PROGRAM AND ABSTRACTS

BELTSVILLE SYMPOSIUM XVIII

Pest Management: Biologically Based Technologies

May 2-6, 1993

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May 2-6, 1993

United States Department of Agriculture
Agricultural Research Service
Beltsville Area Director's Office
10300 Baltimore Avenue
Beltsville, Maryland
20705-2350

WELCOME TO SYMPOSIUM PARTICIPANTS

The value of agricultural products produced in the United States exceeds $150 billion annually. However, pests such as insects, nematodes, microbial pathogens and weeds reduce the quality of these products and cause production losses on the order of 35 percent. The postwar development of effective and economical pesticides has enabled farmers to control many of these pests and to achieve spectacular production efficiencies. In recent years, however, problems with environmental and food safety and pest resistance, have underscored the need to develop alternative and supplemental pest control strategies. This has led to a resurgence of interest in biologically based pest control technologies for use in integrated pest suppression efforts. As with any new and developing technology, strong support and encouragement is vital. This USDA-ARS Beltsville Symposium XVIII is intended to help in promoting these biologically based technologies through bringing together the world's leading experts to share ideas and state-of-the-art approaches.

The subjects to be discussed encompass the full range of approaches from the molecular to the development of holistic IPM systems. The sharing of insights, ideas, experiences, and even disappointments, in forums such as this are critical to both the growth and development of new technologies and the reinforcement of our resolve to confront those obstacles standing in the way to fulfilling the vision of sustainable farming systems. For this reason, the Beltsville Agricultural Research Center is pleased to host this exciting and hopeful symposium.

K D Murrell

K. D. Murrell
Director
Beltsville Area

Sponsors of the Symposium

The Friends of Agriculture Research-Beltsville, Inc. (FAR-B) are co-sponsors of the Beltsville Symposium Series. FAR-B is a non-profit group dedicated to supporting the research and educational programs at the Beltsville Agricultural Research Center. Membership is made up of former and current employees and a growing number of industry supporters. The Symposium Organizing Committee wishes to thank the members of FAR-B for their many contributions to the success of this meeting.

The organizers also wish to thank the following sponsors of the Symposium for their generous financial contributions:

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SYMPOSIUM INFORMATION

Location

Mixer (Sunday), Registration (Sunday), and Posters (Monday evening)
Holiday Inn, Grand Ballroom
10000 Baltimore Blvd. at the Beltway, Beltsville, MD 20740

Symposium: Auditorium, Beltsville Agricultural Research Center (BARC)
Building 003, BARC-West (Baltimore Blvd.), Beltsville, Maryland 20705
Registration, Post Office and Stamp Exhibit: Room 20, Bldg. 003

Registration

Sunday, May 2, 5:00 - 8:00 PM (Holiday Inn, Grand Ballroom)
Monday, Tuesday, and Wednesday, May 3-5, 7:30 AM - 5:00 PM (BARC, Bldg. 003)
Thursday, May 6, 7:30 - 9:00 AM (BARC, Bldg. 003)

Stamp Exhibitors

The organizers of the Symposium express their sincere appreciation to the following scientists for sharing their personal collection of agriculture-related stamps with the symposium participants:

Ronald F. Korcak
Ashok K. Raina
Albert A. Piringer
William P. Wergin

Post Office

A Symposium sub-station of the Beltsville Post Office will be in room 20, Bldg. 003 on Tuesday and Wednesday, May 4 & 5. A special pictorial cancellation will be available. The proceeds from the sale of commemorative envelopes will go to FAR-B.

Slide Preview Area

Room 20, Bldg.003, BARC-West

Sunday, May 2
Mixer
Tickets in Registration Package
7:00 - 9:00 PM
Ballroom A, Holiday Inn
10000 Baltimore Blvd. at the Beltway
Beltsville, MD

Session I Monday, May 3. J. J. MENN, Chairman.

Symposium Opening

8:30 AM Welcome - G. C. MARTEN, Associate Director, Beltsville Area
8:45 AM Friends of Agricultural Research-Beltsville Award Presentation - G. W. IRVING

Plenary Session

9:00 AM Biologically Based Pest Management: Increasing Its Acceptance
R. W. F. HARDY
9:40 AM Parasites and Predators Play a Paramount Role in Pest Management
J. C. VAN LENTEREN
10:20 AM **Coffee Break**
10:50 AM Bt Maize for Control of European Corn Borer
M.-D. CHILTON, M. Koziel, T. C. Currier, and B. J. Miflin
11:30 AM The Role of Biological Control in Pest Management in the 21st Century
R. J. COOK

12:10 - 1:30 PM Lunch

Session II Monday, May 3. D. J. CHITWOOD, Chairman.

Biocontrol Agents for Pest Suppression. Section A.

1:30 PM Viral Satellites, Molecular Parasites for Plant Protection
J. M. KAPER
1:55 PM Viruses for Control of Arthropod Pests
P. V. VAIL
2:20 PM Bacteria for Suppression of Foliar Plant Diseases
S. E. LINDOW
2:45 PM Bacterial Control of Flies in Livestock Operations
E. T. SCHMIDTMANN
3:10 PM **Coffee Break**
3:40 PM Fungi and Fungus/Bioregulator Combinations for Control of Plant-Parasitic Nematodes
S. L. F. MEYER and R. N. Huettel
4:05 PM Arthropods for Suppression of Terrestrial Weeds
R. D. GOEDEN
4:30 PM Introduction of Natural Enemies for Suppression of Arthropod Pests
T. S. BELLOWS

Poster Session and Computerized Applications
7:00 - 10:00 PM
Light refreshments will be served.
Grand Ballroom
Holiday Inn, 10000 Baltimore Blvd. at the Beltway, Beltsville, MD

Session III Tuesday, May 4. R. D LUMSDEN, Chairman.

Biocontrol Agents for Pest Suppression. Section B.

8:30 AM	Multi-faceted Biological Control of Postharvest Diseases of Fruits and Vegetables C. L. WILSON and A. El Ghaouth
8:55 AM	Biological Control of Plant Parasitic Nematodes with Plant-Health-Promoting Rhizobacteria R. A. SIKORA and S. Hoffmann-Hergarten
9:20 AM	Dynamics of *Sporidesmium*, A Naturally Occurring Fungal Mycoparasite P. B. ADAMS and D. R. Fravel
9:45 AM	Managing Soilborne Plant Pathogens with Fungal Antagonists R. D. LUMSDEN, J. A. Lewis, and J. C. Locke
10:10 AM	**Coffee Break**
10:40 AM	Conservation of Arthropod Natural Enemies to Control Insects J. D. DUTCHER
11:05 AM	Augmentation of Parasites and Predators for Suppression of Arthropod Pests E. G. KING
11:30 AM	Progress and Promise in Management of Aquatic-Site Vegetation Using Biological and Biotechnological Approaches L. W. J. ANDERSON

12:00 - 1:30 PM Lunch

Session IV Tuesday, May 4. D. K. HAYES, Chairman.

Biocontrol Agents for Pest Suppression. Section C.

1:30 PM	The Use of Fluorescent Brighteners as Activity Enhancers for Insect Pathogenic Viruses M. SHAPIRO and E. Dougherty
1:55 PM	Plant Growth-Promoting Rhizobacteria as Agents of Induced Systemic Resistance J. W. KLOEPPER, L. Liu, G. Wei, and S. Tuzun
2:20 PM	Mechanisms Involved in Biological Control of Soilborne Plant Diseases by Rhizosphere Bacteria J. E. LOPER
2:45 PM	Naturally Occurring Disease-Suppressive Soils C. ALABOUVETTE
3:10 PM	**Coffee Break**
3:40 PM	New Options for Insect Control Using Fungi A. E. HAJEK
4:05 PM	Foreign Plant Pathogens for Environmentally Safe Biological Weed Control W. L. BRUCKART and N. Shishkoff
4:30 PM	The Past, Present and Future of Insect Parasitic Rhabditids for the Control of Arthropod Pests G. O. POINAR, JR.
4:55 PM	Biocontrol for Insect Pests of Livestock and Poultry J. J. PETERSEN

Posters remain at Holiday Inn for review until 10:00 PM, Tuesday, May 4.

Session V Wednesday, May 5. B. A. LEONHARDT, Chairman.

Natural Compounds in Pest Management

8:30 AM	Microbial Metabolites with Biological Activity Against Plant Pathogens D. M. WELLER and L. S. Thomashow
8:55 AM	Phytochemical Resources for Pest Management W. S. BOWERS
9:20 AM	Maximizing Parasitoid Effectiveness in Biological Management Systems with Semiochemicals J. H. TUMLINSON and W. J. Lewis
9:45 AM	Attracticides for the Control of *Diabroticite* Rootworms R. L. METCALF, L. Deem-Dickson, and R. L. Lampman
10:10 AM	**Coffee Break**
10:40 AM	Applications of Pheromones for Monitoring and Mating Disruption of Orchard Pests R. E. RICE
11:05 AM	Strategies and Tactics for the Use of Semiochemicals Against Forest Insect Pests in North America J. H. BORDEN
11:30 AM	Biologically Based Regulatory Pest Management C. P. SCHWALBE
11:55 AM	Allelopathy for Weed Suppression H. G. CUTLER

12:20 - 1:30 PM Lunch

Session VI Wednesday, May 5. J. D. ANDERSON, Chairman.

Genetic Manipulation of Biocontrol Agents

1:30 PM	Expression of Viral Genes and Viral and Antiviral Proteins in Transgenic Plants to Confer Virus Resistance R. L. JORDAN and J. Hammond
1:55 PM	Recombinant Baculoviruses as Biological Insecticides B. D. HAMMOCK, P. Choudary, and S. Maeda
2:20 PM	Genetics of BT Insecticidal Crystal Proteins and Strategies for Construction of Improved Strains B. C. CARLTON
2:45 PM	Genetically Modified Bacteria for Biocontrol of Soilborne Plant Pathogens D. P. ROBERTS
3:10 PM	**Coffee Break**
3:40 PM	Insect Resistant Transgenic Plants D. A. FISCHOFF
4:05 PM	The Genome of Biocontrol Fungi: Modification and Genetic Components for Plant Disease Management Strategies G. E. HARMAN, C. K. Hayes, and M. Lorito
4:30 PM	Transgenic Beneficial Arthropods For Pest Management Programs: An Assessment of their Practicality and Risks M. A. HOY

Banquet - see next page for details.

Banquet Wednesday evening, May 5

Buses will begin loading at the Holiday Inn and at the front of Bldg. 003, BARC-West at 6:00 PM for the National Aquarium in Baltimore.

Banquet at the National Aquarium begins at 7:30 PM; exhibits are open from 7:00 - 10:00 PM.

Buses leave Aquarium at 10:00 PM for return to Holiday Inn and BARC, Bldg. 003.

Session VII Thursday, May 6. J. L. VAUGHN, Chairman.

Implementation: Needs, Issues, Challenges

8:30 AM	National Perspective on Implementation of Biologically Based IPM Strategies M. S. FITZNER
9:00 AM	New Regulatory Strategies for Pheromones. EPA Policies, Interpretations and Rules P. HUTTON
9:30 AM	Patents and Intellectual Property: An Overview B. U. BUCHBINDER
10:00 AM	**Coffee Break**
10:30 AM	The Development of Gliogard™ for Disease Control in Horticulture J. F. WALTER
11:00 AM	Public Sector Response to Needs, Issues and Challenges E. S. DELFOSSE
11:30 AM	Look to the Future W. KLASSEN
12:00 AM	Panel Discussion

- End of Symposium -

Tour of the Beltsville Agricultural Research Center

The BARC Visitor Center invites all Symposium attendees and their families to join a tour of the Research Center from 2:00 - 3:30 PM, Thursday, May 6. A bus will pick up tour attendees in front of Bldg. 003, BARC-West at 2:00 PM and return there at 3:30 PM. This is an opportunity for an overview of the BARC research facilities and programs. A sign-up sheet will be at the registration desk.

ABSTRACTS

PODIUM PRESENTATIONS

I 01 Biologically Based Pest Management: Increasing Its Acceptance

RALPH W.F. HARDY
Boyce Thompson Inst. for Plant Research, Ithaca, NY 14853

Pest management incorporates the application of products, processes, and information based on physical, chemical, and biological knowledge. This symposium employs those approaches based on biology although it overlaps with chemical and physical ones, and the most successful pest management will integrate optimally all of the approaches into the managed ecosystem. A holistic and integrative concept of biologically based pest management is fundamental to increasing its visibility, importance, financial support, development and adoption. The concept must be holistic for pests—weeds, nematodes, disease, and insects—and for biological approaches—molecular, cellular, organismal, and ecological. The Board on Agriculture of the United States National Research Council (NRC) initiated a study in June 1992, "Pest and Pathogen Control Through Management of Biological Control Agents and Enhanced Natural Cycles and Processes," with the issue of a report projected for spring 1994. The objective of the NRC report is to synthesize information and recommendations so as to increase the importance of biologically based pest management. Based on prior published reports, limitations and opportunities in biologically based pest management will be reviewed and possible options to overcome the limitations will be suggested.

I 02 Parasites and Predators Play a Paramount Role in Pest Management.

J.C. VAN LENTEREN
Laboratory of Entomology, P.O. Box 8031, 6700 EH Wageningen, The Netherlands

Parasites and predators have been recognized as important consumers of pest insects for centuries. More than 95% of the potential pest organisms is kept under control by natural enemies without any manipulation by man. During the past century of modern biological control, 5500 introductions of natural enemies have been made to new areas worldwide. This has led to successful control in 420 cases: 340 by parasites, 74 by predators and 6 by pathogens, so parasites and predators make up 99% of the successful cases. Biological control has shown to be a reliable, economically sound and environmentally safe means of pest reduction.
Application of biological control will be stimulated considerably in the near future because of (1) strongly increased costs for of pesticides, (2) continued development of resistance to pesticides (3) a strong pressure to reduce the use of chemical pesticides. Use of biological control contributes to current policies in the tropics and temperate zones to cut down pesticicde use with 50%.

I 03 Bt Maize for Control of European Corn Borer

MARY-DELL CHILTON*, Michael Koziel*, Thomas C. Currier* and Benjamin J. Miflin@
*Ciba-Geigy Corporation, Agricultural Biotechnology, PO Box 12257, Research Triangle Park, NC 27709, and
@Ciba-Geigy Ltd, Basel CH 4002, Switzerland

European corn borer (ECB), a major pest of maize, has been treated in the past by chemical or biocontrol insecticides or agents. Expression of a CryIA(b) endotoxin from *Bacillus thuringiensis* in transgenic maize affords excellent ECB control under field test conditions. Some advantages of this approach over previous control measures will be indicated. Issues of insect specificity, food safety, pricing, and breeding strategy will be treated. Arguments for and against the possibility of selection of ECB resistant to CryIA(b) will be discussed, as well as strategies for delaying and combatting such resistance if it does occur.

I 04 The Role of Biological Control in Pest Management in the 21st Century

R. JAMES COOK
USDA-ARS, Root Disease and Biological Control Unit, Washington State University, Pullman, Washington 99164

The use of chemical control in pest management is a maturing technology, whereas biological control is still in the infancy stages of development in spite of its long history of use. Integrated pest management is the use of the most effective, economical, safest, and sustainable combination of physical, chemical, and biological methods to limit the effects of pests and improve the yield and quality of plant and animal products. Biological control, particularly the use of natural enemies (or antagonists) of arthropod pests, weeds, plant pathogens, and plant parasitic nematodes, or the use of genes for biological control traits transferred from these beneficial organisms to crop plants, can become the major component of pest management of the 21st century. However, this will only be possible if ways can be found to apply and/or manage populations or genes of thousands if not tens of thousands of site-, pest-, crop-, and/or animal-specific biological control agents. Precedents for this approach have been set with the deployment of thousands of environment-specific crop cultivars and disease-specific vaccines. Moreover, the successes and experiences to date make clear that biological control as the centerpiece of pest management is a technical possibility with no significant compromise in the quantity or quality of plant or animal products, the environment, or the expectations of society. The more urgent issues are whether there is the institutional infrastructure, scientific resources, and socioeconomic resolve to develop this still largely untapped biological and genetic resource in time to make this vision a reality in the 21st century.

II 01

Viral Satellites, Molecular Parasites for Plant Protection

J. M. KAPER, Molecular Plant Pathology Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705.

Viral satellites are small nucleic acid molecules naturally associated with certain plant viruses to which they are sequence-unrelated but which they need for replication support. Satellites are also known for their capability to modify viral symptoms. They can be considered molecular parasites of the viruses they are associated with. There has been a rapidly rising interest in their use for the biologically based control of viral crop disease. Satellites introduced into plants via preinoculation have proven to be effective in large-scale protection of field crops from virus infection. Satellite-transgenic plants with improved resistance to virus disease are under intensive study. Cucumber mosaic virus (CMV) satellites have been at the leading edge of these technologies. This paper defines the replication dynamics and the nested parasitic relationship of satellites and CMV in a susceptible host plant, and shows how satellite-based control of viral disease is emerging as a novel technology for crop protection.

II 02

Viruses for Control of Arthropod Pests

P. V. VAIL
Horticultural Crops Research Laboratory
ARS, USDA, Fresno, CA 93727

The baculoviruses have been the most studied of the many viruses isolated from arthropods. In many cases these DNA viruses are highly virulent and have been demonstrated to be efficacious. Four have been registered by the U.S. Environmental Protection Agency for control of agricultural and forest pests. Others are candidates for registration. Development of baculoviruses to this state has depended on many disciplines including virology, production systems, formulation and applications technology, and microbial control/pest management specialists. Because of their lack of direct effects on other biological control agents they are good candidates for insect control in IPM systems. In order to completely utilize their potential, several areas of research need to be explored.

II 03

Biological Control of Diseases and Frost Damage Caused by Epiphytic Bacteria

S. E. Lindow
Department of Plant Pathology, University of California, Berkeley, CA 9720.

The population size of epiphytic strains of bacteria such as *Pseudomonas syringae* determine the probability of occurrence of disease or frost injury, given suitable environmental conditions. The development of high populations of these bacteria can be reduced by the establishment of antagonistic bacteria on plants prior to the arrival of a target strain. Antagonism of epiphytic *P. syringae* strains is due primarily to preemptive competitive exclusion due to prior acquisition of limiting resources by antagonistic bacteria on leaves. To maximize competitive exclusion, the antagonist and target strains must compete for the same limiting resources on leaves. A single antagonist may therefore not be capable of excluding all strains of *P. syringae* from leaves if this species is ecologically diverse. To test the functional diversity of Ice$^+$ *P. syringae* strains recombinant Ice$^-$ derivatives of *P. syringae* were established in high numbers on potato leaves in the field, and the occurrence of Ice$^+$ strains on the same leaves was measured. Isogenic Ice$^+$ *P. syringae* strains were excluded from leaves better than the populations of indigenous Ice$^+$ strains, suggesting that strains differ in competitive interactions on leaves. Interactions of these strains in the greenhouse using deWit replacement designs reveal that all *P. syringae* stains compete for the same limiting resources but that they differ in their efficiency in acquiring these resources. In contrast, some species of *Xanthomonas* and other species can nearly completely coexist with *P. syringae* strains on leaves.

II 04

Bacterial Control of Flies in Livestock Operations.

E. T. SCHMIDTMANN, D. W. Watson and P. A. W. Martin
Livestock Insects Laboratory, BARC-East, ARS, USDA, Beltsville, MD 20705
Department of Entomology, Cornell University, Ithaca, NY 14850
Insect Biocontrol Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705

The association of immature muscoid flies with bacteria is reviewed relative to the development of a biologically-based management strategy for suppressing muscoid fly populations on dairy farms. Bedding calf hutches with materials alternative to straw, such as inorganic sand and gravel or microbe-limiting ground corncob and sawdust, suppressed immature house and stable fly density from 50 to 99 percent. We also report on the presence of spore-forming bacteria, Bacillus spp., including numerous isolates of insect-pathogenic B. thuringiensis, in calf hutch bedding. Bioassay of this naturally-occurring germplasm indicates that at least several isolates have activity toxic for house fly larvae, hence potential for use in suppressing muscoid fly populations.

II 05 Fungi and Fungus/Bioregulator Combinations for Control of Plant-Parasitic Nematodes.

S. L. F. MEYER and R. N. Huettel
Nematology Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705

Development of new management strategies for control of plant-parasitic nematodes has become increasingly important as effective chemical nematicides have been removed from the market or restricted in use. Biological control organisms could be applied as part of integrated pest management programs, but few are available for commercial applications. Bioregulatory compounds such as pheromones have been utilized for insect control, and may also prove useful for reduction of soil-borne nematode populations. Studies on a mutant fungus strain with enhanced biocontrol potential, on bioregulatory compounds, and on combinations of these agents are being conducted as part of the search for novel means of nematode management.

II 06 Arthropods for Suppression of Terrestrial Weeds.

RICHARD D. GOEDEN
Department of Entomology, University of California, Riverside, CA 92521

The biological control of terrestrial weeds with arthropods in the continental United States and Canada is reviewed from its inception in California during the late 1930's to date. A total of 165 species of natural enemies (insects, mites, nematodes, and phytopathogens) has been released for the biological control of 75 species or congeneric species groupings of weeds. Of these, 117 species have been arthropods, including 114 species of insects and three species of mites. The overwhelming majority of these releases comprised exotic species of insects intentionally introduced for classical biological control of weeds. Major developments, trends, and actions involved in the growth and maturation of biological control of weeds with insects are highlighted and discussed.

II 07 Introduction of Natural Enemies for Suppression of Arthropod Pests

T. S. BELLOWS,
Department of Entomology, University of California, Riverside, CA 92521

Introduction of predaceous and parasitic arthropods (natural enemies) for suppression of arthropod pests has been conducted for more than a century, and currently is practiced throughout the world. The targets of such programs usually have been agricultural pests, but also have included pests of urban settings. The approach has been applied to arthropod pests in many taxonomic groups and has met with substantial success. Approximately two-thirds of all attempts at suppressing arthropod pests have been successful, with half of these resulting in complete control of the target organism. Estimates of economic return on research and development costs of biological control programs are approximately 30:1. The use of arthropod natural enemies is not without risk, although these risks are generally minimal in regards to the physical environment.

III 01 Multi-faceted Biological Control of Postharvest Diseases of Fruits and Vegetables

C. L. WILSON and A. El Ghaouth
USDA, ARS, Appalachian Fruit Research Station, 45 Wiltshire Road, Kearneysville, WV 25430

Public concern over food safety has placed the use of pesticides to control postharvest diseases of fruits and vegetables under scrutiny. A National Academy of Sciences report (National Research Council, 1987) indicates that fungicides pose 60% of the oncological risk from the use of pesticides on our food — a greater risk than insecticides and herbicides together. It is clear that effective alternatives to synthetic fungicides for the control of postharvest diseases of fruits and vegetables are needed. Utilizing a broad definition of biological control, a variety of alternatives to synthetic chemicals for the control of postharvest diseases of fruits and vegetables are available. Among these are the use of: (1) antagonistic micro-organisms; (2) natural plant- and animal-derived fungicides; and (3) induced resistance. Often these methodologies are not as effective alone as synthetic fungicides. However, when utilized in combination, additive and synergistic control can be realized. It is argued that a muti-faceted biological approach toward the control of postharvest diseases is desirable. Evidence is presented to validate this thesis, and suggestions are made for future research.

III 02

Biological Control of Plant Parasitic Nematodes with Plant-Health-Promoting Rhizobacteria.

R. A. SIKORA and Sabine Hoffmann-Hergarten. Institut für Pflanzenkrankheiten, Universität Bonn, Nussallee 9, 5300 Bonn 1, Fed. Rep. Germany.

Plant parasitic nematodes are obligate parasites dependent on the presence of a host plant for development. A strategy for the biological control of nematodes has been developed that is based on the introduction of bacteria colonizing the rhizosphere of the host plant. These plant-health-promoting rhizobacteria (PHPR) adversely influence the intimate relationship between the obligate parasite and its host. The PHPR applied to seed or planting material before planting regulate nematode behavior and parasite developmental cycles during the early root penetration phase of parasitism. Significant levels of control have been obtained with the sugar beet and potato cyst nematodes. Two mechanisms of action are thought to be responsible for reduction in nematode infection 1) production of metabolites which reduce hatch and attraction and/or 2) degradation of specific root exudates which control nematode behavior. Biotic and to a lesser extent abiotic factors influence efficacy.

III 03

Dynamics of *Sporidesmium*, a Naturally Occurring Fungal Mycoparasite.

P. B. ADAMS and D. R. Fravel, USDA-ARS, Biocontrol of Plant Diseases Laboratory, BARC-West, Beltsville, Maryland 20705.

Lettuce drop, caused by *Sclerotinia minor*, was controlled under production conditions by the soil-inhabiting fungus *Sporidesmium sclerotivorum*. In nature, *S. sclerotivorum* is an obligate mycoparasite of sclerotia primarily in species of the Sclerotiniaceae and can survive for at least 5 years in the absence of hosts. Macroconidia of *S. sclerotivorum* germinate in response to the presence of host sclerotia. Mycelium of the mycoparasite can grow out from infected sclerotia as much as 3 cm and infect healthy sclerotia. During this extra-sclerotial growth, an average of 1.5×10^4 new macroconidia of *S. sclerotivorum* are produced. The extra-sclerotial growth and spore production result in spread of *S. sclerotivorum*. Since *S. minor* occurs in an aggregate distribution in soil, infection of a single pathogen sclerotium within an aggregate will ultimately result in destruction of all sclerotia in the aggregate. Most sclerotia of *S. minor* are produced above-ground on lettuce at the end of the growing season. Hence, spraying macroconidia of *S. sclerotivorum* on the crop residue before disking or plowing results in contact of the biocontrol agent with several sclerotia of *S. minor*. By exploiting features of the biocontrol agent, pathogen, host and cropping system, control of lettuce drop was achieved with 0.2 kg/ha of *S. sclerotivorum* applied. *S. sclerotiorum* is thought to be involved in the natural regulation of several diseases.

III 04

Managing Soilborne Plant Pathogens with Fungal Antagonists.

ROBERT D. LUMSDEN, Jack A. Lewis and James C. Locke, USDA-ARS, Biocontrol of Plant Diseases Laboratory, Beltsville, MD 20705.

Agricultural use of commercially available biocontrol products to manage soilborne plant pathogens is an emerging technology. The potential has been shown for fungi to parasitize plant pathogens, form antibiotic metabolites, or compete spatially and nutritionally for ecological niches. Factors are discussed that affected discovery, efficacy, formulation, product acceptance, and registration, for *Gliocladium virens*, the first fungal product developed for greenhouse use in the U.S. (trademark name "GlioGard"). Problems of practical application in the past include variability and inconsistency in product performance. These difficulties were largely overcome by strain selection and innovative formulation and delivery systems for this biocontrol fungus. Additional improvements will be made easier by elucidating the mechanism of action of this and other biocontrol fungi and studying interactions with plant pathogens *in situ*. Saprophytic colonization of organic substrates by *G. virens*, growing from GlioGard™, granules, results in production of the antibiotic gliotoxin. This antibiotic was detected in soil and soilless media amended with GlioGard™. Gliotoxin is active against the soilborne plant pathogens *Pythium ultimum* and *Rhizoctonia solani*, suppressing damping-off diseases caused by these pathogens. Attempts to manipulate the amount and duration of production of gliotoxin by *G. virens* through formulation improvements and genetic manipulation will promote extended applications of the biocontrol fungus for suppression of other soilborne plant pathogens especially in field applications where variability is a problem. Fungal biocontrol agents can, in large measure, replace or reduce the use of synthetic chemical fungicides in the future.

III 05

Conservation of Arthropod Natural Enemies to Control Insects.

J. D. DUTCHER
Coastal Plain Experiment Station,
Entomology Department,
University of Georgia, Tifton, GA 31793

Insect pest outbreaks occur when indigenous predator and parasite populations are reduced and pest populations operate at high rates of increase. Conservation of natural enemies by avoiding the negative effects of: insecticides; reduced plant diversity; secondary predatory generalists; and, secondary parasites, have become increasingly important to agriculture following the era of chemical based crop protection. Orchard systems are conducive to the conservation of natural enemies for several reasons. Plants and insects have coevolved for thousands of years in the native range leading to a diverse set of natural enemies regulating a set of primarily mono- to oligophagous plant pests. Tree and pest phenologies are highly predictable. Trees have a remarkable ability to withstand insect damage and remain productive over a century or more of abandonment and re-establishment. Fruits and nuts are high value crops and growers can afford sophisticated monitoring and control systems. Redesign of the orchard systems by replacing protection with integrated pest management strategies has reduced cost of insect control that outweigh the increased costs of monitoring. Integrated pest management has evolved into integrated fruit production.

III 06

Augmentation of Parasites and Predators for Suppression of Arthropod Pests.

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The number and diversity of predators and parasites inherently are limited in ephemeral agricultural and structural settings. Augmentation of natural enemies by propagation and inoculative/augmentative releases is a rational solution to many of the world's most serious arthropod pest problems. Examples of economic success include control of insect and mite pests in greenhouses, Trichogramma spp. for control of corn borers, Ostrinia spp., in corn, parasites for control of filth-breeding flies in poultry housing, Phytoseiulus persimilis Athias-Henriot for control of the twospotted spider mite, Tetranychus urticae (Koch), in strawberry, and parasites and predators for control of insect and mite pests in orchards. Future opportunities include stored grain pests, Colorado potato beetle, Leptinotarsa decemlineata (Say), with predators and a parasite, Edovum puttleri Grissell, and control of the boll weevil, Anthonomus grandis grandis (Boheman), with ectoparasites--particularly Catolaccus grandis (Burks).

III 07

Progress and Promise in Management of Aquatic-Site Vegetation Using Biological and Biotechnological Approaches.

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Introductions of highly selective, non-native biological control agents over the past 20 years have reduced impacts from exotic aquatic weeds such as alligatorweed, waterhyacinth, salvinia, and waterlettuce. Less selective agents such as the grass carp, tilapia, ducks and geese have resulted in moderate to excellent control of native and exotic aquatic plants. However, reliable efficacy has not been achieved for mycoherbicides, even though several have shown good potential in small scale studies. Infection and virulence may be inhanced by various stressors, but this aproach has been examined only sporatically and needs better focus, including basic research on these processes in problematic freshwater macrophytes. In order to expand and better tailor the use of "non-selective" agents (such as the grass carp), improved "herding" methods and possibly insertion of lethal-genes (to limit longevity) will be advantageous. For long-term management, it will be equally important to identify, selection (through breeding and possibly transgenic plants) macrophytes with desirable establishment and canopy characteristics.

IV 01

The Use of Fluorescent Brighteners as Activity Enhancers for Insect Pathogenic Viruses

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Recently, we demonstrated that certain optical brighteners (*i.e.*, selected stilbenes) act as activity enhancers for the gypsy moth NPV. For the last two years, collaborators have demonstrated significant enhancement under field conditions. Moreover, enhancement could also occur with several homologous and heterologous viruses against the gypsy moth, the fall armyworm, and the corn earworm. The use of brighteners as activity enhancers of insect viruses was awarded a U.S. patent on June 23, 1992, and was licensed to two large companies working with viruses as microbial control agents. The importance and impact of these materials was discussed. From a basic viewpoint, the brightener is a tool which will better enable us to understand the role of the host in the host-virus relationship, affecting both viral activity and viral host range. From a practical standpoint, cooperative research is continuing to determine which brighteners act as enhancers in different host-virus systems. The mode of action of these brighteners was discussed, on the basis of present knowledge.

IV 02

Plant Growth-Promoting Rhizobacteria as Agents of Induced Systemic Resistance.

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Plant growth-promoting rhizobacteria (PGPR) are root-colonizing bacteria which benefit plant production by providing plant growth promotion or biological disease control. Effort to elucidate modes of action for PGPR strains have recently been expanded to examine induced systemic disease resistance. Several lines of evidence support the conclusion that select PGPR strains may enhance host defense mechanisms. Some PGPR stimulate plant growth under gnotobiotic conditions, indicating that the host's physiology was altered. Defense-related compounds, such as lignin, peroxidases, and PR proteins, were found to accumulate in PGPR-treated hosts more rapidly than in nontreated controls. Direct experimental evidence that root colonization by PGPR could lead to reduced incidences of diseases caused by distally inoculated pathogens was reported from three laboratory groups in 1991. These systems and subsequent research directions with PGPR-mediated induced systemic disease resistance will be discussed.

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IV 03

Mechanisms Involved in Biological Control of Soilborne Plant Diseases by Rhizosphere Bacteria.

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Biological control of soilborne diseases achieved through inoculation of seed, soil, or planting material with bacterial antagonists has enjoyed success in greenhouse studies and in certain experimental field trials. Nevertheless, adoption by commercial agriculture of modified disease management practices based on the use of bacterial antagonists has been limited. One obstacle to the agronomic application of bacterial biological control agents is the unexplained variation in efficacy from field to field. Three potential sources of variation influencing efficacy of bacterial antagonists will be considered: 1) Variable establishment of the antagonist in the plant rhizosphere, 2) Variable sensitivities of indigenous pathogen populations to biological control, and 3) Variable *in situ* expression by the antagonist of genes critical to biocontrol activity. Reporter gene systems, in which a gene conferring a phenotype that is readily detected and quantified in natural habitats is placed under the regulation of a gene critical to biocontrol activity, provide invaluable tools for studying the *in situ* expression of genes by bacteria occupying microhabitats on plant surfaces. Identification of the key characteristics contributing to biocontrol activity of bacterial antagonists and of chemical and physical factors determining the expression of such characteristics may enable scientists to better predict the efficacy of these biological control agents.

IV 04

Naturally Occurring Disease-Suppressive Soils

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The incidence of several diseases due to soilborne plant pathogens is limited in naturally-occurring disease-suppressive soils through various mechanisms. In most cases, suppressiveness is fundamentally based on microbial interactions between the pathogen and all or a part of the saprophytic microflora. For example, studies of soils suppressive to Fusarium wilts have shown the involvement of populations of fluorescent *Pseudomonas* spp. and nonpathogenic *Fusarium oxysporum*. Competition for nutrients, especially for carbon and iron, competition for infection sites and induced resistance in the plant are the main modes of action of these antagonists.
The microbial interactions responsible for soil suppressiveness are not independent from cultural practices and soil physico-chemical characteristics. However, as it is difficult to manipulate the soil abiotic factors to induce suppressiveness in conducive soils, most attempts towards biological control are based on introduction of selected strains of antagonists into the conducive soils.
The association of a strain of nonpathogenic *Fusarium oxysporum* (Fo47) with a strain of fluorescent *Pseudomonas* sp. (C7) gave almost total control of Fusarium wilts under experimental conditions. However the development and registration of such a biopesticide faces difficult problems.
Another approach towards biological control would be the enhancement of the level of suppressiveness that exists in every soil. But the choice of cultural practices to limit disease incidence is more difficult to manage than the application of a biopesticide.

IV 05

New Options For Insect Control Using Fungi.

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Epizootics in insect populations provide conspicuous displays of the potential of fungal pathogens to control insects. Experience has demonstrated that simple inundative fungal releases do not always provide control; therefore, innovative methods to promote infection are being developed. Classical biological control efforts are focussed on climatically adapted biotypes and long-lived spores. Augmentative fungal releases are also investigating use of diverse spore types as well as strain improvement. Emphasis is directed toward use of materials to protect fungal spores after application, fungal delivery using baits, and application of fungi to adults that subsequently transfer spores to larvae. Activity of fungal pathogens has also been improved through practices that create micro-climates favorable for infection and through prudent use of pesticides that do not inhibit fungal entomopathogens. Our proficiency with manipulation of fungal pathogens for insect control is growing as these new system-specific approaches are developed.

IV 06

Foreign Plant Pathogens for Environmentally Safe Biological Weed Control.

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Use of plant pathogens for biological control of weeds is a rapidly developing part of the weed management arsenal. Research emphasis is on pathogens that are both highly virulent and environmentally safe. Using criteria for risk assessment, we present evidence that, through proper selection of the pathogen and use of scientific evaluation, foreign pathogens for weed control pose no greater risk than endemic pathogens. Evaluations of the rust fungi Puccinia carduorum and P. jaceae are used to describe a process of risk assessment under containment greenhouse and field conditions. Pathogen selection is the first step. P. carduorum and P. jaceae are excellent candidates, because each in nature is an obligate parasite of one or a few host species. Host range determination is the next step, and for these species infection of plants other than the target either did not occur or was minor. A subsequent set of more critical experiments under optimal conditions for infection indicated neither pathogen could be maintained on species other than the target, and plant damage did not always correlate with disease severity (even with certain target host-pathogen combinations). These data and information from the literature suggest the targets of these pathogens are the only suitable hosts; risk to other species is low.

IV 07 The Past, Present and Future of Insect Parasitic Rhabditids for the Control of Arthropod Pests

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University of California, Berkeley, CA 94720

Since the discovery of the first steinernematid (Steinernematidae: Rhabditida) in 1929 and the ability to commercially produce steinernematids and heterorhabditids (Heterorhabditidae: Rhabditida) for the biological control of soil insects, this discipline has expanded worldwide. At present, there are 12 recognized species of *Steinernema* and four of *Heterorhabditis* but this number will certainly increase. Recent analyses indicate that while representatives of both genera have a similar life cycle in relation to insects and have formed similar associations with mutualistic bacteria, they have evolved independently from each other. The ability to cultivate these nematodes on solid and liquid artificial media has given the field an initial impetus but many challenges still remain. Although many opportunities exist for new strain development through the use of biotechnology, we still have not taken advantage of the variation that occurs in natural populations of these nematodes worldwide. Each of the many strains that have been collected have special attributes in relation to host selection and adaptation to physical and biological factors of the environment. For efficient use of these strains against specific insect pests in select environments, databases containing the characteristics of each strain need to be constructed.

IV 08 Biocontrol for Insect Pests of Livestock and Poultry.

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Biocontrol of insect pests of livestock and poultry is of great interest because livestock and poultry production compose a large portion of farm income, and because of insecticide resistance, increasing restrictions on insecticide use, and concern for the environment. This discussion is limited to house flies, stable flies, horn flies, and face flies because they cause the greatest economic loss to the livestock and poultry industry. Although few scientists are working on alternative control methods for these flies, progress has been made in the past 10 years. This review discusses the status of biocontrol research on flies associated with poultry, pastured livestock, and confinement dairy and beef cattle.

V 01 Microbial Metabolites with Biological Activity Against Plant Pathogens

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For many biocontrol agents, production of metabolites such as antibiotics, siderophores and hydrogen cyanide is the primary mechanism of biological control. Evidence of this includes the demonstration that production of metabolites by some agents correlates with biocontrol activity and the purified metabolite or cell-free filtrate can duplicate the effects of the whole agent. Further, some antibiotics produced by biocontrol agents can be isolated from natural habitats. The application of recombinant DNA technology has provided the most conclusive evidence that certain metabolites contribute significantly to disease suppression. The basic approach involves generation of mutants deficient in a specific trait, complementation of mutants with wild-type DNA and comparison of the biocontrol activity of the parent, mutant and complemented mutant. The antibiotics phenazine-1-carboxylic acid and 2,4-diacetylphloroglucinol have key roles in disease suppression by several biocontrol agents. Putative biosynthetic loci for both of these antibiotics have been cloned from Pseudomonas spp. and expressed in other strains. Some of the recombinants show improved biocontrol activity.

V 02 Phytochemical Resources for Pest Management

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Throughout their co-evolutionary history plants and insects have utilized chemistry for offense, defense, symbiotic and commensal interactions. The success with which plants have been able to defend themselves is best recognized by observing that phytophagous insects are in the minority of insect speciation. Less than half of insect orders feed on living plants while the remainder consume, algae, dead wood, leaf litter, nectar or are parasitic and predaceous. Clearly, most insects avoid feeding on living plants because of their physical and chemical counteradaptations. Plant secondary chemical defenses include not only poisons, but a variety of intrinsically non-toxic compounds that disrupt insect growth, development, reproduction and behavior. Insect pheromones employed for communicative purposes have furnished ideal tools for monitoring insect abundance and for reproductive confusion. Studies of basic insect biology and of their chemical-ecological interactions with plants have revealed a rich resource of phytochemistry that promises to supply environmentally pacific methods for pest management.

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V 03 Maximizing Parasitoid Effectiveness in Biological Management Systems with Semiochemicals.

J. H. TUMLINSON amd W. J. Lewis
Insect Attractants, Behavior and Basic Biology Research Laboratory, Gainesville, FL 32604

Chemical cues enable parasitic wasps to locate the eggs, larvae, or other life stages of the insects in or on which they place their eggs. These chemical signals may be produced by the hosts or by the plants on which the hosts feed. New evidence has shown that wasps exploit semiochemicals emitted by plants in response to insect herbivore feeding. The composition of the chemical signal often differs with the host species or the plants on which they feed. The signal may even vary with different varieties of the same plant species. The wasps learn to respond to the different blends of chemicals that indicate the location of their hosts. With a more thorough knowledge of the factors that regulate the foraging behavior of beneficial parasitic insects it will be possible to develop more effective, balanced, ecologically sound systems for biological management of insect pests. Such a system would avoid the "silver bullet" fallacy that has hampered many attempts at biological control in the past.

V 04 Attracticides for the Control of Diabroticite Rootworms.

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An effective integrated pest management (IPM) alternative to the prophylactic application of broad spectrum, highly persistent soil insecticides for the control of corn rootworms is the use of formulated bait granulars or sprays incorporating cucurbitacin arrestants and phagostimulants with minimal dosages of carbamate or organophosphorus insecticides. Such semiochemical-baits applied at 10 kg per ha have given 90 to 100% control of adult Diabroticite rootworms with quantities of insecticides as low as 10 g per ha. The addition of volatile attractants and structural analogues derived from *Cucurbita* blossoms (eg., indole, cinnamaldehyde, cinnamyl alcohol, 4-methoxycinnamaldehyde, and 4-methoxyphenethanol) improved the efficacy of the cucurbitacin-based baits by at least 3-fold.

V 05 Applications of Pheromones for Monitoring and Mating Disruption of Orchard Pests.

R.E. RICE
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Insect semiochemicals in orchard crop systems are used primarily for pest detection and monitoring in IPM programs, and more recently for control by mating disruption of several key Lepidoptera pests. Pheromone trap data coupled with pest phenology models have enabled growers to reduce pesticide applications significantly in some crops such as peaches and nectarines. As examples, models for the oriental fruit moth, *Grapholita molesta*, codling moth, *Cydia pomonella*, peach twig borer, *Anarsia lineatella*, San Jose scale, *Quadraspidiotus perniciosus*, and California red scale, *Aonidiella aurantii*, optimize timing of single pesticide applications to the most susceptible immature stages of the pest. Each model uses temperature thresholds and heat-unit (day-degree) accumulations that are specific for each species. Mating disruption holds great promise as a biorational pest control technique, but requires more monitoring and attention to application details than conventional pesticide use. Research and development of mating disruption in all crops has been constrained by restrictions on field plot size and consequent inadequate replication, by costs of semiochemicals in comparison to conventional pesticides, and application technology (e.g. sprayable formulations vs. hand-applied dispensers).

V 06 Strategies and Tactics for the Use of Semiochemicals Against Forest Insect Pests in North America.

J.H. BORDEN. Centre for Pest Management, Department of Biological Sciences, Simon Fraser University, Burnaby, B.C., V5A 1S6, CANADA.

Five semiochemical-based strategies and 19 tactics are described. Pheromone-based survey and detection tactics are used to monitor many defoliators, shoot and tip feeders and bark and timber beetles. Disruption of communication through wide-scale deployment of lepidopteran sex pheromones is operational for two species, but for others it is suboptimally effective, possibly because of missing pheromone components. The strategy of removing the pest has been successful using: semiochemical-based containment and concentration of bark beetle infestations followed by logging; mass-trapping; and semiochemical-baited trap trees and logs. Modification of behavior by wide-scale deployment of antiaggregation pheromones has been operationally successful for only one bark beetle species. Improvement in tactics may require blends of semiochemicals or integration with silvicultural treatments. Two promising tactics are to use epideictic pheromones produced by lepidopteran eggs and larvae to deter oviposition and feeding. The tactic of semiochemical-induced competitive displacement of bark beetles is the only promising means for using semiochemicals to enhance natural controls. Future commercial use of semiochemicals will depend on relaxed regulatory policies and an industry willing to grow and survive through the development and marketing of minor species-specific products and services.

V 07 Biologically Based Regulatory Pest Management

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The role of regulatory plant protection in preventing the establishment or mitigating the impacts of invading species is discussed. Examples of programs of exclusion, detection and control of quarantine significant pests utilizing an array of biologically based technologies are given. To an increasing extent, semiochemicals, biological control agents, sterile insect techniques and growth regulators form the technological basis on which these programs are conducted. Declining acceptability of chemical control options and intensifying competition for markets for agricultural products with free trade initiatives dictates that novel approaches to exotic pest exclusion be developed. Scientifically derived risk assessments to support decision-making and designation of pest free zones are quarantine approaches of the future.

V 08 Allelopathy for Weed Suppression

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Microbial Products Research Unit
P.O. Box 5677, Athens, GA 30613

A number of biologically active natural products have been isolated from fungi and higher plants that exhibit either selective or broad spectrum herbicidal activity. These secondary metabolites are diverse in structure and select compounds obtained from fungi control growth and development of lambsquarters, johnsongrass, evening primrose, spotted knapweed, and purple nutsedge. Plant derived phytotoxins, which tend to be more broad spectrum in activity, have been isolated from radish seedlings, Podocarpus, and Magnolia. Additionally, some broad spectrum phytotoxins have been isolated from Pythium, Cochliobolus, and Scopulariopsis. A selection of natural products, their structures and their biological activities are described. In many cases these structures are ideal templates for making derivatized, either by synthesis or biotransformation, useful biodegradable herbicides.

VI 01 Expression of Viral Genes and Viral and Antiviral Proteins in Transgenic Plants to Confer Virus Resistance.

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Florist and Nursery Crops Laboratory, PSI, BARC-West, USDA-ARS, Beltsville, MD 20705.

Although natural genes conferring virus resistance have been incorporated into the genomes of some crop plants by breeding, historically control of plant virus diseases has involved numerous, often combined, strategies to provide durable effective resistance. Recent attempts to confer resistance against plant viruses by genetic engineering and transformation with viral and antiviral genes has provided promising additional strategies for virus control. The major approaches that have been developed to generate transgenic resistance include: (i) expression of viral coat protein (full length or truncated) coding sequences to confer "coat protein-mediated resistance"; (ii) expression of untranslatable sense or anti-sense viral transcripts; and (iii) transformation of plants with other viral gene products, such as truncated or full length replicase components. Other more novel approaches include: (i) the use of synthetic antisense oligonucleotides expressed in plants; (ii) transgenic expression of RNAs with catalytic ribozyme activity; (iii) the expression in plants of human interferon genes; and (iv) the expression of mouse antiviral (anti-coat protein or non-structural protein) antibody genes in transformed plants. Descriptions of the various strategies, as well as several considerations as to how these gene sequences may impart virus resistance, will be presented.

VI 02 Recombinant Baculoviruses as Biological Insecticides.

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Baculoviruses and specifically nuclear polyhedrosis viruses are double stranded DNA viruses showing high specificity to many serious arthropod pests. Although they are used as biological insecticides in a few situations, they generally have failed to compete with classical insecticides commercially. A major reason for this failure is the length of time needed for a wild type virus to kill its host insect. Thus, this and several other laboratories have engineered the virus to lead to rapid kill. Two successful approaches involve the insertion of either the gene for the juvenile hormone esterase from the bollworm or the gene for an insect specific toxin from the Algerian scorpion into the baculovirus genome. The resulting viruses are light stable, orally active, and cause a 30% reduction in time to death with an 80% reduction in food consumed. The use of synergists can lead to a 70% reduction in time to kill making the virus competitive with many classical insecticides. The engineered viruses show no deleterious effects on nontarget organisms tested and greater selectivity than either classical insecticides of the toxin of *Bacillus thuringiensis*.

VI 03 Genetics of BT Insecticidal Crystal Proteins and Strategies for the Construction of Improved Strains

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The insecticidal activities of various strains of *Bacillus thuringiensis* (BT) have been ascribed to proteins produced by the bacterial cells in the form of crystalline inclusions (ICP's). Natural isolates of BT commonly contain several different ICP's. Studies on the cloning, sequencing, and homology comparisons of ICP-encoding genes have revealed a broad diversity of these activities. Parallel developments in genetic approaches have shown that the ICP genes are typically localized on plasmids, many of which are naturally capable of self-transfer between strains. This situation has provided a basis for systematic strain improvement programs. Since individual ICP's have markedly different insecticidal activity spectra, the development of improved BT strains is based on the concept of combining the activities from several ICP's together in a single strain. Construction of improved strains using the non-recombinant approach of natural plasmid transfer or recombinant DNA techniques employing BT-derived plasmid vectors will be described, as well as the limitations of these approaches.

VI 04 Genetically Modified Bacteria for Biocontrol of Soilborne Plant Pathogens.

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USDA-ARS, Biocontrol of Plant Diseases Laboratory, BARC-West, Beltsville, MD 20705.

The consistency of biocontrol performance must be improved before there can be an increase in the use of bacterial biocontrol agents for the control of soilborne plant pathogens. Current research efforts are focused on the identification of traits possessed by the biocontrol agent that function in the biocontrol interaction and the way that environmental conditions impact on the expression of these traits. Genetic analysis of biocontrol bacteria indicates that it may be possible to improve their performance through genetic engineering. Attempts at enhancing biocontrol performance include addition of biocontrol traits and modifying the expression of traits important to biocontrol. Genetic strategies for environmental containment of introduced bacterial biocontrol agents must be developed to allay both the real and perceived fears concerning detrimental effects on the ecosystem due to the introduction of genetically altered biocontrol agents. Two approaches to genetic containment are being developed. With the first approach, a genetically debilitated strain that can not effectively compete in the environment is used. The second approach involves the construction of conditional suicide systems that kill the genetically engineered bacterium under certain environmental conditions.

VI 05 Insect Resistant Transgenic Plants

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Monsanto Company,
St. Louis, MO 63198

Genetic engineering of plants to express insecticidal genes represents one of the newest approaches in the development of biologically based pesticides. The most advanced efforts in this area have used genes from Bacillus thuringiensis (B.t.), a traditional biopesticide. Cotton plants expressing genes for lepidopteran specific insecticidal proteins from B.t. have been tested in the field for three years. These plants show a very high level of protection from damage by cotton bollworm, tobacco budworm, pink bollworm and other lepidopteran insects. Current field tests are focusing on the optimization of overall insect pest management approaches that incorporate these transgenic cotton plants, including the development of appropriate strategies to delay or prevent potential insect resistance to B.t. in plants. Using similar approaches we have also developed potato plants that express a coleopteran active B.t. gene. These plants show an extremely high level of resistance to Colorado potato beetle in the field. Most recently, we have expressed a B.t. gene in corn plants to confer resistance to European corn borer. Taken together, these insect resistant crop plants demonstrate the utility and generality of the transgenic plant approach for creating novel biopesticide products.

VI 06 The Genome of Biocontrol Fungi: Modification and Genetic Components for Plant Disease Management Strategies

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Departments of Horticultural Sciences and Plant Pathology, Cornell University, Geneva, NY 14456

A number of techniques are available for modification of fungal genomes of biocontrol fungi, including mutation, protoplast fusion, and transformation. The genome of *Trichoderma* and *Gliocladium* has unusual organization and mechanisms of variation. The nuclei are haploid, cells are polynucleate, and chromosomes may vary in size and number between different strains of the same species. After plasmogamy is induced between two strains by protoplast fusion or anastomosis, nuclei of one strain may be degraded to release small segments of DNA, and these may be integrated into the genome of the other strain, in a process of interstrain gene transfer. The classical parasexual cycle seems to occur rarely, if ever. The genome of *Trichoderma* and *Gliocladium* contains a number of genes coding for different chitinolytic and glucanolytic enzymes that degrade cell walls of target fungi. These enzymes act synergistically, and combinations of enzymes require very low levels of protein to inhibit fungal spore germination, cell replication, and hyphal elongation. Moreover, the enzymes are strongly synergistic with synthetic and natural fungicides, and may increase sensitivity of fungi to the fungicides by >100-fold. Finally, they are also synergistic with the biocontrol bacterium *Enterobacter cloacae*. Genetic sequences coding for these enzymes have been and are being cloned, and their nucleotide sequence is being determined. The enzymes may themselves be useful in the control of animal and plant pathogenic fungi, and the genes may be used to produce transgenic plants resistant to fungal diseases, or to produce transgenic microorganisms with superior biocontrol capabilities.

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[illegible] patterns of biocontrol fungi, including [illegible]

[illegible] lution, [illegible] conformation. [illegible]

[illegible] general org[illegible]

variation. The [illegible] are haploid [illegible]

chromosomes [illegible]

surface of the [illegible]

between [illegible] by protoplast fusion [illegible]

of one strain may be [illegible] DNA

and the [illegible] is integrated into the genome of the [illegible]

a process [illegible]

cycle [illegible]

and [illegible] a number of genes [illegible]

chitinolytic and glucanolytic enzymes that degrade [illegible]

fungi. These enzymes are [illegible] and

combinations of enzymes require [illegible]

inhibit fungal spore germination, cell replication, [illegible]

[illegible] However, the enzymes [illegible]

[illegible] and natural fungicides, and [illegible]

[illegible] by [illegible]

[illegible] biocontrol bacterium [illegible]

sequences coding for these enzymes have been [illegible]

[illegible] nucleotide sequence [illegible]

[illegible] expression be useful in the [illegible]

[illegible] fungi, and the [illegible]

[illegible] plants resistant to fungal [illegible]

[illegible] microorganisms with [illegible]

VI 07 Transgenic Beneficial Arthropods For Pest Management Programs: An Assessment Of Their Practicality And Risks

MARJORIE A. HOY
Department of Entomology & Nematology
University of Florida, Gainesville 32611

Genetic manipulation projects using beneficial arthropods require efficient and stable transformation methods. Knowledge of appropriate promoters and other regulatory elements are required to obtain effective expression of the inserted gene in both space and time. The number of genes of potential value for pest management programs remains limited, primarily to genes for resistance to pesticides or other toxins. Eventually, we will need genes for other useful traits. Before transgenic arthropods can be developed with these traits, we must understand the underlying mechanisms to identify the critical genes involved. rDNA techniques may be more efficient than traditional selection methods but no one has shown that rDNA techniques yield an effective natural enemy for pest management programs. Until this has been achieved, adequate resources and funding will be difficult to obtain because it is high risk research. Risk assessment of transgenic arthropods will add costs in both time and resources, so it will be important to conduct benefit-cost analyses when transgenic beneficial arthropods are used in pest management programs.

VII 01 National Perspective on the Implementation of Bio-Based Integrated Pest Management

Michael S. Fitzner, USDA, Extension Service, National Program Leader, Integrated Pest Management, Washington, DC, 20250-0900

Today, there is a new sense of urgency in the biological control arena. This urgency springs from a new optimism that bio-based pest management technologies are on the verge of becoming commercially viable on a large scale. Decades of research conducted by scientists in the United States and worldwide have expanded our understanding of the agro-ecosystem and its interrelationship with our environment. Most importantly, many bio-based technologies developed through this research are now ready to be integrated into commercial agricultural production systems. The new optimism for biobased technologies is also due to greater interest in the agricultural industry by the general public. These technologies represent a positive response by agriculture to address the food safety, human health, and environmental concerns being expressed throughout the United States and the world. An examination of the major regional or national constraints limiting adoption of bio-based pest control technologies in four agricultural systems (corn/soybean, cotton, tree fruits, and vegetable crops) was a major focus of the 1992 National IPM Forum. The institutional, policy, regulatory, and research/extension constraints identified before and during the forum will be supplemented by a series of workshops designed to focus more closely on the constraints identified by agricultural producers and other end-users. It is important that appropriate administrative and technical action be taken to address these constraints.

VII 02 New Regulatory Strategies for Pheromones. EPA Policies, Interpretations and Rules.

PHIL HUTTON. Product Manager (18), Insecticide-Rodenticide Branch, Registration Division H7505C, Office of Pesticide Programs, Environmental Protection Agency, 1921 Jefferson Davis Highway, Arlington, VA 22202

The Environmental Protection Agency (EPA) is revising certain policies and establishing new rules and procedures regarding registrations and experimental use permits for pheromones. These new policies and procedures are discussed along with a description of EPA's long-term goals for the regulation of semiochemicals.

VII 03 Patents and Intellectual Property: an Overview.

BARRY U. BUCHBINDER
Life Technologies, Inc., P.O. Box 6009, Gaithersburg, MD 20884

The world of intellectual property as it concerns agriculture is changing. These changes come from new technology, relatively minor changes in patent law, and changes in how companies desire to do business. In this talk, we will look at the differences in the various type of intellectual property: patents, plant patents, plant variety protection certificates, trademarks, trade secrets, and copyrights, with special concentration of on patents. We will also explore the changes, their ramifications, and how they will affect public sector research.

VII 04 THE DEVELOPMENT OF GLIOGARD™ FOR DISEASE CONTROL IN HORTICULTURE.

JAMES F. WALTER, W. R. Grace & Co. -Conn., 7379 Route 32, Columbia, MD 21044.

Recently, W. R. Grace & Co.-Conn. has received registration from the U.S. E.P.A for the microbial fungicide GlioGard™. The development of GlioGard™ involved a cooperative research and development agreement (CRADA) with the Control of Plant Disease lab at the USDA. The active ingredient in GlioGard™, Gliocladium virens (GL-21) has been the subject of extensive investigation and field trials which demonstrates it effectiveness against Pythium and Rhizoctonia in research as well as commercial greenhouses. A recent test market shows the utility of GlioGard™ in the bedding plant, vegetable transplant and container plant production.

VII 05 Public Sector Response to Needs, Issues, and Challenges.

ERNEST S. DELFOSSE
National Biological Control Institute, USDA, APHIS Office of the Administrator, Room 538 Federal Building, 6505 Belcrest Road, Beltsville, MD 20723

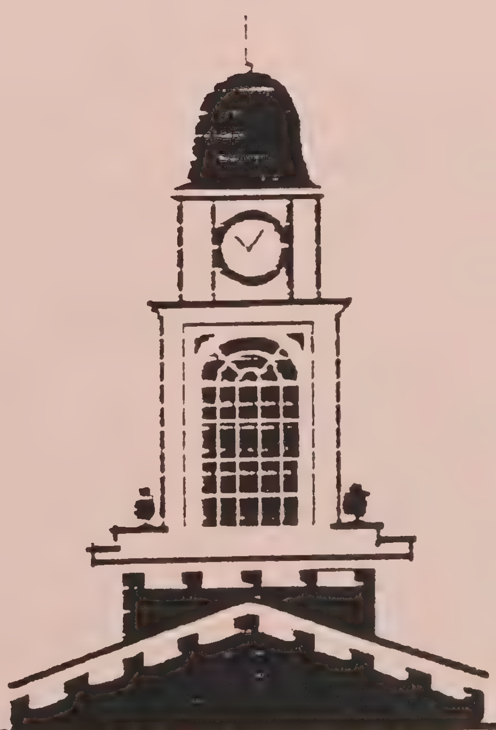
The Federal Government has a critical and multiple role in development and application of biologically-based technologies for pest management. One of the more important needs is an improved regulatory process for biological control. APHIS is renewing regulations to facilitate use of safe biological control agents. A key issue is improving how agencies that have responsibility for biological control can interact more effectively. The Institute is developing strategies to improve communication and implementation of biological control. A main challenge to implementing biological control is to recognize its benefits and limitations. APHIS began to address this challenge by approving a biological control philosophy which states, in part, "APHIS believes that modern biological control, appropriately applied and monitored, is an environmentally safe and desirable form of long-term management of pest species." This philosophy is used as a model of how some of the needs, issues and challenges can be met by the Federal Government, as well as framing policies that safeguard the environment.

VII 06 Look To The Future. WALDEMAR KLASSEN, Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, Vienna, Austria

The paper summarizes some of the Symposium highlights. The Symposium title and several speakers suggest a shift in the paradigms concerning the most fruitful and dominant pathways toward effective and durable ways of coping with organisms harmful in agriculture and to public health. The dominant trend for the foreseeable future is to seek solutions through in depth understanding of biological mechanisms and relationships. The emphasis of the Symposium has been on the development of a variety of methods for coping with pests--many of them novel. Several speakers touched on the need to deploy these methods within the context of strategies that guard against their rapid obsolescence (e.g., the development of resistance), and/or assure the greatest economic benefit. This paper elaborates on considerations in selecting strategies appropriate for deploying biologically based methods. The development and commercial application of these methods is strongly influenced by legal provisions and mechanisms protecting intellectual property. This issue is of overriding importance in the transfer of these technologies among developing countries. The trend in developed countries toward greater private sector involvement in partnerships with public sector institutions for commercial development of biological inventions has increased the disadvantages of developing countries. The Convention on Biological Diversity, signed by more than 150 countries, provides a framework for resolving many of these difficulties. It is likely that progress will be facilitated by the Uruguay round of GATT. Finally, the implications of data pertaining to the growth in markets and the use of biologically based methods are discussed.

ABSTRACTS

VIDEO
AND
COMPUTER APPLICATION
DEMOS



CA01

Fly Management Simulation.
R. C. AXTELL
Department of Entomology,
North Carolina State University,
Raleigh, NC 27695-7613.

Integration of cultural, biological and chemical methodologies in a program for managing manure-breeding house flies in confined livestock and poultry facilities requires considerations of complex interactions which are best visualized with a computer simulation program. FMS (Fly Management Simulator), written in C-language for DOS PC's, provides interacting modules for the house fly (FLYMOD) and its natural enemies: a macrochelid mite predator on fly eggs and larvae (MACMOD), an histerid beetle predator on fly eggs and larvae (CARMOD), and four species of pteromalid parasites (PARMOD) attacking fly pupae. The simulations are temperature-dependent and use variable temperature files which may be provided by the user for local conditions. The user may alter the default values for many biological parameters, such as sex ratios, natural survival, alternate foods, competition, and movement. The user may superimpose the effects of management practices including timing and schedules of manure removal, parasite and predator releases, and the use of pesticides. The toxicities of the pesticide to the fly and its natural enemies, as well as the method and frequency of application and the residual life of the chemical may be specified. Outputs, including all of the values input by the user, are saved in ASCII text files and presented graphically in a screen plot which may be saved also. These interacting simulations of the fly, its natural enemies, and management practices allow predictions of the results of various scenarios so that a practical, effective fly IPM program can be formulated for a particular site.

CA02

A Computerized Data Management and Decision Support System for Gypsy Moth Management in Suburban Parks.

K. W. THORPE, R. L. Ridgway, and R. E. Webb
Insect Biocontrol Laboratory, BARC-East, ARC, USDA, Beltsville, MD 20705

The GYpsy Moth Management Decision Support SYStem (GYMSYS) was developed to address specific data management and decision support needs for gypsy moth management in urban parks and other wooded public lands. These needs include a map-based user interface, simple and reliable data entry, a means to customize the system to a variety of separate and heterogeneous management units and programs, and the use of readily available and inexpensive hardware and software systems. These needs were addressed by incorporating a map image, selected by the user, into the background of the user interface, making it possible for the user to configure the system separately for each individual management unit, and utilizing Apple Macintosh computers and Hypercard software. A knowledge base for making treatment decisions based on egg mass density and size, host tree susceptibility, and defoliation history was encoded into a production rule base.

CA03

Utilization of Computer Molecular Modeling (Chem-X) for QSAR of Trimedlure Isomers

J. D. WARTHEN
Insect Chemical Ecology Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705

This demo illustrates the utilization of computer molecular modeling via Chem-X for the QSAR of the eight, purified isomers (racemic mixtures) of trimedlure. Trimedlure (*tert*-butyl 4- and 5-chloro-*cis*-and *trans*-2-methylcyclohexane-1-carboxylate), a mixture of eight isomers, is used as an attractant for detecting and monitoring the male Mediterranean fruit fly. The relationship between structure and attractiveness is demonstrated by utilizing male medfly field catch on day zero of the individual isomers vs several molecular descriptors: volume, surface area, a torsion angle, and an interatomic distance.

Expert System for the Identification of Fruit Flies (Diptera: Tephritidae) of Economic Importance

CA04

F.C. THOMPSON and L.E. Carroll
Systematic Entomology Laboratory
ARS, USDA, c/o USNM NHB-168
Washington, D.C. 20560

A prototype expert system has been developed to assist non-specialists in the identification of 69 species of fruit fly (Diptera:Tephritidae) of economic importance. This expert system is an interactive, user-friendly, multiple-entry key, which is accompanied by both character and taxon illustrations, as well as help files for both program features and characters. The user may use any character in any order, and may obtain help on character choice from the program. Tentative identifications can be quickly checked by reference to taxon illustrations, and the program will provide lists of diagnostic characters as well as complete descriptions to confirm the identification. The completed version of the expert system will contain data for the identification of some 200 taxa, and is being designed to interface with the Biosystematic Information Database for World Tephritidae, which currently includes 5990 records on taxonomy and nomenclature, 3470 records on host plants and parasites, and 5875 bibliographic records.

CA05 Parental Care Behavior of *Erixestus winnemana* Crawford (Hymenoptera: Pteromalidae) in Eggs of *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) and *Calligrapha* (Coleoptera: Chrysomelidae).

R.F.W. Schroder, A.M. SIDOR and M.M. Athanas, USDA, ARS, Insect Biocontrol Laboratory, Beltsville, MD 20705

Insect parasites exploit a dispersed, disjunct, and mobile resource and are themselves vulnerable to predation and/or parasitism during reproduction. This predation and parasitism is so bad in some insects that the females will not feed in favor of protecting their eggs. This type behavior is very rare among hymenopterous parasites. We report on the first evidence of parental care behavior of Erixestus winnemana, a native pteromalid egg parasite of Calligrapha in the U.S. This behavior is also exhibited on eggs of the Colorado potato beetle. The parasite protects her parasitized eggs from other parasites and hyperparasites.

ABSTRACTS

POSTER PRESENTATIONS

P01 Bacteriophage for control of postharvest diseases. C.G. EAYRE Subtropical Agricultural Research Laboratory, USDA, ARS, Weslaco, TX 78596.

The bacteria Erwinia ananas causes bacterial brown spot of melons, and E. carotovora causes soft rot of potatoes. Phages which attack these bacteria have been isolated from surface water containing agricultural run-off in Florida and another state. Bacteriophages generally have narrow host ranges. For successful biological control of bacterial disease with phages, a phage with a wide host range, or several phages with varied host ranges, are needed. After the phage isolates were purified, host ranges tests were performed using a variety of bacterial strains. The host range pattern for E. ananas and its phages was very different from the host range pattern for E. carotovora and its phages. Phages were found for all E. ananas strains tested, and the host ranges for any two phages either match exactly or did not have any host strains in common. On the other hand, phages were found for many, but not all of the E. carotovora strains tested. Phages of E. carotovora often had overlapping, but not identical host ranges, and clustered into groups with similar host ranges. In preliminary efficacy trials, phages of E. carotovora were found to reduce soft rot of potatoes in a mist chamber. Current work includes testing the efficacy of phages for control of bacterial brown spot of melons caused by E. ananas.

P02 Carbon and Nitrogen Source Utilization by Pseudomonas fluorescens 2-79 for Growth and Antibiotic Formation in Liquid Culture--pH Control Considerations for Mass Production. P.J. SLININGER, M.A. Shea-Wilbur, J.E. VanCauwenberge, and R.J. Bothast National Center for Agricultural Utilization Research, USDA-ARS, Peoria, IL 61604.

Strain 2-79 is a biocontrol agent for take-all, a fungal root disease of wheat. In the rhizosphere, it produces the antibiotic phenazine 1-carboxylic acid (PCA) as the primary means of disease suppression. One barrier to commercial use of strain 2-79 is lack of liquid culture technology for mass producing storable formulations of the bacterium. Batch culture studies were designed to determine C and N source requirements for 2-79 growth and PCA formation. Productions were influenced by pH shifts which varied with C or N sources supplied. When compared in pH 7-controlled fermentors, PCA and cell accumulations were similar on amino acids (AA), urea, $(NH_4)_2SO_4$, and $NaNO_3$, though most rapid growth occurred on AA. Without pH control, ammonium and nitrate caused large pH shifts detrimental to yields. Glycerol, xylose, glucose, fructose, galactose, and mannose supported significant growth and PCA production on urea. Disaccharides lactose, maltose, and sucrose were not utilized. When selected urea-sugar combinations were compared at 16:1 molar C:N (yielding maximum PCA and cells), pH 7 maintenance with automatic acid/base dosing enhanced PCA but reduced cell yields on ~all sugars (on glucose: 2.99±0.33 vs. 4.97±0.72 g/L cells and 0.93±0.14 vs. 0.76±.15 g/L PCA, respectively, were achieved with vs. without acid/base dosing).

P03 Population Dynamics of *Pseudomonas cepacia* in Relation to Control of *Rhizoctonia solani* in Polyfoam Rooting Cubes. D.K. CARTWRIGHT and D.M. Benson. Dept. of Plant Pathology, N.C. State University, Raleigh, NC 27695

A strain of *Pseudomonas cepacia* (isolate 5.5B) effectively controls stem rot of poinsettia, caused by *Rhizoctonia solani*, in polyfoam rooting cubes. The population dynamics of isolate 5.5B or a rifampicin-resistant isolate of 5.5B and subsequent colonization of rooting cubes by *R. solani* was determined. In cubes treated with the wild-type strain of 5.5B, population declined by 52-89, 83-97, 95-98, and 95-99% of the original population after three, seven, 14, and 21 days, respectively. Colonization of the cubes by *R. solani* ranged from 0-0.7, 6-15, 13-28, and 22-42% after three, seven, 14, and 21 days. Colonization of the top 9 mm of the cubes ranged from 54 to 95% after 21 days. In cubes treated with the rifampicin resistant strain, the bacterial population was 34-89, 73-98, 82-97, and 94-99% less after three, seven, 14 and 21 days. Colonization of cubes by *R. solani* ranged from 0-6, 15-22, 22-38, and 31-35% over the same period. Colonization of the top 9 mm of the cubes ranged from 75-100% after 21 days. Colonization of the untreated, infested cubes by *R. solani* was much greater in all tests. Increased colonization of the top 9 mm of the cubes resulted in a greater number of infected poinsettia cuttings. A correlation may exist between population decline of bacterium, colonization of the top of the cubes by *R. solani*, and efficacy of stem rot control.

P04 Exploiting Natural Antagonism in the Genus *Erwinia* for the Control of Bacterial Soft Rot. HAROLD E. MOLINE, USDA, ARS, PPQI, HCQL, Bldg. 002, Rm. 113, Beltsville, MD 20705

Bacterial soft rot of fruits and vegetables is a serious economic problem that reduces both the quality and availability of fresh produce to consumers. Because of increasing consumer resistance to the use of chemical control, new methods of control are being explored. Biocontrol holds the promise of reducing dependency on chemicals, while ensuring an adequate supply of fresh produce. Three *Erwinia* spp. have been recovered from healthy produce that are able to reduce decay caused by soft rot bacteria (*Erwinia carotovora*) in carrot. These antagonistic bacteria appear to act by producing a natural barrier which prevents the decay-causing bacteria access to their source of nutrients, thereby preventing decay. The antagonism seems to be isolate-specific. Not all isolates of the pathogen are affected by a single isolate of the antagonist. Additional studies are needed to determine the feasibility of using these antagonistic bacteria to control bacterial soft rot of fruits and vegetables.

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P05

Development of Neem Seed Oil as a Natural Product to Control Foliar Fungal Diseases.

J. C. LOCKE and J. F. Walter.
Florist and Nursery Crops Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705 and W.R. Grace & Co., Washington Research Center, Columbia, Maryland 21044.

An hydrophobic neem seed oil, solvent extracted from ground, mature seeds of the neem tree (Azadirachta indica) has been clarified and formulated to produce an emulsifiable product suitable for spray application to control a number of foliar fungal diseases. The clarified neem seed oil is a fractionated vegetable oil composed primarily of glycerides (93-95%), free fatty acids (4-8%), and a small amount of inorganic and organic salts. The two major groups of pathogens controlled are rusts and powdery mildews. Control in both greenhouse and outdoor sites has been demonstrated on a variety of crops including; bean, snapdragon, carnation, hydrangea, lilac, and garden phlox. In addition, rose black spot (Diplocarpon rosae) has been significantly reduced in nursery tests. Application rate studies indicate that as low as 0.5 to 1.0% emulsified neem seed oil in water can provide protection equivalent to synthetic fungicides. The mode of action is primarily protective and retreatment is required on a 7-14 day interval depending on the disease pressure and rate of host growth. The activity against powdery mildews also appears to be curative. Although other vegetable oils have shown varying degrees of activity in these tests, none has performed better than the neem seed oil formulation.

P06

Biologically-Based Control of Foulbrood Disease of Honey Bees by Fungal Natural Products.

M. F. FELDLAUFER[1], H. Shimanuki[2], W. R. Lusby[1] and D. A. Knox[2]
[1]Insect Neurobiology & Hormone Laboratory, and [2]Bee Research Laboratory, BARC-East, ARS, USDA, Beltsville MD 20705

Ethanol extracts of mycelia and spores of *Ascosphaera apis*, the causative agent of chalkbrood disease of honey bees was shown to contain an antimicrobial compound active against *Bacillus larvae*, the causative agent of American foulbrood disease. Purification by high performance liquid chromatography and analysis by mass spectrometry identified the active compound as 9,12-octadecadienoic acid (linoleic acid). Various additional saturated and unsaturated free fatty acids were subsequently tested for their antibiotic activity against the foulbrood bacterium. Saturated compounds containing more than 14 carbons were inactive. Antimicrobial activity could be restored to the longer chain length compounds by the addition of one or more double bonds. The use of fatty acids in a prevention and control program aimed at foulbrood disease would be both environmentally sound and cost-effective.

P07

Improvement of a Biocontrol Fungus for Integrated Pest Management Systems by Genetic Manipulation.

SUE MISCHKE, USDA, ARS, BARC, Biocontrol of Plant Diseases Laboratory, Beltsville MD 20705.

Gliocladium virens is a biocontrol agent for several soilborne plant pathogens. Transgenic strains were created by introduction of a mutant *Neurospora crassa* gene for β-tubulin and screened for resistance to benomyl. Although foreign DNA was inserted at multiple sites in their genomes, the expression of phenotypic traits such as growth rate, antibiotic production or enzyme activity was not significantly different from the parent strain. Antagonistic potential of transgenic strains was identified by *in vitro* bioassays, and the ability of a transformant to control Rhizoctonia damping-off of cucumber was tested in growth chambers. The transformant or the parent strain survived similarly in benomyl-free soil however, when co-inoculated, the transformant demonstrated reduced fitness. A transgenic strain which performed at least as well as the parent Gl-21 strain is a potential biocontrol agent suitable for use in integrated pest management situations with benomyl.

P08

Expression Analysis of Gliotoxin Production in the Biocontrol Fungus *Gliocladium virens*.

S.E. WILHITE, R.D. Lumsden, and D.C. Straney
Department of Botany, University of Maryland, College Park, 20782.

Gliotoxin is one of several antibiotics produced by strains of *Gliocladium virens*. Regulation, biosynthesis, and importance of this metabolite in biocontrol are under investigation using strain G20-4VIB (a sub-isolate of GL-21 used in the GlioGard™ formulation). The timecourse of gliotoxin production, as measured by the amount of ^{35}S-incorporated into gliotoxin at intervals between one and four days, demonstrates that synthesis is limited to the initial stages of growth. Mutants lacking gliotoxin production were obtained by a two-step process that involved: (1) uv-treatment followed by a selection process favoring growth of metabolically-altered colonies, and (2) a novel bioassay to screen for gliotoxin-deficient mutants. Nine mutants have been isolated. At least two complementation groups have been identified by cross-feeding. Mutants are being tested for biocontol of *Pythium ultimum* and *Rhizoctonia solani*.

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P09 Alginate Prill Formulations of *Talaromyces flavus* with Organic Substrates for Biocontrol of *Verticillium dahliae*. D. R. FRAVEL and J. A. Lewis. USDA-ARS, Biocontrol of Plant Diseases Laboratory, Beltsville, MD 20705.

Pyrax® (pyrophyllite clay) and milled (0.425 mm pore size) wheat bran, chitin, corn cobs, fish meal, neem cake, peanut hulls, and soy hulls were used to make alginate prill with or without *Talaromyces flavus*. The formulations were compared for their ability to control Verticillium wilt of eggplant in the greenhouse and to increase populations of *T. flavus* in soil. Survival of *T. flavus* in the prill at 5C or ambient temperatures (22-24C), as well as the carbon (C) and nitrogen (N) contents of the prill, were also determined. Three formulations enhanced biocontrol. In treatments without *T. flavus*, half of all plants were wilted by 59 days after transplanting while more than half of the plants treated with *T. flavus* in either Pyrax® or corn cob prill remained asymptomatic 90 days after transplanting. *T. flavus* in soy hull prill delayed the median time for symptom development by about 10 days. Populations of *T. flavus* in soil were greater for the first 4 wk from bran prill compared to other formulations in three different soils. No further differences were detected from 4 wk through 18 wk. Prill with Pyrax® and corn cobs had significantly greater C:N ratios than prill with other substrates. Substrates significantly affected survival of *T. flavus* at 5C and at ambient temperatures. Survival at both temperatures was best in prill formulated with corn cobs, soy hulls and peanut hulls. Temperature did not alter the survival pattern during the 18 wk sampling.

P10 BIOCONTROL OF BOTRYTIS CINEREA ON GREENHOUSE GROWN CONIFER SEEDLINGS Melinda J. Fohn & FRED D. MCELROY. Peninsu-Lab, Poulsbo, WA 98380.

Gray mold, caused by the fungus Botrytis cinerea, is responsible for significant losses of conifer seedlings. The goal of this project was to determine the feasibility of controlling gray mold of conifer seedlings by biological means. Fifty-nine bacterial, fungal and yeast isolates collected from healthy tissue associated with Botrytis infected conifer seedlings were screened on agar plates for ability to inhibit growth of selected Botrytis isolates. Four bacterial, two fungal, and two yeasts were evaluated as biocontrol agents on detached conifer needles and seedlings in a greenhouse setting. Two of the best biocontrol agents as determined by in vitro tests, Trichoderma hamatum and Cryptococcus laurentii, were evaluated in the greenhouse on Douglas-fir seedlings for ability to control two geographically diverse strains of Botrytis cinerea tolerant to two fungicides, thiophanate and iprodione. A single foliar spray of T. hamatum reduced Botrytis infection by 67% compared to Botrytis-only and the fungicide application. Applications of T. hamatum 1 day prior to inoculation with Botrytis and again 14 days later provided an 82% decrease in Botrytis infection compared to 76% in a similar treatment with thiophanate. Three applications of T. hamatum, C. laurentii, and Bacillus megaterium at weekly intervals reduced a naturally established Botrytis infection of 7-month-old Douglas-fir seedlings by 33%, 50%, and 33%, respectively. Similar treatments with thiophanate resulted in a 108% increase in Botrytis infection.

P11 Longevity of Microbial Biocontrol Agents in a Plug Mix Amended with Glomus intraradix S. NEMEC USDA, ARS, SAA, USHRL, 2120 Camden Rd., Orlando, FL 32803.

Commercial sources of Bacillus subtilis, Trichoderma harzianum, Streptomyces griseovirdis, and experimental single isolates of Serratia plymuthica and a Pseudomonas fluorescens parent and its lacZY mutant were mixed into Fafard® Superfine Mix with and without Glomus intraradix. Control treatments were unamended mix and mix amended with G. intraradix alone, and all were put into 128-cell Speedling styrofoam flats and planted with the Sunny tomato cultivar. At 4 to 5 intervals during the growing period (6.5-8 wks) each organism was quantified by dilution plating; G. intraradix infection (%) was determined at the end of each test. Trichoderma isolates increased slightly within two weeks after application and then stabilized through the end of the test. Serratia and Streptomyces declined throughout the test from about 7.75 $\log_{10}$ $cf\mu \cdot g^{-1}$ to numbers at the end (6.2 $\log_{10}$ $cf\mu \cdot g^{-1}$) similar to the controls. Bacillus isolates declined about 1 $\log_{10}$ $cf\mu \cdot g^{-1}$ the first week, but then stabilized. G. intraradix had no influence on numbers of these four genera nor those of their controls. The Pseudomonas parent and its lacZY mutant declined about 1 $\log_{10}$ $cf\mu \cdot g^{-1}$ unit during the test with the mutant higher each sampling period. Propagules of both Pseudomonas isolates were significantly greater with G. intraradix than alone. These data suggest that Bacillus and Trichoderma are the best survivors in a mix for potential use as biocontrols on tomatoes transplanted to the field.

P12 Molecular Phylogenetics and Systematics of Trichoderma (Gliocladium) virens. S.A. REHNER and G.J. Samuels Systematic Botany and Mycology Laboratory BARC-W, ARS, USDA, Beltsville, MD 20705

Trichoderma (Gliocladium) virens is a clonally reproducing soil mold that is a hyperparasite of soil-borne plant pathogenic fungi and is under active evaluation as a biological control agent. The purpose of our research is to determine the relationship of T. virens biocontrol strains to other species of Trichoderma and Gliocladium through evolutionary analysis (cladistic) of nucleotide sequence data. Results of these analyses have revealed the evolutionary position of T. virens and can aid in the identification of novel biocontrol strains. This same approach is being used to determine teleomorphs related to T. virens. A gene phylogeny for a 600bp region of the nuclear large subunit ribosomal DNA has shown that Gliocladium is evolutionarily heterogeneous, elements of which are allied with several teleomorph genera in the Hypocreales. T. virens is shown to be derived from within the genus Hypocrea, whose anamorphs are classified in Trichoderma. Data from ribosomal spacer regions and a single copy nuclear gene, orotidine monophosphate decarboxylase, are being used to infer the relationship of T. virens to species of Hypocrea and their Trichoderma anamorphs.

... USDA-ARS, ... Plant ... Laboratory, Beltsville, ...

pyrophyllite clay) and millet ... corn cobs, fish meal, ... soy hulls were used to make ... *T. flavus*. The formulation ... ability to control ... in the greenhouse and to increase population ... soil. Survival of *T. flavus* in the prill ... temperatures (22-26°C) ... [illegible] ... pH contents of the prill, ... [illegible] ... half of ... plants were ... ing ... than half ... the 90 days after transplanting. *T. flavus* ... days. Populations of *T. flavus* ... the first ... different ... substrates. Substrates significantly ... *T. flavus* ... soy hulls and peanut hulls. Temperature ... survival pattern during the ... months.

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P13 Differential Identification of Wheat Phaeosphaeria Fungal Pathogens by PCR Amplification.
P. P. UENG and E. A. Geiger
Plant Molecular Biology Lab., BARC-West, USDA-ARS, Beltsville, MD 20705-2350

Polymerase chain reaction (PCR) was used to identify Phaeosphaeria nodorum, causal fungus of wheat glume blotch, and to distinguish it from the other related leaf pathogen of wheat, P. avenaria. Stagonospora arenaria, an orchardgrass (Dactylis glomerata) pathogen, and Mycosphaerella graminicola, a wheat leaf blotch pathogen, were included for comparison. Four sets of 20-mers selected from partially sequenced P. nodorum genomic clones were used as primers in the reaction with various genomic DNAs. PCR of P. nodorum genomic DNA with each of the four primer sets resulted in the same products, based on the fragment sizes and the restriction patterns, as the original clones. The primer set 003 specifically identifies P. nodorum, and primer set 103 can distinguish both wheat pathogenic Phaeosphaeria species. Primer sets 062 and 072 can synthesize the same PCR products among wheat Phaeosphaeria species and S. arenaria. It is possible to use these primer sets to monitor the fungal development in plant leaf tissues, and specifically to detect Phaeosphaeria species in mixed fungal infection on wheat.

P14 Integrated Biological and Chemical Control of *Sclerotium rolfsii*. C. D. HOYNES, J. A. Lewis, and R. D. Lumsden. Biocontrol of Plant Diseases Laboratory, BARC-West, ARS, USDA, Beltsville, MD 20705.

The interaction between application of fungal biocontrol preparations to soil and fertilizer or the fumigant metham on survival of sclerotia of *Sclerotium rolfsii* and the damping off and blight the pathogen causes on snap bean are being studied. This includes the feasibility of applying antagonists along with normal rates of fertilizer a week or more before planting to conform with common practices. Generally, ammonium sulfate applied to a loamy sand with fermenter biomass of *Gliocladium virens* (GL-3) prevents disease in the greenhouse as effectively or more effectively than when either component is used individually. Other nitrogen sources decrease, increase, or have no effect on disease compared to the pathogen control. Studies are in progress to determine the effect of liming, soil type, and the nature of the biocontrol fungus propagule on the interaction of fertilizers and GL-3 in disease control. The interaction between the fumigant metham and biocontrol fungi is also being investigated. Conidia of *Trichoderma viride* (TV-1) survive better than chlamydospores in fumigated soil. Studies show that the time of application of the biocontrol agent with respect to soil fumigation with sublethal doses of metham influences the survival of *S. rolfsii* sclerotia. Greenhouse experiments on disease prevention by integrated methods are in progress.

P15 Interstrain Gene Transfer as Mechanism of Asexual Variation in *Trichoderma* spp.
C. K. HAYES, G. E. Harman, and M. Lorito
Departments of Horticultural Sciences and Plant Pathology, Cornell University, Geneva, NY 14456

Parasexuality has been considered for many years to be the primary mechanism by which asexual recombination occurs in Deuteromycetous fungi, and includes the following steps: plasmogamy to form a heterokaryon, then karyogamy to give rise to diploid nuclei containing chromosomes of both strains, and finally, haploidization in which some chromosomes are lost to give rise to a progeny haploid or aneuploid strain containing some chromosomosomes of one strain and some of the other. Rarely, there may be mitotic crossing over to give rise to a chromosome with elements of both strains. In *Trichoderma*, we have prepared and analyzed a great many protoplast fusion progeny, but extensive examination with isozyme, RFLP analysis reveal no evidence for parasexuality. However, protoplast fusion progeny differ remarkably and are unstable, so some genetic alteration must occur. We have evidence for a process of interstrain gene transfer which consists of the following steps: following plasmogamy, most nuclei of one parental strain degrade and release small pieces of DNA. Circumstantial evidence suggests that these are capable of replication. Subsequently, these small pieces of DNA integrate into the genome of the other parental strain. Since cells within thalli of *Trichoderma* are polynucleate and there are many separate DNA segments released from the degraded nucleus, each nucleus of the the progeny may be subtly different from it sister nuclei. Upon selection pressure, nuclei with appropriate genes are selected for; also when conidiation occurs to provide homokaryotic strains, each colony from a separate conidium may be slightly different than those arising from other spores. This process provides a powerful mechanism of adaptation and an unusual procedure for gene flow within the population of this, and probably other related, fungi.

P16 A POTENTIAL BIOCONTROL STRATEGY FOR TRICHURIS SUIS INFECTION IN SWINE, D.E. HILL*, R.H. FETTERER, and J.F. URBAN. BIOSYSTEMATIC PARASITOLOGY* and HELMINTHIC DISEASES LABORATORIES, ARS, USDA, BARC-EAST, Beltsville, MD 20705

A significant decrease in average daily gain and an increase in the feed/gain ratio in pigs accompanies infection with Trichuris suis. Though infective eggs are ubiquitous and long lived, infections are mainly observed in growing pigs. There are few reports on immunity to T. suis in swine. In vitro cultivation of adult T. suis resulted in the production of culture derived excretory/secretory products (ESP). The immunological importance of the ESP was evaluated by immunizing pigs against a combined experimental inoculation and natural exposure. Four groups of 8 pigs each were injected at days 0 and 7 of the experiment as follows: 1) control (uninjected); 2) 2mg ESP in FCA i.m., then 1mg ESP in IFA i.p. (high dose Freunds); 3) 2mg ESP in alum i.p., then 1mg ESP in alum i.p. (high dose alum); 4) o.6mg ESP in alum i.p., then 0.3mg ESP in alum i.p. (low dose alum). Pigs were challenged with 2000 eggs/kg body weight and maintained on a T. suis egg contaminated lot for 52 days. Immunized pigs had increased serum IgG, IgA and IgM antibodies to ESP. Immunized pigs had adult recoveries reduced by 31% in the high dose Freunds group, 86% in the high dose alum group, and 94% in the low dose alum group when compared to unimmunized controls. In addition, immunization with ESP completely eliminated the pathology (weight loss, diarrhea, dehydration) normally associated with trichuriasis in swine.

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P17 Species of the Genus *Pasteuria*, Bacterial Parasites of Nematodes, as Possible Biocontrol agents of Plant Disease.

R. M. SAYRE, Nematology Laboratory, BARC-West, ARS-USDA, Beltsville, MD 20705.

The bacterial genus *Pasteuria*, which consists of an assemblage of mycelial and endospore-forming parasites of nematodes, has some similarities in common with the genus *Bacillus*. The shared traits of their endospores include tolerance to high temperatures, ability to withstand desiccation, and survival for long periods in soils. These endospores, naturally occurring soil, appear to be suitable candidates as biological control agents of plant nematodes. In recent years numerous morphotypes and pathotypes of *Pasteuria* have been observed. This diversity in the *Pasteuria* group indicates a multiplicity of taxa, each being a possible biological control agent of a nematode species. Recent studies on developmental biology of four *Pasteuria* isolates in their respective hosts (i.e., *Meloidogyne incognita*, *Pratylenchus brachyurus*, *Heterodera glycines* and *H. goettingiana*), coupled with documentation of differences in the fine structure of sporangia and endospores, have resulted in establishment of the four following species: *Pasteuria penetrans*, *P. thornei*, *P. nishizawae* and *P. starri*. These four recognized species of the bacterial parasites attack three major nematode pest groups— the root-knot nematodes, root lesion nematodes and cyst nematodes—and offer possible alternatives to our present reliance on chemical nematicides.

P18 Biological Control of Slug and Snail Pests With a Novel Parasitic Nematode

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[1]AFRC Institute of Arable Crops Research, Long Ashton Research Station, Bristol, BS18 9AF, UK.
[2]Agricultural Genetics Company Limited, 154 Science Park, Milton Road, Cambridge, CB4 4GG, UK.

Slugs and snails are important agricultural pests in Northern Europe and parts of N. America; they are especially troublesome in conservation tillage systems and can be particularly damaging to winter wheat, oilseed rape (canola), forage crops and maincrop potatoes. These pests are relatively poorly controlled by current chemical methods, but none of the natural enemies cited in the literature has the potential to be developed as a commercial biocontrol agent. A nematode parasite, *Phasmarhabditis hermaphrodita*, was isolated from UK soils and shown to cause disease in all species of pest slugs tested. The disease in slugs is characterised by swelling of the mantle, inhibition of feeding, and eventually death. The nematode can be mass-produced and formulated using technology developed for entomopathogenic nematodes. In trials the nematode has significantly reduced slug populations and damage to crops and bait plants.

P19 Comparative Studies of Feeding Stimulant Spray Adjuvants with Four Species of Lepidoptera.

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Bldg. 402, BARC-East,
Beltsville, MD 20705

The relative effects of four commercial feeding stimulants, Pheast, Coax, Gusto, and Entice,on four species of lepidopterous larvae, *Lymantria dispar*, *Helicoverpa zea*, *Ostrinia nubilalis*, and *Plutella xylostella* were evaluated. Comparisons were made of feeding stimulants in cellulose-agar media, applied to foliage, and applied to foliage with *Bacillus thuringiensis* (Bt). All feeding stimulants caused feeding on cellulose-agar media, but these effects varied between products and insect species. Similar but smaller differences were seen with material applied to foliage. Feeding stimulants increased mortality due to Bt, with some differences between products. Patterns of differences again varied between insect species. Overall, Pheast most consistently had the greatest effect.

P20 Photoaffinity Labeling of a Transporter for PBAN in the Central Nervous System of a moth, *Helicoverpa zea*.

M.-T. B. DAVIS[1], K. Bhatnagar, G. D. Prestwich, J. Kochansky, R. Wagner, and A. K. Raina.
[1]Insect Neurobiology & Hormone Laboratory, BARC-East, ARS, USDA, Beltsville, MD 20705

Using a radioiodinated azidosalicyclamide photoaffinity analogue of the pheromone biosynthesis activating neuropeptide [^{125}I]ASA-PBAN, we have identified a PBAN binding protein in the central nervous system (CNS), and the pheromone gland of the adult female moth, *Helicoverpa zea* (Lepidoptera: Noctuidae). A 110 kilodalton (kDa) protein in the supernatants of the membrane preparations of the brain-suboesophageal ganglion complex, the thoracic ganglion, the terminal abdominal ganglion, and the pheromone gland was bound specifically to [^{125}I]ASA-PBAN. The specificity of binding was demonstrated by 1) a competitive displacement by 20- to 500-fold excess PBAN, 2) a lack of displacement of labeled PBAN by adipokinetic hormone and Hez-myotropin at concentrations up to 1000-fold excess, and 3) a weak displacement of labeled PBAN by Hez-pheromonotropic-melanizing peptide at concentrations up to 1000-fold excess. The 110 kDa protein recognizes intact PBAN with higher affinity than for partial N- or C-terminal PBAN fragments, as indicated by a lack of strongly competitive displacement of full-length labeled PBAN by N- and C-terminal PBAN fragments. Specific labeling of this 110 kDa protein was observed at the site of PBAN synthesis (the brain-suboesophageal ganglion complex), throughout the CNS, and in the pheromone gland where PBAN has been shown to enhance pheromone biosynthesis *in vitro*. We propose that the transport of PBAN through the CNS is mediated by this 110-kDa transporter. Protein-mediated neuropeptide transport in the CNS appears to be a novel phenomenon in eukaryotes.

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Nematology Laboratory, USDA, ARS, [illegible]
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[illegible] generic genus *Pratylenchus* [illegible] of repeated and unclear [illegible] among [illegible] homonymous morphological similarities [illegible] with the [illegible] The shared use of their [illegible] is temperature [illegible] reproduction and survival [illegible] in culture [illegible], naturally occurring [illegible] as biological control agents [illegible] to morphology and phenotypic [illegible] have been observed. Thus [illegible] the [illegible] because of multiplicity of uses [illegible] have [illegible] control agents of [illegible] nematode species [illegible] developmental biology of four *Pratylenchus* isolates [illegible] respective hosts (i.e., *Meloidogyne* [illegible], [illegible] *brachyurus*, [illegible] [illegible]), coupled with documentation of differences [illegible] structure of sporangia [illegible] [illegible] establishment of the following [illegible] recognized species of the bacterial [illegible] major nematode pest groups [illegible] root lesion nematodes [illegible] possible alternatives to [illegible]

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Using [illegible] monoclonal [illegible] we [illegible] the phenomenon [illegible] [illegible] system (CNS), and [illegible] *Helicoverpa zea* [illegible] 110 [illegible] protein [illegible] the subsets of the [illegible] brain-subesophageal ganglion [illegible] [illegible] the [illegible] cross-reacts with PBAN. The specificity [illegible] was demonstrated by 1) [illegible] to [illegible] excess PBAN, 2) lack of displacement of label [illegible] PBAN or other [illegible] from [illegible] and [illegible] 1000-fold excess, and 3) [illegible] [illegible] labeled PBAN by *Hez*-pheromonotropic-immunoreactive [illegible] concentrations up to 1000-fold excess. The 110 kDa protein [illegible] lower PBAN [illegible] more [illegible] than [illegible]. C-terminal PBAN fragments [illegible] by a lack of strongly competitive [illegible] displacement [illegible] labeled PBAN by N- and C-terminal PBAN fragments. Specific labeling of the 110 kDa protein [illegible] at the site of PBAN synthesis (the brain-subesophageal ganglion complex), [illegible] in [illegible], and in the pheromone gland [illegible] has been shown to enhance pheromone biosynthesis [illegible]. We propose that the transport [illegible] through the CNS [illegible] by the 110 kDa [illegible] [illegible] in the CNS [illegible] novel phenomenon in [illegible]

P21 Peptides in Male Seminal Fluids as Anti-attractants of the Corn Earworm Moth.

T.G. KINGAN[1], W. Bodnar[2], P. Thomas-Laemont[1], D.F. Hunt[2], and A.K. Raina[1] 1, Insect Neurobiology and Hormone Lab, BARC-East, USDA ARS, Beltsville, MD 20705; 2, Dept. of Chemistry, U. of Virginia, Charlottesville, VA 22901

Mating in many species of moths leads to a transient or permanent loss in females of sexual attractiveness and receptivity to new matings. We have shown, in the corn earworm, *Helicoverpa zea*, that a transient (24 hr) loss of attractiveness after mating is associated with a near total depletion from the glandular tissue of her sex pheromone, Z11-hexadecenal. The time required for this loss after separation of the mated pair is ca. 2 hr, while "calling" behavior and the release of pheromone is suppressed immediately after mating. By surgically removing from the (larval) male the testes alone or, from the virgin adult, the testes along with the accessory reproductive glands and duplex (storage site for spermatozoa and seminal fluids), we have shown that the accessory glands/duplex are a necessary source of factors evoking the depletion of pheromone in the mated female. In contrast, a spermatophore alone (devoid of spermatozoa or seminal fluids), is sufficient in evoking the suppression of calling behavior. Extracts of accessory glands and duplexes cause depletion of pheromone in females. Moreover, partially purified pheromonostatic factors are also active in suppressing calling behavior and sexual receptivity in virgin females, suggesting the possibility of redundant mechanisms in evoking the 'mated' state. From extracts of combined accessory glands and duplexes we have purified two polypeptides that are pheromonostatic in females. The amino acid sequence of one, a 57-mer, named "PSP" (pheromonostatic peptide), has been determined. HPLC and mass spectrometry show that the second peptide is nearly identical to PSP. As little as 3 pmol of PSP evokes a 95% depletion of pheromone in our bioassay.

P22 Pheromone Sex Attractant in Male Captures of *Zeuzera pyrina* L. (Lep., Cossidae): Progress Report.

E. PASQUALINI, G. Faccioli, A. Antropoli and M. Molfese.
Institute of Entomology "G. Grandi" University of Bologna (Italy)- 40126-

Various pheromone blends for the capture of *Zeuzera pyrina* L. (Lepidoptera, Cossidae) males were performance-tested in 1990 and 1991 by adding, whether together or separately in various concentrations, the compounds Z2-Z13-18Ac and Z13-18Ac to the main compounds E2-Z13-18Ac and E3-Z13-18Ac (95 and 5%, respectively). The higher catches were obtained with the blend containing Z2-Z13-18Ac (5%), but the difference was not significant. The influence of concentration was tested in 1991 employing a pheromone blend consisting of the two main components only and in 1992 with a three-component blend in dispensers of varying size. At an equal rate (1 mg), the Russell dispensers showed a markedly superior performance to that of the Isagro models, whereas in overall captures the Isagro dispensers baited with 10 mg (small dispenser) proved the same effective. Natural females were by far more attractant than any of the blends (Russell) available at the time of the tests, the ratio being 5.33:1. Both dispenser size and baiting method appear to be of marked importance.

P23 Influence of Guava Ripening on Parasitism levels by *Diachasmimorpha longicaudata* (Ashmead) and Other Parasitoids of *Bactrocera dorsalis* (Hendel)

M. F. PURCELL, C. G. Jackson, J. P. Long and M. A. Batchelor
USDA-ARS Tropical Fruit and Vegetable Research Laboratory, Kapaa, HI 96746

The effects of fruit ripening on the abundance and parasitism levels by *Diachasmimorpha longicaudata* (Ashmead) and other parasitoid species were determined in 3 commercial guava orchards. *D. longicaudata* was released in 2 orchards in Kilauea, Kauai and Hilo, Hawaii. In release orchards, *D. longicaudata* increased to 25% and 54%, respectively of the parasitoids recovered from guavas that were on the ground for 9 and 10 days after harvest. In non-release orchards, *D. longicaudata* accounted for 12% of parasitoids recovered from the oldest fruit. Conversely, less than 1% *D. longicaudata* emerged from guavas collected on the tree or freshly fallen fruit. Psytallia incisi (Silvestri) was absent from tree fruit, but increased to 5% of the parasitoids emerged from 10 day old fruit. The egg parasite, *Biosteres arisanus* (Sonan) was the dominant parasitoid emerging from tree harvested guavas, accounting for 95-98 % of the parasitoids. However, *B. arisanus* decreased 26 - 32% as the fruit aged on the ground. *B. vandenboschi* (Fullaway) was consistently observed from fruit collected on the tree, accounting for 5 % of the parasitoids collected. Large numbers of the eulophid parasitoid, *Aceratoneuromyia indica* (Silvestri), never reported in surveys since its release in the Hawaiian islands, was recovered in guavas that were on the ground 4 to 9 days after harvest. The importance of post-abscission parasitoids for biological control is discussed.

P24 Commercial Examples of "Lure N Kill" Products Using Insecticides and Pheromones

A. R. QUISUMBING and P. K. MacLean
Hercon Environmental Company
Aberdeen Road, Emigsville, PA 17318

Since the mid-1970's, Hercon Environmental has manufactured and marketed controlled release dispensers of insect attractants or insecticides. The latter are used to control household pests. Controlled release attractants, *i.e.*, sex or aggregation pheromones, on the other hand, are used as trap lures for survey, detection, monitoring and mass trapping programs, or as "point sources" to disrupt mating communication. Some recent efforts, however, have been directed towards the development of "attracticides" which are pest management products which combine the insecticide with the pheromone (or a food attractant) in either the same controlled release formulation or end product which can be extremely species-specific, depending on the attractant. Examples of such attracticidal formulations include products for control or suppression of the American cockroach, *Periplaneta americana*; pink bollworm, *Pectinophora gossypiella*; and boll weevil, *Anthonomus grandis*.

P25 Biological Control of Aphids using Parasitoids in European Glasshouses

J.M. RABASSE
Unité de Biologie des Populations
I.N.R.A.- L.B.I., F - 06600 Antibes

Aphids are among the major pests of protected crops. The species are quite diversified but the first targets for biological control are the most polyphagous ones. An indigenous parasitoid, *Aphidius matricariae*, has been successfully used for a long time against *Myzus persicae*, on different crops. Against *Macrosiphum euphorbiae*, on tomatoes, another indigenous species, *Aphelinus abdominalis*, has now been released for 3 years on approximately 50 ha. with good results. The most threatening aphid pest is at the moment *Aphis gossypii* because of its high biotic potential and its resistance to most pesticides. Two polyphagous parasitoids have been tried against this aphid till now: *Lysiphlebus testaceipes*, introduced in France from Cuba in 1973 and *Aphidius colemani*, which also efficiently parasitizes *M. persicae*. Strains of different geographic origins of *A. colemani*, known for their different host specificity, have still to be tested. Both parasitoids proved to be equally efficient but insufficient in the special case of the very fast increasing populations of *A. gossypii* on cucumber in Southern Europe. All these parasitoids are used in the form of inoculative releases of small numbers of individuals as early as possible on low aphid populations. A careful isolation of the rearings from hyperparasitoids is of major importance.

P26 Potential Plant Feeding Attractants for use in Adult Control for Corn Earworm and other Noctuids.

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Crop Insect Pests Management Research Unit
USDA, ARS, SPA, College Station, TX 77845.

During the past few years interest has intensified to identify volatiles from wild and cultivated plants that attract Helicoverpa and other Noctuids to feed or to oviposit. The goal of this project is to identify sources of attractive plant materials, identify volatiles emitted from the attractive plants and to determine the chemical nature of the attractiveness. A free choice olfactometer system has been developed to measure relative attractiveness of flowering plants and emitted volatiles. The olfactometer has been used to confirm field-observed attractiveness of several plant species. Selections of attractive plants for this study were made by direct observations of nocturnal feeding by Helicoverpa and other noctuids and by pollen determination from proboscis of field collected insects. Several chemicals have been identified from hexane extracts of three Guara species (G. longiflora, 24; G. suffulta, 21; and G. drummondii, 12). Also, volatiles obtained have been tentatively identified from Citrus sinensis and Citrus paradisi. All extracts and volatile preparations are being tested for attractancy in the olfactometer.

P27 Plant Hormone-Mediated Insect Resistance in Transgenic *Nicotiana*.

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The bacterial isopentenly transferase (*ipt*) gene involved in cytokinin biosynthesis was fused with a promoter from the proteinase inhibitor II (PI-IIK) gene and introduced into *Nicotiana plumbaginifolia.* Transcripts of the *ipt* gene were wound-inducible in leaves of transgenic PI-II-*ipt* plants. In leaf disks excised from fully expanded leaves, transcript levels increased 25- to 35-fold within 24 hours and by 48 hours were reduced by about 50%. Leaf cytokinin levels in PI-II-ipt plants were elevated by about 70-fold. These plants were used to test for defensive properties of cytokinins against insects. *Manduca sexta* larvae consumed significantly less of the PI-II-*ipt* leaf material than larvae feeding on control plants and *Myzus persicae* nymph development was delayed in comparison to nymphs feeding on normal plants. Exogenous applications of cytokinins further enhanced the level of resistance. The mode of action of the cytokinin gene product on insect feeding is not clear.

P28 Identification of Sucrose Esters from Nicotiana Active against the Greenhouse Whitefly.

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Several species of Nicotiana were observed in greenhouses to be resistant to the Greenhouse Whitefly, Trialeuroides vaporariorum. Preliminary experiments established that materials toxic to the insects could be rinsed off the leaf surfaces of the four most resistant species of the 13 species tested. The assay for toxicity involved topical application of surface component emulsions to whitefly nymphs. Cuticular chemicals of the most effective Nicotiana species, N. gossei, were then fractionated by column chromatography and HPLC in combination with the nymph bioassay to isolate the active insecticidal compounds. The active compounds from N. gossei were identified as 2,3,6'-tri-O- and 2,3,1',6'-tetra-O- acylated sucrose esters with acetyl, 5-methylhexanoyl and 5-methyl-heptanoyl acylation. The isolated sucrose esters were active in causing mortality of early instar greenhouse whitefly nymphs when applied topically. Many of the 65 species of Nicotiana produce sugar esters in different quantities as trichomal exudates with these sucrose or glucose esters having differing amounts of esterification of simple fatty acids of various chain lengths. Related species appear to produce structurally similar groups of sugar esters. However, not all sugar esters have been found to be insecticidal in our investigations.

P29

Neem Tree Compounds for Integrated Control of Mediterranean Fruit Fly Ceratitis capitata.

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A population of Ceratitis capitata (Wied.) (Diptera: Tephritidae), reared in our laboratory, was tested with azadirachtin, extracted from the neem tree (Azadirachta indica A. Juss: Meliaceae). This natural compound produces its insecticide action in different ways, such as antifeedant, interfering with the oogenesis, the larval development and the metamorphosis. Two tests were carried out in laboratory conditions, using climatic chambers (16 hours daylight, T= 27±1°C; RH= 60±5%). A no-choice oviposition and larval development test was set up exposing five new emerged couples in plastic cages, with inside water, one apple, and a dispenser with the solution to be tested. Six different cages (replicated 10 times) were set up: beside the control (sugar-protein solution), one with the neem extract, one without food (only water), and three with different solutions of the neem extract and sugar. In a second test, a choice oviposition and larval development, five couples were transferred in a plastic cage with sugar and protein solution, water, and two apples (one sprayed by a neem solution). The no-choice test showed that complete larval development occurred only in the control and in cage F (sugar and neem separa-ted in two Petri dishes). The choice test indicated up to now that the apple treated with neem can be attacked by the med fruit fly as the control, with probably some differences in the emergence rate.

P30

Site of Action of Fluorescent Brighteners as Enhancers for the *Lymantria dispar* Nuclear Polyhedrosis Virus (LdNPV) in the Gypsy Moth.

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Selected fluorescent brighteners, were found to enhance baculovirus efficacy up to 1000 fold and decrease the LT_{50} by 50% in certain baculovirus-insect systems (Shapiro & Robertson, J. Econ. Entomol. 1992). The gypsy moth, *Lymantria dispar*, and its associated nuclear polyhedrosis virus (LdNPV) were used as the model system. Incubation of both occlusion bodies (OB) and extra cellular virus (ECV) with 1% Tinopal had no effect on virus titer. Combination of either feeding virus and injecting Tinopal or the reverse revealed no enhancement. Likewise, feeding one (virus or Tinopal) followed by various delays with the other agent did not increase efficacy. Both parts of this system were simultaneously required in the midgut. The presence of Tinopal in the hemocoel is doubtful based on a model of hemolymph using Grace's insect tissue culture medium. A significant increase in viremia in Tinopal treated insects was noted when virus titers were measured by *in vitro* assay. Detection of Tdr^3H labeled LdNPV at early times (1-3 HrPI) revealed a non-significant difference of virus uptake in the hemocoel. Autoradiography of tissue sections of larvae infected with Tdr^3H labeled LdNPV showed approximately equal virus uptake in both the cytoplasm and the nucleus of midgut cells. *In situ* hybridization (Genius probes) of similarly treated tissues revealed an absence of virus in midgut cells in quantities significant to produce a positive reaction. Bioassay and REN analysis of progeny LdNPV from control and Tinopal treated larvae failed to distinguish any differences.

P31

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P32

Enhancement of Activity of the Nuclear Polyhedrosis Virus of the Gypsy Moth by Stilbene Disulfonic Acid Adjuvants: Field Confirmation

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The *Lymantria dispar* nuclear polyhedrosis virus (LdNPV) has been registered as a biopesticide since 1978, but high cost of production and delayed kill have limited its actual use. The addition of an adjuvant (one of several stilbene disulfonic acid derivatives widely used as optical brighteners) has been shown to enhance the effect of LdNPV in laboratory tests (Shapiro). To test field performance, formulations of this virus were combined in a tank mix with the adjuvant and applied to forest test plots using ground based equipment. Efficacy against early gypsy moth instars (l^1, l^2) was tested on populations at two different sites: one with a high level of naturally occurring virus (18% to 27% natural mortality) and the other with a low level of natural virus (3% natural mortality). Efficacy against 3rd and 4th instars was assessed at a third site. In all cases, addition of stilbene adjuvant to the tank mix significantly increased percent mortality and speed of kill as measured by LT_{50}.

P33 Construction of a Novel Toxin Delivery System for Biocontrol of Colorado Potato Beetles.

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To develop spiroplasmas as agents for biocontrol of insects, we plan to genetically engineer those spiroplasmas that are gut-inhabiting commensals, such as one that is host-specific for the Colorado potato beetle, to express gut active toxins, e.g., the δ-endotoxin from Bacillus thuringiensis subsp. thuringiensis (Btt). The primary obstacle is the lack of a suitable transformation system, our current research emphasis. To accomplish this we have employed spiroplasma virus SpV1 and plasmids containing gentamycin (Tn4001 in pISM1001) and tetracycline (Tn916 in pAM120) markers to optimize the frequency of spiroplasma transformation by the methods of electroporation, PEG-precipitation and liposome mediated-transformation. Other work will include isolation and characterization of native spiroplasma extrachromosomal elements for use in genetic constructs, construction of a shuttle vector, and development of integrative vectors which exploit homologous recombination-dependent sytems.

P34 Cross-Resistance Among Indianmealmoth Larvae to Specific Crystal Proteins from *Bacillus thuringiensis*

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Resistance has been developed in larvae of the Indianmeal moth (IMM; *Plodia interpunctella*) to several serovars of *Bacillus thuringiensis* that possess a wide variety of crystal protein toxin types. Newly isolated IMM colonies were subjected to continous selection pressure using five different *B. thuringiensis* spore/crystal preparations. Resistance levels following more than 20 generations of selection ranged from 20- to about 300-fold above that of unselected controls. Cross-resistance spectra of each serovar toward the resistant colonies were compared, which tended to reflect the toxin composition of the *B. thuringiensis* serovars used for selection. In addition, the resistant colonies were challenged using individual crystal proteins representing CryIA(a), CryIA(b), CryIA(c), CryIB, CryIC, and CryIIA toxin types. *B. thuringiensis* serovars *aizawai* and *entomocidus* caused a relatively broad-spectrum resistance to all of the individual toxin types, while serovar *kurstaki* produced a narrow-spectrum resistance to CryIA(b) and CryIA(c) toxins. These results suggest that acquisition of resistance in field insects might be accelerated by using a broad-spectrum serovar (such as *aizawai*) and could leave fewer treatment options available once resistance has developed.

P35 Growth parameters for testing interactions between insecticidal bacteria and biocontrol fungi.

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Because of growing interest in using microbials to control pests, we investigated the potential interactions between Bacillus thuringiensis to control insects and Gliocladium virens to control fungal diseases on tomatoes. B. thuringiensis and G. virens have different growth requirements and interactions between these two organisms were dependent on medium and temperature. We initially found interactions ranging from inhibition to synergy. We found that both microbes grew well at 30° on modified potato dextrose agar or an egg yolk based medium. The most interesting interaction involved stimulation of bacterial growth by the fungi. Extracts of fungi showed that the stimulatory activity was preferentially present in the organic extract. This indicated potential for synergistic effects when these two pest control agents are used in the same cropping system.

P36 Comparison of Methods For Quantifying the Efficacy of Aerial Applications of *Bacillus thuringiensis* and Diflubenzuron Against the Gypsy Moth.

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Diflubenzuron and single and double applications of *Bacillus thuringiensis* were aerially applied to four sites in Talbot County, MD in a randomized block design. Treatment effects were estimated from frass (fecal pellet) samples, mortality of larvae collected from the plots, defoliation, counts of pupae under burlap, and post-season egg mass counts. Statistical separation of treatment effects was possible only from frass samples collected 20 or 27 days after the first application. Mortality of larvae collected from the plots, defoliation, and burlap counts were qualitatively similar to the frass counts, and in some cases allowed the statistical separation of treatments and controls, but did not allow the statistical separation of the different treatments. Post-season egg mass counts did not correlate well with the frass samples, probably because of the natural decline of high density populations due to the gypsy moth nucleopolyhedrosis virus.

P37 An Insect Specific Fungus, *Beauveria bassiana*, ATCC 74040, to Control the Sweet Potato Whitefly.

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An insect specific fungus, *Beauveria bassiana* (Balsamo) Vuillemin, ATCC 74040, has pathogenic activity against the boll weevil, *Anthonomus grandis* Boheman, cotton fleahopper, *Pseudatomocelis seriatus* (Reuter), sweet potato whitefly, *Bemisia tabaci* (Gennadius) and other economic insects. Laboratory, greenhouse, and field evaluations of a commercial formulation, Naturalis-L™, against infestations of these insects in cotton, vegetables, and ornamentals have resulted in significant control in studies at our Texas laboratory and with university, federal and private cooperators in California, Arizona, Louisiana, Mississippi, Florida and Texas. During 2 consecutive years of intense insect pressure, cotton was produced with an exclusive Naturalis-L regime. Lint yields was equal to or greater than cotton treated with conventional insecticide programs. An experimental use permit (EUP) with a temporary exemption from tolerance was obtained from the EPA for our 1993 studies.

P38 Potential of Fungal Pathogens for the Control of Pear Psylla.

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Pear psylla, Cacopsylla pyricola L., is the most destructive insect pest of pear in the United States. Chemical control is the primary means of managing this pest but the pear psylla is rapidly developing resistance to many commonly used insecticides. Fungal pathogens of pear psylla and their potential for controlling this pest has not been investigated. Thus, our goal was to investigate the virulence of fungal pathogens to pear psylla. A detached-leaf laboratory assay was developed to evaluate the susceptibility of pear psylla to indigenous and exotic strains of fungi collected from aphids and other psylla species. Two species of Metarhizium and several strains of Beauveria bassiana, Paecilomyces farinosis, and Verticillium lecanii were evaluated. Of these, Beauveria, Verticillium, and Paecilomyces gave 100% mortalities within 5 days after applying spore concentrations of 10^7 spores/ml. regardless of the strain used within species. Both Metarhizium spp. only gave 40% mortalities at the same spore concentration and time. These results indicate that control of pear psylla with fungal pathogens is possible. Further work will be conducted in the field to determine if this is true.

P39 **A Device to Inoculate Flying Insects with Biocompetitors of Toxigenic Fungi, Insect Pathogens or Weed Pathogens.**

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Sap beetles (Coleoptera: Nitidulidae) are adapted to vector toxigenic fungi such as *Aspergillus flavus* and *Fusarium moniliforme*. Toxins produced by these fungi (e.g., aflatoxin, trichothecenes) occur in corn and peanuts, and are a worldwide problem. As a means to reduce mycotoxin contamination in corn, an auto-inoculating device was designed. The device consists of a pheromone-baited trap connected to a cup that allows insects to exit after coming in contact with the biocompetitor. Field experiments using dye demonstrated that insects enter the trap, come in contact with the dye, and exit, carrying the dye to damaged corn ears. In laboratory experiments using two fungal biocompetitors, sap beetles carried a mean of 0.13 mg of *Trichoderma* spp. and 0.9 mg of *Bacillus subtilis* per insect. Insects also carried *Beauveria bassiana* (0.07 mg/insect) and *Bacillus thuringiensis* (0.07 mg/insect). Houseflies and fruitflies also carried dye from the trap. Field experiments using *B. subtilis* as a biocompetitor against toxigenic fungi will be conducted this field season. Traps could also be used for dispersal of weed pathogens.

P40 Entomopathogenic Nematodes for Control of Larvae and Adults of the Sugarbeet Root Maggot (Diptera: Otitidae).

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Entomopathogenic nematodes were evaluated for their ability to infect and reproduce within third instar sugarbeet root maggots (SBRM). Six strains of *Steinernema* representing three species and four strains of *Heterorhabditis bacteriophora* were all capable of infecting and reproducing within third instar SBRM in vitro. Incubation of nematodes at field rates of three billion/acre for 72 h at 24 C resulted in infection and subsequent reproduction of infective juveniles (IJs) within SBRM cadavers. Egress of IJs was observed at 14-21 days post-incubation. Field testing of the six steinernematids indicated that all were capable of infecting SBRM larvae under a typical sugarbeet cropping system. Nematodes were observed to remain viable in soil, as measured by trapping with *Galleria* larvae, for at least two months following application. Adult flies were also found to be susceptible to all six steinernematids tested. *S. glaseri* was found capable of infecting adult SBRM with as little as 2 h coincubation over filter paper containing IJs. Egress of nematodes was observed at 5-6 days post-incubation. Following challenge of diapaused third instar SBRM with steinernematids, pupae were observed to form at an enhanced rate relative to controls. Emerging imagos were found to consist of 25% aberrant individuals. Aberrants had vestigial or absent wings, reduced sclerotization, poor body segmentation, misshapened head capsules and unretracted ptilina. No evidence of nematode infection was observed with these aberrants. They failed to produce eggs when mated to normal flies.

P41 Relationship of the pheromones of some predaceous insects to green leaf volatiles.

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Green leaf volatiles (GLVs) enhance the attractiveness of the pheromones for several phytophagous insects, including boll weevils (Coleoptera), corn earworms (Lepidoptera), and Mediteranean fruit flies (Diptera) (Dickens *et al.*, 1990). Some parasitoids (Hymenoptera) orient to GLVs induced by the feeding of host larvae (e.g. Turlings *et al.*, 1991).

GC-MS analyses and field tests of the pheromones for non-specific predatory bugs (Hemiptera: Heteroptera) indicate that GLVs are synthesized *de novo* as part of the pheromonal blends for these generalized predators. For examples, *(E)*-2-hexenal and α-terpineol released by spined soldier bug males, *Podisus maculiventris* (Pentatomidae), act synergistically as an attractant pheromone; and males of the assassin bug, *Pristhesancus plagipennis* (Reduviidae), produce *(Z)*-3-hexenol and corresponding esters as an attractant pheromone.

P42 Influence of Plants on Insect Predators: a Case Study Using the Insidious Bug.

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Like herbivores, performance of predacious insects may be greatly affected by the food plant of their prey. Yet, plants have rarely been considered in biological control programs. In this study, we compared the performance of the insidious flower bug, *Orius insidiosus* (Say) on three vegetable crops: sweet corn, lima bean, and tomato. *O. insidiosus* is an important predator of thrips, spider mites, whiteflies, and insect eggs and neonates, it is commercially available for greenhouse pest control and it is being considered for use in other systems. Results show that the predator's within plant distribution and searching behavior differed among plant species. Further, *O. insidiosus* females preferred to oviposit in bean and tomato but not in corn. Female oviposition preference seems to correspond with young nymph survival. Whereas newly emerged nymphs survived for about 4 days on bean and tomato plants, they survived for only 2.6±0.5 days on corn. Older nymphs, survived longer on bean than on corn or tomato plants. The ability of nymphs to sustain themselves on plants when prey is scarce may contribute to the efficiency of *O. insidiosus* as a biocontrol agent.

P43 Does the Success of *Euplectrus* Depend on Its Venom Regulation of Host Storage Proteins?

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Investigations were conducted to determine the titer of storage proteins in larvae of the cabbage looper, *Trichoplusia ni* (Hübner), that were parasitized by the ectoparasitoid *Euplectrus comstockii* Howard (Hymenoptera: Eulophidae). A gradual increase was noted in the titer of the storage proteins present in the hemolymph of parasitized third and fourth instar larvae and in the hemolymph of isolated thoracic and abdominal tissues of fourth instar larvae. The final amount present in parasitized third and fourth instar larvae was similar to that found in nonparasitized fifth instar larvae. The stimulation of storage proteins in envenomed larvae demonstrates the ability (competence) of early larval stages to produce a gene product that normally occurs in the last larval stadium of the lepidopteran larval host. The gene expression necessary for storage protein production in isolated tissues may be altered by mechanisms separate from inherent developmental processes and the intact endocrine system.

P44 Rearing of An Endoparasitoid of Heliothis/Helicoverpa Spp. On Atypical Hosts.

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Four potential atypical hosts, Spodoptera frugiperda, S. exigua, Trichoplusia ni, and Galleria mellonella which are less costly to rear than the typical hosts, Heliothis virescens and Helicoverpa zea were studied as potential alternate hosts for Microplitis croceipes. Female parasitoids were induced to oviposit in atypical hosts by treating them with host frass and hemolymph. Oviposition was stimulated at the rate of 1 egg/larva in G. mellonella to 4 eggs/larva in S. frugiperda. In Spodoptera exigua, 100% of the parasitoid eggs were encapsulated 3 days after oviposition; whereas, in T. ni up to 89% of parasitoid eggs were encapsulated at 30° C. Eggs not encapsulated were either undeveloped or the emerging first instar larvae were unable to complete their development. Encapsulation of eggs in S. frugiperda occurred after 6-8 days at a rate of up to 57% at 30° C. A low rate of complete parasitoid development occurred in S. frugiperda with 13% pupating and 12% reaching adult stage. Parasitoid eggs were not encapsulated in G. mellonella; however, up to 77% of the 1st.-3rd instar larvae were encapsulated. Of the unencapsulated parasitoid larvae 24% pupated and 17% emerged as adults.

P45

Status of *Calosoma sycophanta* (L.), an Introduced Predator of the Gypsy Moth.

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Imported from Europe in 1906, the carabid beetle, *Calosoma sycophanta* (L.), became established in New England, where it often has been observed destroying very large numbers of larvae and pupae of the gypsy moth, *Lymantria dispar* (L.). However, no long-term studies have been conducted on the population dynamics of this species, and its current distribution is poorly known. Therefore, we monitored the abundance of *C. sycophanta* (L.) and the gypsy moth in oak stands in southern New Jersey from 1982 to 1992. In addition, adult trapping surveys were made to obtain information of its distribution in the Middle Atlantic States. This species is very well synchronized with the gypsy moth, the adults feeding largely on large larvae in June, and the larvae, on pupae in July. Over time, the predator population responded to changes in prey abundance in a density-dependent fashion; the relationship was direct for larvae and delayed for adults. Spatial responses of predator numbers to prey abundance were weak for *C. sycophanta* adults ($r = 0.431$), but much stronger ($r = 0.709$) for *C. sycophanta* larvae. This differential response appeared to be caused by enhanced reproduction at sites where prey were numerous. The long-lived adults of the predator remained abundant for four years following the year of peak prey abundance. Predation of gypsy moth pupae by invertebrates was correlated positively with abundance of gypsy moth and of *C. sycophanta* larvae. Results of the trapping survey suggest that *C. sycophanta* is missing from much of Pennsylvania, Maryland, and Delaware, and that its distribution lags 100-200 miles behind the leading edge of the gypsy moth infestation.

P46

Cuticular Hydrocarbons of Parasitoids Used in the Biological Control of Stored-Grain Insects.

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Cuticular hydrocarbons were identified from three parasitoids, *Choetospila elegans* (Pteromalidae), *Cephalonomia waterstoni*, and *Laelius utilis* (both Bethylidae) and the hosts on which they were reared (*Rhyzopertha dominica*, *Cryptolestes ferrugineus*, and *Trogoderma variable*, respectively). Effects of gender, life stage, and wing morph were examined for the parasites. In no case did the parasites mimic closely the hydrocarbon profiles of their larval hosts. Some of the parasites showed differences in the hydrocarbon profiles between males and females, whereas others did not. Wing-morph differences were found for *Choetospila elegans*, but no gender based differences were found for this species. The possible ecological roles of the cuticular hydrocarbons on these parasites and their hosts are discussed.

P47

The Definition of Biological Control and its Relationship to Related Control Approaches

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In recent years the advances made in biotechnology have had significant impacts on a variety of disciplines. The incorporation of biotechnological approaches and techniques into efforts to *"biologically control"* pests has resulted in some novel approaches to the control of pest species. The use of transgenic plants which incorporate bacterial genes responsible for the production of toxins lethal to insects, the incorporation of endophytes into plants with similarly lethal properties, and the alteration of insect viruses to carry one or more novel toxins are but a few examples. These approaches, often termed by their developers as biological control. This sentiment has concerned researchers involved in more traditional forms of biological control and has caused significant controversy. This debate has the potential to divide two important parts of the scientific community that inevitably would benefit from collaboration. This initial phase of mutually beneficial interactions may be assisted by a consensus on terminology.

P48

An Effective Parasitoid of the Lesser Cornstalk Borer in Hawaii.

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The eulophid parasitoid, *Horismenus elineatus* Schauf, is now established in Hawaii and is helping to suppress the lesser cornstalk borer (LCB), *Elasmopalpus lignosellus* (zeller). This parasitoid was first released in Hawaiian sugarcane fields in June 1990 and has subsequently been disseminated widely. It is now the most important parasitoid of the LCB.
Field collected larvae of the LCB during 1991 and 1992 indicated that about 25% were parasitized. Also, *H. elineatus* is a gregarious parasitoid with an average of 32.3 individuals produced per LCB larvae. The parasitoid is now widespread and the full impact of its suppressive action on LCB populations should be evident during 1993.

P49 Oriental Fruit Fly Attractants: Effectiveness of Methyl-Substituted Analogs of Methyl Eugenol as Determined from Field Studies.

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Research to develop an environmentally safe attractant to detect and monitor the oriental fruit fly, *Dacus dorsalis* Hendel, was initiated since methyl eugenol (**ME**), the standard detection lure, was reported to be a potential carcinogen. The safety of **ME** is supported by the fact that it is a food flavor, gave a (-) Ames test and is on the GRAS list. Metabolic transformations of natural phenylpropanoids often involve enzymatic oxidation of methylene group and/or epoxidation of the double bond. Hence, modification of the allyl moiety of **ME** is of prime importance. In this study, the effect of introducing a methyl group to various positions of **ME** on attractancy was determined. Relative attractancy was determined from field tests using mean catches related to volatility data. The high attraction observed for three analogs suggested flexibility in the β and γ positions of the allyl moiety for substitution.

P50 Further Structure-Activity Correlations of Medfly Attractants Related to Trimedlure.

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Trimedlure (TML), 1,1-dimethylethyl 4(and 5)-chloro-*trans*-2-methylcyclohexan-carboxylate is an important male-effective lure used worldwide to detect population outbreaks of the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann). Of the three ring substituents, only the ester moiety has been thoroughly investigated in studies correlating structure vs. attractancy. To further define the influence of ring substituents on attractancy, a wide variety of TML analogs reflecting modifications at the 1,2,4 and 5 positions have been synthesized and field tested against the medfly. Data from these tests show that while the 1-*tert*-butylcarboxylate and 4(and 5)-chloro groups can be selectively replaced without significant reduction in attractancy, the 2-methyl substituent is needed for high attraction. Results from extended duration field tests with 14 of the most promising analogs showed that, in four cases, no significant difference in attraction existed between 4-day-old analog-treated wicks and wicks treated daily with TML. (TML treated wicks aged 4-days were almost depleted of lure).

P51 Sterilization of the Gypsy Moth by Disruption of Sperm Release from Testes.

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Knowledge of the mechanisms controlling reproductive physiology of pest insects should provide new targets for designing biologically based pest control methods. In our effort toward this goal, we study the rhythm of sperm release which, when disrupted, causes sterility in male moths.

In gypsy moth males kept in a 16h light: 8h dark photoperiod, release of sperm bundles from the testis displays a daily rhythm which is controlled by a circadian clock at two steps. First, the release of sperm bundles from testis into the upper vas deferens (UVD) occurs only during the 4h period before lights-off. Second, the transfer of sperm bundles from the UVD to the seminal vesicles (SV) takes place within the 2h period after lights-on. Transfer of sperm into the SV is accompanied by a characteristic pattern of contractions by the UVD muscles at this time. Interestingly, in males kept under constant light, this pattern of UVD contractions is absent so that sperm bundles fail to leave the UVD, rendering males sterile.

Rhythmic release of sperm from the testis is initiated several days before adult eclosion, in response to decline in blood ecdysteroid titer. In trying to interfere with the sperm release process by means other than constant light, we have studied the effects of a potent ecdysteroid agonist, RH 5992 which acts specifically on lepidopterous pests. Injection of this compound into developing adults inhibited sperm release from the testis into the UVD in a dose dependent fashion. Nearly complete inhibition of sperm release, lasting for several days, was achieved with 0.3 mM of RH 5992. We plan to study whether sublethal doses of this compound fed to gypsy moth larvae would prevent sperm release in the ensuing adults.

P52 Native Biological Control - The Most Important - and The Most Neglected Component in IPM Programs. An Example in California cotton Pest Control

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Native Biological Control is virtually ignored as a principal mortality component of both key and secondary pest infestations. This is true both historically, and in the present development of IPM Programs. Native biological control is an essential cornerstone for developing area-wide IPM programs particularly in cereal and field crops that serve as extensive sources of natural enemies for high value cash crops like cotton. Examples are given in California cotton pest control programs where multiple insecticide treatments were historically applied to spider mites, lygus bugs, armyworms, loopers and bollworms. Insecticide check and economic threshold studies have been used to demonstrate that insecticide treatments are largely economically unjustified against cotton pests in the central valley of California.

P53

Aspects of Tritrophic Interactions of the Russian Wheat Aphid. **D.K. REED**, J.D. Burd, R.K. Campbell[1] and N.C. Elliott. USDA, ARS, Plant Science Laboratory, Stillwater, OK, 74075. [1]Oklahoma State University, Dept. of Entomology, presently at Plant Quarantine Station, Guam

Classical biocontrol strategies along with resistant germplasm development are now the primary components of an IPM program against Russian wheat aphid *Diuraphis noxia* (Mordvilko). Studies of the interactions between the three trophic levels were initiated using resistant and susceptible wheats, barleys, triticalses and slender wheatgrasses. Tritrophic interactions between these hosts, the RWA and a parasitoid, *Diaeretiella rapae* were studied. Host plants resistant by antibiosis conferred detrimental effects to parasitoids. These included decreased adult emergence, reduced adult size and mummy weight and increased preoviposition periods. Drought stress on plants caused a significantly greater decline in parasitization rates on resistant wheats. A tolerant wheat that had minimal leaf rolling had lower aphid numbers and higher parasite activity, probably being more acceptable for IPM than the highly antibiotic lines. A greater level of coordination between plant resistance and biocontrol programs is needed.

P54

Selective Biological Methods for the Control of the Rice Water Weevil.
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The rice water weevil (RWW), L. oryzophilus, is the most severe insect pest of rice in North America. The larvae of the RWW cause significant damage to the root systems of cultivated rice in the USA. The ultimate goal is the control or suppression of economic plant injury to rice due to insect pests of rice. Biological control measures for the RWW and other insect pests of rice will be explored and developed for field use. Objectives of this research program are: 1) to identify biological control agents (BCA's) i.e., entomopathogens, predators, parasites, effective against rice field insect pests and 2) determine the mechanisms of virulence of BCA's against rice insect pests, and define, clarify, and verify principles and concepts for the use of BCA's against rice insect pests. Adult samples of RWW were collected from overwintering sites and were held in the lab under conditions to identify the presence of diseased individuals. Diseased and dead adults were recovered in a fraction of the adults in the collected samples. A white fungus was observed sporulating from a number of the adult RWW cadavers (presumably Beauveria species). These isolates are currently being investigated as to their pathogenicity to the RWW and their relationship to one another. Insect cell lines to the RWW were establish in serum-free media (Excell 400 and 401) from puparian tissue while larval tissue was used to establish cell lines in M199 (Gibco) containing 10% fetal bovine serum. These cell lines allow for the evaluation of insect viruses as possible BCA's. The control and/or suppression of a destructive insect pest with an environmentally safe, effective microbial insecticide is one goal of this program.

P55

Elcar and Resistant Corn Silks Enhance Mortality of Corn Earworm (Lepidoptera: Noctuidae) Larvae.
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Plant resistance to insects, specifically antibiotic resistance, offers a biologically, economically, and environmentally sound alternative to pesticides for controlling the corn earworm, ***Helicoverpa zea*** (Boddie), in corn, ***Zea mays*** L. Our study included a series of experiments to evaluate the effects of resistant silks incorporated into pinto bean diets, on the infectivity of a nuclear polyhedrosis virus (Elcar) applied to the surface of diet with and without formalin. Neonate, 4- and 8-day-old corn earworm larvae were tested. The compatibility of two biocontrol agents, i.e., resistant corn silks and Elcar, was shown to increase mortality of corn earworm neonates. After feeding on diets containing resistant silks for 4 or 8 days, increased mortality and earlier mortality were associated with lower weight of larvae that were exposed to Elcar. Elcar caused 49% (diet with formalin) and 87% (diet without formalin) mortality of the 8-day-old larvae that had fed on the resistant silk diets as opposed to 0 and 3% mortality for larvae that had fed on diets without the resistant silks prior to treatment with the virus.

P56

Dairy Manure Control of Western Corn Rootworm Damage.
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Research was conducted to evaluate the effectiveness of using manure to reduce lodging and yield losses from western corn rootworm (WCR). Corn hybrids Pioneer 3733 and Cornell 281 were grown in 20 x 60 foot plots treated with 0, 20, 40, or 60 tons per acre of cow manure. Each hybrid/manure combination contained single row plots infested with 0, 500, or 700 WCR eggs per foot or row. Soil CO2 was measured monthly by drawing samples with a syringe from 10, 20, 30, 40, and 50 cm rubber-capped PVC tubes placed within each manure plot. Soil arthropods and nematodes were sampled 9 times throughout the season. Manure did not have a significant affect on root damage, however, root regrowth in the 40 and 60 t/a manure plots was approximately 2 weeks ahead of root regrowth in 0 and 20 t/a manure plots. WCR larval populations were not consistently altered by manure, however, adult WCR emergence was significantly reduced in all manure treatments. Silage yield losses and lodging were reduced or eliminated in manure plots. For all manure rates, Pioneer 3733 was able to tolerate even high density WCR with no observed reduction in grain yield. In contrast, significant grain yield losses due to WCR were reduced or eliminated in manure plots of Cornell 281. Soil CO2 increased with manure level throughout the season and may be connected to increases in root regrowth in manure plots.

P57

Immobilized Microbial Pesticides

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Microorganisms were immobilized or coated inside sterile biogel matrix prepared using agricultural products such as grains, potatoes, beans or their derivatives. Antagonistic microorganisms to plant pathogens or insects were immobilized. They include *Bacillus*, *Pseudomonas*, *Streptomyces* and *Trichoderma*. The spore-forming microorganisms survived over one year at room temperature in the dried gel matrix. One of the *Bacillus subtilis strain* coated in biogel matrix showed good preventative activity against rice blast disease, strawberry grey mold rot, and cucumber seedling damping-off disease.

P58

Hemp sesbania (Sesbania exaltata) Control with the Mycoherbicidal Fungus Colletotrichum truncatum.

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Mycoherbicidal weed control offers effective alternatives to chemical control in some situations. An example is represented by the fungus Colletotrichum truncatum ('COLTRU'), discovered on hemp sesbania seedlings near Stoneville, MS. Hemp sesbania is a serious weed pest in soybeans, cotton, and rice in the lower Mississippi valley region of the U.S. Greenhouse tests revealed that COLTRU is highly virulent, host-specific, and will kill weeds over a wide range of environmental conditions. A moderate period (6-10 h) of free moisture (dew) is required to achieve optimal infection and mortality. However, these dew requirements can be greatly reduced or eliminated by formulating COLTRU in an emulsifiable invert emulsion (EIE). Directed sprays of COLTRU in the EIE applied with air-assist nozzles, or applications of COLTRU formulated in wheat-gluten matrix granules called 'PESTA' have consistently provided effective hemp sesbania control in multi-year field experiments. Weed control levels have approached or equalled those attained with the most effective chemical herbicides recommended. These results demonstrate COLTRU is a biologically effective, ecophilic mycoherbicide for controlling hemp sesbania.

P59

Granules Containing Mycoherbicides and Entomopathogenic Nematodes

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A versatile new formulation concept called "Pesta" has been employed to make granules containing fungal weed pathogens and entomopathogenic nematodes. Wheat flour, fillers, adjuvants, and the biocontrol agent are blended together to make a dough which is then rolled out into a sheet. After drying, the dough sheet is ground into 1-2 mm granules. Pesta granules containing entrapped mycoherbicide agents become covered with new fungal growth after exposure to moisture, resulting in sustained production and release of newly-formed spores. Pesta/Fusarium oxysporum controlled coffee senna, sicklepod, and hemp sesbania in the greenhouse without a dew period. Pre-plant incorporation in soil was the most effective treatment. Pesta/Colletotrichum truncatum controlled hemp sesbania in the field. Pesta granules with entrapped Steinernema carpocapsae soften when wet and the nematodes escape (150,000 to 300,000 per gram) to seek out insect pests. Granules effectively controlled Western corn root-worm in greenhouse tests. Shelf life at 21 C recently has been extended from 4 weeks to about 20 weeks by a formulation additive.

P60

Stability of Colletotrichum truncatum in Culture May Influence Mycoherbicide Potential

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Colletotrichum truncatum, a potential mycoherbicide for control of hemp sesbania, forms spontaneous variants on agar medium. The predominant variant produced more spores than the wild type on potato dextrose agar but the wild type produced more spores on the less nutritious Czapek-Dox agar. Spore yield of both types was greatly reduced on Czapek-Dox. In liquid culture, the variant spontaneously arose and gradually displaced the wild type. Although the variant produced more spores in liquid culture, the variant had reduced virulence. Displacement of wild type strains by similar variants in liquid fermentation, the preferred method of inoculum production, may greatly reduce mycoherbicide efficacy and should be considered during the preparation of C. truncatum inocula.

P61 **Modification of *Colletotrichum* Biocontrol Agents with The Bialaphos Resistance Gene.**

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To overcome the low virulence and narrow host range of some developed or potential mycoherbicides, *Colletotrichum* spp. were transformed with the *bar* gene. The *bar* gene codes for an acetyltransferase that can detoxify bialaphos, a natural herbicide. Successful transformations of *C. gloeosporioides* f. sp. *aeschynomene* (a pathogen of northern jointvetch, *Aeschnomene virginica*) and *C. coccodes* (a pathogen of velvetleaf, *Abutilon theophrasti*) were accomplished using the fungal expression vector pJA4. The transformants demonstrated increased acetyltransferase activity when assayed using acetyl-CoA and glufosinate ammonium as substrates. Five transformants from both *Colletotrichum* spp. displayed stable integration of pJA4 as well as bialaphos resistance (greater than 100 times that of their respective wild types). The five *C. gloeosporioides* f. sp. *aeschynomene* transformants retained their virulence on northern jointvetch. The virulence of *C. coccodes* has yet to be tested. Host and closely related weed species will be treated with the respective transformants and the wild type *Colletotrichum* spp. with and without co-applications of bialaphos to determine the effects on pathogenicity.

P62 **The Influence of Ethylene Pretreatment on Ethylene Biosynthesis in *Nicotiana tabacum* cv Xanthi Treated with a Fungal Xylanase**

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An ethylene biosynthesis-inducing xylanase (EIX) from the fungus *Trichoderma viride* elicited ethylene biosynthesis in *Nicotiana tabacum* cv *Xanthi* leaf tissues. The increase in ethylene biosynthesis was accompanied by the accumulation of 1-aminocyclopropane-1-carboxylic acid (ACC) and an increase in extractable ACC synthase activity. Increased levels of ACC synthase and ACC oxidase transcript were visible within 1 h of EIX treatment. Pretreatment of leaves with ethylene (120 µL/L, 14 h) alone had no measurable effect on ethylene biosynthesis, ACC content, ACC synthase activity or ACC synthase transcript level, but stimulated accumulation of ACC oxidase transcript. Some responses induced by EIX (ethylene biosynthesis, ACC content, ACC synthase activity and ACC synthase transcript level) were greatly enhanced by pretreatment of the leaves with ethylene (12 to 120 µL/L). Eight to twelve hours of ethylene pretreatment were required before the enhancing effects of ethylene were observed. The ethylene pretreatment effect was lost on all responses within 24 h after removing the leaves from the ethylene atmosphere.

P63 ***Bagous affinis*: Biological Control of *Hydrilla verticillata***

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Hydrilla verticillata (hydrilla) is a serious aquatic weed in wetlands in California. In an attempt to biologically control hydrilla, *Bagous affinis* Hustache (Coleoptera:Curculionidae) was imported into California from India in the summer of 1991. The weevil feeds exclusively on hydrilla. The adults feed on hydrilla stems, and the larvae on subterranean turions or tubers during drawdown conditions. The weevils have been released at 2 sites in California in 1991 and 1992. The weevils established temporarily at one site. In addition, the weevil was found to overwinter in northern California.

P64 **Microbial Herbicides for Weeds of Temperate Arable Crops**

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Microbial herbicides have received much attention worldwide as a result of increased focus on integrated pest management and pressure to reduce herbicide usage. Consequently many potential microbial agents have been isolated and a few have been commercialised. None of these are for use in broad-acre arable agriculture on an international scale. Such usage is essential to achieve commercial success.

Long Ashton Research Station has focused on microbial herbicides for use in temperate region arable crops. A long-term programme of isolation, characterisation and establishment of selectivity has identified more than 20 pathogenic fungi with potential for controlling broad-leaved weeds. Particularly promising isolates, strains of Phoma exigua, have been obtained for the selective control of Convolvulus arvensis and Galium aparine, major weeds of several crops in European agriculture. All of the isolates kill their target weeds within 7 to 10 days of application when the weeds are maintained under conditions of high humidity. In natural conditions infection is limited by lack of moisture. Current research emphasises development of formulations to overcome environmental constraints. Experimental formulations of spore inocula in vegetable oil/ water emulsions have significantly reduced dew period requirements and tests of these in field conditions will commence in summer 1993.

P65 Biocontrol of Rangeland Weeds.

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Rangeland weeds targeted for biocontrol in our laboratory include leafy spurge; diffuse, spotted, squarrose, and Russian knapweeds; Canada and musk thistles; St. Johnswort; purple loosestrife; and common crupina. Biological control agents being investigated include introduced insects and nematodes, and extant plant pathogens. Successful biological control of leafy spurge was demonstrated in 1992 after a 1987 release of the copper spurge fleabeetle near Bozeman, MT; leafy spurge canopy cover was reduced from 57% to less than 1%. GPS and GIS have been used to map releases of several species of spurge fleabeetles. Soilborne fungi have been discovered and characterized as agents responsible for fairy rings and disease symptoms on leafy spurge in several states; *Rhizoctonia solani* (AG-4) was implicated. *Agrobacterium tumefaciens* was isolated from Russian knapweed and shown to be highly pathogenic against diffuse knapweed. A seed-destroying weevil, *Bangasternus fausti*, was shown to be complementary to already introduced seed gall flies for diffuse, spotted, and squarrose knapweeds. The Russian knapweed nematode is being mass reared on tissue culture (after methods by A. Watson, McGill U.) and formulated in inverting oil-coated alginate granules for long-term storage and delivery.

P66 The Potential for Biological Control of *Rottboellia cochinchinensis* in the New World.

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Rottboellia cochinchinensis (itch grass) is a pantropical agricultural weed, with an Old World centre of origin. It was accidently introduced into the New World, and it is here that infestations are considered to be the most severe. A collaborative research project has been undertaken to investigate the potential of controlling this weed with fungi; both from a classical and a mycoherbicidal approach. For the classical approach, two obligate basidiomycetes are being considered, a rust *Puccinia rottboelliae* and a systemic head smut *Sporisorium ophiuri*. Both are endemic in Africa and Asia, but absent from the New World. Surveys for non-obligate pathogens resulted in the selection of isolates of *Colletotrichum* sp. nov. near *graminicola* from Asia, for further evaluation. However, the possibilities of importing exotic necrotrophic pathogens into the New World are remote and it is considered that these pathogens could only be used in their endemic range. Nevertheless, the technology developed here could be exploited to develop mycoherbicides for this weed based on adapted neotropical pathogens such a *Fusarium moniliforme*.

P67 Insect Monitoring Systems: Evolutionary Developments in Application.

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Pheromones are used in increasing degrees of sophistication to monitor development of pest populations and have become indispensable as the foundation of robust integrated pest management protocols in horticulture. Prior to commercial implementation, these systems require rigorous season-long testing in each use pattern (pest, crop and geographical region). While acceptance of these systems is continually increasing, wide-spread implementation can only proceed where information is validated in commercial production agriculture. Pheromone companies have limited resources and product development relies on cooperation with the public sector. Will future implementation be compromised through world-wide contraction of public resources?

These monitoring systems have evolved from detection of the pest through to their use in determination of economic thresholds. The position of a pest species on this monitoring time-line is defined by the sophistication of pheromone-based pest management decision making for that species. Increasing commercial use of these monitoring systems will only proceed with development of more temperature driven predictive phenology models, coupled with economic thresholds that allow precision timing of control strategies. This paper will discuss barriers to full utilization of pheromone traps. Future use will expand into area-wide monitoring networks and identification of non-sex pheromone attractants.

P68 Educational Strategies in Support of Biological Management of Pests and Diseases

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Development, acceptance, implementation, and long-term support of biological and biotechnical approaches to the management of pests and diseases will be expedited by effective education of scientists, farmers, manufacturers, agricultural students, legislators, regulators, and the general public. This goal calls for holistic strategies for education on interrelated factors of ecology, genetics, population biology, microbe-plant/animal interactions, soils, technology, environmental quality, energy, sustainable systems, ecosystem management and conservation, and economics. Tactical approaches scaled to specific audiences must build from a basis of clearly presented biological laws and logic and focus on positive examples and long-term benefits so as to correct misconceptions and appeal to reason over emotion. Attention should be given to realistic interim or transitional needs and approaches (such as combined technologies) toward the goal of biological management. Networking and creative problem solving should be integrated with other learning processes. Agriculturalists must take more proactive and participatory roles in general education at all learning levels.

P69 Effects of a Genetically Modified Endophytic Bacterium on Colonized Corn Plants. CECIL F. TESTER, Soil-Microbial Systems Laboratory, Beltsville, MD 20705

The present study was conducted to determine whether the presence of an endophytic bacterium altered the physiological and chemical properties of inoculated corn (*Zea mays* L.) plants and the subsequent decomposition of plant residues in soil. Greenhouse-grown corn (Proprietary hybrid) plants (14 days old) were inoculated with either the endophytic bacterium *Clavibacter xyli* subsp. *cynodontis* wild type strain (MDE1) or a recombinant strain (MDR1.586) transgenic for production of a δ-endotoxin from *Bacillus thuringiensis* subsp. *kurstaki*. Leaf photosynthetic rate, stomatal conductance and transpiration rate of the colonized plants at midday were significantly less than those of the control plants. Leaf surface temperature was significantly higher for colonized than for control plants. Presence of either endophytic strain significantly reduced harvested dry mass of the plants. Leaf residues from plants colonized with MDR1.586 retained significantly more water and contained more C than the controls. Stalks of plants colonized with either endophyte contained significantly less C. Inoculation with either MDR1.586 or MDE1 significantly increased the quantity of N, reducing sugars and some polyamines present in all plant parts. Presence of either endophyte caused no significant differences in decomposition of leaf substrates. In contrast, corn stalks colonized with either strain decomposed significantly more than those inoculated with buffer. On average ca 37% of corn leaf residues and ca 32% of corn stalk residues decomposed.

P70 Control of Codling Moth & Secondary Pests in Pheromone-Based IPM
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Mating disruption is increasingly used in the control of codling moth in apples and pears. Enhanced acceptance will depend on further development and implementation of pheromone-based IPM. Pheromone-based IPM employs mating disruption to effectively manage codling moth. Reduction or elimination of toxic insecticides will enhance biological control of both key and secondary pests. The objective of pheromone-based IPM is to ensure the harvest of a clean crop in an economically viable and environmentally acceptable manner. This objective is best met by employing tactics and strategies within the framework of pheromone-based IPM. In pheromone-based IPM, tactics include the effective use of; (1) mating disruption to manage key pests, (2) monitoring tools and thresholds to assess the pest status of key and secondary pests and, (3) supplementary controls for key and secondary pests. The strategy of a pheromone-based IPM is to "Effectively Manage Key and Secondary Pests in an Economically, Ecologically and Environmentally Acceptable Manner." The acceptance of pheromone-based IPM will depend on the recruitment of informed and committed growers and consultants.

P71 From Research to Reality
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Transferring technology from federal laboratories to the private sector is a nationwide priority that introduces scientific advances and improves the economy. Congress passed several laws to facilitate and encourage partnerships between federal laboratories and business. The goal of this project is to explain to ARS scientists the complexities of the technology transfer process and to furnish information on ways to recognize how basic research developments can become practical products or processes for industry. Before technology transfer can occur an information transfer is needed between the research scientist, a technology transfer facilitator, and business. Three main elements of technology transfer are Cooperative Research and Development Agreements (CRADA), the patent program, and licensing of intellectual property. In some cases, ARS research can and does receive patent protection. A close relationship between the scientist and ARS Patent Advisors may enhance patentability of federally-developed technologies. Patent protection is an incentive to business for investments made under license arrangements. Career enhancement, peer recognition, and royalty income are benefits gained by the scientist from a successful transfer of ARS technology to the private sector.

P72 Ecological Basis of Biological Control of Frost Injury.
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Biological control of frost injury may be achieved by the pre-emptive exclusion of ice-nucleating (Ice$^+$) *Pseudomonas syringae* strains by recombinant non-ice nucleating (Ice$^-$) *P. syringae* strains. In field experiments, indigenous Ice$^+$ *P. syringae* strains were not completely excluded by the Ice$^-$ strain, suggesting that Ice$^+$ *P. syringae* strains exhibit variability in ecological niche requirements and may coexist to different extents with the Ice$^-$ strain. Replacement series experiments, in which *P. syringae* strain pairs were inoculated onto leaves in different ratios but at a constant total density, were used to examine interactions between the recombinant Ice$^-$ *P. syringae* strain and various Ice$^+$ *P. syringae* strains. All of the *P. syringae* strain pairs examined competed for limiting nutritional resources, which were partitioned according to the relative competitive ability of the strains. There was no evidence of niche differentiation among these strains. *P. fluorescens* A506, registered as the biological pesticide FrostbanB, competed for nutritional resources required by *P. syringae*, but was itself able to utilize additional resources. While there was no evidence of niche differentiation among these *P. syringae* strains, if variability in the ecological niche of Ice$^+$ *P. syringae* strains does occur, it could be countered by the use of either a mixture of Ice$^-$ strains with complementary resource utilization profiles, or a strain such as *P. fluorescens* A506 which has a more extensive resource utilization profile than the Ice$^-$ *P. syringae* strain.

P73

The National Environmental Policy Act and Biological Control.

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The National Environmental Policy Act (NEPA) requires that federal decisions be informed by environmental considerations. Agencies are instructed to integrate the NEPA process with other planning at the earliest possible time to ensure that planning and decisions reflect environmental values, to avoid delays later in the process, and to forestall potential conflicts. NEPA is procedural, not deterministic; choice of the environmentally preferred alternative is not mandated. NEPA is typically used as an "umbrella" statute to provide a framework for overall environmental compliance. U. S. Department of Agriculture, Animal and Plant Health Inspection Service (APHIS) believes that modern biological control (BC), appropriately applied and monitored, can be an environmentally safe and desirable form of long-term management of pests. APHIS provides funding for BC research, regulates certain BC agents, and implements BC programs. Early involvement of NEPA facilitates each of these activities by insuring that plans and decisions are environmentally sound and by formalizing and documenting APHIS compliance with NEPA and other environmental regulations.

P74

Evaluation of Botanical Products for Innovative Plant Disease Control.

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The efficacy of several botanical products against foliar and soilborne pathogens was demonstrated when applied prior to inoculation. Foliar application of 1% (v/v) neem seed oil (NSO) controlled *Puccinia antirrhini*, *Uromyces dianthi*, and *Uromyces appendiculatus* on snapdragon, carnation, bean, respectively, and *Erysiphe polygoni* on both hydrangea and bean. NSO at 1% was 81-84% more effective than Sunspray 6E or soybean oil against *P. antirrhini*. NSO, Sunspray 6E, and Funginex (6.5% Triforine) equally reduced the number of *U. appendiculatus* pustules/leaf. Vernonia seed oil (VSO) and NSO at 1% equally suppressed *U. dianthi* on carnation and *E. polygoni* on bean. When all were applied at 14 day intervals, 1% NSO and VSO performed better than Funginex and Sunspray 6E against *E. polygoni* on bean and 2% VSO controlled *E. polygoni* on hydrangea as well as 1% NSO. *In vitro*, 1% NSO reduced severity of blackspot (*Diplocarpon rosae*) on rose compared to the pathogen check and 1% VSO inhibited lesion development. Various solid amendments were incubated for 2 weeks prior to planting in nonsterile, soilless medium and tested against soilborne pathogens (*Rhizoctonia solani* and *Pythium ultimum*) on zinnia. Spent (solvent extracted) and ground neem seed (GNS) at 1 and 2% (w/w) increased stand counts significantly over the R. solani check and 2% GNS resulted in ≤ 60% increase in stand count for *P. ultimum*.

P75

APHIS Plant Pest Permit Procedures

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Researchers and other members of the scientific community often need to transport living pest organisms or related biological control agents. Mishandling may result in a serious insect, disease, or weed outbreak, costing millions of dollars and resulting in the destruction of vital agricultural, plant or water resources.

Three Federal statutes; the Plant Quarantine Act of 1912, the Federal Plant Pest Act of 1957, and the Federal Noxious Weed Act of 1974, provide authority for the Animal and Plant Health Inspection Service (APHIS) to restrict or prohibit unauthorized importation and movement of plant pests and articles that may harbor these organisms.

Authorization to import living pest organisms or ship them interstate may be obtained by an Application and Permit to Move Live Plant Pests and Noxious Weeds (PPQ Form 526). Each application is evaluated individually and the decision to approve or deny the request is made after consultation with State regulatory officials or appropriate specialists.

The applicant must comply with the standard safeguards listed on the reverse side of the permit as well as conditions specified under Section C. Safeguards may vary depending upon pest species, origin, and destination of shipment. Inspection and approval of the premises may be stipulated in the permit.

Information packets will be distributed and questions fielded regarding the regulations, the issuance of permits for plant pests, and the effects of the National Environmental Policy Act (NEPA) on biocontrol researchers.

AUTHOR AND KEY WORD INDEXES

AUTHOR INDEX

KEY WORD INDEX

- NOTES -

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