.m.	Section C Discussion				
10:20 a.m.	Break				
10:30 a.m.	Section E Paper Session	Cindy McKenzie & Greg Walker			
11:45 a.m.	Section E Discussion				
12:15 p.m.	Lunch Break				
2:30 p.m.	Section D Paper Session	Bill Roltsch & Greg Simmons			
4:00 p.m.	Break				
4:15 p.m.	Section D Paper Session (cont.)				
4:50 p.m.	Section D Discussion				
5:35 p.m.	Depart for Boat Taxi to Coronado Island for Dinner (HOTEL LOBBY)				
National Whitefly Conference Agenda					
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## Tuesday, February 12, 2002

7:00 a.m.	Continental breakfast (PACIFIC BALLRO	DOM A)
8:00 a.m.	Section B Paper Session	Judith K. Brown & Robert L. Gilbertson
10:15 a.m.	Section B Discussion	
10:45 a.m.	Break	
11:00 a.m.	Progress Review - Sections A, B, C	Moderated by Section Co-Chairs
12:30 p.m.	Lunch Break	
2:00 p.m.	Progress Review - Sections D, E, F	Moderated by Section Co-Chairs
3:30 p.m.	General Discussion	

Immediately following the General Discussion, the Working Group will meet. The Program Planning and Review Committee will meet immediately following the Working Group.

## Section A: Biology, Ecology, and Population Dynamics Co-Chairs: Jackie Blackmer and David Byrne Sunday Afternoon 1:30-4:20

- 1:30 <u>Keynote Address</u>. Dale B. Gelman, Michael B. Blackburn, Jing S. Hu, and D. Gerling. Whitefly metamorphosis timing, regulation, and influences on the development of its parasitoid, *Encarsia formosa*.
- 2:00 Jackie Blackmer and D. Cross. Response of *Eretmocerus eremicus* to skylight and plant cues in a vertical flight chamber.
- 2:15 Luis A. Canas, Steven E. Naranjo, Peter C. Ellsworth. Seasonal ecology of *Bemisia tabaci* in Arizona: Low temperature and host plant effects on field populations and associated mortality factors.
- 2:30 Thomas P. Freeman, James S. Buckner, and Dennis R. Nelson. What I know about silverleaf whitefly nymph feeding and what I would like to know.
- 2:45 Dan Gerling and Dale B. Gelman. Physiological considerations in the penetration of *Eretmocerus mundus* into *Bemisia* nymphs.
- 3:00 M.S. Palaniswami. Isozyme, vector capability and cross breeding studies to confirm biotypes of *B. tabaci* in India.
- 3:15 Historical Presentation. David N. Byrne. Recounting whitefly biology research since 1992.
- 3:35 4:05 Discussion

4:05 - 4:20 Break

## **Posters:**

Thomas P. Freeman, James S. Buckner, and Dennis R. Nelson. Silverleaf whitefly nymph stylets and feeding characteristics. M. S. Palaniswami. Life table studies of *Bemisia tabaci* Genn. on cassava and laboratory evaluation of potentiality of

predators.

Dennis R. Nelson. Comparison of the feeding process of nymph and adult whiteflies.

Bahram Tafaghodinia. Engineering-based computer simulation for modeling greenhouse whitefly population growth.

### Section F: Integrated and Areawide Pest Management Approaches, and Crop Management Systems Co Chairs: Steve Castle and Peter Ellsworth Sunday Afternoon 4:20-6:30

4:20 C.C. Chu, C.G. Jackson, P.J. Alexander, and T.J. Henneberry. A light-emitting diode equipped CC trap for greenhouse insects.

4:35 Robert T. McMillan, Jr. Tomato yellow leafcurl gemini virus management in south Florida.

4:50 Keynote Address. Steve Castle. Outbreak occurrences: Factors that contribute and tactics that suppress.

5:20 <u>Historical Presentation</u>. Tom Henneberry. Assembling a multi-agency response to *Bemisia* outbreaks: a retrospective.

5:40 Historical Presentation. Nick Toscano. Achievements and management successes in the whitefly wars.

6:00 - 6:30 Discussion

### **Posters:**

Jennifer Jones, Peter Ellsworth, John Palumbo, Kai Umeda, and Pat Clay. Cross commodity research and outreach program. C.C. Chu, A.M. Simmons, P.J. Alexander, and T.J. Henneberry. A light-emitting diode equipped yellow card sticky trap for greenhouse insects.

## Section C: Chemical Control, Biopesticides, Resistance Management, and Application Methods Co Chairs: John Palumbo and Shirley Taylor Monday Morning 8:00 - 10:20

- 8:00 D.H. Akey and T.J. Henneberry. Use of sucrose esters as biorational agents to control silverleaf whitefly in Arizona upland cotton.
- 8:15 D.J. Schuster, L.D. Ortega, S. Thompson, and J.E. Polston. Laboratory comparison of potential repellents for silverleaf whitefly adults.
- 8:30 Eric Natwick. Comparison of four neonicotinoid insecticides for whitefly control.
- 8:45 N.C. Toscano, N. Prabhaker, S. Castle, T. Henneberry. Inter-regional differences in baseline toxicity of *Bemisia* argentifolii to buprofezin and pyriproxyfen.
- 9:00 Keynote Address. Nilima Prabhaker. Insecticide resistance in Bemisia tabaci: Past expectations and present reality
- 9:30 <u>Historical Presentation</u>. John C. Palumbo. Chemical control of *Bemisia* whiteflies: Summary of research accomplishments and technology transfer.
- 9:50 10:20 Discussion
- 10:20 10:30 Break

#### **Posters:**

D.J. Schuster, and S. Thompson. Comparing the susceptibility of the silverleaf whitefly to imidacloprid on tomato in Florida. Cindy McKenzie. Efficacy of sucrose octanoate to whitefly using a plant-based bioassay.

T.J. Henneberry, L. Forlow Jech, D.L. Hendrix, T. de la Torre, and J. Maurer. Effects of Applaud and Knack on sweetpotato whitefly nymph mortality and adult and nymph honeydew production in the laboratory.

## Section E: Host Plant Resistance, Physiological Disorders, and Host-Plant Interactions Co Chairs: Cindy McKenzie and Greg Walker Monday Morning 10:30 - 12:15pm

- 10:30 <u>Keynote Address</u>. Gregory P. Walker. Overview of whitefly feeding behavior: What we know and what we need to know.
- 11:00 Yun-Shu Chen, Wilhelmina T.G. van de Ven and Linda L. Walling. Expression of *SLW3* (silverleaf whitefly-induced) gene in the transgenic plants.
- 11:15 Laura Petro, Rick Redak,, Jim Bethke and Thomas M. Perring. Preference and performance of silverleaf whitefly on selected poinsettia cultivars.
- 11:30 Alvin M. Simmons. Movement of whitefly crawlers on several types of vegetable plants.

#### 11:45-12:15 Discussion

#### **Posters:**

Eric Natwick. Cotton host plant resistance to whitefly transmitted cotton leaf crumple disease.

Gregory P. Walker and Eric Natwick. High level of resistance to silverleaf whitefly in the cotton relative, *Gossypium* thurberi

## Section D: Natural Enemy Ecology and Biological Control Co Chairs: Bill Roltsch and Greg Simmons Monday Afternoon 2:30-5:20

- 2:30 Keynote Address. Don C. Vacek. Application of genetic diagnostics to biological control of silverleaf whitefly.
- 3:00 Steven E. Naranjo. Intraguild predation on Bemisia tabaci parasitoids by three generalist predators.
- 3:15 Jesusa C. Legaspi. Parasitism of the silverleaf whitefly in north Florida.
- 3:30 William Roltsch and Earl Andress. Silverleaf whitefly biological control in Imperial Valley, CA.
- 3:45 Gregory S. Simmons, Kim Hoelmer, Charles Pickett, Eric Natwick, and Earl Andress. Augmentative biological control of silverleaf whitefly infesting melons in California with releases of exotic *Eretmocerus* species, a review of the progress.
- 4:00 4:15 Break
- 4:15 Charles H. Pickett. Releases of exotic parasites in central California.
- 4:30 Historical Presentation. William Roltsch. The value of a collective effort in the biological control of silverleaf whitefly.
- 4:50 5:20 Discussion

## Posters:

Earl Andress. Exotic parasitoids in four crops in the Imperial Valley.

Larry J. Heilmann. DNA based species identification test for parasitic wasps of the whitefly.

James Buckner. Marking pheromones for female Eretmocerus mundus, a parasitoid of Bemisia argentifolii.

## Section B: Viruses, Epidemiology, & Virus-Vector Interactions Co Chairs: Judith K. Brown and Robert L. Gilbertson Tuesday Morning 8:00-10:50

- 8:00 <u>Keynote Address</u>. Judith K. Brown: A twenty year legacy of Begomoviruses and the whitefly vector in the U.S. sunbelt states: 1981-2001.
- 8:25 Keynote Address. Robert L. Gilbertson: Tomato yellow leaf curl virus in the Dominican Republic A case study.
- 8:55 William M. Wintermantel and Arturo A. Cortez. Complementation for transmission by non-vector whiteflies among tomato-infecting criniviruses.
- 9:10 Thomas M. Perring, and Charles A. Farrar. Whitefly-borne virus epidemiology: Lessons learned from the greenhouse whitefly and tomato infectious chlorosis virus.
- 9:25 Jane E. Polston Lack of transovarial transmission of begomoviruses by Bemisia tabaci biotype B.
- 9:45 Judith K. Brown and A.M. Idris. Cloning and sequencing of *Cotton leaf crumple virus* a begomovirus infecting cotton in the Sonoran Desert.
- 10:00 Y.P.S. Rathi. Mungbean yellow mosaic virus-epidemiology and management.

### 10:15 - 10:45 Discussion

## **Posters:**

Judith K. Brown, A.M. Idris, and J. Bird. Cloning and molecular characterization of the A and B components for Macroptilium mosaic virus from Puerto Rico.

## **Appendix D: List of Registered Meeting Participants**

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## **Minutes of the PPRC**

Minutes PPRC Meeting Holiday Inn on the Bay 2/10/02 11:30 a.m.

Meeting was chaired by Tom Perring. PPRC members in attendance were:

Jacke Blackmer	David Byrne	Steve Castle
Thomas Henneberry	Walker Jones	Marla Lawrence
Cindy McKenzie	Tom Perring	Shirley Taylor

Tom Perring discussed the general overview of the program and reminded Section Co-chairs of their assignments during the meeting which included: 1) keeping the scheduled talks on time, 2) leading discussions, and 3) updating year 3 tables during the progress review.

He reminded Section Co-chairs that after the meeting they must submit section summaries with technology transfer and completed year 3 tables to Marla Lawrence.

Marla Lawrence will send co-chairs all copies of abstracts by February 26<sup>th</sup>.

Deadlines for abstracts submission or changes will be February 25<sup>th</sup> and Section Chair information is due to Marla Lawrence by March 10th.

## Five-Year National Research and Action Plan Priority Tables, Research Needs, and Yearly Goals (1997-2001)

Table A. Biology, Ecology, and Population Dynamics

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
Determine life cycle vulnerabilities (life tables) <sup>a</sup> , population development and natural mortality factors, natural enemies on major crops, urban plantings, weeds and predict overwintering potential.	Whitefly and natural enemy sampling in cultivated crops, urban planting and weed hosts.	Determine potential of intercrop weed host & urban planting, movement of whiteflies and natural enemies.	Identify potential low population manipulation on vital host links for survival.	Initiate studies to manipulate host sequences to determine potential influence on whitefly population.	Continue 4 and finalize analysis of the potential of habitat modification as a management tool.
Develop sampling methodology, action and <sup>b,c</sup> economic thresholds for all major crops. Sampling methods and thresholds modified in light of natural enemy levels and existing management strategies.	Initiate whitefly to identify spatial and temporal distributions in major cultivated crops.	Analysis and identification of needed additional sampling research to develop appropriate sampling protocol.	Validate and refine sampling methods.	Implement sampling protocols through cooperative extension outlets and other technology transfer methods.	Finalization, implementation and use in IPM systems.
Develop population models to describe and predict whitefly population growth and spatial and temporal distribution. Develop simple day-degree sub-models for estimating phenology and temporal patterns of whitefly, natural enemies and host crops.	Summarize whitefly biology, ecology and plant phenology to identify whitefly host plant interfaces.	Begin model development to include all biological and plant phenology data in simulation development.	Provide model simulation of whitefly populations and multiple cropping systems.	Identify weak points and needed information to improve model simulations.	Validate and expand effort to provide predictive models capabilities for whitefly population development and crop interfaces.
Develop sampling methods for quality of cotton lint, vegetables and other commodities.	Initiate sampling of seed cotton in the field during the season, at harvest, after picking, moduling and ginning.	Based on 1, expand and repeat sampling protocols as described.	Develop sampling protocol for field and harvest and processing sampling and determine interrelationships.	Extend sampling protocols to textile mill and verify field findings in relation to mill problems.	Modify, refine and complete sticky cotton sampling protocols from the field to the mill.
Quantify whitefly and natural enemy dispersals and contribution to population dynamics.	Review and analyze existing knowledge of whitefly dispersal.	Validate times of whitefly dispersal, environmental factors and identify modifying factors.	Determine proportion of whitefly population that are migratory and their reproductive potential.	Quantify the role of dispersal in population dynamics on different crop systems.	Formulate theory for manipulating and/or using dispersal as a tool in IPM.
Define mating behavior, reproductive isolation, species, biotypes.	Initiate studies on mating, oviposition and other behavior.	Define interspecies interbiotype mating interactions.	Define factors involved in mating, cues, feedback mechanisms, etc.	Develop potential methods of utilizing behavioral information in management strategies.	Review, summarize and propose additional needed research.

#### Table A. Biology, Ecology, and Population Dynamics

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
Validate <i>Bemisia</i> taxa morphology, genetic, biochemical, and biology characteristics.	Continue examination of <i>Bemisia</i> sp. for distinct morphological character differences.	Develop genetic molecular level and acceptable species level separation.	Discuss results, plan additional research, arrive at a consensus decision.	Publish verification of new species or other appropriate taxa.	
Define role of endosymbionts in metabolism, host adaptation, nutrition and survival.	Identify endosymbionts in whitefly.	Determine role of endosymbionts in whitefly biological functioning.	Determine potential for manipulating, interfering with or inhibiting endosymbiont function.	Determine associated enzymes and/or other endosymbionts and whitefly relationships.	Summarize and implement findings with suggestion for additional research.
Characterize nutrient uptake and metabolism	Identify the major carbohydrates, amino acids and other nutrients essential for whitefly growth and development.	Determine the biochemical pathways for metabolism of compounds essential for whitefly development.	Determine the physical and biochemical processes involved in uptake of carbohydrates, amino acids and other essential nutrients.	Determine the potential for blocking key steps in nutrient uptake and/or metabolism.	Implement findings by developing inhibitors of nutrient uptake and/or metabolism.
Develop whitefly artificial diets and natural enemy mass-rearing.	Identify whitefly nutritional components in plant tissue.	Develop whitefly artificial feeding systems.	Conduct addition, deletion studies to identify essential nutritional needs.	Evaluate developed diets on whitefly fecundity/longevity biology, behavioral characteristics.	Develop whitefly rearing system and adapt for production of natural enemies.
<b>**</b> Pursue specific genetic and biological basis for variability in whitefly biotypes, strains, and species; determine impact of different genotypes/phenotypes on whitefly-mediated transmission and on the epidemiology of virus diseases.	Identify differences in species, strains and biotypes with respect to transmission, host range, mating compatibilities, molecular variability, and map the biogeographic distribution of distinct types within the <i>B.</i> <i>tabaci</i> species complex.	Continue to study differences in species/ strains/biotypes with respect to transmission, host range, mating compatibilities, molecular variability. Determine molecular basis of observed variability in biological, molecular, & genetic terms. Infer molecular phylogenies from molecular markers.	Continue with work from previous years. Study impact of biotypes, strains, and species differences in the disease spread, crop damage, and specific control measures to reduce whitefly vector populations. Linkages with biological and chemical control sections.	Identify potential factors related to specific genetic and biological variability that may be manipulated to reduce disease spread. Develop molecular approaches to track biotypes, strains, and species relative to disease spread, based on differential molecular markers.	Summarize results, identify new research needs and make recommendations for implementation or expansion of research.

<sup>a</sup> Natural enemy research complements from Section D, see Table D.
 <sup>b</sup> Action and economic thresholds also apply in Section C, see Table C.
 <sup>c</sup> Sampling technology applicable to all other sections, see Tables B to F.
 \*\* Revised 3/31/2000

Table B.	Viruses,	Epidemiology,	and Virus	-Vector	Interactions
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Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
Identification and characterization of new or emerging whitefly-transmitted viruses and strains	Monitor crops for presence of whitefly- transmitted diseases, and determine relative disease incidence. Begin virus identification and strain differentiation.	Virus identification and characterization. Develop methods for identifying causal agents and for tracking viruses and strains using molecular methods.	Continue etiological studies and virus characterization. Apply molecular diagnostics to virus identification and evaluation of disease incidence and virus distribution.	Continue etiological studies and virus characterization efforts. Apply molecular diagnostics to virus identification and evaluation of disease incidence and virus distribution.	Summarize and review results. Determine areas of new research.
Molecular epidemiology: identification of economic viruses, host plants, and reservoirs, and determination of geographic distribution of viruses.	Monitor and identify host plants, virus reservoirs in affected areas. Linkages to diagnostic methods for virus ID and tracking.	Continue field studies. Determine economic input of diseases on crop production and associated losses.	Establish geographic distribution of viruses and identify sources of inoculum. Assess role of alternative host virus reservoirs on spread of diseases.	Identify and characterize virus involvement in disease establishment and spread. Assess potential methods of reducing virus reservoirs as a method of reducing disease.	Review and make recommendations for further research and potential implementation of results.
Virus-vector interactions, factors affecting virus transmission, and basis for virus-vector specificity; determination of endosymbiont involvement in whitefly-mediated transmission	Initiate studies on virus- vector interactions and on basis for the specificity of whitefly- mediated geminivirus transmission.	Determine specific cellular and molecular factors involved in virus transmission. Study role of endosymbionts in virus acquisition and transmission.	Continue studies in progress to determine specific factors involved in virus transmission, and the role of endosymbionts in virus acquisition and transmission.	Continue virus-vector interactions studies toward the development of approaches for disease control.	Summarize findings and suggest new research needs; implementation of existing knowledge.
Strategies to reduce virus spread by management of cropping systems, reduced transmission frequencies, and other potentially effective approaches.	Develop approaches to managing cropping systems to reduce vector densities to decrease transmission frequency and inoculum sources, taking into account weed and crop reservoirs in disease incidence and distribution.	Continue studies of management approaches for disease abatement. Interdisciplinary studies in conjunction with whitefly control methods in Sections B and C.	Continue studies of management approaches for disease abatement. Focus on interdisciplinary studies in conjunction with whitefly control methods in Sections B and C.	Evaluate strategies for crop management and impact on disease epidemiology.	Evaluate approaches and identify areas of future research for disease control by management of cropping systems. Linkages with IPM approaches.

### Table B. Viruses, Epidemiology, and Virus-Vector Interactions (continued)

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
virus resistant germplasm through r conventional and engineered/molecular I approaches. Define prospective strategies for I selecting candidate viruses, identifying specific v virus diseases to target, and prioritize specific e crops and cultivars for protection approaches.	Define strategies for resistance efforts. Identify target viruses. Identify germplasm with virus resistance. Initiate efforts toward defining prospective engineered resistance strategies. Identify candidate crops and recipient cultivars.	Continue to define suitable strategies for determining target viruses. Isolate and characterize virus- resistant germplasm. Continue work toward engineered resistance in target crops and selected viruses.	Further identification of resistant germplasm and develop new methods of incorporating resistance into crop plants. Evaluate resistance strategies with respect to broad spectrum or virus- specific protection.	Continue development of resistant varieties. Evaluate resistance strategies with respect to broad spectrum or virus- specific protection. Define mechanisms of resistance.	Evaluate resistant plants in greenhouse and field experimentation, and identify additional research. Molecular- based monitoring of transgenes in environment.

\*\* Revised 3/31/2000

	Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
[ <b>m</b> ]	prove insecticide efficacy:					
•	Develop, test, and assist in the registration of insecticides, biorationals, and natural products.	Develop new chemistries and natural products. Develop improved techniques for evaluating efficacy of insecticides. Support registration of desirable new products by providing information to regulatory agencies.	Same as Year 1. Determine new modes of action of effective materials. Elucidate biochemical pathways of synthesis and degradation of natural products.	Same as Year 2. Evaluate the potential for transforming plants with natural product genes.	Same as Year 3.	Same as Year 4.
•	Develop improved methods of application including formulation and delivery of materials to improve control.	Develop spray systems for better underleaf coverage. Evaluate rates, timing, placement in relation to efficacy. Consider formulation, UV protectants, and other means to improve efficacy. Develop improved methods to evaluate application efficacy. Field test under commercial conditions for technology transfer.	Same as Year 1.	Same as Year 2.	Same as Year 3.	Same as Year 4.
Co	nserve insecticide efficacy:					
•	Relate action thresholds to insecticide usage patterns.	Refine action thresholds based on insecticide efficacy and input from other control strategies.	Same as Year 1.	Same as Year 2.	Same as Year 3.	Same as Year 4. Summarize and recommend in IPM systems.

## Table C. Chemical Control Dispecticides, Desistance Management and Application Methods

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
<ul> <li>Elucidate the role of genetic, biochemical and ecological factors leading to insecticide resistance.</li> </ul>	Establish whitefly strains resistant and susceptible to various classes of insecticide. Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide.	Same as Year 1. Evaluate the role of refuge habitats (weeds, tolerant crops, urban areas) to assure input of susceptible genes in whitefly population.	Conduct studies to determine the genetics and biochemistry of resistance and cross resistance to different classes of insecticide. Evaluate the role of refuge habitats (weeds, tolerant crops, urban areas) to assure input of susceptible genes in whitefly population. Evaluate the influence of host plant on susceptibility to insecticides.	Same as Year 3.	Same as Year 4.
Improve insecticide efficacy:					
Improve techniques for monitoring resistance.	Establish baseline data on toxogenic responses of whitefly populations to new insecticides.	Same as Year 1. Expand comparative studies of resistance levels in diverse agro- ecosystems. Evaluate relationship between monitoring results and field efficacy.	Same as Year 2. Summarize, analyze, and produce standardized comparable monitoring systems.	Same as Year 3. Develop standard systems for general use including user friendly techniques to assist growers and extension agents to evaluate susceptibility of whitefly populations to commonly used insecticides.	Same as Year 4.
Develop, evaluate and refine resistance management systems	Evaluate the effects of mixtures and rotations of new and old chemistries to mitigate selection for resistance.	Same as Year 1. Develop methods to evaluate and augment the beneficial influence of refuges as sources of susceptible genes to the population pool.	Same as Year 2. Develop criteria for integration of successful strategies in agricultural systems. Field test resistance management systems as long range components of successful IPM.	Same as Year 3.	Same as Year 4. Technology transfer.

#### Table C. Chemical Control, Biopesticides, Resistance Management, and Application Methods (continued)

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
Integrate chemical control with other tactics.	Evaluate selectivity of synthetic insecticides and natural products to key whitefly natural	Same as Year 1. Test compatibility of biological control with selective synthetic or	Same as Year 2. Integrate systems with host plant resistance and cultural controls.	Test compatibility of biological control with selective synthetic or natural product	Integrate systems with host plant resistance and cultural controls. Summarization and
	enemies.	natural product insecticides as required.		insecticides as required. Integrate systems with host plant resistance and cultural controls.	technology transfer.

#### Table C. Chemical Control, Biopesticides - Resistance Management, and Application Methods (continued)

<sup>a</sup> See Table A for complementary research on thresholds.
 <sup>a</sup> See Table B for complementary research on virus/vector interactions.
 <sup>a</sup> See Table D for complementary research on biological control.
 <sup>b</sup> See Tables E and F for complementary research on systems management.

	Approaches/Goals <sup>a</sup>	Year 1	Year 2	Year 3	Year 4	Year 5
Nat	tural control and conservation:					
•	Develop natural enemy conservation practices to reduce mortality to indigenous and introduced natural enemies.	Conduct life table analyses of indigenous and introduced natural enemies to identify key mortality factors of natural enemy populations.	Identify the spatial scale upon which the key mortality agents are acting.	Conduct manipulative experiments to evaluate the impact of each natural enemy mortality agent on whitefly suppression.	Conduct a feasibility study and economic assessment of altered crop management practices that may enhance the impact of indigenous natural enemies.	Develop and evaluate area wide programs to facilitate full implementation.
•	Evaluate potential of alternate plants it act as in-field refuges or insectaries for natural enemies.	Identify potential plants for natural enemy population development and assess risks of these plants to foster additional pest problems.	Determine refugia plant phenology in relation to cultivated crop phenology.	Conduct field tests to assess whether refuges act of natural enemy and whitefly sinks or sources to adjacent cropping systems.	Conduct field tests to evaluate spacing of refuges necessary to achieve satisfactory whitefly suppression.	Conduct a feasibility study and economic assessment of alterna plantings in terms of entire crop managem program.
•	Assess cues used by natural enemies to locate whitefly to identify potential methods for enhancing natural enemy activity.	Conduct laboratory tests to identify cues used by natural enemies to locate and attack whitefly.	Determine potential methods for manipulating cues as part of a whitefly management program.	Conduct small scale trials to enhance whitefly suppression by manipulating natural enemy location and attack of whitefly.	Conduct large scale field trials and evaluate product development for commercial investment as necessary.	Transfer technology needed) to commerci interests for full implementation.
Au	gmentation of natural enemies:					
•	Develop natural enemy mass-rearing systems.	Identify natural enemies with the highest potential for controlling whitefly in key cropping systems.	Determine nutritional, physiological, and ecological requirements for mass-rearing.	Develop rearing systems on selected hosts and on artificial diets. Determine economic feasibility of the procedure.	Evaluate rearing system effects on natural enemy life history characteristics, behavior, and ability to suppress whitefly populations.	Facilitate transfer of mass-rearing technoloto commercial interest as necessary.

Approaches/Goals <sup>a</sup>	Year 1	Year 2	Year 3	Year 4	Year 5
mportation biological control:					
Develop release technologies to maximize the effectiveness of mass-reared natural enemies in the field.	Identify natural enemies with the highest potential for controlling whitefly in key cropping systems and that may be economically mass produced.	Evaluate the fate of natural enemy life stages under field conditions to identify the appropriate developmental stage to be released.	Develop necessary technology for release of the appropriate natural enemy life stage.	Evaluate release technology effects on natural enemy life history characteristics, behavior, and ability to suppress whitefly populations.	Facilitate transfer of mass-rearing technology to commercial interests as necessary.
Evaluate augmentative parasitoid, predator, or pathogen releases.	Initiate studies on natural enemy augmentation with identified high potential natural enemies.	Conduct releases on selected crop systems at various rates of release.	Identify optimal release strategies for key cropping systems.	Continue evaluation of releases, determine need for additional releases. Compare results in different cropping systems and environments.	Analyze information an make recommendation regarding need for expansion of the approach.
Evaluate the ability of exotic natural enemies to suppress whitefly populations under field conditions.	Identify sites suitable for the release and subsequent evaluation of each candidate natural enemy. Conduct inoculative releases of natural enemies.	Evaluate establishment of exotic natural enemies within target release area. Determine if additional releases are necessary.	Assess spread of established natural enemies and their ability to suppress whitefly populations.	Continue to assess the spread of established natural enemies and their ability to suppress whitefly populations. Evaluate program progress and determine if additional strategies are necessary.	Complete program analysis. Publish program assessment and conduct an economic assessment.
Clarify systematics of predators, parasitoids and pathogens.	Conduct taxonomic studies of species within targeted release sites. Verify taxonomic purity of mass-reared natural enemies. Complete taxonomic work on poorly characterized but important groups. Assist in determining most suitable natural enemies for release through biogeographical analysis.	Provide taxonomic support for importation and mass-rearing programs. Publish keys to assist in species identifications.	Provide taxonomic support for importation and mass-rearing programs.	Provide taxonomic support for importation and mass-rearing programs.	Provide taxonomic support for importation and mass-rearing programs.

#### Table D. Natural Enemy Ecology and Biological Control (continued)

Approaches/Goals <sup>a</sup>	Year 1	Year 2	Year 3	Year 4	Year 5
Systematics, ecology, and population dynamics of natural enemies <sup>b</sup> :					
Determine <i>Bemisia</i> - natural enemy-host plant (Tritrophic) interactions.	Initiate studies to identify mechanisms involved in <i>Bemisia</i> - and natural enemy plant attraction.	Study plant characteristics mediating whitefly population densities.	Study compatibility of characteristics of plant traits mediating whitefly populations with the abilities of natural enemies to suppress whitefly populations.	Assess the implementability of favorable tritrophic interactions within the context of an whitefly management program.	Implement and evaluate large scale crop management programs for suppression of whitefly populations.
Identify the attributes of natural enemy biology and population level interactions to explain biological control successes and failures.	Assess the value of the <i>Bemisia</i> biological control research to evaluate key issues to the science of biological control.	In conjunction with field evaluations, validate predictions made by behavioral and population models important to biological control.	Assess deviations between theoretical predictions and field data.	Evaluate behavioral or population level parameters that may explain observed deviations.	Quantify the impact of basic research on the development of feasible biological control programs for <i>Bemisia</i> and the advancement o the field as a science.

# Table D. Natural Enemy Ecology and Biological Control (continued) Approaches/Goals<sup>a</sup> Year 1

<sup>a</sup> See Table C for complementary research.
 <sup>b</sup> See Table A for complementary research.

Approaches/Goals	Year 1	Year 2	Year 3	Year 4	Year 5
Characterize resistance mechanisms and identify chemical/morphological components, and study effects of insect adaptation.	Identify potential sources of germplasm for disease, plant disorders and whitefly resistance. <sup>a</sup>	Determine physiological and/or morphological basis for resistance, & effects of host-plant history and insect adaptation on plant resistance to whiteflies. Continue to identify resistant germplasm.	Elucidate biochemical and molecular basis for resistance. Continue to identify resistant germplasm.	Determine potential for transfer of resistance traits.	Evaluate potential for incorporating <i>Bemisia</i> , plant disorder and disease resistance into acceptable plant type.
Develop molecular level techniques to produce resistant germplasm.	Identify physiological processes of whiteflies to target for inhibition.	Identify natural products for inhibiting processes.	Isolate the relevant biosynthetic enzymes that encode for natural products inhibiting processes.	Insert genes into plants via plant transformation. b	Evaluate potential of newly transformed germplasm.
Incorporate resistance traits into commercial genotypes.	Identify and isolate genetic sources of resistance for transformation and/or breeding.	Insert genes into plants <sup>b</sup> via plant transformation.	Evaluate potential of newly transformed germplasm.	Continue to refine resistance factors to improve resistance in newly transformed germplasm.	Incorporate other desirable plant characteristics for crop production.
Determine influence of host plant morphology, physiology and phenology on feeding behavior and competition. <sup>c</sup>	Characterize nutritional and other preference properties of various host plants.	Determine the biochemical mechanism regulating adaptation to host plants.	Determine changes in whitefly gene expression in response to host manipulation.	Relate changes in gene expression to whitefly physiology.	Summarize and disseminate results.
Define whitefly feeding and oviposition behavior and investigate approaches for interrupting whitefly feeding and digestion. <sup>d</sup>	Investigate approaches for interruption of feeding, assimilation, development and reproduction.	Identify physiological and morphological mechanisms regulating processes.	Determine biochemical and molecular basis for inhibiting processes.	Determine potential for transfer of resistance traits.	Insert genes into plants <sup>a</sup> via plant transformation.
Study whitefly toxicogenic plant reactions.	Determine effects of whitefly feeding on host plant physiology, morphology and anatomy.	Determine biochemical basis for physiological response of plant.	Elucidate changes in plant gene expression.	Identify resistance germplasm.	Evaluate potential for transferring new germplasm.

#### Table E. Host Plant Resistance, Physiological Disorders, and Host Plant Interactions

<sup>b</sup> Progress at this point may extend to several year research.

<sup>d</sup> See Section A, approach #9.

Approaches/Goals <sup>a</sup>	Year 1	Year 2	Year 3	Year 4	Year 5
Development:					
Study whitefly-crop interactions <sup>b</sup> as cultural components that affect population dynamics, e.g., water, nutrients, plant population, planting/ termination/harvest dates, other farm practices, intercrop relationships.	Identify potential beneficial or exacerbating farm practices or inputs for testing.	Determine nature and character of relationship between interaction and whitefly population dynamics.	Identify mechanisms governing relationship and alter or manipulate factors that suppress whitefly dynamics.	Refine system, add other compatible components, evaluate economic impact; conduct field testing and evaluations.	Conduct economic analyses and determine next level of IPM/ICM systems evaluation. Develop recommendations of best management practices.
Develop behavioral barriers <sup>b</sup> to whitefly colonization and population development, e.g., mulches, trap crops, intercropping, row covers, etc.	Review potential behavioral disrupters and evaluate as potential IPM components.	Conduct field-level trials; quantify impact to crop and whitefly dynamics	Apply promising technologies to high- value crop systems; field test and evaluate.	Refine system, add other components, and conduct economic feasibility analyses.	Summarize and evaluate results; prepare crop systems-specific recommendations.
Integration:					
Develop Integrated Pest Management <sup>c</sup> systems using dual or multiple control tactics, e.g., cultural, biological, chemical, host plant resistance, etc.	Identify candidate dual or multiple control tactic systems, e.g., IGRs and natural enemy conservation.	Initiate field testing of candidate systems.	Continue field testing & evaluate feasibility of large scale testing; add components as necessary.	Initiate large-scale experiments; incorporate economic evaluation.	Evaluate multiple component system as potential deliverable; prepare recommendations.
Integrate sampling with other key components of IPM systems, e.g., thresholds, economics, decision-making, biological control, etc.	Develop or modify sampling systems for new crops; integrate with thresholds and decision-making.	Establish practical utility of system through economic analyses; field efficiencies and costs.	Integrate additional control components into sampling, threshold & decision-making systems	Evaluate in whole field systems. Identify weaknesses; target improvements.	Evaluate redesigned decision systems; continue field testing and economic analyses.
Delivery and Implementation:					
Elevate single field/farm practices to areawide community-based contexts; develop methodology for installing and evaluating areawide control technologies and their impact.	Identify agricultural communities amenable to areawide management; conduct thorough pre- implementation evaluation.	Install control technologies into community; develop systems for evaluation.	Identify additional IPM/ICM compatible components. Re-assess and adapt program. Conduct areawide economic analyses.	Formulate clientele surveys; develop & begin to implement protocols for evaluating areawide technologies.	Refine, reevaluate and identify weaknesses. Formulate recommendations for future areawide management systems. Conduct surveys.

#### Table F. Integrated and Areawide Pest Management Approaches and Crop Management Systems

### Table F. Integrated and Areawide Pest Management Approaches and Crop Management Systems

Approaches/Goals <sup>a</sup>	Year 1	Year 2	Year 3	Year 4	Year 5
Implement and deliver Integrated Pest	Develop and distribute	Conduct whole	Expand sites of testing	Incorporate new	Validate new
Management and Integrated Crop	provisional IPM & ICM	farm/operation	with grower	information and	components; finalize
Management systems or system components to	recommendations.	demonstrations of IPM	cooperators; conduct	economics into	recommendations;
clientele.		systems.	validation studies.	recommendations.	expand to new crops.

<sup>a</sup> See Tables A to E for additional complementary research.
 <sup>b</sup> See Table A for additional complementary research.
 <sup>c</sup> See Table E for additional complementary research.