IOBC / OILB

INTERNATIONAL ORGANIZATION FOR BIOLOGICAL CONTROL
ORGANISATION INTERNATIONALE DE LUTTE BIOLOGIQUE

INTERNATIONAL CONFERENCE :

"TECHNOLOGY TRANSFER IN BIOLOGICAL CONTROL : FROM RESEARCH TO PRACTICE"

CONFERENCE INTERNATIONALE :

"TRANSFERTS DE TECHNOLOGIE EN LUTTE BIOLOGIQUE : DE LA RECHERCHE À LA PRATIQUE"

Montpellier, France, September 9-11, 1996

ABSTRACTS

C.I.L.B.A

COMPLEXE INTERNATIONAL DE LUTTE BIOLOGIQUE AGROPOLIS

IOBC wprs Bulletin / Bulletin OILB srop Vol. 19(8) 1996

Compiled and edited by Christine SILVY,
Head, Common INRA/CSIRO/USDA Documentation Center,
Campus international de Montferrier-Baillarguet,
34982 Montferrier-sur-Lez cedex, France
Fax : (33) 67 59 31 22
E-mail : silvy@ensam.inra.fr

ISBN 92-9067-082-7

IOBC / OILB

INTERNATIONAL ORGANIZATION FOR BIOLOGICAL CONTROL
ORGANISATION INTERNATIONALE DE LUTTE BIOLOGIQUE

INTERNATIONAL CONFERENCE :

"TECHNOLOGY TRANSFER IN BIOLOGICAL CONTROL : FROM RESEARCH TO PRACTICE"

CONFERENCE INTERNATIONALE :

"TRANSFERTS DE TECHNOLOGIE EN LUTTE BIOLOGIQUE : DE LA RECHERCHE A LA PRATIQUE"

Montpellier, France, September 9-11, 1996

ABSTRACTS

C.I.L.B.A

COMPLEXE INTERNATIONAL DE LUTTE BIOLOGIQUE AGROPOLIS

IOBC wprs Bulletin / Bulletin OILB srop Vol. 19(8) 1996

CONFERENCE EXECUTIVE COMMITTEE/ COMITE EXECUTIF DE LA CONFERENCE

CHAIR/ PRESIDENT	E. S. DELFOSSE, President global IOBC/OILB National Biological Control Institute USDA/APHIS 4700 River Road Unit 5 Riverdale MD 20737-1229 (USA) Fax : (1) 301 734 7823 E-mail edelfosse@aphis.usda.gov
MEMBERS	J. COULSON, L. EHLER (USA), F. BIGLER (Switzerland), E. HOEBAUS (Austria), D. J. ROYLE (UK), S. POITOUT (France), P. CASTANERA (Spain), F. FERRER (Venezuela), A. SMETNIK (Russia), H. ZIMMERMANN (S.Africa), R. MUNIAPPAN (Guam)
ORGANIZING COMMITTEE	CHAIRS : J.P. AESCHLIMANN (CSIRO) and S. POITOUT (INRA/OILB)
MEMBERS	N. BOEMARE (INRA), R. BURTON (USDA), G. FABRES (ORSTOM), P. FERRON (INRA/CILBA), P. GUILLET (ORSTOM/CILBA), L. KNUTSON (ARS-USDA), W.M. LONSDALE (CSIRO), J.L. NOTTEGHEM (CIRAD), N. PASTEUR (CNRS), M. PUYGRENIER (AGROPOLIS), L. THALER (Univ.Montp.II), G. THEISSEN (Min. Agric), M. VAISSAYRE (CIRAD)
SCIENTIFIC PROGRAM SUB-COMMITTEE	J.P. AESCHLIMANN (CSIRO), N. BOEMARE (INRA), J. FARGUES (INRA), R. FRUTTOS (CIRAD), M. GOETTEL (INRA), L. LACEY (USDA), W.M. LONSDALE (CSIRO), S. ROUSSOS (ORSTOM)

ACKNOWLEDGEMENTS

The Conference Executive Committee wishes to gratefully acknowledge the generous financial assistance that was received for organizing the Montpellier "Technology Transfer in Biological Control : from Research to Practice", September 9-11, 1996 Conference from :

- AGROPOLIS
- Conseil Régional : Région Languedoc-Roussillon
- District de Montpellier (Montpellier Languedoc-Roussillon Technopole)
- Institut National de la Recherche Agronomique (INRA)
- International Organization of Biological Control (IOBC/OILB)
- International Union of Biological Sciences (IUBS/UISB)
- Ministère des Affaires Etrangères : Direction de la Coopération Scientifique et Technique (DCST)
- Ministère de la Recherche et de la Technologie : Délégation Régionale à la Recherche et à la Technologie (DRRT)
- U.S. Army Research Development & Standardization Group-UK
- U.S. Department of Agriculture, APHIS National Biological Control Institute

Montpellier, August 5, 1996
E.S. DELFOSSE, President global IOBC/OILB
President Conference Executive Committee

TABLE OF CONTENTS

Keynote addresses

Symposia

KEYNOTE ADDRESSES

"Yes, but does it work in the field ?" The challenge of transferring biological control technology

Jeff WAAGE

International Institute of biological Control, CAB International,
Silwood Park, Buckhurst Road, Ascot, SL5 7TA, United Kingdom
Fax : 44 (0) 1344 875007
E-mail : j.waage@cabi.org

Thanks to the efforts and enthusiasm of research scientists, biological control today enjoys considerable popularity. An increasing number of decision makers have high expectations of biological control as the foundation of IPM, as a viable commercial alternative to chemical pesticides, as the self-perpetuating solution to exotic pest problems and as the restorer of parks and endangered habitats threatened by alien invasive species. All of these expectations require that biological control scientists engage a broader community, including farmers, extensionists, environmentalists, regulators, and the public at large, and that they succeed with them to transfer, scale up and implement biological control at a level far beyond its present use.

Increased movement of alien pests due to changing patterns of trade, as well as their emerging importance as threats to biodiversity has increased demand for classical biological control. Meeting this demand effectively will require improved understanding of the risks posed by alien pests and introduced agents, as well as better mechanisms to inform and involve governments and scientists in self-regulation and sharing of benefits. A growing portfolio of commercial biological control agents indicates the potential for inundative methods, but the continued emphasis on developing mass-marketed, pesticide-like agents, increasingly with the help of biotechnology, will eventually limit their application in IPM systems unless a broader approach is taken. Finally, opportunities exist for the transfer of methods for natural enemy conservation on a scale far greater than that of the classical or inundative interventions mentioned above, but realizing these opportunities requires a new approach to research and extension, wherein farmers become active research partners and farmers and scientists explore together the complex and very local dynamics of natural enemy complexes. Thus, for all of these areas of biological control, "making it work in the field" will require taking new directions in scientific research and establishing new partnerships in the transfer and implementation of its results.

Biological control of fungal plant diseases

N. J. FOKKEMA

DLO Research Institute for Plant Protection (IPO-DLO),
P.O. Box 9060, 6700 GW Wageningen, The Netherlands
Fax : 31 8370 10113

Despite of a lot of research on biological control of plant diseases, the number of products available is limited and their market size is marginal. Implementation of biocontrol in agricultural practice is hampered by the costs associated with the registration procedures similar to fungicides. This increases the size of market necessary for an economic production. Major success factors are reliable field performance, cost-effective mass production and the potential market. The latter is not only determined by agricultural aspects such as the number of diseases controlled by one biocontrol product in different crops but also by economic aspects as the availability of competing means of control including fungicides. The future development of low-chemical input sustainable agriculture and organic farming will determine the eventual role of biological control in agriculture.

Research has demonstrated the agricultural potential of biological control. For airborne pathogens as well as for soilborne pathogens similar strategies based on different targets in the life cycle of a pathogen can be distinguished, viz. (1) microbial protection of the host against infection, (2) microbial reduction of pathogen sporulation and (3) microbial interference with pathogen survival. In selecting these targets not only the intensity of the antagonistic interaction but also the duration of such an interaction should be taken into account. Successful examples of control with respect to these targets (1,2,3) include (1) biocontrol of seedling diseases, *Fusarium* wilting disease, foliar and post-harvest diseases (2) biocontrol of powdery mildew and *Botrytis cinerea* (3) biocontrol of sclerotium-forming pathogens. Biocontrol methods aimed at the reduction of dissemination of the pathogen on standing crops or on crop debris and survival of the pathogen resulting in a delay of epidemics may expect wider application than methods aimed at the protection of individual plants because of longer possible interaction times.

Difficulties with cost-effective mass production of the BCA can become a major obstacle for commercialization and this should get sufficient scientific attention at an early stage in the development of a biocontrol method.

3

The Role of parasite and predator production in technology transfer of field crop biological control

C. LEPPLA[1] and E. G. KING[2]

1 : USDA, Animal and Plant Health Inspection Service,
National Biological Control Institute, Riverdale,
Maryland 20737-1229, USA
Fax : 301 734 7823 E-mail : nleppla@aphis.usda.gov
2 : USDA, ARS, Subtropical Agricultural Research Laboratory,
Weslaco, Texas 78596, USA
Fax : 210 565 9584 E-mail : king@pop.tamu.edu

Pest management with limited use of chemical insecticides, less than 50% of current rates, will eventually occur because of the economics of crop production, pest resistance to pesticides, health and environmental concerns, and associated governmental regulation. Annual pesticide revenues worldwide are $24-25 billion and in the U.S. $6-7 billion, 19% from insecticides. Insecticides are used on 93% of U.S. field crop hectarage. Each major pesticide market returns at least $50 million per year and about $2.6 billion is spent on research and development. Conventional pesticides require 8-10 years and $15-30 million to develop and register. Consequently, since 1970, a steady decline has occurred in the number of new pesticides.

The current worldwide market for biological control agents is about $40 million, less than 1/2 of 1% of pesticide sales, and 95% of this is for *Bacillus thuringiensis*. Annually, the public and private sector each invest less than $100,000 million in discovering and developing new biological control agents. Arthropod natural enemies do not require registration and it takes only about three years and $1-2 million to bring a biopesticide to market. Moreover, biological control agents for niche markets may have more of the market share, increased relative profitability, and a longer useful lifetime than chemical pesticides. The biological control industry worldwide is expected to grow at a rate of about 5% per year.

The goals for improving natural enemy production are to reduce costs, increase efficacy and provide additional species for pest management. Nearly 20 examples are given for successfully managing the pests of major field crops by augmentation with natural enemies. Tephritid fruit flies, the European corn borer, and boll weevil are discussed relative to efficacy, cost of production, parasitoid biology, incorporation into IPM, obstacles to their implementation, and recommendations for increasing their use in the future.

Recombinant viral insecticides : delivery of environmentally safe and cost effective products

H. Alan WOOD and Patrick R. HUGHES
Boyce Thompson Institute for Plant Research,
Tower Road, Ithaca, NY 14853, USA
Fax : (607) 254 1242
E-mail : haw5@cornell.edu

Baculovirus insecticides have many attibutes which make them attractive alternatives to synthetic chemical pesticides. However, there have been several economic and agronomic barriers to their widespread use. Two obstacles to commercialization have been competitive cost and speed of action. Biotechnology has contributed several advances toward overcoming these obstacles. New production technologies have recently been developed which improve the cost/benefit ratio of baculovirus pesticides. A versatile *in vivo* production system termed HeRD has been developed to rear many different lepidopterous larvae efficiently at high densities for production of viral insecticides. Production costs for several baculoviruses are equivalent to sprayable *Bt* toxins.
Through recombinant DNA technology, it is also possible to insert foreign pesticidal genes into viral pesticides, resulting in faster time death or, more importantly, time to cessation of feeding. However, the commercial use of recombinant pesticides has raised several potential environmental issues, including healthy safety, ecological interactions, mitigation and genetic stability. Genetic strategies have recently been developed which mitigate most of the potential problems associated with recombinant baculovirus pesticides. Five field tests have been conducted in the U.S. to evaluate these strategies. The laboratory and field results illustrate that the genetic strategies employed ensure environmental safety while also reducing production costs.

5

Experiences in educating rice farmers to understand biological control

Peter A. C. OOI

c/o FAO Intercountry Programme for Integrated Pest Control in Rice
in South and Southeast Asia, P. O. Box 3700 MCPO, 1277 Makati,
Metro Manila, Philippines
Fax : 63 2 8127725
E-mail : IPM-MANILA@CGNET.COM

Traditionally, rice farmers were viewed as ignorant and unable to comprehend biological control as it was too esoteric. This perception was exacerbated by an agro-industry that prefers to maintain a *status quo* to ensure monopoly of information by the priveleged and economically powerful. Hence, between 1950 to 1980, the preferred way of extending technical information to farmers is to package them in "simplistic" messages which ignore ecological realities existing under farm conditions. For example, prophylactic chemical control was the main theme of most extension systems in Asian economies during that period despite evidences to the contrary. Unfortunately, the dogma that chemical control is an integral part of modern agriculture persists to this day in some countries.

In the past two decades, there has been an accumulation of evidence that in Asian rice cultivation and in particular tropical rice, natural enemies are ubiquitous in rice fields and are reponsible for keeping populations of rice herbivores in check. However, this information was largely ignored by the scientific community in favour of a simplistic two-dimensional approach which considered only pests and crops. Such an approach encouraged exaggereted yield losses ; indeed, the rise of the Rice brown planthopper as a serious pest has been linked to the use of insecticides. Outbreaks of this dephacid in tropical rice were related to regular use of chemical poisons which removed effective predators resulting in hopperburn. With the help of biological contol scientists, the FAO Intercountry Programme for Integrated Pest Control in South and Southeast Asia presented the evidence to the President of Indonesia in 1986 resulting in a ban of 56 types of insecticides from rice fields. Promulgating legislative actions was a start and to sustain it an education programme to help farmers understand the importance of biological control was developped. A non-formal education process of learning by experimenting an discovery was formulated. This approach emphasised the need for farmers to understand the rice ecosystem. Hence, the rice field was the classroom for farmers participating in the Farmer Field Schools (FFS). Farmers learned that not all arthropods in the field were pests and that most of the arthropods were "friendly" insects. They discover that these friendly insects eat herbivore. Using insecticide check an exclusion cage experiments, farmers learned of the adverse impact of insecticides and the impact of predators in keeping herbivore populations in check. The curriculum includes crop physiology, agronomy, health risks of insecticides and group dynamics. The principles of IPM emphasised in the Farmer Field Schools are : 1) Grow a healthy crop ; 2) Visit your field weekly to monitor field situation ; 3) Understand and conserve natural enemies ; 4) Farmers become experts in pest management. Adoption of this approach has led to a 60 % dropped in use of insecticides with an estimated 40 % increase in yield. Similar results were achieved in Vietnam and Philippines and this approach is being experimented in other countries within the region. Facilitating farmers to understand biological control through field investigations is a key factor to successful implementation of Integrated Pest Management.

From research to the farmer : the use of extension programs to transfer biological control technology in developed countries

Daniel L. MAHR

Department of Entomology, University of Wisconsin - Madison,
1630 Linden Drive, Madison, WI 53706, USA
Fax : 608 262-3322
E-mail: dmahr@entomology.wisc.edu

Effective use of biological control requires knowledge of the biologies of the pests and natural enemies, and their interactions with their environment and agronomic practices. Manufacturers provide information for products such as microbial pesticides and entomophagous arthropods used in augmentative biological control. However, governmental agencies must often be involved in teaching farmers about process-oriented methods such as classical biological control and conservation of natural enemies. This information is not often available to the farmer. Governmental extension programs are one method for providing practical biological control information, but availability in developed countries varies considerably, with no such programs in some areas.

In the United States, the Extension Service, a branch of the U.S. Department of Agriculture, provides partial funding and coordination for pest management educational programs conducted at the national, regional, state and local levels. Since the1950s, much of the pest management information delivered by extension was related to pesticides. The methods of pesticide use were easily described and the results were dramatic and usually predictable. Although questions arose about the negative aspects of pesticides, their ease of use and reliability still resulted in dominant interest by farmers and therefore recommendations by extension. However, more recent questioning of the use of pesticides by society, along with a generation of more highly-educated farmers, has resulted in increased demand for information on alternatives, including biological controls.

In a twelve-state region of the North Central United States, university extension and research scientists have developed an intensive program to educate county extension personnel, farmers, and private consultants about the use of biological controls in pest management. The details of this model program will be discussed.

The address will be concluded with a discussion of the educational constraints that must be overcome to successfully increase the adoption of biological control.

Evaluating the efficacy of biological control of three exotic Homopteran pests in tropical Africa

Peter NEUENSCHWANDER

International Institute of tropical Agriculture,
Plant Health Management Division, BP, 08 0932, Cotonou, Bénin
Fax : (229) 35 05 56
E-mail : P. Neuenschwander@cgnet.com

To describe criteria and techniques for evaluating biological control, projects concerning Cassava mealybug (CM) (*Phenacoccus manihoti*), Mango mealybug (MM) (*Rastrococcus invadens*), and Spiralling whitefly (SWF) (*Aleurodicus dispersus*), covering large parts of Africa, are described. Against each of the three pests, two hymenopterous parasitoids from the original area of distribution were introduced (serendipitously in the case of the SWF), which brought each of the pest populations under control. Indigenous and exotic coccinellids played a much smaller role,. On MM and SWF, two closely related parasitoids now coexist in Africa. For the two Encyrtid MM parasitoids, partially overlapping niches could be defined. Among the two *A. zoanagyrus* (*Epidinocarsis*) spp. attacking the CM, *A. lopezi*'s niche proved to be superior over the one of *A. diversicornis*. In the field, the latter species was displaced competetively, thereby validating results of a simulation model. SWF parasitoids were not yet studied in detail. In all three projects, control was achieved in areas where the exotic parasitoid(s) had been present for more than two to four years. The impact was documented by 1-exclusion experiments, 2- long-term population dynamics studies based on field samples, 3- results from detailed laboratory and field experiments that were used for the development of simulation models, and - most importantly - 4- quantitative results on multiple variables from large-scale surveys. In many countries, the main introduced parasitoids proved to be the most important factor (among all physical, biotic, and human factors quantified) in contibuting to the decline in the pest populations and recovery of plant growth and yields. Non-target species were only affected in so far as the availability of the homopteran pests as food sources of polyphagous predators was declining. The impact was scale-neutral, benefitting subsistence farmers, commercial farmers, and urban gardeners alike. The population reduction, attributed to the specific parasitoid(s), remained stable and was in the order of XXX times over outbreak levels. In economic terms, excuding ecological and health benefits, both the CM and MM projects returned to the African farmers a multiple of the research an implementation costs (200 x for CM), which had been paid for by various development agencies. The CM biological control project has now spawned a long series of biological control implementations out of West Africa which are all approached in the same holistic manner. The success of these activities is attributed to 1- collaboration among international institutions, universities, etc., 2- a strong committment to support (scientifically, technically, materially) the various African countries in their collaboration with IITA and among each other, 3- a strong training effort, and 4- continued support by donnor agencies. On the political front, an important role was played by the Inter-African Phytosanitary council, under whose auspices all these projects were placed. The solution of many other plant health problems, however, requires an integrated approach. Thus IITA became the convenor of the "IPM Sytem-wide Program". It produced a definition of IPM, in which biological control is the foundation for all other interventions, and sponsors world-wide efforts to coordinate research on combatting particularly reticent pests, diseases and weeds.

Public policy and partners in biological control in France

G. RIBA[1], S. POITOUT[2] and C. SILVY[3]

1 : INRA, Station de Recherches de Lutte biologique, La Minière, 78285 Guyancourt cedex, France
Fax : (33) 30 43 80 97
2 : INRA, Station de Zoologie, Domaine St Paul, Site Agroparc, 84914 Avignon cedex 9, France
3 : INRA, Unité de Recherches en Lutte biologique, Campus international de Baillarguet-Montferrier, 34982 Montferrier-sur-Lez cedex, France

Most public institutions and phytosanitary industries have a positive opinion of biological control, they are concerned by the issue but weakly engaged in its resolution. In spite of this, that concept of plant protection is poorly developed in agriculture, successes of biological control are insufficiently known, competition with chemical means is severe since pesticides are reliable, efficient, specific and more environmentally safe.
In this context, and to impose development of biological control, three actions are proposed :
- a higher interdisciplinarity in research : the agronoms and economists do not sufficiently consider biological control ; the biologists have to take better advantages of concepts of evolutionary biology ; more experiments in real context and large scale must be done ;
- more important and much more integrated objectives : the only success of biological means to control a pest is not a sufficient objective. We absolutely need to consider all aspects of integrated management : pesticides resistance, cultural means, plant breeding, all alternative bio-technical means ;
- more partners : during too many years, only researchers of public institutions had activity on biological control. Today, many partners, including public (Plant Protection Service, Technical Institutes) and private (large and small companies) organisms are engaged in alternative plant protection. To facilitate cooperation between them, a specific committee has been organized in France in order to coordinate experimentations, evaluate programs and promote recommendations for registration and quality control of biopesticides. With such better relationships between all partners, specific arguments and lobbying should be found to convince the great distribution of agronomic products to promote "Integrated Protected Plant Products" and to require specific public supports for promotion of products and research.

The role of public policy in biological control : some global trends

P.W. WELLINGS

CSIRO, Division of Entomology, GPO Box 1700,
Canberra ACT 2601, Australia

The past decade has been a rapid development in the view that contemporary agricultural practices should meet the needs of the present without compromising the ability of future generations. This philosophy has broadened the priorities for research and development in primary production systems. Now there is reduced emphasis on efficiency and productivity, and an increased concern for ecological sustainability. This shift in balance has had a threefold effect on policies for pest management. We now recognise the need to :

1) adopt objectives to promote research and development into sustainability;
2) concentrate on approaches that minimise chemical use in agriculture; and
3) devise strategies for pest management that are integrated as part of regional or whole farm systems.

This has had a consequent impact on policies relating to biological control technologies. Many countries have closely examined their regulatory systems and have attempted to streamline processes for evaluating the risks associated with the introduction of classical biological control agents. Associated policies have been developed to enhance the uptake of biopesticides. The approaches here are divergent because of the range of views on the risks associated with genetically engineered and non-engineered products. In addition, policies on intellectual property rights can have a major impact on routes to effective commercialisation and rates of technology uptake.

In this review, I highlight some of the current trends in public policy and the likely effect these may have in the adoption, implementation and ongoing development of biological control technologies.

SYMPOSIUM #1

BIOLOGICAL CONTROL OF SOIL-BORNE PESTS AND DISEASES

Organizer :

Richard R. SIKORA
Institut für Pflanzenkrankheiten
der Rhein Friedrich-Wilhelms-Universität
Phytomedizin in Bodenökosystemen
Nussallee 9, 53115 Bonn, Germany
Fax : 0228-732432
E-mail : ulp40@ibm.rhrz.uni-bonn.de

12

The integration of nematophagous fungi in management strategies for root-knot nematodes

Brian R. KERRY

Entomology & Nematology Department, IACR-Rothamsted, Harpenden, Herts., AL5 2JQ, United Kingdom
Fax : 44 1582 760981
E-mail : Brian.Kerry@bbsrc.ac.uk

Despite their efficacy in nematode-suppressive soils, nematophagous fungi which attack the sedentary females of cyst and root-knot nematodes and their eggs have proved difficult to exploit in practice. To be effective, these fungi must colonise the rhizosphere of nematode-infected plants and survive throughout the growing season. *Verticillium chlamydosporium* has been selected as a potential biological control agent for root-knot nematodes because of its virulence and ability to colonise the rhizosphere of a range of crop plants. The fungus produces thick-walled chlamydospores which are applied in aqueous suspensions to nematode-infested soils. A single application at planting time may be adequate to establish *V. chlamydosporium* in the rhizosphere. Chlamydospores are not produced in large numbers in liquid media and, at present, solid media are used for their culture and only laboratory-based methods of production have been developed. Plant species differ in their susceptibility to nematode attack and in their ability to support the growth of the fungus in their rhizospheres. *Verticillium chlamydosporium* alone does not control large infestations of root-knot nematodes on susceptible crops. A strategy which incorporates the fungus within crop rotations which utilise resistant or poor hosts for the nematode has been developed for vegetable crops and may lead to more sustainable methods of control for these important pests.

Mutualistic fungal endophytes and plant health promotion - microorganisms targeted for transplant and tissue culture production systems

Richard S. SIKORA

University of Bonn, Soil Ecosystem Pathology, Nussallee 9,
53115 Bonn, Germany
Fax : 0228-732432
E-mail : ulp40@ibm.rhrz.uni-bonn.de

The presence of non-pathogenic fungi in the phyllosphere and rhizosphere tissue of all growing plants is the rule not the exception in nature. More important, a significant number of these saprophytic endophytes have been shown to have plant health promoting activity. Biological control of insects in the phyllosphere occurs in the presence of these mutualistic fungi in leaf tissue. The occurrence of these endophytes in the shoot tissue also simultaneously reduces nematode damage to the root system. More recent evidence shows that mutualistic fungal endophytes commonly found in root tissue are important biological agents for control of plant parasitic nematodes, root pathogens and soil insects. Pre-inoculation of planting material with mutualistic endophytes or the use of application techniques that favor endophyte colonization of the plant before pest and disease infection, are required for acceptable levels of biological control. Commercial use is, therefore, most promising for crops grown for transplanting into the field, plants originating from tissue culture systems or for crops grown in heat pasteurized for fumigated soils e.g. in greenhouses. Research is targeted at developing selection protocols for detecting isolates with broad spectrum activity, high levels of biological control and strong root colonization characteristics. The costs of production are low due to : the limited amount of inoculum need, the small amount of substrate targeted for pre-treatment and because methods of production and formulation of saprophytic fungi already exist. Future acceptance of endophytes for biological control by industry and the grower will depend on consistency of control, economic importance of the soil pests and diseases that can be controlled simultaneously and duration of plant protection activity.

Development of a commercial seed treatment bacterium : the story of *Bacillus subtilis* GB03

Paul A. BACKMAN

Biological Control Institute and Department of Plant Pathology,
Auburn University, Auburn AL 36849, USA
Fax : 334 844 1947
E-mail : pbackman@ag.auburn.edu

Bacillus subtilis strain GB03 was used to inoculate roots of more than 4 million hectares of crops in 1995.
This paper will trace the development of GB03, from the discovery of its parent isolate, A-13, through its present state of commercial development. First isolated from a sclerotium from Australian soil by Priscilla Broadbent in the late 1960's, it was later classified as a spore-forming isolate of *Bacillus subtilis* (A-13) and reported to control several seed and seedling diseases. Typical application methods were as drenches of mixtures of vegetative cells and spores. Results in controlled environments were good, but were much more erratic when the technology was moved to the field. By the mid 1970's after several publications and continued variability of field results, the further development of strain A-13 was abandoned in Australia. Evaluation of A-13 began in the United States in 1980, when a preparation of formulated endospores produced by Abbott Laboratories was applied to peanut seed (*Arachis hypogaea* at 7.0 x 10^7 / seed) for control of seed and seedling diseases. Although there was a significant increase in emergence, the most unexpected results were earlier emergence, and plant vigor improvements that were apparent through midseason. When these trials were repeated it was found that colonization of peanut was always successful, but that vigor differences and emergence improvements were variable. By 1982, a key observation was made : it was found that mixing *B. subtilis* with traditional seed treatment chemicals assured good emergence without compromising colonization. Yield improvements were recorded in most trials, often without signs of either emergence or vigor differences. Evaluation of treated peanut root systems indicated that the root mass was increased, as were the number of *Bradyrhizobium* nodules and levels of several plant nutrients. Damage by *Rhizoctonia solani* AG-4 was frequently suppressed, but was not required for yield enhancement. Variability was much reduced if treatments were limited to seed planted to nonrotated field (high pathogen populations), and/or to seed planted into cool (<22C) soils. A-13 was registered in 1985 for use on peanut. Acceptance was slow because the cost per hectare was high, primarily because of a high seeding rate. It was about this time that Gustafson Inc., a

division of Uniroyal Agricultural Chemicals, took charge of the development of this bacterium. Cotton research with A-13 began in the late 1980's with early results again showing erratic responses. Here, again, treatments were always made in combination with seed treatment pesticides and after the first year a second key improvement was made, the A-13 isolate was adapted to cotton by multiple host-passages (the strain name was then changed to GB03). When data were developed that indicated that root mass was increased, *Rhizoctonia* AG-4 seed and seedling diseases were reduced, plus that there was suppression of *Fusarium* wilt caused by *Fusarium oxysporum f.sp. vasinfectum*, commercial seed vendors were quick to incorporate the GB03 strain into their blend of seed treatment chemicals. Since seeding rates are 1/7 of the levels of peanut, costs were reduced by a similar factor. A third factor facilitating sales of this biological control system was that the companies were not promoting the increase in emergence, nor the vigor differences as the expected product of treatment. Rather, they promoted the product for its improvements in season-long root health and for increases in yield. For the 1996 season, more than 80% of the cotton planted in the USA will be treated with GB03. More than 50% of the cotton will be treated in combination with a second *Bacillus subtilis* (strain GB07). Data points to the second strain reducing *Pythium* damage while also possessing *Rhizoctonia* and *Fusarium* activity. Data from soybean indicates that use of the combination results in more uniform levels of colonization among multiple soybean cultivars. Strain GB03 of *B. subtilis* is a promiscuous colonizer of both monocots and dicots. Although it has detailed registrations on 7 crops in the USA, it can be used on all crops. Research data on soybeans, corn and small grains shows a great potential for continued commercial development. In addition it promotes vegetable growth whether incorporated into the growth media of transplants, drenched in with transplant water, or applied by the seed delivery system. The success of GB03 and that of GB07 as well, comes from the fact that they produce endospores, colonize most plants, are environmentally insensitive, and are extremely tolerant of most agricultural chemicals. It is sold worldwide by the various subdivisions of Uniroyal Agricultural Chemicals.

Biological control of plant parasitic nematodes with rhizobacteria : strategies and technical requirements

Sabine HOFFMANN-HERGARTEN

University of Bonn, Institute for Plant disease,
Soil Ecosystem Pathology, Nussallee 9, 53115 Bonn, Germany
Fax : 0228 1732432

The potential of plant health-promoting rhizobacteria for control of plant parasitic nematodes has been demonstrated repeatedly. The most intensive greenhouse and field studies have been carried out with cyst forming nematodes (*Globodera* spp., *Heterodera* spp.) and root knot nematodes (*Meloidogyne* spp.), both sedentary endoparasites. However, when comparing a bacterial based biocontrol system with a classical chemical control procedure, the direct influence of the bacteria on the plant as well as the antagonistic activity towards the target nematode must be considered simultaneously. In field experiments, it was shown that in spite of subsequent nematode infection the yield of bacteria treated plants reached levels comparable to nematicide treatments. Rhizobacteria are known to have broad activity on different components of the cropping system, *i.e.* the nematode, other deleterious organisms and on plant growth. These multiple interrelationships often compensate for a less than adequate control of the target pest and enhance the attractivity of rhizobacteria as a biocontrol system. Technological transfer of rhizobacteria based biocontrol systems from the experimental scale to practical field conditions, besides leading to good control must also lead to a high consistency in effectivity. The presence of the bacteria in sufficient numbers in the rhizosphere in an early stage of plant growth is undoubtedly important for biocontrol activity. Protection of the seedling stage from nematode attack in many cases is adequate for good plant development and yield. Formulation and application systems have to reflect the requirements of the bacteria and ensure the development of a high initial population. The importance of strategies to enhance population stability in the rhizosphere with time after treatment is based on the mechanism-of-action involved. This may require different levels of root colonization at different times. Long term

biocontrol may be attained even by a low bacterial population due to the induction of resistance mechanisms in host plants.

The Rhabditid nematode *Phasmarhabditis hermaphrodita* for biological control of slugs : from research to biocontrol product

D.M. GLEN[1], M.J. WILSON[1,3], L. HUGHES[1], P. CARGEEG[2] and A. HAJJAR[2]

1 : IACR - Long Ashton Research Station, Department of Agricultural Sciences, University of Bristol, Long Ashton, Bristol BS18 9AF, United Kingdom
Fax : 01275 394299 E-mail : David.Glen@bbsrc.ac.uk
2 : MicroBio Ltd, Church Street, Royston, Thriplow, Herts SG8 7RE, United Kingdom
3 : Current address: Department of Entomology, Rutgers University, New Brunswick, New Jersey 08 903 0231, USA

This paper summarises research leading to the launch of a biocontrol product in 1994, based on the slug parasitic-nematode *Phasmarhabditis hermaphrodita*, for use against slug pests (Mollusca : Gastropoda) in the home-garden market in the UK. Ongoing investigations are also described, which aim to exploit the potential of this nematode for control of slug damage in a wide range of agricultural and horticultural crops.
P. hermaphrodita is a bacterial-feeding nematode which is capable of killing a wide range of pest slug and snail species, but is harmless to other soil invertebrates. Infective larvae enter the shell sac of the host slug and cause the slug to stop feeding, thus preventing slug damage. When the slug dies, nematodes spread and multiply over the host cadaver. The nematode is cultured in liquid fermenters on nutrient-rich media containing a selected bacterium (only certain nematode-bacterium combinations are capable of killing slugs). Infective larvae are harvested and formulated in friable clay, and can be stored under refrigeration for several months. When required for use, nematodes are suspended in water and applied to soil as a drench using a watering can, or as a spray using standard spraying equipment with in-line filters removed. Field trials have shown that the nematode, applied to the surface of moist soil, protects a wide range of crops from slug damage. Timing of application is not critical. Efficacy in drier soil can be improved by shallow incorporation of nematodes. Laboratory studies show that slugs avoid soil treated with nematodes, suggesting that it may be possible to restrict applications to soil around individual plants or along crop rows. Field studies have shown no effects on non-target molluscs living in field margins adjacent to arable soil treated with the nematode. Directions for future research and commercialisation are discussed.

Entomopathogenic nematode technology at a turning point

R.U. EHLERS

Institute for Phytopathology, Christian-Albrechts-University Kiel,
Department for Biotechnology and biological Control,
Klausdorfer Str. 28-36, 24223 Raisdorf, Germany
Fax : 49 4307 7499
E-mail : ehlers@biotec.uni-kiel.de

Entomopathogenic nematodes (*Steinernema* and *Heterorhabditis*) can substitute for chemical insect control in many areas : For example, for control of sciarids in ornamentals and mushrooms, weevils in ornamentals, soft fruit and tree nurseries, grubs in public greens and control of fleas and ticks of domestic animals. State-of-the-art and dimension of production, storage and formulation technology are summarized. An overview of safety and use in integrated pest management is given. International scientific co-operation and results of technology transfer are presented. Costs for research and development efforts and the scale-up of production technologies require financial efforts which are related to the current and future market potentials. In the future, large scale out-door application will make possible the full exploitation of economies of scale in order to reduce product prices significantly. Besides public funding, commercialization in niche markets provides the financial basis for further technical development. In most countries nematodes are exempted from registration. Any kind of activities to regulate nematodes would seriously hinder further technical progress as markets are too small to justify costs related with registration. In the past, minor use provided chemical control measures for small markets like ornamentals, vegetable, fruit, forest and public greens. Nowadays, regulatory officials should limit minor use in order to support the further development of nematode based products which can provide growers with environmentally safe and economically feasible control measures.

Entomopathogenic nematodes : possibilities and constraints in their use as biological control agents

Peter H. SMITS

DLO-Research Institute for Plant Protection (IPO-DLO),
POB 9060, NL-6700 GW Wageningen, The Netherlands
Fax : 31 317410113
E-mail : P.H.Smits@ipo.dlo.nl

Entomopathogenic nematodes are commercially produced and used in practice by growers. The main use is against vine weevil and others curculionids. Other markets are the control of various turfgrass insects, sciarids and ticks. The use of entomopathogenic nematodes is growing but there are constraints that limit expansion of the market. Product costs per hectare are much higher than that of chemical insecticides, although nematodes are getting cheaper. Very high numbers of nematodes are applied, often 1 million/m^2, and there must be room for considerable reduction in these numbers, with better strains and improved application systems. Another constraint is the effectiveness of the strains that are now produced. At present only a single strain of 4-5 different species are available as commercial products. These strains have been selected on criteria such as activity against certain important target insects, but other criteria such suitability to be formulated and suitability to be mass produced in fermenters also played a major role in the choice. Entomopathogenic nematodes are often considered generalists that will kill a whole range of insects. This may be true under Petri dish conditions, but it is certainly not true for all entomopathogenic nematodes and probably completely wrong under field conditions. This means we must not expect a specific nematode product to be effective against a whole range of insect pests in different crops in different climates. More diversification in specialised nematode products selected for efficacy against specific insects under specific conditions would improve field performance and the practical use of entomopathogenic nematodes, but it may in many cases not be economically feasible from a commercial point of view.

Some promising results from field applications of *Pasteuria penetrans* for control of root-knot nematodes

S.R. GOWEN

Department of Agriculture, University of Reading , Earley Gate,
Reading RG6 6AT, United Kingdom
E-mail : aasgowen@reading.ac.uk

As part of an European Community funded project, long term rotations in root-knot nematode infested plots initially treated with *Pasteuria penetrans* (*Pp*) have been conducted in several tropical countries. The objective of these rotations has been to see if under continuous cultivation the levels of Pp will increase and thus suppressiveness will develop. In some locations the incidence of Pp spore attachment on free-living root-knot nematode juveniles in soil samples has been monitored over six crop cycles. Also, the effects of including crops less susceptible to root-knot nematodes in these rotations has been evaluated to observe the conflicting influences of poor hosts on nematode populations and the desired need to promote the increase of the inoculum potential of *Pp*. In all cases, spores of Pp have been mass produced by the standard *in vivo* system, where possible, with *Pp* from the study areas. This has been deployed as a powder produced from the roots of the host plant on which the host nematode developed. *Pp* occurred naturally at the field sites in Ecuador and Trinidad and in consequence, levels of the natural population increased in plots which had not been augmented with the *in vivo*-produced inoculum. In Ecuador, by the sixth cycle of continuous cultivation of root-knot susceptible tomato and bean varieties, the intensity of root galling was significantly less in plots augmented by *Pp* ; numbers of root-knot nematode juveniles decreased, incidence of *Pp* infection increased and the yields in these plots were greater.

Trichoderma harzianum preparation as a biocontrol agent against the root-knot nematode, *Meloidogyne javanica*

Y. SPIEGEL[1], I CHET[2], M. MOR[1], O. KLEIFELD[3], E. SHARON[1] and M. BAR-EYAL[1]

1 : Department of Nematology, Agricultural Research Organization, The Volcani Center, Bet Dagan, Israel 50-250
Fax : 972 3 9604180
E-mail : vpspigl@volcani.gov.il
2 : Department of Plant Pathology and Microbiology, Hebrew University, Faculty of Agriculture, Rehovot, Israel 76100
3. Mycontrol company, Alon Hagalil, Israel

A biocontrol agent against soil-borne diseases based on *Trichoderma harzianum* isolate T-203 has been developed to serve as a control agent against the root-knot nematode *Meloidogyne javanica*. Several preparations based on *T. harzianum*, in combinations with various amendments, other microorganisms and various application techniques are tested and their nematicidal activity is evaluated.
Tests performed in greenhouses and screenhouses indicate improved growth and yield of nematode-infected plants and decrease of the root-galling index and the number of eggs per g root, when nematode-infested soils are pre-treated with the T-203. Speed-seedlings already carried the fungus preparation reveal less activity against nematodes.

SYMPOSIUM #2

LOCUST AND GRASSHOPPER CONTROL

Organizers :

Jacques FARGUES
INRA, Campus international de Montferrier-Baillarguet
34982 Montferrier-sur-Lez cedex, France
Fax : (33) 67 59 31 22
E-mail : fargues@ensam.inra.fr

and

Mark GOETTEL
Agriculture and Agri-Food Canada Research Centre
P.O. Box 3000, Lethbridge, Alberta, T1J 4B1, Canada
Fax : (403) 382 3156
E-mail : Goettel@em.agr.ca

Strategies for Locust and Grasshopper biological control

Roy BATEMAN and Chris PRIOR

International Institute of biological Control, Silwood Park, Ascot, Berkshire, SL5 7TA, United Kingdom
Email: r.bateman@cabi.org

Chemical insecticides applied at ultra-low volume (ULV) rates remain the most reliable and fast acting method of eliminating locust swarms. Concerns about the extensive use of chemicals for locust and grasshopper control have been expressed by environmental organisations (notably in N. America, Australia and Southern Africa) and the donor community (that contributes to control of locusts and grasshoppers, especially desert locusts during outbreaks). Development of an integrated pest management (IPM) approach has therefore been advocated which will include the use of entomopathogens for "strategic control" measures.

Some use of chemicals will continue for the foreseeable future and may always be required for rapid intervention for crop protection. It is therefore advantageous that application techniques for microbial agents should be made as compatible as possible with existing chemical practice. Mycoinsecticides based on *Metarhizium* and *Beauveria* spp. applied as low volume and ULV sprays in oil-based formulations have proved to be the most promising biological agents to date. Field mortality is rarely observed in less than one week, but reductions in feeding and mobility may enhance efficacy.

Slow speed of kill is a key problem that must either be overcome or managed in some way. Some possible strategies are discussed in the light of experience with *Locustana pardalina, Oedaleus senegalensis* and certain grasshopper species. Mobility of the target is a major determinant of strategy: highly mobile hopper bands and swarms present logistical problems which must be overcome. Possible solutions include the use of spray markers, or application techniques that effectively treat infested land at a very high work rate.

Development and field evaluation of *Metarhizium flavoviride* blastospores

D. STEPHAN and G. ZIMMERMANN

Federal Biological Research Centre for Agriculture
and Forestry, Institute for Biological Control,
Heinrichstr. 243, D-64287 Darmstadt, Germany
Fax : 06151 40790
Email: ibp@hrzpub.th-darmstadt.de

Metarhizium flavoviride is a well documented pathogen of locusts and a promising pathogen for biocontrol of African locusts. However, the commercial utilization of this fungus as a biopesticide necessitates fast and efficient mass-production, storage of preparations for long periods and effective formulations which allow a controlled droplet application at low volume rates under desert conditions. Therefore, a submerged culture technique was used in order to produce high concentrations of blastospores of *M. flavoviride* in fermenter with a capacity of up to 350 liter. Using a liquid medium containing 8% composted chicken droppings (Naturpur®) and 4 % sugar-beet syrup, the maximum yield of spores was up to 1×10^9 spores per ml after two days fermentation. Studies on the growth of *M. flavoviride* are discussed. In order to develop a stable biopreparation of blastospores, a special spray-drying technique was developed which did not reduce the viability and pathogenicity. For spray-drying, different carriers and inlet temperatures were compared. Both the carrier and the inlet temperature influenced the viability of spray-dried blastospores. Long-term storage of spray-dried blastospores at 5°C for about two years resulted in a germination rate of 72 %. After one year storage at 20°C and 30°C, the germination rate was 68 % and 38 %, respectively. In Mauretania, different formulations of spray-dried blastospores were tested in semi-field tests against larvae and adults of *Schistocerca gregaria* using an ultra low volume (ULV) application technique. Spray-dried blastospores were highly infective in a water based formulation (20% molasses, 80 % water) against larvae and adults of *S. gregaria*. Additionally, the evaporation of water based formulations was investigated in Mauritania. Therefore, the droplet size was measured with an interactive picture analyzing system. Results are presented. The investigations were carried out within the framework of the GTZ-Project "Integrated Biological Control of Grasshoppers and Locusts".

Development and field evaluation of *Metarhizium flavoviride* conidia

C.J. LOMER
with LUBILOSA project staff and collaborators

IITA-Bénin, B.P. 08-0932, Cotonou, Bénin
Fax : 229 350556
E-mail: c.lomer@cgnet.com

The LUBILOSA (LUtte BIologique contre les LOcustes et SAuteriaux) project produces *Metarhizium flavoviride* in a di-phasic system with rice substrate at IITA Cotonou and AGRHYMET Niamey. Spores are extracted dry and are stable in storage. Spores are readily suspended in different oils to make ULV formulations. We usually apply 100g/ha in 2 litres of oil, approximately $5x10^{12}$ spores/ha. Field population reductions following spore application have been observed in four systems. For *Zonocerus variegatus* in southern Benin, counts of fifth instar or young adult insects were observed to decline by 90% ten days after application. Application rates from 200 to 2 g/ha were tested; even the lowest rate gave satisfactory control. *Hieroglyphus daganensis* at Malanville, north Benin was also controlled by ULV application of *Metarhizium* spores; however, the denser vegetation reduced the direct impact of the spray. This was compensated to some extent by pick-up of spores from the vegetation, resulting in 70% control 14 days after application.

In Sahelian grassland in Niger, three replicate 50 ha plots were treated in 1995 at Maine Soroa, giving 80% control after 12 days in populations of *Oedaleus senegalensis*. Application to desert locust hopper bands in Mauritania resulted in the destruction of treated bands; although population counting of mobile hopper bands was difficult, a significant reduction was observed, and cadaver counts also gave significant results. National programme trials in 1995 were carried out in Chad, Gambia, Senegal, Burkina Faso and Niger, and participatory trials with NGOs in Niger and Mali. Many of the significant biological and socio-economic constraints to the implementation of mycopesticides were identified during these trials.

Development and field evaluation of mycoinsecticides for Grasshopper and Locust control

C. BRADLEY

Mycotech Corporation, P.O. Box 4109, Butte, MT 59701, USA
Fax : 406 782 9912

With deuteromycete entomopathogens, the transition from research to practice requires integration of strain selection and field efficacy evaluations with mass production technology, product development and regulatory compliance. Mycotech selects *Beauveria* and *Metarhizium* isolates for their commercial utility. In addition to efficacy, strain selection includes evaluation of conidia production, stability and toxicology. Grasshopper, locust control involves large geographically dispersed areas. Mass production technology is key to technology transfer. Mycotech constructed a solid substrate production facility with an annual capacity of 500,000 units of formulated *B. bassiana* where one unit contains 1×10^{13} viable conidia. At this scale, mycoinsecticides are price competitive with most chemical insecticides. Mycotech links formulation development and field evaluations. Oil, wettable powder and bait formulation have been field tested using different application equipment and control strategies. Selected results from collaborative trials in Africa and North America provide examples of mycoinsecticide use in grasshopper, locust IPM. Mycoinsecticides are subject to both pesticide and phytosanitary regulations relating to efficacy, safety and potential environmental impacts. Compliance is crucial to technology transfer. Mycotech has obtained registrations for *B. bassiana*-based formulations in the U.S., and approval from all countries where trials have been conducted. The data package submitted to USEPA includes come 40 separate studies covering "product chemistry", toxicology and environmental effects. A number of additional *B. Bassiana*, *M. flavoviride* and *M. anisopliae* isolates have been evaluated in toxicology and non target insect studies prior to field trials with collaborators in Madagascar, Cape Verde and Mali.

Implementing biological control of Locusts : the Madagascar model

Will SWEARINGEN

Department of Entomology, Montana State University, Bozeman, MT, 59717-0302, USA
Fax : (406) 994 6927
Email: ueyws@msu.oscs.montana.edu

Madagascar could be the first country in Africa to implement control of locusts using a natural pathogen. This would be a major departure from the use of broad-spectrum chemicals, which in the past have damaged Madagascar's environment and economy.

Progress toward development of a biocontrol agent has been rapid since 1993, when Montana State University (MSU), with Mycotech Corporation and Malagasy collaborators, isolated nearly 40 distinct pathogens for potential development. Three years later, Madagascar's leading fungal pathogen, *M.f.* SP9, has undergone rigorous laboratory and field efficacy tests as well as toxicology, nontarget-insect, biodiversity-impact, and commercial feasibility evaluations. Entomopathogenic fungi can be formulated in a variety of media, applied with standard equipment, and mass-produced locally.

An efficient selection process was devised to give equivalent emphasis to the technical and commercial characteristics of the pathogens selected for development. This approach is rooted in the understanding that biocontrol agents aim to reduce the use of chemical pesticides and must therefore be competitive both in the field and in the market. Accordingly, strains were selected for conidial yields and storage stability, as well as for efficacy and safety.

Factors which further favor development and implementation of biocontrol technology in Madagascar are : (1) cooperation from the government and (2) environmental factors, notably the geography of Madagascar and the ecology and population dynamics of the principal locust pest, *Locusta migratoria capito*.

Finally, economic studies have bolstered the project's credibility both locally and in the eyes of donors. A commercial feasibility study demonstrated the viability of local production under several demand scenarios, and offers a tool for promoting local investment. Current and future activities aim at increasing operational relevance, developing local initiative, and promoting involvement of the private sector in biocontrol of locusts.

Commercial feasibility of mycopesticides for Locust control

Dan SWANSON

Economic Consultant, Bozeman, MT 59715, USA
Fax : (406) 586 5528
Email: Swanson@gomontana.com

Two recent studies have examined the commercial feasibility of producing and selling entomopathogenic fungi for grasshopper and locust control in Africa. The results of these studies are presented and compared.

The first study examines the commercial feasibility of mycopesticide production in Madagascar, a closed phytosanitary zone insulated from competition from foreign producers. It concludes that local production is feasible using a labor-intensive technology, provided the local market could generate demand for treating 20,000 ha per year at the price of $11 per ha. If, however, the market (including exports) was as big as 80,000 ha per year, prices could drop as low as $9 per ha using a capital-intensive technology.

The second study examines the economic viability of mycopesticides for acridid control in Africa as a whole. This study finds European and African production to be feasible in a few countries with favorable economic signals and trans-African air freight services. Production cost estimates range from $8 to $14 per ha, depending on the level of output, adding an additional dollar per ha for shipping. Air freight shipping would be essential for the viability of the spores, and multiple air-freight routes are necessary for gaining scale economies.

Capital-intensive technologies offer cost advantages in regional production, though labor-intensive technologies could potentially compete in niche or closed markets, or in countries with voids created by gaps in air-freight service. Moreover, labor-intensive technology may precede capital-intensive technology to the African market owing to lower start-up costs and uncertainty about demand.

Both studies assume bilateral donors to be the primary consumers (buyers) of mycopesticides as they have been with chemicals. Donors could channel purchases through the FAO on a set-aside basis at first, as a percentage of historical purchases. Demand is expected to expand owing to local, national, and international goals of reducing environmental damage and avoiding resistance and residue problems. Technical progress is assumed. Economic benefits from biocontrol technology transfer would accrue to Africans in the development and use of the products, and in countries with production and formulation facilities.

Environmental constraints of mycopesticides ; a challenge

Mark S. GOETTEL[1] and Jacques FARGUES[2]

1 : Agriculture and Agri-Food Canada Research Centre,
P.O. Box 3000, Lethbridge Alberta, Canada T1J 4B1
Fax : (403) 382 3156
E-mail: Goettel@em.agr.ca
2 : INRA, Centre de Montpellier, Unité de Recherches en Lutte Biologique,
Campus International de Montferrier-Baillarguet,
34982 Montferrier-sur-Lez Cedex, France
Fax : (33) 67 59 31 22
Email: Fargues@ensam.inra.fr

The entomopathogenic fungi, *Beauveria bassiana* and *Metarhizium flavoviride* are being developed as inundative control agents of grasshoppers and locusts. The success of any microbial control agent will depend on our ability to overcome environmental constraints or to predict windows of opportunity. Microclimatic constraints include solar radiation, temperature and humidity and each of these factors, acting in combination, may have different effects at each step of disease initiation and development. Conidia exposed to UV-B radiation are relatively short-lived. Once conidia have been deposited onto the hosts, humidity does not appear to affect disease initiation. Results of laboratory studies indicate that grasshoppers infected either with *B. bassiana* or *M. flavoviride* preferentially seek temperatures between 40 to 42°C. Such behavior presents an important thermal constraint to the progress of infection. However, there exists a wide range of thermal and solar radiation responses among strains and species. Among the 3 species tested, strains of *M. flavoviride* exhibited the greatest tolerances to both high temperature and solar radiation while strains of *B. bassiana* exhibited greatest tolerances to lower temperatures. In laboratory experiments simulating grasshopper thermoregulation during daylight periods, application of both *M. flavoviride* and *B. bassiana* simultaneously resulted in a final prevalence of disease that was greater than *M. flavoviride* alone in the hot temperature environment, and equal to *B. bassiana* alone in the cool temperature environment. Once microclimatic constraints are better quantified and understood, it may be possible to overcome some through improved formulation, strain selection, genetic or phenotypic manipulation, and inoculum targeting. Identification of microclimatic constraints would also allow development of predictive models which would identify windows of opportunity thereby optimizing efficacious use of these mycopesticides.

Potential non-target effects of mycoinsecticides in Locust control

Ralf PEVELING

Integrated biological Control of Grasshoppers and Locusts Project,
GTZ, 65726 Eschborn, Germany
E-Mail : GTZ-POHASY@GEOD.Geonet.De

Mycoinsecticides against disease vectors and pests have been mainly used in temperate and humid tropical zones. Living in a "microbial world", invertebrates have co-evolved with pathogenic fungi and developed defence mechanisms to prevent infection or to contain the disease, and existing evidence suggests that side-effects on the non-target fauna of humid zones are either negligible or temporary. However, little is known about the impact of fungal control agents on invertebrate communities from arid and semi-arid environments where natural selection for mycopathogen resistance might be less important than in humid environments. Likewise, no data exist on the effect of fungal strains adapted to high temperatures on indigenous vertebrates, particularly reptiles, which are the only vertebrates where natural *Beauveria* and *Metarhizium* mycoses have been confirmed. Despite these reservations, only non-target organisms directly exposed to mycoinsecticides during locust control operations appear to be at risk. Secondary pick-up of viable spores seems unlikely under the prevailing climatic conditions of high insolation and temperature and low humidity.

Towards the development of integrated pest management for desert Locust control

Stephan KRALL

Deutsche Gesellschaft für Technische Zusammenarbeit (GTZ) GmbH
P.O. Box 5180, 65726 Eschborn, Germany
Fax : +49 6196 797413
E-mail : GTZ-POHASY@GEOD.GEONET.DE

The desert locust is well known as the most spectacular and feared pest for thousands of years. Apart from burning hoppers driven into ditches farmers had no real means to fight this insect until the beginning of our century. And there was no substantial impact on the population such as swarms. For the last 50 years synthetic insecticides have been used in the control. Neither the strategy of survey and control nor the kind of products have changed since. The only change has been replacing organochlorines by organophosphates, carbamates and synthetic pyrethroids in the 1980s. Monitoring the populations has been greatly improved through the introduction of the global position system (GPS). However the present control objective is still to locate desert locust populations and control early outbreaks to suppress upsurges and plagues. Despite the obvious failure of this strategy there is no change in sight. At present control decisions are not based on thresholds, which are unknown and only recently have some cost-benefit analyses been attempted. The present strategies do not meet IPM requirements, are difficult to justify on economic grounds and do not appear to be effective. For the last six years some research programmes are developing environmentally sound products to replace the synthetic insecticides, and less than three years ago discussions have started to design new control strategies. This paper reviews the results of these discussions. The strategy aims at developing a cheap monitoring system to detect early upsurges and control these when swarms have already formed. Control should only be initiated when crops are endangered directly. Finally in line with the requirements of IPM it should include alternative products.

SYMPOSIUM #3

COMMERCIALIZATION OF INSECT VIRUSES AND EXPRESSION SYSTEMS

Organizer :

E.M. DOUGHERTY
USDA, ARS, Insect Biocontrol Laboratory, Bldg. 011A,
Rm. 214, BARC-West, Beltsville, MD 20705-2350, USA
Fax : (301) 504-5104

The technical challenges associated with commercialism of non-engineered Baculoviruses

P. V. VAIL

USDA, ARS, Horticultural Crops Research Laboratory,
2021 S. Peach Avenue, Fresno, CA 93727, USA
Fax : 209 453 3088

Interest in the development of baculoviruses as microbial control agents has increased significantly. During the last five years many small and large organizations have been conducting basic and applied research to develop a broad spectrum of baculoviruses into commercial entities. A number of baculoviruses have been registered in Europe and the United States as a result of these endeavors by our commercial counterparts. The importance of technology transfer from the scientist(s) during the process of commercialization cannot be over-estimated. Other factors have also facilitated commercial development. In the United States, registration of non-engineered baculoviruses has been simplified considerably so that these alternative pesticides will have increased use. The EPA has "fast tracked" registration of these organisms by (1) decreasing data requirements, (2) allowing bridging of data at least with the multiply imbedded nuclear polyhedrosis viruses (*M*NPVs), and (3) reducing data turn-around times. Af*M*NPV and Ac*M*NPV were registered for less than $300,000 each in a very short period of time.

The question is, now that we have useful organisms, what do we do with them. Will the market be limited or specialized, *i.e.*, glasshouse-grown crops, vegetable or field crops ? How do we keep decision making within the bounds of the "technical merits" of the organism ? Transfer of this technology involves many steps (potential pitfalls); lack of attention to any one may be disastrous. Based on my observations, companies will try to market a product before the product is actually ready. There often is a dichotomy between marketing and development people. Judgments must be made carefully concerning the efficacy and limitations of the candidate virus or this can lead to some poor decisions, and possibly drastic consequences. It is easy to get caught in what might be called an "efficacy chain". One thing is certain, bad news travels faster than good news in the user community.

Some of the first decisions to be made are those of production methods, standardization and formulation. Once the product is ready for field testing, any number of steps must be taken to achieve a saleable and useful product. Perhaps the single most important decision is the selection of an efficacious dose followed by determination of application frequency based on persistence. Initial field trials must be highly regulated and standardized to obtain these data. Once a company develops it own "comfort zone", the virus then must be tested in larger and more intensive field trials with cooperators and/or Extension personnel. Again, testing protocols must be rigid and evaluations realistic. Once past this step, demonstration type trials must be conducted for the benefit of Extension personnel, pest control applicators, and growers. During this period any specific limitations or requirements for successful use must be thoroughly outlined to minimize the possibility of failure. Only after this developmental process occurs will a product result that is predictable, marketable and that will assure user confidence.

Rotavirus oral vaccines in animals using recombinant Baculoviruses in feedstocks

Robin A. ROBINSON

DynCorp, Molecular Virology Laboratory, Rockville, MD 20850, USA
Fax : 301 738 1109

Rotaviruses are the primary causative agents of viral diarrheal disease in animals and man. These infections are responsible for significant deleterious impacts on worldwide human and animal health in terms of both morality and economy. Additional concerns come from recent evidence of animals being reservoirs of new epidemic rotavirus strains that are human/animal viral reassortants. One approach to control the rotavirus spread has been the production of vaccines comprised of recombinant rotavirus virus-like particles (VLPs) from baculovirus-infected cells. These VLPs were composed of four viral capsid proteins, that form intact particles consisting of a core (VP2), and inner shell (VP6), and an outer shell (VP4 and 7). Chimeric VLPs were produced from insect cells infected with recombinant baculoviruses harboring viral capsid genes from different human and animal rotavirus strains. VLPs were purified on sucrose gradients, examined for particle composition and integrity, and formulated with animal feedstocks for administration as an oral vaccine. Antisera from animals fed for two days on the VLP-containing feeds contained both IgG_2 and IgA antibodies with specificity to human and animal rotavirus capsid antigens. Mucosal immunity was observed, as sIgA antibody to rotavirus capsid antigens was found in fecal samples and vaginal swabs of immunized pigs, and cattle challenged with live rotavirus strains of human and animal origin; however, nonimmunized animals developed a diarrheal disease within 24 hr. post-challenge. Efforts to administer feedstocks containing insect larvae lysates with rotavirus VLPs are on-going. These results demonstrate the utility of rotavirus VLPs produced in insect cells as a multivalent vaccine to limit the spread of rotaviruses among animal populations and to curb reassortment of human/animal rotavirus strains.

Exploitation of baculoviruses as expression vectors and insecticides.

Robert D. POSSEE

NERC Institute of Virology and Environmental Microbiology,
Mansfield Road, Oxford OX1 3SR, United Kingdom
Fax : 44-1865-59902
E-mail : possee@molbiol.ox.ac.uk

The rapid advances in baculovirus molecular genetics in the past 15 years has facilitated parallel developments in their use as expression vectors of foreign genes and genetically virus insecticides. The Institute of Virology and Environmental Microbiology (IVEM) has participated in these areas. Developments in baculovirus expression systems have resulted in the construction of multiple expression vectors able to synthesize up to five gene products in the same insect cell after infection with a single virus. Improvements in the methods for the selection of recombinant viruses have accompanied these advances. Much of this work has been accomplished with "in-house" funding ; permitting later commercialization and benefits from royalties etc. However, the obvious industrial use of such systems has attracted commercial funding which leads to a more directed approach. In concert with the development of expression systems, one of the most rewarding areas of research at IVEM in the last 10 years has been the construction of improved recombinant baculovirus insecticides. These viruses contain insect-specific toxin genes which improve the effectiveness of the unmodified baculovirus. In this presentation, I will summarize the major findings of our work over the last 10 years and who have been our major sponsors. In general, the collaborations we have enjoyed with industry have been very fruitful and given the trend in research within the UK, are likely to continue in future.

Toxin expression and insecticidal effects of recombinant Baculoviruses bearing novel anti-insect scorpion neurotoxin genes

N. CHEJANOVSKY[1], E. GERSHBURG[1,2], D. STOCKHOLM[1], H. RIVKIN.[1], N. ZILBERG[2], O. FROY[2] and M. GUREVITZ[2]

1 : Inst. of Plant Protection, The Volcani Center, Bet Dagan 50250, Israel
Fax : 972 3 9604180
E-mail : VPNJC@VOLCANI.AGRI.GOV.IL
2 : Dept. of Botany, Tel-Aviv University, Tel-Aviv 69978, Israel

Recombinant baculoviruses expressing the Lqh IT and the AaIT anti-insect scorpion toxins (isolated by us and others, respectively), possess enhanced insecticidal activity towards lepidopteran pests. In an effort to develop more potent baculoviruses against those pests we a) isolated recombinant *Autographa californica* Nuclear Polyhedrosis Viruses (*Ac*NPVs), which express novel excitatory (LqhIT1) or depressant (LqhIT2) anti-insect selective toxins and; b) genetically modified the toxin. *Trichoplusia ni* cells infected with the recombinant viruses secreted functional polypeptides. *Spodoptera littoralis* and *Heliothis armigera* larvae injected with the recombinant viruses showed typical symptoms of contraction and flaccid paralysis characteristic of the excitatory and depressant toxins, respectively. *Heliothis armigera* larvae fed with recombinant viral polyhedra showed pathogenic symptoms manifested earlier than the typical effects of baculovirus infection. Feeding was arrested earlier in larvae infected with recombinant virus than in wild type *Ac*NPV-infected larvae. Furthermore, genetic manipulation of the toxin resulted in enhanced insecticidal activity.

Densovirus-based expression vectors

M. BERGOIN, H. BOSSIN and C. ROMANE

Station de Recherches de Pathologie Comparée INRA, URA CNRS 2209,
Unité de Virologie moléculaire, 30380 Saint Christol-lez-Alès, France
Fax : (33) 67 14 46 79
E-mail : bergoin@viro.univ-montp2.fr
et Laboratoire de Pathologie Comparée, Université Montpellier II,
Pl. E.Bataillon, 34095 Montpellier Cedex 5, France

The concept of using densoviruses (DNVs) to express foreign genes in insect cells has emerged from the cloning of infectious sequences of the genomic DNA from the *Junonia coenia* DNV (*Jc*DNV) and the *Aedes aegypti* DNV (*Aa*DNV) (Jourdan *et al*, 1990; Afanasiev *et al.*, 1994). By inserting the procaryotic *lacZ* reporter gene downstream from the promoter driving expression of the structural polypeptides of both viruses, the synthesis of ß-galactosidase in insect cells transfected by these constructs has been achieved (Giraud *et al.*, 1992 ; Afanasiev *et al.*, 1994). Further studies have shown that transfection of the lepidopteran cell line SPC Sl 52 with *Jc*DNV-derived constructs expressing the *neo*^{r} gene led to the selection of G 418-resistant cells, thus providing evidence for their stable transformation. Analysis of the cell DNA showed that the recombinant viral genome maintained either as integrated in the cellular genome if the non structural (NS) genes were present, or as episomal in the absence of NS functions (Rolling *et al.*, 1991). Furthermore, it was shown that the deletion of the NS3 gene led to a tenfold increase of transformation frequency (10^{-4}) (Royer *et al.*, 1995). Owing to these properties, *jc*DNV-derived vectors appear as good candidates both for the constitutive expression of foreign proteins of interest in insect cell lines and for insect transgenesis. Data regarding these two aspects will be presented.

Jourdan, M. Jousset, F.X., Gervais, M., Skory, S., Dumas, B. 1990, *Virology*, **179**, 403-409.
Giraud, C., Devauchelle, G., Bergoin, M. 1992, *Virology*, **186**, 207-218.
Afanasiev, V.N., Kozlov, Y.V., Carlson, J.V., Beaty, B.J.1994, *Expetl. Parasitol.*, **79**, 322, 339.
Rolling, F., Bergoin, M., *Proc. IVth. Internat.Parvovirus Workshop*, 18-22 Aug., 1991,Elsinore, Denmark..
Royer, C., Romane, C., Guerin, M.A., Bergoin, M. *Proc. VIth Internat Parvovirus Workshop*, 10-14 Sept.1995, Montpellier, France.

Insect cell cultures and media for Baculovirus production

Dwight E. LYNN

USDA/ARS, Insect Biocontrol Laboratory,BARC-West, Bld 011A,
Rm 214, Beltsville, MD 20705-2350 USA
Fax : 301 504 5104
E-mail : dlynn@asrr.arsusda.gov

Development of insect cell lines has become a routine process since T.D.C. Grace first established lines from the lepidopteran *Antherea pernyi* in 1962. Within ten years, over 60 insect cell lines had been established and several of the lepidopteran lines had been used to replicate insect viruses, especially nuclear polyhedrosis viruses. Unfortunately, these early successes did not immediately translate into a commercial method for producing biopesticides. While the field of insect cell culture continued to flourish through the 1970's and '80's (by 1989 over 400 lines had been established from insects), the field received it's greatest stimulation with the development of the baculovirus expression vectors (BEVs). The work on Gypsy moth at our lab in the 1980's led to the development of several new cell lines from embryos and one from fat body. The fat body line in particular was substantially better at producing gypsy moth nuclear polyhedrosis virus than previously available cell lines and was closely examined by industry as a tool for producing the virus as a biopesticide. Also in the 1980's, several media manufacturers developed serum-free insect cell culture media for use in BEV systems. Subsequent efforts have been made to replace expensive components of these media for use in biopesticide productions. The history and current status of insect cell culture in the commercialization of insect viruses will be discussed.

Adjuvants for extending the residual activity of entomopathogenic viruses

Michael R. MCGUIRE

USDA-ARS, National Center for Agriculture Utilization Research, Bioactive Agents Research Unit, 1815 North University St., Peoria, IL 61604, USA
Fax : 309-681-6693
E-mail: mcguirmr@ncaur1.ncaur.gov

Adjuvants can be defined as additives to spray tanks that affect pesticide performance. Buffers and drift retardants are used to affect chemical and physical performance, while spreaders, stickers, and feeding stimulants can affect residual activity and overall efficacy. Research conducted with the addition of naturally occurring polymers such as cornstarch, flour, gluten, casein, lignin, etc. to spray tanks containing *Bacillus thuringiensis* has demonstrated that these materials can provide protection from sunlight and/or rainfall thus extending residual insecticidal activity. Upon spraying, the materials from a thin film entrapping the other ingredients within the spray droplet. Recently, the adjuvants have been tested with baculoviruses. Laboratory tests suggest that corn flour and sucrose mixtures can protect viruses from sunlight. For example, after 20 minutes exposure to simulated sunlight, virus without adjuvant lost all activity. Virus with adjuvant retained approximately 50% of original activity. After 80 minutes exposure, 30% activity remained. Similarly, solubilized lignin provided excellent protection from wash-off by simulated rain. Effects from the other ingredients were not as dramatic, and gluten actually was detrimental to the virus. Preliminary field tests from the summer of 1995 on cabbage will be used to build upon for field work in 1996. Aspects of the technology transfer process will be discussed in light of government and company policies with an emphasis on the critical relationships that are necessary between scientists from public and private sectors.

Use of granulosis virus to control potato tuber moth, *Phthorimaea operculella* (Zeller)

J. ALCAZAR

International Potato Center, P.O. Box 1558, Lima 100, Peru
Fax : (51) (14) 351570
E-mail : jalcazar@cip.org.pe

The International Potato Center (CIP) has been looking for non-chemical alternatives to control the potato tuber moth, *Phthorimaea operculella.* As a result, a granulosis virus (*Po*VG) was found affecting larvae of this species in Peru. After a process of research to determine the potential use of this agent, a simple method formulation was designed. This powder can be used to control Potato moth under storage conditions, and a liquid suspension can also be used in potato fields.

Pathogenic studies shown that 20 mg of purified *Po*VG or 20 infected larvae in a litre of water was enough to cause 100 % of larva mortality. Larvae died between 12 and 20 days after the ingestion of virus particles. Comparative studies under field conditions have shown that baculovirus caused 98, 88, 70 and 70 % of infected larvae after 2, 4, 6 and 8 weeks of virus application respectively. Similar results were found using *Bacillus thuringiensis* (Bactospeine) and methomyl insecticide. Another study was carried out to compare the efficiency of baculovirus (powder formulation) to control Potato moth under diffused-light storage conditions. The dose of 5 Kg/t of tuber was enough to protect them and after 120 days of storage, there were 8.5% of damaged tuber and 4 % of damage sprouts. The insecticide treatment (deltametrin) shown a control efficiency of 3 % of damaged tubers and 0.8 % of damaged sprouts after similar time, while the control treatment shown 100 % of both damaged tubers and sprouts.

CIP and Peruvian Ministry of Agriculture started the process of scaling up this technology, and during 1991, around 104t of basic potato seed were treated with baculovirus in different agricultural experimental stations and universities. Treated tubers only shown between 0 and 1.2 % of damage while control treatments shown beween 5.2 and 39.98 % of damage under similar conditions.

Baculovirus has become an important component of the IPM program, in conjuntion with sex pheromones, repellent plants and cultural practices, to control Potato tuber moth especially in Peru, Bolivia and Tunisia.

Process development challenges of Baculovirus production in Insect cells

E. L. WALLS, N. D. DELACRUZ, B. STILES, C. CELERI, M. KHIN, R. CHIARELLO, C. CAMARA, S. HIMMERICH and M.YU

American Cyanamid Co., San Leandro, CA 94577, USA

The cultivation of insect cells for the production of insect viruses to be used as biopesticides presents some unique problems for scale-up. Some challenges such as sterility, passage stability, process monitoring and control are common to many cell culture operations. There are particular problems in making a product for the agricultural market : the diversity of viruses and cell lines, the large volume production and the stringent manufacturing cost restraints. A review of these problems, from the mundane to the complex, will be presented.

SYMPOSIUM #4

GENETIC RESOURCES IN BIOLOGICAL CONTROL

Organizer :

M. VAN MONTAGU
Laboratorium voor Genetica, Universiteit Gent,
K.L. Ledeganckstraat 35, 9000 Gent, Belgium
Fax : 32 9 2645349

Comparative genetic mapping and its applications

K.M. DEVOS and M.D. GALE

John Innes Centre, Norwich Research Park, Colney,
Norwich NR4 7UH, United Kingdom

The value of plant genome projects and their applications in practical breeding programs is well understood and detailed genetic maps are now available for most of the major crops. Until recently, plant genome projects were focused entirely on a specific species, and research on different crops was carried out independently. Indeed, it appeared that little could be gained from interactions between for example maize, wheat, and rice geneticists. In the last few years, however, it has become clear that genes and their organization are highly conserved between species within a tribe, within a family, and possibly beyond the family level.
Colinearity between genomes of different species within a tribe has, unconsciously, formed the basis for crop improvement through the introduction of novel genes from wild relatives, and has been applied by breeders for decades. It was not till the late 1980s, however, that the construction of comparative genetic maps using a common set of restriction fragment length polymorphism (RFLP) markers elucidated genome relationships. Initially limited to species within a tribe - e.g. wheat and rye, maize and sorghum - genome analyses were quickly extended to include the major crops within the grass family - rice, the millet, sugar cane, sorghum, maize, and the Triticeae cereals. The high level of colinearity between genomes within the grass family has allowed their formal alignment, within which rice, due to its small genome size and diploid origin, is emerging as pivotal genome. The immediate benefits of this line of research are perceived to lie in increased availability of markers in any grass crop, the availability of a new precise tool for evolutionary studies, the possibility to isolate genes in any grass by map-based DNA walking in rice, and the potential to predict the locations and mechanisms of key genes in all species from their previous analysis in another. Ultimately, the pooling of all genetic, biochemical and physiological information from grass species will provide a quantum leap in our understanding of the individual major crops.

Understanding and improving mycoparasites for plant protection

Alfredo HERREA-ESTRELLA

Genetic Engineering Department,
Centro de Investigación y de Estudios Avanzados del IPN, Unidad Irapuato,
Apartado Postal 629, 36506 Irapuato, Gto. México

Among the different interfungal relationships perhaps the best exploited by men has been mycoparasitism. An example of such fungus-fungus interaction is that of the soil fungus *Trichoderma harzianum*, a mycoparasitic fungus well known for its use as a biocontrol agent of phytopathogenic fungi. Among other factors, *Trichoderma* produces a series of antibiotics and fungal cell wall degrading enzymes. These enzymes are believed to play an important role in mycoparasitism. Several research groups have recently engaged in elucidating the mechanism of action of this mycoparasite at the molecular level. Our initial approach involved the study of genes coding for its cell wall degrading enzymes. Chitin and β-(1,3)-glucan are the two major structural components of many plant pathogenic fungi, except by Oomycetes which contain cellulose in their cell wall and nearly no chitin. In the fungal cell wall these polysaccharides are embedded in a protein matrix. Thus we believe that chitinases, β-(1,3)-glucanases and proteases may play a major role, among the hydrolytic enzymes, in the control of a wide range of pathogens. We have cloned and characterized three genes encoding lytic enzymes, *prb1* (protease), *ech42* (chitinase), and *bgn66* (glucanase). The pattern of expression of these genes during the fungus-fungus interaction strongly suggests their involvement in mycoparasitism. Among other characteristics, all genes are induced by the presence of a host and repressed by glucose. Preliminary data suggest that induction of the expression of the three genes is independent of the cell wall composition of the host. Furthermore the overexpression of at least one of them in transgenic *Trichoderma* leads to higher biocontrol capacity of the strain. Using as an alternative approach differential display of mRNAs expressed in biomimetic assays, we have been able to identify a series of cDNAs which are expressed at early stages of the interaction. Initial characterization of these cDNAs points towards an important role in cell to cell communication. Finally, we will present data which correlate genome structure in different *Trichoderma* isolates with strain compatibility and biocontrolling characteristics.

Bacillus thuringiensis and its use to engineer insect resistance in crops

Jeroen VAN RIE

Plant Genetic Systems, Jozef Plateaustraat 22, B-9000 Gent, Belgium
Fax : 91 24 06 94

Bacillus thuringiensis has been used for more than two decades as an effective and safe microbiological insecticide. Its insecticidal activity is mainly due to the production of δ–endotoxins or insecticidal crystal proteins (ICPs) during sporulation. Most ICPs known today are active towards larvae of lepidopteran, coleopteran or dipteran insects. The mode of action of ICPs has been studied mainly for lepidopteran ICPs. Following ingestion, crystals are dissolved and the protoxins of about 130 kDa are liberated. These protoxins are proteolytically activated to a protease resistant fragment of about 60 kDa which binds to specific receptors on midgut cells. Subsequent to binding, part of the ICP molecule inserts in the membrane, leading to pore formation, cell lysis and finally death of the insect. Traditionally, *B. thuringiensis* has been delivered as formulated spore/crystal preparations. Since ICPs are monogenic, it seemed straightforward to engineer plants with these genes to confer insect resistance. Genes encoding the toxic fragment resulted in much higher levels of ICP expression and insecticidal activity than genes encoding protoxins. Moreover, by adapting ICP genes for plant expression, ICP levels in transgenic plants could be significantly increased. Such plants show agronomically relevant insect protection. The most serious threat to this new insect control technology is the possibility that insects develop resistance against ICPs expressed in plants. The implementation of resistance management tactics should prevent or delay such resistance build-up and safeguard *B. thuringiensis* as an efficient and environmentally sound insect control agent.

Effect of marker genes on the consumption of transgenic potatoes by *Leptinotarsa decemlineata*, the Colorado potato beetle

Anne LECARDONNEL[1,2], Geneviève PREVOST[1]
and Brigitte SANGWAN[2]

1 : Laboratoire de Biologie des Entomophages, UPJV,
33 rue St Leu, 80039 Amiens, France
Fax : (33) 22 82 78 92 E-mail : genevieve.prevost@sc.u-picardie.fr
2 : Laboratoire Androgenèse et Biotechnologie, UPJV,
33 rue St Leu, 80039 Amiens, France

Rapid advances in recombinant DNA technologies opened new opportunities of crop improvement. Many techniques now available allow us to introduce genes of interest in agronomical plants. The most widespread technic using *Agrobacterium*-mediated gene transfer already gave rise to many transgenic organisms. However, the possible occurrence of genetically modified organisms in the field is giving rise to many questions concerning their possible detrimental effect. In order to discriminate between successful and unsuccessful transformation event, marker genes are of bacterial origin. Therfore, transgenic plants possess proteins resulting from the expression of marker genes in addition to the ones related to the genes of interest. Though no detrimental effect of marker genes was reported in the literature yet, we may wonder about marker proteins which are expressed in transgenic plants and possibly modify its answer to environmental factors. Present study was conducted to evaluate the effects of marker genes present in the genome of transformed potatoes on to the food intake by one very destructive pest, the Colorado potato beetle.

Genetic transformation of potato was achieved using *Agrobacterium tumefaciens*. NptII-Gus genes were introduced into potato genome then transgenic plants were subsequently analysed to confirm the presence of foreign genes and to evaluate their expression rate. Differents lines of transgenic plants were greenhouse grown then submitted to food intake by larvae of the colorado potato beetle.

Results showed a surprising increase of foliage consumption in the transgenic potatoes expressing marker genes. This results demonstrates that genetic transformation may modify the physiology of the plant, and subsequently its response to some environmental factors (here, insect's food intake).

Evaluating the potential of transgenic *Bt*-cotton plants for the control of *Helicoverpa armigera*

K. OLSEN and J. DALY

CSIRO, Division of Entomology, GPO Box 1700,
Canberra, ACT 2601, Australia
Fax : 61 6246 4150
E-mail : joanned@ento.csiro.au

Transgenic plants, expressing insecticidal toxins, provide a new approach to the control of insect pests. The narrower spectrum of activity of these toxins, particularly their inactivity relative to natural enemies enables transgenic plants to be used in integrated pest management systems. Cultivars of commercial cotton, genetically engineered to express the endotoxins from *Bacillus thuringiensis,* are undergoing commercial release in USA and Australia. In Australia, the main targets are the two pests in the genus *Helicoverpa.* A reduction in the use of pesticides in cotton crops through the use of transgenic plants provides a real opportunity to implement biological control for cotton pests.
Variation in the efficacy of the transgenic plants in field trials has increased concern about the development of resistance in *H. armigera* to these plants. Attempts to study factors that impact on field efficacy have been hampered by the lack of bioassay techniques to quantify these changes in cotton. We report on a bioassay method that we developed to quantify the efficacy of *Bt*-cotton plants during growing season. A number of methods were trialed but a leaf mush bioassay in which larvae are fed a mixture of normal and transgenic leaf material was chosen. Because on small amounts of *Bt*-leaf was required and the results were reproducible. We have measured relatively small changes in efficacy of *Bt*-cotton plants, of the order of 3-fold, from young to mature plants. The cause of these changes is not known but we have evidence that the *Bt* toxin in the plants is becoming less available to the larvae in more mature plants.

Integration of molecular markers in poplar breeding programs

Maria Teresa CERVERA[1], Marijke STEENACKERS[2], Veronique STORME[1], Jaqueline GUSMAO[1], An Van den BROECK[2], Marc VAN MONTAGU[1] and Wout BOERJAN[1]

1 : Laboratorium voor Genetica, Flanders Interuniversity Institute for Biotechnology, Univ. Gent, K.L. Ledeganckstraat 35, 9000 Gent, Belgium Fax : 32 9 264 5349
2 : Institute of Forestry and Game Management, Gaverstraat 4, 9500 Geraardsbergen, Belgium

Due to the long generation time of trees, classical tree breeding is a time-consuming process. For poplar, one of the most planted tree species in Europe, only three generations of controlled crosses have been performed. This is in contrast to annual crops which have often been selected for over thousand generations. Tree improvement though is important given the high market position of wood worldwide. The natural European clones, such as *Populus nigra* are sensitive to leaf rust caused by *Melampsora larici-populina*, a fungal pathogen that causes premature defoliation and hence can significantly reduce the growth rate. Because among the American species resistant clones have been found, most commercially used poplar clones in Europe are hybrids between American species or between American and European species. These hybrids show a large heterosis effect. Another important pathogen, *Xanthomonas populi*, causes bacterial canker. Wood of such diseased trees is of very low quality. Using molecular techniques, it is possible to identify molecular markers associated with traits of interest, such as resistance towards pathogens. Amongst others, these molecular markers can be used in marker-assisted breeding programs as a diagnostic method to predict at the seedling stage if a new hybrid has certain characteristics or not. In this way, molecular marker analysis can speed-up breeding programs. A cross has been made between an American species (*Populus deltoides*) that was resistant to three races of *M. larici-populina* and a European species (*P. nigra*) that was susceptible to these races. Half of the progeny of this cross were resistant and half were susceptible. By comparing AFLP™ DNA fingerprints of the resistant with those of the susceptible progeny, molecular markers were identified that were inherited together with the resistance trait. Resistance against the three races of *M. larici-populina* is conferred by a dominant locus. Understanding the genetics of disease resistance should improve future breeding strategies.

[This project is funded by the European Union (AIR1-CT92-0349) and the Flemish Government (BNO/BB/1994)].

How can plant genome studies contribute to the development of new biocontrol techniques

Marc VAN MONTAGU

Laboratorium voor Genetica,
Flanders Interuniversity Institute for Biotechnology (VIB),
Universiteit Gent, K.L. Ledeganckstraat 35,
9000 Gent, Belgium
Fax : 32 9 2645349

Better environmental awareness increases the demand for biocontrol in pest management. Although empirical approaches and judicious use of careful observations by naturalists brought tremendous progress, it is self evident that the availability of molecular techniques can bring the construction of totally novel biocontrol agents. The contributions will come in the forst place from the unraveling of the molecular base of the plant's defense reactions. Pathogen stress has been linked to oxidative stress. H_2O_2 seems to play a role in systemic acquired resistance. The engineering of plants with a lowered catalase activity (Chamnongpol *et al.*, 1996) could bring a good starting tool to study the induction of defense reactions in plants. Understanding how nematodes induce the formation of feeding sites on the plant roots can allow the engineering of plants which no longer respond to the nematode "challenge" and, hence, trap nematodes and can well survive the infection (Gheysen *et al.*, 1996).
Now that the complet sequence of the yeast genome is available, biochemists and cell biologists realize the wealth of information that accumulated through this program. The systematic study of the *Arabidopsis thaliana* genome is accelerating very rapidly. It is easy to predict that the endeavour will bring essential contributions to our knowledge on the structure and function of plant defense genes. Together with the availability of a dense map of polymorphic restriction sites (Cnops *et al.*, 1996) thanks to the development of the AFLP technique (Vos *et al.*, 1995), *Arabidopsis thaliana* can also become a model plant for the study of plant-pathogen interactions. This means gathering of knowledge needed for obtaining better biocontrol agents.

References :

Chamnongpol, S., Willekens, H., Langebartels, C., Van Montagu, M., Inzé, D. & Van Camp, W. - 1996. Trangenic tobacco with a reduced catalase activity develop necrotic lesions and induce pathogenesis-related expression under high light. Plant J., in press.

Cnop, G., den boer, B., Gerats, T., Van Montagu, M. & and Van Lijseberrens, M. - 1996. Chromosome landing at the Arabidopsis TORNADO1 locus using an AFLP-based strategy. Mol. Gen. Genet., in press.

Gheysen, G., Van der Eycken, W., Barthels, N., Karimi, M & and Montagu, M.- 1996. The exploitation of nematode-responsive plant genes in novel nematode controll methods. Pestic. Sci. 47, 95-101.

Vos, P., Hogers, R., Bleeker, M., Reijans, M., van de Lee, T., Hornes, M., Frijters, A., Pot, J., Pleman, J., Kuiper, M. & Zabeau, M. - 1995. AFLP : a new technique for DNA fingerprinting. Nucleic Acids Res. 23, 4407-4414.

Recent advances in the successful field application of the sterile insect technique for the control and/or eradication of *Ceratitis capitata* and *Glossina austeni*

A.S. ROBINSON

Entomology Unit, FAO/IAEA Agriculture and Biotechnology Laboratory,
IAEA Laboratories, A-2444 Seibersdorf, Austria
Fax : (+431) 2060 28447
E-mail : robinson@riall.iaea.or.at

The use the Sterile Insect Technique for the control of certain key agricultural pests is now a conventional approach to rational insect pest management. It can be easily integrated with other forms of biological control to develop environmentally friendly area wide pest management. The Joint Division of the FAO/IAEA in Vienna and the Agriculture and Biotechnology Laboratory of the Agency's Laboratories, Seibersdorf, Austria, have as part of their mandate the development and improvement of this technology and its transfer to operational programmes.

At present, two large projects are being actively supported, one is directed to the eradication of the tsetse fly, *Glossina austeni* from Zanzibar and the other focussing on the eradication of the medfly, *Ceratitis capitata*, from large areas in the Province of Mendoza, Argentina. Recently Chile declared eradication of medfly due partially to an excellent SIT programme in the north of the country. Significant progress is being made in the implementation of these SIT programmes and it will be reported on together with the recent scientific and technical improvements introduced into the programmes.

In Mendoza, the efficiency of the technique has been improved by the introduction of a genetic sexing strain into the mass-rearing facility. This strain is based on a difference in pupal colour between males and females, the male and female pupae being separated by machine. Attempts are underway to utilize the female pupae for the production of parasitoids. On a somewhat smaller scale a genetic strain based on female temperature sensitivity has been introduced into a medfly mass-rearing facility in El Pino, Guatemala.

Tsetse mass-rearing has presented a major bottleneck to the large-scale application of SIT for tsetse control. Flies for release on Zanzibar are now produced mainly at a rearing facility at Tanga, on mainland Tanzania, where a colony of 500,000 female *G. austeni* has been established. This facility has recently introduced changes to the rearing process which has revolutionized the mass-rearing of tsetse. These developments together with

current research at the Agency's Laboratories in this area will be highlighted.
The successful eradication of the screwworm from North Africa using SIT will also be reported on.

Relevance of genetic markers in evaluating biological control by predatory mites

Maria NAVAJAS[1,2] and Howard THISTLEWOOD[3,4]

1 : INRA-ENSA.M, Laboratoire de Zoologie, 2 place Viala, 34060 Montpellier cedex 1, France
Fax : (33) 67 52 15 54 E-mail : navajas@ensam.inra.fr
2 : Institut des Sciences de l'Evolution, Laboratoire Génétique et Environnement, CNRS-UMR 5554, 34095 Montpellier cedex 05, France
3 : European Biological Control Laboratory USDA ARS, B.P. 4168, 34092 Montpellier cedex 5, France Fax : (33) 67 04 56 20
4 : Agriculture & Agri-Food Canada, Vineland, Ontario, Canada

Outdoor release of mites as natural enemies of weeds or arthropods is occurring in many countries but can provide equivocal results and requires an understanding of dispersal and population biology that is often lacking. Factors of interest include variation within species for the selection of candidate strains for introduction, and their establishment, dispersal, and potential exchange with local populations.

We are conducting genetic studies on mites and use the example of *Amblyseius fallacis* (Acari: Phytoseiidae). Since 1990, releases of a mass-reared pesticide-resistant strain of *A. fallacis* have been employed in inundative and augmentative releases on Canadian fruit crops for the biological control of spider mites. We review briefly the results and the major factors affecting the success or failure of releases to date.

We are pursuing two approaches to evaluate releases and provide information on establishment and dispersal of released predators. Insecticide resistance, as has earlier been employed as a genetic marker for this species. Recently, we have developed molecular markers to characterise populations of *A. fallacis* using allozyme analysis by isoelectric-focusing electrophoresis and by analysis of variation in DNA sequences. Using our results, we discuss the potential of these approaches in biological control studies.

SYMPOSIUM #5

BIOLOGICAL CONTROL OF PLANT DISEASES : STRATEGIES AND IMPLEMENTATION

Organizer :

Nyckle J. FOKKEMA
DLO Research Institute for Plant Protection (IPO-DLO)
P.O. Box 9060, 6700 GW Wageningen, The Netherlands
Fax : 31 317 410113
E-mail : N.J.FOKKEMA@IPO.DLO.NL

Strategies for biocontrol of foliar fungal pathogens with emphasis on suppression of sporulation of necrotrophic pathogens

J. KÖHL and N.J. FOKKEMA

DLO-Research Institute for Plant Protection (IPO-DLO),
P.O. Box 9060, 6700 GW Wageningen, The Netherlands
Fax : 31 317 410113
E-mail : N.J.FOKKEMA@IPO.DLO.NL

Three different strategies in biological control of foliar diseases can be distinguished :

1. Microbial suppression of infection. The major role of naturally occurring as well as introduced yeasts or hyphal fungi seems to be the removal of infection-stimulating exogenous nutrients, but also other mechanisms may be involved. The pathogen, however, may rapidly escape from antagonism by penetrating the leaf. Main prospects include wound protection and post-harvest diseases.

2. Microbial suppression of pathogen sporulation. Suppression of the dissemination of the pathogen will reduce the progression of epidemics. This approach allows a long interaction period between the antagonist and the pathogen and is successfully applied in the control of mildews and rusts using mycoparasites. Promising, and relatively new, is this approach in relation to necrotrophs sporulating on dead plant material. Among several saprophytic fungi naturally colonizing plant debris, *Ulocladium atrum* has been selected as a potential BCA because of its ability to suppress sporulation of *Botrytis cinerea* on dead plant tissue under a broad range of fluctuating micro-climatic conditions. Under field conditions sporulation of *B. cinerea* was reduced by 90%. Although the consequences for disease development may vary with the host and cropping conditions, *U. atrum* has the potential to become a broad spectrum BCA because interactions in dead plant tissue are not very specific for host or pathogen species.

3. Microbial suppression of pathogen survival. Mycoparasites may interfere with the formation and vitality of sclerotia in infected above-ground plant tissue and crop remains. Depending on the importance of initial inoculum, treatment of infected plant material may reduce the severity of the disease in the following seasons.

Use of Trichodex (*Trichoderma harzianum* T39) in IPM of *Botrytis cinerea* and other diseases

Y. ELAD

Department of Plant Pathology, ARO, The Volcani Center
Bet Dagan 50250, Israel
Fax : 972 3 9683543
E-mail : VPELADY@VOLCANI.AGRI.GOV.IL

Trichoderma harzianum Isolate T39, originally isolated from a cucumber fruit, was found to be effective under commercial conditions against *B. cinerea* on grapevine and greenhouse vegetables. It was developed as a formulated product (the first biocontrol agent against grey mould to be used commercially) and is now marketed in several countries as TRICHODEX 25 (sprayable powder), for control of grey mould on grapevine and vegetable crops. The case of this commercialized biocontrol agent can serve an example for strategies development and of implementation of biological control of foliar plant diseases. Considerations of control of diseases, effect on pathogen populations and on the plant products will be demonstrated.
Experiments have concentrated on developing integrated biological and chemical control programs. TRICHODEX is sprayed in alternation with common chemical fungicides either on a calendar basis or according to the developmental stage of the crop. Alternation treatments gave more reliable control of grey mould than other treatments ; the variation in percentage of disease control was lower in alternation treatments than in treatments with TRICHODEX or fungicides alone.
More than 150 field experiments on grapevines were completed between 1988 and 1995 throughout the world. Most of these experiments compared TRICHODEX at one or two rates with one or more chemical fungicides. Recently, some experiments were designed to test the efficacy of integrated biological and chemical disease control programs involving alternating applications of TRICHODEX with chemical fungicides. Results of such experiments showed significant control of the disease by TRICHODEX or by the integration treatment. In several experiments, the alternation treatments were compared to reduced chemical treatment programs in which chemical fungicides were applied only at the times when the chemical fungicides were applied in the alternation programs. Disease control in the TRICHODEX/chemical fungicide alternation programs and the reduced chemical fungicide programs were 60% and 40%, respectively.
The biocontrol agent was found effective also against other diseases which attack greenhouse crops, such as *Fulvia fulva* (*Cladosporium*) the causal agent of tomato leaf mould and *Sclerotinia sclerotiorum* the causal agent of cucumber and lettuce white mould. Thus, broader spectrum of diseases is

controlled by TRICHODEX alone or when combined with chemical fungicides. Further, when applied to the root zone in the nursery, TRICHODEX was found by some researchers also effective in control of soilborne pathogens and gave increased growth response in the absence of major pathogens.

In addition to providing disease control, integrated biological and chemical disease control programs can potentially reduce pesticide residues in fruit at harvest and lessen the pressure towards development of fungicide-resistance populations of *B. cinerea*. Furthermore, late season application of TRICHODEX do not affect the quality of white or red wines produced from the berries.

Control of Powdery mildew (*Uncinula necator*) in California vineyards using *Ampelomyces quisqualis*

Doug GUBLER

Department of Plant Pathology, University of California
Davis, CA 95616, USA
Fax : (916) 752 5674
E-mail : WDGUBLER@UCDAVIS.EDU

Powdery mildew is the most important disease of grape in California, both in terms of the dollars spent on disease control and crop loss due to disease. Historically sulfur, in various forms, has been used for disease control. In 1982, DMI fungicides became available for use in California and sulfur use declined; the result was resistance development to triadimefon by 1986. More recently, oils and a commercial formulation of *Ampelomyces quisqualis* (AQ_{10} Ecogen Corp., Langehorn, PA) have shown commercial disease control in California vineyards, particularly when used together. Research trials with AQ_{10} began in 1993 with mixed results, *i.e.* product use in some locations significantly reduced both incidence and severity of disease. In the subsequent years of 1994 and 1995 similar results were obtained. In examining the data, it became obvious that when disease was present before AQ_{10} application, disease quickly got out of control. On the other hand, when AQ_{10} application was initiated prior to infection, disease was controlled effectively for several weeks. In 1996, research trials were established to examine timing of application in relation to *U. necator* ascospore release. Treatment one was initiated at budbreak, treatment two was initiated six hours after ascospore release while treatment three was not applied until leaves were rated as having 14 percent infection, and the control vines were not treated. Disease control was highly effective for 8 weeks (4 applications of AQ_{10}) when applications were made at budbreak or timed with ascospore release. When already infected leaves were treated with AQ_{10},disease control was lost immediately. Growth chamber studies suggest a reduction in inoculum accounts for disease control.

An integrated biological control strategy for foliar bacterial diseases on Tomato

M. WILSON

Department of Plant Pathology, 209 Life Sciences Building,
Auburn University, Auburn, AL 36849-5409, USA
Fax : 334 844 1947
E-mail : mwilson@ag.auburn.edu

Tomatoes are affected by three different foliar bacterial diseases, speck, spot and canker, caused by *Pseudomonas syringae* pv. *tomato, Xanthomonas campestris* pv. *vesicatoria* and *Clavibacter michiganensis* subsp. *michiganensis*, respectively. Control practices for these diseases include the use of pathogen-free seed/transplants and protective applications of copper. Unfortunately, crops derived from pathogen-free seed or transplants are still infected by immigrant pathogen inoculum and copper resistance has appeared in both *P. syringae* pv. *tomato* and *X. campestris* pv. *vesicatoria*; hence, current control practices are inadequate. Therefore, the development of an integrated biological control (IBC) strategy for these diseases is highly desirable. The IBC strategy under development aims to combine foliar applications of copper-tolerant antagonists ; seed-treatment or soil drenches with plant growth-promoting rhizobacteria (PGPR) which induce systemic disease resistance; and/or foliar applications of chemicals which induce disease resistance. Most progress with foliar antagonists has been made with control of speck. Under greenhouse conditions, significant reductions (> 60%) in speck severity have been achieved using foliar applications of *Pseudomonas fluorescens* A506 and the non-pathogenic *P. syringae* strains TLP2 and Cit7. These biocontrol agents were field tested in four different U.S. locations in 1996. Further, cultures of *P. fluorescens* A506 have been re-isolated from the field which exhibit significantly increased levels of copper tolerance and their integration with reduced frequency copper sprays is under investigation. With respect to induced systemic resistance, several PGPR strains of the genera *Burkholderia* and *Bacillus* have been found to significantly reduce speck severity when applied as a seed treatment and soil drench. Additionally, induction of resistance to speck and spot of tomato is being investigated using novel biorational chemistries.

KEY WORDS: tomato, foliar, bacteria, biocontrol, speck

Biological control of *Fusarium* wilts

Claude ALABOUVETTE

INRA CMSE, Flore Pathogène du Sol, BV1540,
21034 Dijon Cedex, France
Fax : (33) 80 63 30 41
E-mail : ala@dijon.inra.fr

Fusarium wilts caused by *formae speciales* of *Fusarium oxysporum* are among the most severe diseases in the world. The available control methods are not efficient or difficult to apply. These difficulties in controlling the disease explain why studies on biological control of *Fusarium* wilts have a long history.
The existence of soils that naturally limit the incidence of *Fusarium* wilts stimulated research on biological control. The aim was to understand the mechanisms responsible for disease suppressiveness in order to reproduce or induce them in conducive soils.
Soils naturally suppressive to *Fusarium* wilts have several characteristics in common, indicating that suppressiveness is microbiological in nature. Competition for nutrients is a mechanism of soil suppressiveness, carbon and iron being limiting elements in suppressive soils. Competition for nutrients is related to the total microbial biomass of the soil but specific populations of antagonists also contribute to the competition. The most consistent results show the involvement of non-pathogenic *F. oxysporum* and fluorescent pseudomonads. Therefore selected strains of non-pathogenic *F. oxysporum* and fluorescent *Pseudomonas* spp. have been used to achieve biological control of *Fusarium* wilts. Introduced alone in conducive soils, they control *Fusarium* wilts more or less efficiently. But the association of a strain non-pathogenic *F. oxysporum* (Fo47) with a strain of *Pseudomonas fluorescens* (C7) always gives good control of *Fusarium* under experimental conditions.
Mechanisms other than competition for nutrients also contribute to the efficacy of these biocontrol agents. Indeed, competition between the pathogen and the biocontrol agents for infection sites at the root surface or inside the plant tissues, and induction of resistance of the host plant have been proved to be involved in mechanisms of disease suppression.
Finally other biocontrol agents such as *Bacillus* spp. and *Streptomyces griseoviridis* have also the capacity to control *Fusarium* wilts, the latter being already registered in some countries.

Use of a colonization-impaired strain of *Enterobacter cloacae* in biocontrol of *Pythium ultimum* damping-off of cucumber

D.P. ROBERTS, P.D. DERY, P.K. HEBBAR and W.L. MAO

USDA-ARS, Biocontrol of Plant Diseases Laboratory,
Beltsville, MD 20705, USA
Fax : 301 504 5968
E-mail : DRoberts@asrr.arsusda.gov

The importance of root colonization for suppressing damping-off by *Pythium ultimum* was studied using *Enterobacter cloacae* strain A-46, a colonization-impaired mini-Tn5 Km mutant of *E. cloacae* strain 501R3 (Roberts *et al.*, 1996. Can. J. Microbiol. 42 : 196-201). Strain A-46 was incapable of increasing in population in cucumber spermosphere in the absence of seed amendments while populations of strain 501R3 increased substantially. Strain A-46 was detected at populations of approximately 10^1 cfu per gram of root tissue 42 days after planting whereas strain 501R3 was detected at populations of approximately 10^8 cfu per gram. Despite being colonization-impaired, strain A-46 provided effective biocontrol of *P. ultimum* damping-off of cucumber grown in potting mix at 24°C. Plant stands from seeds treated with 501R3 or A-46 were similar and stands from both treatments were greater ($P \leq 0.05$) than with the pathogen alone. Biocontrol of *P. ultimum* on cucumber in the absence of effective root colonization may be due to the limited time that cucumber is highly susceptible to damping-off by this pathogen. Cucumber seeds pregerminated for 29 hours or longer before sowing in pathogen-infested potting mix yielded plant stands similar to the healthy control treatment, while plant stands from non-pregerminated seeds were significantly less ($P \leq 0.05$) than these treatments. In a natural field soil containing several *Pythium* spp. and *Fusarium* spp. seed treatments that combined applications of *E. cloacae* A-46 and *Burkholderia cepacia* strains provided superior biocontrol ($P \leq 0.05$) over treatments containing either *E. cloacae* or *B. cepacia* alone. We are currently evaluating the advantages of using the colonization-impaired strain, strain A-46, in biocontrol preparations that contain combinations of biocontrol bacteria targeted against multiple pathogens.

Biological seed coating as a biocontrol strategy

Bernadette A.M. KROON and Rudy J. SCHEFFER

S&G Seeds B.V.,Postbus 26, 1600 AA Enkhuizen, The Netherlands
Fax : 0 228 31 28 18
E-mail : Bernadette.Kroon@gwa.sandoz.com

The use of microbial inocula in agricultural practice has been subject of research for many years. Most widely used are the nitrogen fixing rhizobia on plants grown at low nitrogen levels. Other micro-organisms have been shown to improve growth or yield plants or to control soil-borne diseases. Use of biologicals is especially attractive in those cases where no resistance or chemical alternatives are available. Furthermore, biologicals can replace certain uses of agrochemicals.

Biological inocula applied on seeds are often more effective than inocula applied to the soil. This may be explained by considering that a seed inoculant obviously is present at a relatively high density at the seed and therefore near the emerging rootlet. The micro-organisms colonize the emerging root and can be effective against diseases on the root surface. We demonstrated that several fungi and bacteria applied to seeds are able to suppress damping off caused by *Rhizoctonia* or *Pythium*.

At S&G Seeds a coating with a beneficial *Pseudomonas fluorescens* strain was developed for radish. In greenhouse trials during several years it was shown that his strains applied to the seeds reduced *Fusarium* infections and resulted in higher yields. Successive sowings of coated seeds resulted in an increase in bacterial density on the roots. It is assumed that colonization from the starving root systems of the preceding crop takes place. Rapid spread of the pathogen after steam disinfestation of the soil may be prevented by the beneficial *Pseudomonas*. As long as no completely resistant radish cultivars are available, BioCoat is a valuable tool to help growers produce a healthy crop economically.

Development of *Gliocladium virens* for control of *Pythium* and *Rhizoctonia*

R. D. LUMSDEN

USDA/ARS, Beltsville Agricultural Research Center,
Beltsville, Maryland, USA
Fax : 301 504 5968
E-mail : rlumsden@asrr.arsusda.gov

Commercial development of the biocontrol fungus, *Gliocladium virens* (=*Trichoderma virens*), was initiated with several important criteria established during the early stages of research. These included : 1) use of a screening method for detecting an appropriate biocontrol microorganism for the targeted pathogens, that would translate to actual practice; 2) selection of the targeted pathogens, *Pythium ultimum* and *Rhizoctonia solani,* which cause important damping-off disease problems in commercial greenhouse operations ; 3) selection of microorganisms which were native to the U.S., because indigenous ones might less likely be problems for the U.S. environment ; 4) use of a single strain of a biocontrol agent was desirable for controlling both disease problems, for the sake of simplicity ; 5) use of a commercially-available soilless medium, which was widely accepted in commercial operations ; and 6) selection of high value crops, important to the ornamental plant production industry, for maximum cost effectiveness to private industry. Selection of these criteria was based on the concept that they would enhance the process of registration by regulatory agencies, commercialization of a product by private industry, and acceptance by the end-users, greenhouse grower operators and the general public. From over one hundred different isolates of bacteria, actinomycetes and fungi, most of which had some prior history of biocontrol potential or were originally isolated from survival propagules of plant pathogens, *Gliocladium virens* (strain Gl-21) was selected as the most consistent and effective strain. Processes for fermentation and formulation were tested and selected, based on success in producing an inexpensive product with properties compatible with delivery systems, with a long shelf life (>1 year), and with stable and consistent effectiveness against *Pythium* and *Rhizoctonia* damping-off. A product, SoilGard™, is now on the market for control of damping-off in greenhouse operations.

Prospects of the use of mycoparasites in biocontrol of Sclerotium-forming pathogens

P.H.J.F. van den BOOGERT

DLO-Research Institute for Plant Protection (IPO-DLO),
P.O. Box 9060, NL 6700 GW Wageningen, The Netherlands
Fax : 31 317 410113
E-mail : P.H.J.F.VANDENBOOGERT@IPO.DLO.NL

A crop infected by sclerotium-forming pathogens leaves numerous sclerotia in soil or on plant residues to secure their long term survival. Current control measures of sclerotial diseases are based on the eradication of sclerotia from soil prior to planting or on the protection of the plant with fungicides. Reductions of the size and viability of sclerotial populations is the main objective for biocontrol by mycoparasites.

Mycoparasitic fungi have in common their ability to exploit fungal host cells as a protected food source. Obligate mycoparasites are completely dependent on fungal hosts for growth and development in nature, whilst facultative mycoparasites are primarily saprophytes. Their mycoparasitic ability facilitates rather than overcomes competition for nutrients or sites. Although specific in their host range, obligate mycoparasites possess a highly efficient strategy to infect and kill sclerotia and suppress their formation.

For efficient control timing and targeting of the biocontrol agent at the production phase of inoculum, that is at sclerotium formation, would have the advantage of maximising contact with the sclerotium-producing pathogen. The possibilities for biocontrol of the sclerotium-producing *Sclerotinia* spp. and *Rhizoctonia solani* have been increased recently by the mycoparasites *Coniothyrium minitans* and *Verticillium biguttatum* and comprises the application on a diversity of crops and their residues (lettuce, potato, bean, sunflower) and on crop products during storage (potato tubers, carrots or chicory roots). Although their biocontrol effect is well-established in agricultural practice, several points using the aforementioned mycoparasites are of concern. Firstly, their cost-effective mass-producibility is still a problem, although encouraging progress is being made in fermentation technologies. Secondly, the compatibility with current culture and control measures needs to be verified before integrated control can be implemented. *Verticillium biguttatum* has exhibited this trait in respect to compatibility with other biocontrol agents, including *C. minitans* and with current pesticides. These and other research data will be presented and discussed against a background of prospects of practical application.

SYMPOSIUM #6

USE OF PHEROMONES IN BIOLOGICAL CONTROL

Organizers :

H. ARN
FAW 292, 8820 Wädenswil, Switzerland
Fax : 1 780 63 41
E-mail : Heinrich.Arn@wae.faw.admin.ch

and

P. WITZGALL
SLU, Box 44, 230 53 Alnarp, Switzerland
Fax : + 46 40 46 21 66
E-mail : peter.witzgall@vsv.slu.se

Mating disruption in vineyards : determination of population densities and effects on beneficials

F. LOUIS[1], K.J. SCHIRRA[2] and M. FELDHEGE[3]

1 : Staatliche Lehr- und Forschungsanstalt für Landwirtschaft, Weinbau und Gartenbau, Fachbereich Phytomedizin, Breitenweg 71, 67435 Neustadt/Weinstrasse, Germany
Fax : 0049 6321 67 1222
2 : Chrysopa CI Engelhardt, 1961 Nax, Switzerland
3 : Chembico Chemische und biologische Präparate Produktions GmbH, Hauptstr. 13, 67283 Obrigheim, Germany

For several successive years, investigations were conducted in the viticultural region of Palatinate, Germany. The primary goal of the field trials was to investigate the occurrence of populations of the grape berry moth *Lobesia botrana* Schiff., influenced by artificial pheromone atmosphere. For mating disruption, BASF dispensers were applied in a vineyard area of approx. 50 ha at a rate of 500 dispensers/ha (RAK 1+2 ; 120 g/ha major pheromone component of *L. botrana* ; (*E*,*Z*)-7,9-dodecadien-1-yl acetate). For reviewing the efficiency of pest control by mating disruption, 3 control plots of approx. 150 m^2 each were left untreated by insecticides.

The Palatinate is not only known for its great number of different wine varieties, wine growing has become more and more difficult due to the fact that *L. botrana* can cause considerable economical damage. In one year, infestation rates caused by the pest's second generation reached as much as 674 larvae per 100 bunches of grapes (674%) in the control plots. The degrees of effectiveness within the pheromone treated site increased during the course of the investigations and equalled almost 100% towards the end of the investigations.

It is known, that mating disruption is successful below a "critical density of moths". Above this critical density, mating disruption can no longer be effective due to the fact, that high densities of moths make sexual orientation by pheromone unnecessary. In order to determine the threshold of the critical density, a non-luring trapping system (catching device) was developed and successfully tested during the investigations. The catching results showed, that under present conditions, 8000 moths per ha (4 male and 4 female moths per 10 m^2) is the threshold of the pest's second generation, below which mating disruption is most likely to be successful. Field investigations showed also, that once this critical density of moths of *L. botrana* was exceeded, mating took place in presence of pheromone,

promptly being followed by oviposition. This preliminary critical density gives a rough idea, whether a vineyard area is appropriate for mating disruption or not.
Another important issue was to find an answer to the question, why infestation rates increase towards the borders of pheromone treated areas. Therefore modified light traps were installed in the border zone. The light traps were equipped with funnels pointing in 4 different directions. The trapping results proved, that male moths are highly attracted to a pheromone atmosphere. This change in behaviour obviously resulted in higher mating rates, which led to higher border infestation.
A several years lasting project focused on mating disruption of the grape berry moth *Lobesia botrana* and the European grapevine moth *Eupoecilia ambiguella* and its side-effects on beneficial organisms in viticulture. Investigations were conducted in 4 different vineyards in the Palatinate, Germany. Simultaneously, side-effects of mating disruption on the abundance of occasional pests were investigated. Sampling of relevant arthropods was accomplished by the suction and knocking techniques as well as the vibration and bark analysing method.
Within the vine foliage and the vine trunk, the order of Araneida and Hymenoptera were predominant among the appearing arthropodes. Within the foliage of vines, which had been treated with pheromones, higher abundances of insects and a larger variety of insect species could be found in contrast to insecticide-treated vineyards. Since abundances of predators and grape moths were rather low in general, a direct proof of a biological regulation of grape moth densities could not be furnished. The Araneida species *Salticus scenicus* and *Marpissa muscosa* were predominant at the vine trunks. A regulation of the immigration of larvae of insect pests into the vine trunk area seems possible. Regulating influences of parasitoids on populations of grape insect pests could be proven. Up to 43% of pupae of grape moths were parasitized. The majority of the parasitoids attacking pupae of insect pests belonged to Ichneumonidae of the species *Itoplectis alternans*. To lower degrees, Braconidae and Chalcidoidae were involved as well.
A specific monitoring of other insect pests not being affected by the grape moth pheromones was conducted by choosing the species *Sparganothis pilleriana*. The rate of parasitized pupae equalled 28% (1917 examined pupae). Additionally, up to 25% of pupae of *S. pilleriana* had been prey of predators. Laboratory trials gave evidence, that the traces were similar to those caused by *Forficula auricularia*. This beneficial species often searches for the leaves, which are being wrapped by the larvae of *S. pilleriana*. Therefore, *F. auricularia* is considered the most important predator of *S. pilleriana*.

Effects of mating disruption for the Grape moth on other pest and beneficial populations in Bordeaux vineyards

J.P. STÖCKEL, P. LECHARPENTIER, A. FOS and L. DELBAC

INRA, Unité de Recherches de Zoologie, B.P. 81,
33883 Villenave d'Ornon cedex, France
Fax : (33) 56 84 32 76
E-mail : stockel@bordeaux.inra.fr

Field trials to control the grape moth using the mating disruption technique were carried out in Sauternes region. In this study, each year, since 1989, we equiped a 15 ha vineyard with BASF pheromone dispensers. A progressive decrease in insecticide and acaricide use, since 1990, necessitated a careful monitoring of populations of the green leafhopper *Empoasca vitis*, of the red mite, *Panonychus ulmi* and of its principal predator *Typhlodromus pyri*.
Sampling techniques were adapted for each species. Observations were carried out by counting insect populations directly on the leaves in the vineyard using portable binocular magnifying glasses. In addition, leaves sampled, taken to the laboratory and the insects extracted by brushing, Berlése funnels, or soaking-washing-filtering. Initially we intended to treat for red mites (benzoximate) and green leafhopper (vamidothioin), in the situations where the recommended economic thresholds (red mites : 70% in spring, 30% in summer of leaves infested; green leafhopper: 100 larvae in first generation and 50 larvae in the second generation per 100 leaves) were surpassed. However, during the field trials we temporarily permitted higher populations than recommended by the economic thresholds.
In vineyards that use traditional chemical control programs, the red mite and the green leafhopper are significant pests requiring several sprays per year. In this vineyard in which the grape moth was controlled for 7 consecutive years using mating disruption, the red mite and the green leafhopper are no longer serious pests.
We have 4 possible explanations why the red mite is controlled by the *T. pyri* in vineyards using mating disruption : 1. the existence of native populations of *T. pyri* since 1990, 2. the presence of a favorable environment: woods, hedges, flowering plants, 3. the application of pest management programs including the progessive reduction of insecticides and acaricides and the acceptance of higher economic thresolds, 4. the use of fungicides non-toxic to *T. pyri*.
The reduction of green leafhopper damage could be due to the presence of higher populations of predators such as *Chrysopa carnea*, *Anagrus atomus*... because of the progressive reduction of insecticides. In addition, perhaps the

original economic thresolds were too high to predict damage and must be re-examined to determine if they can be lowered.

This study clearly demonstrates that mating disruption of the grape moth has additional benificial effects for the control of secondary pests such as the red mite and the green leafhopper, and thus should be used to as new tool in vineyards.

Adoption of mating disruption for controlling the Grape berry moth, *Endopiza viteana* (Clemens) (Lep., Tortricidae) in Ontario, Canada

R.M. TRIMBLE

Pest Management Research Centre,
Vineland Agriculture and Agri-Food Canada, Vineland Station,
P.O. Box 6000, Ontario, L0R 2E0 Canada
Fax : (905) 562 4335
E-mail : TRIMBLE@ONRSVI.AGR.CA

The grape berry moth, *Endopiza viteana* (Clemens), is the most important insect pest of grapes in Eastern North America. In the Niagara peninsula of Ontario, *E. viteana* has three generations of actively feeding larvae per year. Traditionally, an average of three annual applications of organophosphorous insecticide have been used to control *E. viteana*. In 1992, a sex pheromone-based mating disruption product was registered for use in Canada for controlling this pest. *E. viteana* is an ideal candidate for control using mating disruption because the technique has proven highly effective and the elimination of insecticide sprays from Niagara vineyards has not resulted in damaging outbreaks of secondary pests. However, the technique is being used on just 4 % of the grape production area. Possible reasons for the low rate of adoption are cost, the method of application and the method used to promote the product. A pheromone-based programme costs from 5-36 % more than an insecticide-based programme and it is occasionally necessary to augment a pheromone-based programme using insecticide. Pheromone dispensers must be applied by hand and pheromone must be used prophylactically, whereas insecticide can be used curatively and in combination with other pesticides. Perhaps most importantly, mating disruption has been promoted as an environmentally friendly alternative to the use of insecticide for controlling *E. viteana* and grape growers, in general, have not regarded this as a benefit to their grape growing business. The cost of using mating disruption might be reduced if it could be used curatively and if it could be applied using conventional pesticide application technology. If mating disruption was introduced as an insecticide resistance management tool for use in a sustainable, cost- effective pest management programme, the use of pheromone for *E. viteana* control would almost certainly increase.

Mating disruption on Pink Corn Borer, *Sesamia nonagrioides* Lef. (Lepidoptera : Noctuidae)

B. FREROT[1], P. BERNARD[2], L. MADRENNES[3], de SCHEPPER[4], F. MATHIEU[1] and M. GUILLON[4]

1 : INRA, Unité de Phytopharmacie et des Médiateurs chimiques, Route de StCyr, 78026 Versailles Cedex, France
Fax : (33) 30 83 31 49 E-mail : frerot@versailles.inra.fr
2 : Service de la Protection des Végétaux, Chemin de la Jasso, 11000 Carcassonne, France
3 : ESAP, 72 voie du Toec, 31076 Toulouse Cedex, France
4 : CALLIOPE-NPP, Route d'Artix, B.P.80, 64150 Noguères, France

For the last three years, field trials have been carried out to monitor populations of *Sesamia nonagrioides*, a severe pest on maize crops all around the Mediterranean area. New dispensers developed by NPP and Elf Atochem, avoiding manual application, sprayable by helicopter or by agricultural machines were used. Pheromone blend loaded in dispensers was a mixture of 4 components according to Mazomenos (1988) sex pheromone identification: Z(11) hexadecenyl acetate (Z11-16:Ac) 69%, Z(11) hexadecen-1-ol (Z11-16:OH) 8%,, dodecanyl acetate (12:Ac) 15%, Z(11) hexadecenal (Z11-16:Al) 8%. In 1995, mating disruption was tested on a 35 ha corn field which was divided in four successive plots. Four formulations of the pheromone were tested.

Two liquid formulations using a new original liquid pheromone developed by NPP were tested. The 11.30 ha plot was sprayed with the complete liquid pheromone (MAZOMENOS, 1988). The 8 ha plot was sprayed with an incomplete liquid blend free of Z11-16:OH and Z11-16:Al. Pheromone rate of application was 100 g/ha for the first spray (25/7/95) and 50 g/ha 15 days later (9/8/95).

Two solid formulations using an adsorbant dispenser developped by Elf Atochem were tested. Pheromone quantity was 100 g/ha. Only one spread was made (25/7/95). The 8 ha and 6.70 ha plots were treated with complete pheromone but containing different quantities of adjuvants. Emission characteristics of the four sex pheromone components as well as the release rate throughout the duration of the field trial were determined by headspace analyses.

The efficiency of different formulations was compared with a 25 ha insecticide sprayed field and with two other untreated fields of 9 ha and 5 ha.

Field trapping with virgin females was used to follow the evolution of *S. nonagrioides* populations, to set up the sprays and to assess the efficiency of mating disruption. Pheromones were applied when first males were caught. At the end of the first generation (11/7/95), *S. nonagrioides* damage were evaluated by counting stem perforations on 1.500 plants on each field. At the end of the second generation (25-27/9/95), corn plants were dissected to check whether mating disruption had occurred, to distinguish between *S. nonagrioides* and *Ostrinia nubilalis* Hbn damage and to analyse larvae instars. We controlled damage on 500 plants for each mating disruption plot, for the three other insecticide sprayed and untreated fields.
First generation damage varied from one field to another. Damage rates within the mating disruption field, the insecticide sprayed field and unsprayed fields were respectively of 2.6%, 6.3%, 1.3%, 4.1%.
Second generation damage rates were also unequal. Complete liquid pheromone successfully prevented *S. nonagrioides* from mating : on the one hand, the lowest infestation rate (1.8%) was recorded ; on the other hand, after spraying complete liquid pheromone, we did not catch any moths from the 25/7 to the 9/8, whereas some males were caught with the same sexual trap on the insecticide sprayed field. There was no significant difference between the incomplete liquid pheromone, the two solid formulations, the insecticide and one of the two untreated fields.
Larvae instar observations showed that no moth mated within the plot sprayed with the complete liquid pheromone, whereas other formulations did not enable mating disruption during the same period.
Complete liquid pheromone efficiency was better than the incomplete liquid pheromone and better than the two microencapsulated formulations. Mating disruption results were also better than the insecticide sprayed. All these results were confirmed not only by male catches but also by larvae instar observations.
Therefore, this new technology opens up in the practice of mating disruption in crop protection, which has, until now, been limited primarily to specialized productions such as orchards, vineyards.

Mating disruption of *Zeuzera pyrina* (Lep.: Cossidae) in apple orchards

H. AUDEMARD[1], B. SAUPHANOR[1] and E. ARMAND[2]

1 : INRA, Station de Zoologie et d'Apidologie, Site Agroparc, 84914 Avignon cedex 9, France
Fax : (33) 90 31 62 70
2 : BASF France, 140 rue J. Guesde, 92303 Levallois Perret, France

The development of biological control of the Codling moth in apple and pear ochards will soon be imposed by the resistance of this insect to various pesticides. Several secondary pests, which were previously controlled by the chemicals applied against this key pest, will need specific control too. Mating disruption trials against the Leopard moth, *Zeuzera pyrina* L., were therefore conducted in three orchards of the Rhône Valley near Avignon. The dispensers (Rak 8, BASF) were loaded with the main component of the pheromone, (E,Z)-2, 13-octadecadien-1-ol-acetate and placed at different densities in the orchards at the end of may 1991 and 1992. The release of the pheromone ranged from 53 to 131 mg/ha during the 18 weeks testing period, according to the test, and was closely correlated with temperatures. The effectiveness of mating disruption was evaluated by recording the primary infestation of young larvae on shoots in summer and the secondary infestation in autumn, on a 250 trees sample for each orchard. The rate of infested trees decreased from 100 % to 17,8% in orchard A, and from 28% to 6,7% in orchard C1 after the first experiment season, while the dispensers released 131 g/ha pheromone. The population was maintained at the same level during the following season, respectively with 80 and 53 g/ha pheromone released in these two orchards. A single trial in 1992 in orchard C2 allowed the control of an higher infestation than in previous orchards with 106 g/ha pheromone. It appears that moderate populations of this species can be controlled by a 15 mg/ha/hour pheromone release. These first trials allowed the definition of experimental conditions and sampling methods to evaluate the mating disruption method against *Z. pyrina*.

Commercial exploitation of mating disruption technology : difficulties encountered and keys to success

E. CASAGRANDE and O.T. JONES

Agrisense BCS Ltd, Treforest Industrial Estate, Pontypridd,
Mid Glamorgan, CF37 5SU, United Kingdom
Fax : 44 1 443 84 115

The use of sex pheromones for mating disruption of Lepidopteran pests of agriculture has, over the last decade, become an important component of Integrated Pest Management packages in a number of crops. Our company has been involved in the commercialisation of at least four major pest management products based on this technology, namely *Pectinophora gossypiella* in cotton, *Chilo suppressalis* in rice, *Rhyacionia buoliana* in forestry and *Keiferia lycopersicella* in tomatoes. A number of other product development projects have been started with other species but have been discontinued for a range of reasons. During the last decade we have observed several common themes which have emerged as problems during the product development and commercialisation process for mating disruption products. These have included factors such as the formulations used, agronomic constraints, the pest's biology and socio-economic factors. This paper examines the problems encountered and suggests keys to how such problems can be overcome, together with indicators of how mating disruption technology may evolve in the future.

Mating disruption in apple orchards

Franco RAMA

Isagro Ricerca s.r.l., Istituto Donegani, Via Fauser 4, 28100 Novara, Italy
Fax : 0039 321 447887
E-mail : ramafra@snefru.comm2000.it

Since mating disruption became a valuable alternative to chemical treatments, growers and agricultural organisations started asking for reliable products in order to implement the method. Industries were hence prompted to supply enough material both to further investigate the agronomic aspects of mating disruption and to start a large scale application of the method.
The work done in the past years allowed to identify the limits of the mating disruption technique, mainly as consequences of orchard organisation and density of starting pest population. Those limits were considered as linked to the recipient of the method. Once the local requirements for a successful application are satisfied, other factors play an important role in the practice of the mating disruption. Such new variables are more bound to the kind of device used to disrupt the matings and must be identified in order to implement the technique at a grower level.
Since 1987 trials have been carried out to evaluate the mating disruption as a control method against Codling moth (*Cydia pomonella* L.) and the leafrollers in apple orchards, using Isagro's dispensers ECOPOM and ECOPOM COMBI. Focusing on these observations, we could modify and optimise several factors, such as the quantity of attractant/ha, the number of dispensers/ha, the amount of attractant/dispenser and the number of applications per season. The persistence of pheromone release in the field and the reproducibility of dispenserís manufacturing process became the most important elements to be defined, and we wish to report here the results of Isagroís efforts toward the development of an effective device for the mating disruption in apple orchards.

Confusion amongst Codling moth fellows continues : a commercial perspective on the implementation of codling moth mating disruption in North America

D.R. THOMSON

Pacific Biocontrol Corporation, 400 East Evergreen Blvd.,
Suite 205, Vancouver, WA. 98660, USA
E-mail : pacbio@mail.pacifier.com

Mating disruption technology is increasingly being used for the control of Codling moth in pome fruit production areas around the world. Some of the countries where Codling moth mating technology is used commercially include the United States, Canada, Argentina, Australia, France, Italy and South Africa. In 1991, Isomate C Plus (Pacific Biocontrol Corp., Vancouver, Washington) became the first commercial formulation of Codling moth pheromone to be registered in the United States. The total pome fruit acreage treated with Isomate C Plus has increased from approximately 1,200 hectares in 1991 to approximately 7,300 hectares in 1995. In 1996, Pacific Biocontrol will treat between 9,500 and 10,000 hectares of pome fruit with Isomate C+.

The successful commercialization of mating disruption technology will depend in large part on the development and implementation of a pheromone-based IPM systems approach. The objective of a pheromone-based IPM program is to effectively manage key and secondary pests in an economically, ecologically and environmentally acceptable manner.

In a pheromone-based IPM system, mating disruption is the major tactic used to control the key pest(s). The subsequent reduction or elimination of insecticides for control of the key pest(s) will promote crop or orchard environments that will support higher populations of natural enemies and thus enhance the biological control of both key and secondary pests. The development of monitoring and sampling techniques in conjunction with economic thresholds are essential in order to accurately assess the biological relationships between key and secondary insects and their natural enemies and to implement supplementary controls if required. Pheromone-based IPM should be presented to growers as a long term approach and commitment to pest management. Growers should be encouraged to define yearly objectives and then identify the strategies and tactics needed to achieve those objectives.

Successful employment of pheromones in apple : exemplary results from Europe

Ulrich NEUMANN

BASF Aktiengesellschaft, Crop Protection Division,
Research and Development, Limburgerhof, Germany
Fax : +49 6236 67192

There is much discussion about the suitability of the mating disruption technique against Codling moth and Summer fruit *Tortrix* in apples.
Based on exemplary results gained in different years and different countries, the effect of the mating disruption technique and its dependency on various influencing factors will be demonstrated.

SYMPOSIUM #7

OPENING THE DOOR FOR AUGMENTATION OF PARASITIC AND PREDACEOUS ARTHROPODS IN AGRICULTURAL SYSTEMS THROUGH ARTIFICIAL DIETS AND AUTOMATED *IN VITRO* REARING SYSTEMS

Organizers :

Donald A. NORDLUND
USDA, ARS, biological Control of Pests Research Unit,
2413 East Highway 83, Weslaco, TX 78596, USA
Fax : 210 969 4888
E-mail : donn@pop.tamu.edu

and

Simon GRENIER
INRA, laboratoire de Biologie appliquée, INSA 406,
20 av. A. Einstein, 69621 Villeurbanne cedex, France
Fax : (33) 72 43 85 11
E-mail : sgrenier@jouy.inra.fr

Introduction : artificial diets and automation prerequisites for the general adoption of augmentation techniques in biological control in extensive agricultural systems

Norman C. LEPPLA

USDA, APHIS, Office of the Administrator,
National Biological Control Institute, 4700 River Road, Unit 5,
Riverdale, MD 20737-1229, USA
Fax : (301) 734-7823 E-mail: nleppla@aphis.usda.gov

Prerequisites for the general adoption of augmentation biological control in extensive agricultural systems are cost competitive with other methods, conclusive demonstration of efficacy, incorporation into integrated pest management (IPM), support for technology development and implementation, and targeted research. The relative cost of augmentation biological control includes not only the initial costs of alternatives but also the non-target effects, occurrence of pest resistance, emergence of secondary pests, disruption of multiple cropping systems, long-term versus short-term control, and so forth. Efficacious techniques can only be developed for appropriate target pests and environments, and more models of success are needed to gain wide acceptance and use of augmentation biological control. IPM based on biological control requires the use of complementary pesticides, other preventative and curative techniques, conservation of natural enemies, and areawide pest management in addition to traditional IPM practices. Technology development could be accelerated by establishing more government/industry partnerships, building municipal and grower cooperatives, and providing greater assistance with permits and registration of commercial products. Research is needed on artificial diets for natural enemies, mass production procedures, strain development using both impact and economic evaluation. Augmentation biological will advance at a pace dictated by investment in these prerequisites to its development and general adoption.

Development of an automated *in vitro* mass propagation system for *Trichogramma* spp. : progress and opportunities

Donald A. NORDLUND, Z. WU and S.M. GREENBERG

USDA, ARS, Subtropical agricultural Laboratory,
biological Control of Pests Research Unit, 2413 East Highway 83,
Weslaco, TX 78596, USA
Fax : (210) 969-4888
E-mail: donn@pop.tamu.edu

Egg parasitoids of the genus *Trichogramma* are used for augmentative biological control of numerous important insect pests. With the exception of some *in vitro* rearing in China, *Trichogramma* are propagated *in vivo* on factitious hosts. *In vivo* propagation systems tend to result in high production costs and limited production capacity. There has been considerable progress in the development of artificial diets for *Trichogramma* spp. We recently reared *Trichogramma minutum* for ten generations on a meridic artificial diet and produced insects of relatively high quality, as compared to insects produced in *Helicoverpa zea* eggs. We have also made progress in development of a system for preparing streached plastic artificial eggs for the collection of *Trichogramma* eggs for use in an automated *in vitro* mass propagation system. With the experience gained during laboratory scale rearing on artificial diet, we developed, conceptually, an automated *in vitro* mass propagation system based on the use of form-fill-seal equipment. This equipment will form larval rearing/adult release cells, fill those cells with a mixture of *Trichogramma* eggs and artificial diet, and seal the cells with a gas permeable material. These cells will be held under appropriate environmental conditions for larval development and eventual shipment to end users for placement in the field. Transfer of *in vitro* mass propagation technology will require the development of a complete production system, not just individual components. Lack of sufficient engineering resources is a major impediment to the development of such a system.

The use of *Trichogramma* spp., reared *in vitro* in China

Li-Ying LI

Guangdong Entomological Institute, Guangzhou 510260 , China
Fax : 86 20 84191709

There are 3 models of manufactured machine in China for automatically *in vitro* rearing of *Trichogramma* spp. The designers of these machines are : the Guangdong Entomological Institute (GEI) ; College of Life Sciences of Wuhan University (CLSWU) ; and the Plant Protection Institute of the Guangdong Agricultural Academy (PPIGAA). The machine of GEI could produce 840 artificial "egg-cards" with 2,520 "host-eggs" and 1000,000 wasps every hour. It is now in improvement. The machine of CLSWU can produce 960 artificial "egg-cards" with 96,000 "host-eggs" and 4320,000 wasps every hour. The machine of PPIGAA can produce 625 artificial "egg-cards" with 62,500 "host-eggs" and 3120,000 wasps every hour. *Trichogramma dendrolimi* and *T. confusum* have been reared *in vitro* for more than 50 generations continuously by the CLSWU and PPIGAA without degeneration of the viability. During the past 10 years, *T. dendrolimi* and *T. confusum*, reared *in vitro*, have been used for controlling sugarcane borers, asian corn borer (*Ostrinia furnacalis*), pine caterpillar (*Dendrolimus punctata*), cotton bollworm (*Helicoverpa armigera*) and soybean pod borer (*Leguminivora glycinivorella*) in a larger scale (in 1995 there was a total area of 2000 ha) with satisfactory effectiveness as *Trichogramma* spp. reared with the eggs of *Antheraea pernyi*.

Artificial rearing of *Trichogramma* : from storage of medium and egg laying stimulation to emergence of normal adults

Simon GRENIER

INRA, laboratoire de Biologie appliquée, INSA 406,
20 av. A. Einstein, 69621 Villeurbanne cedex, France
Fax : (33) 72 43 85 11
E-mail : sgrenier@jouy.inra.fr

To simplify the production of biological control agents such as parasitoids, studies have been conducted to develop *in vitro* culture. Different difficulties have to be overcome for rearing *Trichogramma* in artificial conditions and especially for a mass production. The composition of the artificial media in the different nutriments, can be defined mainly from food analyses (host egg). Egg parasitoids develop in closed systems (egg yolk), consequently for *in vitro* rearing they will need very rich and concentrated diet. Media used can be divided into two main categories according to the presence or the absence of insect components. Hemolymph is often used but could be difficult to obtain. Its replacement by non insect material is advisable but not easy. According to the disponibility of some components or to the possibility to produce high quantity of medium at the same time, it will be useful to store the media. Lyophilization is a good process for a long time storage of artificial media (about one year) without any reduction of the performances of the medium. For normal development, and high yields, *Trichogramma* need a limited quantity of food. It is important to optimize the number of eggs laid by *Trichogramma* females into artificial host eggs. Egg laying stimulation allows to increase this number, often reduced *in vitro*. Three groups of parameters are implicated: the components present on the artificial egg shell, the characteristics of the artificial egg shell, and the composition of the medium. It is necessary to obtain *Trichogramma* that can be considered as normal (compared with parasitoids growing in natural host eggs), as far as their morphological aspects, biochemical content, and behavior are concerned. Total amino acid content of pupae varies according to the composition of the media offered.

Biological control of the Boll Weevil in Cotton by mass propagation and augmentative releases of the Wasp parasite, *Catolaccus grandis*

Edgar G. KING, R. J. COLEMAN, J. A. MORALES-RAMOS, G. M. ROJAS, K. R. SUMMY, Leeda WOOD and Lloyd WENDELL

USDA, ARS, Subtropical agricultural Research Laboratory, biological Control of Pests Research Unit
Fax : (210) 565-9584 E-mail : king@pop.tamu.edu
and USDA, APHIS, Mission Biological Control Laboratory, 2413 East Highway 83, Weslaco, Texas 78596, USA

Release of the wasp parasite, *Catolaccus grandis*, at rates from 350 to 1000 female parasites per week for 6-8 weeks during the boll weevil (*Anthonomus grandis*) F_1 and F_2 larval generations adequately suppressed weevil populations during 1992-1995 in experimental and commercial cotton fields. 1994 parasite-release fields yielded significantly more cotton than IPM-treated fields, and weevil mortality in 1995 parasite-release fields (organically-grown) was 100% for six consecutive weeks ; adult weevil numbers based on captures from traps around the field periphery were significantly less around organically-grown cotton than around the insecticide-treated control (eradication) fields for 10 weeks. During 1995, over 100,000 parasites were produced per week using artificial diet-reared weevils encapsulated in Parafilm®. A chemically-defined artificial diet, devoid of insect components, for the parasite was developed; ARS patent disclosure requests on the diet and *in vitro*-propagation system are pending. *In vitro*-propagated parasites found as many hosts in the field as *in vivo*-propagated parasites. Systems for *in vivo* and *in vitro*-mass propagation were designed. Parasite mass propagation and augmentation technologies are being transferred via a Cooperative Research and Development Agreement to the private sector. *In vitro* propagation reduces parasite-production costs by more than one-half, reduces production complexity, and provides a proprietary product for licensing to the private sector. Aseptic production conditions result in uniformly high quality parasites. Recent outbreaks of the tobacco budworm, *Heliothis virescens*, and beet armyworm, *Spodoptera exigua*, and other secondary pests coincidental with boll weevil eradication efforts jeopardize expansion of Boll weevil eradication in the United States. Augmentations of *C. grandis* could be used *in lieu* of chemicals during early season to spare natural enemies in eradication zones at higher risk to secondary pest outbreaks, as well as reduce insecticide usage for boll weevil suppression in environmentally-sensitive areas and suppress weevils in organically-grown cotton.

Technology transfer potential of artificial media for entomophages

Allen C. COHEN

USDA, ARS, Western Cotton Research Laboratory,
4135 E. Broadway, Phoenix, Arizona 85040, USA
Fax : (602) 379-3524
E-mail : acohen@ix.netcom.com

After 50 years of nutritional research on predators and parasites, there is finally a serious potential for commercially feasible artificial media based (AMB) production of entomophages. The barriers and requirements are discussed for commercial success in AMB entomophage production.
What is required for commercialization is a demonstration that AMB predators or parasites can be produced at lower costs than ones produced on natural hosts. Issues of quality requirements and cost-benefit tradeoffs are discussed in terms of profitability/feasibility matrices.

Lessons learned in commercializing biological control applicable to use with parasitic and predaceous arthropods

Richard M. PARRY, Jr.

USDA, ARS, Office of Technology Transfer, Rm. 358A,
Whitten Federal Building Washington, D.C. 20250-0300, USA
Fax : 202-720-7549
E-mail : parryr@ars.usda.gov

The past century has seen significant investment in basic research on biological processes for ecosystem management. This investment is now yielding new methods to manage pests in agricultural and other environments where biological control is needed. Our challenge is to develop these discoveries into commercial practices in the private sector business environment that looks for secure, risk-free investment opportunities. Augmentative biological control is considered an unproven, high-risk commercial venture despite its remarkable promise. Our challenge is to sufficiently mitigate the technical and regulatory hurdles so that the promise becomes a viable industry. Every new technology faces a huge hurdle crossing the barrier between laboratory success and commercial application. There is no single formula applicable to transfer of technologies, but there are certain factors that can limit the frustrations and time loss in the development process. First, identify the customers/market of the technology and demonstrate its usefulness in real world production systems. Second, form a partnership with a business/private sector cooperator interested and capable of bringing the technology to the market. This partnership may be a small business with limited resources and little technical capability; this may require significant involvement by the scientist to establish a productive partnership. Third, understand the business conditions which the company must operate in for survival; this information will also assist in the identification of the best partner. Fourth, seek opportunities to attract investment in the technology through intellectual property protection or other tools that limit the risk of loss of markets to competitors before development costs are recovered. Fifth, make sure that there are no regulatory barriers that will impede the commercial development process. Specific examples will be described from experiences in transferring biological control technologies that are applicable for use in transferring augmentative release of parasitic and predaceous arthropods.

Importance of genetic variability of entomophagous insects used for pest management

F. POMPANON and M. BOULETREAU

Univ. Lyon I, "Biométrie, Génétique et Biologie des Populations",
UMR CNRS 5558, 43, Bd du 11 Novembre 1918,
69622 Villeurbanne cedex, France
Fax : (33) 78 89 19 41
E-mail : boulet@biomserv.univ-Lyon1.Fr

(CEE grant AIR3-CT9-1433 :
"Genetic and epigenetic improvement of entomophagous insects used in biological control")

Entomophagous insects and especially Hymenopteran parasitoids are widely used as biocontrol agents, and the success of the method strongly depends on their quality. The genetic components of this quality may differ according to the release strategy (inundative or inoculative), but in both situations they must be considered at the individual level (search for the best suited genotypes) and at the population level (search for genetic variation and heterozigosity). Thus any biocontrol program is faced with three basical genetic challenges : how to organize field sampling for catching suited genotypes ? How to keep the genetic background of the strains during mass rearing ? How to organize releases ? Another question is : how to improve the genetic quality of biocontrol agents ?
We give first the theoretical answers which can be given to these questions. Then, taking as examples the fecundity and locomotor activity of Hymenopteran parasitoids (Trichogrammatidae and Eucoilidae), we explain the methods allowing to evaluate the genetical and non-genetical components of the phenotypical variability, and we show the range of genetic variation which can be expected both within- and between wasp populations. Finally we discuss the problems of genetic and non-genetic improvement of biocontrol agents.

Biological control effectiveness strategy of QTL identification in *Trichogramma brassicae* (Hymenoptera, Chalcidoidea)

V. LAURENT , F. VANLERBERGHE-MASUTTI
and E. WAJNBERG

INRA, laboratoire de Biologie des Invertébrés,
Unité de Biologie des Populations, 37 bd du Cap,
B.P. 2078, 06606 Antibes, France
Fax : (33) 93 67 89 55
E-mail : vlaurent@antibes.inra.fr

The existence of genetic variation in quantitative biological traits involved in the reproductive strategy has been demonstrated in the parasitoid wasp *Trichogramma brassicae.* Therefore, it provides the basis of selective breeding for improving the efficiency of *Trichogramma* to control pests in biological control programs.

In this respect, genetic maps represent a powerful tool for accelerating breeding through marker assisted selection. Indeed, identification of the locus controlling the traits involved in biological control efficiency could help breeding by making the screening of such a minute insect easier.

A RAPD genetic map is now under construction for this haplo-diploid species. It will provide evenly spaced markers covering the whole genome for Quantitative Traits Loci (QTL) identification.

As both genotyping and phenotyping is not possible on the same *Trichogramma* individual, a particular strategy of QTL identification has to be used.

In order to locate QTL for fecundity and longevity, two crosses involving parental isofemale lines differing for these two traits were selected for the construction of the QTL mapping population. Two F2 populations of 200 haploid males each will be obtained from virgin F1 females.

F2 males will be scored for molecular markers after being individually crossed with five females from the parental lines. Crossing each F2 male with various females instead of only one will allow to determine the overall value of the male in crossing by analyses of the F3 female progenies performances.

Female F3 progenies will be scored for fecundity and longevity and linkage of these traits with the markers of their father will be checked. In order to limit the number of genotypes to establish, linkage will be checked only for the F2 males whose progeny performances diverge significantly from the mean.

(This work is supported by CEE-AIR3-CT94-1433)

SYMPOSIUM #8

EVALUATION OF THE EFFICIENCY OF BIOCONTROL

Organizer :

Dan GERLING

Tel Aviv University, the George S. Wise Faculty of Life Sciences,
Department of Zoology, Ramat Aviv 69978, Israel
Fax : 972 3 6407830
E-mail : DANGR@post.TAU.ac.IL

Evaluation of biological control : the scope of the problem

Dan GERLING

Tel Aviv University, The George S. Wise faculty of Life Sciences,
Department of Zoology, Ramat Aviv 69978, Israel
Fax : 972 3 6407830
E-mail : DANGR@post.TAU.ac.IL

Biological control is a method used for controlling agricultural pests. Therefore, it is imperative that successes and failures in its implementation be monitored and their causes analyzed. Only thus can future biocontrol projects be scientifically planned for optimal results.

As compared with other pest control methods, bio-control is considered cheap, providing a long-lasting or permanent solution and environmentally safe. Since bio-control attempts to solve agricultural problems, its success should be measured in economic and financial terms. However, because it also has ecological parameters, direct economic assessments are insufficient and long-term pest management and environmental considerations must also be addressed.

The difficulty of devising a proper way to evaluate bio-control projects for both, the short-term economic, and the long term social and environmental benefits is compounded by the fact that many bio-control projects are undertaken without proper attention to later assessment of the results, assuming that any pest population reduction would be beneficial. Projects may also lack an experimental design that would allow to assess the diverse parameters of the project. Finally, since the importance of evaluation is not properly recognized, financial support is often given to the implementation part of bio-control projects and withheld from the evaluation programs.

The purpose of this symposium is to illuminate the different aspects involved in the assessment of bio-control, to discuss the available techniques, and to point to some of the more critical directions of research and implementation destined to improve the present situation.

Economic evaluation of biological control

Karen KLONSKY

Dept. of Agricultural and Resource Economics,
University of California, Davis, CA 95616, USA
Fax : 916 752 3563
E-mail : klonsky@gordy.ucdavis.edu

A classical biological control program remains in effect into perpetuity and encompasses a significant geographic region delineated only by climatic and geographic factors. Exclusion from benefits is impossible. It follows that firms developing the technology cannot accrue all of the benefits and that benefits accrue to people other than direct users. Related research is characterized as long term and very risky. These characteristics of biological control explain the extensive participation by the public sector in biological control research. They also suggest that the scope of an economic analysis of a biological control program will necessarily include society as a whole. The challenges in cost - benefit analysis are first to identify the significant impacts of the program and secondly to assign values to these components in a meaningful way. A listing of factors to be considered follows. Market impacts of biological control include changes in prices of agricultural commodities and substitute commodities. Other market impacts include changes in input prices such as substitute pesticides and land.

Changes in input and commodity prices mean gains for some members of society and losses for others. The redistribution of wealth changes over time with population growth, income distribution, and *per capita* income. Nonmarket impacts are inevitable because successful pest control always has ecological consequences. The use of biological controls impacts water and air quality, farm worker and food safety when substituted for polluting pesticides.

Typically only costs directly attributable to the biological control program are included. Overhead costs such as containment facilities, rearing facilities and laboratories are difficult to allocate to individual projects. Further, the contribution of technology developed in other places is usually ignored.

Economic analysis of biological control serves to justify public and private research expenditures. As such, a complete accounting of benefits and costs, winners and losers is paramount.

Techniques for evaluating bio-control efficacy of parasitoids and predators against Insect pests

Dan GONZALEZ

University of California, Department of Entomology,
Riverside CA 92502, USA
Fax : 909787 3681
E-mail : marcella@ucrac1.ucr.edu

Steps in developing a program for evaluating biological control efficacy of insect parasitoids and predators against insect pests will be discussed. The principal components discussed will include :
1) Defining the terminology used in this field and the parameters that define programs ;
2) outlining sampling procedures to include all relevant species involved, appropriate for various plant substrates ;
3) reviewing and placing in perspective so-called" desirable biological attributes" for estimating the potential efficacy of natural enemies ; and
4) reviewing and criticizing evaluation techniques used in assessing natural enemies in conservation, introduction, and augmentation programs and also in naturally occurring (native) systems.
The discussion will be focused on open-field programs and will be based in part on a survey of information from literature available, and on the author's personal experience. Examples and literature references will be provided as a basis for discussion.

Efficacy evaluation of microbes used for biological control of pests

Raymond I. CARRUTHERS

USDA, ARS, National Program Staff, BARC-West,
Bldg. 005, Beltsville, MD 20705 USA
Fax : + 301 504 5467
E-mail : RIC@ars.usda.gov

Microbial agents and their products are used to control a variety of different pests including insects, nematodes and weeds. Methods of use include conservation of naturally-occurring microbes, augmentation of microbial populations, and introduction and establishment of exotic species. Naturally suppressive soils and parts of the phylloplane have also been found to contain many microbes that can act as antagonists, competitors or parasites to a variety of plant pathogens. Isolation, and identification of microbes and their development as biological control agents is developing quickly, particularly in plant pathology and weed science. Recently, the United States Environmental Protection Agency has streamlined its regulatory procedures to facilitate increased use of microbial agents for pest control as replacements for chemical pesticides. These new registrations may also require increased monitoring to assess potential environmental impacts. These requirements necessitate increased research and technique development in sampling and evaluating the effects of microbes on both target and non-target species.

Assessment of biological control potential and environmental fate of these microbes has followed many different approaches depending upon the specific mode of action of the agent under study and the target host attacked. A range of assessment techniques will be presented including laboratory bioassay and screening procedures, host range evaluations, quarantine testing, field releases, isolate specific detection methods, and impact studies. Physiological *vs.* ecological host range will be discussed, as will methods of field sampling, persistence evaluation of microbial populations, and overall impact assessment. In situations where classical biological control has been conducted using exotic microbes, disease spread and impact on non-target species will be discussed.

Use of microbes for biological control is expanding and will be used against many pests now and in the future. Assessment of their use and impact will also be of expanding interest that will require scientific development both to improve their utilization, and to document their effectiveness and environmental safety.

Use of models to evaluate control efficiency

N.J. MILLS

Department of Biological Control, University of California
at Berkeley, 1050 San Pablo Ave., Albany, CA 94706, USA
Fax : 510 642-0875
E-mail: nmills@nature.berkeley.edu

A wide range of models, from simple analytical models to complex simulation models, have been used to evaluate the effectiveness of natural enemies in biological control programmes. Models have been used most extensively in retrospective evaluations of the effectiveness of imported parasitoids in the classical biological control of insect pests. In retrospective analyses, models are used to verify that observed changes in the pattern of abundance of the pest and its imported natural enemy(ies) in the field can be accounted for by introducing estimates of the biological characteristics of the interacting organisms into a natural enemy-host population model. More recently models have begun to be used for prospective analyses of classical biological control, providing a valuable technique that can be used to predict the outcome of natural enemy introductions and to aid the selection of the most effective biological control agents prior to their introduction. Prospective modelling has also been used in the evaluation of augmentative biological control. In this case the models are used to predict the outcome of inoculative or inundative releases of parasitoids, predators and microbial agents, based upon estimates of the killing power and dispersal of the control agent. In this presentation I will use specific examples to illustrate the benefits of using models of intermediate complexity to evaluate the effectiveness of natural enemies in both classical and augmentative biological control programmes against insect pests.

Evaluation of the efficacy of biological control programs in greenhouse crops

K. BOLCKMANS

BIOBEST N.V., Ilse Velden 18, B-2260 Westerlo, Belgium
Fax : +32 (0) 14/23 18 31

Biological control in greenhouse crops has a history of more than 30 years. "New" beneficial insects and mites are implemented at regular intervals. More than 20 different beneficial insects and mites are currently commercially available and the number is still increasing.
Efficacy or success of biological control programs is determined by several factors. An overview of these factors will be given. One or more effective natural ennemies of sufficient quality have to be commercially available in sufficient quantities for each pest. For those pests for which a natural enemy has not yet been developed, a selective chemical should be available. A good and regular technical support is essential.
Biological control programs ave been developed for almost all important greenhouse vegetables (tomatoes, sweet peppers, cucumbers, eggplants, melons, beans, ...) and for several greenhouse ornamentals (poinsettias, gerberas, ...). Under certain conditions it is now possible in some greenhouse crops to control all pest without the use of chemical insecticides or acaricides.
Developing or "fine tuning" biological control programs and evaluating their efficacy is a never ending, continuous process.

Evaluation of biological control implementation in field crops

Oscar MINKENBERG

Department of Entomology and Center for Insect Science,
University of Arizona, Tucson, AZ 85721, USA
Fax : 602 621 1296
E-mail : PARASITE@CCIT.ARIZONA.EDU

Augmentation has been used only scarcely in field crops such as vegetables, wheat, corn, cotton and soybeans, although these commodities cover most of the agricultural land in the USA. An estimate of use of beneficial arthropods in 1992 indicated application on less than 3% of total acreage. Biological control is still experimental in most of these crops. Herein I will discuss the various methodologies to evaluate natural enemies for use in field systems.
An example of effective augmentative biological control in the field is the use of predatory mites, *Phytoseiulus persimilis* (Acari : Phytoseiidae) against the two-spotted spider mite, *Tetranychus urticae* (Acari : Tetranychidae), a key pest of strawberries. The total area of strawberry production in 1992 was estimated at 20,120 ha. On about 37% of this crop area, growers released beneficial agents. Oatman and McMurtry (1966) conducted the first evaluation studies, which will be described. However, it took 30 years before wide-scale implementation occurred, Recent evaluation studies will be highlighted.
The second example deals with the imported whitefly *Bemisia argentifolii* (Homoptera: Aleyrodidae), which has caused more than one billion dollars in damage to U.S. agriculture since 1991. A research program was initiated to evaluate the use of augmentative releases of the native parasitic wasp *Eretmocerus* sp. (Hymenoptera: Aphelinidae) against this devastating pest on cotton in the Imperial Valley, California. Exclusion cage studies demonstrated that inoculative releases of *Eretmocerus* were effective at a rate of *ca.* 10 parasitoids in total per plant. In the cages with high numbers of parasitoids released, average levels of parasitism reached about 61% and whitefly levels were reduced to 0.0 pupae/cm^2 leaf (Simmons and Minkenberg 1994). However, open field releases in both experimental plots and a grower's field failed to achieve similar levels of parasitism and whitefly suppression (Minkenberg *et al.* 1994).

Evaluation of biological control programs in deciduous orchards

P.J.M. MOLS[1] and L.H.M. BLOMMERS[2]

1 : Dept. Entomology, Agricultural University Wageningen, Binnenhaven7, P.O.Box 8031, 6700 EH Wageningen
Fax : + 317 484821
E-mail : Peter.Mols@MEDEW.ENTO.WAU.NL
2 : Experimental Orchard "De Schuilenburg", Research Institute for Plant Protection (IPO-DLO), Schuilen-burg 3, 4041 BK Kesteren, The Netherlands

In apple and pear orchards one has to deal with approximately 40 potential insect pests, which may occasionally reach the pest status, depending on biotic and abiotic factors. Much research has been carried out to get insight in the factors that lead to or prevent biological control of these species.
For more than 20 years IPM in apple orchards has been shaped around the biological control of the Fruit tree spider mites (*Panonychus ulmi* Koch) and rust mites by phytoseiids either by using selective pesticides against other pests or by selecting pesticide resistant predatory mites. The possibilities to control other pests by natural enemies depend on the strategy applied. More than 20 species of Tortricids including the ubiquitous Codling moth are considered regular apple pests. Natural control including predation by birds, insects and diseases tempered outbreaks and downgraded the pest status of certain leafrollers.
An overview will be presented of the pest and natural enemy complex in orchards and of the methods used to evaluate their efficacy. Special attention will be given to the use of modeling in estimation the role of natural enemies in controlling apple aphids.

Evaluating efficacy of aerially-applied biological control agents in Boreal forests

R.J. WEST

National Research Canada, Canadian Forest Service, Box 6028,
St John's Newfoundland, Canada A1C5X8
Fax : 01 709 772 6019
E-mail : rwest@nefc.forestry.ca

Outbreaks of defoliators require control by aerial application. Products must be delivered at a constant rate and in low volumes to maximize flying times within narrow "windows" defined by the susceptible stage of the insect, and periods of favourable weather. Efficacy evaluations are described and include novel assessments of the delivery and deposition of the product, target effects and persistence of the biocontrol agent, and stand vigour.

Flow-meters and geopositioning systems can provide accurate measurements of product delivery and be presented as swath patterns overlaid on maps of spray blocks. Computer programs using delivery information and weather data can predict deposit patterns on foliage.

Deposit assessments should determine : product distribution and bioactivity at the targetted site. Droplet counts on, and bioassays with, sprayed foliage are common examples. Ground units placed in canopy openings may provide a rough indication of the size, densities and bioactivity of spray droplets.

Inundatively-applied products normally need to reduce feeding or pest numbers to prevent damage during the feeding period. Feeding rates can be determined with the aid of frass trays and bioassays. Bud/shoot/branch sampling and tree beats onto drop cloths yield useful counts of larvae. Larval condition at the time of sampling reflects efficacy (eg., incidence of moribund larvae). For products that have inoculative properties, eg., viruses, it is important to determine if secondary infection has occurred.

Remote sensing techniques provide objective assessments of stand vigour before and after treatments, eg., there is excellent correlation between spectral patterns and defoliation levels. Remote sensing can be used to identify characteristics of stand structure and growth conditions which in turn can be linked to susceptibility and vulnerability to insect attack. Subjective estimates of foliage losses include aerial surveys, and whole-tree and branch assessments and provide a rapid indication of treatment efficacy.

SYMPOSIUM #9

TECHNOLOGY TRANSFER IN BIOLOGICAL CONTROL OF WEEDS

Organizers :

Lloyd KNUTSON
USDA, ARS, European biological Control Laboratory,
Parc scientifique Agropolis II, 34937 Montpellier cedex 5, France
Fax : (33) 67 94 56 20
E-mail : ebcl@cirad.fr

and

W.M. LONSDALE
CSIRO, Division of Entomology, European laboratory,
Campus international de Montferrier-Baillarguet,
34980 Montferrier-sur-Lez, France
Fax : (33) 67 59 90 40
E-mail : lonsdale@csiro.ensam.inra.fr

American and Australian exploration for natural enemies of Eurasian weeds

L. KNUTSON[1] and W.M. LONSDALE[2]

1 : USDA-ARS, European biological Control Laboratory,
Parc Scientifique Agropolis II, 34937 Montpellier Cedex 5, France
Fax : (33) 67 94 56 20 E-mail : ebcl@cirad.fr
2 : CSIRO Division of Entomology, European Laboratory,
Campus de Baillarguet, 34980 Montferrier sur Lez, France
Fax : (33) 67 59 90 40 E-mail : lonsdale@csiro.ensam.inra.fr

The USDA-ARS weed biocontrol lab was established in Rome in 1957 and insect pest biocontrol lab was established in France in 1918. The CSIRO Division of Entomology's weed biocontrol laboratory for Mediterranean weeds was established in 1967 in Montpellier, partly because this was central to the biogeographical origin of the weeds of the Mediterranean biome in eastern Australia, but also because of good transport links with the rest of the region. For the latter reason, and to create a critical mass of researchers working in biocontrol, the two USDA labs were combined in Montpellier in 1991 close to the CSIRO laboratory. USDA uses expeditions around the Eurasian zone and also has maintained substations in Rome and Thessaloniki. By contrast, CSIRO has no substations, using only expeditions to collect at places distant from Montpellier (especially to Turkey, Spain, Portugal and Morocco). As a reflection of the different weed floras and land use in the two countries, there is currently only a small degree of overlap between the species targeted by the two organisations : of 11 genera studied by USDA, and 12 by CSIRO, only two are common to the two organizations, namely *Chondrilla* and *Carduus*. Both USDA and CSIRO attempt to prioritize target weeds in a systematic way, using advice from cooperators at all levels of government, universities, and the end-users to consider scientific, economic, operational, and safety concerns, besides the likelihood of success. However, unexpected discovery of major new natural enemies, or the availability of funds, can elevate a weed's priority. Research information needed to implement an appropriate suite of natural enemies (identity, phenology, life cycle, host range, habitat, and soil and climate regime preferences) is the primary approach for both teams. In pursuing this objective, attention is focused on strategic biocontrol research questions wherever possible.

Regulatory procedures for the introduction of classical biological control agents for weed management in the United States

A.F. COFRANCESCO[1] and J.R. COULSON[2]

1 : Aquatic Ecology Branch, Environmental Laboratory,
Waterways Experiment Station, U.S. Army Corps of Engineers,
3909 Halls Ferry Road, Vicksburg, Mississippi 39180-6199, USA
Fax : 1 601 634 2398
2 : USDA, ARS, Insect Biocontrol Laboratory, biological Control
Documentation Center, Beltsville Agricultural Research Center-West,
Beltsville, Maryland 20705, USA
Fax : 1 301 504 6355 E-mail : bcdcjc@ars-grin.gov

The Animal and Plant Health Inspection Service (APHIS) of the U.S. Department of Agriculture (USDA) has the responsibility of regulating the introduction of classical biological control agents into the United States for weed management. In 1957, an advisory group, later designated the "Working Group on Biological Control of Weeds", was formed jointly by the USDA and U.S. Department of the Interior, to evaluate the safety of proposed introductions of exotic agents. In 1987, USDA reorganized the group and formed the "Technical Advisory Group on Introduction of Biological Control Agents for Weeds" (TAG), composed of 13 individuals from federal, state, and scientific organizations. Proposals were also reviewed by Canadian and Mexican officials, and, thus, proposed introductions were reviewed on a North American basis.

The final decision to permit release of exotic phytophagous agents in the U.S. remains the responsibility of APHIS. However, APHIS directed TAG to review research proposals and the documentation supporting release proposals for correctness and safety, and to provide guidelines for the researcher and recommend actions to APHIS. TAG members represent key federal agencies, including the U.S. Fish and Wildlife Service, which has the responsibility for the protection of endangered or threatened species, under the U.S. Endangered Species Act. The comments of the TAG reviewers are sent to APHIS and to the petitioner with an overall group recommendation. APHIS reviews the comments and makes a decision on whether or not to permit the agents' release. As required by the U.S. National Environmental Policy Act (NEPA), if the agent is to be released, an Environmental Assessment is prepared, currently by APHIS, assisted by the petitioner. Introductions are carefully documented, and copies of APHIS and TAG documents are maintained, by USDA's Biological Control Documentation Center. Procedures for meeting NEPA requirements and APHIS and TAG responsibilities are currently under review.

Introduction and establishment of weed biocontrol agents

Neal R. SPENCER[1] and R.D. RICHARD[2]

1 : USDA-ARS, P.O. Box 1109, 1500 N. Central Ave., Sidney, Montana 59270, USA
Fax : 406 482 5038 E-mail : spencer@ril.usda.montana.edu
2 : USDA-APHIS-PPQ, Bozeman, Montana, USA

A weed target is identified as a suitable target for biological control. The area of origin is established and foreign exploration locates suitable agents for the biological control of the weed target. Proper procedures are used to introduce the agent into the country. In quarantine, identification of the organism is verified and the parasites and diseases eliminated. The critical next step is to establish the agent on the targeted weed host. Conditions under which the agents were found in its native range are used to select initial release sites. Information on soils, climate and vegetation associations are used as indices. Sites are monitored for establishment and impact on the targeted weed. As additional agents become available, either from foreign sources or newly established colonies, additional releases are made using the data from the first successful sites. Indications on the rate of survival, based on the innate ability of the biocontrol agent to increase, will show some sites better than others.

In the early stages of establishment and population increase, the plan is to produce biocontrol agents and not control the weed. Successful control of a noxious weed requires large numbers of one or more control agents. To accomplish this goal, biocontrol agents are placed at release sites which are projected to greatly increase their numbers. Once this technology is developed, it is transferred to an action agency for deployment.

Redistribution of established exotic natural enemies of weeds and the technology transfer to State and local governments, Industry and Land owners

Dale E. MEYERDIRK

Biological Control Operations, USDA-APHIS-PPQ,
Riverdale, Maryland 29737-1236, USA
Fax : 301 734 8192

The redistribution of established exotic natural enemies for the biological control of weeds follows a series of research phases which include the foreign exploration, identification, culturing, host specificity testing, and environmental assessment of the release of a natural enemy candidate. Additional research phases continue following the permit and release of a specific natural enemy, which include evaluation of establishment, impact on the target weed and economic benefit evaluation. The latter research phases can be part of a transitional "classical biological control" implementation process, which potentially includes mass field collection and redistribution techniques that need to be developed or modified for each exotic natural enemy. The redistribution of these natural enemies is a cooperative effort. The technology transfer process shifting from research to implementation on a national scale to the state and local government level, industry and land owners is through a three phase process discussed in this paper.

Establishing linkages, consortia and partnerships

R. A. BUTTS[1], R. A. DECLERCK-FLOATE[1], and A.S. McCLAY[2]

1 : Agriculture and Agri-Food Canada, PO Box 3000 Main, Lethbridge, Alberta ,Canada
E-mail : BUTTS@hg.uleth.ca
2 : Alberta Environmental Centre, Vegreville, Alberta, Canada.

The success of weed biocontrol depends on the development of international linkages. The present classical weed biocontrol program in Canada started in 1960, with Agriculture Canada contracting IIBC to do surveys and host specificity studies. Initially, host tests were solely concerned with showing that economic plants would not be attacked, with only a few other related species being tested. Using this approach insects were cleared for release at a rate of about 0.3 species per year. In 1979, because the need to test native species arose, funds were solicited from other organisations to help fund screening of candidate agents in Europe. With the increased funding, the clearance rate increased to 0.8 species per year in spite of the more comprehensive studies. The need for consortia emerged when costs rose beyond what any one agency was prepared to pay. In addition, historically large contributors cut back funding because of budget restrictions. Thus, in 1991, starting with *Chamaesphecia hungarica*, the spurge consortium was formalised. Since this time consortia have grown to become international, with Canada and the US participating. The benefits of consortia are many. New resources and information can be obtained. Communication improves. Recommendations for future direction are outlined. Basic research is conducted in the target and originating countries. However, there are still issues to resolve. Control over the program is an issue. How much of a say does each member have ? Should the consortia be dictating to the country being contracted ? How do you define what each member is getting for its money ? In Canada, we have used consortia successfully for many years, but the system is still evolving. The most important aspect is open communication between all parties and because we are optimistic about this system we hope to see it grow.

Native pathogens and biological control : field application of *Rhizoctonia solani* and *Fusarium* spp. on *Euphorbia esula*

A.J. CAESAR, N.E. SPENCER and P.C. QUIMBY

USDA, ARS, Range Weeds and Cereals Research Unit,
Culbertson Hall 4th Fl, Bozeman, MT 59717, USA
Fax : 406 994 4526
E-mail : caesar@ril.usda.montana.edu

The rangeland weed leafy spurge (*Euphorbia esula*), an exotic weed introduced from Eurasia, infests c. 2×10^6 ha in the Northern Plains of the U.S, causing hundreds of millions of dollars in economic losses. Native soil-borne pathogens are being investigated as a component of strategies for the control of leafy spurge. *Fusarium* and *Rhizoctonia* spp. are isolated from diseased and dying plants in rarely-occurring natural stand declines in the U.S. or in association with insect damage caused by root-feeding insect biological control agents. Strains of these two species which had been isolated from diseased leafy spurge were applied in field plots in 1992, 1993, 94, and 1995, at three locations in Eastern Montana and one in Western Montana. The fungi were applied as granular formulations either singularly or in combination, with out without pre-wounding of plants, and within proximity or at some distance from areas where such insects as *Aphthona* or *Spurgia* spp. had established or were released. Reductions in stand density ranged from 4-35 shoots m^{-2} over 6-14 months in two field plots where statistically significant reductions were measured.

Technology transfer programmes for biological control of weeds : the New Zealand experience

L.M. HAYES

Manaaki Whenua-Landcare Research, P.O. Box 69,
Lincoln, New Zealand
Fax : +64 3 325 2418
E-mail : HayesL@Landcare.CRI.NZ

Biological control has become a major focus for managing a variety of agricultural and conservation weeds in New Zealand. Successful programmes designed to implement this technology have been in operation for almost a decade. Nucleus populations of control agents are released onto land managed by client organisations, and their staff are trained to manage the control agents. As a result more than 100 field staff throughout New Zealand are now implementing biological control for seven weed species. This network has enabled more than 1300 nucleus populations of biological control agents (insects and mites) to be released, and the fate of these to be monitored. Already 13 of the 15 species released are known to have established, and *Longitarsus jacobaeae* Waterhouse (Coleoptera : Chrysomelidae) and *Trichosirocalus horridus* Panzer (Coleoptera : Curculionidae) appear to be providing control at many sites. Field staff have assisted with research trials and raised the level of understanding about biological control in the wider community.
Recently the technology transfer programmes have been modified to meet the changing needs of people responsible for weed control in New Zealand, to overcome problems encountered, and to increase efficiency. A change in the legislation affecting the control of pest plants has altered the needs of participating organisations. Outputs are now targeted specifically at the needs of clients rather than providing the same service for all. The production of control agents has been streamlined by offering fewer species each year, and by allowing sufficient time for development of mass-rearing techniques before embarking on an intensive release programme. The programmes now support research to identify suitable methods for field staff to collect and redistribute promising agents, assess the effectiveness of well-established control agents, and integrate biological control technology with other control methods.

Technology transfer in biological control of weeds : results dictate the needs

J.H. HOFFMANN

Zoology Department, University of Cape Town,
Rondebosch 7700, South Africa
Fax : +27 021 650 3301
E-mail : hoff@botzoo.uct.ac.za

Some biological control of weeds programmes are remarkably successful and, once initiated, require no intervention to keep the weed under control. However, in most cases, only partial control of the target weed is achieved because the performance of the natural enemies is moderated by the attributes of the environments into which they disperse. The performance of partially-successful agents can be enhanced by integrating biological control with other management practices, a process that usually requires some degree of technology transfer. The biological control programmes against four weed species in South Africa are used to illustrate that : (i) there is no need for technology transfer when the success of the biocontrol agents does not depend on other management strategies, e.g. *Acacia longifolia* and *A. saligna* ; (ii) in some cases, technology can be transferred indirectly through appropriate manipulation of the control agents, e.g. *Sesbania punicea* ; and (iii) convincing recommendations are required when biological control is to be integrated with other control methods, e.g. *Opuntia stricta*. The need for post-release evaluation studies, to determine which model is best for a particular weed, is emphasized.

Community involvement in the distribution and evaluation of biological control agents

D.T. BRIESE[1] and D.A. McLAREN[2]

1 : CSIRO, Division of Entomology, GPO Box 1700,
Canberra, Australia 2601
Fax : +61 6 246 4177 E-mail : davidb@ento.csiro.au
2 : Keith Turnbull Research Institute, PO Box 48,
Frankston VIC, Australia 3199
Fax : + 613 9785 2007 E-mail : D.McLaren@dce.vic.gov.au

A key element for achieving a successful outcome to a biological control program is the effective redistribution of agents throughout the range of infestation of the target weed. Australian biological control workers have enjoyed a good reputation for scientifically-based and systematic studies leading to the selection, release and establishment of control agents. However, in the past, redistribution of agents following establishment has been done in a fairly piecemeal fashion. A recent social phenomenon in Australia has been the development of community groups that have become concerned with, and actively involved in, the remediation of a wide range of environmental problems. Weed biological control researchers are using these groups more and more frequently to develop control agent release networks, ensuring a more systematic redistribution of agents and more rapid delivery of biological control to the end-user. Examples of this, in particular those involving the Landcare program, are given to illustrate how the harnessing of community interest can provide a valuable resource for the classical biological control of weeds, both for the redistribution of agents and their subsequent evaluation.

SYMPOSIUM #10

PRODUCTION AND DELIVERY OF BIOPESTICIDES

Organizers :

Sevastianos ROUSSOS
ORSTOM, Unité de Microbiologie, 911 avenue Agropolis,
B.P. 5045, 34032 Montpellier cedex, France
Fax : (33) 67 54 78 00
E-mail : roussos@orstom.rio.net

and

Alain DURAND
INRA, Plate-forme Biotechnologie, 17 rue Sully,
21034 Dijon, France
Fax : (33) 80 63 32 29
E-mail : durand@dijon.inra.fr

Potential production of biopesticides in Latin America

Rodolfo QUINTERO-RAMIREZ

Institute of biotechnology, UNAM
(Universidad Autonoma del Edo de Morelos), Apto Postal 510-3,
Cuernavaca, Morelos 62271, Mexico
Fax. (52-73) 17 23 88
E-mail : quintero @ ibt.unam.mx

Agriculture in Latin America is facing two large and complex problems : it has to increase its production to feed its growing population and at the same time it must introduce new production methods and techniques to achieve sustainable developement. One of the key factors in the strategy to achieve the goals of sustainable agriculture and increasing output will be to reduce the use of agrochemicals and when possible to replace them by biological products. In this presentation I will review the state of the art of research and development on biopesticides and the possibilities of producing them in the Latin American region :
(a) Main research groups working on biopesticides ; in Latin America there are several groups working on different kinds of biopesticides (bacteria, fungi, viruses, etc.) against a large number of agricultural pests ;
(b) potential markets for specific biopesticides ; for some crops in specific countries a market estimation wille be presented ;
(c) economic evaluation of producing a bioinsecticide (based on *Bacillus thuringiensis*). A case study for producing 200 ton/year in Mexico will be shown.
Most of the discussion and analysis will be on *Bacillus thuringiensis* present and future developments, because this bacterium has been recognized in many countries as a potential marketable bioinsecticide, but so far, all of it is imported.

Formulation of a microbial product, example of *Bacillus sphaericus*

Ole SKOVMAND and Pierre GUILLET

ORSTOM, laboratoire de Lutte contre les insectes nuisibles (LIN),
Parc scientifique Agropolis Bât. 6, 34397 Montpellier cedex 5, France
Fax (33) 67 54 20 44
E-mail : skovmand@orstom.rio.net

Introduction : Most microbes that have potentials for being used for microbial control of pests are never used. Perhaps the scientists started looking at the organism for curiosity and with ideas of benefit for mankind and environment and failed to recognize all other aspects of product development than the initial discovery and efficacy tests. The goal of this presentation is to focus on other aspects of product development that have to be considered and analysed to evaluate microbial candidates for commercial use.

The presentation is based on the situation that a microbial organism has been found to have interesting biological activity, and it should be decided if more time and money should be spent on its development. Analysis starts with the definitions of customer needs, market evaluations, goal setting for the product, informations on target insect behaviour and finally the selection of product formulation type. The goal of the product development is to deliver the active microorganism or metabolite in a form that suits the users requirements. Definition of product parameters comes from a compromise between what possible can be done with current available knowledge, customer needs, insect behaviour and company dedication. These needs have to be properly defined and then transferred into technical descriptions of many different forms: insect-targets, non-target effects, application apparatus, application traditions and developments, storage conditions and shelf life, packaging etc. Then follows considerations of production, choice of preservatives and detergents for fluid or dry formulations. Finally, efficacy studies in the laboratory and then in the field are carried out.

Field tests have the goal to evaluate various formulations and dosages under specified and local conditions against target insects. These tests are performed in the early phases of product development and continue as long as the product is on the market in a steady effort to reach new targets of insects or new markets in other countries. At a smaller scale, field tests are also carried out for evaluating environmental side-effects. For engineered strains, the demands are much stricter and costly to meet. Toxicology and strain identification tests are carried out to meet registration needs. The rules for this area are different in various regions of the world, and very strict rules, thus very expensive, are in contradiction with the environmental need for more specific, biological products. It is stressed that the proper selection of order of all these tests is the key to cost and time reduction.

Materials and methods : A granule was formulated based on primary powder of *Bacillus sphaericus* with low cost ingredients to produce a slow release granule that release the spores in the feeding zone of mosquito larvae.

Results and discussion : In the tropics, field tests showed the granular product to be superior to the existing, commercial fluid concentrate when used in cessponds against *Culex quinquefasciatus* and in sunexposed waterholes against *Anopheles gambiae* at dosages of 03. and 3.0 g/m². The difference in effect is especially pronounced for the tests against *Anopheles*, where the effect of the granule was still near 100 % after 18 days at 3 g/m², when the effect of the fluid concentrate was 0 % control. Further, it was easy to apply and more tolerant to storage under tropical conditions."

Solid state fermentation of *Bacillus thuringiensis* tolworth as a tool for *Spodoptera frugiperda* control in maize

D. M. F. CAPALBO[1], F. VALICENTI[2], I. O. MORAES[3] and C. M. SOARES[1]

1 : EMBRAPA/CNPMA, CP 69, 13820 000, Jaguariuna SP, Brazil
E-mail : deise@cnpma.embrapa.br
2 : EMBRAPA/CNPMS, CP 69, 13820 000, Jaguariuna SP, Brazil
3 : UNESP/ S.J.R.Preto, CP 69, 13820 000, Jaguariuna SP, Brazil

Introduction : *Bacillus thuringiensis* (*Bt*) is a well known entomopathogenic bacterium that produces a spore-crystal complex, which is responsible for the entomopathogenic characteristic and could be obtained by fermentation process, either in liquid or semi-solid substrates. There are a few groups in Brazil that have succeeded in scaling-up the *Bt* fermentation process. Many reasons could be pointed out, but the principal ones are the difficulties in standardizing the resulting product and the low reproductibility of the lab results in the field. A group of researchers from EMBRAPA (CNPMS and CNPMA) / Brazil succeeded in developing a solid-state fermentation for the production of active *Bt* var. *tolworthi* (T09) against *Spodoptera frugiperda* (fall armyworm) in corn fields.

Materials and methods : the *Bt* T09 was grown and sporulated on humidified rice (main component of the culture medium), maintained into polypropylene bags, in controlled chambers (30 ± 2°C). After high sporulation (more than 10^9 CFU/g) was attained, the bags were maintained frozen. Whenever necessary they were defrosted, washed and centrifugated four times, and the pellets were suspended in water to get a 2 x 10^6 spores/ml. The resulting suspension was sprayed via tractor in corn fields in Sete Lagoas, in pre-established maize plants with egg batches. Pulverization took place at 4:00 PM and evaluation was done 48 hours later.

Results and discussion : the LC50 and LT50 were established with laboratory bioassays showing the following values : 1 mg spore-crystal toxin / 2,66 ml, for 2 day-old-larvae; 2,8 days for a 1-mg toxin/6.25 ml, respectively. For a 2-mg toxin/ml it was acomplished a 98% mortality after 24 hours. From all plants in the pre-established field, mortality of neonate larvae was 100% and all larvae were found dead on the leaves. During one cycle of maize crop, two pulverizations took place, and for 70 days after emergence it was not necessary to apply any other insecticide for fall armyworm control.

Improving the production and application of the Potato tuber moth granulosis virus in Egypt

S. ABOL-ELA, H. EL-BOLBOL, A. MONSARRAT and J. GIANNOTTI

Entomovirology Laboratory, Faculty of Agriculture-ORSTOM,
Cairo University, Egypt
Fax : (202) 703 948
E-mail : Thouvene@orstom.rio.net

Introduction : the research on the use of entomoviruses in biological control is recently developed in Egypt. The research policy is oriented to three parallel directions: the choice of the objective which was limited to the important key pests in Egyptian fields, the establishment of specific detection and diagnostic tools, and the pilot production of promising viruses for application.

Material and methods : among different viral groups, nine pathogenic viruses belonging to Baculoviridae, Picornaviridae, Parvoviridae and Bunyaviridae were isolated and characterized. The isolated viruses were found infecting *Phthorimaea operculella, Sesamia cretica, Ostrinia nubilalis, Chilo agamemnon, Pectinophora gossypiella* and *Spodoptera littoralis*.

Results and discussion : the research results permitted us to identify the model of the Potato Tuber Moth Granulosis virus for application. The pilot production was designed to produce sufficient quantity of GV to protect 10 000. The potato tubers are used for the production of GV diseased larvae. The number of eggs laid by 300 PTM females is sufficient to produce 60 000 diseased larvae. This production requires the treatment of 30 kg tubers with 5 OD of nonpurified GV (one diseased larva). The protection criteria of potato tubers is 0 larva per tuber. The GV concentration for treatment should induce mortality to newly hatched larvae before attacking tubers.

The concentration value of *P. operculella* GV required to achieve 95% protection for one kg of stored tubers is estimated by 10 OD (two diseased larvae). Application of this GV concentration in 10 ml of water mixed with 0.001 Tween 20 as a simplified formulation give good result. It is not necessary to apply anti UV protectant in case of application on stored tubers.

Development of a biopesticide (Ostrinil) for fighting against the European Corn Borer

Alain DURAND[1] and Catherine DESGRANGES[2]

1 : INRA, Plate-forme de Prédeveloppement en Biotechnologie,
17 rue Sully; 21034 Dijon, France
Fax : (33) 80 63 32 29
E-mail: durand@dijon.inra.fr
2 : NPP, Calliope, 35 Av. Léon Blum, Parc d'activités Pau-Pyrénées,
64000 Pau, France
Fax. (33) 59 84 89 55

Relationships have been established between INRA (National Institute for Agronomic Research) and an industrial company for the development of a biopesticide (OSTRINIL) able to fight against the European corn borer (*Ostrinia nubilalis*), larvae which can be responsible of up to 20% of losses in corn fields. The partners involved in this development were :
- the Laboratory for biological Control (INRA-La Minière)
- the Platform for Development in Biotechnology (INRA-Dijon)
- the French Society NPP (Natural Plant Protection) belonging to the holding THALIA / CALLIOPE.

The Laboratory for biological Control, specialized in the fight against pathogenic insects of plants, has isolated a strain of *Beauveria* from an infested larva in the Beauce region. This fungus *Beauveria bassiana* infects specifically the corn borer by contact and is not toxic for bees and the auxiliary fauna. The death of the corn borer larvae occurs between 2 and 5 days.

The Platform for Development in Biotechnology has developed a technology and a process for the production and the formulation of this entomopathogenic fungus. The process, which has been patented, consists of cultivating by Solid State Fermentation *Beauveria bassiana* directly on clay microgranules. These clay granules are wetted with an optimal nutritive solution containing the inoculum. The cultivations were carried out in a 1.6 m^3 pilot reactor developed earlier for other applications. This reactor, based on the packed bed principle, has been used for the productions of large quantities for field tests and also for studying the different parameters involved in the optimization of the process. The final product, dried in the reactor, contains mycelium and spores (about $2x10^9$ spores per gram of dry matter). It can be directly sprayed in fields by the usual equipments without formulation (application dose : 25 kg/ha) and with the same efficiency than chemical insecticides.

The French society NPP has been created within THALIA holding for the production of biological products developed by CALLIOPE from INRA patented technologies. After having passed by an approval commission (Temporary Licence for Sale), this product, OSTRINIL, is now produced at industrial level by NPP. Taking into account some drawbacks and limits of the technology developed by INRA, new reactors have been built for the industrial productions.

Formulation and application of *Metarhizium anisopliae* Metch, for the control of insect pests of oilseedrape

A.I. DAVID-HENRIET[1], T.M. BUTT[2], B.J. PYE[1] and L. IBRAHIM[2]

IACR-Rothamsted exp. Stn., Harpenden, Herts AL5 2JQ, United Kingdom
1 : Biological and Ecological Chemistry Dept
2 : Entomology and Nematology Dept
Fax : 44 1582 762595
E-mail : Ana-Isabel.David-Henriet@bbsrc.ac.uk

Introduction : Oilseed rape is a major arable crop in Europe which is attacked by a wide range of insect pests. At IACR-Rothamsted, isolates of *Metarhizium anisopliae* have been identified which are highly pathogenic for selected pests including *Psylliodes chrysocephala, Meligethes* sp., *Lipaphis erysimi* and *Myzus persicae*.

Material and methods : Conidia of *M. anisopliae* were harvested from sporulating cultures grown on SDA, air dried and stored at 4°C and < 15 % relative humidity until needed. The hydrophobic conidia were formulated in either 0.03% aq. Tween 80, mineral, paraffin or vegetable oil and tested against adult mustard beetle (*Phaedon cochleriae*) or larvae of *P. chrysocephala*. Inoculum was applied to flat leaves or field crops using either APE 80 electrostatic or conventional hydraulic sprayers. Yeast extract stimulates germination and was included in the formulations to see if also enhanced virulence.

Results and discussion : Conidia formulated in the paraffin, Shellsol T, proved most efficacious causing 70% and 100% mortality of adult *P. Cochleriae* and *P. chrysocephala* larvae, respectively. Very low volume application (10 l ha -1) of conidia using the APE 80 electrostatic delivery system resulted in a uniform deposition of inoculum on the flat leaf surface. In field trial the delivery system greatly increased deposition on aerial parts of the plant, including the abaxial leaf surface, compared with the conventional hydraulic spraying system. Enhanced virulence of *M. anisopliae* was obtained against *P. cochleriae* by formulating conidia with yeast extract. The reason for the increased virulence is being investigated.

Production and use of *Trichoderma harzianum* as biocontrol agent for *Botrytis cinerea* in grapes

R. SAN MARTIN, E. AGOSIN and B. AGUILERA LATORRE

Catholic University, Department of chemical and Bioprocess Engineering,
Av. Vicuña Mackenna 4860, Santiago, Chile
Fax : (562) 552 4054
E-mail : sanmarti@ing.puc.cl

Introduction : During the last 4 years, we have investigated the production of *Trichoderma harzianum* spores at a pilot plant scale for the control of *Botrytis cinerea* in grapes. This represents an important market in Chile, with approximately 50.00 hectares, and a use of US 10 million of chemical fungicides per year. The project had a funding of US 1.5 million, with direct participation of the University, State funds and the private industry. Based on the results, a production company in Chile will be set up before 1997.
Materials and methods : The production of *T. harzianum* was investigated at a pilot plant level using 60 and 300 Lt. bioreactors (Bioengineering, Switzerland). Biocontrol test were established in the lab, as well as extensive fields tests. The formulation and drying of the spores was given considerable attention to obtain extended shelf life.
Results and discussion : Major achievements are the following: repetitive production of 10^8 spores/ml in 60 h in 250 Lt. working volume bioreactors. Recovery and drying of the product with minimum loss of spore viability (e.g. 10-15%). Formulation of product as a wettable powder, with minimum loss of viability at room temperature, for at least 3 months. Refrigerated, the product shows a stability above 6 months. The product has been tested in the field for 4 seasons, and it shows a performance comparable to traditional chemical fungicides. Due to its compatibility with chemical fungicides, it can be used in IPM programs.
Perspectives : A new company will be set up to carry on the production of the biopesticide in Chile, at a selling price lower than chemical fungicides. The principal market will be export type grapes.

Production of *Paecilomyces fumosoroseus* conidia in submerged culture

H. CARDENAS and M. DE LA TORRE

Biotechnology Department, CINVESTAV-IPN, P.O. Box 14-740,
Mexico City, 07000 Mexico
Fax : (525) 752 05 90
E-mail : mmdelato@mvax1.red.cinvestav.mx

Introduction : Conidia of native Mexican strains of *Paecilomyces fumosoroseus* virulent against adults, nymphs and eggs of the whitefly, *Bemisia tabaci*, are produced in rice-filled plastic bags. However, this method has drawbacks : variation in sporulation, long incubation periods and short spore viability. Submerged cultures are advantageous because allow shorter fermentations, precise manipulation of culture conditions and yield more resistant spores.

Materials and methods : *P. fumosoroseus* PfrD was cultured either in shaked-flasks or a 6.6-l fermenter, in a synthetic medium containing ammonium nitrate and variable quantities of glucose and yeast extract. Glucose, nitrogen and biomass were evaluated by DNS, kjeldahl and dry weight methods respectively, whereas hyphal bodies and conidia were counted under the microscope.

Results and discussion : The effects of temperature stress, organic nitrogen and C:N ratio on sporulation were evaluated. A 24 hour incubation period at 37°C followed by a downshift at 30°C increased sporulation, while incubation at 37°C for one or two hours did not have any significant effect. At 5 g/L of yeast extract, spore production was proportional to glucose concentration from 10 to 50 g/L. However, the produced conidia germinated when residual glucose was noted. Maximal yields of conidia (2.7×10^8 spores/ml) were produced after 5 days of culture, in a nitrogen-limited-medium containing 50 g/L of glucose and 5 g/L of yeast extract. Ninety percent of these spores were conidia virulent against adults of *B. tabaci*. To obtain a high yield of conidia of *P. fumosoroseus*, conidia germination must be prevented by avoiding an excess of remanant glucose at the end of the culture, the growth have to be nitrogen limited and organic nitrogen is required.

SYMPOSIUM #11

NOVEL STRATEGIES FOR THE APPLICATION / INTRODUCTION OF PATHOGENS FOR THE CONTROL OF PEST INSECTS AND WEEDS

Organizer :

Lawrence A. LACEY
USDA, ARS, Yakima agricultural Research Laboratory
5230 Konnowac Pass Road, Wapato, WA 98851, USA
Fax : 1 509 454 5646
E-mail : LACEY@YARL.GOV

Effects of different species and strains of the nematophagous fungus *Arthrobotrys* sp. from West africa on *Meloidogyne* species

Robin DUPONNOIS and Thierry MATEILLE

ORSTOM, laboratoire de Nématologie, B.P. 1386, Dakar, Sénégal
Fax : (221) 32 16 75 (and 32 43 07)
E-mail : mateille@belair.orstom.sn

Different species of the nematophagous fungus *Arthrobotrys* sp. and several strains of *Arthrobotrys oligospora* have been studied for their antagonistic effects against the nematodes genus *Meloidogyne*, important parasites on vegetables.
In vitro, all the fungi trapped *Meloidogyne mayaguensis* and *M. incognita* juveniles but had no effect against the juveniles of *M. javanica.* In pot experiments with *M. mayaguensis*, all these fungi reduced the nematode populations and induced a stimulation of the tomato seedling growth. In a field trial, a strain of *A. oligospora*, isolated in Senegal and incorporated into compost blocks, was efficient in increasing the tomato seedling growth.
Introduction of such nematophagous fungi in compost blocks as a stealthy nematode biocontrol technique adapted for developing countries is discussed.

Formulation and application strategies to overcome environmental constraints in the use of entomopathogenic Hyphomycetes against pest insects in epigeal habitats

Mark S. GOETTEL and G. DOUGLAS INGLIS

Agriculture and Agri-Food Canada, Lethbridge Research Centre
P.O. Box 3000, Lethbridge, AB T1J 4B1, Canada
Fax : (403) 382 3156
E-mail : Goettel@em.agr.ca

Control of a variety of insects by entomopathogenic hyphomycetous fungi can be obtained by application of infective propagules in epigeal habitats. However, success will depend on the use of the right propagule type, formulated in an optimal fashion and applied at the right time to the right place. Microclimatic constraints include solar radiation, temperature, and humidity. Persistence of conidial propagules in epigeal habitats is generally poor. Since a threshold of inoculum is required to cause disease in insects, the poor survival of conidia could seriously decrease the efficacy of conidia applied to foliage by limiting the time that conidial populations remain above inoculum thresholds. Formulation and application strategies that increase persistence and optimize host targeting are important ways in overcoming some of the environmental constraints. The most important parameter limiting conidial persistence in epigeal habitats appears to be sunlight. For instance, conidia deposited within the shaded portion of the canopy survive significantly longer than those deposited on the outer canopy exposed to sunlight. Incorporation of sunlight blockers (e.g., clay) and/or UVB-absorbing compounds (e.g., Tinopal) enhance survival. Formulation of conidia encapsulated as baits can also increase survival of propagules in epigeal habitats by protecting them from light. However, conidia must be liberated prior to ingestion of baits as they rarely incite disease after being ingested. In many cases, insects become surface-contaminated during handling and feeding on baits. Humidity does not always seem to be as major a constraint as previously believed. With some insects, insect body temperature is proving to be a major constraint; infected insects preferentially bask in the sun elevating their temperature thereby escaping disease. Once the environmental constraints for each host-pathogen combination have been identified, it may be possible to overcome these through strain selection, formulation and pathogen targeting. For instance, fungal cocktails may be used to overcome the constraints of temperature in thermoregulating insects. The efficacy of fungal isolates should be tested against host insects in bioassays that incorporate as many pertinent

environmental parameters as possible. Inoculation techniques and the environmental conditions chosen should mimic as much as possible the natural situation and more specifically, conditions at the level of the host microhabitat.

A novel, broad-host pathogen and delivery system for controlling weeds

S. M. YANG and N.W. SCHAAD

USDA-ARS, Foreign Disease-Weed Science Research Unit, Fort Detrick, Bldg. 1301, Frederick, MD 21702, USA
Fax : 301 619 2880
E-mail : syang@asrr.arsusda.gov

Currently, the bioherbicide strategy for control of weeds emphasizes host-specific plant pathogens. We propose a novel strategy for biological control of weeds that uses non- or low-virulent plant pathogens in oil-based carriers as both safe and broad-spectrum bioherbicides. Saprophytic fungi are not virulent and do not cause disease by themselves, and low-virulent plant pathogens cause only slight infection of plants under natural conditions. But these organisms, when used with special carriers, can infect and kill many weed species. An advantage of this strategy is that only plants which come into contact with both the organism and the carrier will be damaged. Drift from oil-based sprays is extremely limited, and the small amount of spray that reaches non-target plants is expected to cause only insignificant damage at most. Results of greenhouse and field studies demonstrate the lethal effects of saprophytes or low-virulent pathogens when applied in oil-based carriers to a broad range of weeds.

Application of entomopathogens using irrigation systems

John D. VANDENBERG and Mark RAMOS

USDA ARS, U.S. Plant, Soil and Nutrition Laboratory
Tower Road, Ithaca, New York, 14853, USA
Fax : 607 255 2459
E-mail : jdv3@cornell.edu

The use of crop irrigation systems for the application of insect microbial control agents dates back many years and includes a broad range of target insects. Organisms from all major pathogen groups - bacteria, fungi, nematodes, protozoa, and viruses - have been applied as the subject of research studies on small grains, corn, cotton, tree fruits, vegetables, and greenhouse and horticultural crops. Habitats of target insects range from leaves to roots to fruits and buds. Irrigation methods used to apply pathogens include overhead sprinkler systems, row flooding, microjets, and drip systems. This presentation will provide a review of past research, but will focus on current research in selected systems. At present, only a few microbial pest control agents are used in concert with irrigation systems for commercial crop production ; these systems will also be featured in the presentation.

Application of microbial pesticides : limitations and a practical solution

Andrew C. CHAPPLE

Koyntoypiotoy 13, Tpiandpia 55337, Thessaloniki, Greece
E-mail : chapple@openet.gr

The long-term success of microbial pesticides requires that they be used extensively in broad-acre crops, *ie*. field grown, normally non-irrigated, minimum labour input crops, *eg*. cotton, corn, cereals, soya, rice, vines, etc. Such crops impose severe limitations on application, particularly foliar applied pesticides. Farmers are unlikely to buy expensive sprayers solely to apply a biological pesticide, irrespective of any "green premium" so gained. Therefore, microbial pesticides must be applied in conventional spray volumes using conventional equipment - *ie*. approx. 200l/ha and boom mounted flat fan or hollow cone nozzles.

With respect to application, biological pesticides have several advantages over conventional chemical alternatives. Their near-uniform distribution of particle size and lack of sub-lethal dose effects allows for the manipulation of the spraying process to enhance their activity. However, current conventional application systems severely limit the scope of exploitation.

A novel application method - a double nozzle system - is described. The system utilises conventional nozzles and equipment, is cheap, versatile, and lends itself to the manipulation of spray clouds to the benefit of the pesticide being applied. It is particularly suited to pesticides that require specific adjuvants to be closely associated with the active component - eg. humectants, anti-evaporants, enhancement agents etc. As such adjuvants are often the main route by which a microbial pesticide is delivered, activated, enhanced, or protected, obtaining the maximum effect from such adjuvants is important.

Autodissemination of entomopathogens

Fernando E. VEGA[1] and Lawrence A. LACEY[2]

1 : USDA, ARS, National Center for agricultural Utilization Research, Peoria, Illinois, 61604, USA
Fax : 309 681 6693 E-mail : vegafe@ncaurl.ncaur.gov
2 : USDA, ARS, Yakima agricultural Research Laboratory, 5230 Konnowac Pass Road, Wapato, WA 98851, USA
Fax : 1 509 454 5646 E-mail : LACEY@YARL.GOV

The autodissemination of entomopathogens consists of using insects to vector microbial control agents to other insects as a mechanism for pest management. Several examples of this concept have been reported, mostly for viruses and fungi. Most autodissemination approaches use devices to which the insects are attracted (e.g., using pheromones and/or other semiochemicals) and in which a formulation containing the entomopathogen is placed in such a way that the insect comes in contact with the agent. These devices, which allow insects to exit and carry the agent to other insects, have been used to spread entomopathogenic viruses by bees and moths (Environ. Entomol. 23:492-501; J. econ. Entomol. 85:710-719; J. Stored Prod. Res. 29:71-74), the fungal entomopathogens *Metarhizium anisopliae*, *Beauveria bassiana*, and *Zoophthora radicans* by different insects (Biocontrol Sci. & Technol. 3:315-320; Biol. Control 5:545-552), as well as protozoans by dermestids (J. econ. Entomol. 70:469-474.).
Autodissemination can also be one of the natural mechanisms of pathogen dispersal. For example, the non-occluded baculovirus of the Rhinoceros beetle *(Oryctes rhynoceros)* is transmitted when insects come in contact, while mating or feeding in palm trees, with material excreted by contaminated insects (J. Invertebr. Pathol. 59:61-68.) Similarly insects can spread viruses (via feces) after feeding on infected hosts, as has been reported for nabids feeding on nuclear polyhedrosis virus (NPV)-infected larvae of the Velvetbean caterpillar *Anticarsia gemmatalis*; (Environ. Entomol. 16:1330-1333), and the Southern corn rootworm (*Diabrotica undecimpunctata howardi*) feeding on cadavers of *Helicoverpa zea* infected with a NPV (J. Invertebr. Pathol. 61:313-314). Parasitoids are also know to spread viruses while ovipositing on their hosts (Ann. Entomol. Soc. Amer. 68:593-594). Transovarial transmission of viruses has been reported when the genitalia of adult females of the Alfalfa caterpillar (*Colias eurytheme*) has been artificially infected with a NPV (J. Insect Pathol. 4:113-121).

Formulation and application strategies for the biocontrol of weeds in crops

W. J. CONNICK, Jr.[1], D. J. DAIGLE[1], C. D. BOYETTE[2] and P. C. QUIMBY, Jr.[3]

1 : USDA, ARS, South Regional Research Center, P.O. Box 19687, New Orleans, LA 70179, USA
Fax : (504) 286 4419 E-mail : wconnick@nola.srrc.usda.gov
2 : USDA, ARS, SWSL, P.O. Box 350, Stoneville, MS 38776, USA
3 : USDA, ARS, RWL, Culbertson Hall, Montana State University, Bozeman, MT 59717, USA

Formulation and application technology must address shortcomings in the efficacy and shelf life of bioherbicides. Invert emulsion and vegetable oil emulsion carriers were developed that reduced fungal weed pathogens' (mycoherbicide agents) dependence on dew, were less phytotoxic to crops, and were easy to spray. Adjuvants such as water-soluble weed filtrates and fruit pectin altered host specificity of a mycoherbicide agent and broadened its weed control spectrum. Weed control efficacy also was improved by incorporating adjuvants such as starch, corn flour, casamino acids, and sucrose into spray formulations. The proven alginate/fungus encapsulation technology was applied to aquatic weed control. New "Pesta" wheat flour-based granules entrapped mycoherbicide agents in a wheat gluten matrix and, with proper drying, preserved agent viability for one year or longer. An oil/absorbent/sucrose encapsulation technique preserved the viability of bacterial herbicides for spray application. A nonionic organosilicone surfactant facilitates stomatal penetration by bacterial weed pathogens, causing infection in the absence of wounds or free moisture on the leaves. These and other recent advances in bioherbicide formulation and application technology will be presented.

Application of entomopathogens for the control of soil dwelling pests

Trevor A. JACKSON

AgResearch, OP Box 60, Lincoln, New Zealand
Fax.(0064)3 325 2946
E-mail : jacksont@agresearch.cri.nz

Soil dwelling insect pests pose some of the most intractable problems in pest control. Many chemicals, which were used in the past for soil insect control, have become ineffective due to pest resistance and resurgence, are no longer economical or cause environmental pollution. A promising alternative to chemical pesticides is microbial control. The soil is rich in microorganisms, some of which are pathogenic to insects and have the potential to be used as bioinsecticides. While several insect pathogens have been produced in bulk and commercialised, success against soil pests has proven difficult to achieve, often due to lack of appropriate application methods.

While pathogens are often well adapted for survival in the soil, they must be delivered to a position within the soil profile where they will come into contact with the target insects. If applied to the soil surface they will be subjected to desiccation and ultra violet light and may only survive for a few hours. Researchers from several different programmes have independently concluded that subsurface application is essential for the success of microbial pesticides against insects feeding in the soil. Successful establishment of bacteria, nematodes and fungi for scarab larvae control has been achieved by application of the microorganisms into the soil through modified seed drills. Fungi coated on wheat grains are applied directly through the seed drill, while bacteria and nematodes have been applied as liquid suspensions. Once established within the soil, the pathogens can initiate a cycle of infection and recycle through the pest population.

The use of specialised equipment for application of microbes to the soil adds significantly to the cost of application. This extra cost is not always warranted and thus low cost application strategies may be needed. Pathogens can be introduced as an inoculum in spot applications or spread naturally through the activity of mobile stages of the target insect. They can be applied at planting by incorporation into a seed coating or can be introduced into the soil by irrigation. The potential and problems of these application methods will be discussed.

Use of *Beauveria bassiana* as an endophyte of corn for control of the European corn borer

Leslie C. LEWIS

USDA-ARS, Corn Insects Research Unit, Genetics laboratory,
Ames, Iowa 50011, USA
Fax : 515 294 2265
E-mail : leslewis@ia.state.edu

Beauveria bassiana, an ubiquitous fungus, has a long documented history of association with the European corn borer, *Ostrinia nubilalis.* It has been applied as a crop protectant by many researchers, isolated from wild specimens, defined in epizootic logical studies and more recently discovered to form an endophytic relationship with corn, *Zea mays*, and provide season-long suppression of the European corn borer.

Conidia of *B. bassiana* placed in the whorl of a vegetative-stage corn plant or behind the sheath-leaf collar of an anthesis stage plant, will adhere, germinate and penetrate the corn tissue establishing as an endophyte. Once the fungus has entered the plant it can routinely be isolated from the nodal plates and internodal tissue. Larval corn borer mechanically placed within the plant following invasion by the fungus exhibit a mycosis within 7-10 days. These data show that *B. bassiana* can persist inside the corn plant throughout the growing season while maintaining its pathogenicity toward the European corn borer.

Many fungi are known to colonize plants, usually as plant parasites or symbiots. Endophytic fungi that do show toxicity towards insects may produce feeding deterrents and/or antibiotics, but are not insect pathogens as defined by Koch's postulates. *Beauveria bassiana* is different by (i) being an entomopathogen that colonizes a plant and (ii) not having been documented as a plant pathogen. In addition, there is no evidence of *B. bassiana* coevolving with the corn plant or interacting specifically with the corn plant. Therefore, this endophytic relationship between this entomopathogenic fungus and the corn plant is unique. The role of this unique relationship in fungal epizootics in corn ecosystem will be discussed.

Control of Black field crickets, *Teleogryllus commodus* (Orthoptera : Grylloidea), with an experimental bioinsecticide containing conidia of *Metarhizium anisopliae*

Richard J. MILNER[1] and Lisa MILLER[2]

1 :CSIRO, Division of Entomology, GPO Box 1700, ACT 2601, Australia
Fax : (06) 246 4042 E-mail : richardm@ento.csiro.au
2 : Agriculture Victoria, 83-85 Gellibrand Street, Colac,
Vic. 3250, Australia

The black field cricket, *Teleogryllus commodus*, is a serious pest of pastures in western Victoria, Australia. The insect has an annual life cycle with the eggs hatching in December. The nymphs live in cracks in the gound and emerge, mainly at night, to feed on pasture foliage and seeds. They are particularly damaging in autumn (March/April) and they reduce the ability of the pasture to grow in response to the rain which marks the end of the summer drought. At present control is achieved using with a malathion/cereal bait which is often applied from fixed-wing aircraft to many thousands of hectares. In response to farmer demands for a biological alternative, we have been evaluating the efficacy of *Metarhizium anisopliae* for cricket control. This disease occurs naturally but rarely kills more than 5% of the population. A number of isolates obtained from black field crickets in various parts of Victoria were available at the start of the study. These isolates were compared using RAPDs and found to be all very similar. One of these isolates, FI1099, was selected for use in field trials in 1995. In the field application of 4 x 10^{13} conidia/ha in a high volume water/oil emsulsion gave 60-70% control after about 3 weeks compared with 80% control using malathion/cereal baits. At another site a lower dose was tested (2 x 10^{13}/ha) and found to be less effective giving about 30-40% control. However at this second site a pure oil ULV spray was compared with the high volume oil/water emulsion. The ULV spray was more effective. In 1996, another isolate FI1037 was tested in the field using 2 application methods, the oil ULV spray and a rice bait. For both treatments the dose was 1.5 x 10^{12} conidia/ha. The ULV spray reduced the population in 13 days from 6.4 crickets/bag to 1.4 crickets/bag while the malathion bait reduced the population from 6.4 to 1.9 over the same period.

SYMPOSIUM #12

BIOLOGICAL CONTROL OF POSTHARVEST DISEASES : MAKING IT A REALITY THROUGH TECHNOLOGY TRANSFER

Organizer :

Charles L. WILSON
USDA, ARS, Appalachian Fruit Research Station,
Kearneysville, WV 25430, USA
Fax : 304 728 2340
E-mail : cwilson@ASRR.ARSUSDA.GOV

A broader biological control concept for plant pathology

Charles L. WILSON

USDA, ARS Appalachian Fruit Research Station, 45 Wiltshire Road,
Kearneysville, WV 25430, USA
Fax : 304 728 2340
E-mail : cwilson@ASRR.ARSUSDA.GOV

Entomologists developed their first paradigm for biological control of plant pests around a "one on one" interaction between an insect and parasite or predator. Plant pathologist initially followed this concept, but in recent years there have been attempts to broaden the definition of biological control of plant diseases. If we define biological control of plant diseases as "the control of a plant disease by a natural biological process or the product of a natural biological process" three major avenues become available to pursue biological control :
(1) control with antagonistic microorganisms through competition ;
(2) control with natural plant- or microbe-derived pesticides (the product of a natural process) ; and
(3) control with induced resistance.
In our study of antagonists which control postharvest diseases of fruits and vegetables, we have discovered that a single antagonist may elicit all three forms of disease control. We have found that combinations of biological control agents (e.g. antagonists plus natural compounds) can be synergistic.

An ecological basis for the use of natural antagonists to control postharvest diseases

Mark WILSON

Department of Plant Pathology, 209 Life Sciences Building,
Auburn University, Auburn AL 36849-5409, USA
Fax : 334 844 1947
E-mail : mwilson@ag.auburn.edu

Biological control of postharvest pathogens using natural antagonists has become one of the more successful areas of biocontrol. Recent emphasis in selection protocols has been on antagonists which compete with the pathogen for nutrients required for either spore germination or hyphal growth. The selected biocontrol agent has to colonize the various habitats present on stored produce, including unwounded fruit surfaces and fruit wounds, each of which exhibits a different chemical and physical environment. In order to be effective in each habitat, the biocontrol agent must survive, multiply and, potentially, consume resources required by the pathogen. Unfortunately, to date very little is known about the microbial or chemical ecology of fruit surfaces or wounds. In contrast, however, substantial progress has been made in understanding the microbial and chemical ecology of leaf surfaces. Epiphytic microbial populations on leaf surfaces are known to be nutrient-limited. Inhibition of epiphytic phytopathogenic bacteria by non-pathogenic bacteria can be achieved through pre-emptive (prior) use of growth-limiting nutritional resources. Similarly, inhibition of conidial germination can be achieved by competition for exogenous nutritional resources by indigenous or introduced microorganisms. The nature of these limiting nutritional resources apparently depends on the C:N ratio of the habitat. In such systems, the effectiveness of biocontrol based on nutrient competition appears to be related to similarity in resource use between the biocontrol agent and the target pathogen. Such information, derived from ecological studies in the phyllosphere, can contribute significantly to the development of postharvest biocontrol and has already facilitated the development of strategic approaches to the selection of mixtures of antagonists for superior biocontrol efficacy.

How natural antagonists work to bring about biological control

Michael WISNIEWSKI[1] and Samir DROBY[2]

1 : USDA, ARS, 45 Wiltshire Road, Kearneysville, WV 25430 USA
Fax : 304 728 2340
E-mail : wisniew@asrr.arsusda.gov
2 : Agricultural Research Organization, The Volcani Center, Bet Dagan, Israel

The recent discovery, development, and commercialization of yeast antagonists to control postharvest diseases of citrus and pome fruit (e.g., Aspire^{TM}, Ecogen) has been accompanied by an intensive effort to elucidate the mechanisms by which the antagonists are able to exert their biocontrol activity. A full understanding of this process is essential for the further development of biocontrol strategies. To date, we have discovered that both direct and indirect interactions of the yeasts with postharvest pathogens, as well as the commodity itself, play an integral role in the biocontrol activity. In contrast to many synthetic chemical fungicides, it is hypothesized that the complex interactions exhibited by the yeast should make it difficult for the pathogen to develop resistance. We have reported that nutrient competition, attachment of the yeast to the pathogen, induced resistance, and cell wall hydrolases, are all involved directly or indirectly in the biocontrol activity of the yeast. In addition, we have discovered and patented a unique, biocontrol activity exhibited by the extracellular polysaccharides secreted by the yeast cells. Additionally, we have characterized the resistance of the yeast to agents, such as calcium and deoxyglucose, that in turn are inhibitory or toxic to the postharvest pathogens. Such research has laid the groundwork for developing improved biocontrol strategies for the near-future.

Natural fungal, plant and animal-derived antimicrobials

Horace G. CUTLER[1], Stephen R. PARKER[2] and Robert A. HILL[2]

1 :USDA, ARS, Richard B. Russell Center,
950 College Station Road, Athens, Georgia, USA
Fax : 706 546 3250
E-mail : HCutler876@aol.com
2 : HortResearch, Natural Products Group, Ruakura Research Center,
Hamilton, New Zealand

During the course of examining fungi for biologically active natural products, the non-phytopathogenic *Trichoderma koningii* was isolated from the soil-line of a thrifty *Diffenbachia* plant. Upon semi-solid fermentation, subsequent extraction and column chromatography, the organism yielded a number of novel compounds that were significantly inhibitory to etiolated wheat (*Triticum aestivum*) coleoptiles. Among these were koninginins A, B, C, and E. In addition, the known compounds cyclonerodiol and 1-monolinolein, which also exhibited inhibitory growth, were extracted. Koninginins A, B, and C proved to be active against the cereal phytopathogen *Gaeumannomyces graminis* var *tritici*: E could not be tested because of insufficient quantities. 1-Monolinolein, a common component of vegetable oils and partially digested animal fats, was also active in the wheat coleoptile assay, but was relatively inactive against the growth of greenhouse-grown bean, corn, and tobacco plants. It was also inactive against *G. graminis*. However, the literature indicates that the material is active against bacteria and encapsulated viruses and this has been attributed to its membrane disruptive property.

Another *Trichoderma* metabolite, specifically from *T. harzianum*, is 6-pentyl-α-pyrone. This secondary metabolite was inhibitory to wheat coleoptiles at 10^{-3}M and inhibited the growth of *Aspergillus flavus*, the producer of the aflatoxins, at dilution rates of 1:40 when assayed *in vitro*. It has been field tested against *Armillaria novae zealandiae*, by injection into infected kiwifruit vines, where it fully controlled that organism. In addition, it has been very successfully employed to control *Botrytis cinerea* in stored kiwifruit and extensive trials have shown that treated fruit are plump, firm and juicy after one year in storage. 6-Pentyl-α-pyrone occurs in fruits, most notably in peach (*Prunus persica*) and it has been extracted from pure peach essence. Hence, two small research groups have developed a natural product fungicide from the basic discovery to practical field application.

A multifaceted biological control strategy

Ahmed EL-GHAOUTH[1] and Joseph ARUL[2]

1 : USDA, ARS, Appalachian Fruit Station, Kearneysville, WV 25430, USA
Fax : 304-728-2340
2 : Département de Science et Technologie des Aliments, Université Laval, Québec, G1K7P4, Canada

Postharvest diseases of fruits and vegetables cause major losses in food production. Presently, fungicides represent the primary means of controlling postharvest diseases. Public concerns over the presence of chemical residues in the food chain, the possible deregistration of some of the more effective fungicides, and the development of fungicide-resistant strains of postharvest pathogens have generated an urgent need for the development of safer alternative technologies. Recently we have witnessed an increase interest in the development of non-antibiotic producing antagonistic yeasts for the biological control of postharvest diseases and a number of antagonistic microorganisms have been patented and some are commercially available. Although the potential of the use of antagonists to control postharvest decay has been demonstrated with several commodities, under commercial conditions this emerging technology often do not offer consistent disease control comparable to that obtained with synthetic fungicides. For biological control methods to emerge as an economically viable option, their consistency and efficacy in reducing postharvest diseases must be enhanced to a level comparable to that of synthetic fungicides.

This paper presents an overview of the different components of a multifaceted strategy and its advantages. The fundamental basis and the potential of this novel approach will be discussed with special reference to two biological products developed in our laboratory (Bioactive coating and Bioenhancer).

Bridging the gap between laboratory results on biological control of postharvest diseases and commercial application

E. CHALUTZ and S. DROBY

Department of Postharvest Science of fresh Produce, Agricultural Research Organization, The Volcani Center, P.O. Box 6, Bet Dagan, 50250 Israel
Fax : 972 3 968 3622
E-mail : vtfrst@volcani.agri.gov.il

Several factors indicate that in the postharvest environment, chances for success in the development of biological control measures may be high : the partially controlled environment may help shift the balance of the interactions between the host, the pathogen, and the antagonist in favor of the antagonist; the restricted and well defined site of application of the biocontrol product, the relatively high cost of the commodity and the fact that protection against postharvest diseases may sometimes be needed for a limited time only, are all contributing factors. On the other hand, other factors suggest specific difficulties in controlling diseases after harvest by biological means. These include the extremely high level of disease control required, food safety considerations, and the relatively small potential market for use of the biofungicide.
During the past five years we have been involved in an effort to commercialize a biocontrol product of postharvest diseases of citrus fruits. The yeast biocontrol agent *Candida oleophila* has shown promising results in laboratory tests. Together with a private company, Ecogen Israel Partnership, approximately 150 experiments have been performed in a pilot packinghouse located at the Volcani Center. In 1995, the yeast product has been registered under the name of Aspire™, which opened the way to test the product under commercial conditions. Aspire alone consistently reduced the level of mold rots caused by *Penicillium digitatum* and *P. italicum* by about 50% when compared with water control. When applied following a pre-wash of SOPP, mold rots were reduced by 90%. Application of Aspire with 50 to 200 ppm TBZ (3-10% of commercial levels) resulted in control levels equivalent to that of commercial treatments. These results will be discussed in relation to the commercialization of the product as a viable biofungicide of postharvest diseases of citrus fruit.

Scientist/Industry partnerships in the commercialization of biologically-based technology for the control of post-harvest diseases

Raphael HOFSTEIN and Richard A. DAOUST

Business Directors, Ecogen Inc., 2005 Cabot Boulevard West, P.O. Box 3023, Langhorne, PA 19047, USA
Fax : (1) 215 757 2956

ASPIRE is a biofungicide which was developed by Ecogen for the control of post-harvest rot disease on a variety of crops and primarily citrus and pome-fruit. Concerted effort made by scientific teams coming from various disciplines allowed Ecogen to develop ASPIRE into a commercially viable product.

Ecogen became actively involved in the development process only after completion of the first phase in which the scientific teams of Wilson and Chalutz developed a screening method to identify the ultimate, naturally occurring, yeast organism and following preliminary efficacy testing of the "active ingredient" on apples, pears and citrus. The second phase addressed the development of fermentation and formulation processes, most of which have been conducted by Ecogen's product development teams. Such a biphasic program has created the most productive atmosphere for an expedited process from insemination of an idea to the final stages of product commercialization.

The main emphasis in our R&D program during the last two years has been placed on the adaptation of ASPIRE to fit the existing regimes of disease control of packing houses in Israel, Europe and the USA. The main conclusions drawn form these tests are that Ecogen has created a product which is user-friendly and is well tailored to be applied onto fruit in the currently existing pome-fruit and citrus packing-lines. The most consistent performance has been attained when the product was applied following a sanitary treatment with any of commonly used disinfectants and in conjunction with low rates of chemical fungicides. For treatment of apples, pears, grapefruit and oranges, ASPIRE has been applied by spraying or by drenching in an aqueous solution and prior to waxing. However, due to compatibility acceptance on lemons, prior to long term storage ASPIRE has been applied within the wax.

The results of our studies clearly indicate that biological fungicides should become a prominent component of Integrated Pest Management programs, which remain the only economically feasible approach for pest control.

Industry's perspective on new biologically-based controls for postharvest diseases

J. D. FROYD

American Cyanamid Company, Princeton, NJ 80543-0400, USA
Fax : (609) 275 3569
E-mail : froyd@pt.cyanamid.com

Agricultural chemical companies do not have a problem in developing and selling biological pesticides along with synthetic pesticides. However, three basic criteria must be met before a company will decide to invest time and money on a biological product. First, is there a market need for this product? For postharvest citrus disease control in the USA, fungal resistance to the major synthetic fungicides is common and compromises fruit quality. Also, some previously effective fungicides are no longer available and no new postharvest fungicides are under development. Second, is the product effective and safe ? The level of disease control provided by the biological agent must be similar to that of existing products or the fruit processor will not use it. The shipper must deliver a sound product to the retailer, who in turn wants to sell good looking, healthy fruit to the consumer. If the shipper, retailer, and/or consumer are dissatisfied with the fruit quality, the fruit processor will use a different product for disease control next time. Third, will the product be profitable ? What are the development costs and potential market size ? Fortunately, registration costs and time in the USA are relatively low for biologicals compared with synthetic pesticides, and the current regulatory climate is favorable for approving new biologicals. However, the market size for postharvest fungicides is not large (less than $40 million in the US), and profits will be small relative to other fungicide markets. A crop protection company might still find this market opportunity attractive because (1) the biological product could serve as an entrant into a new market, (2) controlled fruit storage conditions favor performance of the biological agent, (3) few existing fungicides and none under development minimizes competition, (4) fungicide resistance has created a demand for products with novel modes-of-action, and (5) the lower cost of developing a biological product makes profitability more possible.

Partnerships for technology transfer : scope for international collaboration

Ruxton VILLET

USDA, Agricultural Research Service,
Office of International Research Programs, BARC West,005,
Beltsville, MD 20705, USA
Fax : 301 504 5298

In research program design, a useful criterion for choosing projects and committing investment, is the potential for creating technologies and products which could excite demand in the marketplace. Depending on the mode of technology transfer, royalty streams can be generated for a research laboratory. Biological control technologies and products have attractive features. They are significant components of natural product and green technology portfolios increasingly sought after commercially. The high value, niche-product character can contribute to minimizing investment risk. To spirit such technologies to the marketplace, alliances of various kinds are essential even at early stages of technology creation. Examples are exclusive relationships with a commercial partner and also consortium type structures involving several industrial partners in a broad coalition of interests. Patents, industrial partnerships, patent licenses, for example, are significant components of technology transfer mechanisms. International networks of collaborating scientists offer attractive possibilities for accelerating technology development and transfer. Success can be shared in the form of co-patenting and technology management plans mutually agreed upon. In its selection of international patterns for certain research collaborations, ARS Office of International Research Programs has an explicit policy : joint ventures are designed to be coherent with institutional program goals on both sides, and with a frank emphasis on prospects for technology commercialization to the benefit of both venture partners. This can provide budget efficiencies and build mutual research value in times of funding constraint.

SYMPOSIUM # 13

PRODUCTION OF BIOLOGICAL CONTROL AGENTS IN DEVELOPING REGIONS

Organizer :

Martin J. DOWNES
St Patrick's College, Department of Biology,
Maynooth, Co Kildare, Ireland
Fax : + 353 1 7083845

Biological control in India : from lab to land

T.M. MANJUNATH

Bio-Control Research Laboratories, a division of Pest Control (India) Ltd., P.O. Box 3228, R.T. Nagar, Bangalore 560 032, India
Fax : 0091 80 333 4316

Several major native pests in India have defied chemical control. In the absence of suitable exotic natural enemies for introduction, augmentation of indigenous species, either exclusively or as part of IPM programmes, holds out promise to check such pests. The success of such augmentation programmes depends mainly on the economic production, ready availability, timely application and adequate releases of well-adapted biocontrol agents. Commercial insectaries and other biocontrol centres play a key role in promoting such programmes.

Mass-production techniques for several promising natural enemies and their hosts have been standardised. This has resulted in successfully transferring the biocontrol technology from lab to land in India.

This paper discusses some of the important biocontrol agents that have been well-researched and successfully transferred from lab to land for control of several major insect pests and weeds in India. These include the annual production of about 3,600 million *Trichogramma* spp. for control of Cotton bollworms and Sugarcane borers ; 0.6 million *Cryptolaemus montrouzieri* and other ladybird beetles for control of mealybugs infesting grapevine, coffee, etc.; 5.0 million *Chrysoperla carnea* against soft-bodied sucking pests; 0.5 million larval parasitoids, *Goniozus nephantidis* and *Bracon brevicornis*, against Coconut black-headed caterpillar; and 4.0 million LE nuclear polyhedrosis viruses (NPVs) of *Helicoverpa armigera* and *Spodoptera litura* each for their control.

The status of exotic natural enemies that have been introduced and field-released for control of certain insects pests and also weeds like *Eichhornia, Parthenium*, *Salvinia* and *Chromolaena* is also discussed.

The planning involved in the production and procurement of natural enemies, methods of packing, application techniques and other common problems encountered in transferring the technology from research to practice are also discussed.

Insect rearing methods for biological control efforts in resource-poor areas of Africa

D.E. CONLONG[1] and C.F. MUGOYA[2]

1 : SA Sugar Association Experiment Station, Private Bag X02,
Mount Edgecombe, Natal, 4300, South Africa
Fax : 00 27 31 595406
2 : Uganda National Council for Science and Technology,
P.O. Box 6884, Kampala, Uganda

In general, biological control of insect pest and weeds using beneficial insects in resource poor areas has not been very well supported. The main reason behind this has been a lack of finance for importation of classical biological control agents, quarantine facilities, effective research in resource poor areas and resources such as rearing and research laboratories.
Donor agencies, and commercial and semi-commercial enterprises in a number of African countries have, however, been able to contribute to biological control efforts using beneficial insects by providing some of the resources needed. This has led to biological control becoming a real possibility to control insect pests and weeds in resource-poor areas in numerous countries.
Examples will be provided by the authors of the "spin-offs" of these programmes for resource-poor areas in South Africa, Zambia, Kenya, Malawi, Benin and Nigeria. In addition to conventional insect rearing in laboratories, insect conservation and habitat management ideas will be discussed.

Insect handling and distribution in resource-poor areas

J. BLEHM

Buena Biosystems, Inc., Ventura, California 93007, USA
Fax : 00 1 805 525 6058

In many areas of the developing World, beneficial arthropods are successfully used in pest management programs. The now decade-old term "Appropriate Technology" is quite applicable to handling and distribution of these organisms in Less Developed Countries (LDC).
As a general rule, the substantially lower marginal labor costs and lack of investment capital in LDCs are the major economic factors that shape the mix or materials and methods used in biological control programs. Processing and packaging functions are done by hand when a very simple machine costing a few hundred dollars or readily available material would be used in regions such as Europe or North America. However, resource scarcity can lead to innovations not considered in more economically advanced regions.
My intention is to give an overview of some general concepts, how they may differ between economic regions, and that a more simplistic, labor-intensive system is probably the best solution for the area with a different resource mix. An example would be, why should we make an investment in time/labor saving equipment when a substantial proportion of a local population is under-employed ? There are disturbing examples of centuries-old sustainable agricultural systems being disrupted by the introduction of technological advancements from highly developed countries. We must be cautious that technology transfers from the first World to the third World are "economically viable, environmentally sound and socially just".

Indigenous for the production of biocontrol agents for the control of *Eichhornia crassipes* (Mart) Soms (Water Hyacinth)

K. JAMIL

Biology Division, Indian Institute of Chemical Technology,
Hyderabad 500 007, India

Control of *Eichhornia crassipes* (Mart) Solms, the world's worst aquatic weed has been possible by utilising a combination of biocontrol agents consisting of insects and microbes. The biocontrol agents are very specific to the target weed and their mass multiplication and mass culturing have been possible by the following techniques.

The fungi which exhibited specificity and virulence against *Eichhornia* were:

a) *Alternaria eichhornae*
b) *Fusarium solani*
c) *Cercospora rodmandii*

The production *Alternaria* was possible in both liquid media (Nutrient broth) and solid media (Zapexdox or potato dextrose agar media). Solid state fermentation of *Alternaria* gave very good production of the spores which could be harvested and diluted with water before spraying with sprayers of appropriate nozzles. The mass production of *Fusarium* was possible in liquid media which was diluted in water before spraying. The pH of the growth medium for both the microbes was 5.6 to 5.8 and the temperatures required were 24-27°C. Avoiding culture contamination was an art, and could be carried out with proper autoclaving and careful inoculations. Only experienced personnel could mass produce the cultures for field applications.

Mass multiplication of weevils

Two exotic species from Venezuela were imported from CSIRO, Australia and from Dr. Ted Centre in Florida. These were *Neochetina eichhorniae* and *N. bruchi*. Continuous attention to transfer the plants containing 20-30 eggs of the weevils were monitored and larval mortalities were avoided by rescuing them from rotting plants and re inoculating in the petioles of fresh plants. The plants were inspected for pupal formations in the root before discarding them. These precautions gave us good results and thousands of weevils could be collected from the stock cultures for releases.

The release strategy of the biocontrol of agents and a successful case study of the control of hyacinth will be presented.

Production of entomopathogenic fungi in resource-poor areas

H.E. JENKINS[1], G. HEVIEFO[2], J. LANGEWALD[2], C.J. LOMER[2] and C. PRIOR[1]

1 : IIBC Silwood Park, Ascott, Berkshire SL55 7AT, United Kingdom
Fax : 00 229 350 556
2 : IITA Biological Control Centre for Africa, Cotonou, BP08-0932, Benin

The LUBILOSA (LUtte BIologique contre les LOcusts es SAuteriaux) programme has been developing a low capital cost, high labour input, intermediate technology system for the production of aerial conidia of *Metarhizium flavoviride*. This is a two stage system which utilises locally available substrates throughout and may be employed by minimally equipped in resource poor areas for the production of fungal conidia. In 1995, average output from the LUBILOSA production unit at IITA, Benin, was 2 x 10^{14} conidia/week (sufficient to treat 40 ha at the current application rate of 5 x 10^{12} conidia/ha), maximum production was 80 ha in one week, but could not be sustained over a longer period due to shortage of space.
Key aspects of the LUBILOSA mass production system are stringent quality control standards of the final product and contamination monitoring throughout the procedure to identify the source of contamination if found. In this respect, production of fungi in resource poor areas should not fall below quality standards expected of industrial production.
The building of a new production unit with a projected output of >300 ha/week has now been completed at IITA, Benin, and is in the process of being commissioned. The design of this building, the development of the production system and the integration of quality control standards are presented. The potential of such a low capital cost, high labour input system is discussed with reference to recent economic feasibility studies carried out by D. Swanson in 1995.

Appropriate methods of Insect virus production in resource-poor regions

K.A. JONES, D. GRZYWACZ and A. CHERRY

Natural Resources institute, Central Avenue, Chatham Maritime, Chatham, Kent ME4 4TB, United Kingdom

At present insect virus production has only been achieved on a large-scale using *in vivo* technologies. This is also the most appropriate method for resource-poor regions. There are three production methodologies currently being employed in these regions.

1. Rearing infected larvae in a production plant. This is suitable for host insects that can be easily reared under artificial conditions. The process requires the optimisation of insect rearing and harvesting conditions in order to maximise production and minimise cost. Examples are the production of *Helicoverpa armigera* and *Spodoptera* spp. NPVs in India and Thailand.
2. Collection of infected larvae from the field, which are then centrally processed. This is suitable where large numbers of larvae can be easily located, infected and collected. An example is the collection of NPV-infected *Anticarsia gemmatalis* in Brazil.
3. Self-production by farmers. This is particularly suited to areas where access to outside inputs are limited, or expensive. An example is the distribution of seed inoculum of *Spodoptera exigua* NPV in Brazil.

There is a fourth alternative for *supply* of viral insecticides to resource-poor regions, this is production of the virus in resource-rich areas and export to resource-poor areas. There are a number of situations where this may be the most attractive option, for example large-scale production (and storage) for control of an intermittent migratory pest or where the target pest is of world-wide significance such as the *Heliothis/Helicoverpa* complex.

The choice of production (or supply) technologies is dependent on a number of factors, including host insect (ease of rearing or collection), market size (national and international) and quality control. All the above methodologies can be cost-effective and can produce products that compete in terms of price with conventional chemical pesticides.

Biological control agents, such as entomopathogenic nematodes, and the business of food security in resource-poor areas

M.J. DOWNES[1] and A. RYAN[2]

1 : Department of Biology,
Fax : + 353 1 7083845
2 : Centre for Adult and Community Development,
St Patrick's College, Maynooth, Co Kildare, Ireland

Global food production needs to grow by 75% over the next 30 years (FAO, 1996) : the world's ability to produce enough food is less in question here than its ability to put the food in the hands of those who need it. Food produced by those who need it is in the right place already. Although some estimates are higher, about 30% of potential global food, fiber and feed is widely believed to be lost to pests, disease and weeds (NRI, 1992). The ability to produce of natural enemies for inundative application, and to develop businesses based on this, can be an important element in food security of developing regions.
Biocontrol scientists also offer some robust production technologies that are amenable to use by small or medium scale businesses in resource poor areas. Among these technologies is the production of entomopathogenic nematodes. Such nematodes are currently produced in liquid fermentation, or on solid media (a matrix such as foam plastics soaked in a nutrient broth), or in insect hosts.
This paper will review experience of the last two techniques in the tropics. It will conclude that each of these production systems works, that each can be improved by local R&D aimed at production based on local materials and that suitable local materials can sometimes be of little other economic value. Benefits can include the ability to use locally isolated nematode strains, the avoidance of hard currency costs, reduction of chemical usage, increased environmental and human safety and local small business development with the empowerment of disadvantaged groups.
This form of pest control can be developed as part of an integrated, community managed, pest control strategy. Involving the community in the development itself, can be used to ensure the immediate applicability of the research and the likely long term sustainability of the strategy under local conditions.

Production of *Bacillus thuringiensis* in resource-poor regions

I.O. MORAES

Universidade Estadual Paulista - UNESP, CP 136, DETA/UNESP/SJRP,
13130010 Sao Jose do. Rio Preto, SP, Brazil
Fax : 00 55 172 248692

Bacillus thuringiensis (*Bt*) was used in Brazil for the first time in 1960. In the beginning of the 1970's, studies on *Bt* production by submerged fermentation were initiated. A technological patent was deposited in 1976, relative to the fermentation process. Following these studies it was visualised that in developing countries, mainly in technologically resource-poor areas, it would be better to use semi-solid fermentation using regional raw materials as carbon and nitrogen sources for *Bt* development. Some groups in Brazil, therefore, began to study and develop appropriate technologies to be transferred to the rural or small scale producer of agricultural goods. In 1985, another production process patent was deposited.

This paper will present some case studies of *Bt* development in Brazil for use against Lepidoptera and Diptera.

These papers treat the problems concerned with the large territorial extension of the Country which contribute to the expensive rates of transporting finished products from one State to the other. So, adapted technologies that permit both local production using a minimum of scientific and technological knowledge and the use and training of the local manpower, with the benefits that come from the use of raw material, residues and waste waters rich in carbon, nitrogen, vitamin and mineral salts, are highly desirable.

Another considerable difficulty is the price of the imported products now in use in Brazil. This paper will discuss some research groups that adopt appropriate technology to produce the bioinsecticide of *Bacillus thuringiensis*.

Production and on-farm evaluation of biological control agents in resource-poor areas

M.O. ODINDO, A.M. NOUR, S.O. AJALA, K.G. KIROS, P.O. CHITERE, K. AMPONG'-NYARKO, M.A. MOHAMED ALI and Z.O. NGALO

The International Centre of Insect Physiology and Ecology,
P.O. Box 30772, NAIROBI, Kenya
Fax : 00 254 2 803360

Bacillus thuringiensis var. *kurstaki* was isolated locally, and produced in a simple and readily available medium based on soybean and molasses as the main ingredients. The preparation was evaluated on elite sorghum and maize genotypes in farmers' fields. Farmers were invited to the trial sites at different stages of crop phenology so as to participate in the process of evaluation of the various pest management technologies, including plant resistance, intercropping, and biological control.
The sites where the trials were held had been selected on the basis of their variation in soil type, level and distribution of rainfall, vegetation type, and other physical and climatic factors. The study sites were also selected as points for introduction of sorghum, a hitherto alien crop in the region.
All the plots had a natural infestation of mixed populations of the spotted stem borer *Chilo partellus* and the coastal stem borer *C. orichalcociliellus.* A 2% pathogen suspension was applied directly into the leaf whorl at the rate of 50 l/ha, using a 5 l shoulder-held sprayer. The second strip was treated with a chemical insecticide (Furadan, carbofuran) applied as a 5% granular formulation, at the rate of 9 kg per ha. The third strip was not treated (infested control plots).
Foliar application of B. *thuringiensis* reduced borer infestation on the sorghum and maize genotypes tested in all areas where *C. partellus/C orichalcociliellus* were prevalent. Up to 17.4% increase in yield was recorded in sites of high borer incidence. Proportion of plants showing foliar damage, or deadheart, was 29.4 and 1.2% in *B. thuringiensis*-treated plants compared to 31.5 and 2.1% in the non-sprayed control plots, respectively. Insecticide-treated plots gave higher yield in some sites, although there was no significant difference between insecticide- and pathogen-treated plots in others. *B. thuringiensis* was therefore shown to be a viable alternative for the control of cereal stem borers in the tropics.

SYMPOSIUM # 14

PRIVATE-PUBLIC SECTOR COOPERATION IN THE DEVELOPMENT AND USE OF MASS-PRODUCED MULTICELLULAR ENEMIES

Organizers :

Richard L. RIDGWAY
USDA, ARS, Insect Biocontrol Laboratory, Beltsville,
MD 90705 USA
Fax : (301) 504 9077
E-mail : rridgway@asrr.arsusda.gov

and

J.M. RABASSE
INRA, laboratoire de Biologie des Invertébrés,
Unité de Recherche de Biologie des Populations,
B.P. 2078, 06606 Antibes
Fax : (33) 93 67 89 55

Requirements for development and use of mass-reared natural enemies

John T. TRUMBLE

University of California, Department of Entomology
Riverside, CA 92521, USA
Fax : (909) 787 5624 +33
E-mail : John@citrus.ucr.edu

Intuitively, one would think that the development and use of a natural enemy would be a simple and straightforward process. At a minimum, several important traits would be needed. The natural enemy should (1) provide effective control of a pest, (2) be tolerant of local conditions, (3) be easily and inexpensively reared and (4) provide a measurable cost-benefit to the user. Remarkably, some of the best known commercial natural enemies do not meet any of these conditions. Mass-reared arthropods used in the home gardening industry are particulary likely to fall into this category. However, even in commercial agriculture, cost-effectiveness is not always demonstrated prior to use. The resulting mistakes can be costly. Relatively few studies provide an in depth economic analysis of natural enemy use. Even fewer publications document the economic benefits of integrating beneficial arthropods with chemical and cultural controls. The public sector development of an integrated pest management program for strawberries is an exception. This program integrates mass releases of predaceous mites with pesticide applications for spider mite suppression and provides a detailed economic analysis which documents the strategy yielding the best net profits for the strawberry producer. Although sequential releases of predaceous mites during the season result in a net benefit of up to $4,315 per hectare over the untreated controls, the economic returns from two applications of fenbutatin-oxide ($9,146) or abamectin ($15,802) range from two to three times higher. However, the best monetary profit is generated by abamectin in combination with releases of predaceous mites ($19,705). The benefits of this program are considerable from both the agricultural and societal points of view. Providing unambiguous evidence of the economic advantages is critical for promoting development and use of mass-released natural enemies.

Practical use of predaceous mites to manage Twospotted spider mites on strawberries

Glenn SCRIVEN

Biotactics Inc.
Riverside, CA 92501, USA
Fax : (909) 781 9154

The use of *Phytoseiulus persimilis* in California strawberry fields, to aid in the control of twospotted spider mite, *Tetranychus urticae*, began in earnest in the late 1980s. This was the result of the key miticide, Plictran, being removed from the market. Earlier, university research studies had indicated that *P. persimilis* could be effective ; however, growers were reluctant to commit to a program relying on biological control alone and an integrated pest management (IPM) program for strawberries had not been developed. Large scale use of *P. persimilis* on a California field crop had not been tried before, although, use of *P. persimilis* in protected cropping, especially in Europe, was well established by this time. By trial and error, the growers, with the help of pest control advisors, predator producers and university Extension personnel, developed IPM programs that saved the industry from serious losses. In order for the growers to succeed with an IPM program using *P. persimilis*, it was necessary for them to make a paradigm shift from treating mites when they approached the economic threshold to planning ahead and releasing the predators very early when the mites appeared in the field. Costs of managing spider mites have declined over 50% since the use of *P. persimilis* began in 1988. Grower success in using predators varies considerably. This is primarily due to quality of the predators used, timing of the releases, and the degree of understanding of predator dynamics. Eighty percent of the California strawberry growers now integrate predators into their mite control programs. Some growers have successfully used only *P. persimilis* to control their spider mite problems. The essential elements of a successful IPM program using predaceous mites are use of quality unstressed predators ; field scouting ; proper timing of predator releases ; adequate release numbers based on field counts of the spider mites ; and care in using pesticides that may affect the predators. Not only are growers saving money but they are also having fewer problems with phytotoxicity, soil compaction, worker reentry, pesticide residue concerns, and pesticide permits.

Development and use of an entomopathogenic Nematode with the aid of licensing

James R. RAULSTON[1] and Ramon GEORGIS[2]

1: USDA, ARS,Weslaco, TX 78596, USA
Fax : (210) 565 6133
2 : Biosys, Columbia, MD 21046, USA
Fax : (410) 381 3840

Cooperative Research and Development Agreements (CRADA's) between the federal and private sector provide a mechanism for developing and commercializing technology produced or discovered through federal research efforts. Within the Agriculture Research Service (ARS) of the U.S. Department of Agriculture, the Office of Technology Transfer plays a vital role in forming these agreements. As its name implies, the role of this office is to ensure that technology initiated in the federal sector is made available for use by the public. CRADA's provide licensing opportunities (first right of refusal) to the cooperating private sector entity for products resulting from the cooperative research. This mechanism was employed to develop and commercialize the use of an entomopathogenic nematode, *Steinernema riobravis*, recently discovered in the subtropical Rio Grande Valley of Texas. This nematode was initially observed parasitizing *Helicoverpa zea* and *Spodoptera frugiperda* pre-pupae and pupae in corn fields. Ecological studies showed the nematode to be the most important factor contributing to mortality of these pests in the life stages inhabiting the soil. Application of *in vivo* reared *S. riobravis* to post-silk corn plots resulted in 95-100% mortality of naturally occurring populations of *H. zea*. Concomitant applications of *S. carpocapsae* resulted in no mortality which was attributed to the high soil temperatures (ca. 40°C.). These initial tests indicated the efficacy of *S. riobravis* applied to the soil for controlling *H. zea*. Personnel at biosys Inc., a company with expertise in *in vitro* production and formulation of entomopathogenic nematodes, were contacted to determine their interest in pursuing cooperative research on *S. riobravis*. A CRADA was formed between ARS and biosys Inc. to further the development of *S. riobravis* as a biological control agent. Research and subsequent licensing under the CRADA has currently resulted in development of commercial formulations of *S. riobravis* for control of mole crickets in turf grass and the citrus root weevil complex in citrus.

The future of mass-released natural enemies in glasshouses in Europe

Willem RAVENSBERG

Koppert Biological Systems,
Berkel en Rodenrijis, 2650 AD The Netherlands
Fax : (31) 10 511 21 57
E-mail : W.Ravensberg@Koppert.NL

The future for the use of natural enemies for pest control in greenhouses in Europe is uncertain. While the proliferation of beneficial suppliers in the past decade has provided greater choice to growers, the increased competition also threatens to hinder further success, as diminishing profit margins reduce resources available for both user guidance and research and development of new products. The most significant new element affecting the health of private companies, however, is the increased importance of political factors. These include regulation, registration requirements, bans on non-indigenous species, in-country production rules, ownership issues (biodiversity), decreased public spending on research and extension, and privatization. Although governmental programs to reduce the use of pesticides in agriculture have been successful, they have scarcely increased the use of biological alternatives. The failure of both public policy and the market to encourage biological control has to be recognized and ways have to be found to promote biological control. Cooperation between the public and private sector needs to be strengthened in all possible ways to increase (or even secure) the use of natural enemies. At present the only real incentive for growers to use biological control is the market, but consumers are not paying anything more for integrated pest management grown produce. Well defined research programs have to be developed by both sectors to come to total biological control programs which are recognizable for consumers and can be used in the market.

Development and use of Trichogramma in the field for Insect control in Western Europe

Firouz KABIRI[1] and Franz BIGLER[2]

1 :Biotop, Route de Biot, D4, 06560 Valbonne, France
Fax : (33) 93 12 14 06
2 : Swiss Federal Research Station for Agroecology and Agriculture
Reckenholzstr. 191, Postfach, CH-8046, Zurich, Switzerland
Fax : +41 (0) 1 377 72 01

The European corn borer (ECB), *Ostrinia nubilalis*, is widely distributed in Europe. Its pest status is well documented from the southernmost edges up to approximately the 50th latitude. It has become a new pest in many regions of Central and Western Europe during the last 25 years by following the expansion of the maize crop. The ECB, an important pest in France for decades, only invaded most maize-growing regions of southern Germany and Switzerland after 1970.

Between 1973 and 1976, research programmes for biological control of the ECB with the egg parasitoid, *Trichogramma brassicae*, started in France, Germany and Switzerland. In France, the development of commercial use of *Trichogramma* required about 15 years. During this period, the National Institute for Agricultural Research (INRA), a government institution, investigated the possibilities of setting up large-scale production of the parasitoid and its factitious host, including storage procedures (e.g., diapause of *Trichogramma*). The commercial and industrial aspects were developed by a private partner. The Plant Protection Service, another public organization, participated in field research, especially for the determination of optimal release periods and efficacy experiments. They also supported the private company during the registration procedure of *Trichogramma*. Due to a number of different conditions (structure of farms, price of products, political support, attitude towards environmental concerns) in Switzerland and Germany, the experimental period there lasted only three years. The *Trichogramma* projects got a high priority within federal agriculture research institutions in the two countries, and as in France, cooperation with private companies started at a very early stage. Monitoring of the pest, field efficacy experiments (necessary for registration of *Trichogramma* in Switzerland) and farmers' training were performed by the plant protection services. Release systems were developed in collaboration with private industry, the federal research institutions and the *Trichogramma* production companies.

Two main factors proved to be critical for the acceptance of *Trichogramma* by farmers : (1) the costs of the product and (2) the efficacy. Efficacy is comparable to that of insecticides but costs are still higher than those for insecticides; therefore, *Trichogramma* are subsidised by governments of several German counties. Experience over 20 years has shown that the development of *Trichogramma* for field crop use needs strong support by government institutions and close cooperation between public and private bodies.

Successful transference of technology in biological control in sugarcane and corn crops in Venezuela

Francisco FERRER

Servicio Biologico, C.A. Barquisimeto, Venezuela
Fax : (58-51) 316253-547486

Two examples of technology transfer-biological control of the borers, *Diatraea* spp., in sugarcane and of the fall armyworm, *Spodoptera frugiperda*, in corn - were developed in Venezuela. Since 1975, the Asian braconid, *Cotesia flavipes*, has been introduced several times for control of *Diatraea* spp. on sugarcane, with effectiveness first demonstrated in 1987. From 1988 to 1994 after a program of releases of *C. flavipes* in nearly 30,000 ha, parasitism increased in the area of Rio Turbio (Estado Lara) from 5.8 to 29.7%. This was higher than that provided by *Metagonistylum minense*, which had traditionally been used since 1952. A complementary effect was demonstrated, with total parasitism of over 34% and a reduction in infestation from about 75% to 2.5%. At the present, *C. flavipes* is widespread in Venezuela, with a contribution to general parasitism of 90%. The reduction in the infestation resulted in a cost-benefit ratio of 1:20.

Corn is one of the most important crops in Venezuela and is the basis of the population's diet. However, this crop is seriously affected by *S. frugiperda*, which is the main factor in losses in yield. In the past, control of this pest has been achieved mainly with chemicals, which recently have become less effective because of resistance and the destruction of natural enemies. These circumstances increased costs and drove the farmers to seek new alternatives. An integrated pest management program (IPM) was begun with pest monitoring and releases of the scelionid, *Telenomus remus*, which was introduced in 1979 from the Commonwealth Institute of Biological Control in Trinidad. Starting in 1979, techniques were developed for its mass production in *S. frugiperda* eggs, but it was not shown to be effective for controlling this pest until 1987 in a demonstration area. Although 90% parasitism was observed after the 6th week, acceptance by the farmers was slow. In 1989, positive results were again demonstrated, and the use of this parasitoid increased by 1991 in four states of Venezuela : Lara, Yaracuy, Portuguesa and Guáric, with 5 million *T. remus* released on about 900 ha, with promising results. For 1992, optimistic plans were made to expand the treatment to 5,000 ha; however, only 600 ha were available, due to economic conditions such as lack of credits, high interest rates, and low crop prices. However, from 1994 to 1995 under a better strategy, the area of the IPM program was increased to nearly 4,000 ha, and for 1996 it is proposed to reach nearly 6,000 ha. Success now has been demonstrated with 50% savings in use of pesticides.

Augmentative biological control of Mealybugs on Citrus : the Israeli experience

Shimon STEINBERG[1], Zvi MENDEL[2] and Shmuel GROSS[3]

1 : Biological Control Industries, Sede Eliyahu,
M.P. Beit Shean 10810, Israel
Fax : (972) 6 580908
2 : Department of Entomology, ARO, Volcani Center,
Beit Dagan 50250, Israel
3 : Division of Plant Protection, Ministry of Agriculture,
Extension Service, Israel

Mealybugs, particularly the citrus mealybug, *Planococcus citri*, have become important pests of Israeli citrus in the past few years. Since IPM has been used in Israeli citrus for years, there was an incentive to try non-chemical alternatives to control mealybugs. Thus, seasonal inoculative releases of citrus mealybug natural enemies have been carried out since 1993. The natural enemy complex is comprised of the exotic parasitoid, *Leptomastix dactylopii*, the indigenous parasitoid, *Anagyrus pseudococci*, (applied since 1996), and the predatory lady beetle, *Cryptolaemus montrouzieri*. Three major bodies, from the public and private sectors, are involved in the project : research, extension service and industry. Typically, these bodies interact among themselves and with the growers. The research develops monitoring tools for the citrus mealybug and methods for estimating the recovery of the mealybug's natural enemies and establishes the rate of mealybug control. This is done in 10–15 study plots (pairs of release vs. control plots, ca. one hectare each, 0.5–1.5 km distant from each other) scattered in the major citrus growing regions in the country. Extension serves as the link to the growers by organizing training sessions where all the updated information from research and industry is presented. Furthermore, extension provides technical assistance to the research by : (1) choosing the appropriate growers and plots for the study and (2) being involved in sampling of field material. Industry mass produces the mealybug's natural enemies, supplies live material back-up for research, develops shipment and release methods and screens the effect of pesticides on the natural enemies. Parallel to the research, industry offers biological control of the citrus mealybug on a semi-commercial scale for 100–150 hectares of citrus. This year is the last season for research. We expect a breakthrough in augmentative biological control of the citrus mealybug, as timing of release and dosage of parasitoids has been optimized. We foresee that from now on, extension and industry will lead the project on a fully commercial scale, for the benefit of all.

Opportunities for increased cooperation between the private and the public sectors

Richard L. RIDGWAY and May N. INSCOE

USDA, ARS, Insect Biocontrol Laboratory, Beltsville, MD, 20705, USA
Fax : (301) 504 9077
E-mail : rridgway@asrr.arsusda.gov

Alternatives to conventional insecticides are needed to replace those that are being lost, if an abundant, safe, and affordable food supply is to be ensured for generations to come. Several groups of biological products could contribute significantly to the range of alternatives that may be needed; one such group is the mass-reared multicellular natural enemies (mostly arthropods and nematodes). This group made up about 15% of the estimated $340 million world market for biologically based insect control products in 1991. Over eighty species of arthropods and nematodes were reported to be available for purchase in North America, and there is considerable potential to expand the use of mass-reared natural enemies. However, the characteristics of these organisms, including (1) narrow spectrum of activity, (2) short shelf life, (3) relatively small market size, (4) limited opportunities for proprietary protection, and (5) relatively high costs of production, suggest that institutions differing from those that have developed and delivered conventional insecticides are needed. A framework for a total discovery, development, and delivery process that can be applied to biological products is presented. The review of four case studies documents economically viable pest control in North America with mass-reared natural enemies. These studies, plus the many documented potential uses of mass-reared natural enemies, point to the merits of expanded coordinated efforts to create increased inquiry into the role of mass-reared natural enemies in insect management. Such inquiry, by both the public and private sectors, is needed to identify gaps and those responsible for filling those gaps and to jointly plan specific actions for filling the gaps, if the natural enemy industry is to flourish and to contribute in a substantial way to the future of insect management.

Future strategies for the private sector

Melvin FIDGETT

Ciba Bunting, New Road, Aldham, Colchester,
Essex CO63PN, United Kingdom
Fax : (44) 1206 243209

The use of natural enemies in crop protection plays a significant role in glasshouse crops and some field crops, but their value compared to the total crop protection market worldwide is insignificant at less than 0.2%. Their use has been limited to glasshouse crops or some niche field crops where no viable chemical solution is available for registration or resistance reasons. Changes towards sustainable agriculture through "Integrated Crop Management", however, are increasing the interest and demand for use of biological control methods in crop protection. Despite these developments, the use of natural enemies on new crops in new regions has stagnated. As producers of beneficial arthropods compete for market share and sales volume we are only seeing marginal changes in existing markets. New alternatives are rushed to the market, very often with little testing and of poor quality, looking for this season's advantage. An over-supplied and unprofitable market has resulted which is not generating the funds required for major research and development projects.

If natural enemies are to become a more important element of crop protection, worldwide fundamental changes will need to occur. Development focus should be moved away from solely new species towards field development. In the harsher environment outside the greenhouse, crop programmes which utilize chemical treatments, naturally occurring and introduced natural enemies, other biological products and cultural methods will need to be developed. Recommendations for use of natural enemies will need to be more robust. This means higher introduction rates at lower cost and higher quality products. To achieve this, new factory-based production methods are required. Mechanical application methods will also be a key success factor where labor is limited. The huge investment required to develop complex crop programs presents a dilemma as the products have limited, if any, patent protection. Technologies which can be patented or protected through registration, industry quality standards, and marketing contracts with customers will be needed so that the industry can generate the required return to fund these developments.

The competition for natural enemies remains low-cost chemicals, transgenic crops and other biological solutions. Natural enemies will have to be priced to compete with these products if they are to become a major element in crop protection programmes. With today's production and application techniques, they will always remain a fringe niche player.

SYMPOSIUM #15

BIOLOGICAL CONTROL OF *BEMISIA*

Organizers :

W.A. JONES
USDA, ARS, Southern Plains Area,
Biological Control of Pest Research Unit, 2413 E. Hwy 83,
Weslaco, Texas 78596, USA
Fax : 210 969 4888
E-mail : w-jones@pop.tamu.edu

and

M. BENUZZI
BIOLAB, Centrale Ortofrutticole, Via Masiere 1,
47023 Cesena (FORLI), Italy
Fax : 0547 380 637

Use of Mycotrol® (*Beauveria bassiana*) for control of *Bemisia argentifolii* (Homoptera : Aleyrodidae) in field crops

S.P. WRAIGHT[1] and C.A. BRADLEY[2]

1 : USDA, ARS, Subtropical Agricultural Research Laboratory (SARL), Weslaco, Texas 78596, USA
Fax : 210 969 4888 E-mail : swraight@pop.tamu.edu
2 : Mycotech Corporation, 630 Utah, Butte, Montana 59702, USA

Research on the use of the fungus *Beauveria bassiana* for control of *Bemisia argentifolii* Bellows & Perring (= *B. tabaci* strain B) was initiated by USDA-ARS-SARL in 1990. Initial laboratory and field tests indicated considerable biological control potential, and a collaborative agreement was established with Mycotech Corporation for mycoinsecticide research and development in 1993. In small-scale field trials in cantaloupe, cucumbers, and tomatoes, approximately five applications of *B. bassiana* made at 4-5 d intervals at a rate of 1-2 x 10^{13} conidia $acre^{-1}$ using portable air-assist and hydraulic sprayers produced 80-95% control of immature whiteflies. Aqueous suspensions of the hydrophobic conidia were readily prepared using organosilicone surfactants at 0.03-0.04%. On the basis of these positive results, Mycotech submitted a wettable powder formulation of *B. bassiana* strain GHA (trade named Mycotrol® WP) to the U.S. Environmental Protection Agency in 1995, and registration was granted in March 1996. Mycotech is now building a production facility in Butte, MT; current small-scale production is supporting field trials in the southern USA and Mexico. Tests employing conventional tractor driven spray equipment identified spray coverage as the most important predictor of efficacy. Weekly applications producing deposits of 1200 versus 200 spores per mm^2 on the lower surfaces of melon leaves resulted in 66 and 20% infection, respectively. Studies are in progress to maximize coverage through manipulation of various spray parameters. Using hydraulic sprayers, best coverage of cucurbit foliage was achieved by spacing nozzles 20 cm apart, lowering them to within a few cm of the crop canopy, directing them at a 45° angle to the ground, and increasing pressure to 28 kg cm^{-1}. Using ceramic, hollow-cone nozzles (capacity 820 ml min^{-1} at 28 kg cm^{-1}) and applying 0.5 lb Mycotrol $acre^{-1}$ (1 x 10^{13} spores $acre^{-1}$) this spray configuration produces deposits of between 1000 and 2000 spores per mm^2 on the lower surfaces of melon foliage and reduces larval whitefly numbers by 70%.

Progress in the use of *Delphastus pusillus* and *Paecilomyces fumosoroseus* for managing *Bemisia* (Homoptera : Aleyrodidae) in ornamentals

L. S. OSBORNE

University of Florida, Institute of Food and Agricultural Sciences,
Research and Education Center, 2807 binion Road, Apopka, FL 32702, USA
Fax : 904 392 9359
E-mail : lso@gnv.ifas.ufl.edu

The progess in the development of *Paecilomyces fumosoroseus* (PFR) has far surpassed the use of *Delphastus pusillus*. Problems associated with using *D. pusillus* are similar to those for any other predator. Pesticide sensitivity is a critical factor and the need for relatively high prey densities has severely limited the use of *D. pusillus* in many ornamental greehouses. Secondly, a problem with the identification has resulted in some confusion about what we are actually dealing with. It is currently unclear which species of *Delphastus* we are working with. It appears that the species we have been evaluating are *D. catalinae* and not *D.pusillus*. PFR has also experienced a number of problems, the most important being its lack of registration in Europe and the United States. The most progress we have made with PFR is in the use of it during specific windows in the production of ornamental plants. Ornamental foliage plant production can be divided into distinct phases. The first phase is the production of new plants. These new plant come from either tissue culture, stock plants and, to a very limited extent, seeds. During this phase the humidity is often manipulated. High relative humidity is needed to acclimatize plants that have been removed from tissue culture vials or to enable cuttings taken from stock plants to survive while they develop roots. Controlling these pests during the rooting or propagation phase has had mixed results. It appears the PFR might be limited by the constant presence of free moisture. During the production phase, the humidity is again manipulated in order to reduce plant pathogenic fungi and bacteria. The reduction in humidity does impact PFR but it does not preclude its use. A number of trials have demonstrated that this fungus can be utilized as part of an overall IPM program for whiteflies attacking ornamental plants. During the final phase in the production cycle, finished plants are packaged and shippes. During the period there is another window within which the humidity reaches levels that favor the development of insect pathogenic fungi. We have made significant progress utilizing PFR to manage both whiteflies and mites during shipping. Levels of control have exceeded 90% for both *Tetranychus urticae* and *Bemisia argentifolii*.

The selection, evaluation and release of an *Eretmocerus* sp. (Hymenoptera : Aphelinidae) from Spain for biological control of *Bemisia argentifolii* (Homoptera : Aleyrodidae) in the USA

A.A. KIRK[1], M.A. CIOMPERLIK[2], J.A. GOOLSBY[2],
D. C. VACEK[2], L.E. WENDEL[2], W.A. JONES[3] and L.A. LACEY[1]

1 : USDA, ARS, European Biological Control Laboratory,
Parc Scientifique Agropolis, 34397 Montpellier cedex 5, France
Fax : (33) 67 04 56 20 E-mail : EBCL@CIRAD.FR
2 : USDA, APHIS, PPQ, Mission Biocontrol Center, P.O. Box 2140,
Mission, TX 78873-2140, USA
3 : USDA, ARS, Subtropical Agricultural Research Laboratory,
1413, Hwy. 83, Weslaco, TX 78596 USA

The USDA system for introducing safe effective biological control organisms from the field to release is discussed using the example of an *Eretmocerus* sp. collected in Spain for the biocontrol of *Bemisia argentifolii.* Since 1991 the silver leaf whitefly (SLWF) has caused annual losses in excess of $500 million to vegetable crops, melons, cotton and ornamentals in the USA. Foreign exploration for *Bemisia* natural enemies has been carried out in 25 countries including SE Spain. The hot dry climate and heavily treated cotton and vegetable crops are similar to parts of the southern USA where *Bemisia* is a pest. Cotton leaves infested by parasitized *Bemisia* were sent to the APHIS Biocontrol Center quarantine at Mission Texas in November 1991. Cultures of the emerging *Eretmocerus* sp. were started on *Bemisia* infested *Hibiscus* sp. plants. The population was identified using the genetic diagnostic method RAPD-PCR and given a unique number (*Eretmocerus* nr. *mundus* M92014 - Spain). Evaluation in both the laboratory and in the Lower Rio Grande Valley on cantaloupe melons, kale, and broccoli showed it parasitized more SLWF than the native *Eretmocerus* spp. and other exotic parasitoids tested. It has been recovered from SLWF infesting numerous crops, ornamental and weedy hosts at several release sites through 14-96. RAPD-PCR is used to identify and distinguish *Eretmocerus* sp. M92014 from native and other exotic species of *Eretmocerus* In addition evaluation experiments showed that M92014 was more tolerant of selected pesticides than native U.S. *Eretmocerus* spp. On the basis of these results mass rearing of this species for area wide field release is recommended.

The efficacy of indigenous parasitoids in the biological control of biotype "B" of *Bemisia tabaci* (Homoptera : Aleyrodidae)

J.C. ONILLON and P. MAIGNET

INRA, Laboratoire de Biologie des Invertébrés, U.R. sur les Parasitoides d'Aleurodes, B.P. 2078, 06606 Antibes, France
Fax : (33) 93 67 89 55

Surveys carried out in several localities in Provence and the Cote d'Azur have shown that a fairly diverse parasitoid fauna can develop from the larvae of Biotype "B" of *Bemisia tabaci*. Three species have been collected, *Encarsia pergandiella*, *Encarsia hispida* and *Eretmocerus mundus.* These three species have been tested as well as the 2 *Encarsia* spp., *E. formosa* and *E. tricolor*, which have shown their efficacity against *Trialeurodes vaporariorum*. *E. tricolor* showed no affinity to *B. tabaci* larvae. Used at a rate of 3/1 parasitism/predation did not exceed 13.8%. *E. formosa* exhibited variable efficacy. A rate of 3/1 and reared for 20 generations on *T. vaporariorum*, resulted in 16.5% parasitism ; reared on *B. tabaci* and used at the same ratio, it gave 43% parasitism. *E. pergandiella* showed a strong affinity to larvae of *B. tabaci.* Used at a ratio of 3/1 like the other species, it gave 55-60% parasitism in the first generation. In the second generation, this level remained constant but 30% of the *B. tabaci* larvae were hyperparasitized. This hyperparasitism was produced at the expense of itself as well as that of other species. *E. hispida*, tested at the ratio of 1/1 gave 40% parasitism in the first generation without showing hyperparasitism. A very large variability is nevertheless observed in the level of efficacy of this species. Lastly, *Eretmocerus mundus* showed high efficacy in the parasitism of whitefly larvae. Used at the rate of 1/1, this species gave 80% parasitism as early as the second generation.

A user-controlled system for storage of Lacewing eggs

Y.Q. FAN, F.L. PETITT and D.O WIETLISBACH

The Land, Epcot, Walt Disney World, Lake Buena Vista, FL, USA
Fax : 407 560 6793
E-mail : fred_petitt@wda.disney.com

Lacewing (*Chrysoperla rufilabris* Burmeister) egg eclosion was evaluated after mechanical egg harvesting vs. sodium hypochlorite harvesting. The average survival to first instar from six shipments was significantly higher for mechanically harvested as compared to sodium hypochlorite-harvested eggs. Eclosion of sodium hypochlorite-harvested eggs was more variable across shipments than for mechanically-harvested eggs. Although survival of *C. rufilabris* eggs did not diminish with up to 9 d. of storage in an insectary (approx. 10°C), egg development continued during storage. From the perspective of a user who is trying to time egg applications so that egg eclosion occurs just after application to plant foliage, the inability to predict the time of eclosion creates numerous problems. In this study, a storage system was developed to enable the user to control the timing of egg eclosion. Lacewing eggs oviposited within a 24 h-interval were harvested and shipped about 24 h later. Upon arrival through overnight delivery, the eggs were taken out of the shipping box and allowed to develop at 25°C to a stage about 12-24 h before hatching. These "ready-to-hatch eggs" were stored at 7-9°C for up to 10 d. without negative impact on their survival. Within this period, the eggs can be released any time and will hatch within 12-24 h after the release. Thus, the user can predict the hatching time of these eggs and delay their hatching if conditions for the release are not favorable. This storage system also allows the user to reduce the number of orders required, thereby significantly reducing shipping costs.

Current situation of *Bemisia tabaci* (Genn.) in Spain study on its native parasitoids

F. BEITIA[1], A. ADAN[1], J.R. ESTEBAN[1], P. GUIRAO[2] and J.L. CENIS[2]

1 : C.I.T.-I.N.I.A., Protección Vegetal, Carretera de La Coruña, Km. 7, 28040 Madrid, Spain
E-mail : BEITIA@CIT.inia.es
2 : C.I.D.A., Protección Vegetal, Estación Sericícola, 30150 La Alberca (Murcia), Spain

Bemisia tabaci (Gennadius, 1889) is an insect well known in Spain since 1943, when it was described for the first time by Gómez-Menor. However, it has not been considered as an important pest till last ten years, when the insect was detected producing several damages to greenhouse crops.
As in other regions of the world, this situation has been imputed to the presence of a new biotype of the species, the B type, which is easily recognized by producing "silverleaf" symptoms on squash leaves. Recently, this biotype has been described as a new species, *Bemisia argentifolii* Bellows & Perring, n. sp.
In Spain, we have considered very important to know the status of *B. tabaci*, before starting the study of its native parasitoids in order to use them for the biological control of the pest. Actually, we have verified the presence of, at least, two different biotypes of this whitefly (a B type and a non B type) by using several procedures : the production of "silverleaf" on squash, the RAPD-PCR technique and the biological development of individuals, principally. Now, we are continuing this study in order to know the distribution of the two biotypes in Spain.
On the other hand, we have just started the study on the development of several parasitoid species on the two biotypes, with the aim to know if that parasitism is different depending on the whitefly biotype. At the moment, we are working with two species:

Current programs on the biological control based management of *Bemisia tabaci* by the USDA APHIS in the USA

L.E. WENDEL, M.A. CIOMPERLIK and J.A. GOOLSBY

USDA, APHIS, PPQ, Mission Biological Control Center, P.O. Box 2140, Bldg. 6410, Moore Air Field, Mission, TX 78572, USA
Fax : 210 580 7300
E-mail : a3481wendel@attmail.com

The discovery and implementation of biological control based technologies to manage *Bemisia tabaci* are essential for the long-term solution for producers. The Lower Rio Grande Valley (LRGV) of Texas is a very productive vegetable, fiber and grain producing area that provides favorable hosts for *B. tabaci* the majority of the months throughout the year. A 20 acre demonstration farm representing the major crops grown throughout the year in the LRGV has been initiated. This provides an opportunity to investigate various management strategies for *Bemisia* under field conditions. Parasitoids of *B. tabaci* have been screened on the primary hosts grown in the area. A system of field cages is being utilized to mass produce the most promising parasitoids. Releases of parasitoids are timed to coincide and enhance other management practices currently used by producers. Costs of production, yield and quality information are being collected to quantify the economics associated with technology prior to transfer to the growers.

Using predator gut content immunoassays to expedite the search for indigenous, foreign, and augmented natural enemies of *Bemisia tabaci* (Homoptera : Aleyrodidae)

James R. HAGLER

USDA-ARS, Western Cotton Research Laboratory,
4135 East Broadway Road, Phoenix, Arizona, 85040, USA
Fax : 602 379 4509
E-mail : haglerj@primenet.com

A pest-specific monoclonal antibody (Mab) and an indirect enzyme-linked immunosorbent assay (ELISA) have been developed to detect sweetpotato whitefly, *Bemisia tabaci* (Gennadius) egg remains in the guts of predaceous arthropods. This Mab has been used to identify key indigenous predators of the sweetpotato whitefly and to monitor the efficacy of an augmentative predator release targeted for this pest. The gut contents of over 10,000 individual predators indigenous to Arizona cotton fields were examined for the presence of whitefly egg antigens by ELISA. Four predator species with frequent positive assays for whitefly egg remains were (% positive) : *Collops vittatus* (Say) (55%), *Geocoris* spp. nymphs (39%), *Orius tristicolor* (White) (38%), and *Hippodamia convergens* Guerin-Meneville (33%). A novel immunomarking ELISA that is superior to DayGlo dust marking and elemental marking has also been developed. This immunomarking technique can be used in concert with our predator gut content ELISA. The efficacy of this procedure was evaluated using commercially-reared *H. convergens*. Commercially-reared predators were labelled with either rabbit or chicken IgG. Those labelled with chicken IgG were released into a cantaloupe field and those labelled rabbit IgG were released into an adjacent cotton field. Both fields were vacuum sampled several days after release and the all of the *H. convergens* collected were assayed for (1) the presence of whitefly egg antigen in their gut, (2) the presence of the rabbit IgG label, and (3) the presence of the chicken IgG label. Results from this study showed that these immunoassay procedures can be very useful for identifying key predators of the whitefly egg stage and for monitoring the dispersal characteristics of augmented predator populations. Gut content immunoassays in combination with immunomarking allow researchers to simultaneously monitor the dispersal patterns of augmented predator releases (will they remain at the target site ?), determine the impact of an augmentative predator release on *B. tabaci* (can they reduce *B. tabaci* populations ?), and compare the prey profiles of commercially-reared predators with their wild counterparts (do augmented predators feed on

B. tabaci as frequently as their indigenous counterparts ?). The ELISA techniques described here offer a precise, rapid, economical, and sensitive method for assaying large numbers of predators (*i.e.*, 1,000 per day) without disrupting normal feeding behavior. This system eliminates tedious and time-consuming direct observations. Moreover, the technology described here can expedite both the search for key indigenous predator species and the monitoring of augmentative biological control agents.

Technology transfer and the use of parasitoids in managing *Bemisia* (Homoptera : Aleyrodidae) in desert field crops

K.A. HOELMER[1] and G.S. SIMMONS[2]

1 : USDA, APHIS, PPQ, Phoenix Methods Development Center, Phoenix, AZ, USA
Fax : 619 344 7951 E-mail : hoelmer@mail.sdsu.edu
2 : Western Region, Irrigated Desert Research Station, Brawley CA, USA

Outbreaks of silverleaf whitefly in southwestern U.S. deserts, where susceptible crops are in continuous production, have caused substantial crop losses and losses in sales and employment. In these areas, whitefly populations increase rapidly in spring melons, then move into cotton and alfalfa. Seedling cole crops are impacted in the fall. Winter temperatures limit the whitefly's development rate, and populations are greatly reduced by spring when melons germinate. Where melons are not grown, heavy infestations occur later during the summer. Desert plants, agricultural weeds and many hosts in urban areas also harbor whiteflies, although populations increase only after migrations from nearby fields when crops are harvested. Urban populations are valuable reservoirs for natural enemies. Naturally-occurring biocontrol by indigenous species is inadequate at key points in the whitefly population cycle. Greater activity of enemies in melons is essential during spring, when whiteflies increase in abundance. Large numbers of inexpensive enemies are needed for releases in commercial field crops to be feasible. Several non-indigenous parasitoids of *Bemisia* cultured in the U.S. show promise for improving whitefly management in desert regions (e.g. *Eretmocerus* species or geographic races from Spain, India and Israel). High levels of parasitism by two of these were achieved in spring melons with augmentative releases in 1994 and 1995. Within-field reproduction also multiplies parasitoids in the field, increasing the likelihood of permanent establishment. Two years of operation has shown that local mass-rearing with parent material obtained from different sources is feasible and can be used to support augmentative, inoculative and other research projects. Multiple agencies cooperated in this effort; individual growers have also expressed interest in establishing their own insectaries. Other approaches include transplanting seedlings inoculated with parasitoids, redistribution of field-reared parasitoids, and promotion of conservation refuges. Growers are becoming more receptive to the use of natural enemies. The prospects for integrating parasitoids with selective pesticides, such as imidacloprid and buprofezin, are good. However, specific information on using parasitoids is lacking and remains a research priority.

SYMPOSIUM #16

PROBLEMS IN THE PRODUCTION, SALES AND DISTRIBUTION OF BIOLOGICAL CONTROL AGENTS

Organizers :

Richard A. DAOUST
Ecogen Inc., 2005 Cabot blvd. West, Langhorne, PA 19047, USA
Fax : 215 757 2956

and

Jean-Paul AESCHLIMANN
c/o CSIRO, Campus international de Montferrier-Baillarguet,
34980 Montferrier-sur-Lez, France
Fax : (33) 67 59 31 22

Problems in the production, sales and distribution of pheromone-based control products for insect control

Guy D. LAUDE

Biosytèmes France SARL, Parc d'activités des Bellevues, B.P. 227,
Eragny, 95614 Cergy Pontoise Cedex, France
Fax : (33) 34 48 99 27

The insect control technology using pheromone-based control products, has been developping graduallyy over the past eight years. However its development is facing problems that have limited the use of these safe and effective pest control agents. Difficulties met in the transfer of these products from the research laboratory to their commercial use are discussed :

* Problems in the production :

The manufacturing of large volumes of pheromone active ingredients is raising many particular problems due to the special chemistry of the pheromones (undesirable by products, related compounds with possible adverse effect, purity and quality control for large volumes, volatility and stability of the compound produced, storage ability, complicated production process involved). At the industrial level, the pheromone production remains very small in volume when compared to the production of insecticides. High investments for sythesis equipments and moderate quantities involved lead to increased costs ans reduced number of interested manufacturers. The same concern applies to the pheromone carrying dispensers (cost for research and production of easy-to-use long term dispersers with improved field performance is of basic importace). The technology developped for the dispenser may be subject to better patent protection.

* Problems in the sales and distribution :

Contrary to most of the insecticides, ready-to-use pheromone blends for mating disruption are used against one insect pest only, strongly limiting the potential markets of such a product. When several pests are met, the need for more and deeper crop inspections becomes obvious and the necessary corresponding highly qualified technical assistance devoted to each crop/field environment becomes an expensive must for distributors, resellers and end-users. As far as registration is concerned, not all countries are giving the same consideration to pheromone based products. In some cases, registration is not needed. In general, the toxicology studies requested are the same as for any insecticide. The commercial use of these products may face difficulties in finding good crop environment conditions for best results (large and even crop areas). It is hoped that progresses particularly at the dispenser level will be made in the future to solve some of the above problems affecting the development of pheromones.

Poblems in the development and commercialization of *Bt*-based products and other biocontrol agents in Europe

Alessandro NATALI

Intrachem (International) SA, 34 Quai de Cologny,
1223 Genève, Switzerland

The introduction of biocontrol agents in Europe must face several difficult problems. The first concerns the development of the product in the different European climatic conditions that can greatly influence the efficacy of biological products. The second big problem regards the registration of the microbial agents, in particular the cost of the required toxicological data, often unjustified on a scientific point of view, and the lack of guidelines specific for biological products for the official efficacy trials, that very often strongly penalise them in the comparison with the chemical pesticides. Finally, the constraints to the commercialization of biocontrol agents regard the short shelf-life of the products, the necessity to deal with well technical prepared distributors and farmers, the difficulty to introduce them in a global integrated pest management system, and the cost of the products, generally more expensive than chemical competitors.
Different examples of these difficulties taken from the practical experience in various European countries will be discussed.

Problems in introduction and marketing of Nematodes for control of insects and slugs

Graeme GOWLING

Microbio Limited, Dales Manor Business Park, Babraham Road, Sawston, Cambridge, CB2 4LJ, United Kingdom
Fax : (01223) 830861

Anyone who has worked with parasitic nematodes will be only too aware of the difficulties of producing and marketing them.
Fermentation of any multicellular organism is still at the leading edge of technology. The process is further complicated by the fact that we are fermenting a bacterial species in the same medium and vessel, at the same time. Solid state production is still being done commercially but is prohibitive in terms of labour costs.
Just as crucial to economically viable yields from the fermenters in the downstream processing : washing and extracting from the spent medium. Losses at this stage can easily be 40 % of fermenter yield.
Formulation in a clean, stable carrier is the next stage. Inert, absorptive, clay-based materials offer the simplest opportunities for stable formulations and ease of use. However, these products do require refrigeration and are prone to contaminating fungi and bacteria unless the nematode washing process is extremely thorough.
Packaging needs to be moisture retentive but gas permeable, allowing outflow of CO_2 and inflow of O_2.
The marketing of nematode-based products too is fairly specialized. They have to compete in the overall crop-protection market which means, at current production capacities and costs, they are largely restricted to riche markets in the horticultural sector- chiefly protected crops and gardens.
Distribution and delivery systems need to be tailored to the restricted shelf-life and requirement for low temperature storage. Finally, and most importantly, the products need close attention in terms of promotion and after sales servicing. There are only a limited number of companies able to deliver this service and, in our experience, these are not the Agrochemical majors.
Over-optimistic label claims have engendered a reputation of low efficiency in some markets. Price erosion too has been an added disincentive to the manufacturers.

Opportunities and challenges in the production, sale and distribution of Spo-X GH, a viral insecticide for control of Beet armyworm in greenhouses

I.A. SCHONHERR[1], S.A. COLLINS[2] and M.S. DIMOCK[3]

1 : Brinkman B.V., P. O. Box 2,
2690 AA' s-Gravenzande, The Netherlands
Fax : 0174 411280
2 : Agrisense BCS Limited, Treforest Industrial Estate, Pontypridd,
Mid Glamorgan, CF37 5SU, United Kingdom
3 : Biosys, Inc., 10150 Old Columbia Road, Columbia, MD 21046, USA

Spod-X GH was the first insect virus to be registered for insect control in the Netherlands in 1993. The active ingredient of Spod-X GH is the *Spodoptera exigua* nuclear polyhedrosis virus, which is highly specific for Beet armyworm, *Spodoptera exigua*. In the Netherlands Beet armyworm is an important pest in greenhouse crops, including a range of cutflowers and potted plants, and sweet pepper.
Spod-X is produced in living insects on artificial diet (*in vivo*). *In vivo* production involves mass-rearing of high-quality insects, timely inoculation, and maintenance of continued insect growth to maximize virus replication and bioactivity. To prevent contamination, strict procedures are followed to insure healthy and disease-free *S. exigua* colonies.
Variability in the product contents and microbial contamination are constraints in the production. Quality assurance tests are performed to check ingredient limits and to insure that no toxic contamination will occur in the end product. To insure high quality of the product in the hands of the user, the product is labeled with an expirationd date and storage under cool, dark conditions is recommended.
Spod-X GH sale in the Netherlands exceeded all expectations. Key causes were the growing Beet armyworm problem, the absence of effective chemicals, and warm summers in 1994 and 1995. The non-toxicity to beneficial insects is an advantage for growers that use biological control.
Education of growers appeared to be important.
The biology of the pest, the relatively long time before an effect is visible and the need for early applications require from the grower some patience, pest monitoring and determination, and good spraying techniques.
We are pursuing EC registration of Spod-X GH under guideline 91/414 in order to maintain registration in the Netherlands, expand the label to vegetables, and to obtain registration in other EC contries.

Industrially acceptable production of biopesticides preparations containing air-borne spores of fungi

Miloslav NESRSTA and Jan KYBAL

FYTOVITA Co.Ltd., facilities Blatnice 855,
696 71 Blatnice, Czech Republic
Fax : +42 631 331539

The mycopesticide preparation BOVEROL - containing an active agent of air-spores of the entomopathogenous fungus *Beauveria bassiana* and SUPRESIVIT - containing an active agent of air- spores of the antagonistic fungus *Trichoderma harzianum* were officially registered in the Czech Republic.
Both of the preparations are dispersable powders containing $1\text{-}3x10^{10}$ spores/g when 70% of these spores must be germinable. The products contains no toxins.
The preparations are produced by Czech original technology of surface fermentation in plastics bags. In this conditions fungus grown on the surface of thin layer of liquid cultivation medium, creates conidiophores that form spores into the air environment, as in natural environment. By constitution of cultivation medium we limit growth of mycelium and stimulate production of spores. During finalisation, the cultivation medium is separated so the final product doesn't contain any dead weight material, only spores and residues of mycelium. For standardisation inert material (active silicondioxide) is used.
The preparations are dispersable in water and they can be applicated by a common application equipments without any problems.

Constraint to the development of biofungicides : the example of "AQ10", a new product for controlling Powdery mildews

R. HOFSTEIN[1] and J.P. AESCHLIMANN[2]

1 : Ecogen Inc., 2005 Cabot boulevard West, P.O. Box 3023, Langhorne, PA 19047-3023, USA
2 : c/o CSIRO, Campus International de Baillarguet, 34980 Montferrier-sur-Lez, France

Over recent years the company ECOGEN Inc. has been successfully developing a biofungicide known under the trade name "AQ10" in the USA, where it has been authorized in 1994 for controlling powdery mildews on various host plants. This new product is based one use of *Ampelomyces quisqualis,* an hyperparasitic fungal species infesting representatives of Erysiphaceae (powdery mildews) only. The production process now allows for the hyperparasite to be mass-cultured and formulated into an easily water-dispersed dry powder which reserves spores viability at room temperature for a period in excess of 12 months. Upon spraying, the conidia germinate rapidly to effectively attack any developing powdery mildew colonies. Efforts are now concentrating on experimental application schedules in vineyards aimed at defining optimal positioning of the biofungicide within existing disease control programs. The pilot trials are being conducted over a large number of wine-producing areas under a Mediterranean climate around the world where powdery mildew represents the major pest problem. Results obtained so far show that in order to achieve an optimal, cost-effective, and consistent disease control, the biofungicide had to be applied before the incidence of powdery mildew has reached high levels, and at any of the three most susceptible growth stages of grapevine, *i.e* : bud break, beween flowering and bunch closure, and just before veraison. The biofungicide may therefore be used as a curative to slightly preventative control agent likely to fit into any IPM strategies. The various technical problems associated with the development and industrial production of this new biofungicide are largely solved. Constraints relating to designing comprehensive, user-friendly IPM programs which include "AQ10" still require adjustment in terms of defining spraying dates and intervals according to a range of agricultural practices, climatic changes likely to occur during the season, and compatibility for tank-mixing with other pesticides. A number of additional constraints relate to official registration in various countries, distributors' and farmers' acceptance of a new "living" product : these are still more difficut to address because they claim for a profound change in the users' general attitude towards controlling diseases of cultivated plants.

Problems in the production, sales and distribution of biological control agents for pest control in greenhouses

K. BOLCKMANS

BIOBEST N. V., Ilse Velden 18, B-2260 Westerlo, Belgium
Fax : +32 -(0) 14/23 18 31

More than 20 different biological control agents are currently commercially available for insect and mite control in greenhouse crops.
Once a biological control agent has been selected, a reliable and economical mass production system has to be developped. Because development of mass production systems is rarely adressed in scientific research programs, a producer of biological control agents has to develop his own mass production systems.
Before a biological control agent can be transferred from the research laboratory to its commercial use, a release system or "formulation" has to be developed ; the release rate or dosage, optimal moment of release and frequency of release have to be determined ; and the best way to distribute the biological control agent in the greenhouse has to be investigated. Knowledge on the side effects of pesticides which are used in greenhouses has to be accumulated as early as possible.
Beneficials have a very short shelflife. Therefore correct storage conditions, a suitable packaging, a fast transport system to the client or distributor and a quality control system are essential. Distributors are typically supplied with products at least once every week.
A key factor for success is the system of combined sales and advice of biological control agents. Technical information has to be transferred from the producer to the user through the local distributor. The distributors are trained by the producer which keeps them informed about new developments.
Generally no registration is required for beneficial insects and mites although some countries started to establish registration. Registration of microbial biocontrol agents is required. The high cost and long lead time of registration of microbial pesticides and the limited greenhouse market seems to hamper the implementation of microbial pesticides in greenhouse crops.

Ciba's way to a biocontrol product

H.J. KEMPF

Ciba-Geigy AG, CH-4002 Basle, Switzerland
Fax : 0041616973383
E-mail : hans-joachim.kempf@chbs.mhs.ciba.com

Ciba is on its course in developing microbial biocontrol agents for the control of plant diseases.
A screening has been set up in order to identify bacteria which are able to antagonize damping-off pathogens, mainly *Rhizoctonia solani*, but also *Pythium* and *Fusarium* species.
Initial activity has to be demonstrated in greenhouse pot assays some of which already involve natural field soil. Then, further trials are performed in the field. Besides field crops like cotton, beans, potatoes, and cucumbers, also covered crops like ornamentals and other vegetables are included in the tests.
In addition to the studies on biocontrol activity, also fermentation and formulation of the selected biocontrol strains are investigated.
Mode of action studies can lead the way for improving the performance of the original strain, either by conventional methods or by a genetic modification of the wildtype.
Ciba is currently investigating a *Pseudomonas* strain and its genetically engineered derivatives. Those genetically improved mutants overproduce antifungal metabolites like pyrrolnitrin which is responsible for a major part of the biocontrol activity.
The major challenge in developing *Pseudomonas* is the achievement of a long term formulation stability at ambient temperatures.
A superior biocontrol product can be brought to the market where an intelligent screening concept and a sound research on strain activity, mode of action, and stability integrates with the experience of an agro company like Ciba.

Practical biological control in Japan and its registration system

Tetsuo WADA

Tomen Corporation, 14-27 Akasaka 2 Chome, Minato-ku,
Tokyo, 107, Japan
Fax : 03- 3588-9930
E-mail : tetsuo-wa @aix.or.jp

In March 1995, long awaited registration, of *Encarsia formosa* and *Phytoseiulus persimilis* was granted.
It took almost 4 years and a half to obtain registration since bioefficacy test had started in 1990. Biological agents are regarded as biological pesticides according to the pesticide regulation law in Japan.
Currently it seems to be the most stringent registration for biologicals in the world and it will require more environmental impact data.
The guideline is not established but will be enforced within 5-6 years.
Major requirements are 6 valid bioefficacy data over 2-3 years, quality control, impact to environment, and other relevant data.
Uses of biologicals are gradually increasing. The acreage where *Encarsia* and *Phytoseiulus* were used is approximately 50 ha in 1995-6 and it is only 1 % of tomato acreage in Japan.
But the future trend for using biologicals depends on their performance.
Low temperature in winter is a major concern for *Encarsia*. There are strong requests from growers to realize early registration of aphids, *Liriomyza*, Thrips control agents. *Phytoseiulus persimilis*, *Aphidius colemani*, *Orius laevigatus*, *Dacnusa sibirica*, which are used for long time in Europe, may be target for criticism.
Spontaneous occurrence of indigenous species sometimes interfer the semi-open greenhouse field test results.

POSTERS

Production of *Bacillus thuringiensis* Berliner on locally available raw materials for control of the Spotted stem borer, *Chilo partellus* (Swinhoe) (Lep. : Pyralidae)

M.A. MOHAMED ALI[1], M. ODOUR ODINDO[1],
M. MAKAYOTO[1], E.M.E. A. EL-TOUM[2], A. MUBARAK NOUR[3]

1 : The International Center of Insect Physiology and Ecology,
Nairobi, Kenya
Fax : 254 2 803360
E-mail : icipe@cgnet.com
2 : Faculty of Agricultural Sciences, University of Gezira,
WadMedani, Sudan
3 : The Agricultural Research Corporation, Wad-Medani, Sudan

Powders of blood, fish, horn and hoof, meat and bone, and soybean meals, were assessed for production of Kenyan isolate of *Bacillus thuringiensis*. These meals were used as the sole protein source, supplemented with cane sugar molasses as carbohydrate source. Soybean meal supported a relatively high level of *B. thuringiensis* production with cell count of 3.72 x 10^8 ml^{-1} and spore count of 1.85 x 108 ml^{-1}, whereas *B. thuringiensis* grown on blood meal was found to be the most toxic with median lethal concentration of 2.04 x 10^6 colony forming units g^{-1} diet. This was evidenced by the test against second-instar *Chilo partellus* larvae. Soybean medium was taken to a 10-litre working volume fermenter in order to scale up the production of *B.thuringiensis* for the field trials. Scale-up production in the fermenter using soybean gave good growth and sporulation. Cell count and spore count after 18 h were 3.06 x 10^8 ml^{-1} and 2.07 x 108.ml^{-1}, respectively.

Development of *Ascochyta caulina* as a mycoherbicide for control of *Chenopodium album*

J.R.F. ALLEN[1], E. PERRIN[1], P. O'CONNELL[2], M. ENZMANN, W. BÄTTIG[3] and P.C. SCHEEPENS[4]

1 : Ciba, Stein 433, Switzerland
Fax : 0041 62 868 64 39
2 : Ciba, Basel, 4002, Switzerland
3 : Ciba : EZA,Münchwilen 4333, Switzerland
4 : AB-DLO, Wageningen, 6700, The Netherlands

The pathogenic fungus *Ascochyta caulina* selectively infects *Chenopodium album*, which is an important weed in arable crops. The potential of this organism as a mycoherbicide was investigated in growth chamber and field studies. Weed control following post-emergent foliar applications was found to be strongly dependent on growth stage of weeds and on climatic conditions. The requirement for high air humidity for successful foliar infection is a constraint to the commercial potential as a mycoherbicide. Attempts to reduce the moisture requirement by adding humectants, stickers and an oil emulsion to spore suspensions sprayed on to plants failed to significantly improve activity in growth chamber or field trials.

Introduction of the aphid parasitoid *Aphelinus abdominalis* (Hymenoptera : Aphelinidae) into unheated tomato plastic greenhouses

O. ALOMAR[1], R. GABARRA[1], C. CASTAÑE[1], R. ALBAJES[2] and J.M. RABASSE[3]

1 : IRTA, Unitat d'Entomologia aplicada. 08348 Cabrils (Barcelona), Spain
2 : Universitat de Lleida-IRTA, 25006 Lleida, Spain
Fax : (93) 753 39 54 E-mail : Alomar@cabrils.IRTA.es
3 : INRA,Laboratoire de Biologie des Invertébrés, 37 bd du Cap, 06606 Antibes, France

Application of chemical treatments for aphid control in unheated IPM tomato greenhouses may affect effectiveness of other introduced natural enemies. *Aphelinus abdominalis* Dalman (Hym.: Aphelinidae) was released in 4 tomato plastic greenhouses in order to evaluate the effectiveness of this parasitoid in establishing in unheated tomato IPM greenhouses and controlling populations of the aphid *Macrosiphum euphorbiae* (Thomas) (Hom.: Aphididae). In order to assure early detection of aphid infestation and precise localization of major aphid foci, we established a grid of quadrats within the greenhouse. Aphid populations were assessed with the use of visual abundance classes. Weekly monitoring of aphid infestation was done by random sampling and regular monitoring of fixed aphid-infested plants.

Results show how *A. abdominalis* does establish in unheated tomato greenhouses in Catalonia and mummies can be found in the greenhouse three weeks after parasitoid releases. Aphid populations did increase in some foci but no sooty-mold developed. The presence of indigenous predators (*Macrolophus caliginosus* Wagner, *Dicyphus tamaninii* Wagner, *Aphidoletes aphidimyza* Rondani) and parasitoids (*Praon* sp., *Aphidius* sp.) may have contributed to overall control of the aphid populations.

10 years of experience with biological control of *Cydia pomonella* and *Adoxophyes orana* by granulosis viruses

Martin ANDERMATT and Markus ZUBER

Andermatt BIOCONTROL AG, Unterdorf,
6146 Grossdietwil, Switzerland
Fax : 063 59 21 23

The poster presents an overview of the development, registration, production and marketing of the two granulosis virus products MADEX (*Cydia pomonella* granulosis virus) and CAPEX (*Adoxophyes* granulosis virus). A summary of all the field tests made in different countries worldwide is given. Advantages and disadvantages of microbial control products based on granulosis viruses are discussed.

A geographical information system (GIS) for the application of the sterile insect technique (SIT) against the Medfly

R. BALDUCCHI[1], U. CIRIO[1] and F. MUSMECI[2]

1 : ENEA CRE Casaccia- Biotechnologies and Agriculture Division, Via Anguillarese 301, 00060 S.M. di Galeria, Rome, Italy
Fax : 0039 6 30486624
2 : ASIA Application and Development of Advanced Informatics, Via Armando Diaz 3, 00061 Anguillara Sabazia, Rome, Italy

In Southern Italy, the Sicily Region is planning to build, an insect factory of 250 million/week capacity, with the support of the EU, for the control of the Medfly by rearing and the release of only sterile males.
One of the main problems related to the wide use of the Sterile Insect Technique (SIT) is the need of a extensive amount of information on the ecology of the species in the island.
The Italian National Agency for New Technologies, Energy and the Environment (ENEA) has developed a Geographical Information System (ECOMED), to overcome the obstacle related to the ecology of the insect, before and during the application of the SIT, for the control of the *Ceratitis capitata* Wied. in the Sicily Region.
In this paper the utility of a GIS for the application of the SIT is examined, in relation to :
a) Medfly population dynamics in the different agro-eco-systems, before and after the release of the sterile flies;
b) planning of the mass-rearing of the Medfly
c) evaluation of the general costs of the SIT application.

Analysis of parasitoid species (Hymenoptera : Aphelinidae) of *Bemisia tabaci* (Homoptera, Aleyrodidae) by using RAPD-PCR

F. BEITIA[1], A. ADAN[1], J.C. ONILLON[3], P. GUIRAO[2] and L. CENIS[2]

1 : C.I.T.-I.N.I.A., Protección Vegetal, Carretera de La Coruña, Km. 7, 28040 Madrid, Spain
Fax : 34 1 3572293 E-mail : BEITIA@CIT.INIA.ES
2 : C.I.D.A., Protección Vegetal, Estación Sericícola, 30150 La Alberca (Murcia), Spain
3 : I.N.R.A., Laboratoire de Biologie des Invertébrés, 37 Boulevard du Cap,. 06606 Antibes, France

In last fifteen years *Bemisia tabaci* (Gennadius, 1889) has reached the status of an important pest in ornamental and vegetable crops in greenhouses, all around the world. This situation could be explained by the identification of a new biotype (**type B**) of the species, which has recently been described as a new species (*Bemisia argentifolii* Bellows & Perring, n. sp.). In Spain, at least, we have verified the presence of two different biotypes of *B. tabaci* on several vegetable crops : a **B type** (possibly *Bemisia argentifolii*) and a **non B type**.

In order to implement and improve the biological control of this pest by using parasitoids, it is very important a quick and easy identification of the species founded parasitizing the whitefly. The molecular technique RAPD-PCR has demonstrated its ability as an useful tool for the identification of several species and/or biotypes of different insects, included *B. tabaci* and its parasitoids.

In the present work, we show preliminar experiments developed to find molecular markers for parasitoid species of Spanish *Bemisia* types, belonging to two genera of *Aphelinidae* : *Eretmocerus* and *Encarsia*, by using RAPD-PCR. Moreover, we try to verify the possibility to distinguish several populations of a same species from different geographical zones and parasitizing different whitefly biotypes.

Science plus Politics equal Action

Stephen BIRDSALL

Imperial County Agricultural Commissionner,
150 South Ninth Street, El Centro, CA 92243, USA
Fax : (619) 353 9420
E-mail : impagcomm@aol.com

In 1987 the Silverleaf Whitefly appeared in Florida greenhouses and tomato fields. It rapidly migrated through natural and artificial means across the United States to San Diego, CA. From San Diego it is believed to have been artificially introduced in the Southwestern Desert on holiday ornamental plants. Nationally, it is responsible for the destruction of more than $1 billion in ornamental plants, high cash value vegetable crops, staple crops such as cotton and important forage crops such as alfalfa. It has now established itself in the lower Colorado River basin of California, Arizona and Mexico where it has continued to migrate in a North South direction. In the fall of 1991, it infested our farming region of 450,000 acres, causing a preliminary crop loss of $130 million with a cumulative four-year crop loss that has reached well beyond $400 million.

We immediately realized that for agricultural to survive as we knew, it would require the cooperative efforts of the entire agricultural community. Growers formed the Imperial County Whitefly Management Committee (ICWMC) to do battle with the Silverleaf Whitefly.

This committee followed the "5-Year National Research and Action Plan for Development of Management and Control Methodology for Silverleaf Whitefly." Through private donations, grants and a grower self-imposed water assessment, we raise more than $250,000 per year for research to run in parallel with the federal effort. These private funds have been responsible for generating at least an additional $1 million annually toward local research performed by the United States Department of Agriculture, California Department of Food and Agriculture and the University of California

The Office of the Agricultural Commissioner of Imperial County is a clearing house and action center, including liaising with numerous other whitefly-afflicted areas in the U.S. and Northwest Mexico. The committee has among other things been responsible for public education of the whitefly, been a driving force behind local research, effectively lobbied for federal whitefly research funding and administered local whitefly control programs. This committee is also an effective vehicle for transferring research technology to the growers. The development and implementation of a resistance monitoring program, home gardener biological control program and volunteer crop sanitation program are just a few examples.

On November 30, 1995, the California Environmental Protection Agencies Department of Pesticide Regulation recognized our efforts in the area of IPM by presenting the ICWMC with their IPM Innovator Award for our leadership and creativity in advancing reduced-risk programs for pest management in agriculture.

Biofertilizers with protective properties against vermin and diseases

R.V. BOROVICK, G.A. ZHARIKOV, N.I. KISELYOVA and N.R. DYADISHCHEV

Research Centre of Toxicology and hygienic Regulation of Biopreparations (RCT&HRB), 102A, Lenin str., Serpukhov, Moscow region, 142283, Russia
Fax : (0967) 72 55 78
E-mail : kostrikova@mx.ihep.su

The report is devoted to the preparations which combine properties of growth stimulants, organic fertilizers and they protect the plants against diseases and vermin. It is grounded in the need for permanent plant protection from the pre-sown processing and through the period of active vegetation to the seeds of next harvest. Such approach is called by the authors "babyseed" technology. This approach is realized via a number of successive stages, so the preparations must be multifunctional. They must possess disinfectant, growth stimulant properties and protect the plants and their harvests as well as being an ecologically safe fertilizers.
The observance of the ecological principle is the obligatory condition to realize the above-mentioned approach. So, environmental contaminations should be reduced and ecologically pure production can be achieved.
As a basis of such preparations, the vermicomposts are used. They are obtained through earthworms. These vermicomposts are enriched by humic substances, nitrogen-fixed bacteria, microelements and they contain the strains of fungi and bacteria such as *Bacillus subtilis, Bacillus sphaericus, Trichoderma, Verticillium* etc, which protect-plants against root rots, phytonematodes and insect pests. It has shown effectiveness and the outlook for use of such preparations in plant protection is excellent.

Biological control through auxiliary insects : Search for information produced using scientometric tools

Geneviève BRANCA-LACOMBE and Françoise COTTE

INRA, Centre de Recherches d'Antibes,
Unité régionale de la Direction Information et Communication
B.P. 2078, 06606 Antibes Cedex, France
E-mail : lacombe@antibes.inra.fr and cotte@antibes.inra.fr

The INRA research teams at Antibes working on biological control wanted to know the major trends in international publications on "biological control" through auxiliary insects". To provide a "cartography" of the scientific and economic aspects in synthesised fashion, we used the Dataview (1) software to analyse a corpus of data (4000 references) from international databases on agriculture (CAB and Agricola), the environment (Environline and Geobase), the economy and patents (WPIL, Science Citation Index, Chemical Industry Notes, Biobusiness, NTIS).

The INRA Centre in Antibes is particularly well-structured for this type of study, because on the same site there are research laboratories, a unit for transferring research results and a biological factory, the company Biotop (a subsidiary of UNCAA). It was therefore easy to set up a group of experts (2) to interpret and validate the results obtained. The analysis by this group of experts enabled us to bring out a certain number of points which came up frequently, in particular :

- for publication countries, the preponderant position of the USA, and the minimal presence of France ;
- the preponderant role of Entomophaga, which was found in all the databases examined ;
- for the key-words, the major families of insects used (Coleoptera, ladybirds, Hymenoptera), the target plants and the techniques used ; the theme regarding *Lymantria dispar* on which numerous American research programmes have been launched for several years.

On the other hand, they infrequently provided interesting new elements, like:

- the astonishing presence of cosmetology publications ;
- the appearance of a new theme, insect-eating fishes feedind on mosquito larvae.

Finally, as regards patents, their low number seems to prove that the practice of research transfer on this subject is more "know-how". The USA (USDA) is the largest holder. France is well represented with 8 INRA patents, one in partnership with the company Biotop.

The Enhancement Department of INRA would like this study to be continued in other aspects of research, such as microbiological control, biological control by acclimatisation, the use of bacteria, viruses, to provide greater and more precise information on research directions and their application to whole subject.

(1) Collaboration with the University of Marseille CRRM
(2) Group of experts consisting of INRA researchers (Biological Control Unit, Enhancement Department) and researchers from Biotop company.

Evaluation of the "genetic control" efficacy in preventing the *Trialeurodes vaporariorum* (Homoptera : Aleyrodidae) population outbreaks under greenhouse conditions

M. CALVITTI, M. BUTTARAZZI, C. GOVONI and U. CIRIO

ENEA CRE Casaccia- Biotechnologies and Agriculture Division
Via Anguillarese 301, 00060 S.M. di Galeria, Rome, Italy
Fax : 396 3048 6624
E-mail : CALVITTI@CASACCIA.ENEA.it

Trials have been carried out in order to evaluate the efficacy of the "autocidal technique" in preventing the "greenhouse whitefly" population outbreaks under commercial greenhouse conditions on squash (*Cucurbita pepo*) plants.
Two demographic trends have been analysed and compared : one related to normal whitefly populations, the other one related to normal whiteflies interacting with radiosterilized insects according to the ratio normal:sterile=1:6.
We have been able to compare the first and second generations produced in experimental greenhouses by means of periodic counts.
The results obtained have showed a remarkable reduction (about -40%) of the total F1 progeny in greenhouses treated with sterile insects. Moreover, we have found a prevailing male off-spring with peaks of about 90 per cent.
As consequence of these two combined results the differences related to the second generation have been found to be more evident especially in quantity.
The results obtained have confirmed the encouraging ones obtained under laboratory conditions and they take us to consider the "Sterile Insect Technique" a new available methodology able to improve the global IPM strategy usable both for "greenhouse whitefly" and *Bemisia tabaci* Genn. control.

Prospects for management of pear Thrips [*Taeniothrips inconsequens* (Thysanoptera : Thripidae)] in Sugar Maple (*Acer saccharum*) forests in the Northeastern United States

E. Alan CAMERON[1], David A.J. TEULON[1,2] and L.H. McCORMICK[3]

1: Penn State University, University Park, Department of Entomology, 501 ASI Building, PA 16802, USA
Fax : 1 814 865 3048
E-mail : ALAN-CAMERON@AGCS.CAS.PSU.EDU
2 : Current Address : Crop and Food Research, Private Bag 4704, Christchurch, New Zealand
3 : School of Forest Resources, 201 Ferguson Building

The annual value of maple syrup, produced from sugar maple (*Acer saccharum*) trees in northeastern North America, is over $US 100,000,000. Moderate to heavy defoliation of a tree can cause the loss of the equivalent of a full year's production during the three years following defoliation. Thrips populations are notoriously erratic ; numbers depend on close synchrony between emergence in the spring from the soil of overwintering adults and development of new buds, and especially of flowers, on the tree.
At least two different techniques to monitor thrips populations have been developed to guide management decisions ; transferring this technology to users will be critical to the eventual success of management programs. Biological agents such as parasitoids (e.g., *Ceranisus* sp.), nematodes, and fungal pathogens (e.g., *Verticillium* sp.) have been identified as having potential value for thrips management ; research is continuing, but programs are not yet ready for implementation. The use of prescribed burning has shown promise as a management tactic to disrupt synchrony between adult thrips emergence and maple bud expansion; its consequences on other biological agents has not yet been investigated. Reliable strategies for management of this pest of sugar maple forests and of sugarbushes remain to be developed and transferred to user groups.

Biological control agents of Musk thistle, *Carduus nutans* L. (Asteraceae) in the United States

G. CAMPOBASSO[1], P. E. BOLDT, LLOYD KNUTSON[2]
and G. TERRAGITTI[1]

1 : USDA, European biological Control Laboratory Rome Station,
Via Colle Trugli 9, 00132 Rome, Italy
Fax : (06) 39 207 90 86
E-mail : EBCL.RomeSubstation@agora.stm.it
2 : USDA, ARS, European Biological Control Laboratory,
B.P. 4168 Agropolis, 34092 Montpellier cedex 5, France

In classical biological control of immigrant weeds, natural enemies (insects, mites, nematodes, pathogens) from the native area of the weed are introduced to reduce its abundance in the invaded area. Musk thistle, *Carduus nutans* L. complex (Asteraceae), is a spiny, noxious herbaceous plant that chokes pastures, rangelands, and roadsides areas. In the United States this weed is successfully controlled in many mid-Atlantic and mid-western states by insects collected in Italy by the European Biological Control Laboratory, USDA-ARS. Two weevils, *Rhinocyllus conicus* (Froelich) and *Trichosirocalus horridus* (Panzer), are responsible for most of the reduction in musk thistle density. Two flies, the syrphid *Cheilosia corydon* (Fallén), and the tephritid, *Urophora solstitialis* (L.) are recent introductions for use against musk thistle in other areas of the country, but are not yet well established. A strain of the rust fungus, *Puccinia carduorum* Jacky, also was released and has spread within several mid-Atlantic states. A leaf feeding chrysomelid, *Psylliodes chalcomera* (Illiger), has been host tested and the results will be submitted for approval for its release in the United States next year. It is expected to be effective especially in warmer areas.

Evidence for sexual reproduction in native populations of Skeleton weed (*Chondrilla juncea* L.) : implications for biological control

Pierre CHABOUDEZ

18, rue des tilleuls, 30900 Nîmes, France

Increasingly, a number of studies has demonstrated that high levels of polymorphism in clonal plant species are usually associated with some residual or facultative sexuality. Here, I report evidence for sexual reproduction within the agamospermous composite *Chondrilla juncea* L. (skeleton weed).

Extensive surveys made in the mediterranean area show that sexual diploid populations occur in Central-Western Turkey. This area also comprises high levels of genotypic variability arising from both high clonal diversity and sexual recombination in populations of the weed.

The relevance of these results to the biological control of *C. juncea* with the rust pathogen *Puccinia chondrillina* Bubak & Syd. is discussed.

Impact of pest management strategies on the biological control of Sunflower insect pests

Laurence D. CHARLET

USDA, ARS, Northern Crop Science Laboratory, Box 5677,
University Station, Fargo, ND 58105-5677, USA
Fax : 701 239 1346
E-mail : charlet@ars.usda.gov

Sunflower is the only row crop in North America coexisting with its native ancestors. Wild sunflower, a complex of fifty *Helianthus* species native to North America, evolved with a large number of insects. Although over 150 phytophagous insect species occur on wild sunflower, only about six species have moved to cultivated sunflower and become economic pests in the United States and Canada. Indigenous natural enemies have been a significant factor in preventing many insects from becoming significant pests. The Sunflower beetle, *Zygogramma exclamationis* (F.), the Banded sunflower moth, *Cochylis hospes* Walsingham, the Red sunflower seed weevil, *Smicronyx fulvus* LeConte, the Sunflower stem weevil, *Cylindrocopturus adspersus* (LeConte), and the Sunflower moth, *Homoeosoma electellum* (Hulst) have been important pests of cultivated sunflower in recent years. Research has focused on the biology and population dynamics of the major sunflower insects and on integrated approaches to provide for their management. The biological control of the pests has also been investigated as an important component of the integrated pest management of the sunflower cropping system. Knowledge about the interaction of the different management strategies, especially cultural and chemical control, with the action of the parasitoid complex of the pests is essential in developing a successful management program. Research has shown that the use of different planting dates has reduced infestations levels of certain pest species without significant changes in rates of parasitism. In addition, some sunflower lines have shown resistance to insect infestation. Parasitization rates also varied among lines, but high parasitism did occur in some lines with low pest densities. The use of insecticides often has a deleterious effect on the parasitoid fauna of the sunflower pests and this strategy should be utilized only when economic thresholds have been reached in order to conserve the natural enemy complex present in the sunflower agroecosystem.

Caracterisation of the biotic potential of *Psyllaephagus euphyllurae* SILVESTRI (Hymenoptera, Encyrtidae), endoparasitoid of the Olive tree psylla *Euphyllura olivina* (COSTA) (Homoptera, Aphalaridae)

B. CHERMITI

Laboratoire de Zolologie agricole, E. S. H. E.,
Chott Mariem, 4042, Sousse, Tunisie
Fax : 216 3 248 275

The females of *Psyllaephagus euphyllurae* SILVESTRI are able to parasitize their host *Euphyllura olivina* (COSTA) at all its development stades. However, a preferential laying is observed on larvae of fourth and fifth stages.

In the range of studied temperatures, the laying capacity of *P. euphyllurae* is maximum at 22°C. On the other hand, the females of *P. euphyllurae* have a very low power of discrimination and come back on the infested hosts and consequently have a tendency to become superparasites.

Alternaria alternata, a new fungal pathotype pathogenic to Aphids

C. CHRISTIAS, Alexandra DARA, A. KALLIAFAS
and G. CHRYSANTHIS

Department of Biology, School of Natural Sciences,
University of Patras, 265 00 Rion, Patras, Greece
Fax : +30 61 997 494

A new, as yet unreported aphid pathogen was isolated from diseased or dead aphids, from samples collected from the areas of Aetoloakarnania, Salonica, Crete and Western Peloponnese, Greece. The new fungus which proved to be a very aggressive aphid pathogen was identified as *Alternaria alternata*. Its pathogenic nature was verified using Koch's postulates. Inoculation of healthy aphids with spores of the pathogen always resulted in death of the insects within 48 hours. The percentage of successful inoculations resulting in death was always 100%. The pathogen was able to infect the aphid species *Myzus persicae*, *M. nicotianae, Aphis fabae*. *A. nerii*, *Brevicoryne brassicae*, *Macrosiphum euphorbiae*, *Uroleucon* spp. and at least twelve other unidentified species of aphids. Pathogenicity tests, using *Ceratitis capitata* and *Drosophila melanogaster* showed that *A. alternata* was unable to infect the above two insect species.

The disease development started with spore germination on the surface of the insect exoskeleton and proceeded with the penetration of the infection peg and mycelial development and ramification in the soft tissues. All the insect parts were susceptible to infection, namely, legs, antennae, head, thorax, abdomen, syphons. A brown discoloration followed the initial infection which spread progressively from the infection point to the entire insect. Immediately after artificial inoculation the insects became uneasy and started moving in a haphazard manner. Six to 12 hours later the insects became immobile and moved some of their legs and antennae only occasionally and very sluggishly. Eventually the insects died. Death was followed by external mycelial development and profuse sporulation.

Optimum conditions for infection were : temperature 20-25°C, relative humidity above 75% and photoperiod of 12h light and 12h darkness. This new aphid pathogen offers new opportunities for its exploitation a as biocide for the biological control of aphids in the field.

Feeding on plant resources by natural enemies : a desirable trait for biological control ?

Moshe COLL

Department of Entomology, Faculty of Agriculture,
The Hebrew University of Jerusalem,
P.O. Box 12, Rehovot, 76100, Israel
Fax : 972 8 9466768
E-mail : Coll@Agri.Huji.ac.IL

Many species of predators and parasitoids consume not only animal tissues but also plant materials - a phenomenon termed omnivory. Two types of omnivory are of particular importance for biological control. The first type, in which plant and prey resources are consumed at different life stages of the omnivore, is found in parasitoids and some predators (e.g., Chrysopidae). The second type, in which the omnivore switches between plant and prey feeding during a single life stage, is exhibited by predators, primarily by heteropteran and coccinellid species. Much work has been devoted to the influence of nectar-feeding on parasitoid performance. However, it is not clear how feeding on plants influence predator-prey population interactions. Data show that feeding on plants is important not only in sustaining the predators in the absence of prey, but also in increasing the predators' fitness when it supplements prey diet. A simple model shows that feeding on plants by predators destabilizes the system, through both enhanced predator reproduction and increased predator-prey competition. Thus, prey populations are likely to go extinct, at least locally, when the predator feeds also on suitable plants. The habit of many predators to feed on plants must be considered if their effectiveness as biocontrol agents is to be maximized. Early releases of plant-feeding predators could enhance their effectiveness because they may sustain themselves on plant resources when prey density is still low. We must, however, test carefully for potential negative effects of plant-feeding predators on yields. The compatibility of plant-feeding predators with the use of systemic pesticides also needs to be assessed. Finally, plant materials could be utilized for feeding and as oviposition and delivery media in mass-production and release programs.

Problems associated with the production and use of *Helicoverpa* NPV in India

S.E. COWGILL

International Crops Research Institute for the Semi-Arid Tropics,
Patancheru 502 324, A .P., India
Fax : +91 40 241239
E-mail : COWGILL@ICRISAT.CGNET.COM

Helicoverpa armigera is a major crop pest in Asia. In India it causes substantial losses in cotton and is the dominant pest of several grain legumes. In chickpea and pigeonpea, it has been estimated to cause annual losses in excess of $600. Traditionally these crops were grown as subsistence crops but recent increases in their market price have resulted in a shift to commercial production. This has resulted in an increase in the amount of insecticide applied to the crops. However, organophosphate, cyclodiene and pyrethroid resistance are ubiquitous in *H. armigera* populations in the Indian subcontinent and management of the pest is often not achieved. In 1987/1988 the combination of high population pressure and high resistance levels resulted in control failures and financial losses for farmers. As a result, there is increased interest in the development of alternative management options to deal with the pest. These have included the use of *H. armigera* nuclear polyhedrosis virus.

Both commercial and Governmental organizations are involved in *Ha*NPV production in India. Examination of samples from both sources have shown products to be highly contaminated and of low concentration. Poor product quality has contributed substantially to the observed variability in the level of effectiveness provided by *Ha*NPV. However, the lack of information on the most appropriate application rates and procedures has also played a role. This paper summarizes the difficulties experienced by producers of *Ha*NPV and describes the results of studies to identify appropriate production techniques and develop recommendations for the field use of *Ha*NPV in India.

Technological transference of the integrated House fly management in poultry farms of Argentina

D.C. CRESPO[1,2] and R.E. LECUONA[1]

1 : Instituto de Microbiología y Zoología agrícola,
IMYZA - CICA - INTA Castelar,
C.C. 25 (1712) Castelar, Bs. As. Argentina
Fax : 0054 1 6210670
2- CONICET, Buenos Aires, Argentina.

The house flies reach high population levels during spring and summer seasons in confined poultry productions. This insect is a known vector to several human diseases. Up to now, the exclusive use of chemical products has been unsuccessful to control this pest and contributed to the selection of flies with resistant genotypes. For this reason, in Argentina, the Instituto Nacional de Tecnología Agropecuaria (INTA) worked out an integrated system that considers different alternatives of control. The technology is mostly based on the adequate poultry house management (including manure), the use of parasitized pupae by *Spalangia* spp. and *Muscidifurax raptor,* and occasionally the employ of chemical (sexual lures and paints). Several trials were performed in poultry farms with approximately 500.000 laying-hens under high levels of houseflies. Three weeks after the implementation of this technology, the fly population was reduced from 300 to 5 flies per sampling unit. INTA trained the farmers on methods to dry the manure, to correct the poultry house management problems, to use fast sampling methods and about the release of benefical insects. The farmers learned to storage and release the parasitoids, as well as to evaluate *in situ* the parasitism level. There were periodic farms surveys by INTA to check up the activities. The success of this approach raised the demand for this technology making necessary to increase the parasitoid production. Consequently, the automation of different production steps is under development to diminish the costs of massive rearing. INTA obtained funding from the Secretaría de Ciencia y Técnica from Argentina and Banco Interamericano de Desarrollo, to develop the first biofactory of these parasitoids. Now the objective is to produce large quantities of insects with high quality and at lower costs.

Phytophagous insect candidates for biological control : evaluation of the impact of interspecific competition on their effectiveness

M. CRISTOFARO[1], G. CAMPOBASSO[2], A.C. PASTRINO[3] and L. KNUTSON[2]

1 : ENEA, C.R. Casaccia, Rome, Italy
Fax : +39 6 30486545
E-mail : ENTOM@SUNGAZ.CASAX+CCIA.ENEA.IT
2 : USDA-ARS, European biological Control Laboratory,
B.P. 4168 Agropolis, Montpellier cedex 5, France
3 : University of L'Aquila, L'Aquila, Italy

The establishment of a biological control agent of a weed depends on several factors, including competition with other established herbivorous insects, associated with the same weed. This study attempts to set up a procedure for the evaluation of interspecific behavioral relationships beween phytophagous insects occurring on the same plant species. Laboratory tests and field observations were conducted during a study of the relationships beween two noctuid leaf-feeding moths (*Simyra dentinosa* and *Oxicesta geographica*) and two gall midges (*Spurgia esulea* and *Dasineura sp. nr.capsulae*), selected as biological control agents against leafy spurge (*Euphorbia esula* "complex").
Three approaches were used : i) field observation, recording qualitative and quantitative data on the distribution of the target plant and the related entomofauna for a prolonged period ; ii) competition tests, in the field and laboratory ; iii) behavioral tests, in the laboratory, using an olfactometer, wind tunnel, and event recorder software, to record and analyse the response of one species in the presence of the chemical signs of the feeding of another species on the same host plant.
The results of competition and behavioral tests indicate a mechanism of repellence in both cases in the presence of the gall midge the gall tissue induces a negative suitability for feeding by the moth larvae, while the presence of the moth feces inhibits oviposition in the gall midge. These results, confirmed by the field observations showed a mechanism that allows for release and coexistence of the gall midge and noctuid moth species on the same host in the same area.

Milsana (extracts from *Reynoutria sachalinensis*) protects cucumber against Powdery mildew by inducing plant resistance

F. DAAYF[1], A. SCHMITT[2] and R.R. BELANGER[1]

1 : Laboratoire de biocontrôle, Centre de Recherches en Horticulture, Envirotron, Université Laval, Ste-Foy, Québec, G1K 7P4, Canada
E-mail : phytodaf@vml.ulaval.ca
2 : Federal Biological Research Center for Agriculture and Forestry, Institute for biological Control, Heinrichstraße 234, 64287 Darmstadt, Germany

Milsana, a commercial formulation of leaf extracts of *Reynoutria sachalinensis*, reduces the incidence of cucumber powdery mildew (*Sphaerotheca fuliginea* Schlecht). The product is not known to have any direct antifungal activity but peroxydases, polyphenoloxidases, and PAL, enzymes involved in the production and metabolism of phenolics, were previously shown to be activated in cucumber by such a treatment. This enzymatic activation, implying the capacity of the plant to stimulate phenylpropanoid reactions, may have a key role in explaining an indirect action of Milsana, but no phytoalexins nor hypersensitive reactions, involving phenolics, could be detected before in cucumber treated with Milsana. Recently, however, we showed that Milsana stimulated the production of many compounds of phenolic nature, displaying fungitoxic activity in their aglycone form. Some of them presented characteristics of cinnamic acid derivatives. We recently identified one of these compounds as p-coumaric acid methyl ester, which was also present in control plants but in smaller quantities. Some of these antifungal compounds can be found in planta in their free form within a few days following treatment. Such changes in metabolism, associated with reported enzymatic activities indicate that the phenylpropanoid metabolism is activated in cucumber by Milsana. Considering differences in metabolic changes between control and treated plants, associated with protection against powdery mildew, and since Milsana does not act directly, phenolic pathway activation appears to be the main phenomenon involved in protection.

Producing Pesta, a delivery system for biocontrol agents, by twin-screw extrusion

D.J. DAIGLE, W.J. CONNICK, M. LOVISA, K. WILLIAMS, C.D. BOYETTE and M. WATSON

USDA, ARS, SRRC, P.O. Box 19687,
New Orleans, LA 70179, USA
Fax : (504) 286 4367
E-mail : ddaigle@nola.srrc.usda.gov

"Pesta" granules in which fungal propagules are encapsulated in a wheat gluten matrix were prepared in pound quantities by twin-screw extrusion. The c.f.u. of these granules were compared to the c.f.u. of laboratory samples. In the dough formulation of wheat flour, clay filler, fungus, and water, low water concentration reduced the viability of *Colletotrichum truncatum*. This loss of viability was moderate with *Alternaria conjuncta/infectoria* and minimal with *Aspergillus flavus* and *A. parasiticus*. Drying the product to approximately 7 % moisture content similarly reduced the viability of the aforementioned fungi. In the greenhouse, twin-screw extruded (1.5 mm dia. x 2 mm) granules containing *C. truncatum* [5 x 104 c.f.u. per gram] caused high levels of infection and mortality in the target weed, hemp sesbania.

A Seedling test for evaluating resistance induced by *Penicillium oxalicum* to *Fusarium* wilt in tomato

A. DE CAL, R. GARCIA-LEPE and P. MELGAREJO

Area de Protección vegetal, CIT-INIA, Carretera de La Coruña,
Km 7. 28040 Madrid, Spain
Fax : 07 34 1 3572293
E-mail : melgar@cit.inia.es

An *in vitro* test is described for determining resistance induced by *Penicillium oxalicum* against *Fusarium oxysporum* f.sp. *lycopersici* race 2 in tomato. Tomato seedlings previously induced with *P. oxalicum* were grown in sterile flasks containing different doses of spore suspension of *F. oxysporum* f.sp. *lycopersici* in Hoagland solution. Seedlings were grown for 20 days at 22-28°C in a growth chamber. Disease severity, AUDPC, stunting, weight of plants and consume of the nutrient solution were compared. All the parameters were independently correlated to consume of the nutrient solution (R 0.88). Plants non-inoculated with *F. oxysporum* f.sp. *lycopersici* (induced or not) consumed the same amount of nutrient solution. However, induced/inoculated plants consumed more nutrient solution than non-induced/inoculated plants. The consume of the nutrient solution can provide with a significant measure to compare the resistance to *Fusarium* wilt. Resistance appears to depend on the control of the water stress caused by the pathogen in tomato plants.

Susceptibility of *Spodoptera littoralis* (Lep.: Noctuidae) and *Helicoverpa armigera*, (Lep.: Noctuidae) laboratory and field strains to *Bacillus thuringiensis* formulations and insect growth regulators

A. de COCK, P. COUWELS and D. DEGHEELE

Laboratory of Agrozoology, Department of Crop Protection,
Faculty of agricultural and applied biological Sciences,
University of Gent, Coupure Links 653, 9000 Gent, Belgium
Fax : 00 32 9 264 62 39

As a part of the EC-project "Improving aphid and noctuid pest control in the framework of integrated pest management programmes for vegetable crops", a cooperation between Belgium, France, Italy and Spain, *B. thuringiensis* formulations and insect growth regulators (IGRs) were evaluated for their effect on the noctuids *S. littoralis* and *H. armigera.* A comparison was made between the effect on strains reared in the laboratory for several years and recently collected field strains originating from Spain and Sicily.
The *B. thuringiensis* formulations, bactospeine, dipel and thuricide were not active against *S. littoralis* field and laboratory strain (LC50 > 1500 mg/l), dipel was moderately active against the laboratory strain but not against the field strain. All the *B. thuringiensis* preparations were active against *H. armigera*, dipel was the most potent one.
Teflubenzuron, hexaflumuron, flucycloxuron and flufenoxuron can provide an effective control for *S. littoralis.* The Sicilian field strain was generally more susceptible to IGRs as compared to the laboratory strain. Teflubenzuron was not active against *H. armigera*, the other compounds had a good activity against this insect. The laboratory and the Spanish field strain had about the same susceptibility.
Our results indicate that cross resistance and/or resistance against these compounds is not yet to be expected.

Development of a biopesticide based on *Bacillus sphaericus* and technology transfer of production process to a Brazilian industry

J.M.C. de S. DIAS[1], P.de T.R. VILARINHOS[2], C.S. HONDA[3] and R.G. MONNERAT[1]

1 : Centro Nacional de Pesquisa de Recursos Genéticos e Biotecnologia - CENARGEN/EMBRAPA- PO Box 02372, 70849-970 Brasília DF, Brazil
Fax : +55 61 340 3624 E-mail : cabral@cenargen.embrapa.br
2 : Instituto de Saúde do Distrito Federal, Brasília, DF; Brazil
3 : GERATEC Biotecnologia Aplicada, Cruz Alta, RS, Brazil

Mosquitoes are an important public health problem in Brazil. In 1995, more than 400.000 malaria cases were recorded. Filariasis is endemic in Recife and other cities of the Northeast and dengue outbreaks have been registered in many states. With problems arising from the continuous use of chemical pesticides, like pest resistance, toxicity to man and non target fauna, associated with environmental concerns, the search for more friendly alternatives to control mosquitoes has been opportune. *Bacillus sphaericus* is a spore-forming bacteria discovered in 1904, whose larvicidal activity against mosquitoes was determined in 1964. The larvicidal activity is associated with pro-toxins produced during the bacterial sporulation phase that, exposed to the mosquito larval gut conditions are transformed into toxins. These toxins are active against mosquitoes from the genus *Culex* and *Anopheles* mainly and some *Aedes* species. Strain 2362, from serotype H5a5b recommended by WHO was used to develop the biopesticide. Lab experiments were conducted in CENARGEN to determine alternative culture medium, growth conditions (pH, shaking, aeration and temperature) and its relation with toxicity to mosquito larvae. Initial development was carried out at CENARGEN and final formulation and scale up were done by GERATEC, through a technology transfer contract. Field tests were conducted under real use conditions, mainly in urban areas where the mosquito *Culex quinquefasciatus* was predominant. During this tests it was shown that the biopesticide could replace the chemical products used in routine control successfully. GERATEC submitted registration of the product in 1993 to Health Ministry and in January 1996 a commercialization licence was released. During this year the company will start production and commercialization of the biopesticide, for use in urban areas mainly.

An azadirachtin formulation on *Ceratitis capitata*(Wiedemann) (Diptera : Tephritidae) : effects on the oogenesis and the life cycle

V. DI ILIO[1], M. CRISTOFARO[1], D. MARCHINI[2], P. NOBILI[1] and R. DALLAI[2]

1 : ENEA - C.R Casaccia, Rome, Italy
Fax : 39- 6- 304 86545
E-mail : ENTOM@SUNGAZ.CASACCIA.ENEA.IT
2 : University of Siena, Sierta, Italy

The effects of azadirachtin, a terpenoid compound extracted from the neem tree (*Azadirachta indica*), were evaluated on the oviposition and the longevity of the Mediterrean fruit fly [*Ceratitis capitata* (Wiedemann)], under laboratory conditions. Four tests were set up in no-choice conditions to define the oviposition rate and the longevity of the medfly adults in relation to the concentration of the active principle and to the stage of development. The results showed a drastic decrease in the oviposition rate, due to a loss od the development of the ovaries in the female, and a low insecticide action. Histological and ultrastructural studies confirmed the damage on the female gonads. Subsequent tests showed the irreversibility of the process, even after a relatively short insect exposure to the compound. These promising results may suggest a possible application in open field, in combination with specific baits and attractants.

Methods of application the biological control

Eva DUSKOVA

Research Institute of Crop Production, Drnovska 507,
161 06 Praha-Ruzynà, Prague, Czech Republic

Fax : 42 2 36 5228

This year, the Czech register of pesticides has a separate section for biological preparates. The first part of this section deals with mycobacterial preparations ; those of Czech production such as Boverol (*Beauveria bassiana*), Ibefungin (*Bacillus subtilis*), Polyversum (*Pythium oligandrum*), Supresivit (*Trichoderma harzianum*) ; the others imported, mostly different forms of *Bacillus thuringiensis*. The second part of the section deals with macroorganisms that are used to control pests of Czech production and imports.

The problem of assured efficacy is still the most important impediment of common use of biopreparates in practice. The variability of resistance of plants, virulence of pathogens, environmental conditions etc. all influence efficacy, and methods of application must take each of these factors into account. Methods for pest control by chemical pesticides are the same or similar all over the world. In contrast, biological preparates are specifically effective against e.g. a certain genus, variety and/or race of pathogen, and under certain specific, characteristic and often local conditions. It is, therefore, impossible to simply replace chemical pesticides with biopreparates, and to use methods for the former to apply the latter. Completely new methods have to be developed, and applied with flexibility.

As an example of successful application, the biocontrol in propagation of geraniums by the producer (in a greenhouse) may be presented here :

During November and December losses of young plants to damping-off increased and reached 45 - 60% . We analysed the situation (soil and other conditions, technology, diseased plants affected mostly by *Pythium* spp.) and recommended to change the composition of the substrate, and changed the technology (after removal of the plants all substrate has to be removed before planting new cuttings). The biological fungicide Supresivit was drenched into the substrate at a dose of $1 g/m^2$ one to five days before planting the cuttings. We obained an improvement, but damping-off still ranged beween 13-15 %. *Fusarium oxysporum* diagnosed as the cause of it, and its source were the mother stocks. Infection by *F. oxysporum* was latent at the mother plants, and at the low temperature in winter did not manifest itself, but caused damping-off during rooting of the cuttings at the higher temperature of the propagation period. Accordingly, negative selection of the mother stock was done to exclude all plants with weak or wilted branches.

Damping-off then decreased to a level of 2-3 %, by a completely non-chemical control.
Before use of a biopreparate it is very important to diagnose all pathogens, pests and harmful abiotic factors, to test the efficacy against certain pathogens (*in vitro*), bearing in mind that the efficacy may differ with different isolates of the pathogen. Control of other pests or diseases must not affect the bioagent. The growing technology must be adjusted to create an accord of optimal conditions for both plant and biopesticide.

Experimental models in toxicological tests of pesticides

N.R. DYADISHCHEV, S.P. RYBALKIN and A.I. MARCHENKO

Research Centre of Toxicology
and hygienic Regulation of Biopreparations (RCT&HRB)
102A, Lenin str., Serpukhov, Moscow region, 142283, Russia
Fax : (0967) 72 55 78
E-mail : kostrikova@mx.ihep.su

It is advisable to use alternative methods *in vitro* in toxicology which limit or eliminate animal test. These methods are physical and chemical, mathematical modeling, molecular modeling with computers and application to of invertebrates, plants, bacteria, cell culture , etc.
Human and animal cell cultures take the most important place among experimental models in medical and biological researches.
Research into the uses of cell culture in toxicological tests is carried out in RCT and HRB. Toxicity of antigenic complexes of nonpathogenic *Escherichia* and *Pseudomonade* has been tested on human cell cultures (Hela, Hep-2, M-20) and animal cell cultures (L 929, BHK-2r, Vero). As a result, subtoxical and minimal doses were defined ; the correlation between toxicity of bacterial antigens for cell cultures and laboratory animals was determined (r = 0,73).
The data obtained makes it possible to prepare indicator tests that can define toxic levels of bacterial antigens if pesticides are present.
Aternative methods of prognosis are made possible by created mathematical models and accumulated experimental data. These alternative methods allow us to determine the toxicity of chemical and biological substances that are supposedly dangerous to humans, and to obtain additional information about the influence of any toxic agents found.

Implementation of adjusted relative humidity for integrated control of Two spotted spider mite *Tetranychus urticae* Koch under plastic tunnel conditions in Egypt

Ahmed Yousry Mohamed EL-LAITHY

National Reserarch Center, Dept. of plant Protection ,
12622, Dokki, Cairo, Egypt
Fax : 202 3370931

Microclimatic conditions were monitored in cucumber crops grown under unheated plastic tunnels during spring time in three consecutive years 1990/1992 in Egypt. Relative humidity prevailing in the plastic tunnels constructed in either sandy or clayey soil fields did not exceed 30% during mid day time, while average temperature did not drop below 30°C. Such conditions favored the population build up of Two spotted spider mite *Tetranychus urticae* Koch, were not favored for the population of the predatory mite *Phytoseiulus persimilis* Athias-Henriot.
Three plastic tunnels growing similar cucumber cultivar (Hana) were selected. Relative humidity in two tunnels were maintained constantly around 50% during the day using a spraying nozzels, the third one was kept as check. Incidence of mildew diseases induced by leaf was wetness was characterised by percentage of infested plants. Both of the predatory mites *P. persimilis* and *P. longipes* were released in either of the sprayed tunnels, while acaricides were applied for the check. Nonsignificant difference took place between sprayed and unsprayed tunnels in the ratio of downy mildew infested plants, 25.8, 23.0 and 23.1 for check. Unless fungicides were applied the rate of infestation increased significantly in sprayed tunnels. At the begining of May population of *T. urticae* covered the apical part of cucumber plants, which required two acaricidal applications as predatory mites did not migrate to the higher leaves of cucumber plants, which were very warm and dry. In sprayed tunnels more than 80% of the yield was produced without any acaricidal application besides the reduction of frequent fungicide applications as a prophylactic treatment.

Biocontrol of Mulberry Two spotted spider mite *Tetranychus urticae* with introduction of *Phytoseiulus persimilis*

A.Y.M. EL-LAITHY[1], M.S. SALAMA[2] and M.A. MADKOUR[3]

1 : National Research Center, Dept. of Pests and Plant Protection, 12622 Dokki, Cairo, Egypt
Fax : 202 3370931
2 : Fac. of Science, Entomology Dept., Ain-Shams Univ., Cairo, Egypt
3 : Agricultural genetic engineering Research Institute (AGERI), 12619-Giza, Egypt

Infestation of mulberry leaves reared in plastichouses with the Two spotted spider mite *Tetranychus urticae* Koch is the main feeding constraint of Silkworm *Bombyx mori* especially during spring season.
Two exotic variety of mulberry shrubs Canva 2, *Morus alba* and kukoso 20, *Morus latifolia* were transplanted into a rearing plastichouse. Infestation spots of *T. urticae* were seen on new leaves late Feb. Leaves of kukoso 20 appeared to be less favored to *T. urticae* feeding compared with canva 2. Population density in control shrubs reached 456 individual/leaf in the later variety, while it did not exceed 20 individual/leaf of the former one. The predatory mite *Phytoseiulus persimilis* Athias-Henriot was released at the rate of 1:50 predator-prey. The predacious mite successfully suppressed population density of *T. urticae* from 14o.4 to 0.3 individual/leaf within 4 weeks. An indigenous predatory mite *Amblyseius swirskii* A.-H. was also introduced in the same rate , but individuals disappeared away from released shrubs. Water was sprayed in the experimental plastichouse during mid day time to improve its climatic conditions. Spraying program maitained relative humidity about 45% and temperature between 30-40 °C.

Biological Control of Citrus Mealybug, *Planococcus citri* in Turkey

Lerzon Bakircioglu ERKILIC[1] and Ahmet OZKAN[2]

1 : Plant Protection Research Institute, PK 21 01321, Adana, Turkey
Fax : (90) (322) 322 48 20 E-mail : Uygunnd@pamuk.cc.edu.tr
2 : Greenhouse and Citrus Research Institute, Antalya, Turkey

Citrus mealybug, *Planococcus citri* (Risso) is one of the most common plant protection problem of citrus growing. Besides feeding injury on plant tissue, the secretion of honewdew cause 30-60 % damage on fruit quality. Broad spectrum pesticides, which is not recommended, were stimulated the damage by side-effects on natural enemies. Because of safety and long live effects the biological control is prefer Citrus mealybug and other citrus pests in Turkey.

For this aim, *Cryptolaemus montrouzieri* Muls and *Leptomastix dactylopii* (How) were introduced from USA by Antalya Biological Control Station in 1965. Since 1970s these beneficials have been reared and distributed to citrus growers by Adana Plant Protection Research Institute and Antalya Citrus Research Institute. In the first years we had some problems on both rearing and distributing. Particularly, citrus growers interest was very low compared to the recent years. The production capacity of Adana Plant Protection Research Institute is approx. 1.5 million *C. montrouzieri* and 3.0 million *L. dactylopii*. From this production 1.2 million *C.montrouzieri* and 2.5 million *L. dactylopii* were distributed to the citrus growers in East Mediterranean Region of Turkey in 1995. This biological control activity takes a special part of citrus IPM studies. There is no broad spectrum pesticides recommended for any other citrus pests and most of them have their natural balance in the orchard. According to the sufficient results of biological control, recent years demand of citrus mealybug natural enemies increased drastically. Rearing capacity may also need increasing.

Microbial control of the Maize stem borer *Sesamia cretica* Lederer (Lep. : Noctuidae) with a granulosis virus in Egypt

G. FEDIERE[1], M.A.K. EL-SHEIKH[1], A.M. SEMEADA[2], A. EL-HEFNY[1], M. MASRI[1] and S. EL-SHERIF[2]

1 : Entomovirology Laboratory, ORSTOM,
Dept. of econ. Entomol. and Pesticides, Fac. of Agric.,
Cairo Univ., Giza, Egypt
Fax : (202) 35 13 941
2 : Corn Borers Research Laboratory, Dept. of econ. Entomol. and Pesticides, Fac. of Agric., Cairo Univ., Giza, Egypt

The Pink Borer, *Sesamia cretica* is one of the important corn borers in Egypt. Until now, no biocontrol agent has been tested against this pest. Pilot experimental field trials were conducted using a recently isolated Granulosis Virus, (*Sc*GV, FEDIERE *et al.*, 1993) in the north of the Nile Delta, at Sakha Agricultural Research Station, Kafr El-Sheikh Governorate, where heavy natural infestation rates of this Corn borer often occur.

The efficacy of spray application of *Sc*GV was field tested and compared with the currently recommended chemical insecticide (Lanet 90% S.P.) as two different control strategies against *S. cretica* larvae on maize. The viral insecticide is a highly purified suspension of granules in water plus 10% molasses as a sticker and feeding stimulant additive. Sprays containing 0.1 g of granules /litre (*i.e.* 1. 24 X 10^{10} I.B./ml), applied 2 times at rates of 1.25 and 2.5 litre/1/100 feddan (1 feddan = 42 m^2) at 2 weeks interval, starting 3 weeks after sowing. The chemical insecticide was used at the same times, at a concentration of 3g/litre/ 1/100 feddan. All treatments were made using a portable knapsac sprayer. Additional plots were left as a control treatment.

Results revealed that both tested treatments (microbial and chemical pesticides) noticeably reduced the numbers of *S. cretica* larvae. As regards yield, the mean weight of seeds was higher for the plots which were treated by either virus or insecticide than untreated ones. However, the chemical insecticide treatment was more effective than the formulated virus treatment. The corrected yield in ardab (1 ardab = 145 Kg) / feddan were 20. 11, 16. 04 and 12. 79, for chemical insecticide, viral insecticide and control treatments, respectively.

It is concluded that *Sc*GV proved to be pathogenic to the field population of *S. cretica* and may be considered a promising biocontrol agent to support the Integrated Pest Management Programme of such an important Corn Borer. This work will further include the utilization of another virus, the *Mythimna loreyi* Densonucleosis Virus, *Ml*DNV (FEDIERE *et al.*, 1995).

References

FEDIERE, G., TAHA, A.A., ABOL-ELA, S, LERY, X., ZEDDAM, J.L., VEYRUNES, J.C., V GIANNOTTI, J. - 1993. Mise en évidence d'un virus de granulose chez *Sesamia cretica* Led. (Lepidoptera Noctuidae) principal ravageur du maïs d'Afrique du Nord-Est: Caractérisation de l'ADN génomique et diagnostic viral. C.R. Acad. Sci., 316, 1350- 1354.

FEDIERE, G., EL-SHEIKH, M.A.K., ABOL ELA, S., SALAH, M., MASRI, M., VEYRUNES, J.C. - 1995. Isolation of a new Densonucleosis Virus from *Mythimna loreyi* Dup (Lep. Noctuidae) in Egypt. Bull. Fac. Agric.Cairo, 46, (4), 693 - 702.

Laboratory selection of Trichogrammatidae as biological control agents using multivariate analysis

F. FOURNIER[1] and G. BOIVIN[2]

1 : Les Services Bio-Controle Inc., 430 boulevard Gouin, Saint-Jean-sur-Richelieu, Québec, J3B 3E6, Canada
Fax: 514-346-7740
E-mail: FOURNIERFR@EM.AGR.CA
2 : Agriculture and Agri-Food Canada, Québec, Canada

A multi-variable selection among 40 species and strains of *Trichogramma* and *Trichogrammatoidea* (Hymenoptera : Trichogrammatidae) was initiated to select biological agents for three Lepidopterous pests of cruciferous crops, the Imported cabbageworm (*Artogeia rapae* L.), the Diamondback moth (*Plutella xylostella* L.) and the Cabbage looper (*Trichoplusia ni* Hübner). Parasitism, egg abortion, host adequation, progeny sex ratio and host preference were evaluated on the three hosts in no-choice and choice experiments. Strain longevity was measured at 25°C as well as their parasitism and survival at temperatures ranging from 10 to 35°C. Productivity was also measured on eggs of a factitious host, *Ephestia kuehniella* Zeller. An important inter- and intraspecific variability was observed on all the variables measured. The first three axes of the principal component analysis explained 49,4% of the total variance (26,5%, 14,5% and 8,4%, respectively). The first principal axis was positively determined by host acceptation, parasitism and progeny sex ratio on *P. xylostella*, strain longevity, fecundity on *E. kuehniella*, parasitism at 15 and 35°C and survival at high temperatures. The second principal axis was negatively determined by host acceptation and parasitism of *A. rapae* and positively, by parasitism of *T. ni* and the abortion rates on the three hosts. Acceptation and parasitism of *A. rapae* and progeny sex ratio on *P. xylostella* and *T. ni* had positive coefficients on the third axis, while longevity and adequation of *P. xylostella* had negative coefficients. Seven groups of strains with similar attributes were obtained after an average linkage clustering analysis. Two of these groups, one exlusively made up of thelytokous strains, were poor candidates for either the control of the target pests, or production on the factitious hosts, or both. Another group of two species was rejected due to their poor longevity and low adequation on *A. rapae* and *P. xylostella*. The four remaining groups contained promising candidates from which 12 strains were selected for further evaluation in greenhouse experiments.

Natural enemies associated to Lepidoptera pests in IPM tomato fields

R. GABARRA[1], J. RIUDAVETS, J. ARNO[1], C. CASTANE[1] and R. ALBAJES[2]

1 : Unitat d'Entomologia Aplicada, IRTA, Centre de Cabrils, 08348 Cabrils, (Barcelona), Spain
Fax:(93) 753 3954 E-mail : Gabarra@cabrils.IRTA.es
2 : Area de Proteccio de Conreus, Centre UdL-IRTA, 25006 Lleida, Spain

This work evaluates lepidoptera eggs and larvae mortality due to natural enemies in order to incorporate the rates of natural parasitism and predation into the insecticide treatment decisions. Eggs and larvae of *Helicoverpa armigera* and *Chrysodeixis chalcites* were weekly asmpled during 1994 and 1995 in seven pole tomato fields where an IPM program was applied.
Telenomus spp. and *Trichogramma* spp. were the egg parasites identified during both years. In 1995 *H. armigera* egg parasitism exceeded 11 % in all the fields monitored and in one field was higher than 50 %. In 1994 parasitism was lower (0 % to 7.5 %) than in 1995. For *C. chalcites* in 1994 the 0 % of eggs were parasitized and ranged beween 7.5 % to 21 % in 1995. The natural parasitism of *H. armigera* larvae was very variable : 28 % in 1994 and 4 % in 1995. Parasitism of *C. chalcites* larvae was 0 % in 1994 and 0.6 % in 1995. In both year and for eggs and larvae the natural parasitism of *H. armigera* was always higher than that for *C. chalcites*.
The most abundant polyphagous predators in these tomato fields were *Macrolophus caliginosus* and *Dicyphus tamaninii* (Heteroptera : Miridae). These predators were reponsible, in one field, for to 75 % of egg mortality during several weeks.

Nosema disease of the House fly parasitoid *Muscidifurax raptor* and its impact on fly IPM programs

Christopher J. GEDEN[1], James J. BECNEL[1], Stefan J. LONG[2] and Donald A. RUTZ[2]

1 : USDA, ARS, Medical and Veterinary Entomology Research Laboratory, 1600 SW 23rd Dr., Gainesville, FL 32604, USA
Fax : 352 374 5912 E-mail : cgeden@nervm.nerdc.ufl.edu
2 : Department of Entomology, Comstock Hall, Cornell University, Ithaca, New York 14853,USA

IPM programs for house flies that include releases of the parasitoid *Muscidifurax raptor* can be highly effective, economical, and reduce pesticide usage by 80%. Such programs are compromised when parasitoid colonies are infected with the microsporidium *Nosema muscidifuracis.* Infection with this microorganism results in a debilitating, systemic and chronic disease that reduces the effectiveness of the parasitoids by 90%. Surveys of colonies have revealed the presence of disease in virtually every colony of *M. raptor* and *M. zaraptor* in the world, some colonies of *Urolepis rufipes*, but no *Spalangia* species. Natural prevalence rates in the field are low (1-11%), but disease is rapidly amplified to 100% under mass-rearing conditions because of superparasitism and cannibalism among parasitoids. Adult female parasitoids transmit the pathogen transovarially (100%) and both males and females can become infected (16-25%) by feeding on wounded, infected parasitoid immatures within host puparia. Mass-release of diseased parasitoids has the perverse effect of undermining biocontrol efforts by introducing massive amounts of *Nosema* inocula into natural systems where it is otherwise rare. The disease can be managed by immersing infected parasitoid eggs in a hot (47°C) water bath for 30-60 minutes, but parasitoid mortality is high. Eliminating disease from an established colony of *M. raptor* resulted in a ten-fold increase in parasitoid fecundity.

Mathematical modelling supporting evaluation of biological control efficacy

G. GILIOLI[1], M. CALVITTI[2] and G. DI COLA[1]

1 : Dipartimento di Matematica, Università degli Studi di Parma, 43100 Parma, Italy
Fax : +39 521 902350
E-mail : DICOLA@PRMAT.MATH.UNIPR.IT
2 : ENEA, INN-BIOAGR, CRE Casaccia, 00060 S. Maria di Galeria, Roma, Italy

In order to explore the role of mathematical modelling to guide the biological control practices and evaluation of their efficacy the use of the predatory mite *Phytoseiulus persimilis* Athias Henriot as control agents of *Tetranychus urticae* Koch in Mediterranean greenhouses is analyzed.
We propose a mathematical model for a realistic simulation of the local dynamics, in terms of biomass variation, of this acarine predator-prey system. The parameter introduced in the model are production and elimination rate and control efficiency ; the parameters which are not directly observable are estimated by using a simulation model of the population dynamics, which takes into account the stage structure of prey and predator.
The representation of trophic interactions in prey-predator system is based on composite models described by a system of ordinary differential equation in terms of demographic and metabolic parameters. The model is able to represent, given a specific infestation level, the effects of quantity and timing of predators introduction on prey dynamics ; so the knowledge obtained with this model provides insight into predator-prey interactions with implications for management strategies and may represent the fundamental component of a Supporting Management System.
The results of simulation experiments are then subjected to experimental validation by means comparing simulating population dynamics with population dynamics observed in experimental greenhouses.
It is possible to produce an extension in the model range of application. In fact the model, associated with a data base, may be able to produce knowledge suitable for different agroecosystem management involving acarine predator-prey systems (including different type of cultivation and environmental and field conditions).
Furthermore the results may be summarized in a diagram for a more manageable use by farmers. These graphs show both the results of simulation studies and the empirically derived decision rules, to indicate whether biological control is sufficient to control mite populations.

Leafminers (*Liriomyza* sp.) in Portugal : a natural control strategy

Maria GODINHO and António MEXIA

Secção autónoma de Protecção integrada,
Instituto superior de Agronomia, Tapada da Ajuda,
1399 Lisboa Codex, Portugal
Fax : 01 363 50 31

Studies conducted in several greenhouses in the Oeste Region of Portugal, aproximately 40-50 Km north of Lisbon, revealed a gradative situation related whith the economic importance of the leafminer pest species on vegetable crops.
The parasitoids complex appears to be the key-factor responsable for the differences between agroecosystems. According to this, the studies were orientated to assess the role of the parasitoids complex in such pest impact.
The pests species present, their relative importance and especially the patterns of the different parasitoids complex either in chemical control greenhouses or in greenhouses under IPM systems are presented and the strategies for the leafminers control in Portugal are discussed.
These results are the basis to discuss the importance of the local studies on the development of biological control techniques in different agricultural systems. This situation revealed particular importance in mediterranean climate greenhouses since natural enemies are present in abundance especially leafminers parasitoids.

Possibilities of using natural parasitoids to control the Sugarcane internode borer *Chilo sacchariphagus* (Lep : pyralidae) in Reunion Island

R. GOEBEL, J.M. BEGUE and M. LATCHOUMAYA

CIRAD-Réunion, Laboratoire d'Entomologie, Station de la Bretagne,
97487 Saint Denis cedex, la Réunion, France

The Spotted stem borer *Chilo sacchariphagus* is a major pest for the sugarcane farmers in Reunion Island and Mauritius. Heavy infestations can cause important losses is cane yield and sugar recovery. In these traditional sugar countries, entomological programmes are focused on biological control for many years because of relative inefficiency of chemical control and because of environmental considerations.
Use of natural or introduced parasitoids against graminaceous stem borers is a widespread practice, with varying results. In Reunion Island, common natural species on *C. sacchariphagus* are egg parasitoid *Trichogramma chilonis*, larval parasitoids *Cotesia flavipes* and pupal parasitoid *Tretrastichus howardii*. Release of introduced tachinids flies from Asia in the seventies have failed. Paradoxically, little or none studies were carried out on indigenous species and inondative experimental releases have never been tested.
Possibilities of using a natural parasitoid complex as a suitable biological control in sugarcane farms are discussed in this paper.

Problems of the introduction of the biological control in the palm grove in Elche, Spain, against the Red scale insect of the date palm tree : *Phoenicococcus marlatti*, Cockll.

S. GOMEZ VIVES

Estacion Phoenix, Hort del Gat., 03203 Elche, Spain
Fax : 34 65423706

Phoenicococcus marlatti. Cockll. is an homopterous insect widely spread all over the palm groves in the world. Usually considered as a minor pest but, occasionally, like in Elche, it produces several and grave damages in the palm date tree : drying of leaflets and rachis, weakening of the tree, drooping of the date production, introduction of other pests like fungus.
The scale is found deep inside the crown, massing the tight leaf bases, protected from dry and heat. This location makes difficult a chemical control of this pest with pesticides of contact or inhalation, moreover the use of systemics is known to have a low efficiency.
The particular biotope of this scale and the fact that the palm grove in Elche is closely interweaving with the urban structure of the city makes essential the research on biological control against this pest.

Problem specification workshops as a tool for determining opportunities in the development of an integrated pest management program for greenhouse crops in Australia

Stephen GOODWIN, Frances PARKER and Larissa WILSON

NSW Agriculture(Goodwin), Horticultural Research and Advisory Station,
PO Box 581, Gosford NSW 2250, Australia
Fax : 61 43 481910
E-mail : goodwis@agric.nsw.gov.au

To date, pest management practices in greenhouse crops (nursery, cutflower and vegetable) in Australia, continue to rely on regular pesticide application. Growers have not endorsed an IPM approach, and crop monitoring practices are almost non-existent. A national greenhouse crops IPM program involving multi R&D agency collaboration in the States of Queensland, New South Wales and Victoria has been developed.
Problem Specification Workshops (PSWs), which aim to involve end users in the design of an IPM program, were conducted in each of the above States at the commencement of the project. Each PSW was comprised of a group of approximately 30-35 key stakeholders including growers, consultants, chemical industry, marketing, greenhouse manufacturers, biocontrol producers, R&D fund providers and R&D providers. The workshop process, facilitated by staff of the Cooperative Research Centre for Tropical Pest Management (CTPM), involved a single day where problem definition, identification of opportunities and constraints for improvements, and action plans to facilitate key opportunities were discussed. The PSW is a technique developed by the CTPM, Brisbane specifically for the development and implemented of IPM programs.
The PSW process is regarded as a key element in the construction of a successful IPM program, and to obtain broad adoption of the IPM processes. The results of the PSW and subsequently developed plans for the adoption of IPM in greenhouse crops in Australia are presented.

Reliance on the use of IGRs - a major obstacle to the success of IPM on Citrus in Southern Africa

V. HATTINGH and B.A. TATE

Outspan Citrus Centre, PO Box 28, Nelspruit 1200,
Republic of South Africa

IGRs have been widely promoted as being ideal IPM compatible pesticides. The IGR pyriproxyfen has been widely used in southern African citrus as an effective treatment for red scale *Aonidiella aurantii,* which is resistant to organophosphates. Field experience indicated that this has had considerable direct and indirect detrimental effects on IPM. Bioassays confirmed that this and other IGRs are highly detrimental to certain coccinellids and hymenopteran biocontrol agents of economic importance. Therefore certain IGRs cannot be considered compatible with IPM on citrus in Southern Africa. The extreme persistence and widespread contamination resulting from the agricultural use of some of these products present a major obstacle to maximisation of biocontrol in IPM and the success of future biocontrol projects. It also gives rise to concern about potential environmental contamination. Use of the more disruptive products is being phased out and comprehensive determination of the non-target effects will be required before any further registrations of IGRs can be supported.

Evaluation of parasitoid attributes for successful biological control : an adaptation of some parasitoids for maintaining their reproductive capability during periods of host deprivation

Y. HIROSE

Institute of Biological Control, Faculty of Agriculture,
Kyushu University, Fukuoka 812-81, Japan
Fax : 81 92 642 3040
E-mail : hirose@grt.kyushu-u.ac.jp

Long-lived parasitoids are known to delay oviposition during periods of host scarcity. A question is how a level of the delayed reproduction is maintained. This may depend on the length of the prolonged periods, but levels of the delayed reproduction in many species of long-lived parasitoids appear to be reduced greatly after the long period of host absence. However, there is evidence that females of some species of *Telenomus* (Hymenoptera : Scelionidae) retain their ability to reproduce at high levels for a long period, and that this contributes to two successful cases of biological control.

The first case is that of classical biological control of the geometrid pest *Oxydia trychiata* by *T. alsophilae* introduced from North America into Colombia. Females of this parasitoid live over four months and retain their ability to reproduce at high levels for such a long period.

The second case is seen in naturally-occurring biological control of the Pine moth *Dendrolimus spectabilis* by *T. dendrolimi* in Kashima district in Japan. Females of this parasitoid live during two month period of host absence between two host generations to cause a high rate of parasitism in these generations, suggesting that they retain the ability to reproduce at high levels for the host absence period. *T. dendrolimi* is pro-ovigenic and has a high r (intrinsic rate of natural increase), but this parasitoid is long-lived and maintains its reproductive capability for a long period of host absence. The compatibility of pro-ovigenic mode of reproduction and high r with great longevity in *T. dendrolimi* may be ensured by its adaptation for maintaining reproductive capability during a long period of host deprivation. Parasitoid attributes, such as longevity, fecundity, mode of reproduction and r, in the two *Telenomus* species and other many parasitoid species are discussed in relation to the success in biological control.

Compatibility between a parasitoid and a fungal pathogen of *Bemisia tabaci* (Homoptera : Aleyrodidae)

W.A. JONES[1] and T.J. POPRAWSKI[1,2]

1 : USDA, ARS, Subtropical Agricultural Research Laboratory, Biological Control of Pests Research Unit, 2413 E. Hwy. 83, Weslaco, TX 78596, USA
Fax : 210 969 4888 E-mail : w-jones@pop.tamu.edu
2 : Texas A&M University, Texas Agricultural Experiment Station, Weslaco, TX 78596, USA

Laboratory tests were conducted in 1994-1995 to determine if parasitoids are compatible with the use of *Beauveria bassiana* for managing the Sweetpotato whitefly, *Bemisia tabaci*, Biotype B (= *B. argentifolii*). The lethal and sublethal effects of direct sprays of fungal spores (at 1000 spores/mm^2 of strain GHA) to whitefly nymphs previously parasitized by an *Eretmocerus* sp. from Texas was tested 1, 2, 3, 9, and 13 d. after parasitoid egg deposition. Treatments were : fungus in an aqueous carrier containing 0.02% Tween 80 surfactant, 0.02% Tween 80 solution without fungus, water alone (control), and fungus spray to unparasitized hosts. Tests were conducted using rooted sweet potato leaves as substrate. The results clearly demonstrated that parasitized *B. tabaci* are immune to infection beginning three days after parasitization. Significant infection was recorded one and two days after parasitization, apparently before eclosion of parasitoid eggs. Resistance to infection was not due to inhibition of spore germination. Measurement of sublethal effects showed that longevity of emerged parasitoids surviving spore applications at 9 and 13 days as immatures, was slightly but significantly lower, but no different when applied at three days old. Four-day fecundity of mated females surviving the 13-day-old treatments was not significantly different from the water treatment.

Biological Control : an important component of the IPM approach to control the Citrus leaf miner *Phyllocnistis citrella* (Lepidoptera : Gracillaridae) in two govenorates of Egypt

Hans KAACK, Ahmed R. HAMED, Werner GASSERT, N.Z. SOLIMAN and Makram S. BARRSSOUM

Egyptian-German IPM Project c/o GTZ-PAS,
4d El Gezira Street, Zamalek, Cairo, Egypt
Fax : 0020 2 360 3972
E-mail : 103205.305@compuserve.com

Citrus is the most important fruit crop grown in Egypt. In 1993 approximately 160.000 ha were planted. Since 1994 the crop has been attacked by the Citrus Leaf Miner, *Phyllocnistis citrella* (Lepidoptera: Gracillaridae). The larvae of the small butterflies mine on young leaves, which causes damage to citrus especially in the young plantations and in the nurseries. As new flushes are found all year round the CLM has ideal living conditions.

Since 1994, the Egyptian-German Integrated Pest Management Project, has been working on Integrated Control of this pest in the govenorates of Beni Suef and Ismailia. Three indigenous parasitoids of the CLM were found and identified (*Pnigalio sp., Baryscapus sp., Cirrospilus pictus*). All of them are unspecific parasitoids of leaf mining Lepidoptera, Diptera and Coleoptera. Larval parasitization seldom exceeds 30 %. In order to increase the parasitization, the Egyptian Ministry of Agriculture has banned the use of all insecticides except summer oil, which has proven to be nearly as effective as synthetic insecticides but less harmful to the natural enemies.

The attempt to increase the natural parasitization, is accompanied by awareness campaigns (posters, handouts, field days) at the farmers level, in order to prevent the use of insecticides.

To enhance these efforts, a programme of classical biological control was started in spring 1996. The larval endoparasitoid *Ageniaspis citricola* (Hymenoptera: Encyrtidae) was imported, mass reared and released initially in the two Govenorates Beni Suef and El Fayoum in Upper Egypt.

A. citricola is host specific to *P.citrella*. The polyembryonic larval parasitoid has a much higher multiplication potential (1-6 adults from 1 CLM larva) than the indigenous larval ectoparasite (1 adult from 1 CLM larva). It is hoped that *Ageniaspis citricola* will establish quickly in Egyptian citrus orchards.

Maximising parasitoid fitness during mass production : effect of size of the mealybug *Pseudococcus viburni* (Hom., Pseudococcidae) on survival, sex ratio, developmental duration and size of the parasitoid offspring in *Leptomastix epona* (Hym., Encyrtidae)

Filitsa KARAMAOUNA and Mike COPLAND

Department of biological Sciences, Wye College, University of London,
Ashford, Kent TN25 5AH, United Kingdom
Fax : 01233 813140
E-mail : m.copland@wye.ac.uk

The solitary koinobiont endoparasitoid wasp *Leptomastix epona* was tested on five different size classes of the mealybug *Pseudococcus viburni*. Each class represented predominantly one host stage (first, second and third instar nymph, young adult, preovipositing adult). The classes were exposed, one at a time or with the other size classes, to female wasps before and after they had gained experience of host size. Results showed that mealybugs larger than 0.5 mm (second instar nymph) are susceptible, but crawlers are not attacked. Both susceptibility and preference of the host increases with the ascending host size classes ; larger hosts (adult stage) are mummified in significant greater numbers than smaller ones (third instar nymph). Survival of the parasitoid does not differ between the size classes of the mealybug. Sex ratio in emerging parasitoid offspring is female biased in larger hosts (1.83-3.33 mm). Wasp experience of the host sizes (24 h) does not seem to influence preference, efficacy or sex allocation strategy of *L. epona*. Both male and female parasitoids develop faster in larger than in smaller hosts. Larger wasps emerge from larger hosts and with higher numbers of mature eggs present. In terms of mass production, the young adult stage (1.83-2.33 mm) of *P. viburni* seems to be the most suitable for *L. epona* development; with greater successful parasitism, shorter developmental duration and a higher female biased sex ratio.

Combining classical biological control and augmentation : "Multiple releases" of an exotic parasitoid [*Cales noacki* (Hymenoptera: Aphelinidae)] for controlling *Aleurothrixus floccosus* (Homoptera: Aleyrodidae) in Greece

P. KATSOYANNOS[1] and D.C. KONTODIMAS[2]

1 : N.AG.RE.F. at Benaki Phytopathological Institute,
145 61 Kifissia, Greece
Fax : 3018077506
2 : Benaki Phytopathological Institute, 145 61 Kifissia, Greece

A small colony of 40-50 individuals of the parasitoid *Cales noacki* was imported from Valencia, Spain, into Greece in November 1991 for the biological control of *A. floccosus* which had invaded Attica earlier in the same year. The imported *C. noacki* were initially kept and reproduced in the laboratory, then mass-produced in the insectary, until adequate numbers were obtained for implementing a strategy of "multiple releases" dictated by the speed and direction of *A. floccosus'* dispersion.

In 36 districts of Attica, 10-20 thousand *C. noacki* were released at each of 74 sites, four times between mid March and early October 1993. Observed rates of parasitism in March 1994 ranged between 50-97% in 21 districts, 30-50% in 12 districts and 10-20% in 3 districts. By December 1994, rates of parasitism were between 50-99% in 30 districts and 40-50 % in the remaining 6.

In other parts of Greece, *C. noacki* was released when and where *A. floccosus* was discovered infesting citrus. Thus, *C. noacki* was released at 33 sites in 8 different regions, using the method described above, between mid May and late October 1993. Observed rates of parasitism in March 1994 ranged between 60-76% in 4 regions, 38-43% in 2 regions and 1-2% in 2 regions in the department of Corinthe which had been excessively treated with chemicals. During the following year, the same method was used for releasing *C. noacki,* between mid March and early October 1994, at a total of 345 sites in 27 different regions of Greece. Observed rates of parasitism in March 1995 and thereafter ranged between 50-99% in 21 regions and between 12-44% in the remaining 6.

In addition to its widely aknowledged economic and ecological benefits for Greece, the spectacular success of this operation was also a countrywide demonstration of the practical importance and efficiency of biological control.

Plutella xylostella (Lep. Yponomeutidae) with special reference to its parasitoids in South Africa

Rami KFIR

Plant Protection Research Institute, Private Bag X134,
Pretoria 0001, South Africa
Fax : +27 12 3235275
E-mail : PPRIRK@PLANT4.AGRIC.ZA

Diamondback moth (DBM), *Plutella xylostella* (L.) (Lep.: Yponomeutidae), which is considered to be of European origin, is the most destructive pest of cole crops in the world. In many countries the pest has developed resistance to most insecticides including *Bt*. Most biological control efforts against DBM in these countries have failed. The pest status of DBM in South Africa is lower than in other countries with similar climates. As very little work has been done on DBM in Africa a research project was initiated. Moths, larvae and parasitoids were active throughout the year. Infestations were low from January to September and higher during October-December. Parasitism levels were high reaching 90-100% on several occasions. Seventeen parasitoids and hyperparasitoids were identified:
Egg-larval parasitoids : *Chelonus curvimaculatus* Cameron (Braconidae) ; *Chelonus* sp.
Larval parasitoids : *Cotesia plutellae* (Kurdjumov) (Braconidae) ; *Apanteles eriophyes* Nixon (Braconidae), known only from South Africa ; *Peribaea* sp. (Tachinidae).
Larval-pupal parasitoids : *Diadegma* sp. (Ichneumonidae), an undescribed species ; *Oomyzus sokolowskii* (Kurdjumov) (Eulophidae).
Pupal parasitoids : *Diadromus collaris* Gravenhorst (Ichneumonidae), males were abundant whereas in Europe it is a uni-parental species; *Tetrastichus howardi* (Olliff) (Eulophidae); *Brachymeria* sp. A (Chalcididae); *Hockeria* sp. A (Chalcididae). Hyperparasitoids : *Mesochorus* sp. (Ichneumonidae); *Pteromalus* sp. (Pteromalidae); *Aphanogmus fijiensis* (Ferrière) (Ceraphronidae); *Eurytoma* sp. (Eurytomidae); *Brachymeria* sp. B; *Hockeria* sp. B; *Proconura* sp. (Chalcididae).
The large number of parasitoid and hyperparasitoid species associated with DBM in South Africa indicate a very long association between the parasitoids and the pest in the region. In addition, the fact that 175 species of Brassicaeae have been recorded in South Africa is a further indication that DBM might have evolved in southern Africa. It is suggested that some of these parasitoids would be considered for introduction as biological control agents into countries where DBM is a serious pest.

Prospects of controlling Blackleg of Canola/Rapeseed with a bacterium

P.D. KHARBANDA[1], J. YANG[1], P. BEATTY[2], S. JENSEN[2] and J.P. TEWARI[2]

1 : Alberta Environmental Centre, Vegreville, Alberta, T9C 1T4, Canada
Fax : 403/632 8379
2 : University of Alberta, Edmonton, Alberta T6G 2E9, Canada

Blackleg (*Leptosphaeria maculans*, conidial state: *Phoma lingam*) causes serious economic losses to canola/rapeseed (*Brassica napus* and *B. rapa*) in Canada and several other temperate parts of the world. A bacterium (*Bacillus* sp.) recovered from canola roots was found to be inhibitory to *L. maculans* in culture. Subsequently, several studies were conducted in the laboratory and growth chamber to characterize the antifungal metabolite and determine disease control potential of the bacterium. The antifungal material is found associated with the *Bacillus* sp. cells and can be extracted with methanol or dilute acid. Initial characterization of the metabolite suggests that it is a small lipophilic peptide with a molecular weight of about 2,000 Da and a blocked N terminus. Preliminary studies suggest that the antifungal agent works synergistically with the fungicide propiconazole in reducing the amount and survivability of *L. maculans* on overwintering infected canola stubble. Studies on the spectrum of activity indicate that the antifungal metabolite is active against several other fungi pathogenic on canola, *i.e. Alternaria brassicae, Fusarium avenaceum*, *Pythium* sp., *Rhizoctonia solani* AG2-1, and *Sclerotinia sclerotiorum*. None of the fungicides, *i.e.* propiconazole, prochloraz, or iprodione, or the herbicides, *i.e.* clopyralid, ethametsulfuron-methyl, or sethoxydim, which are registered for use on canola were deleterious to the growth of the bacterium on nutrient agar. Studies are underway to develop bacterium-infested compost suppressive to the blackleg disease. These strategies may lead to the development of integrated control of blackleg and other diseases of canola.

Optimising biological control of glasshouse leafhoppers (*Empoasca decipiens*) by computer simulation

Neil A.C. KIDD

University of Wales, School of pure and applied Biology,
Cardiff CF1 3TL, United Kingdom
Fax : 44 1222874305
E-mail : sabnk@cardiff.ac.uk

The Green leafhopper, *Empoasca decipiens*, has in recent years become an important pest of glasshouse sweet peppers in both Holland and the United Kingdom. When leafhopper numbers are high in midsummer, their feeding produces highly visible blemishes on the skin of the fruit, reducing its quality and saleability. Control of the leafhopper has to involve methods which do not conflict either with existing biological controls against other glasshouse pests, with the use of bees in glasshouses to improve pollination, or with the demands from supermarkets for reduced pesticide usage. The target of this project was therefore twofold : (1) to find one or more suitable biological control agents of the Green leafhopper and (2) to find the best strategy for introducing them in order to maximise control.
Since the larval and adult stages of the Green leafhopper can move very rapidly over plant surfaces, insect predators currently used against other glasshouse pests are unlikely to prove effective. One egg parasitoid species (*Anagrus atomus*, Mymaridae), however, was found to have considerable potential as a biological control agent. It is relatively easy to culture and also to mass-release using a simple delivery system.
Employing a user-friendly, interactive computer simulation model, the grower can precisely time the introductions to optimum effect. The most effective parasitoid release strategy involves five mass-releases at five-day intervals during August. Use of the model avoids the need for both insecticides and long-term detailed monitoring of leafhopper numbers in the growers' glasshouses.

Plant age and the resistance of sugarbeet to aphids

N.B. KIFT and A.F.G. DIXON

IACR Broom's Barn, University of East Anglia, Higham,
Bury St Edmunds, Suffolk, United Kingdom
Fax : 01284 811191
E-mail : N.KIFT@uea.ac.uk

The collapse of virus transmitting aphid populations on Sugar Beet (*Beta vulgaris* L. cv. Saxon) from late June through July has been shown to coincide with population peaks of coccinellid predators (Van der Werf *et al*, 1992) and other studies have shown the exclusion of predators has increased aphid numbers (Wratten and Pearson, 1982).

Whilst predation plays a role in determining aphid abundance on sugar beet there are a number of other factors that effect aphid mortality on the crop such as weather and plant quality.

During June and July sugar beet growth changes from mainly from above ground development to increased root production (Milford and Watson, 1971. Milford, 1973). These changes are thought to effect aphid mortality, with adult mortality per day in late June and July being twice that seen in early June.

This paper describes the negative effects of plant quality from early June until mid July on the survival of adult *Myzus persicae* (Sulzer) when feeding on fully expanded leaves of sugar beet. Mortality is increasingly associated with the occurrence of a dark deposit in the aphid mid gut as plants develop. The nature of this deposit is organic, however, little else is known about its composition.

The decline in host quality is probably due to secondary plant compounds in sugar beet and is currently being investigated.

Testing of *Trichoderma* strains from the former Soviet Union for efficacy against *Pythium ultimum* and *Rhizoctonia solani*

E. KOCH

Federal Biological Research Centre for Agriculture and Forestry,
Institute for biological Control, Heinrichstrasse 243,
64287 Darmstadt, Germany
Fax : 06151 407 90

The potential of fungi belonging to the genus *Trichoderma* as biocontrol agents of soilborne diseases is well known. However, use of *Trichoderma* on a commercial scale is still very limited. Low and unreliable efficacy of *Trichoderma* products are often seen as major obstacles for commercial use.
In the Soviet Union, preparations based on *Trichoderma spp.* were widely used in vegetable production in glasshouses. Within the frame of an INTAS project, *Trichoderma* isolates from the Institute of Biological Plant Protection (former All-Union-Institute) in Kishinev (Moldavia) were tested in the glasshouse against *Pythium ultimum* Trow on cucumber and against *Rhizoctonia solani* Kühn on peas. The four isolates obtained were designated as *T. lignorum* (two isolates), *T. harzianum*, and *Gliocladium virens*.
For glasshouse testing, the fungi were raised in a mixture of wheat bran, vermiculite and Czapek-Dox medium. The inoculum thus obtained was mixed (fresh or after drying and grinding) into the potting substrate. In preliminary tests with different concentrations of the antagonists, the isolates designated *T. harzianum* and *G. virens* gave a dose response. In further tests with these two isolates, the efficacy against *P. ultimum* on cucumber varied between 10 and 53 %. The efficacy against *R. solani* on peas was 41 to 82%. Parallel to the increase in plant stands, the shoot dry weight increased significantly.
The two isolates designated *T. lignorum* gave only weak disease control.
In pathogen-free potting substrates, all isolates caused a reduction in shoot dry weight which was, however, statistically not significant. At 10 - 26°C, the isolates designated *T. harzianum* and *G. virens* grew more slowly on agar media than isolates prepared from the commercially available *Trichoderma* products BINAB, SUPRESIVIT and TRICHODEX.

Selection of antagonists inhibiting *Erwinia amylovora* and Ina$^+$ *Pseudomonas syringae* in Czech Republic

B. KOKOŠKOVÁ, V. KÙDELA, J. ZÁMEÈNÍK and A. BILAVÈÍK

Research Institute of Crop Production, Drnovská 507,
161 06 Prague 6, Czech Republic
Fax : 422 365228

More than 1000 isolates of yellow saprophytic bacteria obtained from rosaceous plants (hawthorn, apple, pear, mountain ash) were evaluated as potential biological control agents against *Erwinia amylovora* (*Ea*). The antibiotic ability of the isolates was tested on solid medium in petri dishes; 47-57 % of the strains obtained from various plants showed antibiotic properties. Biocontrol ability of selected bacterial strains and several chemicals (including streptomycin) was tested in the immature pear fruit assay, on blossoms of rosaceous plants (on detached branches in a growth chamber) and on apple and pear blossoms (on trees growing in a nethouse). Incidence and intensity of blossoms blight was evaluated 7 days after inoculation with *Ea* and expressed as % effectiveness.

Three of the six selected bacterial strains, determined as *Erwinia herbicola* (*Eh*) exhibited considerable activity against blossom blight of apple trees (66%) and pear trees (47%). The best activity (73%) was obtained with strain *Eh* 170. Among chemicals tested streptomycin showed the best effectiveness (80%) and fosetyl-Al (Aliette 80 WP) only 34 %.

About 85 isolates of fluorescent pseudomonads and saprophytic erwinias were screened for inhibitory activity against INA$^+$ *Pseudomonas syringae* (INA$^+$*Ps*) on solid medium in petri dishes. About 40% of the tested pseudomonad and erwinia isolates exhibited some inhibitory activity against INA$^+$*Ps*. When sprayed on detached blossoms of apple in chamber the best of the selected antagonistic isolates shifted the ice nucleation activity of INA$^+$ *Pseudomonas syringae* by 4°C and thereby avoided injury by freezing of blossoms. However, the blossoms connected with spurs froze at -4.3°C due to intrinsic ice nuclei in spurs.

Are releases of generalist predators always the best way in the biological control of phytophagous pests ? Some case studies in French vineyards

S. KREITER[1], P. AUGER[1], M.S. TIXIER[1], G. SENTENA[2], M. WEBER[3] and G. SALVAT[4]

1 : ENSA. M/INRA, laboratoire de Zoologie, 2 place Viala, 34060 Montpellier cedex 01, France
Fax : 67.52.15.54 E-mail : Kreiter@ensam.inra.fr
2 : ITV (Centre technique interprofessionnel de la Vigne et du Vin), 6 rue du 16[e] Chasseur, 21200 Beaune, France
3 : Service régional de la Protection des Végétaux (SRPV) Languedoc-Roussillon, rue d'Alco, 34034 Montpellier cedex 01, France
4 : CIVAM Corse, Station "Les Cazelles", 21230 San Giuliano, France

"Generalist", *i.e* those species which consume a wide variety of foods, including some of plant origin such as pollen, are the dominant members of the phytoseiid mite (Acari, Phytoseiidae) fauna in some perennial crops.
Biological attributes commonly evident in "generalist" in contrast to "specialist" phytoseiids include a lower reproductive potential on mite prey, close association with certain plant species and sometimes a kind of specificity or specialisation for the host plant, population increases in the absence of mite prey, lower responses to kairomones emitted by preys and intraplant distribution unrelated to that of preys. In French perennial crops, the main species of phytoseiid mites are generalist predators as shown by a ten years survey of phytoseiid mites. The two most important species are *Typhlodromus pyri* and *Kampimodromus aberrans.* Inundative and augmentative releases of generalist phytoseiid mites as natural enemies of phytophagous mites have occurred in many countries all around the world. In perennial crops in France, experiments of field releases have shown various results from success to failure. Major factors affecting success or failure in such crops are reviewed. Limits of such classical biological control strategies in French vineyards are also given concerning research and practices view points.
Case studies of important colonizations of vineyards by the two main phytoseiid predators from surrounding vegetation or crops are presented, mainly in Burgundy and Languedoc-Roussillon. These colonizations have occurred in vineyards sprayed with non toxic pesticides for phytoseiid mites. In such vineyards, phytoseiid mite densities increase rapidly. The phenomenon seems very quick and concerns generally very wide areas. Interests and limits of colonization vs. release in agricultural practices are discussed. Prospects of studies of origins, mechanisms and determinisms of the colonization for better understanding and management are also given.

Biological control of the larvae of Turnip saw-fly (*Athalia rosae* L.) by *entomopathogenic nematode, Steinernema carpocapsae*, in Hungary

Attila LUCSKAI and Gyula SARINGER

Pannon University of Agricultural Sciences,
Georgikon Faculty of Agriculture, Plant Protection Institute,
8360 Keszthely, Deák F. u. 57, Hongrie
Fax : 36/ 83/ 315105

The Turnip saw-fly is one of the major pests in the culture of rape. The larval stages cause damages three times per year in our rape cultures.The largest harm is in autumn. Entomopathogenic nematodes of the family Steinernematidae are among the common insect parasites in the field. They attack mainly insect stages living in the soil, because moisture conditions are favourable for a prolonged survival of infective nematode larvae. The entomopathogenic nematode, *Steinernema carpocapsae* Weiser (Rhabditida: Steinernematidae), shows potential for biological control of Turnip saw-fly in Hungary.

The tests against the larvae of Turnip saw-fly were carried out in laboratory and in small parcels. The infective jeveniles of *S. carpocapsae* were applicated against the larvae of the Turnip saw-fly. In the laboratory tests, the concentration of 100 nemas/ml was effective in the control of the larvae of target pest. In the parcel tests, 47 % larva mortality was observed using 4.9×10^{10} nemas/1 m^2 dose on the plant leaves. Higher larva and pupa mortality was achieved at the same nematode concentration in treatment on the soil surface.

In September and in October is the optimal time of application of the entomopathogenic nematode against the Turnip saw-fly. In this time the temperature and soil moisture is favourable for the pest control with entomopathogenic nematodes. Even this season is caracterised by the dew formation for help the nematode treatment on the plant leaves.

Our preliminary results of reducing the larval damages of the Turnip saw-fly by using nematodes of the species *S. carpocapsae* suggest that the rate of mortality of the pest larvae can be increased by an introduction of the entomopathogenic nematode. In the following years, this method has a perspective in the control of an outbreak of Turnip saw-fly.

Extension biological control educational materials and programs in the North-Central United States

Susan E. Rice MAHR[1], Daniel L. MAHR, Douglas A. LANDIS[2], Robert O'NEIL[3], Clifford S. SADOF[3], Robert N. WIEDENMANN[4] and Robert J. WRIGHT[5]

1 : Department of Entomology, University of Wisconsin, Madison, WI 53706, USA
Fax : 608 262 3322 E-mail : smahr@entomology.wisc.edu
2 : Department of Entomology, Michigan State University, East Lansing, MI 48824, USA
3 : Department of Entomology, Purdue University, West Lafayette, IN 47907, USA
4 : Illinois Natural History Survey, 607 East Peabody Dr., Champaign, IL 61820, USA
5 : University of Nebraska, SC Res & Ext Center, PO Box 66, Clay Center, NE 68933, USA

Numerous reports indicate technology transfer is critical to implementing biological control. Historically in the United States, pest management recommendations have focused on chemical control. Although there is a well developed biological control technology, the actual adoption of these practices into IPM programs lags far behind the implementation of other approaches, especially pesticide use. A program to bridge the existing gap between developed biological control technologies and actual implementation in IPM programs was initiated in the North Central United States. Information designed for pest management practitioners (crop consultants, extension agents, state extension specialists, etc.) is provided in three main ways : 1) state and regional biological control conferences; 2) regional publications on biological control, and 3) a monthly newsletter on biological control.

A two to three day regional conference on biological control has been held annually at different sites since 1993. Additional state conferences modeled on the regional one have also been held. The conferences discuss the biology, identification and use of natural enemies, and various workshops demonstrate principles for specific crops.

A series of extension bulletins to foster the use of biological controls in various crops is being produced. The first is a general introduction to biological control. The others focus on a specific commodity : crucifers, greenhouse crops, alfalfa, apples. Each is designed as an in-depth practical guide to provide sufficiently detailed information for implementation.

An 8-page insect biological control newsletter, Midwest Biological Control News (MBCN), is sent to all county extension offices in the 12 North Central states as well as extension entomologists and extension IPM coordinators nationally, and to over 100 private subscribers. The newsletter contains practical information to aid extension personnel in the development of their pest management educational programs.

Use of biological control in tropical tree crops : CIRAD research results

D. MARIAU, B. DECAZY, S. QUISLICI, B. MALLET,
J.P. MORIN, M. ARBONNIER and R. PHILIPPE

CIRAD, B.P 5035, 34032 Montpellier Cédex, France
Fax : 33 67 61 57 93
E-mail : mariau@cirad.fr

Research undertaken by CIRAD entomologists, specialized in tree crops, has concentrated on palms, oil palm and coconut, coffee, cocoa, fruit trees and forest species. Much of the research was intended to improve knowledge of abiotic and biotic factors that limit pest population, particularly entomophagous insects (parasitoids and predators) and entomopathogenic organisms (fungi and viruses), olfactory trapping techniques using chemical mediators (pheromones and allelochemical compounds), and visual trapping techniques.
All these investigations have made it possible to opt for particular entomophagous organisms, or develop a biological control method using an entomopathogen or a pheromone substance.
Several parasitoids and predators introductions have been successful and in some cases it has been possible to introduce cultural techniques unfavourable to certain pests. Lastly, the use of tolerant material can be recommended in a few cases.
Wherever it has not been possible to propose a biological control method, research has at least provided advice to users regarding rational chemical control methods that are more environment-friendly.

The perspectives on the transfer of production technologies of biopreparations for control of mosquitoes in resource-poor regions with a tropical climate

V.E. MATVEEV[1], V.V. SHEVTSOV[2] and O.A. KRAINOVA[2]

1 : Russian Federation Commitee of Chemistry and Oil-Chemistry
20, Myasnitskaja str., Moscow 101851, Russia
Fax : (095) 921 78 73
2 : Biotechnological Innovation Centre, 102A, Lenin str., Serpukhov,
Moscow region, 142283, Russia

Two new biopreparations Sphaerolarvicide and Kombilar for the control of mosquitoes-vectors of malaria, japanese encephalitis, filariosis and others have been developed. Sphaerolarvicide on the basis of *Bacillus sphaericus (Bs)* has advantages in comparison with biopreparations used on the basis of *B.thuringiensis* sv. *israelensis* (*Bti*).

- It produces a long larvicide effect through reproduction of *Bs.*
- It shows activity in contaminated pools but it is the most effective in a hot climate.
- It possesses more selected action than *Bti,* and it shows high activity in regard to mosquito *Culex, Anopheles* and it is inactive in regard to *Aedes.*

Kombilar on the basis of selected efficient combination of *Bs* and *Bti* has advantages :

- It produces a fast effect in regard to mosquito *Culex*, *Anopheles* and *Aedes,*
- It produces a prolonged effect through reproduction of *Bs*,
- It prevents the development of resistance in mosquitos to biolarvicides.

Different versions of equipment and technologies delivery for biopreparation production on small-scale plants in countries wich have poor resources and tropical climate using the regional raw materials and waste products of the food industry are offered. Besides, it is possible to produce biopesticides by feeding antibiotics and other biotechnological products to confuse your employeers during a year on this equipment.

Welsh Office regional enterprise grant support for biological control project using the Green Lacewing (*Chrysoperla carnea*). A successful example of State assisted technology transfer

Peter McEWEN[1] and Peter ROSSITER[2]

1 : School of Pure and Applied Biology, PO Box 915,
University of Wales, Cardiff, CF1 3TL, United Kingdom
Fax : 44 1222 450538
E-mail : SABPKM1@cf.ac.uk
2 : "Insect Habitat", Ty Pellaf, Pant-y-deri, Boncath,
Dyfed SA37 OJB, United Kingdom

Although the biological control of insect pests has great potential and is widely perceived to be of increasing importance, it can be extremely difficult to actually persuade people to part with money for this type of research. Furthermore, when the research has been carried out, funds are rarely available to exploit the ideas and techniques developed, meaning that there is a failure to transfer technology to the market. In this paper we report on our experience of state assisted technology transfer in Wales for a biological control device using the Green lacewing (*Chrysoperla carnea*).

The green lacewing is the most important lacewing with regard to its impact on arthropod pest populations in agricultural ecosystems, and is used extensively in biological control. A single adult female is able to produce several hundred eggs and each emerging larva is capable of consuming hundreds of individual prey (such as aphids, whitefly, thrips) during its lifetime.

Two basic observations of lacewing biology form the basis of our work. Firstly, adults locate suitable habitats and hosts by responding to chemical cues (semiochemicals) and secondly lacewings need to find a suitable habitat in which to survive the winter in a semi-active physiological state known as diapause which comes about in response to changes in daylength and temperature. Recorded sites for diapause include unheated parts of houses, stables and barns, the underside of bark, leaf-litter and abandoned wasps nests, and clusters of large numbers of individuals can be found at such sites. The project evaluates artificial chambers which mimic normal diapause sites, and which incorporate attractive chemicals into their design in order to maximise the rate of lacewing colonisation. The chambers will subsequently act as portable pest control units to assist in the control of insect pests in agricultural, horticultural and glasshouse systems.

The efficacy of coating bioformulations for promotion of postharvest control of fruit decay prive

Raymond G. McGUIRE

U.S.D.A., ARS, 13601 Old Cutler Road, Miami, FL 33158, USA
Fax : 305 238 9330

Fruits and vegetables are frequently coated postharvest to restore natural films lost during processing and to artificially improve color and sheen as well as reduce moisture loss during storage. Coatings, however, can also be substrates for microorganisms such as yeasts that antagonize pathogens and promote biological control of postharvest decay. Shellac and wax coatings are most commonly applied to fresh produce, but commercial products typically contain concentrations of alcohols and bases that quickly kill yeast antagonists. Reducing the amounts of these solvents, and modifying other constituents, can allow yeasts to survive in the liquid formulation until it has dried on the fruit. From this initial inoculum, yeast populations on fruit surfaces can be maintained over six months at levels between 10^5 and 10^6 cfu/cm^2, which are required for effective competition. Incorporation of the yeast epiphyte *Candida oleophila* Montrocher into these modified shellac and wax coatings, and into other films based upon cellulose or sucrose esters, can significantly reduce losses in the storage of citrus attributable to green mold caused by *Penicillium digitatum* Sacc. Use of such bioformulations should not require the need for additional stages during fruit processing, but, whether an antagonist is added within a fruit coating or applied separately, coating constituents must be compatible with the antagonist to allow populations to develop for effective biocontrol.

Biological control of Blackberry and Ragwort in Victoria

D.A. McLAREN and E. BRUZZESE

Department of Natural Resources and Environment,
Keith Turnbull Research Institute, Frankston, 3199 Australia
CRC for Weed Management Systems
Fax : 03 97852007
E-mail : dam@dca.UIC.gov.au

The blackberry rust fungus, *Phragmidium violaceum* has been having a significant impact on reducing blackberry infestations in Victoria. Trends show that blackberry biomass has been declining since 1985. This has occurred through reductions in cane length and root biomass. Daughter plant production has decreased dramatically since 1984. A regional survey shows that the rust is having its greatest impact in the higher rainfall areas of south eastern Victoria and in the North East of the state.
Of five biological control species released to control ragwort, three have established. The Ragwort flea beetles, *Longitarsus flavicornis* and *L. jacobaeae* have established (10% and 18% respectively) but have only controlled ragwort in some isolated locations and their rate of spread has been extremely slow. The ragwort crown boring moth, *Cochylis atricapitana* has been recovered from 36% of sites of which 17% have established. *C. atricapitana* is yet to control ragwort populations in Australia.

Predatory and parasitic activity of *Aphelinus asychis* after treatment with *Paecilomyces fumosoroseus* and incubation under different humidity regimes

Antonio L. M. MESQUITA[1,2], Lawrence A. LACEY[1], Cornelia-Svetlana CEIANU[3] and Rémi DABIRE[4]

1 : USDA, ARS, European Biological Control Laboratory, B. P. 4168, Agropolis II, 34092 Montpellier, France
Fax : (33) 67 04 56 20 E-mail : ebcl@cirad.fr
2 : Permanent address: Empresa Brasileira de Pesquisa Agropecuaria-EMBRAPA/CNPAT - Rua dos Tabajaras, 11, Praia de Iracema, 60060-510, Fortaleza, Ceara, Brasil
3 : Cantacuzino Institute/Medical Entomology Laboratory
4 : Institut National d'Etude et de Recherche Agricole, B.P. 910, Bobo-Dioulasso, Burkina Faso

The effect of the Hyphomycete *Paecilomyces fumosoroseus* on the predatory and parasitic activity of *Aphelinus asychis,* a common parasitoid of *Diuraphis noxia* (Russian wheat aphid) was investigated under different humidities. Three lots of 20 female *A. asychis* were treated with 5.2 x 10^4 spores/cm^2 (two times the LC_{95} for *D. noxia*). Each female was then individually maintained in a Petri plate provided with 3 barley leaves and 20 3rd instar *D. noxia*. The lid of each dish had a ventilation window covered by gauze and the edges were sealed with parafilm. Lots of 20 treated females were each held at one of the following relative humidities along with untreated control insects : 67-73% RH, 85-89% RH and 94-98% RH. Every 24 hours during one week, the surviving parasitoids were transferred into Petri dishes provided with fresh leaves and 20 aphids. After each 24 hour period of exposure and for each parasitoid, dead aphids were counted and the leaves holding live aphids were placed on separate barley plants to permit the re-establishment of the aphids. Nine days after the transfer of aphids, the mummies that had formed were counted. The total number of aphids consumed (304) and mummies formed (596) per treated groups was significantly lower than the control groups of parasitoids (588 aphids consumed and 1158 mummies formed) held at 94-98% RH due to a significant decline in predation and parasitism per female per day and due to a progressive reduction of number of surviving parasitoids each day. The emergence of the F1 generation of treated *A. asychis* was reduced (85.7%) at 94-98% RH relative to controls (90.5%) probably due to an infection by the fungus transmitted by the female at the moment of oviposition. The total

mortality of treated parasitoids submitted to 94-98% RH 7 days after treatement and the high level of mycosis in cadavers show the susceptibility of adult *A. asychis* to *P. fumosoroseus* at high relative humidity. However the compatibility of the fungus and parasitoid at lower humidities indicates good potencial for their combined use.

Host-acceptance and host-suitability of non-target tephritids as determinants of environmental risk for fruit fly biocontrol

Russell H. MESSING and Jian J. DUAN

University of Hawaii, Dept. of Entomology, 7370 Kuamoo Rd., Kapaa, Hawaii 96746, USA
Fax : 808 822 2190
E-mail : MESSING@HAWAII.EDU

Both laboratory and field host-response data using extant opiine braconid parasitoids of Asian origin were gathered to use in estimating environmental risk to non-target tephritids in Hawaii. In the lab, gravid females of *Diachasmimorpha longicaudata* and *Psytallia fletcheri* were confined with larvae of the gall-forming tephritid *Eutreta xanthochaeta* in several substrates. Females of both species showed positive ovipositional responses to larvae, but oviposition and subsequent parasitization were influenced significantly by exposure substrate. Intact galls received significantly fewer stings and ovipositions than damaged galls or normal hosts (*B. dorsalis*) in artifical diet. *D. longicaudata* on *E. xanthochaeta* developed normally with an F_1 female:male sex ratio of 1.8:1, but eggs of *P. flecheri* failed to eclose, although there was no evidence of melanization nor encapsulation.

In $21m^3$ field cages, single-choice and two-choice bioassays were used to evaluate *D. longicaudata*'s response to habitual host-habitats (*i.e.*, guava) *vs.* non-target host-habitats (*i.e.*, lantana) containing *B. dorsalis* and *E. xanthochaeta*, respectively.

The number of observed ovipositor probes and percent parasitism (as determined both by larval dissection and rearing) were significantly higher in all cases for *B. dorsalis*. No parasitoids were reared from *E. xanthochaeta*, nor did dissections reveal a single parasitoid eggs or cadaver.

Thus, although laboratory tests (similar to those normally used in quarantine) revealed *E. xanthochaeta* to be physiologically suitable for *D. longicaudata* development, field tests demonstrated that behavioral adaptations to host-habitat finding preclude successful parasitism. The utilization of these data in issuing parasitoid release permits is discussed.

Control of *Elasmopalpus lignosellus* and *Aspergillus flavus* by *Bacillus thuringiensis* reduces aflatoxins in peanuts

William MOAR[1], Kira BOWEN[2], Peiming HUANG[3], Narendra SINGH[3], Joe CHERRY[3], Cecile RANG[4] and Roger FRUTOS[4]

1 : Auburn University, Department of Entomology, Auburn, AL 36849, USA
Fax : (334) 844 5005 E-mail : wmoar@ag.auburn.edu
2 : Department of Plant Pathology, "
3 : Department of Botany/Microbiology, "
4 : BIOTROP-IGEPAM CIRAD, Montpellier, France

A strain of *Bacillus thuringiensis* (633) with lepidopteran activity was selected for high chitinase activity using an *in vitro* assay. Incubation of this strain with N-methyl-N'-nitro-N-nitrosoguanidine resulted in several colonies exhibiting clearing zones after 24 hr. when plated on nutrient agar containing chitin. One colony was selected (634) and assayed *in vitro* for fungicidal activity against *Aspergillus flavus* and *Alternaria solani*. In both cases, 634 inhibited growth of the fungi, whereas 633 did not. In peanut field trials, initial seed treatments using 634 significantly reduced *Aspergillus* spp. fungal colonization and aflatoxin concentration of the harvested peanuts when compared to the untreated control. Results of glasshouse assays of peanuts grown in pots demonstrate that 634 (used as a seed treatment) also significantly reduced plant damage due to the lesser cornstalk borer, *Elasmopalpus lignosellus*. This soil insect causes direct damage to the peanut peg and pod which also allows easier penetration by *Aspergillus*. Current research has focused on enhancing the fungicidal activity of 634 by expressing the antifungal protein, osmotin. Results of these studies and the possibility of using *B. thuringiensis* for concurrent control of more than one type of pest will be discussed.

Comparative study on reproductive activity of *Bemisia argentifolii* (Hom. : Aleyrodidae) on winter weeds

M. MUÑIZ and F.G. ZALOM

Centro de Ciencias medioambientales (CSIC),
c/Serrano 115 Dpdo., 28006 Madrid, Spain
Fax : (34)/1 5640800

Amsinckia intermedia F & M, *Brassica kaber* (DC.), *Capsella bursa-pastoris* (L.) Medic., *Lactuca serriola* L. and *Malva parviflora* L. were germinated in a growing chamber and placed in a greenhouse (25:15 °C; 70-80% R.H.). Adults of *Bemisia argentifolii* Bellows & Perring were collected from a stock colony on *Poinsettia pulcherrima.* 2 males and 2 females were introduced into small clip-cages attached to the leaves of the plants. Eggs, nymphs, pupae, new emerged adults and adult survival (egg-adult) were counted on each plant. There were significant differences in the reproductive capacity of *B. argentifolii: M. parviflora* hosted more number of eggs, nymphs and pupae than *C. bursa-pastoris*, *B. kaber* and *L. serriola.* No reproduction was observed on *A. intermedia.* The highest number of adults was obtained on *M. parviflora,* followed by *C. bursa-pastoris* and *B. kaber;* a low number of adults was obtained on *L. serriola* (less than 1 adult/female.day). The highest male and female survival proportion was obtained on *C. bursa-pastoris* followed by *M. parviflora* and *B. kaber.* 0% male survival was observed on *A. intermedia and only 5% was recorded on L. serriola*; females did not survive on these weeds. These results demonstrate that the best host in terms of the reproductive capacity of *B. argentifolii* is *Malva parviflora,* followed by *Brassica kaber* and *Capsella bursa-pastoris. Lactuca serriola* can be considered as plant hosting whiteflies in terms of those reproductive parameters. Reducing the overwintering population of *B. argentifolii* on these plants, especially *M. parviflora,* by selective methods such as biological control, in a given area might be expected to influence its relative abundance the subsequent season.

Comparative ecology of *Orius sauteri* and *O. tantillus*, predators of *Thrips palmi*, with special reference to their effectiveness as biological control agents in greenhouses

Y. NAKASHIMA and Y. HIROSE

Institute of Biological control, Faculty of Agriculture,
Kyushu University, Fukuoka, 812 Japan
Fax : (81) 926423040
E-mail : nksm@grt.kyushu-u.ac.jp

In Japan, *Orius sauteri* is an effective biological control agent *against Thrips palmi* which is a serious pest of vegetables both in the field and in greenhouses. However, *O. sauteri* cannot be applied to controlling *T. palmi* under short-day conditions because its diapause is induced under these conditions.

Orius tantillus is known to be a predator of *T. palmi* in the Philippines. It is a tropical and subtropical species and widely distributed in the Indo-Pacific. In Japan, *O. tantillus* is distributed in southern parts of Okinawa island where adults and nymphs of this species were found in a short-day season. Thus, *O. tantillus* is a candidate for biological control agents against *T. palmi*.

We examined biological and ecological characteristics of *O. sauteri* and *O. tantillus* to evaluate the effectiveness of these two species as control agents against *T. palmi* in greenhouses.

The results of the study showed that 1) developmental threshold of *O. tantillus* is lower than temperatures in winter greenhouse production in Japan, 2) reproductive diapause of *O. tantillus* is not induced at short-day conditions, and 3) prey consumption rates and egg production rates of both sexes of *O. sauteri* adults decrease more slowly with prey density than that of *O. tantillus* adults.

From these findings, we conclude that *O. sauteri* could be an effective control agent against *T. palmi* in greenhouses from summer to autumn when reproductive rate of *T. palmi* is high since *O. sauteri* is more excellent in depressing high population densities of *T. palmi* than *O. tantillus*. Non-diapausing *O. tantillus* can be applied to control of *T. palmi* from autumn to next spring when reproductive diapause of *O. sauteri* is induced.

Biological control of agricultural pests in Thailand

Pimolpom NANTA[1], Suapa DISTHAPOM[2] and Rut MORAKOTE[1]

1 : Biological Control Research Group, Entomology and Zoology Division, Department of Agriculture, Chatuchak, Bangkok 10900, Thailand
Fax : (662) 5614875
2 : Biological Control Institute, Department of agricultural Extension, Chatuchak, Bangkok 10900, Thailand

Research and extension on biological control of agricultural pests in Thailand have been widely promoted from 1991 till present due to the policy of Ministry of Agriculture and Cooperatives on the minimization of agricultural pesticides use for supporting the national policy on quality of life and environmental protection. The research on production and utilization of important biological control agents such as nuclear polyhedrosis virus (NPV), *Bacillus thruringiensis* (*Bt*), *Metarhizium anisopliae* (fungus), *Steinernema carpocapsae* (Nematode), *Anastatus nr. japonicus* (Parasitoid), *Trichogramma* spp.(Parasitoids) and *Eocanthecona furcellata* (Predator), besides, conservation pratices for Integrated Pest Management Programme in many important economic crops have been emphasized. For extension programme, some of biological control agents have been already adopted by Thai farmers such as NPV, *S. carpocapsae*, *B. thuringiensis*, *Trichogramma* spp. and *A. nr. japonicus*. The extension activities have been transferred to farmers through training, demonstrations, campaign days, exhibition, study tour and mass media. Up to now, there are about 30.000 farmers who participated in biological control projects. The main problem for extension is the limitation of commercial production to serve the demand of farmers, and the knowledge on beneficial and use of these natural enemies are much needed.

Effects of insecticidal management of *Bemisia tabaci* on abundance and impact of indigenous natural enemies

Steven E. NARANJO and C. C. CHU

USDA-ARS, Western Cotton Research Laboratory,
4135 East Broadway Road, Phoenix, Arizona 85040, USA
Fax : (602) 379 4509
E-mail : SNARANJO@ASRR.ARSUSDA.GOV

Studies were conducted in 1994-1995 to measure the effects of insecticide use frequency in cotton on populations of general predators and parasitoids of *Bemisia tabaci* in Imperial Valley, California, USA. Replicated plots were sprayed with a pyrethroid + organophosphate mix when densities of adult whiteflies exceeded 2.5, 5, 10, or 20 per leaf. Untreated plots served as controls. Weekly sweepnet samples estimated the abundance of general predators, and leaf samples estimated the abundance and activity of whitefly parasitoids. *Geocoris punctipes* was consistently the most abundant predator species, but increasing insecticide usage significantly reduced populations. In contrast, *Orius tristicolor* was moderately abundant and insecticide applications had little effect. Insecticide use frequency also had little effect on *Collops* spp., *Spanogonicus albofaciatus*, assassin bugs (*Zelus* and *Sinea* spp.), and *Lygus hesperus*. Spiders and *Hippodamia convergens* declined with increasing insecticide use. Populations of *Chrysoperla* spp. actually increased with greater insecticide use in both years. Populations of *Eretmocerus* and *Encarsia* parasitoids and immature stages of whitefly were progressively lower in plots receiving more frequent insecticide use. However, there were no significant differences in percent parasitism regardless of insecticide usage. Percent parasitism often exceeded 60% throughout much of August for *Eretmocerus*. Computer simulation and recent results from serological analysis of predator feeding activity were used to evaluate the overall impact of natural enemies on whitefly dynamics. The potential integration of biological control by indigenous natural enemies with current grower practices will be discussed.

Problems and prospects in large scale production and use of Trichogramma against apple and pomegranate insect pests in Iran

A.A. NASROLLAHI[1], M. SHOJAEII, F. GHAVAM[2],
Y. LABBAFI and M.R. ZIAII

1 : IROST, Division of biological Control, P.O. Box 1581-3538,
Tehran, Iran
2 : P.O. 91746, Koohasangy Ave., Alandast Garden, Mashhad, Iran
Fax : (98) 21 8838340

Codling moth *Carpocapsa pomonella* is a major pest of commercial apple production and carob moth *Ectomyeloise ceratoniae* is a major pest of pomegranate in Iran. Both pests cause great damage to fruit production and the latter pest can not be controlled chemically on pomegranate because it lays its egg inside pomegranate crown, and also pomegranate is sensitive to insecticides. Right now the only choice is then its biological control by egg parasitoids. For this reason and also production of healthy fruits, we started two separate biocontrol projects one in Khorassan and one in Yazd provinces. In both regions the Trichogramma wasps produced at large scale level and released in pomegranate and apple orchards at interval of 10 to 20 days respectively at the rate of 150000 wasps per ha. During the job many problems arose, especially in mass rearing, because there was not enough resources to work with easily and the subject were totally different from research practices. The following problems were the major problems which we had to deal with.

1. Lack of suitable space for large scale parasitoid factitious production, and cold storage room to store produced trichocards.
2. Lack of suitable trichocards due to robbing of parasitised eggs on trichocards by ants pomegranate orchards and desiccation of parasitised eggs in apple orchards.
3. Dry climate and low humidity less than 10 % in Yazd Province and unstable climate in Khorassan Province especially in April and May.

Resistance of growers and authorities to new methods. These and some other problems were faced by us, but by working hard and courageous action of some authorities and orchard owners most of the problems were solved. In apple orchards 10 times of spray stopped because other pests such as mites and leaf minors were controlled by natural enemies. Damages of both pests in apple and pomegranate orchards reduced to almost 10 % which is very promising for commercial production of natural enemies, and we expect to set up two commercial insectariums in both regions for the coming year.

Integrated pest management of strawberry arthropods with selective pesticides and *Phytoseiulus persimilis* Athias-Henriot.

H. NEMOTO

Saitama horticultural Experiment Station, Rokumanbu,
Kuki, Saitama 346 , Japan
Fax : +81 480 29 1021

As for a natural enemy, registration which procedure is same as a chemical pesticide is necessary in Japan. Data of efficacy in 6 points for 2 years, toxic examination for a man and fish, and data that a natural enemy doesn't give influence to environment, are demanded. However Japanese government doesn't show how prepare deter in the presentation about investigation technique. The registration of natural enemy doesn't progress, and a farmer can't choose a natural enemy among a such thing.

In case of strawberry, the natural enemy registered with is only *Phytoseiulus persimilis* Athias-Henriot. The Cotton aphid, *Aphis gossypii* Glover, the Greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood), and the Common cutworm, *Spodoptera litura* (Fabricius) occur other than spider mites in a strawberry greenhouse, but insecticides are used for those. The insecticides have a lot of the things that *P. persimilis* includes bad influence, and the insecticide that there is not bad influence is expected in *P. persimilis*. So I evaluated influence for *P. persimilis* on solution of DEP emulsion, solution of water dispersible powder of fluvalinate, Asetamiprid and imidacloprid, and imidacloprid granules. As a result there were few influence of imidacloprid granules. The application of imidacloprid granules at the time of planting enabled to control the cotton aphid for more than 3 months after planting. The plants in this study which were treated with granules at the time of planting, then released *P. persimilis* afterwards, displayed a significantly lower degree of infestation with spider mites. The control of only one species of pest is not relevant for strawberry that require simultaneous protection from other pests. Imidacloprid granules thus may be effectively exerts a minimal adverse effect on *P. persimilis*. This method could be applied in the IPM system.

Genetic effects of host species on *Trichogramma* sp. (Hym.: Trichogrammatidae)

L. NETO* and B. PINTUREAU

INSA, UA INRA 203, Biologie 406, 20 av. A. Einstein,
69621-Villeurbanne-cedex, France
Fax : (33) 72 43 85 11
E-mail : lba@insa.insa-lyon.fr
*Current address : rua de Berlim, n°7, 8 000-Faro, Portugal

The host species has numerous epigenetic effects on *Trichogramma* (size, fecundity, ...). Although essential for biological control, no information is available on possible genetic effects of the host species. If hosts used in rearings exert selective pressures on *Trichogramma* which are different from those exerted by natural hosts, it can lead to a decrease of the biological control efficiency.
Evolution of the frequency of 3 genes was followed up in different experimental populations reared on 2 hosts: *Ephestia kuehniella* Zeller and *Galleria mellonella* L. (Lep.: Pyralidae) (Ek and Gm, respectively). Genes Est $1^{0.09}$ and Est $1^{0.13}$, encoding for esterases, were studied in *T. evanescens* Westwood; 4 populations (initial frequency of Est $1^{0.09}$: 10% or 90%; host: Ek or Gm) with 2 replicates were followed up from G0 to G20. Genes Bh and bh, encoding for the parasitized host colour, were studied in *T. fuentesi* Torre; 4 populations (initial frequency of bh: 10% or 90%; host: Ek or Gm) with 3 replicates were followed up from G0 to G24. Genes Db and db, encoding for the body colour, were studied in *T. turkestanica* Meyer ; 4 populations (initial frequency of db: 20% or 80%; host: Ek or Gm) with 3 replicates were followed up from G0 to G14.
No evolution of Est 1 was found, this gene being probably neutral in most environments. The bh gene frequency did not evolve when Ek was the host, but it stabilized near 0.6 when Gm was the host (convergence of populations with a bh initial frequency equal to 10 or 90 %). The db gene frequency underwent an evolution with the 2 hosts: stabilization near 0.2 when Ek was the host and near 0 when Gm was the host.
So, the host species is not neutral for the *Trichogramma* genome. It exerts a selective pressure on parasitoids and we have to take this phenomenon into consideration in intensive rearings.

The effect of locally available vegetable oils on *Bemisia* spp. and its parasitism in cassava, *Manihot esculenta* Crantz in Malawi

G. K. C. NYIRENDA[1], D. C. MUNTHALI[2] and G. S. N. PHIRI[3]

1 : Bunda College of Agriculture, P.O. Box 219, Lilongwe, Malawi
Fax : 277364
2 : Chancellor College, P.O. Box 280, Zomba, Malawi
3 : Makoka Research Station, Private Bag 3, Thondwe, Malawi

Three locally available purified vegetable oils extracted from groundnuts, *Arachis hypogea,* sunflower, *Helianthus annuus* and cotton, *Gossypium hirsutum* were applied at 3 per cent to Mbundumali, a local variety of cassava *Manihot esculenta* Crantz. Dimethoate at 0.05 per cent a.i and the untreated were included as controls. 30 ml of each oil were mixed with 1ml of an emulsifying/wetting/spreading agent Agral (containing 900g alkyl phenol ethylene oxide condensate per litre, to produce an emulsifiable concentrate. The concentrate was made 2-3 days before application. On the day of application the concentrate was mixed with water to make a total of 14 litres. The oils were applied with a knapsack sprayer mounted with a tailboom and the trial was designer as a balanced latin square. Whitefly adults were monitored with yellow plastic traps quoted with a sticky substance, tanglefoot. Immatures of the whitefly were counted under the microscope on sampled leaves. Dimethoate gave the best control of immatures but all the vegetable oils gave better control of the immatures than the untreated control. Sunflower oil gave consistently better control than the other two oils. The vegetable oils were also more friendly to parasitoids than dimethoate. Vegetable oils may have a role in the integrated management of whitefly.

Introduction of *Encarsia lahorensis* Howard (Hym. : Aphelinidae) into the countries of the former USSR for control of Citrus whitefly, *Dialeurodes citri* Ashmead (Hom. : Aleyrodidae)

A.D. ORLINSKI[1] and T.V. BASSOVA[2]

1 : European and Mediterranean Plant Protection Organization (OEPP/EPPO), 1 rue Le Nôtre 75016 Paris, France,
Fax : (33) 1 42248943 E-mail : hg@eppo.fr
2 : State Plant Quarantine Inspection Service, Moscow, Russian Federation, Eastern Palearctic Regional Section of IOBC

The Citrus whitefly *Dialeurodes citri* Ashmead is one of the most important citrus pests of field crops on the Black Sea coast of the Caucasus and on the Caspean Sea coast of the Talysh Mountains ; it is also the most important citrus pest of trench cultivation in Central Asia and in glasshouses in Russia and some other parts of the former USSR. The parasitoid *Encarsia lahorensis* Howard was introduced in 1983 into Russia (Moscow) from Pakistan and was reared in a laboratory of the All-Russian Plant Quarantine Institute. The parasitoid was released in the following years in different parts of the country: in 1985 in Adzharia (Batumi); in 1987 in Abkhasia (Sukhumi) and in a glasshouse in Russia (near Moscow); in 1988 in Uzbekistan (Namangan); in 1990 in Tadzhikistan (Kurgan-Tyube). It established well in all places of release. The rate of parasitization of the whitefly by *Encarsia* reached 50-52% in the orchard in Batumi five years after the first release, 56-60% in the glasshouse in Moscow three years after the first release, and 50% in trenches in Namangan two years after the first release. Levels of the pest population decreased dramatically in the greenhouse and trenches. The efficiency of the additional feeding of *Encarsia* adults on the host larvae was evaluated to be 26-60%. It was found that *Encarsia* is able to establish in areas with very different ecological and climatic conditions but it is, in general, much more efficient and spreads much faster in trenches and glasshouses than in the field. *Semidalis aleyrodiformis* Stephens [Neuroptera : Coniopterygidae], found naturally occurring in trenches in Namangan, showed promise in preliminary trials as a potential agent for biological control of the Citrus whitefly.

Technology-transfer constraints of biological control on the potato crop in Peru

O. ORTIZ, H. FANO, M. PALACIOS, J. ALCAZAR and H. TORRES

International Potato Center, av. la Universidad s/n La Molina, Apartado 1558, Lima, Peru
Fax : 51 14 351570 E-mail : O.ortiz@cgnet.com

There are few examples in which biological control has been implemented in a sustainable manner in developing countries. The specific case of biological control on the potato crop in Peru provides an opportunity to analyse the constraints of the technology transfer process. Biological control for two of the main potato pests in the Andean Region was developed by the International Potato Center (CIP) and the National Agricultural Research Institute (INIA) during the late 1980s. These biological control components were the use of a fungus (*Beauveria brongniartii*) to control the Andean potato weevil (*Premnotrypes* spp.), and the use of a granulosis virus to control the Potato tuber moth (*Phthorimaea operculella*). Both of them have been included within an integrated pest management (IPM) program which was tested at farmer level in rural communities in Peru. Several institutions, especially non-governmental ones, were interested in this approach and started the technology transfer process of IPM. However, biological control has been one of the most difficult components to be implemented in a sustainable way. Some of the main constraints for its implementation are : the conflict between short-term control (farmers'goal) *vs.* long-term control (biological control goal), the conflict beween production of biological control agents at community level *vs.* production of such agents by private companies, the scarcity of credit an lack of interest of private companies to invest in the production biological control agents, low relative profitability of biological control *vs.* chemical control, the strong competition of insecticide-sellling companies, legal problems to register this kind of products, promotion of bio-control based on political or institutional objectives instead of technical and socio-economic analysis ; the promotion of biological control without suitable technical assistance for farmers, the believe that biological control is a "new way" of chemical control instead of being a component of an IPM program.

Encapsulation of *Hirsutella rhossiliensis*

A.V. PATEL[1], C. ISEMER[1], J. MÜLLER[2] and K.D. VORLOP[1]

1 : Institute of Technology, Federal Agricultural Research Centre (FAL),
Bundesallee 50, 38116 Braunschweig, Germany
Fax : +49 531 596 363 E-mail : patel@tec.fal.de
2 : Institute of Nematology and Vertebrate Research,
Federal Biological Research Centre for Agriculture and Forestry (BBA),
Toppheideweg 88, 48161 Münster, Germany

In previous experiments it was found that encapsulation of the nematophagous fungus *Hirsutella rhossiliensis* enhances its effectivity against *Heterodera schachtii*. We then focussed on the construction of a formulation consisting of the capsule as such, biomass, medium and other additives.

On petri dishes filled with autoclaved sand we tested the growth of fungus from capsules as a function of wet biomass content (5 and 15%), medium (none, 15% corn gluten, 15 % corn gluten and 0.5% yeast extract), capsule size (1.5 and 2.5 mm), soil humidity (10 and 14%) and conservation method (none, drying).

Furthermore, bioassays were conducted where encapsulated and free fungus was added to pots containing 96 ml autoclaved soil and a sugar beet seed each. After 1 week incubation 1000 nematodes were added and after another week, invasion of nematodes into plantlet roots was examined.

The experiments on petri dishes resulted in a capsule containing 15% biomass, 15% corn gluten and 0.5% yeast extract. Growth from 1.5 mm capsules was observed to be less than from 2.5 mm capsules, possibly due to early desiccation. Growth at 10% soil humidity was less than at 14%. It could also be shown that the fungus grew well from dried and rehydrated capsules (after 14 days 75% of mycel compared to fresh capsules).

Bioassays at 14% soil moisture showed that capsules containing 15% biomass, 15% corn gluten and 0.5% yeast extract could reduce the infection of sugar beet plantlets by 92% compared to controls whereas free mycel reduced the infection just by 50%. Interestingly, effectivity was retained at 10% soil moisture.

Ethylene release under field conditions for the management of the Olive bark beetle

A. PEÑA, A.J. SANCHEZ-RAYA, C. LOZANO and M. CAMPOS

Estación experimental del Zaidín (CSIC), c/ Profesor Albareda,
1. E-18008-Granada, Spain
Fax : (34) 58 129600
E-mail : arancha@eez.csic.es

Ethylene is a plant growth regulator, whose evolvement depends greatly on the plant physiological cycle (*i.e*, flower and fruit formation), on biotic (*i.e*, drought, flooding, high or low temperatures) and biotic factors (*i.e*, viral, bacterial and fungal diseases). Ethylene release has been also observed in several plants after insect infestation. The Olive bark beetle, *Phloeotribus scarabaeoides* Bern. (Coleoptera, Scolytidae), is a pest of olive trees which reproduces in logs left over from the pruning. The ethylene released by the logs acts as an attractant for these scolytids both in laboratory bioassays and in field experiments. GC-FID analyses have shown that when olive logs were sprayed with ethrel® (a formulation containing 2-chloroethyl phosphonic acid, that releases ethylene) they produced higher quantities of ethylene. Therefore, the attraction of *P. scarabaeoides* to the olive logs was increased. However, the increase in ethylene release, although very important, lasted only for about two weeks. A new dispenser has been optimised, which consists of a plastic vial with an aqueous ethrel solution to which a small piece of olive log is added. Field tests have been carried out to compare the results obtained with this vial, with ethrel spraying and with a combined treatement with an insecticide to kill the attracted olive beetles. The first results show that the dispenser is very effective, is not washed off by rain and lasts longer, at least for 4 weeks, without needing a change.

Insecticidal bioactivity of crude extracts of *Aglaia* species (Meliaceae)

C. PHOLPUNTHIN[1], M.B. ISMAN[2], P. WIRIYACHITRA[3] and G.H.N. TOWERS[2]

1 : Department of Biology, Faculty of Science, Prince of Songkla University, Hat-yai, Songkla, 90112, Thailand
Fax : 66 74 212917 E-mail : schutama@ratree.psu.ac.th
2 : Department of Plant Science and Botany, University of British Columbia, Vancouver, B.C., V6T 1Z4, Canada
3 : Faculty of Pharmacy, Chiangmai University, Chiangmai 50002, Thailand

Crude foliar extracts of 19 species of *Aglaia* (Meliaceae), mostly of Indo-Malaysian origin were screened for larval growth inhibiting and insecticidal effects on the polyphagous lepidopteran *Peridroma saucia* (Noctuidae). Extracts of at least seven of these species significantly reduce larval growth of *P. saucia*. *Aglaia odorata* yielded the most inhibitory extracts, but there is significant (35-fold) geographical variation in the bioactivity of the extracts within this widespread species. In addtion, extracts of bark are significantly more active than foliar extracts. Foliar extracts significantly deter neonate larvae, but nutritional analyses of fourth instar larvae fed artificial diets laced with *A. odorata* extracts indicate that both diet consumption and dietary utilization are impaired. These results are similar to that obtained with pure (-)-rocoglamide, an insecticidal principle isolated from *A. odorata*. This species should provide a useful starting point for the development of a botanical insecticide.

Study of the semiochemicals for the Olive bark beetle, *Phloeotribus scarabaeoides* Bern. (Coleoptera, Scolytidae)

M.T. PLAZA[1], I. IZQUIERDO[1], M. RODRIGUEZ[1], F. RIVAS[2], A. PEÑA[2], M. CAMPOS[2] and C. LOZANO[2]

1 : Department of Organic Chemistry, Faculty of Pharmacy, University of Granada, 18071 Granada, Spain
Fax : 958/243845
2 : Estación Experimental del Zaidín, CSIC, 18008 Granada, Spain

Olive growing is an important agricultural activity in Southern Europe and Northern Africa. It has been estimated that in Europe alone 2.5 million families directly depend on the olive cultures. In Andalucia, olive groves represent more than 30% of the total agricultural area, being an economical support to a great percentage of its human population.
The Olive bark beetle, *Phloeotribus scarabaeoides* is one of the pests of olive trees throughout the Mediterranean coast, causing important economical damage.
In order to identify the semiochemicals involved in the behaviour of *P. scarabaeoides*, volatile components of healthy and infected woods and of frass (produced by the beetles during the formation of the galleries) from olive logs have been analyzed. The samples came from an olive grove close to Granada, Spain. The air volatiles were collected according to Browne et al.[1]. GLC analyses indicated a similarity in the composition of extracts from different sources. On the other hand, the combined GC-MS technique allowed the identification of C_6-C_{11} saturated aldehydes, borneol (or isoborneol) and its acetate, 2-hexanone, 2-ethylhexanoic and nonanoic acids and some sesquiterpene derivatives. Some other components are currently being investigated.

Reference :
1.- Browne, L.E., Birch, M.C. and Wood, D.L. (1974) J. Insect Physiol. 20: 183-193.

Genetic influences on Aphid parasitoid host selection

W. POWELL and A. REHMAN

Dept. of Entomology and Nematology, IACR-Rothamsted, Harpenden, Herts., AL5 2JQ, United Kingdom
Fax : 44 (0) 1582 760981

Genetic factors influence the host recognition and attack behaviour of the closely related aphid parasitoids *Aphidius ervi* and *Aphidius microlophii* (Powell & Wright, 1988 ; Pennacchio *et al.*, 1994). Recent mother/daughter regression studies of the aphid attack behaviour of the parasitoids *Praon volucre* and *Praon myzophagum* also suggest that behavioural traits involved in host selection have a genetic basis.

In this paper the implications of these genetic factors for the use of aphid parasitoids in pest control are reviewed and the following topics discussed :

a) The importance of conserving genetic diversity in field populations of aphid parasitoids living in agricultural ecosystems. Does intraspecific genetic diversity promote long-term population stability by ensuring flexibility in host species exploitation ?

b) The comparative importance of genetic influences on host selection for generalist and specialist species. Do genetic factors play a greater or lesser role in host selection by parasitoids with a broad host range compared with more oligophagous species ?

c) The role of genetic factors governing host recognition in the evolution of host range and in speciation. Do populations gradually become reproductively isolated through behavioural adaptation to specific hosts ?

d) The potential effects of introducing parasitoids to new geographic regions on their host range. Do introductions stimulate rapid genetic changes leading to adjustments in host range ?

Pennacchio, F., Digilio, M.C., Tremblay, E. & Tranfaglia, A. - 1994. Host recognition and acceptance behaviour in two aphid parasitoid species: *Aphidius ervi* and *Aphidius microlophii* (Hymenoptera: Braconidae). Bull. entomol. Res., 84, 57-64.

Powell, W. & Wright, A.F. - 1988. The abilities of the aphid parasitoids *Aphidius ervi* Haliday and *A. rhopalosiphi* De Stefani Perez (Hymenoptera: Braconidae) to transfer between different known host species and the implications for the use of alternative hosts in pest control strategies. Bull. entomol. Res., 78, 683-693.

Utilization of *Nudaurelia*-β virus for biological control of the Oil palm leaf-eating nettle caterpillar *Setothosea asigna* Van Eecke (Lepidoptera : Limacodidae) in Indonesia

S. PRAWIROSUKARTO[1], Dj. PARDEDE[1], J.C. VEYRUNES[2], F. COUSSERANS[2], R. PHILIPPE[3],
R. DESMIER DE CHENON[3], D. MARIAU[3] and M. BERGOIN[2]

1 : Indonesia Oil Palm Research Institute, Jl. BrigjenKatamso 51,
Kp. Baru, P.O. Box 1103, Medan 20001, Indonesia
2 : Laboratoire de Pathologie comparée, Université de Montpellier II,
Pl. E. Bataillon, cc 101, 34095 Montpellier cedex 5, France
Fax : (33) 67 14 46 79
3 : URDDC, CIRAD-CP, Av. du Val de Montferrand, BP 5035,
34032 Montpellier Cedex 1, France

Among of the Oil palm leaf-eating nettle caterpillars in Indonesia, *Setothosea asigna* Van Eecke is the most important species, attacking indifferently the young and the old plantations.
We have recently isolated from infected-dead larvae of this defoliator collected in North Sumatra a small RNA virus that we characterized as belonging to the *Nudaurelia-β* group. This virus proved to be highly pathogenic for *S. asigna* larvae in laboratory conditions. In order to develop a method for the biological control of this pest based on the spraying of this virus in the oil palm plantations in Indonesia, we have prepared appropriate diagnosis tools *i.e.* : polyclonal antibodies against purified virions for ELISA tests and a cDNA probe for dot-blot analysis. Both diagnostic methods proved to be efficient for detecting the presence of the virus in infected caterpillars.
The sensitivity of larvae according to the instar and to virus concentration was determined in laboratory conditions. Field applications of viral preparations were performed and the comparative results of larval population dynamics in treated and untreated plots will be presented.

Controlling brown locusts with a myco-insecticide

R.E. PRICE[1], E.J. MÜLLER[1], H.D. BROWN[1] and R.P. BATEMAN[2]

1 : Locust Research Division, Plant Protection Research Institute, Agricultural Research Council, Private Bag X134, Pretoria, 0001, South Africa
Fax : (012) 329 3278
E-mail : RIETMEK@PLANT2.AGRIC.ZA
2 : International Institute of biological Control, Silwood Park, Buckhurst Rd., Ascot, Berks, SL5 7TA, United Kingdom

Recurring outbreaks of brown locust, *Locustana pardalina* (Walker), in the semi-arid Karoo region of South Africa and Namibia necessitate continual chemical control intervention. However, increasing concern from landholders and conservationists, regarding the possible impact these pesticides have on the Karoo ecosystem, has highlighted the need for alternative, non-chemical, control strategies. Building on the discovery that certain strains of the entomopathogenic fungus, *Metarhizium flavoviride*, show great control potential when formulated in oil, the PPRI in collaboration with IIBC, began testing the standard strain of this locust-killing fungus in 1992.

A number of ground trials, using a hand-held Micron Ulva spinning disc sprayer, were undertaken against brown locust nymphal bands in the Karoo in 1994. Additionally, in what was to prove the first such aerial application against an African locust species, 10 ULV aerial trials using a microlight spray aircraft, fitted with Micronair AU 7000 atomisers, were undertaken against nymphal bands in the following year. Effective control was achieved in both cases: mortalities averaged >90%. Median lethal time was however slow: averaging 10.3d in the ground application and 13.4d in the aerial application.

Although the level of control produced by the myco-insecticide equalled that achieved by any of the pesticides in current use in the Karoo, its rate of action was too slow. Brown locust nymphal bands are highly mobile and require more rapid control than the myco-insecticide technology can presently offer. Control operators in the Karoo are consequently likely to experience problems distinguishing between treated and untreated targets if these take too long to die.

Although further R & D is required to optimise this technology, it does provide us for the first time with a biocontrol agent that consistently achieves a high level of control. Product registration aimed at implementing the myco-insecticide in conservancy areas and Game Reserves, where speed of kill is not so critical, is therefore planned.

From research to practice : biological control of stored product pest moths (Lepidoptera : Pyralidae) in organic food processing industry with *Trichogramma evanescens* (Hymenoptera : Trichogrammatidae)

S. PROZELL[1], M. SCHÖLLER[1], S.A. HASSAN[2], B. WÜHRER[2], and C. REICHMUTH[1]

1 : Institut für Vorratsschutz, biologische Bundesanstalt für Land- und Forstwirtschaft, Wonigin Luise Str. 19, 14195 Berlin, Germany
Fax : 0049 30 8304 284
E-mail : adler@zedat.ju-berlin.de
2 : Institute of biological Pest Control, Darmstadt, Germany

The pyralids *Ephestia kuehniella* Zeller and *Plodia interpunctella* (Hübner), are the major stored product lepidoptera pest in the food processsing industry in Central Europe. Field trials with *T. evanescens* egg parasitoids were conducted in organic food processing industry, namely bakeries, stores and cereal processing departments. In these facilities no synthetic chemical insecticides are used.

Prior to the field trials, the spatial distribution, response to different light regimes and the penetration depth of *T. evanescens* into bulk grain was investigated in the laboratory.

In the field, *T. evanescens* was released weekly from cardboard egg-cards in the areas where moth infestation was detected. The pest populations were monitored from 1994 to 1996 using TDA baited funnel and delta traps. Integrated control comprising *T. evanescens* and a hygiene programme started in 1995.

Compared to the year without integrated control, the number of moths trapped in 1995 was reduced by one third. Moreover, parasitisation was found to be influenced by the habitat. The control programme did not interfere with the production process. The acceptance of biological control by the workers was very encouraging. No complaints by the consumers about the use of egg parasitoids have been reported.

The field data from 1994 to 1996 will be compared and the limits of scaling up previously conducted laboratory results will be discussed.

The present state of research and application of biological control in the EPS/IOBC territory

Stefan PRUSZYNSKI

Plant Protection Institute, ul.Miczurina 20, 60318 Poznan, Poland
Fax : 676 301

Thirty five scientific institutes and organisations connected with plant protection from 13 countries of Central and Eastern Europe are presently the members of East Palearctic Regional Section of IOBC established 25 years ago.
On the territory of Section activity there exists a long lasting tradition of researches on the application of biological method in plant protection. At the same time, the use of chemical plant protection products in the agriculture and horticulture was and still is considerably lower on such the territory. This situation favourized the development of biological method and its implementation to plant protection practice. The researches developed and carried out were connected with all the aspects of biological method, in this number also with bacterial, viral and fungal products for pest and disease control, with the release of beneficial species, with the researches on local species of parasites and predators as well as with the methods of their rearing and application. The results of researches were successfully implemented and applied in plant protection practice.
Only on the territory of former Soviet Union in 1987 biological method has been applied on 35 millions ha of plant cultures (5 species of egg parasite Trichogramma on the acreage of 15 millions ha and the bioproducts containing *Bacillus thuringiensis* Berliner on the acreage of 2 millions ha).
Very characteristic for this region was the establishment of so called "bio-factories" producing the parasite *Trichogramma* in and some bioproducts for the needs of plant protection in smaller districts or regions. The number of such the factories only in former Soviet Union was estimated as about 600.
In all the member countries biological control or pests of glasshouse crops has been implemented on a large scale ; and the bioproducts produced in other countries have been officially admitted to be used and sold. Legal backgrounds of the authorization of bioproducts as well as parasite and predator application meet international requirements.
Political and economical changes which took place in last years in the region of Section activity in first stage infavourably affected general situation of science and economy. Nevertheless, they did not change the approach to the application of biological method as well as to the researches on this field. Biological factors are more and more frequently applied in integrated pest management and in integrated technologies of horticultural and agricultural production.

Management of Citrus pests in Egypt : from research to practice

ALY. H. RASMY

National Research Center, Dokky, 12622, Cairo, Egypt
Fax : (202) 360 1877

A successful natural control of the citrus pests was achieved after 3 years of discontinuing pesticide applications in an orange orchard. The population of the Citrus brown mite *Eutetranychus orientalis*, a key pest, initially increased to damaging levels after discontinuing pesticides, then declined and was maintained at a very low level. This coincided with a gradual increase in the populations of the predacious mites. Regarding other citrus pests, the populations of the Fruit fly, *Ceratitis capitata* and aphids were maintained also at low levels, but the scale insect, *Chrysomphalus ficus* increased to a noticeable number at the 3rd year. Thereafter, a pest management program based on maximizing the role of the natural enemies and minimal use of pesticides was applied to maintain the citrus key pests at low levels, but the problem in how to mitigate damage caused by the Citrus leafminer, *Phyllocnitis citrella* invaded Egypt citrus in 1994. Field observations indicate that indigenous parasitoids of the Citrus leafminer can give insufficient pest control.

The use of molecular markers for detecting the geographical origin of *Ceratitis capitata* populations

A. REYES and M.D. OCHANDO

Dpto. de Genética, Facultad de CC. Biológicas, Universidad Complutense, 28040- Madrid, Spain
Fax : 34 1 394 4844

Nowadays, many molecular techniques are currently used in population genetic studies. Nevertheless, these techniques have only recently been applied to pest species, even though it is known that they can provide useful information for designing and monitoring biological pest control programme. In this way, we have employed three different molecular techniques, enzyme electrophoresis, restriction fragment length polymorphism in mitochondrial DNA, and random amplified polymorphic DNA (RAPD-PCR), in order to deduce the geographical origin of a population of the Mediterranean fruit fly, *Ceratitis capitata*. For this purpose, we analysed the genetic variability of three Spanish natural populations of this pest from different geographical origins : Atajate (South), Madrid (Center), and Campo Arcis (East), as well as a problem population, *i.e.* a population that was established under laboratory conditions more than 20 years ago and whose origin was to be determined (although it was known it came from the East).

The results show that when using isozymes, the problem population is located in a wrong area (Center), while by means of the analysis of mitochondrial DNA or RAPD-PCR the population clusters in the correct position (East). The discussion of these results is focused on the understanding of this discrepancy and on the possible use of these methodologies for detecting the geographical source area of a pest species that could systematically colonize the surrounding areas.

Potential of *Bacillus* species against rice diseases

V. RUKACHAISIRIKUL[1], S. PHONGPAICHIT[2]
and W. JUTIDAMRONGPHAN[3]

1 : Department of Chemistry,
2 : Department of Microbiology,
3 : Department of Biochemistry, Faculty of Science,
Prince of Songkhla University, Hat-yai, Songkhla, 90112, Thailand
Fax : 6674 212918
E-mail : rvatchar@ratree.psu.ac.th

Antagonist *Bacillus* species, isolated from rice seeds and rice soils in the southern part of Thailand, inhibit mycelia growth of two pathogens, *Rhizoctonia solani* and *Pyricularia grisea*, in *in vitro* dual culture plate assays. The scanning electron micrographs revealed that *Bacillus* caused distorting and extensive bursting of affected hyphae. The antagonist activities of 9 *Bacillus* isolates were compared. All isolates inhibited the growth of *P. grisea* while 7 out of 9 isolates showed the inhibitory activity against *R. solani* in the range of 78-100% at 1:2 dilution.
Bacillus MK007, one of the effective antagonist isolates, was further investigated. Ethanol extracts of cell-free culture filtrates, containing antibiotics, efficiently suppressed the growth of both pathogens to a similar extent. Thin-layer chromatography demonstrated that the extracts contained three major antibiotics which were visible, after treatment with the Ehrlich reagent, as white spots. Their antagonistic activity against rice diseases will be presented in this paper.

A cooperative extension initiative by commercial producers of beneficial arthropods in Australia

Andy RYLAND and Robert SPOONER-HART

University of Western Sydney, Hawkesbury, Richmond, NSW 2753, Australia
Fax : 045 701314
E-mail : a.ryland@uws.au

Australia holds a significant place in the history of modern biological control. The management of the pest, Cottony cushion scale, *Icerya purchasi*, in California in 1888 by the Australian native Vedalia beetle, *Rodolia cardinalis,* was one of the earliest successful cases of planned biological control, and was a catalyst for the development of modern approaches to classical biological control. The most notable success of this approach used in Australia in the last sixty years was the control of the exotic weed prickly pear, *Opuntia inermis,* by the moth larvae, *Cactoblastis cactorum,* in the 1920's. However inundative releases of mass reared beneficial species is a more recent phenomenon developing in Australia over the last 15 years. In 1992, Australasian Biological Control Inc., the Association of Beneficial Arthropod Producers, was established. This non profit association brought together all six commercial breeders of beneficial insects and mites established in the country at that time. The association's main goal is to encourage the reduction of pesticide use by the adoption of integrated pest management (IPM) strategies. The association provides a forum for both research and extension initiatives in the production and use of biologicals and a pool of expertise in their mass rearing. All members of the association are involved in on going research and development, both individually and with government and university researchers. By adopting IOBC quality control recommendations for commercial production of beneficials, the association intends to set a standard to which new producers in the region will conform. It also gives the industry a united representation to public policy makers. The first initiative in industry wide extension by the association has been the production of a reference book providing information and photographs of parasites, predators and their uses, currently commercially available in Australia for biological control. The book is intended to be a useful resource for growers, researchers and students.

International information system on biological control

L.A.N. de SA[1], G.J. de MORAES[1], N.C. GATTAZ [1]
and D. CANHOS [2]

1 : Laboratório de Quarentena, CNPMA/EMBRAPA, 13820-000
Jaguariúna - SP, Brazil
Fax : (019) 867 5633 E-mail : l.sa@phx.ftpt.br
2 : Fundação Tropical de Pesquisas e Tecnologia "André Tosello",
13087-000 Campinas - SP, Brazil

As for any other science, quick communication is becoming increasingly important for biological control specialists. Exchange of information facilitates adoption and adaptation of developed techniques, and may promote the exchange of valuable live biological material to be used in classical control projects. Quick global exchange of ideas may also have a positive impact on the continuous development of working rules and regulations of international acceptance related to biological control. An international information system on biological control that can be accessed trough the Internet was established in May 1994. It is physically based at the "Fundação Tropical de Pesquisas e Tecnologia 'André Tosello'", in Campinas-SP-Brazil, and is managed by a group of biocontrol workers and computer specialists from CNPMA/EMBRAPA and Fundação "André Tosello" . The main objective of this system is to facilitate contacts between biocontrol specialists around the world. The system, composed of different data bases related to biological control can be accessed at http://www.bdt.org.br/bdt/biocontrol. This web server also has pointers to other relevant information system related to biological control and a read-only searchable version of the discussion list Biocontrol-L. This list now has about 500 subscribers from over twenty countries. New members are welcome to join Biocontrol - L, and to participate actively in the discussions raised daily by different participants world-wide.

Protocols for testing host specificity of agents for biological control of arthropod pests

D. P. A. SANDS

CSIRO, Private Bag No. 3 Indooroopilly, Queensland 4068, Australia
Fax : (617) 3214 2885
E-mail : don@brs.ento.csiro.au

Before exotic organisms are released into countries as biological control agents for weeds or arthropod pests, their safety and acceptability must first be demonstrated. For weeds, protocols ensure that potential agents are sufficiently host specific and will not have any detrimental impact on crops, ornamentals or native plants but for arthropods pests, protocols to assess any development of agents on non-target taxa have not been so widely adopted. Agents for many arthropod pests have been released when known to be relatively less specific than most agents for weeds. When any feeding or development by potential agents occurs on beneficial or native organisms, such biological control agents must be assessed to ensure that their establishment will not significantly affect abundance or the survival of non-target taxa. When development of an agent does occur on non-target taxa the benefits of biological control of a target pest must be weighed against any real detrimental affects. Methods for testing agents for arthropod pests often differ from weeds for example, chemical compounds from a host held in close proximity may induce atypical oviposition on a species otherwise not utilised as host, using choice tests similar to those for effectively assessing specificity of agents for weeds. The number and range of non-target arthropods to be tested with an exotic agent should not be based on the expectations of the number of plants traditionally exposed to agents in weeds projects. The logistics of maintaining a range of species in culture when the life histories of so many are unknown, limits the number that can reasonably be tested. However, limited selection of taxa with appopriate relationships to the target species, can provide the meaningful information necessary to assess the host specificity of potential agents. Guidelines are proposed as a basis for assessing the safety and acceptability of agents for biological control of arthropod pests.

Biological control of phytopathogenic diseases of potato and cabbage strain *Bacillus mycoides*

T.G. SHABASHOVA[1], S.I. BELSKAYA[1] and F.A. POPOV[2]

1 : Institute of Experimental Botany, Academy of Science, F. Skorina st. 27, Minsk, 220733, Belarus
Fax : 375 (172) 220 91 25
2 : Institute of Plant Protection, Belarus

The search of highly active biological means for vegetable plants protection against diseases and nide introduction of their into agropractice could yield ecologically safe produce. So far the range of bacterial preparations is extremely limited. Researchers from laboratory of Mycology, Institute of Experimental Botany carried out scrinning of microorganisms - antagonists of phytopathogenic fungi and bacteria, and isolated potential biocontrol agents. Special attention was focused on strain *Bacillus mycoides* 683 possessing antagonistic action towards most deleterious pests of agricultural crops : *Fusarium sambucinum, Phoma lingam, Alternaria brassica, Botrytis cinereae, Phytophtora infestans, Erwinia carotovora* v. *atroseptica, E. aroideae, Xanthomonas campestris.* The investigation of antagonistic activity of strain *B. mycoides* has showed that intoduction of tested cell suspension with bacterial and fungal infections into potato tubers resulted in decrease of pathological process by 40-68%.

In collaboration with laboratory of vegetable cultivars, Institute of Plant Protection were examined of antogonistic properties of strain *B. mycoides* to control of cabbage diseases. Experiments with steeping cabbage seeds in cells suspension of *B. mycoides* have demonstrated increased number of germinated seeds -7.5% and 6.6% rise upon 6h and 24h exposure, respectively. In variant with reference phytolavin-300 the corresponding figures equalled 12.5% and 8.5%. As to reduction of seed bacterial infection, in both cases steeping in suspension containing $1*10^9$-$1*10^7$ cells were decreased the level of seeds colonization of pathogenic bacteria by 37.5-56.7% and 78.7-81.2% in reference variant. The seed treatment with suspension of *B. mycoides* cell ($1*10^9$ cell/ml) exerted promoting and protection action on cabbage sprouts. Leaf surface was enhanced by 39.6% as compared to the control, and damping of spread was lower by 30.1%. Test batches of biopreparation based on strain *B. mycoides* were manufactured in liquid and dry forms. Field trials demonstrated its stimulating effect on potato shoots and cabbage sprouts. Cabbage yields averaged 307 centners per hectare in contrast to 273 centners/ha at control plots. Treatment of potato tubers with tested biopreparation raised harvests by 20% on the average. Currently the research efforts are concentrated on elaboration of process regulations pack age and further field tests on various vegetable crops.

The perspective on the transfer of biopesticide production technologies

V.V. SHEVTSOV, E.V. SCHYOLOKOVA and V.L. ISCHENKO

Biotechnological Innovation Centre, 102A, Lenin str.,
Serpukhov, Moscow region, 142283, Russia
Fax : (0967) 72 55 78
E-mail : kostrikova@mx.ihep.su

Some very active strains of *Bacillus thuringiensis* serovars *thuringiensis, galleriae, dendrolimus, kurstaki* for control of various pest insects were selected. Physiological and biochemical properties of the strains were studied. The strains were tested by the state commission and used to production commercial preparations Lepidocide, Dendrobacillin, Bitoxibacillin in form of wettable powder.
The technologies for production of bioinsecticide liquid formulations at Biotechnological Plants and small-scale regional factories with use of native raw materials and food industry waste were elaborated.
The technologies for production of the most ecologically safe bioinsecticides based on asporogenic strains of *B.thuringiensis* serovar *kurstaki* for control of pests which cause damage of forest, agricultural and medical crops in silkworm breeeding regions, water-meadow and food storages.
There are offered the different variants of transfer technologies (strains, know-how) and design of small-scale regional manufacturies.

The effect of temperature and humidity on the development, fecundity and survival of *Trichogramma dendrolimi* (Hym. : Trichogrammatidae)

Zu-hua SHI and Shu-Sheng LIU

Department of Plant Protection, Zhejiang Agricultural University,
Hangzhou 310029, P.R. China
Fax : 0086 571 6049815

Trichogramma dendrolimi is a widely used biological control agent against both agricultural and forestry insect pests in China. We report here a detailed climatic study of this egg parasite under controlled conditions. The development, survival and fecundity of the parasite reared on *Corcyra cephalonica* (Stainton), were determined under 13 temperature and humidity regimes. Normal development of the whole immature stage was observed within the temperature range of 13.5°C to 30°C, and the relationship between the development rate and temperature fitted to a logistic curve with both low and upper boundaries. The development threshold temperature and thermal constant for the whole immature stage were estimated to be 10.1±0.03 C and 140.2±0.29 day degrees, respectively. The survival rate of the immature stage was highest under about 26°C. Longevity of the female wasp decreased with increase in temperature. The fecundity was highest at the range 23-28°C. Both low and high levels of relative humidity showed decelerative effect under low temperature and accelerative effect under high temperature on rate of development. The survival rate of the immature stage was reduced under the combinations of low temperature and high humidity or high temperature and high humidity. Under both low and high temperatures, adult survival and fertility were higher under 75% RH than those under lower and higher levels of relative humidity. At 33 C, no female progeny were produced at all levels of relative humidity tested. The combined effects of temperature and humidity were shown by changes of the intrinsic rate of increase.

Biodiversity of Phytoseiid mites associated with biological control of Cassava spider mites *Mononychellus* spp. in Northern South America (Acari : Phytoseiidae, Tetranychidae)

L. SMITH, E.L. MELO, J.R. ESCOBAR and G.J. de MORAES

International Center for Tropical Agriculture, A.A. 6713, Cali, Colombia
Fax : 572 445 0073 E-mail : L.Smith@cgnet.com
and
Centro Nacional de Pesquisa de Monitoramento e Avaliaçao de Impacto Ambiental, EMBRAPA, Caixa Postal 69, CEP 13820-000, Jaguariúna, Sao Paulo, Brasil

The cassava spider mites, *Mononychellus tanajoa* (Bondar) and *Mononychellus caribbeanae* McGregor, feed on leaves and growing tips of cassava, *Manihot esculenta* Crantz, an important tropical root crop. These mites are native to the neotropics, but *M. tanajoa* became established in Africa in the early 1970s, where it causes extensive damage. It is also a serious pest in the dry region of northeast Brazil, where there are few species of predators. Explorations were conducted in Colombia, Ecuador and Venezuela to find phytoseiids suitable to release in Africa and northeast Brazil. Ecological data collected during explorations and climatic data from GIS databases were used to describe the ecological niches of the predators. At least 84 species of predaceous phytoseiids have been found in this region, and 50 were in association with cassava.

Multiplication of *Zoophthora radicans* (Brefeld) Batko (Zygomycetes: Entomophthorales) on natural substrates

S. STEWART[1]; M.S. ZERBINO[1]; R. ALZUGARAY[1] and J. EILENBERG[2]

1 : INIA La Estanzuela, CC 39173, Colonia, Uruguay
Fax : 598 522 4061 E-mail : stella-@inia.org.uy
2 : Dept. Ecol. Molec. Biol., Royal Vet. Agric. Univ., 1870 Frb.C., Copenhagen, Denmark

The presence of *Zoophthora radicans* occurring naturally in Uruguay was reported in 1992. The fungus has been observed killing larval populations of *Epinotia aporema* (Walsingham) (Lepidoptera: Tortricidae) in pasture fields every year since then. Prevalence in the field has been evaluated showing high variation between years. During 1995 lab trials were conducted to multiply the fungus on natural substrates. Substrates evaluated were mowed corn, orange peel, rice (both common and parboiled) and wheat bran, previously inoculated with the fungus grown on liquid media. Mycelium growth was only observed on both rice media in a period of time comparable to its growth on SDAY (around 14 days). Different methods to store the inoculum on these media were unsuccesfull. High mortality rates were achieved from spore shower discharged by the inoculated rice. Low production costs of *Zoophthora* on rice without using sophisticated equipment and its proven pathogenicity on larvae encourages further studies to achieve a succesfull method of storing the inoculated rice.

Effect of light intensity upon flight activity of parasitic wasps, *Dacnusa sibirica* (Hym.:Braconidae) and *Diglyphus isaea* (Hym.:Eulophidae)

T. SUGIMOTO

Entomological Laboratory, Kinki Univ., Nakamachi 3327-204,
Nara 631, Japan
Fax : 0742-43-1155

Parasitic wasps of *Dacnusa sibirica* Telenga and *Diglyphus isaea* (Walker) have been used as a biological control agent to larvae of *Liriomyza trifolii* (Burgess). In the insectarium (350x170x220cm) of 20°C, 70% RH and the illumination of the fluorescet light of 40W (about 70 lux on the floor) adults of *D. sibirica* could fly normally among plants, while many eulophid wasps including *D.isaea* could not so. *D. isaea* only jumped repeatedly after flying down from plants on the floor in such an insectarium. This makes it hard for us not only to study its foraging activity among plants but also to evaluate its efficacy as a biological control agent, in such an insectarium. This study was carried out from some aspects to determine the releaser of the normal flight activity of *D.isaea*. Finally, we found that females of *D. isaea* could fly well only above 300 lux. Also males of both species flew more actively regardless of the light intensity.

Development of a monoclonal antibody-based kit for the identification of whitefly species and biotypes

W.O.C. SYMONDSON[1,2], T. GASULL[1,2], J.E. LIDDELL[2] and P.A. LANGLEY[1]

1 : School of pure and applied Biology, University of Wales Cardiff, P.O. Box 915, Cardiff CF1 3TL, United Kingdom
Fax : 01222 874305 E-mail : SYMONDSON@CARDIFF.AC.UK
2 : School of molecular and medical Biosciences, University of Wales Cardiff, P.O. Box 911, Cardiff CF1 3US, United Kingdom

Whiteflies are major pest of both horticultural crops and ornamental plants in glasshouses and polythene tunnels. They damage plants directly, through feeding on sap, causing stunted developments of foliage and flowers. More significantly, perhaps, they cause massive growth of sooty moulds on the honeydew they produce which, in addition to the presence of the whiteflies themselves, and their scales, causes severe cosmetic damage. In addition to all of this they are known to be vectors of a number of plant diseases. Identification of the species of whitefly infesting plant material is important for a number of reasons. The species *Bemisia tabaci* Gennadius has recently invaded a number of European glasshouses, and attempts are made by customs authorities and growers to restrict the spread of this species to new areas. To do this it is important that non-specialists should be able to easily distinguish between this species and the ubiquitous glasshouse whitefly, *Trialeurodes vaporariorum* (Westwood). In addition, different species or strains of biological control agents, such as parasitoid wasps, are used against each species of whitefly, and therefore prior identification is essential. Even where *B. tabaci* is know to be present, it can be important to know whether or not the 'B biotype', that attacks 600+ species of cultivated plants, or one of the 'non-B biotypes', with a more restricted host range, is present. A grower with the latter would not wish to bring plants infested with the 'B biotype' into his glasshouses. This report describes progress towards developing a kit based upon a dipstick assay, incorporating monoclonal antibodies. Details are given of the characterisation of antibodies created to date that will distinguish between these species and biotypes. This kit is the first in a series that are planned for the identification of invertebrate pests. The kits are being developed in association with Insect Investigations Ltd., a company owned by the University of Wales Cardiff.

Evaluation of introduced natural enemies in classical biological control : a case study on successful multiple species introduction of exotic parasitoids

M. TAKAGI

Faculty of Agriculture, Kyushu University, Fukuoka 812 Japan
Fax : (81) 926423040
E-mail : mtakagi@grt.kyushu-u.ac.jp

One long-standing debate on introduction strategies in classical biological control has been that between "multiple species introductionist" and "single species introductionist". Evaluation of competitive natural enemies introduced for biological control of a pest will contribute to settle the debate.
In 1980, two Aphelinid parasitoids, *Aphytis yanonensis* and *Coccobius fulvus*, were introduced from China to Japan for biological control of the Arrowhead scale, *Unaspis yanonensis.* After the introduction of the parasitoids, population density of *U. yanonensis* rapidly decreased. Now this pest, once been a defoliating pest of citrus in Japan, is controlled under the subeconomical level. Multiparasitism and interspecific competition of these two parasitoids are easy to occur in the laboratory. However, both species coexist in citrus orchards in Japan and multiparasitism is seldom observed in the field.
One of the parasitoids, *A. yanonensis* is higher in the reproductive capacity than another parasitoid, *C. fulvus* because of the shorter generation time and uniparental reproduction. Moreover, *A. yanonensis* is superior in interspecific competition over *C. fulvus* in multiparasitizd host. This parasitoid is good at increasing its number at the pest population outbreak. On the other hands, *C. fulvus* has longer longevity and its fecundity is larger than that of *A. yanonensis.* This parasitoid can use wider size range of *U. yanonensis* than *A. yanonensis* and is good at maintaining its own population even when the host population is at low level and intermittent.
Field population dynamics of *U. yanonensis* after releases of the parasitoids indicates that two parasitoids perform as complementary natural enemies. It is suggested that *A. yanonensis* is good at control an outbreak of the pest and that *C. fulvus* is a better control agent for maintaining the pest population at low level. Thus the multiple introduction of these competitive parasitoids is considered to be reasonable strategy for biological control of the Arrowhead scale.

Presentation of the Research Station for Biological Control at Loos-en-Gohelle (Pas-de-Calais, France)

C. TROUVE[1], O. CREPIN[2], S. LEDEE[3] and S. DUVAUCHELLE[4]

1, 4 : Service régional de la Protection des Végétaux Nord-Pas-de-Calais, 81 rue Bernard Palissy, B.P. 47, 62750 Loos-en-Gohelle, France
Fax : (33) 21 43 97 72
2, 3 : Fédération régionale de Défense contre les Ennemis des Cultures Nord-Pas-de-Calais

The studies and the experimentations carried out for 3 years by the Research Station for Biological Control at Loos-en-Gohelle (Pas-de-Calais) are displaid.
They concern 3 levels of investigations :
⇨ Technical support with growers (managed control, environment protection)
⇨ Technology transfer in biological control at the regional crops.
⇨ Research of new techniques in laboratory, rooms under controlled conditions and in the field.
The main studies are given for hops, fruit growing, vegetable and ornemental growings and potatoes.

Biological and integrated pest management (IPM) in nursery stock in the Netherlands

R.W.H.M. van TOL and M.J. van der HORST

Research Station for Nursery Stock, P.O.Box 118,
2770 A.C. Boskoop, The Netherlands
Fax : (0) 172 219717
E-mail : TOL@BPO.AGRO.NL

Research on IPM in nursery stock started in 1992. In a cooperation between our Research Station, the Plant Health Service and the Advisory Services, the introduction of IPM was extended to 22 nurseries in 1994. In this presentation the results of three introductions of IPM will be presented. These are introduction in a non-heated greenhouse, container field outside and in the field outside. In the greenhouse we were able to reduce the total chemical control with 96% and in the outdoor nurseries with 50 to 60%. In the greenhouse we were able to reach these reductions by active biological control of pests like thrips, aphids, whitefly and two-spotted spider mites. In the field we reached these reductions by active biological control and mostly by the built-up of natural populations of beneficial insects like hoverflies, ladybirds, anthocorids, lacewings and parasitic wasps that were able to control several insect pests in the field. Savings up to 87% on insecticide use were reached in the field. Control strategies and results for different pest-plant combinations will be discussed as well as the costs of crop protection in IPM on these nurseries. Promising results in field experiments to control spider mites with the predatory mite *Amblyseius californicus*, *Metaseiulus occidentalis* and *Phytoseiulus persimilis*, aphids with *Harmonia axyrides* and *Chrysoperla carnea* and black vine weevil with insect-parasitic nematodes will be presented also.

IPM Strategy for the Control of *Hoplochelus marginalis* (Col : Melolonthinae) : from field application to industrial process and commercial patent

B. VERCAMBRE[1], M. GUILLON[2], R. GOEBEL[3], C. NEUVEGLISE[4], P. ROBERT[4] and G. RIBA[4]

1 : CIRAD, Unité de recherches d'Entomologie appliquée, 2477 ave du Val de Montferrand, B.P. 5035, 34032 Montpellier cedex 1, France
Fax : (33) 67 61 56 66 E-mail : vercambre@cirad.fr
2 : Calliope-NPP, Route d'Artix, B.P. 80, 64150 Noguères, France
3 : CIRAD-Réunion, laboratoire d'Entomologie, Station de la Bretagne, 97487 Saint Denis cedex, la Réunion, France
4 : INRA, Station de Recherches de Lutte biologique, La Minière, 78285 Guyancourt cedex, France

The research programme against the white grub *Hoplochelus marginalis* (Col : Melolonthinae) in the Reunion Island is an exemplary model of IPM strategy, particularly from an industrial and commercial point of view, against a soil insect pest.
Following the accidental introduction of this species to the island, probably in the early 70s, significant damage has been caused since 1981 and 1989, chemical control was encouraged. From onwards full antagonists wer introduced without success.
The following outlines the development of this biological control programme using a *Beauveria brongniartii* strain :
- Discovery of the strain in its country of origin, Madagascar (CIRAD-INRA, 1987)
- Purification of a monospore strain (*Bt* 96) and laboratory bioassays to check the virulence of the fungus strain (INRA-CIRAD, 1988).
- Production of a registered product (BETEL^{R}) :
    a) Field trials with *Bt* 96 mass produced on rice (CIRAD) and on clay granules produced in prototype pilot (INRA-CALLIOPE 1989-1993).
    b) Toxicological files (1993)
- Setting up an efficient molecular test to monitor a characteristic pattern for *Bt* 96 (INRA, 1993)
- Scaling up of industrial production (NPP 1993 - 1996), in order to offer large scale biological control, *i.e.* a single mixed treatment with a reduced rate of Suscon^{R} (6 kg/ha instead of 28 kg/ha) and Betel^{R} biocide (50 kg clay granule formulation containing 0.2 x 10^9 spores/g dry weight) at the same price of the treatments using chemical insecticides alone. The ecological balance remains satisfactory for both the spores produced by saprophytic development and those produced by pathogenic development in adult insects.

Biological control of the major postharvest diseases on apple with *Candida sake*

I. VIÑAS[1], J. USALL[1], N. TEIXIDÓ[2], J. OCHOA DE ERIBE[3], and V. SÁNCHIS[2]

1 : UdL-IRTA Center, Postharvest Area, Pathology Unit,
2 : UdL, Dep. of Food Technology./ Rovira Roure, 177. 25198 Lleida, Catalonia, Spain
Fax : 34 73 238301 E-mail : usall@lleida.irta.es
3 : SIPCAM INAGRA SA Research and Development, Prof. B Báguena 5, 46009 Valencia, Spain

Biological control using microbial antagonist has been considered as a desirable alternative to the use of chemicals. The control of major postharvest pathogens through application of biological agents was reported for stone fruits, apples, citrus, and other fruit.
The yeasts are a particular interest, they can colonize the surface of the fruit for long periodes of time, producing extracellular polysaccharids which enhance their survival and limit the places of colonization and the germination of the fungic propagules, owing to the fact that they use the available nutrients more rapidly.
A screening program was initiated in 1989 to isolate from plant surfaces more than 1200 naturally occurring bacteria and yeast having potential as biocontrol agents effective against postharvest disease fungi found in the Mediterranean area, including *Penicillium expansum* Link, *Botrytis cinerea* Pers. and *Rhizopus stolonifer* (Ehrenb.) Lind.
The objective of this research was to investigate the biocontrol potential of a new strain of *Candida sake* (CPA-1), isolated from the surface of apple, against the three major postharvest pathogens of apples in the Mediterranean area, and their capacity of growh and retain their effectiveness under conditions of low temperature (1°C) and low-oxygen atmosphere. Complete control of *Botrytis cinerea* and *Rhizopus nigricans* (10^4 conidia/ml) was obtained with a 7. 10^6 ufc/ml of *C. sake* on apples incubated 7 days at 20°C. At the same conditions, the control of *Penicillium expansum* (10^4 conidia/ml) was upper 80%.
This strain of *C. sake* also provides excellent results of control the rot development under cold storage conditions.
The strain of *C. sake* show great growth in aerobic conditions on apples, at two temperatures, ambient (25°C) and a cold storage (1°C), whether in ambient atmosphere (21 % oxygen), and in controlled atmospheres with 3% and 1% oxygen, which are inherent to the storage in fruit-horticultural centres.

Biological control research on fire ants by the USDA-ARS

David F. WILLIAMS[1], Sanford D. PORTER[1] and Juan A. BRIANO[2]

1 : USDA-ARS, Medical and Veterinary Entomology Research Laboratory, P.O. Box 14565, Gainesville, Florida, USA
Fax : 352 374 5984 E-mail : DFW@NERVM.NERDC.UFL.EDU
2 : USDA-ARS, South American Biological Control Lab., Hurlingham, Argentina

Biological control research on fire ants in the USDA-ARS involves 3 highly specific natural enemies that will stress imported fire ant populations sufficiently to allow native ants to better compete. These are, (1) *Thelohania solenopsae*, a microsporidian that kills a high percentage of infected IFA colonies, (2) *Solenopsis* (Labauchena) *daguerrei*, a parasitic ant that attaches to the IFA queen and redirects IFA workers to tend the brood of the parasite, and (3) *Pseudacteon* spp., phorid flies that are strong antagonists and parasites of IFA.

Laboratory studies in Argentina with *Thelohania solenopsae* showed that healthy colonies lived longer than infected ones. After 12 weeks of artificial rearing, 50% of the healthy colonies died versus 93% of the infected ones. In field studies in Argentina, the size of infected colonies was smaller than noninfected ones and the overall density of the infected populations declined by more than 80%.

The presence of *Solenopsis daguerrei* in fire ant colonies has detrimental effects on colony growth and the proportion of sexual reproductives produced in the colony. *S. daguerrei* queens enter fire ant colonies and attach themselves to the mother queen. Previous studies have demonstrated that this parasite inhibits the fire ant mother queen and her egg production, thus causing the ant colony to collapse and eventually die out. Since *S. daguerrei* queens attach themselves to fire ant queens, this species could be especially useful in controlling the multiple-queen form of the fire ant.

At least 18 species of *Pseudacteon* flies have been found attacking fire ants in South America. Different species attack different sizes of fire ants. These flies are common and active throughout most of the year, but different species are more active at different times of the day. Most species appear to be broadly distributed across a wide range of habitats and climates. The *Pseudacteon* species that attack fire ants appear to be specific to fire ants. The host specificity of several parasitic *Pseudacteon* flies in South America was tested in the field with 23 species of ants from 13 genera. As expected, these flies were attracted only to *Solenopsis* fire ants.

Biological studies of the 'B strain' of *Bemisia tabaci* (Genn.) on Poinsettia (*Euphorbia pulcherrima*) and other potplants grown under glass

Matina ZAROKOSTA and Mike COPLAND

Department of Biological Sciences, Wye College, University of London,
Ashford, Kent TN25 5AH, United Kingdom
Fax : 01233 813140

In order to determine the host-plant suitability of 'B strain' of *Bemisia tabaci* (Genn.), the developmental time, pre-adult mortality, fecundity, sex ratio, morphology and size of immature stages have been studied on a range of host plants comprising Poinsettia (*Euphorbia pulcherrima*), Cotton (*Gossypium hirsutum*), Tobacco (*Nicotiana tabaccum*), Okra (*Hibiscus esculentum*) and Aubergine (*Solanum melongena*). The developmental rates were studied at 18±0.5°C, 21.5±0.5°C, 26.5±0.5°C and 31±0.5°C; 65-75% R.H. and 16:8 D:L photoperiod. The shortest developmental period for 'B strain' (oviposition - adult emergence) was 22.4 days on Poinsettia at 26±0.5°C. This period increased with lower temperature. The incubation period and the adult survival was also recorded. Not only the external morphology (number of setae and shape) but the size of the immature stages was also influenced by the host plant in the following order : Cotton > Okra > Aubergine > Poinsettia. Although the species is arrhenotokous, the sex ratio of offspring of mated females was in favour of females. The ratio of females was highest at lower temperatures. Not only the plant species but the maturity of the leaves have a significant influence on the longevity and fecundity on 'B strain'. The daily fecundity on young leaves was higher (8.1) on Tobacco followed by Aubergine, Poinsettia and Okra (4.5) while it was highest on Okra (4.5) followed by Tobacco, Poinsettia and Aubergine (2.3) when the pest was reared on mature leaves. The oviposition rate of 'B strain' of *B. tabaci*, gradually increased during the first few days reached a maximum for a short period, then declined to a constant level before the death of the female.

Biological control available for practical use in Switzerland

Markus ZUBER and Martin ANDERMATT

Andermatt BIOCONTROL AG, Unterdorf, 6146 Grossdietwil, Switzerland
Fax : 063 59 21 23 (after 9/11/96 : 062 927 21 23)

The poster presents an overview of the measures for biological control available and registered in Switzerland up to 1996. It focuses on recent products/possibilities for arbori- and viticulture such as NeemAzal-T/S against aphids as well as mating disruption technique against *Cydia pomonella*, *Adoxophyes orana*, *Grapholitha funebrana*, *Lobesia botrana* and *Eupoecilia ambiguella*. Moreover, it contains a list of the beneficial insects registered for greenhouses and vegetable crops.

"TECHNOLOGY TRANSFER IN MATING DISRUPTION"

Meeting of the IOBC/WPRS working group
"Use of pheromones and other semiochemicals in integrated control"

The smell of victory : Current and future roles for pheromones and pheromone research in IPM development in Indonesia

Nello ANGERILLI[1] and Agus Dana PERMANA[2]

1 : Dept of Biological Sciences, Simon Fraser University, Burnaby, B.C. V5A 1S6, Canada
E-mail : nello@server.indo.net.id
2: Dept of Biology, Bandung Institute of Technology, Bandung, Indonesia

The isolation, identification and application of pheromones in pest management programs in agriculture and forestry in Indonesia is only beginning. As the fourth most populous country in the world, home to some of the largest remaining tropical rainforests and undergoing rapid industrialization, there is substantial domestic and international pressure for Indonesia to develop and implement IPM programs that are effective, readily available and user-friendly. Recent examples of pheromone-based IPM programs in agriculture and forestry demonstrate the utility of the approach ; obstacles to both development and application and future needs.

The need for standardization in using pheromones for pest control

Heinrich ARN

Swiss Federal Research Station, 8820 Wädenswil, Switzerland
Fax : 1 780 63 41
E-mail : Heinrich.Arn@wae.faw.admin.ch

People using pheromones to monitor or control insect pests complain about insufficient reproducibility of results obtained with certain commercial products. In many cases, variability of chemical composition or release rate seems to be the cause.
The release of chemicals from dispensers is strongly dependent on temperature and other meteorological factors. This is critical in mating disruption. Manufacturers should specifiy composition and amounts per unit of time of chemicals released from a formulation under standard conditions. This is important not only in the beginning of the season, but mainly at the time of highest risk of infestation, when most dispensers are near depletion. Ageing procedures for simulating exposure under various climatic conditions need to be worked out.
Variability of lures can be caused by changes of chemical composition. Although attractant blends have been described in detail for most pest species, their efficacy can be impaired by impurities present in the synthetic material. Selection of the proper synthetic route and subsequent purification can help to avoid inhibitors, but the ultimate test is always the field bioassay. Therefore, the only method to provide a continuous supply of reliable lure is to produce a batch of synthetic product and test it before use. This procedure may go beyond the possibilities of most commercial enterprises but is conceivable as an international cooperative effort.

Pheromones mesaurements by air sampling

A.C. BÄCKMAN

ABSTRACT NOT AVAILABLE

A novel controlled-release device for disrupting sex pheromone communication

Thomas C. BAKER[1], Agenor MAFRA-NETO[1], Timothy DITTL[2] and Marlin E. RICE[1]

1 : Department of Entomology, Iowa State University, Ames, Iowa, USA
2 : Ocean Spray Cranberries Inc., P.O. Box 155, Necetah Rd, Babcock, Wisconsin, USA

A novel MSTRS system (Metered Semiochemical Timed Release System) was developed that consists of aerosol canisters loaded with gram amounts of pheromone that are timed to discharge their aerosol spray-burst onto a cloth pad, with the pad serving as a high-emission-rate point source that is rejuvenated with fresh pheromone every 15 minutes (Mafra-Neto A. and Baker T. C., 1996, J. Agr. Entomol. 13, 149-168). Pheromone discharged at a rate of 50µg/spray disrupted all mating of freely flying virgin *Cadra cautella* (Walker) males and females present in 3x3x2.5m storage rooms at moderate to high densities of a 2:1 male/female ratio for 24 hours. Up to 92% mating suppression was achieved at high moth densities (100 males, 50 females) over a 72 hour period in these rooms. Disruption was measured directly by recapturing females and examining their bursa copulatrices for spermatophores. MSTRS devices emitting only 5µg/spray caused greater than 60 percent mating disruption. One MSTRS device per room was as good as two per room at high moth densities and the 50µg/spray rate. Field tests were also commenced against the European corn borer, *Ostrinia nubilalis*, in grassy aggregation sites adjacent to Iowa corn fields, and against the Cranberry blackheaded fireworm, *Rhopobota naevana* (Hubner) in commercial cranberry bogs in Wisconsin. MSTRS devices emitting either *O. nubilalis* or *R. naevana* pheromone disrupted males' abilities to locate calling virgin females or female equivilent doses of synthetic pheromone by greater than 90% in most plots, at densities of 0.4, 2, or 3.2 canisters/hectare.

Goading growers towards mating disruption : the South African experience with Oriental fruit moth (*Cydia molesta*) and Codling moth (*C. pomonella*) (Lepidoptera : Tortricidae)

B.N. BARNES and T.L. BLOMEFIELD

Stellenbosch Institute for Fruit Technology, Private Bag X5013,
Stellenbosch, South Africa
Fax : +27 21 883 8669
E-mail : BRIAN@infruit.agric.za

Oriental fruit moth, *Cydia molesta* (Busck), and codling moth, *C. pomonella* (L.), are key pests in South African stone and pome fruit orchards respectively. *C. molesta* was accidentally introduced into the Western Cape in the late 1980's, where it has up to six generations per year. By 1991, despite comprehensive insecticide programmes, it had assumed epidemic proportions in canning peach orchards to the extent that some growers were considering removing productive trees. An area-wide mating disruption programme with Isomate-M was initiated during the 1991/92 season in 1 200 ha of peaches and nectarines in the Tulbagh Valley, involving 60 growers. While research entomologists acted as advisers, the local canning company coordinated the execution of the programme. The venture was an outstanding success. By the end of the season only 49 *C. molesta* adults had been caught in pheromone traps, shoot strikes were limited to an estimated 5%, while not a single infested fruit was recorded from the treated area.
C. pomonella has three generations per year in South Africa and a high reproductive potential. It has recently increased in pest status due to the development of a high degree of insecticide resistance. As an exercise in population reduction and resistance management, an Isomate-C+ mating disruption programme with supplementary insecticide intervention was implemented in pome fruit orchards. By the end of the third season, insecticide applications had been reduced by an average of 40%, and fruit infestation limited to an average of 0.5%. However, the acceptance by growers, compliance with recommendations and continuation of use of mating disruption of both pests has been disappointing, despite technology transfer efforts by experienced personnel. Main reasons include economic considerations, lack of sophistication of some growers, inadequate knowledge and technical backup, low profit margins, and the high reproductive potential of both pests. Some proposed solutions are discussed.

Confusion sexuelle contre le carpocapse des pommes : expérimentations du Pool ARBOTECH (UNCAA)

Pierre BECID

UNCAA (Union nationale des Coopératives agricoles d'approvisionnement),
83-85 avenue de la Grande Armée, 75782 Paris cedex 16, France
Fax : (33) +1 45 00 76 49

Face à la montée en pussance de ce ravageur dans les vergers français, les coopératives du Pool ARBOTECH (Pool Technique Phytosanitaire de l'U.N.C.A.A., spécialisé en arboriculture fruitière) ont, très vite, intégré dans leur programme expérimental la recherche de stratégies de lutte contre le carpocapse des pommes (*Cydia pomonella*). Parallèlement à l'élaboration de stratégies insecticides "classiques", l'évaluation prospective d'une méthode de protection par confusion sexuelle a été menée. Cette communication présente 4 expérimentations conduites sur ce thème en 1995 par trois coopératives du Pool ARBOTECH : la C.A.P.L. (84), ALPESUD(05) et UNION SET (37). Le matériel testé est fabriqué par la Société ISAGRO : il s'agit des diffuseurs ECOPOM (encore non homologués en France).
Les résultats obtenus démontrent que, bien utilisée, la confusion sexuelle peut être une voie d'avenir. Cependant, il convient d'insister sur les points suivants : la confusion sexuelle contre le carpocapse des pommes reste pour l'instant expérimentale; d'autre part, elle est davantage une technique qu'un produit : sans une formation des distributeurs et des utilisateurs, des échecs cuisants sont possibles. Enfin, loin d'être la panacée universelle, elle doit être intégrée (au sens "lutte intégrée") dans une stratégie globale de protection de verger.

Control of *Mythimna loreyi* (Duponchel, 1827) (Lepidoptera, Noctuidae) by pheromone traps in spinach crops

A. BENACHES, T. PINA, J. LÓPEZ, J. MORENO
and R. JIMÉNEZ

Departamento de Biología Animal (Entomología). Universitat de Valencia.
Dr. Moliner, 50, 46100 Burjasot, Spain
Fax : 34 6 386 4651
E-mail : Ricardo.Jimenez@uv.es

Mythimna loreyi (Duponchel, 1827) (Lepidoptera, Noctuidae) is known to be a pest in several countries of mediterranean area. It is especially linked to grass cultures, and present a variable incidence. Previously recorded in Spain, it had not been considered as a pest for our cultures, although some works pointed it out as a potential pest due to the damages caused in other European countries. This species does not cause direct damages to the spinach plant. However, its presence produces serious problems during the manufacturing process freezing, since adult population during the harvest time is high, and remains attached to the spinach leaves, causing rejection problems in the consignment destined to manufacture. In view to the protection of spinach crops our has been designed a pheromone mass-trapping method for *M. loreyi* control. The results obtained in this study show that the synthetic pheromone has a high efficacy and that the use of pheromone traps is a good method for protection of spinach crops of this noctuid.

Understanding mating disruption in the Pink bollworm

R.T. CARDE[1], R.T. STATEN[2] and A. MAFRA-NETO[1]

1 : Dept. of Entomology, University of California, Riverside, CA 92521, USA
2 : USDA-APHIS, Phoenix AZ, USA

The mechanisms responsible for achieving mating disruption of moth pests with the broadcast application of formulated pheromone have remained undefined, largely because of the difficulties involved both with the direct observation of moths in the field and the creation of lab assays that mimic field conditions. We have employed walk-in field wind tunnels set out in cotton fields to establish how various types of formulation alter male attraction of Pink bollworm moths (*Pectinophora gossypiella*) to point sources of pheromone. Males are released at the tunnel's downwind end, and success of orientation is guaged by capture in pheromone-baited traps and direct observation with video. Male orient to very high dose lures, and long-term exposure to pheromone seems to have little effect on a male's ability to locate a point source of pheromone. Other tests suggest that males are unable to locate point sources of pheromone when there is a high background of airborne disruptant. An off-ratio of pheromone, which is not attractive, may be as effective a disruptant as the natural ca. 1:1 blend. Disruption seems to rely on several mechanisms which vary with the type of formulation used.

Control of codling moth *Cydia Pomonella* L. with an attract and kill formulation

P.J. CHARMILLOT[1] and D. HOFER[2]

1 : Swiss Federal Research Station for Plant Production of Changins, 1260 Nyon, Switzerland
Fax :(41 22) 363 43 79
2 : Ciba-Geigy Limited, Plant Protection Division, 64002 Basle, Switzerland

An average drop is 0.1 ml in size and contains 0.16 mg of codlemone and 6 mg of permethrine. Males contacting a drop die within hours. Thus, reproduction is inhibited. The formulation is applied by hand with a specially developed application system.
In the two trials carried out in isolated orchards in 1995, 1200 and 2700 drops per hectare were applied. At harvest, both treatments kept the larval attack of Codling moth below a threshold of one percent. The hibernating population stayed at a low level.
Another trial was carried out to study the effect of 100 *versus* 5000 drops per hectare and the addition or respectively lack of an insecticide in the formulation. The trial was evaluated based on male trap catches and copulation of tethered females. Using drops without insecticide, trap catches were reduced to 50 % compared to the untreated check, independent whether 100 or 5000 drops/ha were applied. In both treatments the copulation of tethered females was practically not reduced. Standard A&K formulations reduced trap catches by 55 % and 94 % and copulation of tethered females by 54 % and 84 % at dose rates of 100 and 5000 drops/ha, respectively.
Trials show, that SIRENE^{R} CM in fact attracts and kills the males and therfore has a different mode of action from the disruption technique and is an effective agent in controlling Codling moth.

Sex pheromone identification of the leaf roller, *Bonagota cranaodes* (M.) (Lepidoptera : Tortricidae)

Alvaro E.EIRAS[1], Peter WITZGALL[2], C. Rikard UNELIUS[3], Marie BENGTSSON[2], Aadécio KOVALESKI[4], Evaldo F. VILELA[5], Anna-Karin BORG-KARLSON[3] and J.P. CHAMBON[6]

1 : Setor Semiquimcos, LPP-CCTA, Universidade Estadual do Norte Fluminense, Campos, RJ, 28045-620, Brasil
Fax : +55 247 263746
E-mail : ALVARO@UENF.BR
2 : Swedish University of Agricultural Sciences, Alnarp, Sweden
3 : Royal Institute of Technology, Stockholm, Sweden
4 : CNPUV/Embrapa, Vacaria, Brazil
5 : Universidade Federal de Viçosa, Brazil
6 : INRA, Station de Zoologie, Route de St Cyr, 78026 Versailles cedex, France

The leaf roller, *Bonagota* (*Phtheochroa*) *cranaodes* Meyr., is the most important lepidopteran pest of apple orchards in southern of Brazil. The larvae shelter between the fruit and leaves and are therefore difficult to control by insecticides. Monitoring of the seasonal flight period will increase the efficacy of such treatments. Traps baited with live virgin females have been used successfully, but the development of a synthetic lure is required for routine applications. The pheromone compounds produces by *B. cranaodes* females were identified by chemical analysis (GC/MS) and electroantennography (GC/EAD). The main compound, (E,Z)-3,5-dodecadienyl acetate has been identified for the first time in family Tortricidae.Traps baited with 100 µg (E,Z)-3,5- dodecadienyl acetate were attractive over 15 weeks in the field and were as effective as baited traps.

Applying sex pheromones of Douglas-fir tussock moth, *Orgyia pseudotsugata*, and their control potential when integrated with virus application

Michael HULME and Tom GRAY

Pacific Forestry Centre, 506 West Burnside Rd.,
Victoria, V8Z 1M5, Canada
E-mail : MHULME@A1.PFC.Forestry.CA

When the sex pheromone of the Douglas-fir tussock moth, *Orgyia pseudotsugata*, was applied to a forest stand infested with this insect, mating was completely disrupted and no eggs were laid. Application of a nuclear polyhedrosis virus also controls outbreaks of this pest. Integrating the two methods promises to be more useful for the forest manager than either method alone. We transfer this technology to the user in several ways, including the use of video.

Use of pheromones in biological control against *Zeuzera pyrina* L. on hazel-nuts in Spain

J. ISART *et al.*

LEAAM-Agroecology, CID,
Consejo Superior de Investigaciones Científicas
Jordi Girona 18-26, 08034 Barcelona, Spain
Fax : 34.3.2045904
E-mail : leaam@cid.csic.es

Zeuzera pyrina L. (Lepidoptera, Cossidae) is a pest of hazel nuts (*Corylus avellana* L.) and appel (*Pyrus malus* L.) in Spain. A study of the methods of control against this pest was conducted in Tarragona (province of Spain) on hazel nuts. The results of mass trapping during 1995-1996 are given.
Success in this method of control is studied.

Amazing Daze : tales of futures past

P. KIRSCH

IPM Technologies, PO Box 509, West Linn OR 97068, USA
Fax : 001 503 557 5527
E-mail : semiochem@aol.com

Over the past 30 years, considerable attempts have been made to commercialize semiochemicals for pest management. Success has been realized in many different crops, and on several continents, in applications ranging from monitoring tools, through mating disruption and mass trapping. The industry is characterized by constant change: in practise and in products.
This paper will attempt to review global semiochemical applications, and discuss some of the limitations and new developments. The author will also offer his comments on the future of this nascent technology.

Behavioural effects of mating disruption dispensers on Codling moth

A.L. KNIGHT, T. WEISSLING and H. HILL

USDA-ARS, Wapat, Washington 98 951, USA

Various studies that address the behavior of Codling moth within orchards treated with mating disruption have been conducted since 1992. Habituation of male response to high levels of sex pheromone was demonstrated in the laboratory, but was not an important factor at levels present in a disrupted orchard. Using a passive interception trap we have demonstrated that male movement within the canopy and among trees in disrupted orchards is profoundly affected, but female distribution patterns remained unaffected. Exposure to high levels of pheromone increased the calling frequency of virgin females, but did not affect rates or timing of oviposition. However, in choice experiments, females oviposited a significantly higher number of eggs on surfaces not treated with sex pheromone. Efforts to clarify the mechanisms of mating disruption continue.

Over several years we have found that approximately 50% of females are mated in disrupted orchards. The much higher reduction in fruit injury observed does not appear to be due to an increase in egg predation. Delay of mating by females of four days or more in the laboratory significantly reduces viable oviposition. Field trials attempting to demonstrate the importance of mating delay are continuing.

Pheromone measurements by field EAG in apple orchards

Uwe T. KOCH[1], Lüder WOLFGANG[1], Stephan CLEMENZ[1]
and Liliana I. CICHON[2]

1 : FB Biologie, Universität Kaiserslautern, 67653 Kaiserslautern, Germany
2 : INTA Alto Valle, 8332 Gral. Roca (R. N.), Argentina

Measurements of airborne pheromone concentrations can be an important contribution to the further development of mating disruption methods. Investigations of wind effects, plant-surface interactions and concentration profiles in vertical and horizontal dimensions all require the acquisition of many measurement data points in time intervals which can be considered short in comparison to the time course of changes in temperature or wind. While methods using the air sampling and subsequent GC analysis offer absolute average concentration values, they are not fast enough to yield a sufficient number of measurements in the required time.
Field EAG measurements of pheromone concentrations can offer reproducible concentration values on a relative scale which can be calibrated to absolute values using GC measurements. In order to avoid erroneous interpretations of the EAG signals, it is important to continuously calibrate the antenna using at least two calibration standards of different concentration. The calibration yields two independent antenna parameters, sensitivity and threshold. Antennal responses to plant odours and other non-pheromone stimuli can be compensated by using a signal superposition technique and appropriate evaluation methods.
Our EAG system uses three calibration sources for the reconstruction of the dose-response curve of the antenna repeated every 50 s. The measurement probe is fully remote controlled and can be operated on a pole up to 5 m high. Wind velocity and direction are recorded in 40 ms intervals by two vector anemometers, one mounted on the EAG-probe, the other at 5.7 m height.
Pheromone concentration measurements were made in various apple orchards treated for mating disruption of *Cydia pomonella*, in Southern Sweden, investigating effects of tree size and spacing on pheromone distribution as well as simultaneous EAG and air sampling measurements. "Blank" background signals, fluctuating "odour plumes" and a continuous "fog-like" pheromone atmosphere have been measured in direct sequence due to fast wind shifts, which were recorded simultaneously. These recordings provide insights into the power of field EAG measurements in combination with high resolution vector anemometry.

Optimization of pheromone mixtures for monitoring some species of Noctuidae

K.V. LEBEDEVA[1], Yu. B. PYATNOVA[1], V.V. BULEZA[2] and V.S. VASIL`EVA[2]

1 : Institute of Plant Protection Chemicals, Moscow, Russia
Fax : 095 208 49 70
E-mail : sasha@teluk.msk.ru
2 : Severtsov Institute of Ecology and Evolution, Moscow, Russia

Our method for making pheromone mixtures takes into account the following phenomena : 1) the main pheromone components in related species of Noctuidae are often the same; 2) the activity and specificity of pheromone mixtures depend upon the presence of minor components and the ratio of their concentrations in respect to that of the main component; 3) the effect of the pheromone mixture upon the recipient sex is dose-dependent.

The procedure comprises several steps : collection of volatiles from calling females, identification of natural pheromone by means of GLC/MS, synthesis of original pheromone components, their laboratory screening including EAG and behavioral tests in the wind tunnel, field tests of a set of mixtures composed of natural components with various concentration ratios and dosages in geographically distant populations.

We found that the main component of the pheromone of five species among seven studied is Z11HDA. The optimal mixtures for monitoring of *Mamestra suasa* and *M. oleracia* in three geographical regions were found to be different in ratio of components or in doses. Mixtures of 2-3 components found in pheromone of *Amathes C-nigrum* from two different districts were effective in four geographically distinct populations with different dosages and component ratios.

The pheromones of *Agrotis segetum* from Usbekistan and central Russia were found to consist of 11 and 10 components respectively. For each of the five regions where the field tests were conducted, we found an active mixture of five components, four of which were the same in all populations. Their activity was enhanced by the fifth which was population specific.

In Ussury district one can perform monitoring of *Mythimna separata* and *Scotogramma trifolii* with the help of mixtures comprising 3 components of their natural pheromones each.

Significant interpopulation variation characteristic for pheromone structure of *Mamestra brassicae* is correlated with variability of mixtures for this species monitoring.

Mating disruption of the Olive moth (*Prays oleae* Bern) by the major sex pheromone component (Z)-7-tetradecenal

B.E. MAZOMENOS

Institute of Biology NCSR, "Demokritos", 153 10 Aghia Paraskevi, Attikis, P.O Box 60228, Greece

The mating disruption method to control the Olive moth *Prays oleae* (Bern) [Lepidoptera : Hyponomeutidae] was tested from 1992 to 1995 in olive groves in Greece. The major sex pheromone component (Z)-7-tetradecenal was formulated in β-Cyclodextrin (β-CD) and polyvinyl chloride polymers (PVC, AgriSence-BCS, Ltd). Both formulations proved to be effective dispensers of the pheromone in the field. Pheromone trap captures were reduced to 96-100% in the treated plots and fruit infestation remained at commercially acceptable levels. During the first year of the mating disruption programme a treatment with *Bacillus thuringiensis* (var. kurstaki) was required to reduce the first generation larvae. The effectiveness of the method depends on the availability of oviposition sites and dispenser density. In low fruiting years fruit damage is higher compared to those of high fruiting years. Three dispensers per tree gave better results than two with the same dose of pheromone per hectare. Multi-annual application of the mating disruption devices in the same olive groves progressively reduced moth populations from one year to the next.

Sex pheromone traps as a tool to study population trends of the principal predator of a scale insect and to select potential predators for biological control

Z. MENDEL, K. ADAR, D. NESTEL and E. DUNKELBLUM

Plant Protection Institute, Agricultural Research Organization,
The Volcani Center, Bet Dagan, 50250, Israel
Fax : 972 3 9604180

At present *Elatophilus hebraicus* (Heteroptera: Anthocoridae) is a unique example of a predator species attracted to the sex pheromone of its prey. The attraction of *E. hebraicus* to traps baited with the sex pheromone of *Matsucoccus josephi* (Homoptera: Matsucoccidae), permitted to examine the population trends of the predator as related to those of its only prey, a pernicious scale pest of pine forests in the Near East. Over 27 months traps baited with the sex pheromone of *M. josephi* were exposed at monthly intervals in pine stands of *Pinus halepensis* and *P. brutia* ssp. *brutia* in several regions in Israel. A Fourier series was used to modulate population trends. Both insects are multivoltine. The population density of *M. josephi* increased in March-April and during August and October. These peaks coincided with the main period of cambial activity of the pine. A steep increase in catch of *E. hebraicus* was noticeable during May and June. The rise of the predator population was only partially related (r2=0.375) to the increase in prey density in the previous spring. An inverse relation was found between the densities of *M. josephi* and *E. hebraicus*. Population trends of both prey and predator varied slightly among regions, but not between host plant species, despite their different susceptibility to *M. josephi*. We also exposed pheromone -baited traps in other areas of the Palaeartic region where *Matsucoccus* spp. occur. The objectives were to determine the geographic distribution of *E. hebraicus* and to identify other potential predators of *M. josephi* on the basis of their kairomonal response to the scale's sex pheromone. It was found that the range of *P. brutia* ssp. *brutia* in the East Mediterranean coincides with the natural area of *E. hebraicus* and *M. josephi*. *Elatophilus crassicornis* and *Hemerobius stigma* (Neuroptera: Hemerobiidae) are also significantly attracted to the pheromone; both predators being caught only in *Pinus pinaster* stands in Portugal. These insects are now under consideration as a mean to augment the enemy fauna of *M. josephi* in Israel. *E. hebraicus* is also attracted by the sex pheromone of two other allopatric bast scales, *M. feytaudi* and *M. matsumurae*. Hence, *E. hebraicus* may be considered a candidate to improve the biological control in extensive pine forests injured by these scale insects.

Technological problems associated with use of Lepidopteran pheromones in Insect management

Jocelyn G. MILLAR[1], Harry SHOREY[1] and Richard E. RICE[2]

1 : Dept. of Entomology, University of California, Riverside CA 92521, USA
Fax : 001 714 787 3086 E-mail : Millar@ucraci.ucr.edu
2 : Dept. of Entomology, University of California, Davis CA 95616, USA

Sex pheromone components were identified for many of the major lepidopteran pests of concern to agriculture a number of years ago. However, despite intensive efforts, implementation of large-scale, effective, and reliable insect control strategies based on pheromones remains elusive for many pest species. For example, pheromone-based mating disruption of codling moth still suffers from problems which have prevented the widespread adoption of mating disruption for control of this world-wide pest, in spite of continuous research efforts spanning 20 years. During this time, our knowledge and understanding of possible causes of unsatisfactory performance of pheromones in insect pest management has increased substantially. It is now commonly acknowledged that site characteristics such as the slope of a site and edge effects associated with adjacent cropping systems can contribute to the failure of mating disruption. However, field researchers have focused less attention on technological factors such as pheromone dispenser performance, pheromone purity, and pheromone degradation under field conditions. Both field researchers and growers are usually dependent on commercial outlets for supplies of both pheromone and dispensers, but the pressure on commercial suppliers to market product may result in products being released before adequate efficacy tests under field conditions have been conducted. In this paper, we will summarize results from several studies which examined pheromone dispenser characteristics, pheromone purity, and pheromone degradation under field conditions. The results will be discussed in terms of the performance of several pheromone products in different agricultural systems.

Pheromones of Lepidoptera : Research and business in the Dutch way

Albert MINKS and Simon VOERMAN

Research Institute for Plant Protection (IPO-DLO),
P.O. Box 9060, NL-6700 GW Wageningen, The Netherlands
Fax : 0031 8370 10113
E-mail : A.K.Minks@IPO.DLO.NL

Pheromone research was started at the then called Laboratory for Research on Insecticides (LIO) at Wageningen in 1967. Leafroller moths were chosen as subject for our studies, because they formed at that time a major problem in pome fruit orchards in the Netherlands. After we had shown that live female traps worked effectively, we set our first goal, namely the identification of the sex pheromone of the Summer fruit tortrix, *Adoxophyes orana*. The publication of the pheromone identification of the closely related Red-banded leafroller, *Argyrotaenia velutinana*, by Roelofs & Arn in 1968 came right on time and helped us in convincing our research leaders to go ahead with our plans. A fruitful collaboration between LIO and TNO at Delft (F.J. Ritter & C.J. Persoons) was initiated and continued until 1980. Pheromone identifications of several leafrollers and other moth species followed, starting with that of *A. orana* in 1971.

In addition to the identification studies, we developed our own lines of research: a chemical line (S. Voerman) and an entomological line (A.K. Minks). Our major objective was to make pheromones usable in agricultural practice, according to the institutional policy, which we fully endorsed. We both preferred a down-to-the-earth approach in our research, probably due to our Friesian origin, and we have tried to keep things straightforward e.g. by taking our chemicals directly into the field for activity testing. During all these years we kept our believe in the great potential of pheromones. These substances are unique in their species specificity and low toxicity, which are very favourable characteristics for pest control agents in integrated control systems. The great potential has been illustrated by the large number of pheromone traps used nowadays for monitoring. Since more than 30 years tens of thousands of traps have been used worldwide in many different cultures and this number is still growing.

Our institute has made a significant contribution to that development along the chemical line (through the systematic production of potential pheromone compounds of high quality), as well as along the entomological line (through studies on the relationship between trap catches and moth numbers actually present in the field). The chemical line eventually resulted in the operation of the IPO pheromone shop, which was first meant just as a service to colleagues, but in recent years more and more developed to a

commercial enterprise. The entomological research line shifted attention to mating disruption in the eighties and was mainly occupied in efficacy testing of various commercially available dispensers, first on orchard pests, and more recently also on noctuid pests in greenhouse cultures. The renewed collaboration with TNO should be mentioned here (J.J. de Vlieger & J. Klijnstra), since TNO polymer dispensers are used in the greenhouse tests. Highly fruitful is also our cooperation with Syntech (J.v.d. Pers), in which portable EAG and SSR equipment is being developed that enables direct measurement of pheromone concentrations in ambient air.
At the meeting we will present a more detailed account of our experiences, but we can conclude right now that in pheromone research, close cooperation between chemists and entomologists is indispensable.

The key to success of mating disruption

K. OGAWA

Shin-Etsu Chemical Co., Fine Chemicals Dept., 6-1,
Ohtemachi 2-chome, Chiyoda-ku, Tokyo 100, Japan
Fax : 0081 3 3246 5371

There are a number of factors influencing the effect of mating disruption. We would like to discuss on the following factors: pheromone concentration, weather condition, field location, population density and natural enemies.
An adequate dispenser system is essential for reliable and cost-effective maing disruption. Dispenser life and release rate, stability of active ingredient, and the number of dispenser per area are important factors. We would like to discuss on the ideal dispenser system.
For pheromone applications, growers and researchers should observe safety margins, with respect to applying large enough amounts of active ingredient, sufficiently early in season. However, most growers focus on a reverse direction to get a cheaper product.
When pheromones are applied on a large surface, mating disruption works very well and the populations of target pests decrease year by year. Even the minor pests are usually controlled by their natural enemies.
In some cases, the importance of minor pests increases as a result of the reduction of insecticide sprays. This can be overcome by combining pheromone compounds of several species, as for example, codling moth and leaf rollers.

Pheromones to control *Zeuzera pyrina* L. and *Cossus cossus* L. (Lepidoptera Cossidae) : status of the art

E. PASQUALINI, S. CIVOLANI, G. GALZORALI
and S. VERGNANI

In 1992, 1993, and 1994, a number of experimental studies on *Zeuzera pyrina* L. (Lepidoptera : Cossidae) (Leopard moth) were conducted in some pear and apple orchards in the Provinces of Ferrara and Bologna, in order to identify the more suitable trap to study the application of the mass trapping method.
Therefore, this work evaluated the efficacy of a number of traps (glue-and funnel-type) with suitable improvements over standard models. Another field of inquiry was evaluation of the influence of trap colour on captures.
During 1995, both for *Z. pyrina* and *Cossus cossus* L.(Lepidoptera, Cossidae) (European goat moth), were carried out some trials to evaluate the attractivity of several kind of dispensers.
The results showed that the funnel traps capture more *Z. pyrina* adults than do glue traps. In funnel traps, the absence of barriers between the roof of the trap and the funnel proved a determining factor in improving performance.
Trap colour does not appear to influence catches, although the greatest number of adults were observed in white traps.
In the orchards in which the various tests were carried out, and in which the traps were present for three consecutive years, a decrease of capture rate of approximately 60-70 % was observed between the first and third year.
About the comparison of different dispenser types for *Z. pyrina*, the captures obtained were insufficient to have definitive conclusions. This few catches was due essentially to unfavourable climatic conditions, even though one dispenser (type C-TNO) was more attractive than the others. For *C. cossus*, the most effective dispenser was the Isagro type. About the trap number/hectare, no differences in captures were observed between 5 and 10 traps.

Efficacy of mating disruption pheromones in paraffin emulsion dispensers

R. E. RICE[1], C. A. ATTERHOLT[2], M. J. DELWICHE[2] and R. A. JONES[1]

1 : University of California, Kearney Agricultural Center, Parlier, CA 93648, USA
Fax : 209 891 2593 E-mail : rerice@uckac.edu
2 : University of California, Dept of biological and agricultural Engineering, Davis, CA 95616, USA

Pheromones of Oriental fruit moth, *Grapholitha molesta*, Peach twig borer, *Anarsia lineatella*, and San Jose scale, *Quadraspidiotus perniciosus*, were mixed into sprayable emulsions of water and paraffin and applied to stone fruit trees for mating disruption of these pests. Pheromone/emulsion blends were applied twice for OFM and PTB during the 1995 season, using 49.4-76.6 g a.i. per ha. per application; SJS pheromone was applied only once at 74.0 g a.i. per ha. Efficacy of mating disruption was measured by pheromone trap (OFM, PTB) or sticky tape (SJS) collections and infested fruit.
Results of these field trials showed that treatments using OFM pheromone were comparable to commercial mating disruption dispensers, with monitoring trap shutdown for 8-12 weeks, and reduction of fruit infestation of 80-90 percent. The PTB pheromone treatments were less effective, with increased collections of PTB moths in monitoring traps and no differences in infested fruit compared to untreated checks. Mating disruption of SJS using pheromone in paraffin emulsions showed some reduction in crawler populations for two generations after treatment, but the cost of pheromone is prohibitive for commercial use.

Research regarding use of *Ostrinia nubilalis* pheromone in warning and control of ECB (*Ostrinia nubilalis* Hb.) in Romania

I. ROSCA, P. WITZGALL,V. BRUDEA, E. BUCUREAN, F. MURESAN, A. UDREA and M. VOICU

Research Institute for Cereals and Industrial crops,
8264 Fundulea, Romania
Fax : 40 1 310722

The resuls reveal the relatively high number of *O.nubilalis* males captured through the period under study, this number varying according to the locality, year and pheromone variant used. It is to stress that it was undoubtedly for the first time that in Romania both pherotypes of this species, cis (Z) and trans (E) exist , this later being not recorded in the centre and north-east of the country.
Pheromone traps is used to drow up flight curves of *O.nubilalis* males in the first and second generation.
There are presented results regarding disorientation of males with pheromone and mass capture of males for control of pest in isolated field plots.
Releasing and recapture of marked moths was used in order to compare behavior of F-1 males, released in field 10 days before appearing of wild moths in field and recapture in pheromone trap situated at 25, 100 and 200 m in 1993 and at 100, 200 and 300 m in 1994 far from releasing site in N,S,E and W direction. The marked moths have to be checked individually for the red color of abdomen.
Using of pheromone for mass capture of male moths has not succeed, percentage of attacked stems, number of larvae/attacked plant was bigger in 1993 than for control, thus it was observed an increasing of population of pest with 31 %, only in 1994 it was a slight reducing of pest population with 8.5 % and in 1995 it was the same. Even that male disorientation of male moths seems to determine a reducing of pest population with 43.7 % in 1993, the results were no confirmed in 1994 and 1995, when it was registered an increasing with 0.3 % respectively 22.3 %.
Mass capture of ECB males was made by 16 sticky traps/ha (25/25m). The lure cis (Z) was changed weekly and total number of males captured was 231 in 1993 and 325 in 1994. Mating disruption (male disorientation) was made by 100 cis (Z) lure/ha which were changed weekly during the first flight of moth (15 june-30 july).

The use of pheromone traps in Spruce budworm integrated pest management : from research to operational use

C.J. SANDERS

Canadian Forest Service, P.O. Bo 490, Sault Ste Marie, Ontario, P6A5M7, Canada
Fax : 0017057595700
E-mail : sanders@fcor.glfc.forestry.ca

The Spruce budworm is a cyclical pest with 35-40 years between outbreaks. As with many other forest pests, control relies on detection of defoliation and then aerial spraying to kill sufficient larvae to allow the trees to survive. As forest management becomes more sophisticated, integrated pest management techniques are being developed that depend upon the early detection of outbreaks followed by harvesting of the most vulnerable and valuable stands. Between outbreaks Spruce budworm can become extremely scarce and conventional larval sampling techniques become prohibitively expensive. However, sex pheromone traps have proved to be extremely efficient and effective for monitoring low density populations. They have now been deployed at more than 700 sites throughout North America for 10 years as a method of monitoring low density populations to determine when more intensive larval sampling is warranted. On a local scale (forest management units) catches of over 100 moths per season are used as a threshold to trigger more intensive larval sampling. On a provincial and national scale trap catches are converted from point sample data to contour maps by a geostatistical process called 'kriging'. The resultant maps are being compared with those from previous years by use of Geographic Information Systems to locate areas where populations are showing significant changes in density.

The role of foliage in mating disruption in apple orchards

D.M. SUCKLING and G. KARG

HortResearch, PO Box 51, Lincoln, New Zealand
Fax : +64 3 32 52 946
E-mail : sucklingm@hort.cri.nz

Apple foliage affects atmospheric concentrations of pheromone by absorbtion and release, according to a) electroantennogram (EAG) measurements, b) behavioural studies with *Epiphyas postvittana* (Lepidoptera: Tortricidae), and c) chemical analysis. EAGs indicated that apple leaves exposed to an airstream containing pheromone reached a constant level of release within three minutes. Leaves from a pheromone-saturated environment released pheromone over a period > 24 hr. Leaves sampled near a pheromone source in the orchard had detectable pheromone levels for a distance of 25 cm from the source. Concentrations in a treated orchard were higher near foliage, but with fewer fluctuations, than above the grassed inter-row. Removal of dispensers from treated plots resulted in a rapid drop in the mean pheromone concentration, within minutes. The pheromone plume structure in continuous EAG recordings disappeared as soon as dispensers were removed.

Leaves exposed to pheromone were effective as lures in sticky traps, but the catch decreased logarithmically over three nights. Removal of dispensers from orchard plots - so that leaves were the sole source of disruptant - continued to reduce catch for one to two days. On three nights following the removal of 10 dispensers/tree, trap catch to synthetic lures was 0, 10 and 15% of the control. On the three nights following removal of one dispenser/tree, catches were 35, 40 and 80% of the control. Release of marked naive male moths into apple orchard plots following the removal of dispensers resulted in significantly lower catches in traps baited with virgin females in blocks which had been treated, compared to controls.

Recovery of pheromone by solvent washing of leaves, followed by coupled gas chromatography-electroantennogram analysis showed that leaves held in a saturated atmosphere were loaded with 0.045 ± 0.007 µg (E)-11-tetradecenyl acetate/cm^2. Leaves taken from a pheromone-treated tree up to one metre from the dispenser showed a logarithmic decay of the pheromone load with increasing distance from the dispenser, as indicated by EAG. At 4 cm from the source of pheromone, leaves contained an estimated 0.014 ± 0.009 µg of (E)-11-tetradecenyl acetate.

Codling moth mating disruption field trials with TNO dispensers from 1991 up to 1995

Jan de VLIEGER and J.W. KLIJNSTRA

TNO Institute of Industrial Technology, P.O. Box 6031, NL-2600 JA, Delft, The Netherlands
Fax : +31 15 269 65 01
E-mail : J.deVlieger@kribc.tno.nl

In five consecutive years (1991-1995), TNO has tested the method of mating disruption with a new type of dispensers against the Codling moth (*Cydia pomonella*) in apple orchards in Spain, The Netherlands, Switzerland, France, Italy, Hungary, California, Washington, Israel and South Africa. Codling moth dispensers in the form of flat square wafers, releasing 70 g of Codlemone (E,E-8,10-dodecadien-1-ol)/ha/season were applied at densities of 500/ha. Because average temperatures can vary in different parts of the world, controlled release dispensers were made, adapted to local circumstances. Here, we present the results of the biological efficacy of the mating disruption method as well as the release characteristics of the dispensers sampled at the different locations.
In all cases the mating disruption treatments resulted in season-long satisfactory control of Codling moth. Examples and release patterns will be given of using the same technology in mating disruption of other insect species.

Three years of large-scale control of Codling moth by mating disruption in South Tyrol, Italy

Walther WALDNER

South Tyrolean Advisory Service for Fruit- and Winegrowing,
39011 Lana, Italy
Fax : +39 473 564 220

In 1990 we noticed, at first only in a few places, that Codling moth populations were increasing in orchards treated with difubenzuron. In 1992 it was proven that some Codling moth strains had developed a strong resistance to chitin synthesis inhibitors. The resistance appeared after about 25 CSI applications. After having thus lost an important component of our pesticide list, there was the danger that we would not be able to make an IFP-programme that would come up to international standards. Therefore we introduced the mating disruption method on a large scale.
In 1991, we started with RAK 3+4 on 110 ha. As in 1992 RAK 3+4 were not available, we made a new start with Isomate-C dispensers on 232 ha in 1993. In the following year, the disrupted area expanded to 2500 ha and reached 4500 ha in 1995. In 1996, the treated area is 3500 ha.
The paper contains the results obtained with different dispenser types (Isomate-C, Isomate-C-Plus, Isomate-C-Special, RAK 3+4, Ecopom Combi) against codling moth and leafrollers. Mating disruption was successful even on hillsides and smaller plots. Finally, organization problems concerning the planning of mating disruption projects will be dealt with in the paper.

Mating disruption of Pea moth, *Cydia nigricana*, using a blend of sex pheromone and attraction antagonists

Peter WITZGALL

Swedish University of Agricultural Sciences, Box 44, 23053 Alnarp, Sweden
Fax : +46 40 46 21 66
E-mail : peter.witzgall@vsv.slu.se

The sex pheromone of Pea moth, *Cydia nigricana*, is *E*8,*E*10-12Ac; the geometric isomers *E,Z*-, *Z,E*-, and *Z*8,*Z*10-12Ac are strong attraction antagonists. The males are attracted to fresh dispensers releasing 98% pure *E*8,*E*10-12Ac. After isomerization of *E*8,*E*10-12Ac, males are no longer attracted to the dispensers, but fly out of the treated pea fields. Male moths respond to synthetic pheromone up to one hr before the onset of female calling. For mating disruption, it may be more effective to repel males by attractant/antagonist blends during this premating flight period, rather than to attract them.

Attraction of males to calling females, and to traps releasing ca. 100 times more E8,E10-12Ac than calling females, was entirely suppressed at aerial pheromone concentrations ranging from 2 to 7 ng/m3 within pea canopy (average release rate: 110 mg/ha/day; 6 to 9% of E,Z; Z,E; Z,Z isomers). At lower release rates of 45 and 14 mg/ha/day, resulting in pheromone concentrations of 3 and <2 ng/m3, reduction in trap catch was 87 and 68%, respectively, and a few males were attracted to cages with calling females.

Mating disruption treatments can be specified by a few basic parameters. These must be assessed in order to interprete and optimize applications. They can be measured with current techniques, but are most often incompletely available from literature: (1) chemical composition of the disruptant and its behavioral effects at low doses, compared to female pheromone, (2) dispenser placement, release rate, aerial concentration and dispersal of disruptant, (3) communication disruption, as measured by attraction to traps or females, in relation to population density. The measurement of these parameters is prerequisite for the comparison of applications and for the identification of the behavioural mechanisms of mating disruption.

AUTHOR INDEX

—A—

—B—

—C—

—D—

—E—

—F—

—G—

—H—

—I—

—J—

—Q—

—R—

—S—

—T—

—U—

—V—

—W—

—Y—

—Z—

Due to some last minute problems, some pages related to keywords may be wrong, please just see the previous or the following page.

KEYWORD INDEX

—A—

—B—

—C—

—D—

—E—

—F—

—G—

—H—

—P—

—Q—

—R—

—S—

—T—

—U—

—V—

—W—

—X—

—Z—

Le CILBA ou Complexe International de Lutte Biologique Agropolis est une structure fédérative d'Agropolis, pôle international de recherche et d'enseignement supérieur dont la spécificité est l'agronomie et le développement des régions chaudes, méditerranéennes, tropicales et subtropicales. Neuf établissements d'enseignement supérieur ou de recherche, établis à Montpellier, ont souhaité organiser l'information réciproque et le développement d'actions communes de recherche, de développement et de formation dans le domaine de la lutte biologique, suivant le concept de développement durable.
Ces neuf établissements fondateurs sont les centres de Montpellier du CIRAD, du CNRS, de l'INRA et de l'ORSTOM, l'Ecole Nationale Supérieure Agronomique de Montpellier (ENSA.M), l'Université de Montpellier-II, les laboratoires américains et australiens de lutte biologique implantés également à Montpellier (USDA et CSIRO), ainsi que la Mission de Coopération Phytosanitaire du ministère de l'agriculture (MCP).

CILBA (International Center of Biological Control Agropolis) is a center of research and higher education made up of a group of institutions in Agropolis, an international center for mediterranean, tropical and subtropical agronomy and development. Nine institutions of higher education and research, located in Montpellier, cooperate in exchange of information and engage in common activities in research, development and education in the area of biological control.
These nine founding institutions are the Montpellier centers of CIRAD, CNRS, INRA, ORSTOM, the Ecole Nationale Supérieure Agronomique at Montpellier (ENSA.M), Montpellier-II University, the American and Australian biological control laboratories established at Montpellier (USDA and CSIRO) and the Office of Phytosanitary Cooperation of the French Ministry of Agriculture (MCP).

For further information contact
P. FERRON, President of CILBA,
INRA-URLB, Campus International Agropolis de Montferrier, Baillarguet,
34982 Montferrier-sur-Lez, France,
telephone 04 67 59 31 20, fax 04 67 59 31 22

IOBC / OILB
40 years
for agriculture & environment

C.I.L.B.A

MINISTERE DE
L'ENSEIGNEMENT SUPERIEUR
ET DE LA RECHERCHE

AGROPOLIS

INRA

MONTPELLIER DISTRICT

CONSEIL
GENERAL
DE L'HERAULT