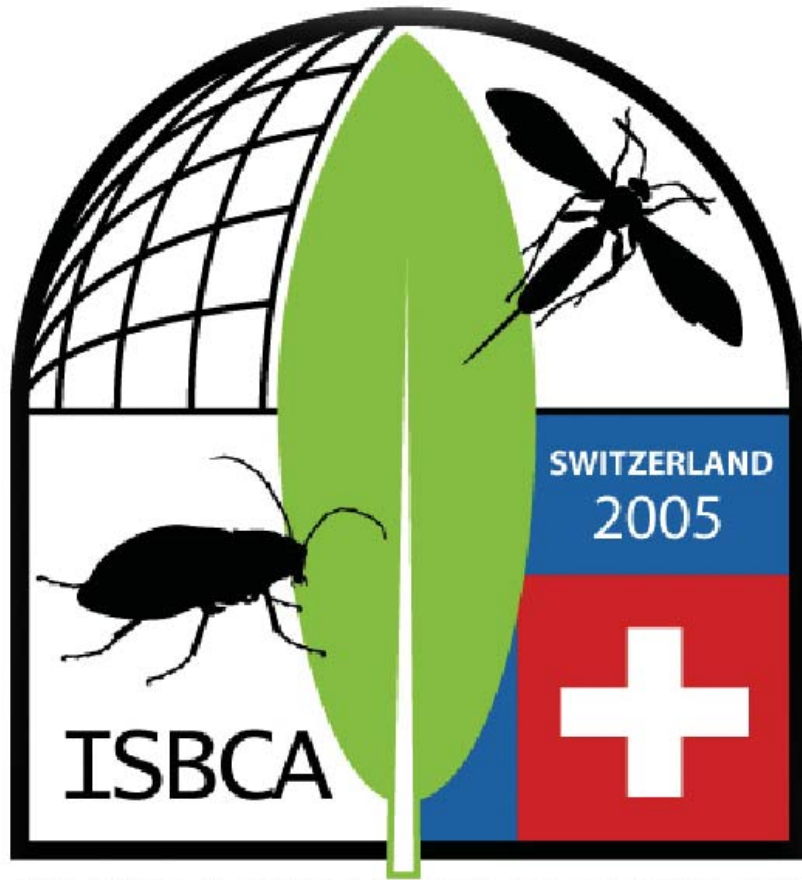


Forest Health Technology Enterprise Team

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TRANSFER

Biological Control



INTERNATIONAL SYMPOSIUM ON BIOLOGICAL CONTROL OF ARTHROPODS

September 12-16, 2005

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**SECOND INTERNATIONAL SYMPOSIUM ON
BIOLOGICAL CONTROL OF ARTHROPODS**

**DAVOS, SWITZERLAND
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SECOND INTERNATIONAL SYMPOSIUM ON THE BIOLOGICAL CONTROL OF ARTHROPODS

The Second International Symposium on the Biological Control of Arthropods held in Davos Switzerland builds upon the foundation laid at the first meeting in Hawaii in January 2002. The intent of the ISBCA meetings is to create a meeting for practitioners, a forum for information exchange, an event to build cohesion among the research community, and to foster discussions of issues effecting biological control work, particularly pertaining to the use of parasitoids and predators as biological control agents.

To this end, a 14 session conference with invited has been designed to address the most interesting and relevant research topics that have broad international application. The oral sessions have been complimented with unsolicited poster presentations prepared by over 100 different scientists from around the world. Topics covered at ISBCA II are diverse and include invasion biology and application to biological control, biological control of arthropod pests of conservation importance, the role of biological control for pest management in developing nations, and emerging experimental protocols and legislation for assessing natural enemy specificity and safety.

The printed ISBCA II conference proceedings are large, indicating the great interest in the content of this meeting. The two volume proceedings only include the articles prepared by invited speakers. The accompanying CD has an electronic version of the conference proceedings and the abstracts of approximately 115 posters that were presented at the meeting and perused by over 200 meeting attendees representing the international biological control community.

ACKNOWLEDGEMENTS

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SCIENTIFIC SESSION ORGANIZING COMMITTEE MEMBERS

Session 1: Sandy Smith, University of Toronto, Ontario Canada, and Heikki Hokkanen, University of Helsinki, Helsinki Finland. **Session 2:** Mark Hoddle, University of California Riverside, California U.S.A., and Matthew Cock, CABI Bioscience, Delémont Switzerland. **Session 3:** Nick Mills, University of California Berkeley, California U.S.A., and Jacques Brodeur, Université de Montréal, Quebec, Canada. **Session 4:** Steve Wratten, Lincoln University, Canterbury New Zealand, and Geoff Gurr, University of Sydney, Orange NSW, Australia. **Session 5:** Juan Manuel Alvarez, University of Idaho, Aberdeen Idaho, U.S.A., and Shu-sheng Liu, Zhejiang University, Hangzhou, P.R. China, China. **Session 6:** Janny Vos, CABI Bioscience Nederland, Leusden The Netherlands, and Peter Ooi, FAO, Bangkok Thailand. **Session 7:** Joerg Romeis, Agroscope-FAL, Zurich-Reckenholz Switzerland, and Tony Shelton, Cornell University Geneva, New York U.S.A. **Session 8:** Felix Wäckers, NL Royal Academy of Sciences, Harlem The Netherlands, and Henry Fadamiro, Auburn University, Alabama U.S.A. **Session 9:** Bob Pfannenstiel, USDA-ARS, Weslaco Texas, U.S.A., and Matthew Greenstone, USDA-ARS Beltsville, Maryland U.S.A. **Session 10:** Steve Naranjo, USDA-ARS Phoenix, Arizona U.S.A. and Patrick de Clercq, Ghent University, Ghent Belgium.

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Session 1: Invasion Biology and Lessons for Biological Control

**PHYLOGEOGRAPHY AND GENETIC STRUCTURE OF THE
INVASIVE WHEAT STEM SAWFLY, *CEPHUS CINCTUS* NORTON,
(HYMENOPTERA: CEPHIDAE) IN NORTH AMERICA: NEW
INSIGHTS INTO THE BIOLOGICAL CONTROL MANAGEMENT
OF THIS PEST**

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The wheat stem sawfly, *Cephus cinctus*, which was originally recovered from wild grasses, has become a chronic pest of wheat in the semi-arid steppe region of the North American Great Plains. Traditional pest management practices are not effective, therefore biological control options are being investigated. To provide the background necessary to manage this pest with biological control, it is important to clarify the geographical history and population structure of this species so that source areas for biological control agents can be more accurately located. In addition, such study may settle a current standing debate about the origin of *C. cinctus*: is it synonymous with an Asian species, and introduced from Northeastern Asia during historical times, or is it indigenous to North America?

Due to the lack of Asian specimens, we began our study with North American samples, using cytochrome oxidase subunit I mitochondrial DNA genealogies obtained from 57 North American populations to assess the extent of the phylogeographic structure in this morphologically monotypic insect. Mitochondrial sequences uncovered 25 haplotypes, thirteen of which were present in Montana, five in Canada (Alberta and Saskatchewan), and the remaining ones in Wyoming, Nebraska, North Dakota, and Idaho. The results showed highest nucleotide diversity in Montana and absence of structuring of mtDNA variation within the wheat continuum that is represented by these two provinces of Canada and Montana. The phylogeographic pattern observed did not reflect the population bottleneck that likely would have occurred during a single recent colonization event of new regions. However, the present analysis is providing multiple inferences on the reconstruction of the invasion routes and origin of the pest that will be discussed, and a further refinement of the biological control program proposed.

Session 1: Invasion Biology and Lessons for Biological Control

LOCAL ADAPTATION OF A SOUTH AMERICAN PARASITOID FOLLOWING ITS INTRODUCTION TO NEW ZEALAND

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Invasion biologists and classical biological control practitioners share a keen interest in the factors which regulate: (i) the establishment of non native species in new regions; and (ii) the extent of their subsequent impacts on resident communities. The potential of a species to adapt to its new environment is expected to be a key regulatory factor, and intraspecific genetic variation is therefore thought to play a major role in the outcomes of new introductions, both purposeful and unintentional. In classical biological control, efforts to maximise the probability an introduced natural enemy to adapt to its new environment are reflected in common practices such as endeavouring to obtain a large sample of the natural enemy genetic diversity present in the native range, minimising both inbreeding and selection for laboratory fit populations during culturing, and releasing as much genetic variation as possible in the new environment. Another common practice, climate matching, seeks to minimise the evolution required for an introduced species to become locally adapted by sourcing it from locations where it may already possess climatic adaptations appropriate to the intended region of introduction. However, despite widespread acceptance that populations should evolve to become locally adapted following establishment in new regions, there has been little or no supporting evidence. Here, research is described which shows that an asexually reproducing South American insect parasitoid, *Microctonus hyperodae* Loan (Hymenoptera: Braconidae), has undergone rapid, adaptive evolution since its introduction to New Zealand for biological control of an exotic pest, *Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae). Morphometric, protein and molecular analyses have all shown the *M. hyperodae* collected from South America and released in New Zealand comprised two genetically differentiated biotypes. Therefore, explicit comparisons were made between the parasitoid genetic variation which was released in New Zealand and that which survived. Of the two biotypes which were released together at each New Zealand site, one exhibited the highest fitness at nearly all release sites, with the other usually becoming rare or absent within two years of the release. There was a highly significant probability this change in the genetic composition of the released populations was due to selection. A population dynamics study at a New Zealand site where both biotypes were relatively common has provided a strong indication of why one has performed well, and the other poorly, at most New Zealand sites. This contribution therefore describes evidence of rapid, adaptive evolution following the introduction of a species to a new region, and demonstrates the importance of intraspecific variation to the outcomes of new introductions.

Session 2: Biological Control of Arthropods of Conservation Importance

PARASITES AS AN IMPORTANT FACTOR OF POPULATION DYNAMICS OF *LITCHOCOLLETIS POPULIFOLIELLA*, A SERIOUS PEST OF POPLAR SPECIES IN THE URBAN ECOSYSTEMS OF SIBERIA

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The most popular tree species of urban Siberian ecosystems is poplar. The species is highly ornamental and adaptable, promptly gains foliage, and actively absorbs air pollutants. Nevertheless, it suffers very much from a lepidopteran pest, *Litchocolletis populifoliella* Tr. whose larvae mine the leaves which promotes premature shedding.

A complex of favorable circumstances produced by urban ecosystems is the main reason for *L. populifoliella* outbreaks. The peculiarity of the pest life style (permanent residence by larvae within poplar leaves) protects larvae from adverse influences of urban environment pollution. Adults overwinter in human dwellings which guarantees high survival rates of pest populations during periods of extreme cold. More than 30 species of parasites attack the leaf-mining larvae. Hymenoptera from at least five families including Braconidae, Encyrtidae, Eulophidae, Ichneumonidae, Pteromalidae families are known from *L. populifoliella*. This parasitoid complex can not regulate pest populations in the urban ecosystems because of a population imbalance between numbers of parasites and *L. populifoliella* populations on mines.

Investigations in Krasnoyarsk, one of the biggest Siberian cities, has been shown that there is statistically significant negative correlation between density of pest mines on poplar leaves (p) and density of parasites which were found in mines (n): $\ln(n-1)=6.03-1.26*\ln(p)$, $R^2=-0.81$. This result suggests that leaves with the high densities of pest mines reduces individual risk of parasitoid attack for each larva.

Mortality rate of *L. populifoliella* larvae from parasites was seven times higher in urban area where leaf litter was saved than in areas where leaf litter was removed. The reason for this observation is that parasite pupae and adults overwinter in leaf litter, inside of *L. populifoliella* mines. Conservation of leaf litter leads to an increase in parasite survival rates and therefore decreases the probability of pest outbreaks. If this is correct, one of the main reasons of permanent outbreak of *L. populifoliella* in Siberian urban ecosystems is the tradition clearance of public garden parks of dropped poplar leaves that harbor parasitoids.

BIOLOGICAL CONTROL OF THE CITRUS LEAFMINER IN ISRAEL: SUCCESSSES AND FAILURES IN THE ESTABLISHMENT OF THE INTRODUCED PARASITIDS

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4 The citrus leafminer, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae: Phyllocnistinae) (CLM), invaded Israel in 1994, as part of a worldwide expansion of the pest from its origin in Southeast Asia. Between 1994 and 1998 we imported eight parasitoids from Thailand, Taiwan, China, Australia and South America. They included the endoparasitoid *Ageniaspis citricola* Logvinovskaya (Hymenoptera: Encyrtidae) (two strains, one from Thailand, the other from Taiwan), which oviposits in the pest's eggs and first instar larvae. Also 7 Hymenoptera: Eulophidae parasitoids; the larval ectoparasitoids *Citrostichus phyllocnistoides* (Narayanan), *Quadrastichus* sp., *Semielacher petiolatus* (Girault), the endoparasitoid *Teleopterus* sp., the pupal ectoparasitoids *Cirrospilus ingenuus* Gahan, *Galeopsomyia fausta* LaSalle and the endoparasitoid *Zaommomentedon brevipetiolatus* Kamijo. All were reared in the laboratory and released in citrus groves. Three parasitoids became established. The most common parasitoid during 1997–98 was *C. phyllocnistoides*, followed by *S. petiolatus* and *C. ingenuus*. *Ageniaspis citricola*, *Quadrastichus* sp. and *Z. brevipetiolatus* were released over a period of 4 years, with no evidence of establishment. *Teleopterus* sp. and *G. fausta* were released between 1998 and 2000; only the first was ever recovered, but without evidence of establishment. Several factors appear to have affected the successful establishment of the introduced parasitoids. These included (1) The climate in the parasitoids' country of origin - *Citrostichus phyllocnistoides* and *S. petiolatus*, which are abundant in China and Australia, respectively, became established. *Cirrospilus ingenuus*, which is abundant in Asia, does not require high relative humidity, likewise became established. However, parasitoids that are important in tropical regions and were imported from Thailand and South America, failed to establish despite continuous releases since 1994. The sensitivity of *A. citricola* to low relative humidities could be the cause for its failure to establish in Israel, and the low tolerance of *Quadrastichus* sp. to low temperatures had the same effect. (2) Competition between species - In addition to its sensitivity to low temperatures, *Quadrastichus* sp. might also have failed to establish due to being outcompeted by *S. petiolatus*. The advantages of the latter are that it attacks a wider range of the host stages, it oviposits female eggs in smaller hosts and that it has a higher

oviposition rate. (3) Facultative hyperparasitism - *Quadrastichus* sp. and *C. ingenuus* are known to be facultative hyperparasitoids, although there is no evidence that this phenomenon prevented the establishment of other parasitoids. (4) Methods and number of release sites - Releases in various climatic areas and during different seasons might have influenced the chances of successful establishment. Releasing parasitoid pupae within host-infested seedlings could improve the chances of success more than the release of adults. Is the number of released parasitoid individuals an important criterion for successful establishment in a new habitat, or are the number of releases, their timing, and the number of release sites of greater significance? These factors, based on the species' ecological and biological characteristics, which relate to our methods of rearing and releasing parasitoids, will be discussed.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

CLASSICAL BIOLOGICAL CONTROL OF THE ALEYRODIDAE (HETEROPTERA): AN ANALYSIS OF ECONOMIC AND SOCIAL IMPACT

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Numbered among the Aleyrodidae (Heteroptera) are some of the most destructive pests known, both in agriculture and in urban settings. In addition to destructive traits, members of the Aleyrodidae are among the most commonly moved pests on the globe, and invade new areas with relative ease. Classical biological control has been attempted against several of aleyrodid species. In this paper, the economic damage and impact on society of several species is reviewed, and where possible the cost or other quantification of the impact of such pests is calculated. In two recent cases of successful biological control against two different whitefly species, information on the actual cost of the biological control effort is also available. Thus, in these two cases we have quantified information on (1) the cost of the damage caused by each pest both before and (2) after the introduction of natural enemies, together with (3) the cost of the biological control program itself. In these two case studies, then, we are armed with a critical, three-pronged approach to assessing the value of a classical biological control program. The analyses presented in this paper include first an assessment of the immediate economic impact of the invasion of each pest and the subsequent impact on society, and then an evaluation of the cost of the biological control program. Having this quantitative information, a marginal rate analysis leads to an economic assessment of the cost-to-benefit ratio of the biological control program itself. The lessons learned appear to apply broadly to classical biological control, and demonstrate that classical biological control is of significant value to society, even when such quantitative assessment information is not available.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

ESTABLISHMENT IN NEW ZEALAND OF *PSEUDAPHYCUS MACULIPENNIS* (HYMENOPTERA: ENCYRTIDAE), AND ITS IMPACT ON OBSCURE MEALYBUG, *PSEUDOCOCCUS VIBURNI* (HEMIPTERA: PSEUDOCOCCIDAE)

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Pseudaphycus maculipennis is one of the few known encyrtid parasitoids of *Pseudococcus viburni*. It was imported into quarantine in New Zealand in December 1997, before new environmental legislation (the Hazardous Substances and New Organisms Act) was implemented. After host testing in quarantine, application for approval to release was made to the Environmental Risk Management Authority in April 1999. Approval to release was given on 9 August 2000, after extensive consultation, provision of further technical information, and a public hearing. The first *P. maculipennis* were released in February 2001. By 2004, approximately 765,000 parasitoids had been released to nearly 50 pipfruit orchards in Hawke's Bay, Nelson/Motueka and Auckland, and to the Wellington Botanic Gardens. Using 'trap' mealybugs around the release orchards, *P. maculipennis* was recovered from up to 85% of the properties after 3 years and had dispersed at least locally at a rate of about 200 m/year. It is concluded that the species has established in New Zealand. Preliminary studies of the impact of the parasitoid on *P. viburni* populations in pipfruit orchards are described.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

BIOLOGICAL CONTROL OF THE PINK HIBISCUS MEALYBUG IN BAHIA DE BANDERAS, NAYARIT, MEXICO

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A combination of classical and augmentative biological control program was established in the valley of Bahía de Banderas, Nayarit, Mexico, to control the recent invasive pest the Pink Hibiscus Mealybug (PHM) *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae) through the introduction of three biological control agents, predator *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae), and parasitoids *Anagyrus kamali* Moursi and *Gyranusoidea indica* Shafee, Alam & Agarwal (Hymenoptera: Encyrtidae). PHM was detected in Bahía de Banderas in February 2004. Afterwards an Integrated Pest Management program was implemented by the Federal Government to contain the spread and development of this pest into commercial agriculture, urban and forest areas, including physical, cultural, chemical, legal and biological control. *C. montrouzieri* were introduced from Canada and California, USA commercial insectaries, while parasitic wasps were obtained from Belize and Puerto Rico as a cooperative program sponsored by a Central America agency OIRSA. The effectiveness of *C. montrouzieri* to control PHM and the establishment of *A. kamali* on teak were evaluated. Although, currently a total of 672,900 predators and 155,925 *A. kamali* have been released over an area of 250 hectares on this valley, this evaluation was carried out only on three commercial plantations: guava, soursop and teak. On the guava plantation (2 hectares) 2,500 and 9,500 predators were released on June and July 2004, respectively. On the soursop plantation (5 hectares) 4,500 and 5,000 predators were released on June and July 2004, respectively; while on the teak plantation (2 hectares) 4,000 and 1,000 predators were released on August and September 2004, respectively. No individuals of *A. kamali* were released at the teak plantation. Samples were taken in weekly bases from 50 trees on each plantation.

Guava. On this crop the starting PHM density recorded was 27.9 individuals/ vegetative shoot, then on August, 77 days after the first predator release the PHM density decrease drastically where the predator population density reach its maximum activity 4.3 individuals/ vegetative shoot, this resulted in a maximum decrease of 99% on the PHM density.

Soursop. On this plantation PHM density before predator release was 29.2 individuals/ vegetative shoot. Seventeen days after predator release PHM density decrease rapidly in a 78%. The maximum reduction on PHM density was 3.6 individuals/ shoot meaning an 87.6%, at this point there were 0.16 predators/ shoot.

Teak. On this plantation PHM presented the highest population densities with 119.5 individuals/ shoot. As a result of the predator activity, PHM start to decrease, up to 88.9% (13.3 individuals/ shoot) 48 days after predator first release, at this time predator density was 3.4 individuals/ shoot. The maximum reduction on PHM density 96.4% was detected 69 days after predator first release with a predator density of 0.14 individuals/ shoot. On this crop it was possible to detect *A. kamali* establishment, nevertheless this parasitic wasp was not released on this plantation but around 1 mile away on other commercial orchards and released six months before on the valley. At this time a density of 1.8 mummies/ shoot and 4.88 PHM/ shoot were detected.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

ESTABLISHMENT OF *APROSTOCETUS VAQUITARUM* AND *QUADRATICUS HAITIENSIS* (HYMENOPTERA: EULOPHIDAE) IN THE U.S.A. AS MORTALITY FACTORS OF THE DIAPREPES ROOT WEEVIL (COLEOPTERA: CURCULIONIDAE)

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The weevil, *Diaprepes abbreviatus* L. (Coleoptera: Curculionidae) invaded Florida during the 70's becoming one of the most important agricultural pests of the state. Classical biological control efforts were initiated but were not successful. During 1997 a new biocontrol program was initiated. Thirteen species of egg parasitoids were introduced and tested under quarantine conditions. Among these parasitoids, the egg parasitoids *Aprostocetus vaquitarum* (Wolcott) and *Quadrastichus haitiensis* Gahan (Hymenoptera: Eulophidae) were introduced

from the Caribbean Region between 1999 and 2000. Both parasitoid species were released in different areas of Florida and their recovery assessed during 3 years. Both parasitoids have become established in the southern portion of the Florida peninsula [tropical climate, winter temperatures not lower than 18°C, constant host densities), but have failed to establish in areas with sporadic host densities and winter temperatures lower than 15°C. In the area where the ectoparasitoid, *A. vaquitarum* is established, it appears to be more prevalent than *Q. haitiensis* causing 60-100% mortality to *Diaprepes abbreviatus* eggs. Parasitoid dispersion has been observed within a 4-mile ratio from the nearest dispersion site with host mortality levels between 70-90%. It is not known if the parasitoid would be able to colonize areas of the state north of the southern tip of the peninsula.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

BIOLOGICAL CONTROL OF THE REDGUM LERP PSYLLID IN MEXICO

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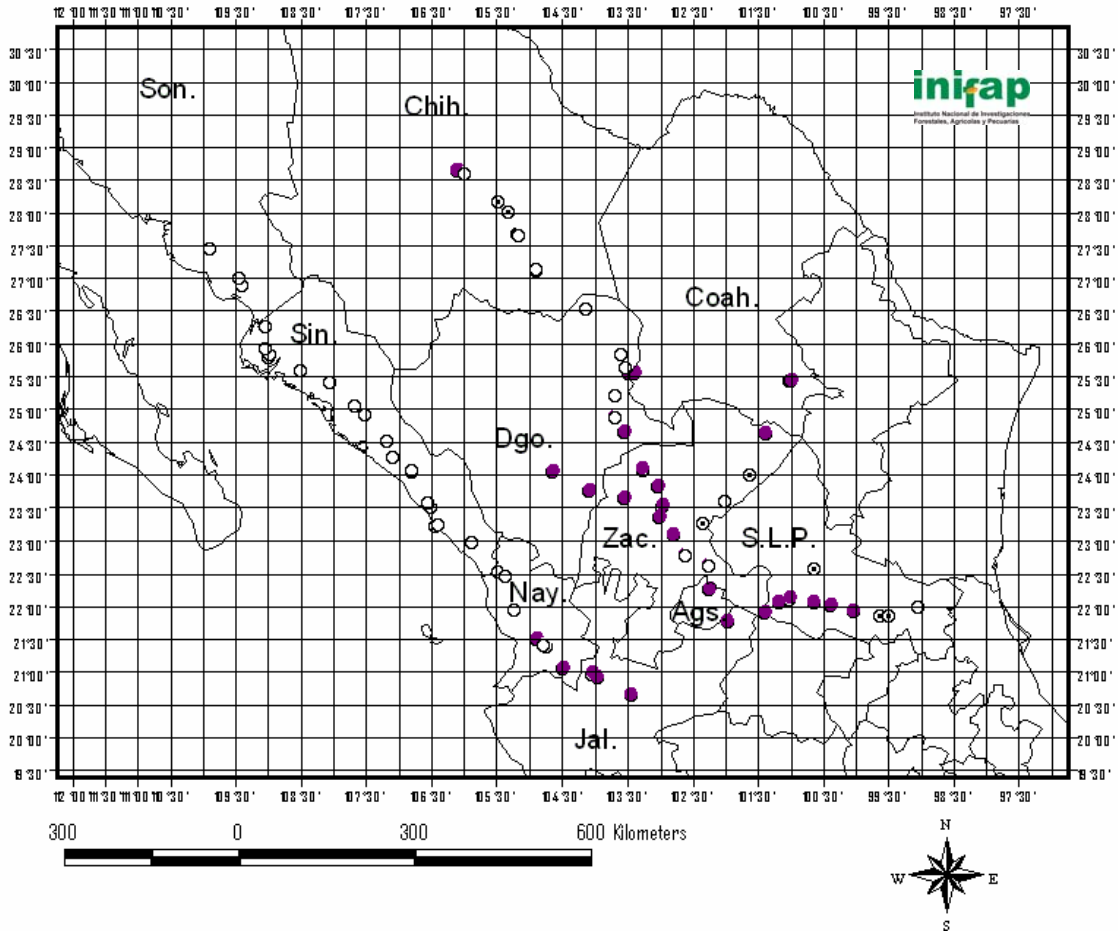
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Since the beginning of the 20th century, *Eucalyptus camaldulensis* Den. (Myrtales: Myrtaceae) and a few other *Eucalyptus* species have been widely planted in Mexico. For a long time *E. camaldulensis* was a pest free tree. However, in year 2000 the redgum lerp psyllid (*Glycaspis brimblecombei* Moore) (Homoptera: Spondyliaspidae) was detected. In 2001, this psyllid became a nationwide problem. On January of 2002, a quarantine program was implemented on 21 states, and sanitary actions were mandated by law.

Two main approaches were taken for direct control of the red gum lerp psyllid:

- 1) **Biological control with local natural enemies.** Several predator insects were evaluated: *Harmonia axyridis* Pallas, *Olla v-nigrum* Mulsant (Coleoptera: Coccinellidae) *Geocoris punctipes* Say (Hemiptera: Lygaeidae) and *Chrysoperla* spp. (Neuroptera: Chrysopidae), were tested in Guadalajara, Jalisco and Pabellon de Arteaga, Aguascalientes. Additionally, the control effect of the ant *Anoplolepis longipes* (Jerd.) (Hymenoptera: Formicidae) was evaluated in Guadalajara. Only *A. longipes* appeared to exert some control. For the other species, results were equivocal.
- 2) **Classical biological control.** This strategy was started by a forestry agency in Guadalajara (FIPRODEFO), in collaboration with D. L. Dahlsten (University of California) and the National Secretariat of Environment and Natural Resources. The parasitoid *Psyllaephagus bliteus* Riek (Hymenoptera: Encyrtidae) was brought from California (USA) (previously imported from Australia) to Guadalajara in May of 2001. At that time, 75 couples were released on a recreational park. The parasitoid was reared at FIPRODEFO facilities and released at the same park in October of 2001, and later on nearby areas. Insect dispersion was monitored with yellow sticky traps. Parasitoid catches were difficult to observe at first. However, from May to July of 2002, a high percentage of parasitism by *P. bliteus* was found on tree foliage by personnel of the National Research Institute on Forestry, Agriculture and Livestock (INIFAP), at 32 locations, ca. 250 km apart from the original release point. The parasitoid was also collected at other locations more than 500 km apart from the original release point. Because of the high parasitism observed at very distant points, soon after the first releases, the senior author hypothesized that *P. bliteus* could have entered into Mexico together with the psyllid, or spread down from early releases in California (USA). However, regardless of the origin, this parasitoid is now well established in Mexico. A regional survey made by INIFAP during 2004, reports *P. bliteus* in nearly all places where the psyllid is present. Infestation levels were very low in 2004 in comparison with 2002. As by October of 2004, the psyllid was completely absent in two formerly infested coastal states and partially absent in two other states. Other factors appear to be affecting *G. brimblecombei* as well; however, *P. bliteus* is a specific parasitoid and it has found in Mexico suitable environments for its establishment.



- Both, *Glycaspis brimblecombei* and *Psyllaephagus bliteus* present.
- ◐ *Glycaspis brimblecombei* present and *P. bliteus* absent.
- *Glycaspis brimblecombei* absent.

Current distribution of *Glycaspis brimblecombei* and *Psyllaephagus bliteus* within 10 estates of Mexico. Each circle indicates the locations where insects were sampled. (INIFAP-CONAFOR-CONACYT 2004).

UPDATES ON THE BIOLOGICAL CONTROL OF THE CALIFORNIA RED SCALE IN SICILY

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The California Red Scale [*Aonidiella aurantii* (Maskell) (Homoptera: Diaspididae)] is considered to be one of the most injurious pests to citrus groves in the Mediterranean basin. Continuous investigations on the population dynamics of the scale as well as on its main mortality factors have been carried out.

The natural enemies complex is constituted by the parasitoids *Aphytis* spp. and *Encarsia perniciosi* (Tower) (Hymenoptera: Aphelinidae) and by the predators *Chilocorus bipustulatus* (L.), *Rhyzobius lophantae* Blaisdell (Coleoptera: Coccinellidae), *Cybocephalus rufifrons* Retter (Coleoptera: Cybocephalidae) and *Lestodiplosis aonidiellae* Harris (Diptera: Cecidomyiidae). The following species of *Aphytis* are recorded in Sicily: *chilensis* Howard, *chrysomphali* (Mercet), *lignanensis* Compere, *maculicornis* (Masi), *melinus* DeBach and *proclia* (Walker). After the introduction of *A. melinus* in Sicily during the 60's, this parasitoid and *A. chrysomphali* were the predominant species in Sicilian citrus groves, but following studies have shown the substitution of *A. chrysomphali* with *A. melinus*, which was proven to be well adapted and spread out in the main citrus growing areas in Southern Italy. In 1988-90 the parasitoid *Comperiella bifasciata* Howard (Hymenoptera: Encyrtidae) was introduced and since then it has been only occasionally recovered. The results of a field survey on the presence and diffusion of *A. chrysomphali* and *C. bifasciata* in Sicily are presented.

***Aphytis chrysomphali*.** The observations were conducted on orange (cv. “Tarocco”, “Valencia Late” and “Washington Navel”) and lemon (cv. “Femminello”) in Eastern Sicily during 2003. Infested fruits, leaves and twigs of each variety were monthly collected and 500 specimens of California Red Scale per sample were observed and the *Aphytis* spp. pupae found were identified. The data collected showed that in the investigated areas, the species *A. melinus* and *A. chrysomphali* were present; the first one was predominant while the second species was occasionally recovered on colonies of California Red Scale infesting lemon trees. Furthermore, the two species showed a seasonal alternation: *A. chrysomphali* was found especially during wintertime, while *A. melinus* mainly in warmer periods.

***Comperiella bifasciata*.** The aim of the survey, which started in 2003 and is still continuing, was to confirm the establishment of the encyrtid and to draw a map of its diffusion, 15 years after its first introduction. Infested fruits (20) and twigs (4 meters, 1-2 years old) were col-

lected monthly in 10 groves in South Eastern Sicily (Siracusa province). Half of the sample was observed and the parasitized instars were isolated and reared until the adult parasitoids emerged. The remaining 50% was kept into emergence boxes and the emerging parasitoids were collected and identified. The presence of the parasitoid was also monitored using pheromone traps for the California Red Scale in different citrus groves. The data collected showed that the encyrtid is well adapted and has colonized a wide area, 50km, on average, far away from the first introduction site. The survey will be continued and expanded in the next years in order to acquire quantitative data on the parasitic activity of the encyrtid.

Session 3: Recent Successes of Classical Biological Control: An Impact Analysis

**RELEASES, RECOVERY, AND DISSEMINATION IN
NORTH AMERICA OF *LATHROLESTES ENSATOR*,
A BRACONID ENDOPARASITOID OF THE APPLE SAWFLY,
*HOPLOCAMPA TESTUDINEA***

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Introduced accidentally in New York State in 1939, the apple sawfly, *Hoplocampa testudinea* Klug (Hymenoptera: Tenthredinidae), gradually invaded apple orchards of Northeastern North America. Native from Europe *Lathrolestes ensator* Brauns is a braconid endoparasitoid that has only one host: the apple sawfly. As the apple sawfly has one host plant, the apple tree, and as no soft methods or natural enemies were known in North America, the apple sawfly was a

prime target for a biological control program, although presently no rearing methods are known. Collected in various localities in Western Europe, *L. ensator* has been shipped as parasitized cocoons to be released annually, after a quarantine check, in an insecticide-free apple orchard at Frelighsburgh, Qc, Canada, from 1995 to 1999. Collections in 1999 demonstrated that *L. ensator* had successfully established in North America. Further collections in 2000 demonstrated that the braconid parasite had extended his local distribution. In 2002 parasitized cocoons were collected in Frelighsburgh and released in a non-treated orchard of New Hampshire. In 2003 and 2004 parasitised cocoons were collected and released in two organic orchards in Quebec, one non-treated orchard in Ontario and one non-treated orchard of New Hampshire. Dissection of cocoons determined that parasitism rates in Frelighsburgh were variable, and as high as 69.7%. At such rates, apple sawfly populations would be under control. Recovery studies are planned for May 2006.

Session 4: Cultural Manipulations to Enhance Biological Control

ATTRACTIVENESS OF FLOWERING PLANTS TO APHIDOPHAGOUS HOVERFLIES: SUITABILITY AS INSECTARY PLANTS TO ENHANCE BIOLOGICAL CONTROL

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Insectary plants are increasingly being used in Conservation Biological Control to enhance predators in crops. In this study, relative attractiveness of twenty four plant species to aphidophagous hoverflies has been evaluated in an experimental field. Plant species were selected according to published results of previously examined plants and flowering time. Attractiveness was assessed by conducting timed observations of visit frequencies and also recording the observed behaviour in each plot. Plants were also inspected for aphids and syrphid larvae.

SELECTIVE RESOURCE SUBSIDIES AND BIOLOGICAL CONTROL

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Biological control as an ecosystem service is one of the most sustainable strategies to reduce reliance on pesticide use. In particular, conservation biological control is an important component. It involves the provision of resource subsidies, such as flowers, which can provide nectar, pollen and shelter which many natural enemies, such as parasitoids and predators, need. As a consequence, the local abundance and fitness of natural enemies may be enhanced, leading to the improvement of “top-down” control of pest populations. However, a potential dilemma arises when pests, natural enemies and antagonists of the third trophic level forage on the provided floral resources. Therefore, the provision of selective resource subsidies may have major consequences for the population dynamics of pest/natural-enemy systems. There is a hierarchy of levels at which flowers can be deployed selectively to favour natural enemies. Availability of resource subsidies only to a natural enemy may be achieved via: (1) the morphology of the flowers, (2) the quality of nectar and pollen, (3) floral attractiveness and (4) the morphology of insects targeted. A relative advantage to the natural enemy may further be provided by: (5) a greater fitness improvement of the natural enemy compared with the pest and (6) the natural enemy benefits from a prey/host of improved quality more than does the prey/host itself. Also, the effectiveness of a natural enemy may be enhanced by (7) changes in its sex ratio and (8) a relatively greater enhancement of its fitness than that of its fourth-trophic-level antagonists. Resource subsidies may have additional selective effects (9) within a natural enemy guild, (10) within a natural enemy species and (11) between fitness traits of a natural enemy.

In future, conservation biological control measures should consider this hierarchy of ways in which the provision of resource subsidies might affect the population dynamics of pest/natural-enemy systems. Biological-control practitioners need to be aware of the complexity of this hierarchy because the selectivity of resource subsidies may act across four trophic levels. Therefore, only rigorous laboratory and field experiments can assess the outcome of a conservation biological control measure which aims to favour natural enemies more than pests. Moreover, the concept of a hierarchy of selectivity levels can be generalised to

other components of conservation biological control. These include, for example, relative differences in the use made of shelter by pests and their natural enemies and the effects of reduced-rate insecticide applications. In consequence, biological control approaches that incorporate concepts of “selectivity hierarchies” are likely to become more important as “ecosystem engineering” to enhance biological control becomes more widely practised.

Session 4: Cultural Manipulations to Enhance Biological Control

A NEW CULTURAL CONTROL PRACTICE TO ENHANCE BIOLOGICAL CONTROL OF THE CAROB MOTH BY OPPORTUNISTIC PREDATORS IN CALIFORNIA ‘DEGLET NOOR’ DATE GARDENS

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‘Deglet Noor’ dates, *Phoenix dactylifera* L. (Arecales: Arecaceae), undergo a natural fruit abscission during the summer in California, U.S.A. date gardens. Many of the abscised dates become lodged in the date bunch and become heavily infested by the carob moth, *Ectomyelois ceratoniae* (Zeller) (Lepidoptera: Pyralidae). Carob moth mortality due to natural enemies is minimal when abscised infested fruit remain in the tree. We found that shaking abscised fruit to the ground significantly increased carob moth mortality. Mortality in these dropped fruit was attributed to predation by two native ant species, the desert fire ant, *Solenopsis aurea* Wheeler (Hymenoptera: Formicidae), and the California harvester ant, *Pogonomyrmex californicus* (Buckley) (Hymenoptera: Formicidae), in concert with extreme summer ground temperatures. Infested dates on the ground, rapidly increased in temperature, which resulted in larvae either exiting the fruit (exposing them to ants) or dying within the fruit. Shaking abscised dates from bunches in the tree is a cultural control strategy which augments the effectiveness of naturally occurring opportunistic predators by increasing the amount temporal and spatial overlap with carob moth larvae.

USE OF SOWN WILDFLOWER STRIPS TO ENHANCE THE PARASITISM OF LEPIDOPTERAN PESTS IN CABBAGE CROPS

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Many agroecosystems are unfavourable environments for natural enemies due to high levels of disturbance. Fragmentation and loss of suitable habitats has caused natural enemies to decline in species diversity and abundance, and has even resulted in extinctions and loss of biocontrol function. Therefore a diversification through habitat manipulation is essential to create a suitable ecological infrastructure within the agricultural landscape to resources such as food for adult natural enemies, alternative prey or hosts, and shelter from adverse conditions. The aim of this study was to investigate, if the use of wildflower strip is an appropriate tool to reduce key lepidopteran pests in cabbage crops?

We have investigated the parasitism rates of cabbage lepidopteran pests in relation to presence or absence of sown, species rich wildflower strips, on four farms at different sites in arable landscapes of Western-Switzerland.

On one hand naturally occurring caterpillars and eggs of *Pieris rapae* L. (Lepidoptera: Pieridae), *Plutella xylostella* L. (Lepidoptera: Plutellidae) (only larvae) and *Mamestra brassicae* L. (Lepidoptera: Noctuidae) have been analysed as well as exposed batches of reared eggs of the two last mentioned pests in unsprayed cabbage crops. Three treatments have been tested within the naturally occurring pests: (1) Cabbage field directly adjacent to a wildflower strip, (2) cabbage near to a wildflower strip (at least 15 m), and (3) control cabbage field without wildflower strip or any other semi-natural habitats nearby. Using batches of reared eggs the spatial pattern of parasitism in relation to the strip was analysed in a grid of 3m to 3m (288 resp. 384 points/plants). On two sites two blocks with a adjacent strip and a control plot without any strip were investigated.

Microplitis mediator Haliday (Hymenoptera: Braconidae), *Cotesia rubecula* (Marshall) and *Diadegma semiclausum* (Hellén) (Hymenoptera: Ichneumonidae) were the most abundant larval parasitoids of totally twenty found species (incl. two hyperparasitoids) and egg parasitoids *Telonomus* sp. (Hymenoptera: Scelionidae) and *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae). Caterpillars of *M. brassicae* and *P. rapae* were parasitised at a higher rate in cabbage fields near to the wildflower strip. In contrast, caterpillars of *P. xylostella* were significantly more parasitised in the field without strip. Parasitism rates of *M. brassicae* eggs nearby the wildflower strip were higher, with a range of 17 % to 63 %, than in both isolated fields with 0 %. Finally, we got contrasting results with the grid trial showing no clear relation to the distance to the strip.

Session 4: Cultural Manipulations to Enhance Biological Control

HOW DO PLANT AND HOST TRAITS INFLUENCE THE REPRODUCTIVE SUCCESS OF A PARASITOID?

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Legume seeds provide one of the primary sources of dietary protein particularly in developing countries and control of post harvest loss of legume seeds is a serious problem worldwide. Stored legumes are endangered by bruchids (Coleoptera) throughout the tropical belt, with the Common Bean Weevil, *Acanthoscelides obtectus* Say and the Mexican Bean Weevil, *Zabrotes subfasciatus* (Bohemann) (Coleoptera: Chrysomelidae), being the major storage pests of the common bean (*Phaseolus vulgaris* L. (Rosales: Fabaceae)), and *Callosobruchus* sp. (Coleoptera: Bruchidae) of cowpeas (*Vigna unguiculata* Walp (Rosales: Fabaceae)).

18 Limiting bruchid damage is being approached using plant breeding in combination with biological control. For example, recent studies indicate that the combination of certain bean characteristics and biological control by the parasitic wasp *Dinarmus basalis* (Rondani) (Hymenoptera: Pteromalidae) is a promising integrated approach to control *A. obtectus*. Since resistance of bean cultivars to bruchid attack can either be based on physical, or chemical bean parameters, both of which may interfere with either bruchid larval penetration or development, any altered plant characteristics may also influence the performance of parasitoids. On the other hand different host traits such as oviposition behaviour of the bruchids, larval development can also affect the reproductive success of a parasitoid. A parasitoid with a relatively narrow host spectrum confined to bruchids such as *D. basalis* can exploit a variety of different bruchid hosts, the efficiency, however, depends on not only the suitability of the host but also the host plant.

Using the tritrophic system of bean, bruchid and parasitoid, we investigate which plant and host traits influence the reproductive success of the larval ectoparasitoid *Dinarmus basalis* in regard to its biological control potential. The oviposition behaviour and success of *D. basalis* was studied on different legume seeds infested with two bruchid hosts: *Acanthoscelides obtectus* and *Callosobruchus maculatus* (Fabricius). These bruchids display different oviposition behaviour – *A. obtectus* distributes its eggs loosely among the seeds with the first instar

larvae actively infesting a seed, whereas *C. maculatus* glues its eggs on the surface of the seed with the hatching L1 penetrating the seed directly from the egg. To date results indicate that while choice of host does not negatively influence the parasitism success. The characteristics of the legume seed strongly influences parasitism. The implications for biological control approaches are discussed.

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USING BUCKWHEAT TO INCREASE PARASITISM RATES OF A TORTRICID PEST IN A NEW ZEALAND VINEYARD

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In this study, the effects of floral resources on parasitism rates of the lightbrown apple moth, *Epiphyas postvittana* Walker (Lepidoptera: Tortricidae) by one of its most important larval parasitoids, *Dolichogenidea tasmanica* (Cameron) (Hymenoptera: Braconidae) was examined in an organic vineyard in Marlborough, New Zealand. Potted grapevines (cv. Sauvignon Blanc) were seeded with either 20 or 100 first-instar larvae of the lightbrown apple moth. Vines were then placed in vineyard blocks planted with buckwheat, *Fagopyrum esculentum* Moench (cv. Katowase) or with no flowering plants (control) five times from January to April to compare parasitism rates by *D. tasmanica* in the two treatments and to examine the effect of host density on parasitism rates. The potted vines were collected two weeks after placement and the *E. postvittana* larvae were removed and reared to either adult moths or parasitoids. Parasitoids were identified and proportional parasitism was compared between the buckwheat and control treatments. When all of the dates were pooled, results indicated that parasitism rates by *D. tasmanica* were significantly greater in the buckwheat blocks compared with the controls. Parasitism rates by *D. tasmanica* were greater on the vines seeded with 100 larvae compared with vines seeded with 20 larvae; however this result was not statistically significant. These results support previous work, which showed that parasitism rates of *E. postvittana* could be increased by the use of floral resource subsidies. The results of this study and implications of these results on the biological control of the lightbrown apple moth and other pests are discussed.

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EFFECT OF A SORGHUM TRAP CROP ON THE SOUTHERN GREEN STINK BUG, *NEZARA VIRIDULA*, AND ITS PARASITOID, *TRICHOPODA PENNIPES*, IN COTTON

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Phenology of stink bugs is closely bound to the pattern of crop phenology and seasonal succession of host plants. In the southeastern states, a farmscape can consist of wooded areas, weedy field edges, grassy meadows, and a variety of agricultural crops including corn (*Zea mays* L. (Cyperales: Poaceae)), peanuts (*Arachis hypogaea* L. (Rosales: Fabaceae)), and cotton *Gossypium hirsutum* L. (Malvales: Malvaceae). In this farmscape, the southern green stink bug (SGSB), *Nezara viridula* (Linnaeus) (Heteroptera: Pentatomidae), can disperse from alternate host plants into cotton at the vulnerable fruiting stage. Addition of an attractive trap crop at the right time and place in this farmscape could lead to a concentration of the SGSB in this crop, protecting cotton. The problem that natural enemies of these pests can have in shifting their habitats to follow the seasonal movement of their hosts could be mitigated by incorporating a trap crop to provide the basic resources, including hosts, of the natural enemies in relatively close temporal and spatial association within the farmscape. Therefore, the ability of sorghum (*Sorghum bicolor* (L.) Moench (Cyperales: Poaceae)) to serve as a trap crop for the SGSB and enhance the tachinid *Trichopoda pennipes* (Fabricius) (Diptera: Tachinidae), an important parasitoid of the SGSB, in cotton was investigated in 2002 and 2003. Sorghum trap crops and equally sized cotton trap crops were planted in a strip between a corn and cotton field. Field cotton was divided into field cotton plots associated with sorghum trap crop plots and field cotton plots associated with cotton trap crop plots. For both years, a greater number of SGSB adults were present on sorghum panicles (2.25 and 1.22 per 0.91-m sample in 2002 and 2003, respectively) than on cotton plants in trap crop cotton plots (0.05 and 0.11 per 0.91-m sample in 2002 and 2003, respectively) and field cotton plots (0.004 and 0.025 per 0.91-m sample in 2002 and 2003, respectively) associated with the trap crop plots. In sorghum trap crops, mean parasitism of SGSB adults by *T. pennipes* females was moderately high, 57.1% and 52.8% in 2002 and 2003, respectively. Parasitization of SGSB adults was highly correlated to the density of SGSB adults in sorghum suggesting that aggregation of SGSB adults in sorghum trap crops resulted in high parasitism by *T. pennipes* females. Overall, mean percentage parasitization of SGSB adults by *T. pennipes* was higher on sorghum panicles than on field cotton (13.0% and 15.6% in 2002 and 2003, respectively). However, on dates when SGSB adults were present in substantial numbers in field cotton, *T. pennipes* females were present in field cotton and parasitized these pests at levels similar to those for sorghum indicating that the trap crops were sources, not sinks, for the parasitoid.

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BOOSTING BIOLOGICAL CONTROL: DO WE USE A 'BOUQUET' OR A 'KISS' APPROACH TO WIN A NATURAL ENEMY'S HEART?

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The dependence of many natural enemies on nectar and/or pollen raises the opportunity for enhancing biological control via the provision of flowers around or across fields. A 'Keep It Simple, Stupid' ('KISS') approach is a contentious idea when applied to the provision of flowers in this way. Some biological control practitioners recommend that land managers use a single species of flowering plant (i.e., the 'KISS' approach) to conserve resident natural enemies of crop pests. Proponents of conservation biological control argue that species-rich patches of flowers (i.e., a 'bouquet') support a high arthropod biodiversity, which improves functional diversity and therefore herbivore suppression. The use of plant mixtures (i.e., the 'bouquet' approach) may result in improved pest suppression because plant mixtures may support an assemblage of natural enemy species, each differentially benefiting from the plant species present in the complex mixture. The complex flower mixture may extend the supply of floral resources through time, both within a day and over the growing season. Preliminary laboratory results from a 'bouquet' approach indicated that the lifespan of parasitoid wasps with access to a combination of two flowering plants may be higher than when provided with each plant species alone.

Laboratory and field experiments are being used to compare the 'bouquet' and 'KISS' approaches for boosting biological control. In the laboratory, single and mixtures of flowering plant species are ranked for their effects on the lifespan of adult hoverflies (Diptera: Syrphidae) and parasitoid wasps (Hymenoptera: Braconidae). In replicated field experiments, the role of single plant species and simple or complex plant mixtures are being assessed when planted adjacent to broccoli or lucerne crops in Canterbury, New Zealand. Assessments of the density and impact of pest and natural enemy populations are being conducted in the flowering patches and the adjacent crop during the growing season. It is predicted that the density of natural enemy populations will be higher, and conversely pest populations lower, in the crop with a patch of the complex plant mixture. The results gained will allow recommendations to be made on optimal habitat management strategies available to land managers.

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Session 4: Cultural Manipulations to Enhance Biological Control

FUNCTIONAL BIODIVERSITY FOR SUSTAINABLE AGRICULTURE

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Establishing flowering field margins is often propagated as tool to support predators and parasitoids and thereby enhance the efficacy of biological control of pest arthropod. When deciding on the composition of these field margins, usually only the requirements of beneficial insects are taken into account, while the risk of unintentionally promoting pest insects is often neglected.

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In order to make strategic use of botanical biodiversity to boost biological control, we aim at identifying flower species that selectively fulfil the needs of beneficial insects without supporting pests. For two cabbage pests and their parasitoids we investigated the following parameters:

Plant attractiveness. We used direct observations and sweep-netting on standardized flower plots to study differences in herbivore flower visitation. The cabbage moth *P. xylostella* (Lepidoptera, Plutellidae) was found on nearly all flowers, while the cabbage white *Pieris* spp (Lepidoptera: Pieridae) was restricted to two flower species with more hidden nectar.

Accessibility and suitability of nectar in different plant species. Nectar accessibility was tested by weighing food deprived individuals before and after exposure to nectar sources. Nutritional suitability of nectar was tested in longevity experiments. Flowers differed with respect to their impact on longevity of the cabbage pests and their parasitoids.

Gustatory and longevity response to nectar sugars. We measured the gustatory response of starved individuals to a range of nectar sugars (0.5M) and established their longevity when feeding on these sugars. Overall, the effect of sugar supply had a much greater effect on the parasitoid *Diadegma semiclausum* (Hellén) (Hymenoptera: Ichneumonidae) as compared to the cabbage moth *Plutella xylostella* (L.).

Impact of nectar plants on biological control under (semi-) field conditions. Under semi-field conditions parasitoids with access to nectar lived 20 times as long as individuals

without access to nectar and parasitized 60 times more *P. xylostella* larvae. Under field conditions we found parasitism rates of 80% (2002) and 65-70 % (2003) irrespective of the field margin treatment (flower/control).

Variation in response to nectar sugars and differences in flower suitability among pests and their parasitoids show the potential for application of selective food sources. Results from (semi-) field experiments demonstrate the great potential of using specific nectar plants to boost parasitoids as biocontrol agents. Fine-tuning the composition of flowering field margins with respect to key pests and their natural enemies can optimize the impact of functional biodiversity on sustainable biological pest control.

Session 4: Cultural Manipulations to Enhance Biological Control

RESPONSES OF APHID NATURAL ENEMIES TO THE SPATIAL CONFIGURATION OF EXPERIMENTAL ALFALFA MICRO-LANDSCAPES

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The spatial configuration of agricultural landscapes may influence the arrival and establishment of natural enemies to prey patches. Habitat subdivision and isolation should generally negatively affect natural enemies more than their phytophagous prey, with a resulting increase in pest populations. Here we report the abundance of aphid natural enemies (ladybirds, carabids and parasitoids) and the percentage of aphid parasitism in experimental alfalfa micro-landscapes that varied in the level of fragmentation (0, 4 or 16 fragments) and/or habitat loss (0, 55% or 84% of habitat loss). In August 2003 we sowed 20 alfalfa landscapes (30 x 30m), and in December we removed vegetation from 15 of them. As a result, we ended up with four kinds of landscapes, replicated five times each: i) unfragmented control landscape, ii) 4fragments-55% loss, iii) 4 fragments -84% loss, and iv) 16 fragments -84% loss. In the fragmented landscapes, the matrix was composed of bare ground and the distance between fragments was 6 m.

Natural enemy abundance. Contrary to what we expected, two of the five more frequent species of ladybirds were more abundant in the fragmented landscapes, with the other three showing no difference between landscapes. Carabids were more abundant in the more fragmented landscapes (16 vs. 4 fragments). Parasitoids and aphid parasitism, in general did not differ between landscapes, although occasionally parasitism increased in the more fragmented landscapes (16 vs. 4 fragments).

Aphid abundance. Aphid abundance was similar in all landscapes, but they were more abundant at the fragment edges of the more fragmented alfalfa landscapes.

The higher perimeter-to-area ratio in more fragmented landscapes and the lower amount of remaining habitat may explain why natural enemies are more concentrated in those fragments.

Session 5: Contribution of Biological Control to the Global Agenda

MASS-REARING, RELEASES AND ESTABLISHMENT OF *PHYMASTICHUS COFFEA* LASALLE FOR THE CONTROL OF THE COFFEE BERRY BORER *HYPOTHENEMUS HAMPEI* (FERRARI) IN COLOMBIA

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The coffee berry borer (CBB) is widely distributed in all coffee-growing regions of Colombia and is considered to be the country's number one coffee insect-pest. CBB causes serious economic losses and directly affects the economy of more than half a million families in Colombia. As an alternative to the use of highly toxic synthetic insecticides to control CBB in Colombia, *Phymastichus coffea* LaSalle (Hymenoptera: Eulophidae), a gregarious endoparasitoid of CBB females, was introduced from Africa to Colombia in 1996. Since

then, *P. coffea* has been mass-reared at the Centro Nacional de Investigaciones de Café Cenicafé, in Chinchiná, Colombia. *P. coffea* was first released in Colombia in 1997 with promising results. Thereafter, field studies conducted in 2001 confirmed this parasitoid as a successful candidate for biological control of CBB. The extent of the parasitism was significantly affected by the developmental stage of the coffee berries, as well as by the position of the insect pest inside the berries at the time of the various releases of the parasitoid. Levels of parasitism of up to 85% were recorded when adults of CBB were confined in sleeve cages in coffee branches to infest 150-day old coffee berries. *P. coffea* preferred to parasitize CBB adults that were beginning to bore into the coffee berries prior to damaging the endosperm. The establishment of *P. coffea* in the coffee-growing region of Colombia is being evaluated. Initially, about 641,000 adults of *P. coffea* were released in 44 small farms in the Colombian Central Coffee area, and one year later they were recovered in 84% of these farms. Levels of parasitism varied from 4 to 28% in coffee fields that maintained low levels of coffee berry borer infestations (0.1 to 9.2%). However, a preliminary examination in two plots after 3 years following *P. coffea* releases has not documented the presence of this parasitoid in the field. A permanent establishment of *P. coffea* in the field is rather difficult because of (i) the short lifespan of the parasitoid females, and (ii) the limited temporal availability of hosts, as *P. coffea* mostly parasitizes CBB females that have not reached the endosperm of the coffee berries. Hence, inundate releases of *P. coffea* in coffee plantations are proposed for biological control of CBB in Colombia, particularly as a new automated mass-rearing technique for both hosts and parasitoids, using artificial diets, is under development. This will reduce costs and increase the efficiency of the mass-rearing of *P. coffea*.

Session 5: Contribution of Biological Control to the Global Agenda

OLIVE FLY AND NATIVE NATURAL ENEMY POPULATION DYNAMICS IN INDIGENOUS AND COMMERCIAL OLIVE HABITATS

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26 Olive fly, *Bactrocera oleae* (Gmelin) (Diptera: Tephritidae), is a key pest in olive-production worldwide. Specimens of olive fly natural enemies collected in the South Western Cape Province in South Africa showed promise as bio-control agents for olive fly control in areas other than South Africa. In order to determine the importance of some of the major parasitoid species found during this initial exploration, a year-long population dynamic study was conducted on olive fly as well as its natural enemy populations in indigenous wild olive stands, *Olea europaea* L. subsp. *africana* (Miller) (Oleaceae) as well as orchards of various commercial cultivars. The habitats selected for this study were; wild olive stands and commercially cultivated olive orchards in close proximity to the wild olive stand as well as two conservation area stands of wild olives, one in a mountainous area and another along a river. The first two study areas were regularly sprayed with full-cover insecticide sprays and the cultivated orchards further had human intervention such as pruning, fertilization, weeding and irrigation. The third and fourth areas were subjected to limited human intervention during the past 50 years.

As expected, bi-monthly samples of olives and olive fruit fly trap counts from each of these areas indicated the highest incidence of both olive fly and natural enemy species in the third and fourth areas, followed by the first and then second areas. The dominant parasitoid reared from field collected olive samples in all of the areas was a *Psytalia* sp. (Hymenoptera: Braconidae). The highest percent parasitism in summer months was found in the areas where human intervention was limited, followed by the cultivated areas. Early indications show that percent parasitism in wild olive stands remained relatively high through colder winter and spring periods in the undisturbed areas compared to areas which were more disturbed. The season-long availability of fruit in wild olive stands is believed to greatly influence the incidence of olive flies and its natural enemies as well as percent parasitism throughout the year. The area along the river had larger, less disturbed wild olive stands and produced a

virtually continuous source of fruit throughout the year compared to the other wild olive stands which displayed more erratic olive production. This correlated to lower incidence of both groups of insects in these areas. Measurements of olive fruit sizes from each of the areas indicated that the wild olive fruit stands along the river produced larger wild olive fruit which probably more readily supported the successful development of both olive fruit flies and parasitoids. Ovipositor length of the collected parasitoid species were measured on specimens from each of the collection areas and these measurements were correlated with fruit sizes in order to determine differences in species composition based on fruit size.

Session 7: Compatibility of Insect-Resistant Transgenic Plants with Biological Control

STUDIES ON THE COMPONENTS AND DIVERSITY OF THE ARTHROPOD COMMUNITY IN TRANSGENIC *Bt* COTTON (FOUR BOLLGARD VARIETIES) IN JIANGSU COASTAL REGION

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In order to overall evaluate the ecological safety and planting risk of insect resistant transgenic Bt cotton, the composition and diversity of the arthropod community in 4 Bollgard varieties: '109B', '154', '690', and '972' was studied by means of the quantitative analysis method in Dafeng of Jiangsu coastal region. The results showed: no marked difference in arthropod species was found between each of Bollgard varieties and traditional variety check (Simian-3). The dominant species and their abundance were very close among them. The parameters of arthropod community structure include species richness (S) total individuals (N) community diversity (H₂) evenness (J) and concentration © in each of Bollgard varieties were fluctuating near the ones in check. The path analysis of H₂† with other parameters showed that it all was dominant C and J that mainly decided the change of H₂ for arthropod community in 5 treatments. H₂ would be reduced with increasing of dominant C and decreasing of J for all treatments. The coefficient of species similarity (V₁) and the coefficient of community similarity (V₂) between each of Bollgard varieties and check was determined by the Sorenen's formula and all very high. The arthropod community average value of V₁ is 0.5758±0.7742 whereas the average value of V₂ is 0.4961±0.6484 respectively. From the results above, it is clear that the stability of the arthropod community of 4 Bollgard varieties is not significantly different than traditional cotton and its risk increase by planting Bollgard varieties is not obvious.

DO TRANSGENE PRODUCTS IN HONEYDEW POSE A RISK FOR AN APHID PARASITOID?

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An important aspect in the assessment of the environmental risks of genetically modified crops is their potential impact on non-target organisms. These include parasitoids and predators that are important for natural pest regulation. A gene encoding snowdrop lectin (*Galanthus nivalis* agglutinin, GNA) has successfully been engineered into a number of crops, causing partial resistance to sap-sucking insects such as aphids. However, also non-target insects may unintentionally be exposed to the insecticidal compound. Several studies have shown that sap-sucking insects excrete GNA in their honeydew, an important carbohydrate source for many parasitoids and predators. Here we address whether this route of exposure poses a risk for the aphid parasitoid *Aphidius ervi* Haliday (Hymenoptera: Braconidae).

Our study shows that ingestion of GNA dissolved in a sucrose solution has a direct negative effect on the longevity of adult *A. ervi*, similar to other parasitic wasps (Romeis *et al.* 2003). However, honeydew itself can be a suboptimal food source, which may affect the impact of the transgene product (Hogervorst *et al.* 2003). To address this issue we have investigated the effect of honeydew from two aphid species [*Sitobion avenae* (Fabricius) and *Rhopalosiphum padi* (L.) (Hemiptera: Aphididae)] feeding on GNA-expressing or non-transformed wheat plants on *A. ervi* longevity. We found a significant effect of aphid species on parasitoid longevity, but there was no significant plant effect. This was probably due to the low GNA expression level in the transgenic plants resulting in low GNA concentrations in the aphid honeydew. In addition, the honeydew was analysed for carbohydrate and amino-acid composition, factors that are of importance for the nutrition of honeydew-feeding insects. HPLC analysis showed some slight differences in honeydew sugar and amino-acid composition between the two aphid species but no effect due to the GNA expression was apparent.

To increase the amount of GNA in the honeydew, *R. padi* were fed on either pure artificial diet or on diet containing 0.1% GNA. Collected honeydew was fed to *A. ervi*. Honeydew from *R. padi* reared on a diet containing 0.1% GNA reduced longevity of *A. ervi* by

almost 50% when compared to parasitoids that had been fed with honeydew from aphids reared on pure diet. In contrast, a 2M sucrose solution with 0.1% GNA reduced the lifespan of the parasitoid by about 30% when compared to a 2M sucrose solution.

From these results we can conclude that if *A. ervi* is exposed to GNA through honeydew, this will negatively affect their lifespan. This will have potential implications for biological control of aphids.

Session 7: Compatibility of Insect-Resistant Transgenic Plants with Biological Control

EFFECTS OF GENETICALLY MODIFIED CROP-DERIVED *BACILLUS THURINGIENSIS* TOXINS ON SOIL INVERTEBRATES

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Bacillus thuringiensis (Berliner) (*Bt*) is a gram negative soil bacterium that produces a range of endotoxins during sporulation. These endotoxins have entomopathogenic properties enabling *Bt* formulations to be used as microbial insecticides for more than 50 years by organic farmers. More recently *Bt* genes have been manipulated and inserted into several plant species to create insect resistant crops (*Bt* plants) thus reducing reliance on chemical pesticides. Though controversial, this method of biocontrol has many advantages over *Bt* insecticidal sprays including season-long expression of the proteins whilst protecting the proteins from degradation by UV light. Recent research has shown that the *Bt* plants introduce the endotoxin into the soil via root exudates, post-harvest incorporation of plant residues and pollen. The endotoxin can bind to soil, especially clay and humic acid particles, which protect it from degradation whilst remaining active against target pests. This has led to concerns about a build up of the toxins in the environment to levels which are potentially harmful to non-target organisms or would increase the chance of resistant target organisms emerging.

We investigated the effects of genetically modified crop-derived *Bt* endotoxins on the soil biotic community using genetically modified *Brassica oleracea* var. *italica* (L) (Brassicaceae: Cruciferae) producing the Cry1Ac lepidopteron-active toxin. Effects on both target lepidopteron larvae and non-target soil-dwelling organisms, including insects, molluscs and nematodes, were investigated using microcosm studies and enzyme linked immunosorbent assay (ELISA) techniques to track the endotoxin. Results so far indicate that the *Bt* toxin can have a direct effect on non-target organisms but effects are species specific. Indirect effects on predators have also been measured. Implications for the widespread use of these pest-resistant plants, within integrated pest management systems, are discussed.

ASSESSING THE EXPOSURE OF ARTHROPOD PREDATORS TO CRY1AB TOXIN IN *BT* MAIZE FIELDS IN SPAIN

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30 A major concern regarding insect resistant transgenic plants is their potential impact on non-target organisms, including beneficial arthropods such as predators. In order to assess the risks that transgenic plants pose to predatory arthropods, several factors have to be taken into account. An important factor is the degree of exposure of these arthropods to the insecticidal protein expressed by the plant. In the case of transgenic maize expressing the *cry1Ab* gene derived from the soil bacterium *Bacillus thuringiensis* (Bt), predators may be exposed to the Bt toxin when preying on herbivores containing the toxin after feeding on transgenic plants. In addition, they can be directly exposed when feeding on plant parts (e.g. pollen) containing the toxin. So far, assessment of the exposure predominantly has been done in laboratory studies. To date, little is known about the extent to which beneficial arthropods are exposed to Bt toxin under field conditions.

We collected the most relevant arthropod groups (both, herbivores and predators) in Bt maize (Event 176) fields in Spain at different periods in the season and analysed their toxin content using an immunological test (ELISA). Our results revealed that Bt toxin levels in most important herbivores (aphids, thrips, cicadellids) and consequently in predators were extremely low or not detectable before pollen shed started. The pattern changed when pollen was available. Nymphs as well as adults of *Orius* spp. (Heteroptera: Anthocoridae) contained significant levels of toxin. Similar levels were found in adults of *Chrysoperla* spp. (Neuroptera: Chrysopidae) during that period. After pollen shed, spider mites (*Tetranychus urticae* Koch (Acarina: Tetranychidae)) occurred in the maize fields. This herbivore was shown to contain levels of toxin that exceeded those expressed by Bt maize plants by a factor of three. Accordingly, significant toxin levels were found in *Orius* spp. and larvae of *Chrysoperla* spp. The highest levels among predators were found in the larvae of the specialist spider mite feeder *Stethorus punctillum* Weise (Coleoptera: Coccinellidae).

Our findings confirmed that the Bt toxin can be transmitted from lower to higher trophic levels and thus that predators are actually exposed to it. However, the exposure strongly depended on the feeding ecology of predators as well as on the sampling time in the season, that is to say on the presence of pollen or spider mites in the field. This study provides information about the feeding behaviour and toxin exposure of important predators in the field and represents a further step towards an ecological risk assessment of Bt maize.

Session 7: Compatibility of Insect-Resistant Transgenic Plants with Biological Control

COMPATIBILITY OF *APHELINUS HORDEI* (KURDJUMOV) (HYMENOPTERA: APHELINIDAE) WITH RUSSIAN WHEAT APHID RESISTANT CULTIVARS IN SOUTH AFRICA

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Host plant resistance and biological control are effective methods of insect pest control. Since both methods are generally inexpensive, self-perpetuating and non-pollutant they are rendered as desirable and sustainable components of integrated pest management. Tritrophic studies involving the interactions between plant, herbivore and natural enemy indicated that the application of host plant resistance and biological control to an insect pest could give significantly better or worse control than expected from each factor alone.

Both host plant resistance and biological control have been applied against the Russian wheat aphid, *Diuraphis noxia* Kurdjumov (Homoptera: Aphididae), in South Africa. This is the most serious insect pest of wheat in the summer rainfall areas of the country. To date 12 (non- transgenic) cultivars, containing different levels of plant resistance, have become available to farmers in the country. More than 70 % of the wheat farmers in the eastern parts of the Free State Province are planting resistant cultivars, which resulted in a reduction of ca. 36% in insecticide application between 1990 and 1996. During this period the parasitoid *Aphelinus hordei* (Kurdjumov) was introduced from the Ukraine into South Africa and released in wheat fields. Field trials were conducted in 1998 and 1999. Trials were planted in two major treatment blocks about 700m apart and in only one block parasitoids were released. Within each block, three cultivars (two resistant and one susceptible) were planted in plots and replicated four times. *Aphelinus hordei* (2000/plot) was released augmentatively once early in the season and the aphid population development was followed on a weekly basis.

As expected percentage infested tillers on the resistant cultivars was significantly lower than on the susceptible cultivars. Where *A. hordei* was released a further reduction in the percentage of infested tillers was observed in the susceptible and the resistant cultivars (Fig. 1A and B). This is an indication that *A. hordei* is compatible with these cultivars.

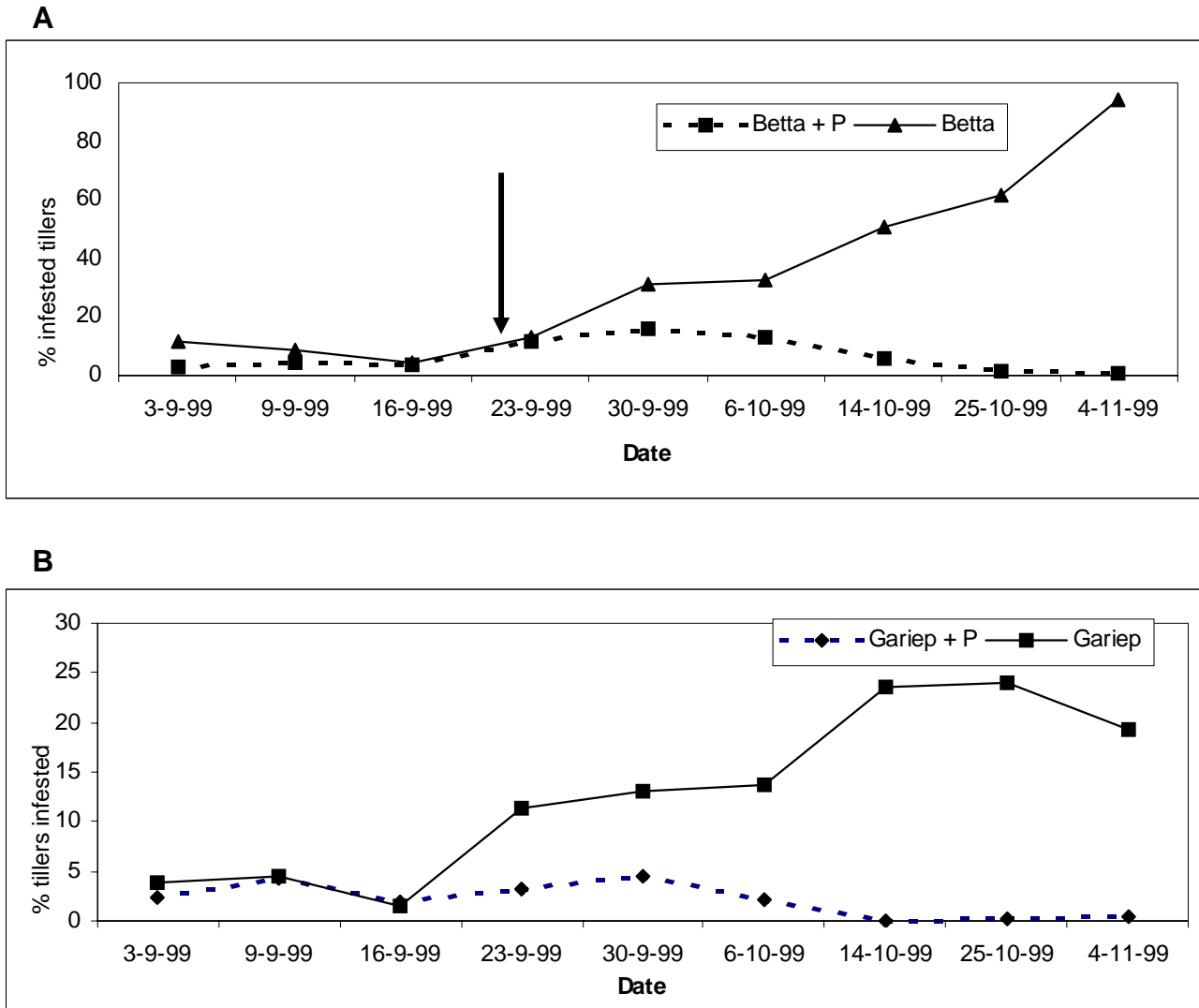


Figure 1. Percentage infested tillers per plot in the presence of the parasitoid *A. hordei* (+P) or in the absence of parasitoids on: (A) the susceptible cultivar Betta and (B) the resistant cultivar Gariep. Arrows indicate the date of parasitoid release.

EFFECTS OF GARLIC LEAF LECTIN ON *APHIS CRACCIVORA* AND ITS PREDATOR *COCCINELLA SEPTEMPUNCTATA*

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A number of crop plants have been genetically modified to express lectins, carbohydrate-binding proteins, with the potential to protect the crop from sap-sucking pests such as aphids. The lectin that has received most attention is the mannose-binding snowdrop lectin (*Galanthus nivalis* agglutinin, GNA). Currently efforts are being made within the Indo-Swiss Collaboration in Biotechnology (ISCB) to insert the gene that encodes for another mannose-binding lectin, the garlic (*Allium sativum* L. (Amaryllidales: Alliaceae)) leaf lectin (ASAL), into chickpea (*Cicer arietinum* L. (Fabales: Fabaceae)) (Romeis et al. 2004). The primary target is the aphid *Aphis craccivora* Koch (Hemiptera: Aphididae), which is the transmitter of a number of viral diseases. ASAL has been reported to affect Hemipteran species including *A. craccivora* in artificial diet bioassays (Majumder et al. 2004). In aphids, lectins appear to act primarily by reducing growth, development, and fecundity rather than causing mortality. This makes the additional impact of antagonists such as predators and parasitoids necessary for pest control. We have therefore compared the insecticidal properties of ASAL for the target pest and one of its predators in the Indian chickpea system, the ladybird beetle *Coccinella septempunctata* L. (Coleoptera: Coccinellidae).

Effects of ASAL on *A. craccivora*. The biological activity of mannose-binding lectins can be confirmed by their agglutination properties of rabbit erythrocytes. The agglutination properties of ASAL and GNA were compared and artificial diet tests were performed to assess the effects on *A. craccivora*. Although agglutination properties of both lectins were shown to be similar, strong differences in toxicity towards *A. craccivora* were found. When aphids were fed with ASAL at concentrations ranging between 0.0001% and 0.1 (w:v), mortality after 8 days ranged between 50 and 100%. In contrast, only 14% mortality was observed when aphids were reared on a 0.1% (w:v) GNA diet.

Effects of ASAL on *C. septempunctata*. Feeding 1st instar *C. septempunctata* with ASAL dissolved in a sugar solution (0.1%, w:v) resulted in a significant decrease in predator

longevity, which appeared to be due to feeding repellency rather than toxicity. No effects were apparent when the predator larvae were fed with aphids kept on artificial diet containing ASAL.

Implications. The fact that ASAL shows a high activity towards the target pest, but has little effect on the non-target predator species makes it an interesting compound to deploy in transgenic chickpeas. To fully assess the potential of this transgenic approach it has to be verified whether ASAL expressing chickpeas would eventually protect the crop from virus infection.

Session 8: The Role of Food Supplements in Biological Control

THE EFFECT OF FOOD SUPPLEMENT ON LIFESPAN AND NUTRIENT METABOLISM IN A PARASITIC PHORID FLY

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The phorid fly, *Pseudacteon tricuspis* Borgmeier (Diptera: Phoridae) has been released in many parts of southern United States for biological control of red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae). However, little is known about the nutritional ecology and foraging behavior and of phorid flies and the impact of sugar feeding on their lifespan. We investigated the effect of sugar feeding on the longevity of female and male *P. tricuspis* along with temporal patterns of nutrient accumulation and mobilization in flies provided different diet regimes.

Parasitoid longevity. Flies fed 25% sucrose continuously throughout their lifespan have greater longevity than completely starved (provided no water and no sugar solution) flies, sugar starved (provided water only) flies, or flies fed sugar solution only on their first day of adult life (see Table 1). Flies fed 50% sucrose have similar survivorship as those fed 25% sucrose. There was no significant effect of sex on longevity. Sex, however, had a significant effect on wing length with females being larger than males. *P. tricuspis* adults are also able to utilize floral nectar from buckwheat and honeydew produced by cotton aphids, suggesting that availability of supplemental sugar sources near its release sites may enhance the efficacy of *P. tricuspis* as a biological control agent of imported fire ant.

Patterns of nutrient metabolism. The temporal patterns of nutrient accumulation and utilization are also compared in *P. tricuspis* fed different diets: sugar-starved, sucrose-fed on the first day of adult life only, and sucrose-fed continuously. Adult *P. tricuspis* emerge with no gut sugars, and only minimal amounts of body sugars and glycogen. While the levels of body sugars and glycogen decline gradually in sugar-starved flies, a single day of sugar feeding resulted in the accumulation of maximum amounts of gut sugars, body sugars, and glycogen. High levels of these nutrients are maintained in female and male phorid flies feeding sucrose continuously over the observation period, whereas nutrient levels decline in flies fed only on the first day of life, beginning 1 day postfeeding. Honeydew-fed phorid flies also show a slight increase in the levels of body sugars. Female and male *P. tricuspis* emerge with an estimated $12.3 \pm 2.3 \mu\text{g}$ and $7.2 \pm 1\mu\text{g}$ lipid reserves per fly, respectively. These teneral amounts represent the highest lipid levels detected in adult flies, irrespective of their diet and are maintained over the life times of sucrose-fed female and male flies, but declined steadily in sugar -starved females. Wing length was positively correlated with the amounts of gut and body sugars in female and male flies, and also with glycogen levels in male flies. However, no significant relationship between wing length and lipid amounts was detected in female and male flies, suggesting that the amount of lipid reserves at emergence is not a function of size. These data suggest that adult *P. tricuspis* are capable of converting dietary sucrose to body sugars and glycogen, but not lipids.

Table 1. Mean longevity (days \pm SE) of female and male *P. tricuspis* subjected to different feeding regimes.

Diet	Females	Males
Completely starved ¹	1.2 \pm 0.1 c	1.3 \pm 0.1 d
Sugar starved	3.0 \pm 0.2 b	3.3 \pm 0.2 c
Sugar fed day 1 only	4.2 \pm 0.3 b	5.0 \pm 0.3 b
Sugar fed throughout lifespan	7.9 \pm 0.8 a	8.9 \pm 0.9 a

¹Completely starved = flies provided with no water and no sugar solution. Means within the same column having different letters are significant ($P < 0.05$).

Session 8: The Role of Food Supplements in Biological Control

POTENTIAL EFFECT OF FLOWERING PLANTS ON THE ACTIVITY OF THE EGG PARASITOID *TRICHOGRAMMA BOURARACHAE* PINTUREAU AND BABAULT (HYMENOPTERA, TRICHOGRAMMATIDAE), A CANDIDATE FOR BIOLOGICAL CONTROL IN OLIVE CULTIVATION

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The egg parasitoid *Trichogramma bourarachae*, isolated from sentinel eggs in an olive grove near Alexandria, Egypt, and experimentally released in an olive grove near Cairo for control of Lepidopterous olive pests, was significantly attracted by flowering vegetable islands. These islands were introduced in the olive grove to enhance the productivity of the beneficial fauna. They were distributed in 2 x 2 m plots alternating between olive trees in four rows and included six different treatments: 1- Uncovered soil as control, 2- Summer squash, *Cucurbita pepo* L. (Magnoliopsida: Cucurbitaceae), 3- Radish, *Raphanus sativus* L. (Capparales: Brassicaceae), 4- Roquette, *Eruca sativa* Miller (Capparales: Brassicaceae), 5- Turnip, *Brassica rapa* L. (Capparales: Brassicaceae), and 6- Wild Groundsel, *Senecio vulgaris* L. (Asterales: Asteraceae). Each island was sown at two or three different dates to create continuous flowering of vegetables from April to July. *T. bourarachae* was mass-reared in the laboratory and released in several applications from March to August 2004 at a rate of 9.000 wasps/tree. *Trichogramma bourarachae* could be frequently collected from sentinel eggs, distributed on olive trees neighbouring vegetable islands. Captures occurred more than 60 m apart from the releasing area suggesting an attractiveness of flowering vegetation for this egg parasitoid, which was not autochthonous distributed in this olive grove before release. The potential use and effect of *Eruca sativa*-flowers as food source for *T. bourarachae* was studied in laboratory and greenhouse tests by evaluating fecundity, longevity as well as searching capacity of this species in the presence of flowers in comparison to wasps which were offered honey or water.

**THE EFFECT OF FOOD RESOURCES ON THE LONGEVITY OF
GONATOCERUS ASHMEADI, *G. TRIGUTTATUS*, AND
G. FASCIATUS, EGG PARASITOIDS OF THE GLASSY-WINGED
SHARPSHOOTER (*HOMALODISCA COAGULATA*)**

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The effect of water only, 3:1 honey-water solution, floral and extra-floral nectars from five different plants [buckwheat (*Fagopyrum esculentum* Moench (Polyganles: Polygonaceae)), alyssum (*Lobularia maritima* L. (Capparales: Brassicaceae), *Phacelia tanacetifolia* Benth (Solanales: Hydrophyllaceae)), dill (*Anethum graveolens* L. (Araliales: Umbelliferae)) and broad bean (*Vicia faba* L.), honeydew from *Coccus hesperidum* L. (Homoptera: Coccidae) and *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae), a commercially available food supplement (Eliminade) and citrus foliage on the longevity of male and female *Gonatocerus ashmeadi* Girault, *G. triguttatus* Girault and *G. fasciatus* Girault (Hymenoptera: Mymaridae), was determined in the laboratory. Additionally, the sugar composition of each food resource was determined using high performance liquid chromatography (HPLC) analysis. Honey-water and *F. esculentum* significantly increased longevity of male and female *G. ashmeadi*, *G. triguttatus*, and *G. fasciatus* up to 1760%, 1223% and 1359%, respectively, compared with water.

These results indicate that resource procurement maybe extremely important for enhancing the survival of these parasitoid species in agroecosystems for *H. coagulata* biological control. For both sexes and all three parasitoid species, survival on citrus foliage and *P. tanacetifolia* flowers was equivalent to that on water only. *Vicia faba* extrafloral nectaries increased survival of both male and female *G. fasciatus* by 263% and 376%, respectively but did not significantly increase the survival of *G. ashmeadi* and *G. triguttatus* compared with water only. *Lobularia maritima* and *A. graveolens* significantly enhanced survival of female *G. ashmeadi* and *G. triguttatus* by up to 670%, but not *G. fasciatus*. The longevity of both sexes of *G. ashmeadi* and *G. triguttatus* was up to 439% higher on Eliminade compared with water only, however there was no significant effect of Eliminade on survival of *G. fasciatus*. *Coccus hesperidum* honeydew increased survival times up to 565% for all mymarid species compared with citrus foliage alone, whereas, parasitoid survival on *H. coagulata* excrement was equivalent to that on citrus foliage which was comparable to longevity estimates when parasitoids had access to water only.

These results suggest that in citrus orchards, *H. coagulata* excrement is not an adequate food supply for *Gonatocerus* parasitoids, and that low non-damaging *C. hesperidum* populations may be beneficial for enhancing parasitoid survival and could augment biological control of *H. coagulata*. HPLC analysis showed that food resources that were most beneficial to *Gonatocerus* parasitoids (honey-water, *F. esculentum* and *C. hesperidum*) possessed a high proportion of glucose (up to 44.3%) and fructose (up to 52.5%), and results suggested that sucrose may not be important for parasitoid survival since honey-water resulted in maximum parasitoid longevity, yet did not contain sucrose. Honey-water and *C. hesperidum* were very beneficial for parasitoid survival and were the only food resources that contained maltose (3-9%), thereby indicating maltose may be beneficial for parasitoid survival. Citrus and *P. tanacetifolia* contained favorable proportions of glucose and fructose, and possessed a significantly higher (up to 255.2%) total sugar concentration compared with *C. hesperidum* excreta and *F. esculentum*, which both promoted greater longevity of these parasitoids. Therefore, the inability of *Gonatocerus* spp. to benefit from nectar produced by citrus and *P. tanacetifolia* may be related to flower morphology.

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CONSERVATION BIOLOGICAL CONTROL TO ENHANCE THE EFFICACY OF A PARASITOID OF THE ARGENTINE STEM WEEVIL

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Conservation biological control aims to enhance the efficacy of biological control agents, partly by providing them with 'resource subsidies'. *Microctonus hyperodae* Loan (Hymenoptera: Braconidae) is a parasitoid of *Listronotus bonariensis* (Kuschel) (Coleoptera:

Curculionidae), a major pasture pest in New Zealand. To understand the delivery of benefits to *M. hyperodae* in the field, carbohydrate metabolism in the parasitoid was investigated using high performance liquid chromatography (HPLC). Lifetime patterns of carbohydrate metabolism were compared in the laboratory between unfed and honey-fed *M. hyperodae*. These patterns were compared with field-collected parasitoids.

Laboratory-reared parasitoids. The honey-fed individuals lived twice as long as did parasitoids given water only. Fed and unfed parasitoids could be distinguished by measuring total sugar levels. Furthermore, the glucose/(glucose+fructose) ratio [g/(g+f)] of honey-fed *M. hyperodae* was always lower than that of the unfed and newly-emerged individuals.

Field-collected parasitoids. The HPLC analysis of field-collected parasitoids from near buckwheat (*Fagopyrum esculentum* Moench cv. Katowase) plots showed that a high proportion of parasitoids near flowering plants had fed from buckwheat. Those which fed had a low [g/(g+f)] ratio and varying levels of total sugar in their body. Recently-fed individuals had high levels of total sugars whereas previously-fed had low total sugars. The field-collected and laboratory-reared unfed parasitoids showed similar total sugars and [g/(g+f)] ratios.

These results can be very useful for determining the nutritional state and feeding history of field-collected *M. hyperodae* and the analytical procedure tested here can be used to help evaluate the benefits to parasitoids through the provision of plant 'resource subsidies' in the field.

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EFFECT OF FLOWER PRESENTATION METHOD ON PARASITOID WASP SURVIVAL

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The first stage in developing a conservation biological control strategy typically involves ranking the suitability of various plant species as food resources for the target natural enemy species in the laboratory. It is unclear whether excised flower shoots in water will generate similar effects on the life history traits of a natural enemy compared with those using flowers remaining attached to a rooted plant. Both methods have been used in previous studies, yet none has quantified this effect. It is possible that plant nectar quality and quantity is altered as a result of changes in the physiological condition of the excised flowers. Laboratory tests are being conducted to assess the influence of flower presentation method for 10 plant species on the lifespan of *Aphidius ervi* Haliday (Hymenoptera: Braconidae), a potentially important parasitoid wasp of lucerne aphids, *Acyrtosiphon kondoi* Shinji and *Acyrtosiphon pisum* (Harris) (Homoptera: Aphididae). It is predicted that parasitoid wasp survival will be lower on excised compared with intact flowers for each plant species, but that the overall ranking of the plant species tested will remain unchanged. Results and recommendations for the design and interpretation of future laboratory studies are provided.

BIOLOGICAL CONTROL OF CITRUS RUST MITE USING INDIGENOUS AND INTRODUCED GENERALIST ACARINE PREDATORS IN ISRAEL

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Citrus rust mite (CRM), *Phyllocoptruta oleivora* (Ashmead) (Acarina: Eriophyidae), is a major pest of citrus in Israel. Two approaches were evaluated in order to improve CRM biological control.

1. Classical biological control - five species of predatory mites: *Amblyseius herbicolus* Chant, *Euseius victoriensis* (Womersley), *Euseius elinae* (Schicha), *Typhlodromus rickeri* Chant and *Euseius stipulatus* (Athias-Henriot) (all Acarina: Phytoseiidae), were imported. Mite rearing and sampling methods were developed and improved, and the predators were released. Although some recovery was recorded, only *E. victoriensis* became established in the north of Israel. Despite the latter's establishment CRM control was not improved.
2. Conservation of indigenous acarine predators - damage caused by CRM is usually negligible in minimally to unsprayed isolated groves located in the central coastal plain of Israel. Assuming that resident natural enemies were responsible for this situation, we monitored the pest's potential predators in five unsprayed citrus plots, and concurrently determined their feeding habits in the laboratory. In the field *Iphiseius degenerans* (Berlese) and *Amblyseius swirskii* Athias-Henriot (both Acarina: Phytoseiidae) were the main predators found, the former being dominant during the critical winter and spring

months, the period of low pest populations. In the laboratory, when solely CRM was offered, the decline in pest numbers was similar in leaf arenas containing either phytoseiid or the stigmatid *Agistemus cyprius* Gonzalez, but *I. degenerans* killed fewer CRM in the presence of pollen. While the cessation of pesticide applications during two years was insufficient for reducing CRM populations, observations suggest that a three year break from broad spectrum pesticides would be the turning point for the reestablishment of *I. degenerans*, the postulated major winter time predator. Our field and laboratory data indicate that a complex of indigenous, generalist predators could be responsible for the control of CRM in isolated, unsprayed citrus groves on the central coastal plain of Israel.

Session 9: Role of Generalist Predators in Biological Control

STAGE PREFERENCE AND FUNCTIONAL RESPONSE OF *EUSEIUS HIBISCI* TO *TETRANYCHUS URTICAE* (ACARI: PHYTOSEIIDAE: TETRANYCHIDAE)

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The aims of this study were: a) determine the prey stage preference of female *Euseius hibisci* (Chant) (Phytoseiidae) at constant densities of different stages of *Tetranychus urticae* Koch (Tetranychidae), b) assess the functional response of the predatory females to the varying densities of eggs, larvae, or protonymphs of *T. urticae*, and c) estimate the functional response of *E. hibisci* when pollen of *Ligustrum ovalifolium* Hassk. (Oleales: Oleaceae) was present as well. We conducted experiments on excised pieces of strawberry leaf arenas (*Fragaria ananassa* Duchesne (Rosales: Rosaceae)) under laboratory conditions of $25\pm 2^\circ\text{C}$, $60\pm 5\%$ RH and 12 h photophase. Our results indicated that the predator consumed significantly more prey eggs than other prey stages. Consumption of prey deutonymphs and adults was so low that they were excluded from the non-choice functional response experiments. The functional response on all food items was of type II. The two parameters of the functional response were estimated for each prey type by means of the adjusted nonlinear regression model. The highest estimated value of a' (instantaneous rate of discovery) and the lowest value of T_h (handling time, including digestion) were found for the predator feeding on prey eggs, and a' was lowest and T_h highest when fed protonymphs. Using the jack-knife method, the values for the

functional response parameters were estimated. The values of a' and T_h produced by the model were similar among all prey types except for the eggs, which were different. Using pollen simultaneously with prey larvae decreased the consumption of the latter over the full range of prey densities. The suitability of this predator for biological control of *T. urticae* on strawberry is discussed.

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POPULATION INTERACTION BETWEEN *PHYTOSEIULUS LONGIPES* AND *TETRANYCHUS PACIFICUS* ON BEAN PLANTS

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The predator-prey interactions between *Phytoseiulus longipes* Evans (Acari: Phytoseiidae) and *Tetranychus pacificus* McGregor (Acari: Tetranychidae) and also between *Phytoseiulus persimilis* McGregor (Acari: Phytoseiidae), and *T. pacificus* were studied on bean plants. The plants were fertilized with Shultz-instantä, and RA.PID.GROä. Young potted lima bean plants, each having 7-8 leaves, were placed in wooden boxes (60x46x10cm) surrounded by a water barrier. Newly formed leaves on the plants were removed every 2-3 days to prevent excessive growth. The plants were watered every 2-3 days and were fertilized every two weeks. Experiments were conducted under constant conditions. Due to excessive time required to make counts of prey and predators, this study was conducted in three parts:

Part 1. Twenty five mated female *T. pacificus* were introduced to each of 10 plants by placing the mites on a single leaf at the base of the plant. When about half of the female prey had moved to the adjacent leaves (after about three hrs.), one 3-4 day old mated female *P. longipes* was introduced to each of the first 5 plants by placing the predator on a single leaf at the top of the plant. The additional 5 plants containing only prey individuals served as predator-free controls.

Part 2. Five plants were each infested with 25 *T. pacificus* females and one *P. persimilis* female in the same manner as in part one.

Part 3. Five plants were each infested with 25 *T. pacificus* females and two *P. longipes* females in the same manner as in part 1 and 2. The plants were placed in the wooden tray and spaced so that they did not touch each other. All stages of both predator and prey were counted on each plant every four days.

Spatio-temporal relationship between predator and prey populations. The parameter m^*/m (Lloyd, 1967) was used to estimate the patchiness or degree of aggregation between the populations of the prey and the predators, where m is the mean density and the m^* is the mean crowding, i.e., number/individual of other individuals/quadrat. The degree of spatial overlapping in the distribution between the populations of prey and each of the predators was estimated using the Kuno's overlapping index (Kuno, 1968). The value for each of the above two indices was calculated for the interaction between populations of the prey and each of the predators, on the basis of data gathered from the first 4 population censuses after the start of experiment, and the mean values for each index was calculated at each population census. Both predators required an average of 12-16 days to suppress the prey population. The degree of spatial overlapping between the distribution of both predators and their prey declined with time and decreasing prey density. The degree of aggregation of the prey increased while that of both predators decreased with prey decline and time. Regression between mean crowding and mean density revealed a clumped distribution in the population of the prey and the predators.

Session 9: Role of Generalist Predators in Biological Control

GENERALIST PREDATOR COMMUNITIES EXERT TOP-DOWN CONTROL OF A NEW INVASIVE PEST, THE SOYBEAN APHID (*APHIS GLYCINES* MATSUMURA)

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The soybean aphid (*Aphis glycines* Matsumura) (Heteroptera: Aphididae), is a major new invasive pest of soybean in North America. In 2003, over 42 million acres of soybean in the North Central US were infested and over 7 million acres were treated with insecticides to control soybean aphid. A complex of generalist predators including both native and previously introduced natural enemies, is abundant in soybean fields and may contribute to soybean aphid regulation. Alternatively, host plant quality, pathogens and abiotic conditions may combine to influence the abundance of this pest. During 2003, we conducted field studies to study the relative impact of generalist predators (top-down regulation) and agronomic practice (bottom-up regulation) on *A. glycines* by contrasting aphid growth under zero, low and conventional high-input production practices with and without access by predators. In addition we investigated the role of the generalist predator community in suppressing aphid

populations at three spatial scales; patches within a field, plants within patches, and within-plant distribution. In both studies, the major natural enemy species observed included *Harmonia axyridis*, *Coccinella setempunctata* (Coleoptera: Coccinellidae) and *Orius insidiosus* (Heteroptera: Anthocoridae).

Experiments reveal a significant top-down effect of generalist predators on aphid abundance with 3 - 10 fold higher aphid numbers in exclusion cages over sham or control treatments. There was also a smaller (1.5 - 2 fold) bottom-up effect when predators were excluded, indicated by a significant interaction between natural enemies and agronomic treatments. In 2004, we conducted a similar experiment in a field under conventional high-input production practices and found a 200 fold reduction in aphid numbers in sham or control treatments over exclusion cages. This season-long control resulted in a significant biomass increase of 2 fold and yield increase on the plants exposed to naturally occurring generalist predators over predator exclusion cages. These results present strong evidence of top-down regulation due to predator assemblages coupled with comparatively weak bottom-up effects. At the field scale, predators rapidly detected high-density patches of aphids and reduced their abundance and net replacement rates. At the plant level, the community of generalist predators reduced aphid populations to similar low levels, despite initial aphid density. Finally, at the within plant level, the proportion of aphids at each node was shifted lower on the plant in the presence of predators indicating that these areas may constitute a refuge from predation allowing aphid persistence in the field. As a whole, these studies document the critical importance of the generalist predator community in regulating soybean aphid abundance and suggest practices that may conserve and enhance their effectiveness.

IMMATURE PREDATORS: AN UNDERAPPRECIATED RESOURCE FOR BIOLOGICAL CONTROL

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The vast majority of arthropod predators that are considered important natural enemies in agriculture and forestry are predaceous not only as adults but also as larvae. However, for all but a few groups, such as coccinellids, chrysopids, and syrphids, we know almost nothing about the significance of the immatures in biological control. This ignorance arises from difficulties in identification and sampling. Identification of adult predators is challenging enough when closely related species are found at a given locality, but identification of the immature stages is especially problematic, because distinguishing morphological features are lacking or difficult to use. Using data from carabids and spiders, we show that immatures can be correctly identified by using polymerase chain reaction (PCR) primers to amplify cytochrome oxidase I sequences of immatures and matching them to those of correctly identified adults. The sampling problem is less tractable: many sampling technologies, such as sweeping and pitfall trapping, do not yield absolute density estimates, but rather some index of activity and density that is affected by stage- and species-specific responses to weather and catchability. Also, adults and immatures may tend to be found in different strata, e.g., carabid beetles, whose adults are mainly epigeal or foliar while the larvae tend to be found in litter and soil, sometimes to depths of several centimeters.

Here we present previously unpublished absolute density data from cotton and potato, and review the quantitative literature from other cropping systems. We show that immature predator densities can be considerable, often rivaling or exceeding those of adults. Absolute density data, coupled with PCR-based species identifications and molecular gut analysis, will enable an appreciation of the biocontrol potential of immature arthropod predators.

CARABID BEETLE ASSEMBLAGES IN INSECTICIDE-SPRAYED AND UNSPRAYED POTATO FIELDS

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Aphids, which cause direct sucking damage and transmit virus diseases, are the most important pest insects in northern Japan, and the farmers usually spray insecticides for this pest 3 – 4 times on average during a cropping season. However, a recent study (Ito et al., in press) showed that no aphid outbreaks occurred in the field when no insecticides sprayed due to the activity of indigenous predators (i.e. *Harmonia axyridis* (Pallas), *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) and *Orius* spp. (Heteroptera: Anthocoridae)), nor other leaf-eating pests such as *Mamestra brassicae* (L.) (Lepidoptera: Noctuidae) were conspicuous. It is considered that generalist predators are also contributing to leaf-eating pests control in the unsprayed potato field. Therefore, ground beetles assemblages were surveyed in the insecticide-free (=N) and conventionally sprayed (=S) plots. From early July to mid-August, total of 13 and 11 carabid species were collected in N and S plots, respectively. The most abundant species was *Bembidion morawitzi* Csiki (Coleoptera: Carabidae) in both plots (84.8% of the total catches in N plot and 77.1% in S plot) (see Table). Several dominant carabids (i.e. *B. morawitzi*, *Pterostichus planicollis* Motschulsky, *P. haptoderoides japonensis* Tschitscherine, *Amara chalcites* DeJean, *Chlaenius pallipes* Gebler and *Campalita chinense* (Kirby)) were experimentally confirmed that they fed lepidopteran larvae. Diversity indices calculated showed that S plot was more diverse than N plot. This might be due to the smaller catches of the most abundant species, *B. morawitzi* in S plot, though the species richness did not differ much. Since *B. morawitzi* is smaller in size, this species is likely to be more susceptible to insecticides than other species, but details are unknown. These results indicate that carabid beetle assemblage and diversity do not remarkably change even if conventional aphid control was carried out, unlike the case of aphid predators.

Total number of carabid beetles collected by pitfall traps in the potato field (July 3 – August 13, 2004).

Species	Applx. Size	N 0(%)	S (%)
<i>Bembidion morawitzi</i>	4 mm	870 (84.8)	356 (77.1)
<i>Pterostichus planicollis</i>	11 mm	54 (5.2)	28 (6.1)
<i>Amara chalcites</i>	9 mm	30 (2.9)	35 (7.6)
<i>Chlaenius pallipes</i>	14 mm	13 (1.3)	8 (1.7)
<i>Anisodactylus signatus</i>	12 mm	12 (1.2)	18 (3.9)
<i>Bembidion paediscum</i>	3 mm	12 (1.2)	3 (0.6)
<i>Clivina fossor sachalinica</i>	6 mm	10 (0.9)	6 (1.3)
<i>Campalita chinense</i>	30 mm	7 (0.7)	4 (0.9)
<i>Dolichus halensis</i>	19 mm	7 (0.7)	0 (0)
<i>Pterostichus haptoderoides japonensis</i>	9 mm	5 (0.5)	2 (0.4)
<i>Amara plebeja</i>	6 mm	3 (0.3)	1 (0.2)
<i>Bembidion semilunium</i>	6 mm	2 (0.2)	1 (0.2)
<i>Chlaenius micans</i>	16 mm	1 (0.1)	0 (0)
Total		1026 (100)	462 (100)

STUDY ON POPULATION DYNAMICS OF EUROPEAN ELM SCALE (HOMOPTERA: ERIOCOCCIDAE) AND ITS PREDATOR, EXOCHOMUS UNDULATUS (COLEOPTERA: COCCINELLIDAE) IN ISFAHAN, IRAN

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The European elm scale *Gossyparia spuria* (Modeer) (Hemiptera: Eriococcidae) is one of the most serious pests of the elms in Iran, Isfahan. An investigation of the population dynamics of European elm scale and its natural enemies revealed that the two coccinellid species, *Eriococcidae undulatus* Weise and *Chilocorus bipustulatus* L. (Coleoptera: Coccinellidae), were important predators of this pest. Of these two predators, *E. undulatus* had the highest density. This study was conducted by weekly sampling during 2000 and 2001. Twenty elm trees were randomly selected and the coccinellids on 10 infested branches of each elm tree were collected and counted. In order to investigate the population changes of different life stages of European elm scale, four branches (approximately 30 cm length and 2 cm diameter) were taken weekly from the four cardinal directions of selected trees and the number of each stage was recorded. Adults of *E. undulatus* were active in mid March and began to feed on overwintered *G. spuria* nymphs. The coccinellid population increased rapidly with the increase of temperature and peaked in early June. The population changes of *E. undulatus* were synchronized with the population changes of different stages of *G. spuria* including adults, first nymphal instar and eggs. The coccinellid population decreased rapidly in summer and fall, whereas high populations of *G. spuria* second nymphal instars were observed. In essence, this predator could be regarded as a potential biological agent to control *G. spuria* in the long term and it will be necessary to continue these studies to observe the efficiency of this predator.

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WHICH ARE THE KEY INVERTEBRATE PREDATORS OF EGGS AND LARVAE OF SCARABS?

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Larvae of the Scarabaeidae (Coleoptera) are amongst the most abundant and most widespread soil-living pests. They feed on roots and thereby cause considerable damage in grassland, arable land and forestry. Augmentative natural control by bacterial pathogens, fungi and entomopathogenic nematodes are of restricted use due to their sensitivity to environmental conditions and the high production costs compared with the crop value. Conservation natural control by soil-living invertebrate predators may be an inexpensive alternative strategy to regulate white grub populations in the long term. However, up to now, the key invertebrate predators of the most abundant European scarab pests, *Melolontha*, *Amphimallon* and *Phyllopertha*, are unknown.

We developed a PCR-based approach to detect scarab-DNA in the gut of soil-living predators. The new method has been tested in laboratory feeding experiments with one of the most abundant soil living generalist predators in grasslands, *Poecilus versicolor* (Sturm) (Coleoptera: Carabidae). The detection times differed among prey species and depended on the length of the amplified nucleic acid sequence. The half-lives of detectability ranged from 20 to 32 hours.

In contrast to earlier studies on trophic interactions with DNA-based methods, copurified inhibitors caused considerable problems in PCR. Widely used extraction and purification methods failed to eliminate the inhibitors or resulted in an unacceptable high loss of nucleic acids. The inhibition could be reversed by adding BSA in high concentrations to the amplification cocktail. To exclude false negative results in field caught predators a multiplex PCR was developed: the combination of predator-specific primers and prey-specific primers allowed testing for inhibition and detection of prey DNA simultaneously.

For the correct interpretation of field derived data it is highly important to distinguish between scavengers and active predators, as the former group has no or only an indirect influence on the prey population. Feeding experiments showed that carrion prey is equally well detected from the gut of predators as fresh prey. This means additional approaches have to be included to estimate the magnitude of scavenging. First results on field caught soil-living predators including larvae of Carabidae and Staphylinidae as well as Geophilidae will be presented.

Session 9: Role of Generalist Predators in Biological Control

IMPACT OF GENERALIST PREDATORS ON SURVIVAL OF EGGS OF *CHRYSOPERLA CARNEA*

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One of the natural enemies of aphids is *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysoptera), the green lacewing, which, in biological programs, is usually released in the egg stage. Predators of the egg have direct effect on the survival of *Chrysoperla carnea* eggs. To compare the effect of an ant as a generalist predator on the survival of lacewing eggs, three methods of egg release were conducted 1) chrysocard, 2) chrysobag and 3) eggs mixed with sawdust, all in 5 replications. Lacewing eggs were released on cabbage plants where ants were present. After 24 hours the remaining healthy eggs were counted. In chrysocard all eggs were healthy, but in chrysobag 35.6 and 0% and with the sawdust method 59.6 and 7.6%, respectively. The results showed that the chrysobag method protects eggs better than other methods.

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SEASONAL ABUNDANCE OF PREDATORS OF THE ELM APHID, *TINOCALLIS NEVSKYI*, IN SHAHREKORD, IRAN

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The seasonal abundance of predators of the elm aphid, *Tinocallis nevskyi* Rem., Que. and Heie (Homoptera: Aphididae) was studied in Shahrekord. The predators were sampled with a standard sweep net. The samples of 100 sweeps were taken each week in spring and summer 2003. The elm aphid was sampled on 20 randomly selected trees once per week. Eight shoots were removed from the two heights at ground level and four cardinal directions of selected trees. The number of elm aphids per five randomly selected leaves per shoot was recorded. Seasonal fluctuations of the aphid and predators consisted of Coccinellidae (four species), Miridae, Chrysopidae and spiders were compared. Peaks of populations of Chrysopidae and spiders as well as total population of predators were coincided with the peaks of aphid. Peaks of an active coccinellid, *Oenopia conglobata* L. were observed after the peaks of elm aphid. This lag was due to the developmental time of the coccinellid from larval to adult stages. In spite of decrease of aphid population in summer, some predator populations were increased. This was probably due to increase of other preys such as scale insects on elm trees. The findings can be used in assessing predator potentials in biological control of the elm aphid and choosing suitable selective insecticides and timing of applications.

RAPID SCREENING OF THE GUTS OF INVERTEBRATE PREDATORS TO DETECT DNA FROM MULTIPLE PREY

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It is now possible, for the first time, to rapidly screen large numbers of invertebrate predators to identify the whole range of prey that may be present in their guts, simultaneously. In the past molecular approaches (protein electrophoresis, monoclonal antibodies, PCR) have concentrated upon screening potential natural enemies to confirm trophic links with one or a few major crop pests. Little attention was given to the role of non-pest prey as alternative food for the predators and their effects on the dynamics of the predator-pest interactions. Alternative prey both help sustain predator numbers and, potentially, divert predators away from feeding on pests. Here we describe two approaches that allowed us to study the complex food webs involved.

We developed a multiplex PCR to analyse the gut contents of carabid beetles for 10+ prey targets simultaneously. The prey included various species of aphids, weevils, earthworms and molluscs. During the PCR DNA from many different prey were amplified in the same PCR reaction. Use of fluorescent-labelled primers allowed the PCR products to be screened using a fragment size analyser (an ABI377 or ABI3100 sequencer). As each amplicon for each prey species was a different size or labelled with a different marker, software could assign an identity to each prey DNA fragment. Prey choice could be analysed by comparing the ratios of different species present in the field and in the guts of the predators.

Our multiplex PCR depended upon pre-designing prey-specific primers for each target species. Any prey in the guts for which we did not have primers would go undetected. However, in research concerned with predator responses to prey diversity, 'diversity' becomes the parameter of primary interest. We developed a temperature gradient gel electrophoresis

(TGGE) approach using general invertebrate primers, or conserved primers that amplified groups of prey, such as all aphids or all earthworm species. Small difference between sequences of as little as a single base pair could be clearly separated by TGGE. Although we could run prey standards to confirm species identity, all species within groups would be detected, even those for which we had no sequence information. Prey diversity within the guts of predators can be compared with diversity in the field, revealing predator responses to diversity amongst prey resources.

SESSION 9: ROLE OF GENERALIST PREDATORS IN BIOLOGICAL CONTROL

A TALE OF TWO ENEMIES: WILL THE INTRODUCTION OF A GENERALIST PREDATOR IMPROVE OR DISRUPT BIOLOGICAL CONTROL?

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Generalist predators may act as either an additional source of pest mortality and therefore increase pest suppression or they may suppress more specialised control agents, thus releasing the pest from control. In New Zealand, the recent decision to mass rear and release a predatory ladybird against the eucalyptus tortoise beetle, *Paropsis charybdis* Stål (Coleoptera: Chrysomelidae), has provided an opportunity to examine the effect of a generalist predator on a pest and its specialist parasitoid.

P. charybdis was accidentally introduced into New Zealand from Australia in the early 1900s. Adults and larvae feed on eucalypt foliage and this species is a serious pest of eucalyptus plantations in New Zealand. A coccinellid predator, *Cleobora mellyi* Mulsant (Coleoptera: Coccinellidae), was introduced from Tasmania on several occasions from 1979 to 1987. *C. mellyi* became established at only one release site in the South Island (Maori Bay, Marlborough Sounds) and the species persists there today. While *C. mellyi* feeds readily on *P. charybdis* eggs it is not specific to this pest, requiring additional prey, particularly psyllids, to mate and

reproduce successfully. Only one psyllid species was present in New Zealand at the time of introduction so there may have been insufficient prey for *C. mellyi* to establish more widely. Several new eucalyptus psyllid species have established in New Zealand since the original release of *C. mellyi*. A more specific control agent, *Enoggera nassau* (Girault) (Hymenoptera: Pteromalidae), which is a solitary egg parasitoid of *P. charybdis*, was first introduced to New Zealand from Western Australia in 1987 and 1988. An additional release of a cool-adapted strain from Tasmania took place in 2000. This parasitoid is well established in New Zealand and attacks a high proportion of *P. charybdis* eggs.

The possibility that *C. mellyi* will establish more readily now that prey sources on eucalypts have increased will be tested following additional releases. Adults and larvae were collected from the Maori Bay population and mass reared over summer for field releases in the North Island in 2005. We will use this opportunity to examine intraguild interactions between the two agents. Parasitised eggs of *P. charybdis* are exposed to predation longer than unparasitised eggs, because *E. nassau* takes longer to hatch than *P. charybdis*. Feeding preferences of *C. mellyi* for parasitised and unparasitised eggs of *P. charybdis* were measured in comparison with psyllid prey, under laboratory and semi-field conditions. The possible repercussions of widespread establishment of this predator on the biological control of *P. charybdis* is discussed.

Session 9: Role of Generalist Predators in Biological Control

GENERALIST PREDATORS – SPECIES COMPOSITION, MIGRATION, AND MOLECULAR DETECTION OF APHID PREDATION IN SWEDISH SPRING SOWN CEREALS

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Spiders and carabid beetles are generalist predators and consume a large number of different arthropods including major agricultural pests. An important pest in cereals in Sweden is the bird cherry-oat aphid, *Rhopalosiphum padi* L. (Homoptera: Aphididae). If generalist predators are to prevent the population of *R. padi* from growing beyond economic thresholds, the predators have to find and consume aphids as soon as the pest starts colonizing the field. Thus, it is important that the predators occur in the fields early in the season. Knowledge about the abundance and distribution of generalist predators at the time of aphid establishment together with quantification of insect predation by these predators will contribute to our understanding of the role of generalist predators for pest control.

The timing of spider and carabid dispersal, their abundance, and distribution were investigated for 3 weeks directly after sowing in cereal fields around Uppsala, central Sweden, in 2003 and 2004. Information was also generated about the species richness of generalist predators on the different farms. Data from 2003 has not yet been completely processed. Results from 2004 showed that both spiders and carabids were present in the field immediately after sowing. No migration movement from surrounding habitats was detected. Different species showed different distributions in the fields. Many spider- and carabid species were bound to the field edge. However, some species were found uniformly spread in the field or even more abundant in the field than at the edge. This group of species included *Pardosa agrestis* (Westring) (Araneae: Lycosidae), *Oedothorax apicatus* (Blackwall) and *Meioneta rurestris* (Araneae: Linyphiidae), and *Bembidion lampros*, *Pterostichus cupreus* and *P. melanarius* (Coleoptera: Carabidae). Twelve Lycosidae, 36 Linyphiidae, and 41 carabid species were found in the fields. Also, an additional 10 spider families were found.

We are also investigating the biological control efficacy of the dominant predators. A method for quantification of insect predation is detection of prey DNA in predators using the Polymerase Chain Reaction (PCR). In order to draw reliable conclusions about predation rates in the field when using this method, it is critical to ascertain how long after a meal it is possible to detect prey DNA remains. As an initial step the half-life (the time after which only half of the meals eaten can be detected) of DNA from *R. padi* was determined for *P. cupreus*, *Bembidion* spp. and *P. agrestis*. Field collected predators were starved and then fed one *R. padi*. After consuming the prey, predators digested their meal for different time periods. Total DNA was extracted followed by PCR reactions with *R. padi* and aphid specific primers.

The entire analysis is not yet complete but preliminary results show that it is possible to detect DNA from one single *R. padi* in all three predatory species. However, the number of positive individuals per time period seems to decline rapidly. In future experiments the effect of temperature on DNA half-life will be examined. This information is essential for design of field collection methods.

Session 9: Role of Generalist Predators in Biological Control

PREDATION RATE AND FUNCTIONAL RESPONSE OF THE PREDATOR *MACROLOPHUS PYGMAEUS* ON THE APHID *MYZUS PERSICAE*

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The predation rate of the predatory bug *Macrolophus pygmaeus* (Rambur) (Hemiptera: Miridae) was studied for all instars of the aphid *Myzus persicae* (Sulzer) (Homoptera: Aphididae). A leaf of eggplant was placed in a petri dish with 4, 8, 12, 16, 20, or 24 aphids of each instar. One, fifth instar nymph of the predator was introduced into the dish after a 24hr exposure to an eggplant leaf without prey. Numbers of attacked aphids were recorded 24hr after the introduction of the predator into the dish. The experiments were conducted at 20°C and 25°C, a L16:D8 photoperiod, and 65 ± 5% R.H. For each aphid density, 10 replicates (i.e., 10 predators) were tested. The predation rate was found to be higher at 25°C than 20°C, whereas among the aphid instars, predation was highest on the first instar and decreased proportionally with increasing aphid instar. Apart from attacked aphids, aphids dying from causes other than predation were also recorded, and this number was positively related to aphid instar. Results were used to estimate the functional response curves for *M. pygmaeus*.

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ENCOUNTERS BETWEEN PREDATORS AND PREY: MOLECULAR DETERMINATION OF WHEN PREDATION OCCURS

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58 PCR-based gut contents analysis is increasingly used to track predator-prey interactions within invertebrate foodwebs. The gut contents of predators can be analysed using species-specific primers to amplify prey DNA and confirm trophic links. The retention time for DNA within the gut of a predator during digestion is potentially affected by factors including temperature, quantity ingested and size of the target DNA molecule. There is increasing evidence that large DNA fragments are digested relatively quickly within insect guts and even with advanced PCR methodologies small DNA fragments (< 300 bp) are usually targeted to ensure successful amplification of prey DNA. We tested the hypothesis that prey DNA retention/detection time is inversely proportional to fragment length and that this effect can be calibrated to determine, though *post-mortem* analysis, when prey are consumed.

A limiting factor in PCR gut contents analysis is that each predator needs to be analysed with primers that target many different prey amplicons, even though the mean number of targets (usually different prey species) in each gut sample may be low. This requires a large number of separate PCRs and is potentially time consuming. The problem has largely been solved by using a multiplex-PCR approach. Here we found a similar multiplex approach to be effective for amplifying many different sized DNA fragments (71-350 bp) from the same target prey species (the aphid *Sitobion avenae* F. (Homoptera: Aphididae)). This approach successfully amplified degraded DNA from the guts of predators and could amplify and detect many mitochondrial DNA fragments simultaneously.

Primers were designed and selected that could, in combination, detect 13 different sized amplicons using multiplex PCR. Predators (carabids and spiders) were fed on aphids and then killed after a range of time periods. From this it was possible to calculate the time at which 50 % of beetles tested positive, the median detection period (T_{50}), for each DNA frag-

ment. This analysis confirmed that larger DNA fragments decayed more quickly in the predator gut and the relationship between T_{50} and fragment size modelled using regression analysis. The time at which a predator consumed its last aphid meal could, therefore, be determined from the T_{50} of the largest detectable aphid DNA fragment still present in its gut.

The multiplex-fragment analysis approach was then applied to beetles caught fortnightly (May-July) using pitfall traps in a winter wheat field to measure the effects abiotic (time of year, temperature, day length) and biotic factors (aphid numbers) on the time of day when most aphids were encountered and consumed.

Session 9: Role of Generalist Predators in Biological Control

IMPACT OF NATURALLY OCCURRING GENERALIST PREDATORS ON *APHIS PUNICAE* IN A POMEGRANATE ECOSYSTEM

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The pomegranate (*Punica granatum* L.) (Myrtales: Punicaceae) is a commercial fruit tree in parts of Asia, tropical Africa and Europe. The aphid, *Aphis punicae* Passerini (Homoptera: Aphididae) a serious pest infesting pomegranate, attracts a host of general predators. The study was carried out in an unsprayed 12-year old orchard, for three years (2000 – 2002) at Bangalore (12°58'2" N, 77°35'2" E), S. India. For sampling the predators, each week 1600 shoots were randomly examined (80 shoots/tree x 20 trees). Concurrently, the aphids, weather parameters and crop phenological stages were also quantified. The data were subjected to suitable statistical analyses.

Species diversity. The diversity of predators consisted of three coleopterans two dipterans and a neuropteran. These were *Cheilomenes sexmaculata* (Fab.), *Scymnus* sp., *Pseudaspidemerus circumflexa* (Motsch.) (all Coleoptera: Coccinellidae); *Paragus serratus* (Fab.), *Ischiodon scutellaris* (Fab.) (both Diptera: Syrphidae) and *Chrysopa* sp. (Neuroptera: Chrysopidae).

Species abundance. The significantly most abundant predator was *C. sexmaculata* in all the three years, while others predators showed no difference statistically at $p = 0.05$. *C. sexmaculata* showed a high significant positive correlation with total predator counts ($r = 0.87, 0.94$ and 0.99 in 2000, 2001 and 2002, respectively). On an abundance scale, *C. sexmaculata* was followed by *Chrysopa* sp., *Scymnus* sp., *P. serratus*, *P. circumflexa* and *I. scutellaris*.

Numerical response. This was measured using correlation co-efficient 'r' ($p = 0.05$) and it was found that the predator *C. sexmaculata* had positive numerical response with the prey, *A. punicae* in 2000 and 2002 while *Chrysopa* sp. showed positive correlation in 2002. So, only these could be potential predators of *A. punicae* and the advantage is that both are amenable to mass rearing and release.

Control. Positive numerical response of predator numbers (x) brought about excellent reduction in prey population (y); ($y = 0.0027x + 0.0067, R^2 = 0.81$).

Weather and crop phenology effects. All the predators showed no correlation with major weather factors like temperature, relative humidity, wind speed and rainfall, implying that their occurrence was influenced more by the prey. However, the full mature leaf stage showed significant negative correlation with predator numbers; this again may be linked indirectly to fall in prey number, as at this stage, *A. punicae* density also showed a negative correlation with full mature leaf stage. Interestingly, at senescence stage of the tree, the predator numbers showed a surge with significant positive 'r' value opening new vistas of exploration; do yellow leaves act as cues or are predators responding to other homopterans?

The results of this long-term field study will be discussed in terms of sustainable biological control of *A. punicae* and conservation of the predators.

BIOLOGICAL CONTROL OF THE ROSY APPLE APHID – AN UTOPIAN DREAM?

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The rosy apple aphid, *Dysaphis plantaginea* Pass. (Homoptera: Aphididae), is one of the worlds most detrimental insect pests in apple production. Already single individuals of *D. plantaginea* are causing irreversible damage to leaves, branches and fruits which leads to severe yield losses. The common strategy to control the rosy apple aphid is to spray broad-spectrum insecticides in early spring. However, intense spraying resulted in the appearance of resistance and moreover suppressed the natural enemy complex of the aphids. Thus, the Research Institute of Organic Agriculture (FiBL, Switzerland) was involved in the development of alternative control strategies against the rosy apple aphid. Research was focused on the augmentation of the natural enemy complex of *D. plantaginea*. The aims of our studies were (1) to identify the most suitable control agent, (2) to investigate cost-effective mass-release strategies (eggs versus larvae and adults), (3) to define the optimal predator-prey ratio, (4) to determine the ideal date of release, (5) to understand the behaviour of the predators released and overall (6) to assess the effectiveness of augmentative releases.

Laboratory, semi-field and field screenings showed that the indigenous ladybeetle *Adalia bipunctata* L. (Coleoptera: Coccinellidae) was the most promising aphidophagous antagonist. Mass-releases of *A. bipunctata* pointed out that eggs are not suitable for the release in early spring. However, the release of larvae significantly reduced aphid density, but failed to suppress pest population below the economic threshold level, even at very high predator-prey ratios. Thus, research was focused to target the autumn generation of *D. plantaginea*. Once again the cold weather conditions prevented the hatching of *A. bipunctata* eggs. But the release of larvae decreased the deposition of over-wintering aphid eggs and consequently fewer aphids were observed the following season. Moreover, the release of adults indicated that *A. bipunctata* tended to stay on trees where they were released and significant reductions of aphids could be achieved. Overall, all attempts to reduce *D. plantaginea* below the economic threshold failed. However, the very low threshold value of one rosy apple aphid per fifty buds is a major challenge for any control strategy.

We conclude that the augmentation of natural enemies by mass-releases of ladybird beetles should be considered as a component of an integrated control strategy for *D. plantaginea* in the future. Augmentative release in combination with conservation biological control measures and cultivation of resistant apple varieties could lead to a more sustainable control strategy against *D. plantaginea*, in particular in regions (e.g. Belgium) where insecticide-resistance has evolved.

Session 10: Augmentative Biological Control in Outdoor Annual Crops

CHILO SACCHARIPHAGUS BOJER (LEPIDOPTERA: CRAMBIDAE) IN MOZAMBICAN SUGARCANE-A CASE FOR AUGMENTATION OR CLASSICAL BIOCONTROL, OR BOTH?

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In 1999, the spotted stalk borer *Chilo sacchariphagus* Bojer (Lepidoptera: Crambidae) was identified from bored sugarcane at two estates in Sofala Province, Mozambique. This borer, originating from Southeast Asia, is a major sugarcane pest in the Indian Ocean Islands of Mauritius, Madagascar and Reunion. It is the first time it has been recorded as a pest on mainland Africa, and poses a threat to the sugar industries of Mozambique and surrounding countries.

In 2000, a biological control programme against it was requested. As a result, regular surveys of infected sugarcane were implemented in order to determine the levels of infestation, seasonal occurrence and presence of any indigenous parasitoids attacking the different life stages of this borer. It soon became apparent that less than 1% of the larval and pupal stages were attacked by indigenous parasitoids, with a *Stenobracon* sp. (Hymenoptera: Braconidae) and *Cotesia sesamiae* Cameron (Hymenoptera: Braconidae) occasionally being found from parasitised large instar larvae. No pupal parasitism was recorded. In contrast, in excess of 90% of *C. sacchariphagus* egg batches were parasitised by *Trichogramma bournieri* Pintureau and Babault (Hymenoptera: Trichogrammatidae).

In 2001, releases of a pupal parasitoid *Xanthopimpla stemmator* Thunberg (Hymenoptera: Ichneumonidae), an indigenous natural enemy of *C. sacchariphagus* obtained from Mauritius (who originally obtained their population from Sri Lanka) commenced, as it was recognised that an empty niche existed for an effective pupal parasitoid. In addition, the South African Sugarcane Research Institute had a strong laboratory colony of this parasitoid. Population reductions of *C. sacchariphagus* of up to 60% were measured in the release fields within a year of releases commencing. Two years after releases, *X. stemmator* adults were collected from surrounding fields, as were parasitised pupae. It is thus apparent that *X. stemmator* is established on its aboriginal host at both sugar estates.

The decision now is whether to pursue the classical biocontrol approach, as used with *X. stemmator*. A larval parasitoid, *Cotesia flavipes* Cameron (Hymenoptera: Braconidae), has been reported as effective against *C. sacchariphagus*. A collection of this parasitoid from a population of *C. sacchariphagus* in Southeast Asia most closely related to the Mozambican population, could be imported into Mozambique in order to fill the empty larval parasitoid niche which is available in this new outbreak area, and thus increase parasitoid biodiversity. Alternatively, *T. bournieri* could be reared on a factitious host for release at times of the year when its population is low, in order to augment the field population already there, to make it a more effective parasitoid.

In the light of information already obtained about the population dynamics of *C. sacchariphagus* and its parasitoid complex in Mozambique, arguments for the single use of both approaches will be presented and discussed, as well as a case for the complementary use of both approaches together.

AUGMENTING LEAFROLLER PARASITOIDS IN APPLE ORCHARDS

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The obliquebanded leafroller, *Choristoneura rosaceana* (Harris), and three-lined leafroller, *Pandemis limitata* (Robinson) (Lepidoptera: Tortricidae), are important pests of apples in southern British Columbia, Canada. The two sympatric and generally bivoltine leafroller species overwinter as second or third instar larvae in bark crevices and begin to feed on apple foliage and blossoms when the latter are in the pink stage of bud development. The larvae shelter within blossoms or rolled leaves and fruit damage is caused when the sheltering leaf is tied to the fruit and larvae feed on the surface of the apple.

Over thirty species of parasitoids have been identified from these two leafroller populations in organically managed apple orchards in western Canada. Up to 25 % parasitism was recorded in the spring generation of overwintered leafroller hosts and up to 68 % in the summer generation. It would be advantageous to better understand the biology of this complex in order to allow augmentation of leafroller parasitism in orchards where previous pest management practices have deterred parasitoid establishment.

Laboratory bioassessments of common key leafroller parasitoids indicate that several species in the complex should contribute towards significant suppression of host populations in the field. Female *Apophua simplicipes* (Cresson) (Hymenoptera: Ichneumonidae) are long lived, parasitize a large number of obliquebanded leafroller larvae and act as predators of, and induce escape reactions in early instars of both leafroller species. The parasitoid emerges from the ultimate and penultimate host instar; however, feeding is significantly reduced in late instar parasitized hosts. *Apanteles polychrosidis* Viereck (Hymenoptera: Braconidae) parasitizes both leafroller species and emerges from third to fourth instar hosts, thereby killing the leafroller before it causes significant fruit damage. Both endoparasitoid species were mass reared and introduced as adults into separate commercial orchards under spring conditions, when wild members of these species would still be developing in overwintered leafroller hosts. Spring release was chosen to target generally higher population densities of host larvae in susceptible instars prior to significant damage occurring to the fruit. Potted trees, infested with early instar obliquebanded leafrollers, were used to monitor parasitoid activity in each orchard. *Apophua simplicipes* parasitized 7 to 29% of the sentinel larvae in the orchard in which this parasitoid was released and 21 and 9 % of wild leafrollers collected on release and non-release trees respectively. *Apanteles polychrosidis* parasitized 6 to 9 % of the sentinel larvae in the two orchards in which it was released and 7 % of wild leafrollers collected on release trees.

Releases of both parasitoid species were also carried out under fall conditions in commercial and research orchards, when leafroller hosts would be entering diapausing locations in bark crevices. Spring collections of wild leafroller larvae will be carried out in 2005 to assess augmentation of the two parasitoid species in overwintering host populations.

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EVALUATION OF CERTAIN EXOTIC APHID PARASITOID SPECIES AGAINST CEREAL APHIDS UNDER LABORATORY, FIELD CAGE, AND OPEN WHEAT FIELD CONDITIONS

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Aphids are the most serious insect pests attacking cereal crops, particularly wheat, barley and corn in many countries worldwide. Aphid parasitoids' importation and colonization have a great potential as a classical and effective biological control method. Through an Egyptian/American collaborative project carried out during 1997-2001, four cereal aphid exotic parasitoid species were imported from different countries to provide additional mortality factors to the indigenous ones, against key cereal aphid species in Egyptian and American wheat fields. A search for exotic cereal aphid parasitoid species or biotypes was carried out in Syria, Morocco, and Iran, in localities near the reported areas of the origin of cereal species and from habitats of climatic patterns similar to those in Upper Egypt and Southern California, USA. *Aphidius matricariae* Haliday (Syria), *Diaeretiella rapae* M'Intosh (Morocco), *Aphidius rhopalosiphi* De Stefani (Hymenoptera: Aphidiidae) and *Aphelinus albipodus* Hayat & Fatima (Hymenoptera: Aphelinidae) (Iran) were the parasitoid species introduced and evaluated under laboratory, field cage and open field conditions. Identification of the exotic species was confirmed by Dr. P. Stary, Institute of Entomology, Academy of Science of the Czech Republic. For each species, no. of parasitoids released, timing of release, wheat cultivar, wheat plant growth stage and location were recorded. The exotic parasitoid species showed different performances under several tested conditions. Generally, slight significant differences among the developmental periods of the released parasitoid species under the field cages at different environmental locations were recorded. *A. matricariae* parasitized significantly greater numbers of the oat-cherry aphid species, *Rhopalosiphum padi* L. (Homoptera: Aphididae) than the numbers parasitized by *D. rapae* and *A. rhopalosiphi* not only under the laboratory conditions (22+2°C and 50-60% R.H.) but also under field cages and in open fields, as well when compared with the indigenous *A. matricariae*.

CHOOSING (OR CREATING) PARASITOIDS FOR TEPHRITID FRUIT FLY CONTROL

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Inundative releases of mass-reared fruit fly braconid parasitoids have significantly suppressed Mediterranean fruit fly (*Ceratitis capitata* (Wiedmann)), *Anastrepha suspensa* (Loew) and *A. ludens* (Loew) (all Diptera: Tephritidae) populations, with reductions of up to 95%. The combination of inundative parasitoid releases and the Sterile Insect Technique (=SIT) is synergistically efficacious, and inundative parasitoid releases, unlike SIT, can be fully integrated with insecticide bait-sprays. Rearing methods and expenses vary with species, but production methods that reduce costs make the technique practical under more circumstances. These include the following:

- 1) Choice of optimal parasitoid. The numbers of parasitoids available for inundative release has recently expanded. While efficacy is paramount, reproductive potential under mass-rearing conditions, and ultimately cost, is an important consideration for its use in integrated control programs. With any particular parasitoid, those species with a higher r are “competitors” for inclusion in augmentative releases. Other characteristics to considered are: 1) host vulnerability as a function of ovipositor length; 2) optimal macro and micro-environments; 3) host range and stage attacked; and 4) capacity to forage well at declining host densities.
- 2) Multispecies rearing through serial exposure. Depending on species, parasitism of larvae in mass-rearing facilities is often ~50%. However, the unparasitized pupae remaining can be exposed to pupal parasitoids to increase production and provide a means of attacking immature fruit flies that escaped braconid parasitism. For example, the diapriid *Coptera haywardi* (Oglobin) is an endoparasitoid tephritid specialist that does not hyperparasitize braconid larval parasitoids. Serial parasitism can double production and in field cage trials *C. haywardi* killed substantial numbers of buried pupae.
- 3) Creation of parthenogenic strains. Sex ratios of mass-reared braconids are typically 50%. If quality female parasitoids could be reared in thelytokic strains then the production costs would be automatically halved. Such strains might be created by exploiting sex ratio-distorting microbial endosymbionts. Bacteria in the genus *Wolbachia* can produce sex-ratio distortions in Hymenoptera through induced diploidy. Collection and screening of naturally-occurring infested insects may provide thelytokic females. In some parasitoid genera, parthenogenic and bisexual females occur sympatrically, but there

have been few surveys searching for thelytoky. At present, we have one parthenogenic line of a figitid species, a possibly parthenogenic, but uncolonized, braconid and a braconid colony that has extreme female biases. *Wolbachia* infections are largely transmitted vertically through female offspring but horizontal transmission, both intra and inter specific, can occur during super- and multi- parasitism. Infections can also be passed through microinjection and we hope to develop both a *Wolbachia* “library” and means of transmission.

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LONG-TERM EFFECTS OF NATURAL ENEMY AUGMENTATIVE RELEASES ON VINE MEALYBUG PEST POPULATIONS

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Vine mealybug (VMB), *Planoccocus ficus* (Signoret) (Homoptera: Pseudococcidae) is one of the key pests of grape growing areas in South Africa, the Mediterranean and more recently California in the USA. The cryptic behavior and waxy excretions produced by this pest result in inefficient control when using conventional pesticides. In order to develop an alternative, environmentally safe pest management tool, the mealybug parasitoid, *Coccidoxenoides perminutus* (Timberlake) (Hymenoptera: Encyrtidae) was laboratory-reared and mass-released in VMB infested vineyards with low, medium and high pest infestation levels situated in three environmentally different grape-growing areas in the South-Western Cape Province in South Africa. Mass-releases were done for a four-year period in release plots which were left without pesticide treatment during this period and compared with control plots where VMB were controlled with the use of conventional pesticides. Control and release plots were separated by buffer plots aimed at limiting the spread of mass-released parasitoids from the release plots to the control plots. A physical monitoring system with known levels of error coupled with degree-day information for VMB was used to determine the start date and number of natural enemies that needed to be released in each of the areas. Mass releases were done at monthly intervals starting during the beginning of summer in each of the areas and continued until the end of summer.

VMB infestation levels as well as crop loss assessments were done during the season and at harvest respectively. Season-long monitoring was done in order to determine the presence of natural enemies as well as percent parasitism in each of the three areas. Biological control of this pest was not hampered by the presence of ants in the first two areas, but the third had high incidence of *Anoplolepis custodiens* (Smith) and *A. steingroeveri* (Forel) (Hymenoptera:

Formicidae), both aggressive and difficult to control pugnacious ant species. Analysis of the above data sets indicated differences in season-long VMB infestation levels between release and control plots for the four-season period and higher VMB infestation levels were found in the control plots compared to the release plots and similar findings were made during crop loss assessments. Crop losses were lower in the release plots compared to the controls. Higher numbers of the parasitoids were found in the release plots compared to the control plots. The plots where ants hampered bio-control had lower percent parasitism compared to the plots which did not have ants. VMB infestation levels gradually decreased during the four-year period in all three areas in both the control and release plots, possibly due to the movement of released parasitoids from release plots to the surrounding control plots. The lowest decline in VMB season-long population levels were encountered in the plots which had the highest VMB infestation levels as well as ant presence. A cost analysis was done for each of the three areas and this information indicated that mass releases of natural enemies against VMB was more cost-effective in the areas which had no ant interference and displayed lower initial VMB infestation levels.

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IMPROVING KNOWLEDGE AND MANAGEMENT OF NATIVE NATURAL ENEMIES FOR BIOLOGICAL CONTROL OF COLORADO POTATO BEETLE

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Several native natural enemies of Colorado potato beetle (*Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) (CPB)) provide biological control of this key pest of potatoes, tomatoes, and eggplant in North America. These native predators and parasitoids of CPB present a spectrum of biological opportunities and management options which range from conventional augmentation (laboratory mass-rearing and release), through field-based augmentation (in-situ natural enemy nursery) to conservation biocontrol tactics involving habitat modification to favor natural enemy populations and to “welcome” the natural enemies by providing critical resources to retain and propagate them within the agroecosystem.

Two species are more or less generalist predators, *Podisus maculiventris* (Say) (Heteroptera: Pentatomidae), and the polyphagous *Coleomegilla maculata* (De Geer) (Coleoptera: Coccinellidae). Both of these species are generalists in terms of habitat and also in their catholic feeding habits, which include significant consumption of plant juices and pollen respectively. This limits the usefulness, for these species, of simple augmentative tactics which may result in insufficient specialization in prey or in habitat, to meet the desired function of pest suppression. Instead it requires consideration of manipulation of predator movement and/or provision of properly-timed trophic supplements.

Three specialist natural enemies of CPB are less known, in part because of challenges in rearing. *Lebia grandis* Hentz (Coleoptera: Carabidae) is a carabid predator and parasitoid of *Leptinotarsa* which is well-synchronized with the CPB life-cycle but which is apparently limited both by conventional cultural practices and by climatic factors. We have succeeded in developing *Lebia* rearing techniques both for research and for augmentation objectives. The first instar carabid, the host-seeking life-stage, is short-lived and sensitive to climatic extremes. This puts a premium on tailoring cultural and chemical practices to favor the predator-parasitoid in the agricultural setting. Two specialist tachinid species, *Myiopharus doryphorae* (Riley) and *M. aberrans* (Townsend) (Diptera: Tachinidae), can be abundant in the field especially in the late season. Building on discoveries of the overwintering stage of the flies, which is within the adult chrysomelid host, we hope to develop in-field sorting techniques to favor the survival of tachinid-parasitized, compared to nonparasitized, host beetles.

Each natural enemy has specific requirements which limit the success of a conventional “lab rear and release” augmentation paradigm. This in turn requires species-specific modifications in biocontrol tactics, to include possible combinations of lab- and field-based rearing and conservation tactics.

PARASITOIDS OF *CHROMATOMYIA HORTICOLA* (GOUREAU) (DIPTERA: AGROMYZIDAE) ON GARDEN PEA IN JAPAN

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The leafminer, *Chromatomyia horticola*, which infests garden pea, is exposed to various parasitoids. We studied the biological control of *Liriomyza* leafminer species, especially *L. sativae* Blanchard and *L. trifolii* (Burgess) (Diptera: Agromyzidae), on glasshouse tomatoes using parasitoid complexes from *C. horticola*. Garden pea and tomato plants were collected in Shizuoka Prefecture, Japan, between April and May 2004. The numbers and species of leafminers and the parasitoids emerging from them were determined.

Species of leafminers and parasitoids. *Liriomyza sativae*, *L. trifolii*, and *L. bryoniae* (Kaltenbach) were collected from tomato plants, whereas *C. horticola* was mainly collected from the garden pea. *Liriomyza* leafminers were very rare on garden peas and their occurrence was limited to summer. The predominant parasitoids on the garden pea were *Diglyphus isaea* (50.5%), *Chrysocaris pentheus* (25.6%), *Neochrysocaris formosa* (10.8%), and *C. pubicornis* (7.5%) (Hymenoptera: Eulophidae). The former three parasitoid species were also frequently collected from tomato plants, suggesting that parasitoids of *C. horticola* also commonly parasitize *Liriomyza* leafminers.

Emergence of parasitoids. About 15 parasitoids emerged per 10 leaves. Ninety percent of parasitoids had emerged from garden peas on the 24th, 18th, and 15th day at 20°C, 25°C and 15°C, respectively (Table 1).

In conclusion, *Liriomyza* leafminers can be controlled by the parasitoids that emerge from *C. horticola* when garden pea plants are cut and introduced into tomato-growing glasshouses.

Table 1. Percentage of parasitoids emerging from leafminers on garden pea and tomato in Shizuoka Prefecture, Japan, between April and May 2004.

Species	Garden pea* n = 14,266 40 fields	Tomato* n = 148 7 fields
<i>Diglyphus isaea</i>	50.5%	6.4%
<i>Chrysocaris pentheus</i>	25.6%	19.1%
<i>Neochrysocaris formosa</i>	10.8%	32.0%
<i>Chrysocaris pubicornis</i>	7.5%	0.3%
Others (<i>Opius</i> sp., <i>Dacnusa sasakawai</i> , etc.)	5.6%	42.3%

*Average percentage of parasitoids per field.

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REPRODUCTIVE BEHAVIOR OF THE GENERALIST PREDATOR *MACROLOPHUS CALIGINOSUS*

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Macrolophus caliginosus Wagner (Heteroptera: Miridae) is a polyphagous predator native to the Mediterranean region where it spontaneously colonizes open field and protected vegetable crops. Several companies produce this natural enemy that it is widely used in vegetable crops in Europe. Nevertheless, its use in greenhouses is somehow unpredictable and a slow installation of the population is observed in some circumstances. Due to a lack of knowledge of its main biological traits it is difficult to overcome many of the difficulties that appear in the general practice of releasing this predator. Reproduction is one issue not well known and that affect the performance of the predator both in the greenhouse and in commercial mass rearing. In this work we have studied the copulatory behavior and the ovary maturation of this predator with the aim of implementing its use in augmentative releases in greenhouses.

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INTRAGUILD PREDATION AND FEEDING PREFERENCES IN THREE SPECIES OF PHYTOSEIID MITES USED FOR BIOLOGICAL CONTROL

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72 There has been much recent debate about the ecological impact on native insect and mite species by introduced arthropod biological control agents. This study investigated the ability of the non-native predatory mite *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) to feed on the native *Typhlodromus pyri* (Scheuten) (Acari: Phytoseiidae) and *vice versa*, as both species now co-occur in UK orchards. *Typhlodromips montdorensis* (Schicha) (Acari: Phytoseiidae) is a candidate for introduction into the UK as a glasshouse biological control agent. The ability of *T. montdorensis* to feed on the widely used *N. californicus* was investigated to identify possible intraguild predation, which may impact positively or negatively on the effectiveness of either or both species as predators of *Tetranychus urticae* in the glasshouse. Both *N. californicus* and *T. pyri* consumed larval stages of each other, but in choice experiments both showed a preference for *T. urticae*. Both *N. californicus* and *T. montdorensis* also fed on each other, but whereas *N. californicus* again showed a preference for *T. urticae*, *T. montdorensis* fed equally on *T. urticae* and *N. californicus*. Interactions between *N. californicus* and *T. pyri* and *N. californicus* and *T. montdorensis* are discussed in relation to their effectiveness as biological control agents in the glasshouse and the natural control of spider mite in the field.

DETERMINING THE EFFICIENCY OF TWO METHODS OF RELEASING LACEWING EGGS IN GREENHOUSES

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Lacewings are predatory insects that are widely used in biological control programs. Among them, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) is a promising biological control agent for use against pests in greenhouses and fields. In this study, two lacewing egg release methods were compared: 1- Distribution of eggs mixed with sawdust as an egg carrier and 2- dispersal of eggs using a Chrysobag (special net bags). Experiments using randomized block design were conducted using the cotton aphid, *Aphis gossypii* Glover (Homoptera: Aphididae), on 4 cucumber plants, *Cucumis sativum* L, in each net cage. A first egg release showed no significant difference between the two methods. An approximately 80-84% reduction in population density of aphids was observed compared with a control treatment. However, a second egg release, 7 days after the first release, caused a 94-95% reduction in aphid populations. Better control of aphids was observed with repeated egg release. The two methods had the same effect in controlling aphids in experiments conducted in cages without generalist egg predators (such as ants). Although the two methods had the same efficacy in the absence of ants, if eggs are released in greenhouses and fields, the Chrysobag would protect eggs from ants and other predators. Therefore, the Chrysobag is a more effective method, with a high efficiency and without the negative effect that sawdust has on host plants. We suggest that the Chrysobag method should be used in commercial greenhouses and in the field.

INFLUENCE OF LIGHT ON THE EFFICACY OF BIOCONTROL AGENTS IN GREENHOUSE ENVIRONMENTS

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74 A key issue affecting adoption of biological control in protected culture is the variability in effectiveness of natural enemies. Growers need natural enemies to provide predictable and reliable control of a pest. Predictable biological control requires that we have an understanding of how environmental conditions affect both the pest and biocontrol agent. Greenhouses are unique in that environmental conditions can be manipulated. The greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) (Homoptera: Aleyrodidae) and western flower thrips, *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) are important pests of greenhouse vegetable crops worldwide. It has long been recognized that the effectiveness of biological agents in the winter in northern temperature regions is reduced. Short day length, low light intensity and low temperatures have been suggested as possible explanations for this reduced efficacy. Little information is known about the influence of light on the behaviour of natural enemies. We present results from studies investigating the influence of light quality (intensity) and light quantity (day length) on the feeding and oviposition activity of: 1) two aphelinid parasitoids, *Encarsia formosa* Gahan and *Eretmocerus eremicus* Rose and Zolnerowich (Hymenoptera: Aphelinidae) on greenhouse whitefly and, 2) a predacious mite, *Neoseiulus cucumeris* (Oudemans) (Acarina: Phytoseiidae) on western flower thrips. Results are put in context of greenhouse management in Canada.

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THE INFLUENCE OF FORMULATION AND PACKAGING ON PERFORMANCE OF BENEFICIAL ARTHROPODS

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Research into biological control agents often focuses on the characteristics of the species being researched, such as intrinsic rates of increase, predation rates and host range. One to one comparisons in laboratory or semi-field situations will show that a novel species may be superior to one already in use. This approach fails to account for various factors which will influence the commercial production and use of an agent. Excellent predators may be impossible or expensive to produce in sufficient quantities, and so fail to displace weaker predators which can be economically mass produced. The formulation of products and the way they are presented in crops can turn a relatively weak predator into a viable means of control. Changes in formulation and product design can further improve performance and extend the range of crop situations in which a given product can be used. Whilst the species used is a key component of a product, and can be regarded as the active ingredient, it is by no means the only element which defines performance.

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ESTIMATING PARASITISM LEVELS IN *OSTRINIA NUBILALIS* HÜBNER (LEPIDOPTERA: CRAMBIDAE) FIELD POPULATIONS USING MOLECULAR TECHNIQUES

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Accurate detection and identification of parasitoids are critical to the success of IPM programs to detect unusual variations of the density of these natural enemies, which may follow changes in agricultural practices. For such purposes specific molecular markers to detect *Lydella thompsoni* (Herting) and *Pseudoperichaeta nigrolineata* (Walker) (Diptera: Tachinidae) within the european corn borer, *Ostrinia nubilalis* (Hübner) (Lepidoptera: Crambidae) have been developed. Primers amplifying fragments of the mitochondrial cytochrome oxidase I (COI) gene were designed following alignment of comparable sequences for a range of parasitoid and host species. Each of the primer pairs proved to be species-specific to one of those tachinid species, amplifying DNA fragments of 191 and 91 bp in length for *L. thompsoni* and *P. nigrolineata*, respectively. This DNA-based technique allowed to perform a molecular detection of parasitism in natural populations of *O. nubilalis*.

Molecular evaluation of parasitism was compared with the traditional method of rearing ECB populations in controlled conditions before breaking off the diapause. This method is both labour intensive and slow, taking easily up to 8 months, too long to be compatible with

effective modifications of cultural practices that would be required if unusual variations of densities of parasitoids are detected. Fifth-instars diapausing ECB were collected in maize stalks at twelve sites in three geographical areas of France: three in the South, three in the center and six in the North. For each site, diapausing larvae were randomly assigned in two sets. The first set (fifty larvae per population) was used to estimate the mean percentage of parasitism using this molecular method. The second set was used to estimate the mean percentage of parasitism using the traditional rearing method – i.e., by recording the number of tachinid flies emerged from the ECB larvae. This set contained between 76 and 217 diapausing larvae depending on the population. Obtained percentages of parasitism of both tachinid species were higher – approximately three times – using this molecular method, suggesting an underestimation of the traditional rearing protocol. This study confirms molecular methods as very promising for a correct detection and identification of parasitoids in natural field populations.

Session 12: Environmental Risk Assessment of Invertebrate Biological Control Agents

RISK-ASSESSMENT IN OMNIVOROUS PREDATORS

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The fact that many predaceous arthropods can occasionally or regularly feed on plants or plant products is of some concern as if it would inevitably lead to crop damage. However, facultative herbivory does not necessarily cause economic loss in crops. Economic loss is the result of complex interactions between the morphological, physiological and behavioural traits of the predator and environmental features including crop plants. In this poster we summarize the complexity of plant feeding by predators when drawing guidelines for risk assessment of introducing arthropod biological control agents into a new environment.

EVALUATING THE RISKS TO NON-TARGET SPECIES ASSOCIATED WITH INTRODUCING A STAPHYLINID PARASITOID INTO THE CANADIAN PRAIRIES

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78 The Eurasian parasitoid *Aleochara bipustulata* L. (Coleoptera: Staphylinidae) is a prospective classical biological control agent for the cabbage root maggot, *Delia radicum* (L.) (Diptera: Anthomyiidae) in Canada. The pest feeds on the roots of *Brassica* plants, and is a growing concern to Canadian canola producers. Prior to introduction, the risks of negative effects on population levels of non-target species will be evaluated by studying the parasitoid's fundamental and ecological host range.

Contemporary protocols for studies on host range of a prospective biological control agent recommend beginning with a review of the literature. The literature review revealed the habitats in which the parasitoid has been found and the host range of the prospective agent and two congeneric species. Researchers have found the parasitoid in fifteen different habitats, and complete development reported within puparia of fly species from seven families of Diptera Cyclorhapha. This information was used to compile a list of dipteran species that both overlap with the habitats of the parasitoid and may support complete development. Species of conservation concern and beneficial species were appended as safeguards. This list was then filtered to include only those species likely to be found in sufficient numbers to permit rigorous testing. This filtered list of non-target species and the habitats reported for the parasitoid in the literature were used to develop a programme for evaluating the risk to non-target species.

Since some reports of habitat use may have been based on improper identification of the parasitoid, pitfall traps will be used to determine if it does in fact exploit some of these equivocal habitats. Collections will be made from natural populations of species on the filtered list to assess the parasitoid's ecological host range. Finally, lab-based studies using species from the filtered list will be used to investigate the parasitoid's fundamental host range.

 Session 12: Environmental Risk Assessment of Invertebrate Biological Control Agents

EVALUATING PARASITOID HOST RANGE USING MOLECULAR TECHNIQUES: A NEW APPROACH TO NON-TARGET RISK ASSESSMENT STUDIES

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Accurate identification of natural enemies is the cornerstone of biological control and methods that can accurately identify potential biological control agents are essential, particularly when morphological variation among species is slight. Conventional rearing and dissection methods for detecting and identifying parasitoids within their hosts can be tedious and time-consuming; however, these techniques are commonly used in host range and risk assessment studies. Molecular methods for detecting and identifying parasitoids within their hosts have the potential to be both rapid and accurate, and may expedite the ecological studies necessary prior to introducing potential biological control agents. In order to determine the utility of molecular diagnostics in risk assessment studies on European parasitoids of *Lygus* Hahn (Hemiptera: Miridae), a single-step multiplex PCR assay was developed for three *Peristenus* Förster (Hymenoptera: Braconidae) species. The availability of a single-step multiplex PCR assay to detect and identify immature stages of *Peristenus* species within mirid nymphs may facilitate host range and non-target risk assessment studies by eliminating mortality issues encountered using rearing techniques, expediting identification, and providing additional information not available by dissection (e.g. species composition) or rearing (e.g. multiparasitism). The specificity and sensitivity of this assay were tested, and the PCR primers were shown to be highly specific for their respective species and capable of detecting single parasitoid eggs within parasitized *Lygus* nymphs. The single-step multiplex PCR assay was applied to DNA extracted from field-collected target and non-target mirid nymphs from different habitats in northern Germany, and parasitism levels and parasitoid species composition based on rearing, dissection and molecular analysis were compared.

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ASSESSMENT OF SPECIES COMPOSITION OF NATIVE *ORIOUS* SPECIES IN JAPAN USING MOLECULAR MARKERS

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The anthocorid bugs, *Orius* spp. (Heteroptera: Anthocoridae), are the most promising biocontrol agents of various minute insect pests, such as thrips. Five species in the genus are commonly distributed in Japan: *Orius sauteri* (Poppius), *O. minutus* (L.), *O. strigicollis* (Poppius), *O. nagaii* Yasunaga, and *O. tantillus* (Motschulsky). *Orius strigicollis* are mainly used commercially for the control of thrips in greenhouses in Japan. However, conservation ecologists are apprehensive that composition of species might be disturbed by the augmentative release. In this study, we developed molecular markers to identify species easily, and surveyed the species composition of *Orius* communities in the field.

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Development of molecular markers to identify *Orius* species. By multiplex-PCR techniques using five primers simultaneously, we could identify the five *Orius* species easily. The DNA region amplified was Internal Transcribed Spacer 1 (ITS1) of nuclear ribosomal RNA gene.

Species composition of *Orius* communities. Because *O. strigicollis* has been used in greenhouses throughout Japan recently, released individuals may disperse to the surrounding area. By PCR assessment on over 30 communities however, the species could not be found in the areas where they had not been reported (Fig. 1). This suggests that released *O. strigicollis* has not yet been established under natural conditions.

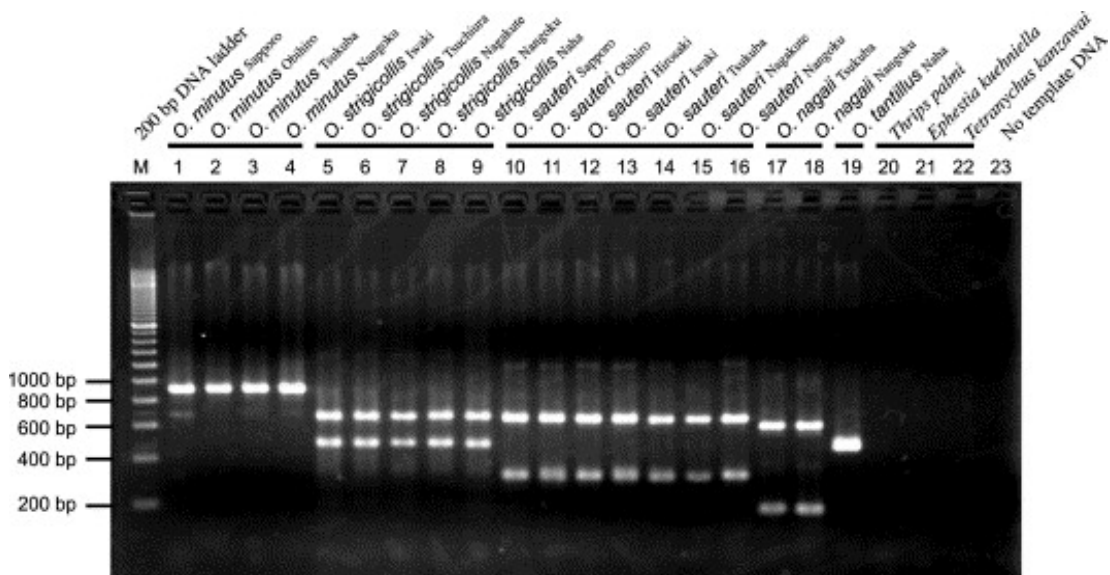


Figure 1. Electrophoretogram of multiplex PCR of five *Orius* species and three prey species. Samples were electrophoresed on 1.6% agarose gels.

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RISK ASSESSMENT OF A POTENTIAL BIOLOGICAL CONTROL AGENT OF CHERRY BARK TORTRIX

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Cherry Bark Tortrix (CBT), *Enarmonia formosana* Scopoli (Lepidoptera: Tortricidae), is a bark-boring pest of several tree species, including *Prunus*, *Malus* and *Pyrus* (Rosales: Rosaceae). Since its accidental introduction from Europe into North America, its high densities pose a threat to nursery and orchard industries as it spreads across British Columbia, Washington State and Oregon State. CBT population levels are consistently low across Europe indicating that this pest has a suite of natural enemies in its area of origin. It was reported that the parasitoid species, *Campoplex dubitator* Horstmann (Hymenoptera: Ichneumonidae), in particular appears to have a substantial impact on European CBT populations and was therefore proposed as a suitable candidate for introduction into North America for the biological control of invasive CBT.

Current regulatory procedures in Canada state that biological control agents must undergo risk assessment prior to their introduction in order to verify minimal risk to non-targets. A new initiative was therefore set up to specifically assess the ecological and physiological host range of *C. dubitator* and to assess potential non-target effects. Experiments were designed according to recently developed guidelines for the risk assessment of biological control agents of arthropod pests and initial tests have been conducted to investigate *C. dubitator* acceptance of selected orchard tortricid species.

It has been indicated in the past that *C. dubitator* shows a strong response to CBT frass and potentially uses it for host location. Preliminary experiments have therefore also been performed with *C. dubitator* to investigate the potential role this indirect cue may play in parasitism of non-target species occupying the same niche as CBT.

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HISTORY AND FUTURE OF INTRODUCTION OF EXOTIC ARTHROPOD BIOLOGICAL CONTROL AGENTS IN SPAIN: A DILEMMA?

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The first documented introduction of an exotic invertebrate biological control agent (IBCA) in Spain occurred in 1908. Since then, 69 additional species have been introduced. We summarize information, both previously recorded and original data, on the species introduced for pest control. Most of the introduced IBCA's focused on citrus pests, and homopterans clearly predominate among target phytophagous species. Success has been more frequent for natural enemies introduced in seasonal inoculative strategies (56.0% of cases) than for those used in classical biological control programs (19.0% of cases). Concerns about potential non-target effects of such species are increasing, but post-release evaluation has often been insufficient to draw conclusions about non-target effects. Most of the beneficial species introduced into Spain were parasitoids (n = 59), and the remaining species (n = 10) were predators. Only 4 of parasitoids are considered specialized monophagous natural enemies. The mean number of host species parasitized by parasitoids is 15.5, whereas the mean number of prey species attacked by predators is 21.2. So, polyphagy seems quite common in the introduced IBCA's in Spain. The rationale guiding many of these introductions in the past would not be acceptable nowadays. Because biological control is such a valuable strategy for pest control, straightforward protocols to evaluate exotic candidate species are urgently needed.

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CAN A PATHOGEN CONSTRAIN THE HOST SPECIFICITY OF A BIOLOGICAL CONTROL ORGANISM?

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Biocontrol is a powerful technology for reducing harm caused by invasive species. It is neither a panacea nor risk-free. Investigators have recently warned that arthropods introduced for biological control of weeds might be harmful to native, nontarget plants, the environment, and public health. Here we ask whether biological control of an arthropod introduced for biological weed control can mitigate the harm it causes to native, nontarget plants. Our case study centers on the decisions to introduce and redistribute the European cinnabar moth *Tyria jacobaea* L. (Lepidoptera: Arctiidae) in North America for biological control of ragwort *Senecio jacobaea* L. (Asterales: Asteraceae). First we show that introduction and redistribution of the cinnabar moth in North America has increased the incidence of herbivory on native, nontarget plants. Second, infection by the microsporidian *Nosema tyriae* reduces the performance (growth, development, survival, pupal mass) of cinnabar moth on test plants in the laboratory. We plan further studies of the incidence and severity of disease in cinnabar moth populations as possible constraints on host use in the field. Finally, we use an epidemiologic approach (spanning 25 sites and 3 years) to show how use of native host plants by this insect is constrained by abiotic conditions, quantity and quality host resources, other specialist and generalist herbivores, as well as natural enemies (*Nosema*). We conclude that biological control introductions are generally irreversible, but it may be possible to mitigate their harmful side-effects.

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ASSESSING PARASITOID HOST RANGES OF CLOSELY RELATED *CEUTORHYNCHINAE* TO AVOID CONFLICTS BETWEEN WEED AND ARTHROPOD BIOLOGICAL CONTROL

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Classical biological control of insect pests and weeds can lead to potential conflicts, particularly if the insect pest and weed agents are closely related. Such a conflict may occur in biological control of the cabbage seedpod weevil, *Ceutorhynchus obstrictus* Marsham (Coleoptera: Curculionidae), which belongs to the same subfamily, *Ceutorhynchinae*, as a number of agents introduced or proposed for introduction against invasive alien weed species in North America. The cabbage seedpod weevil is a serious pest of canola and rapeseed (*Brassica napus* L. and *Brassica rapa* L., respectively) in North America. As the future availability of insecticides is uncertain, there is a critical need to more effectively utilize biological control. Several hymenopteran parasitoids of cabbage seedpod weevil are known from Europe. Some of those European parasitoids such as *Trichomalus perfectus* Wlk. (Hymenoptera; Pteromalidae) and *Mesopolobus morys* Wlk. (Hymenoptera; Pteromalidae) show greatest potential for incorporation into an integrated pest management system for cabbage seedpod weevil in North America. Prior to importation, the host specificity of candidate European parasitoids has to be determined in their native cultivated and non-cultivated habitats to estimate potential non-target risks. The aim of this study is to assess host plant – *Ceutorhynchinae* – parasitoid associations and time of occurrence of *Ceutorhynchinae* and their parasitoids in the fields. This will help to determine potential non-target risks to other *Ceutorhynchinae* implemented as weed biological control agents in North America. A survey in selected crop and non-crop habitats for potential non target insect hosts of *C. obstrictus* parasitoids was conducted at sites in Germany, Switzerland, France, Austria and Hungary between 2002 and 2004. Results of the field surveys as well as parasitoid complexes associated with each selected non-target host will be presented. Results show that *T. perfectus* would probably not attack any native species, or introduced or candidate biological control agent. However, *M. morys* was found parasitizing species such as *Ceutorhynchus turbatus* Schltz., which is a candidate biological control agent of Whitetop (*Lepidium draba* (L.) Desv.). Further studies are being carried out to understand how these results might affect the future decisions in term of introduction of candidate biological control agents into Canada.

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RETROSPECTIVE STUDIES TO ASSESS THE IMPACT OF NATURAL ENEMIES USED FOR CONTROL OF HIBISCUS MEALYBUG, *MACONELLYCOCCUS HIRSUTUS* (HOMOPTERA: PSEUDOCOCCIDAE), ON NON-TARGET ORGANISMS

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The hibiscus mealybug has since spread to most islands of the Caribbean and has also been reported in northern Mexico, Belize, Central America and southern California. In most of these countries the HMB has also been kept under control by *A. kamali* and *C. montrouzieri*. A second exotic parasitic wasp *Gyranusoidea indica* Shafee, Alam and Agarwal (Hymenoptera: Encyrtidae) was released in some islands in the Caribbean region to control the HMB. It was introduced in St. Kitts and Nevis, Puerto Rico, Grenada and the United States Virgin Islands through the USDA-APHIS programme. *G. indica* was not officially released in Trinidad, however it was found in HMB infested plant material in Trinidad in the Las Lomas, Arima and St. Augustine areas in early 2000. CABI Bioscience confirmed its identification. At present its distribution is more widespread. The presence of *G. indica* in this country will certainly impact on the HMB Control Programme of the Ministry of Agriculture. Classical biological control is claimed to provide the basis for an environmentally sound solution to pest problems. Thus the general assumption is that there is no risk (non-target effect) associated with biological control since nature is being used to fight nature. Adequate data to defend this has not been systematically gathered. To this end a study has been undertaken in post release monitoring of these natural enemies to determine their efficacy, interactions and impact on non-target species.

The objectives of this study are the determination of the:

1. Impact, if any, of introduced natural enemies on non-target species (Mealybugs) and
2. In the process other Mealybug fauna and their Natural Enemies would be determined.

A general field survey has already been conducted in Trinidad to determine the natural enemies of the local mealybugs. Then this will influence the design of the specific field surveys that are to be conducted to determine the impact of the exotic natural enemies on non-target organisms.

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MOLECULAR DETECTION OF PREDATION BY SOIL MICROARTHROPODS ON NEMATODE BIOPESTICIDES

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Parasitic nematodes have been used as biological control agents on a number of different pests in recent years, including successful control of vine weevils, chafer larvae, leatherjackets, Japanese beetles and slugs. Although effective biological control agents, a number of factors have inhibited the widespread use of nematode biopesticides in agriculture and horticulture. One of these factors has in some cases been the poor persistence of the dauer larvae in the soil soon after application. The persistence of nematode biopesticides in the soil has a large impact on both the short and long term effectiveness of nematodes as biological pest control agents. Although UV light and desiccation are commonly attributed to being the main cause of decline in viable nematode numbers, predation by micro-arthropods in the soil may also play a role. Two of the most common micro-arthropods encountered in soils are mites and collembolans.

Our aim was to develop a set of species-specific DNA primers for the detection of three common parasitic nematodes utilized in biological control, *Phasmarhabditis hermaphrodita* Schneider (Nematoda: Rhabditidae), *Steinernema feltiae* Filipjev (Nematoda: Steinernematidae) and *Heterorhabditis megidis* (Poinar) (Nematoda: Heterorhabditidae), and test their ability to detect the presence of these nematodes in the guts of micro-arthropod

predators. A 510-bp fragment of mitochondrial CO1 DNA was amplified from each species using general primers and sequenced. From this information, species-specific primers were designed and tested for specificity against a range of non-target organisms.

Feeding trials were conducted using two model organisms representing the micro-arthropod predators, the collembolan *Folsomia candida* (Willem) (Collembola: Isotomidae) and the mesostigmatid mite *Stratiolaelaps miles* (Berlese) (Acari: Laelapidae). After exposure to the nematode prey, DNA from the whole micro-arthropods, including the gut contents, was extracted. Detection of nematode DNA within the gut of both micro-arthropods was possible for a maximum of 12 hours with a half life between 8.1 and 10.2 hours in the guts of *F. candida*. Prey choice trials were then conducted with *F. candida* on soil, in which they were offered three nematode species simultaneously at a range of nematode densities.

The use of PCR and species-specific primers has enabled detection and quantification of predation on scales that were not possible using traditional techniques. Soil ecosystems contain many cryptic food chain links and direct observations of such interactions within the soil are not feasible. The study has proved that predation by soil micro-arthropods on nematode biopesticides can occur in semi-natural conditions and that it is possible they may be a limiting factor on nematode biopesticide persistence in soils.

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ENVIRONMENTAL FACTORS INFLUENCING THE INFILTRATION OF INTRODUCED BIOLOGICAL CONTROL AGENTS INTO INDIGENOUS HABITATS

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During recent years there has been an increase in concern regarding non-target impacts of purposely introduced biocontrol agents on native and desirable species. Considerable effort is being expended to understand the causes of host range expansion. Less effort has been made to address reasons for habitat range expansion. There is a need to elucidate the causes of infiltration of native habitats and the potential disturbance of the community of native arthropods.

Environmental factors influencing parasitoid community and parasitism levels. This study aims to identify key environmental factors that might influence the parasitoid community and parasitism levels by biocontrol agents and adventive parasitoids in remote native habitats in Hawai'i. Two endemic non target insect species, *Udea stellata* (Butler) (Lepidoptera: Crambidae), and *Spheterista infausta* (Walsingham) (Lepidoptera: Tortricidae) are being investigated to quantify the level of parasitism by exotic parasitoids and to explore the effects of environmental variables on the occurrence of the parasitoids. *Pipturus* spp. (Urticales: Urticaceae), are the host plants of these insects. These endemic plant species are distributed across a wide range of habitats in Hawai'i, giving the opportunity to investigate various environmental gradients.

CANOCO analysis. We used canonical correspondence analysis (CANOCO) to analyze community structure across gradients (such as elevation, level of disturbance, host plant density, plant species richness, etc). Adventive parasitoids occurred across all environmental gradients, and were most strongly associated with moderately disturbed habitats. Purposely introduced parasitoids were frequently associated with the most pristine and remote habitats. Further analyses are to be conducted to elucidate which environmental factors determine these differences. The results of this study will contribute to the development of risk assessment models for assessing non-target risks posed by potential biocontrol introductions, and will contribute to providing tools to identify habitats that are particularly susceptible to alien parasitoids.

Session 13: Predicting Natural Enemy Host Ranges: Strengths and Limitations of Lab Assays

STRATEGIES FOR EVALUATING NON-TARGET EFFECTS IN ARTHROPOD BIOLOGICAL CONTROL

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Examining possible non-target effects of biological control agents is becoming a more common requirement for many biological control programs targeting arthropod pests. Currently, for classical biological control of weeds, the Wapshere method provides an excellent means for eliminating possible natural enemies that could cause harm to non-target plants. However a rigorous, reliable, and broadly applicable testing standard for arthropod biological control is currently lacking. No-choice and choice testing strategies are a common way to

test for possible non-target effects of new biological control organisms. However, these lab studies are often carried out in small testing arenas where the study organism is forced onto the host which may be adequate for determining physiological host range but may seriously overestimate its ecological host range in nature. Our research involves the use of rigorous testing strategies utilizing standard Petri dish test arenas, coupled with larger-scale entire plant test arenas in no-choice and choice comparisons. As retrospective studies in ongoing biological control programs can yield valuable information on non-target impacts, we chose the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata* Say (Hemiptera: Cicadellidae), classical biological control program in California as model for our non-target studies. We are examining the possible non-target impacts of the self-introduced *Gonatocerus ashmeadi* Girault and the recently introduced *G. fasciatus* Girault (Hymenoptera: Mymaridae), egg-parasitoids of the GWSS, and three sharpshooters native to California, U.S.A.: (1) the smoke-tree sharpshooter (STSS), *Homalodisca liturata* Ball; (2) blue-green sharpshooter (BGSS), *Graphocephala atropunctata* (Signoret); and (3) green sharpshooter (GSS), *Draeculocephala minerva* Ball (all Hemiptera: Cicadellidae). Our study, along with the use of small-scale Petri dish studies and larger-scale full plant studies are supplemented with sentinel plants and habitat surveys to determine the invasiveness of GWSS parasitoids. Since very little is known regarding the native sharpshooters and their native natural enemies, this research, in part, also focuses on surveying and classifying the native parasitoid fauna of the native sharpshooters. Two parasitoids, *Gonatocerus latipennis* Girault and a *Polynema* sp. (Hymenoptera: Mymaridae) were reared from field collected BGSS eggs and sentinel plants. Reciprocal tests of these two egg-parasitoids confirmed their associations with BGSS eggs. Additional reciprocal tests of the *Polynema* sp. yielded no parasitism of GWSS and STSS eggs. Collectively the *Polynema* sp. and *G. latipennis* constitute the first documented parasitic natural enemies of BGSS eggs.

Session 13: Predicting Natural Enemy Host Ranges: Strengths and Limitations of Lab Assays

BIOLOGICAL CONTROL OF OLIVE FLY: BALANCING PARASITOID EFFECTIVENESS AGAINST NON-TARGET IMPACTS

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Olive fly (OLF), *Bactrocera oleae* (Diptera: Tephritidae), has become an important pest of California olives, and the target of a classical biological control program. We report here on the pre-release screening of imported parasitoids (Hymenoptera: Braconidae), conducted in the University of California, Berkeley, Quarantine Facility. Tested material included *Psytallia concolor* (Szépligeti) "Tunisia," *Diachasmimorpha kraussii* (Fullaway), and *D. longicaudata* (Ashmead) (Diptera: Tephritidae), which are known generalist parasitoids that were originally derived from insectary colonies using the Mediterranean fruitfly as host material. We also studied six suspected OLF specialists, which were originally reared from OLF that were field collected in Africa or Pakistan, these are: *Bracon celer* Szépligeti (Hymenoptera: Braconidae), *Psytallia concolor* "Kenya," *Psytallia lounsburyi*, *Psytallia* sp. nr *concolor* "South Africa," *Utetes africanus* (Szépligeti), and *Psytallia ponerophaga*.

Non-target studies. We conducted non-target studies to determine the potential host range of each parasitoid species. Species of native and exotic tephritids used for non-target impact studies include: *Rhagoletis indifferens* (western cherry fruit fly), *Rhagoletis pomonella* (apple maggot), *Euphranta canadensis* (currant fly), *Chaetorellia succinea* (yellow star thistle fly), *Euaesta aequalis* (cocklebur fly), and *Parafrentreta regalis* (Cape ivy fly). Priority was given to *C. succinea* and *P. regalis*, which are beneficial tephritids used to control weeds in the U.S.A.

Generalist parasitoids. All tested species except *P. lounsburyi* and *U. africanus* were generalists, attacking OLF and either the non-target beneficial or endemic tephritid species.

Parasitoid biology. To help assess the field-potential of each imported parasitoid species to suppress OLF, we conducted laboratory studies to determine the parasitoids' basic biological parameters when reared on OLF.

Potential parasitoid effectiveness. We found that the generalists (*D. kraussii*, *D. longicaudata*, *B. celer*, and the *P. concolor*-complex) were often the more aggressive and effective OLF parasitoids under laboratory conditions. Of the probably specialists, *P. lounsburyi* was relatively effective parasitoid, while *U. africanus* proved to be difficult to rear and, therefore, quarantine-screening remains incomplete for this species.

Quarantine-screening impact on biological control. Based on these results, field tests will be conducted initially with *P. lounsburyi*. Further foreign exploration and importation will continue. Our cross-mating experiments with *P. concolor* found populations that exhibit variation in phenological color patterns may be conspecifics, suggesting that further taxonomic information is needed to better predict its impact in biological control programs. Many of the generalist species attacked beneficial parasitoids (*C. succinea* and *P. regalis*) and we use these data to discuss potential non-target impacts from large regional projects on the conservation of endemic fauna or disruption of biological control.

HOST RANGES OF SIX SOLITARY FILTH FLY PARASITIDS (HYMENOPTERA: PTEROMALIDAE, CHALCIDIDAE) FROM FLORIDA, EURASIA, MOROCCO, AND BRAZIL

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Pupal parasitoids are among the most important and common natural enemies of filth flies associated with animals and humans. Augmentative releases of parasitoids can provide satisfactory suppression of fly populations, but in many cases parasitoid releases have had little impact on fly populations or parasitism levels. Although the factors that determine success of augmentative releases are unclear, failures may be due to an imperfect understanding of how to match candidate parasitoid species with target pests and breeding conditions. A better understanding of niche characteristics of parasitoid species could assist in the process of matching parasitoid species with release sites. Different parasitoid species have been evaluated in recent years with regard to temperature-dependent attack rates, development time, manure moisture preferences, and the effect of habitat type and depth on foraging behavior. Host range is another niche characteristic that may vary among species of fly parasitoids. Most of the available literature on muscoid fly parasitoids has involved trials with house flies and stable flies. By comparison, information on parasitism of other species such as the horn fly (*Haematobia irritans* (L.) (Diptera: Muscidae)) is limited, and mostly deals with field surveys of flies in various locations. The objectives of this present study were to compare parasitoid attack rates and progeny production on different host species and to determine whether there were distinct differences among parasitoid strains from different locations.

Attack rates, progeny production, sex ratios and host utilization efficiency of *Muscidufurax raptor* Girault and Sanders, *Spalangia cameroni* Perkins, *S. endius* (Walker), *S. nigroaenea* Curtis, *S. gemina* Boucek (Hymenoptera: Pteromalidae) and *Dirhinus himalayanus* (Hymenoptera: Chalcididae) were evaluated in laboratory bioassays with five dipteran hosts: house fly (*Musca domestica* L.), Stable fly (*Stomoxys calcitrans* L.), horn fly (*Haematobia*

irritans (L.), black dump fly (*Hydrotaea aenescens* (Weidemann)) (Diptera: Muscidae) and a flesh fly (*Sarcophaga bullata* Parker) (Diptera: Sarcophagidae). *M. raptor*, *S. cameroni*, and *S. endius* readily attacked and produced progeny on all five host species, with substantially lower production from *S. bullata* than from the muscid hosts. Rates of host attacks by *S. nigroaenea* and *S. gemina* were similar on house fly, stable fly and black dump fly hosts, with lower rates on horn fly; almost no progeny were produced by *S. nigroaenea* on *S. bullata* hosts. *D. himalayanus*, a large-bodied chalcidid parasitoid, had highest rates of host attacks and progeny production on *S. bullata* and *H. aenescens*, followed by stable fly and house fly hosts; very few progeny were produced by this species on horn fly hosts. Overall differences among different geographic strains of parasitoids (from Russia, Kazakhstan and Florida) were generally small, although the Florida strain of *M. raptor* was superior to the two Eurasian strains. The results with horn fly hosts support field survey data suggesting that several of these species may be promising biological control agents for this pest.

Session 13: Predicting Natural Enemy Host Ranges: Strengths and Limitations of Lab Assays

EVALUATION OF THE EFFICIENCY OF AN ECOPARASITOID OF CYDIA SPECIES – IMPORTANCE OF LAB BIOASSAYS!

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The oriental fruit moth (*Cydia molesta* (Bousck) and the codling moth *Cydia pomonella* L. (Lepidoptera: Tortricidae)) are important pests in apple orchards. Since current control methods are not always successful in controlling these pests, alternative solutions are needed. The use of antagonists as biological control agents might be a suitable approach to complement existing IPM strategies, particularly in regard to pest population reduction and anti-resistance strategies.

The parasitic wasp *Hyssopus pallidus* (Askew) (Hymenoptera: Eulophidae) is a gregarious ectoparasitoid of larval instars of *Cydia* species (*C. pomonella* & *C. molesta*). Caterpillars are parasitized inside the apple and larvae develop by feeding on the caterpillar's body, resulting in the death of the caterpillar. Thus, reduction of primary *Cyda* infestation in early summer could help to prevent heavy infestation later in the season.

In this study, comparative life history traits of *H. pallidus* reared on the two different *Cydia* sp. was assessed and evaluated in regard to the optimal condition for effective mass rearing systems. The implications for biological control are discussed. Furthermore, behavioural aspects of the host location mechanism and parasitism success in mixed infestations are elucidated in order to optimize field release. Accomplished parasitism rate will help to exploit the parasitoid's biocontrol potential in reducing *Cydia* population.

DO LABORATORY BIOASSAYS REFLECT ECOLOGICAL HOST RANGE AND PREDICT POST-INTRODUCTION NATURAL ENEMY HOST RANGE?

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Using the retrospective case study of *Peristenus digoneutis* Loan (Hymenoptera: Braconidae) introduced in the early 1980s in the United States for biological control of native *Lygus* plant bugs (Hemiptera: Miridae), laboratory assays and field studies in the area of origin were conducted to evaluate pre-introduction host range and to analyze if predictions would match actual post-introduction natural enemy host range. Twenty years after release in North America, the host range of *P. digoneutis* has been evaluated in its area of origin in Europe by using data from the literature, conducting laboratory assays to assess the fundamental host range as well as assessing the ecological host range based on field collections. Data from host range assessment in Europe were then compared to literature data from an eight years post-introduction study on the host range of *P. digoneutis* in the United States. Seven non-target mirid species in the area of origin in Europe were studied in the laboratory to define the fundamental host range of *P. digoneutis*. Selection criteria for non-target hosts included phylogenetic relatedness, habitat overlap, phenology and abundance in the field. Laboratory choice and sequential no-choice tests demonstrated that all non-target species studied were accepted by *P. digoneutis* and suitable for parasitoid development. However, only one out of the seven test species was accepted with the same frequency as the target host species, *Lygus rugulipennis* (Popp.). To confirm that the results from the laboratory host range assays were valid, the ecological host range of *P. digoneutis* in the area of origin was investigated by a three-year study of the parasitoid complexes of common mirids. *Peristenus digoneutis* was reared from ten hosts, including three *Lygus* species and seven other species in the subfamily Mirinae. When comparing the fundamental and ecological natural enemy host ranges, the fundamental host range of *P. digoneutis* did not completely match its ecological host range in Europe. The fundamental host range results indicated only that *P. digoneutis* would attack non-targets less often, but did not suggest the total absence of this parasitoid in field collections of two of the tested non-targets. However, the proportions of *P. digoneutis* relative to the total number of parasitoids emerging from field collection of each non-target host were less than 5%, a finding that corresponded with decreased preference in sequential no-choice and choice tests in the laboratory assays. Published findings of post-introduction host range assessments in the

United States corresponded with ecological host range findings in Europe, showing that despite the number of native *Lygus* species available only a few non-target host species were parasitized at a very low level in the northeastern United States. This case study shows that ecological host range field surveys in the area of origin are more important than fundamental host range laboratory assays for predicting post-introduction natural enemy host ranges.

Session 14: Legislation and Biological Control of Arthropods: Challenges and Opportunities

IMPORTATION AND EXPORTATION OF LIVING ORGANISMS FOR SCIENTIFIC PURPOSES: LEGISLATION AND REGULATIONS IN THE UE AND PROCEDURES IN USE AT EBCL, AN OVERSEAS USDA-ARS LABORATORY IN FRANCE (UE)

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Introduction of exotic pests into a territory must be controlled to prevent serious economic losses, drastic health problems, and associated environmental problems. Certain harmful organisms, plants, plant products and other objects, may be introduced into the European Union, for trial or scientific purposes, but only under specified conditions.

International conventions, European Union provisions, French legislative provisions, French regulation provisions and French administrative provisions provide obligations and govern the movement and the confinement of certain harmful organisms, plants, plant products and other objects in France.

Within its research activities, the European Biological Laboratory (EBCL) imports live organisms, macro and micro organisms and plants on the French territory. All of these species are imported according to the legislation and regulations in use at International level (ICPP, CITES, Code of Conduct), by the European Union (Directive 95/44/CE, Directive 2000/29/CE), and by France (Decree 2003-768, Ordinance of June 10, 1998, Ordinance of November 22, 2002). EBCL has two quarantines; the quarantine activity is approved by Prefectoral Ordinance, and each organism is imported after being declared to official plant protection authorities (French Regional Service of Plant Protection).

These organisms are imported by EBCL researchers from missions abroad, or by their foreign counterparts who send these organisms via an airfreight or express transport company. Importations of live materials are accompanied by documents such as the Official Letter of Authority (for regulated living material) or the submission of a Declaration of Intention of Importation of living organisms and other objects for trial or scientific purposes, delivered by the Director of the Regional Service of Plant Protection of Languedoc-Roussillon Region.

The living material exported via airfreight has no commercial value but it has an inestimable scientific value. Once sent to our cooperators via airfreight it is subject to repeated radiation and X-rays that can be harmful to the material. Consequences to the use of these treatments can be either a drastic reduction of the reproduction rate or sterility. These living organisms can also be affected by prolonged shipment delays due to 24 - 72 hours safety devices at each airport transit.

Since 2000 the EBCL laboratory holds a certificate of conformity mentioning the title of "Known Client" within the French Civil Aviation Management Department (DGAC) and it has implemented a safety program for abroad shipping of living organisms.

In 2002 due to the International context the legislation changed and EBCL implemented prescribed training programs and established a secure chain between EBCL and the airport. Concomitantly, a security program was delivered to the French authorities and in 2004 the EBCL Laboratory got a five year agreement as "Known Loader" from the French authorities. All the parcels containing beneficial insects for cooperators are qualified for airfreight.

Session 14: Legislation and Biological Control of Arthropods: Challenges and Opportunities

A DIGITAL COMMONS FOR BIOCONTROL: AN OPEN ACCESS CYBERINFRASTRUCTURE FOR ENHANCED REGULATORY DECISION MAKING

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This project will create and evaluate an open access information infrastructure to enhance regulation and research of arthropod biological control agents for agriculture. Cyberinfrastructures, or world wide web information infrastructures, are revolutionizing many branches of science. Biocontrol researchers, private industry practitioners, and public officials are hampered by constraints on access to behavioral and ecological knowledge about arthropods so that they can make informed decisions about their importation, rearing and release. A digital open access cyberinfrastructure will provide public agency scientists improved access to more comprehensive knowledge to make enhanced importation, quarantine, and regulatory decisions about arthropod biocontrol agents. This resource will also open up greater access to extant scientific knowledge for government scientists, and enhance and expedite decision making.

This project will assemble a team of biocontrol entomologists, computer scientists, and science & technology researchers to create an information infrastructure for biological control, tied to the U.S. National Biological Information Infrastructure (NBII). Entomologists at the University of California Center for Biological Control will provide expert knowledge necessary to make the infrastructure useful to researchers, practitioners and regulators, and make available in open access form previously unpublished databases. Computer scientists at the University of California – Davis Information Center for the Environment will provide a framework for semantic web technologies, drawing from their experience managing the California node of the NBII, and their invasive species open access digital infrastructure. Researchers at Santa Clara University's Center for Science, Technology & Society will facilitate collaboration between these research communities; organize outreach and training sessions for all stakeholders to stimulate greater understanding and use of the infrastructure; evaluate the use, impacts and benefits of this infrastructure; and convene an advisory team of scientists, practitioners, stakeholders, and regulators to ensure that as it develops it will meet their needs. This project will establish the first ever template for biological control cyberinfrastructure, and it will ultimately make this available for the International Organization of Biological Control (IOBC).

This project seeks to provide greater coherence for the biocontrol knowledge system for regulators and researchers; create an open access information source for biocontrol research of agricultural pests in California, which will stimulate greater international knowledge sharing about agricultural pests in Mediterranean climates; and facilitate the exchange of information through a cyberinfrastructure among government regulators, and biocontrol entomologists and practitioners. It seeks broader impacts through: the uploading of previously unavailable data being made openly accessible; the stimulation of greater interaction between the biological control regulation, research, and practitioner community in selected Mediterranean regions; the provision of more coherent and useful information to enhance regulatory decisions by public agency scientists; a partnership with the IOBC to facilitate international data sharing; and progress toward the ultimate goal of increasing the viability of biocontrol as a reduced risk pest control strategy.

No Designated Session Theme

**BIOLOGY OF *CIRROSPILUS INGENUUS* GAHAN
(HYMENOPTERA: EULOPHIDAE), AN ECTOPARASITOID OF THE
CITRUS LEAFMINER, *PHYLLOCNISTIS CITRELLA* STANTON
(LEPIDOPTERA: GRACILLARIIDAE) ON LEMON**

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The citrus leafminer (CLM), *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae) invaded the Jordan Valley in 1994 and was able to spread throughout Jordan within a few months of its arrival. It was the most common parasitoid from 1997 to 1999 in the Jordan Valley. An increase in the activity of *C. ingenuus* was observed in autumn and the highest number of emerged *C. ingenuus* adults was in November 1999. The reproductive capacity of *C. ingenuus* at three constant temperatures showed that this agent could be considered as a promising candidate for successful biological control of CLM.

Parasitoid biology. *C. ingenuus* is an ectoparasitoid that prefers the fourth instar larvae of CLM as revealed by the preliminary studies. Adults have uniform orange yellow color. Both males and females are similar in size of about 1227 μ m long and 470 μ m wide. Females are easily distinguished by the presence of a black ovipositor.

Longevity and pre-oviposition periods. Male longevity was significantly higher at 20°C than at 25°C and at 30°C. No significant differences were obtained between female lon-

gevity at 20°C and at 25°C, but female longevity was significantly higher at those two temperatures than at 30°C. No significant differences were obtained in the pre-oviposition period at 20, 25 and 30°C.

Fecundity and sex ratio. A female *C. ingenuus* paralyzed the leafminer larva first and then laid eggs beside or on the host. It was rare to observe two eggs per host and if this happened, only one of them completed its development. Significantly higher numbers of eggs were laid by female *C. ingenuus* at 25°C than that at 20°C and at 30°C. Percentage of emerged females was higher at 20 and 25°C than at 30°C.

Male longevity, female longevity, and pre-oviposition period (\pm SE) of *C. ingenuus* on CLM at three different temperatures

Temperature	Male longevity (days)	Female longevity (days)	Pre-oviposition period (days)	Female fecundity (eggs/ female)	Sex ratio Female: Male
20°C	7.56a \pm 0.32	17.20a \pm 0.52	2.95a \pm 0.12	35.00(10)#b \pm 1.15	1.38: 1 (19)
25°C	6.50b \pm 0.24	16.11a \pm 0.32	2.72a \pm 0.09	43.11(9)a \pm 0.99	1.36: 1 (26)
30°C	4.58c \pm 0.20	9.75b \pm 0.23	2.71a \pm 0.07	29.75(12)c \pm 0.89	1: 1.2 (22)

Means within columns with the same letter are not significantly different using LSD test at 5 % level. (Number tested in sample.)

Development periods. Eggs of *C. ingenuus* had protracted shape and transparent colour. The incubation period was significantly longer at 20°C than at 25°C and at 30°C. Larvae with white colour were seen beside or on the paralyzed host larvae. The development period of larva was significantly longer at 20°C than at 25 °C and at 30°C. Pupae were at first colourless, then light brown and finally black. The development period of pupa was significantly longer at 20°C than at 25°C and at 30°C.

Development periods of egg, larva, and pupa (\pm SE) of *C. ingenuus* at three different temperatures

Stage	Development period (days)		
	20°C	25°C	30°C
Egg	4.20(30)*a \pm 0.20	3.08(30)b \pm 0.07	2.32(30)c \pm 0.06
Larva	7.14(22)a \pm 0.11	6.41(270)b \pm 0.06	5.20(25)c \pm 0.10
Pupa	8.29(19)a \pm 0.10	7.33(26)b \pm 0.10	4.91(22)c \pm 0.08

* Values in parenthesis represents the number of the tested sample.

Means within rows with the same letter are not significantly different using LSD test at 5 % level.

No Designated Session Theme

INTERACTIONS AMONG BIOCONTROL AGENTS OF THE BALSAM GALL MIDGE, *PARADIPLOSION TUMIFEX*, ON CHRISTMAS TREES

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With a production cycle of approximately 10 years, balsam firs planted as Christmas trees constitute a semi-natural, dynamic agroecosystem that is highly suitable for biological control. The balsam gall midge *Paradiplosis tumifex* Gagné (Diptera: Cecidomyiidae) is a major pest of balsam fir foliage, causing aesthetic damages. The pest exhibits cyclic outbreaks lasting 3-5 years, which are likely driven by foliage suitability to insect infestation interacting with pest mortality inflicted by natural enemies. We documented the impact of natural biological control of the gall midge during its recent outbreak in southwestern Québec, Canada. Field data collected on half grown to mature Christmas trees in three different localities in 2002 revealed 20-60% incidence of usurpation of *P. tumifex* galls by the inquiline midge *Dasineura balsamicola* Lintner (Diptera: Cecidomyiidae). This species cannot induce its own gall on fir needles, but invades and takes control of *P. tumifex* galls once developed and nutritionally functional, thus eliminating the initial gall resident. We also documented the incidence and phenology of a complex guild of endo- and ecto-parasitoids of the balsam fir midge, which in combination with *D. balsamicola* likely caused gall midge populations to crash to near zero levels in 2003. We clarified the taxonomic identity of the predominant parasitoids of the balsam gall midge system, and their host relationships in the gall.

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No Designated Session Theme

STUDIES OF THE RED IMPORTED FIRE ANT, *SOLENOPSIS INVICTA*, INFECTED WITH THE MICROSPORIDIA *VAIRIMORPHA INVICTAE* AND *THELOHANIA SOLENOPSAE* IN ARGENTINA

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After their accidental introduction into the United States more than 80 years ago, the red and black imported fire ants, *Solenopsis invicta* Buren and *Solenopsis richteri* Forel (Hymenoptera: Formicidae) became aggressive pests for people, domestic animals, agriculture, and wildlife. A classical biological control approach has been considered since the 1970's and emphasized since 1988 with an intensive search of biological control agents in South America. The pathogens *Vairimorpha invictae* Jouvenaz and Ellis (Microsporidia: Burenellidae) and *Thelohania solenopsae* Knell, Allen and Hazard (Microsporidia: Thelohaniidae) are obligate intracellular microorganisms specific to fire ants. They were originally discovered in *S. invicta* in Mato Grosso, Brazil, and later found in other species of South American fire ants such as *S. richteri*, *S. quinquecuspis* Forel and *S. macdonaghi* Santschi. They are important components of the complex of natural enemies that attack fire ants in South America; however, their effect on red imported fire ant populations in their native land was never reported. Thus, the objective of this study was to document the long-term effects of both pathogens on field populations of the red imported fire ant. Fire ant populations and microsporidia prevalence were monitored 3-5 times per year for 4 years in eight field plots in northern Argentina. The mean population index per plot showed an overall reduction of 69%. The percentage of infection with *V. invictae* and *T. solenopsae* showed fluctuations that ranged from 29.2 to 1.4% and 13.6 to 2.6% respectively. The highest infection rates were observed at the beginning of the study. A total of 394 colonies were sampled during the study, 325 (82.5%) were healthy and 69 (17.5%) were infected with microsporidia. The proportion of infected colonies with brood was 81% (56/69), similar to the proportion of healthy colonies with brood, 78% (255/325). The proportion of infected and healthy colonies in the population index categories was significantly different. Of the infected colonies with brood, 49.3% was medium and 1.4% was large in size. In contrast, healthy colonies were larger, with 29.7 and 10.4% being medium and large, respectively. The general environmental conditions in the area of the plots were appropriate for fire ant population growth; consequently, they do not explain the overall reduction in the populations. These results, combined with additional evidence reported previously, suggest that the infection with *V. invictae* and *T. solenopsae* has deleterious effect on native populations of *S. invicta*.

No Designated Session Theme

PARASITISM OF THE SILVERLEAF WHITEFLY FEEDING ON TEN SOYBEAN GENOTYPES IN SINALOA, MEXICO

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The silverleaf whitefly SLWF *Bemisia argentifolii* Berrows & Perring (Hemiptera: Aleyrodidae) is a key pest of several crops in northwestern Mexico. Plant resistance and Natural Biological Control are one of the tactics that may be used to reduce populations of this pest. However, genotypes and natural enemies must be identified and evaluated before they are effectively used to suppress SLWF populations. Soybean is a preferred SLWF crop host.

Parasitoids identified. *Eretmocerus californicus* Howard (Hymenoptera: Aphelinidae), is the only parasitoid that has been obtained, attacking last instar nymphs, after two soybean crop seasons. Higher number of parasitism has been observed on July reaching near of 50% (Fig. 1 and 2).

Parasitism on soybean genotypes. Percent parasitism was variable depending of the soybean genotype, but it was not consistent in the two crop seasons evaluated.

Soybean yield. Yield was significantly different among soybean genotypes ($P=0.01$) in both 2003 and 2004 seasons. Suaqui D-1-2-M showed the highest yield and Hutcheson the lowest.

Figure 1. Season 2003.

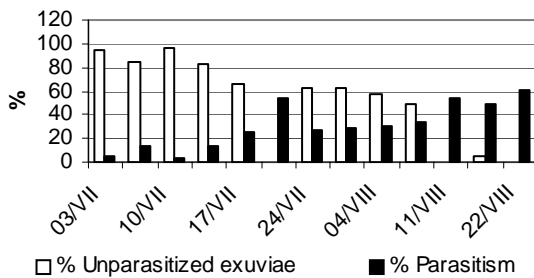
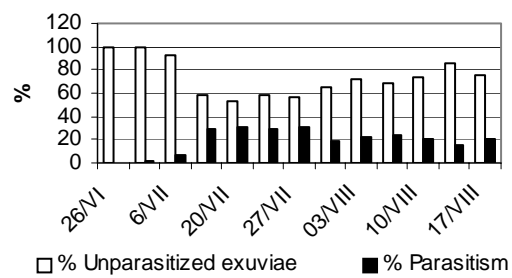


Figure 2. Season 2004.



Figures 1 and 2. Percent of parasitized and unparasitized exuviae of SLWF on soybeans.

No Designated Session Theme

NATURAL PARASITOID COMMUNITY OF THE GREEN BUD MOTH *HEDYA NUBIFERANA* (HAWORTH) (LEPIDOPTERA: TORTRICIDAE) IN ROMANIA

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Certain secondary pests, including the green bud moth, can be found in most commercial orchards, but rarely cause economic damage. However, they can act as alternative hosts for parasitoids of primary pests, such as *Cydia* spp (Lepidoptera: Tortricidae). Presence of secondary pests and their suite of parasitoids can therefore contribute to the natural regulation of primary pests. Between 1992 and 2004, we collected immature stages (eggs, larvae and pupae) of the green bud moth, *Hedya nubiferana*, attacking different fruit trees (apple, pear, plum, cherry) in mostly organic orchards in eastern Romania. Parasitoids were reared to adult under laboratory conditions and identified to species level.

We obtained 47 parasitic species from the following orders and families with the number of species in brackets: a) Hymenoptera - *Ichneumonidae* (22), *Braconidae* (9), *Chalcididae* (2), *Eulophidae* (6), *Encyrtidae* (1), *Torymidae* (1), *Pteromalidae* (1), *Eupelmidae* (2), *Trichogrammatidae* (1); b) Diptera - *Tachinidae* (1); and c) Nematoda - *Mermithidae* (1). Most of the species (32) behave as primary parasitoids, 12 are secondary parasitoids and three act as both. Most of the primary parasitoids (24) attack the larval stage of *H. nubiferana*, three attack larvae and pupae, three only pupae and one species was reared from eggs.

From 1992 to 1995, parasitism rates of both larval and pupal parasitoids reached 30.4% in pear orchards, 21.4% in apple, and 9.6% in plum orchards. The most important agents were parasitoids from the family Braconidae, contributing to 28.6% of parasitism in pear

orchards, 16.6 % in apple, and 7.8% in plum orchards. The braconid wasps *Macrocentrus pallipes* (Nees), *Apantheles xanthostigma* (Hal.) and *Apanteles longicauda* (Wesm.) were the most efficient species in limiting the number of the green bud moths. Our survey revealed more than 50 trophic relationships, most of them new for Romania and almost 30 of them new to science.

No Designated Session Theme

EFFECTS OF REGULAR INTRODUCTIONS OF *DINARMUS BASALIS* RONDANI (HYMENOPTERA: PTEROMALIDAE) ON THE EVOLUTION OF BRUCHIDS POPULATIONS WITHIN TRADITIONAL STORAGE SYSTEM IN SAHELIAN AREA

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Bruchidius atrolineatus Pic. (Coleoptera: Bruchidae) and *Callosobruchus maculatus* Fab. (Coleoptera: Bruchidae) are, in Sahelian area, Nigeria, the most important pests of cowpea beans, *Vigna unguiculata* (L.) Walp (Fabales: Fabaceae). Cowpea infestation by these two species of bruchids starts in the field at the beginning of the plants fruit bearing and continues during storage when damage can be high if no control action is taken. *B. atrolineatus* adults emerging in the stores from December to January are in reproductive diapause and the *C. maculatus* population is small. Numbers of *C. maculatus* increase substantially from February onward when temperatures and humidity rise.

In field as well as in stores, bruchids are associated with two larval parasitoids: *Eupelmus vuilleti* CRW (Hymenoptera : Eupelmidae) and *Dinarmus basalis* Rondani (Hymenoptera: Pteromalidae) which is the most efficient natural enemy of the bruchids. The number of *D. basalis* is low at the beginning of cowpea storage in December and because of the interspecific competition between the two parasitoid species, it cannot control the *C. maculatus* population, which therefore causes important seed weight losses.

In this study we investigated the effect of regular introductions of *D. basalis* on the evolution of bruchid populations within traditional storage systems. The experiment consists of a comparison between the evolution of a bruchids population in a standard jar and in one which received 200 couples of *D. basalis* adults every two weeks. Releases of *D. basalis* adults were made in stores containing varieties of cowpea which had been infested naturally in the fields before harvest. The results obtained from this study point out that the number of bruchids eggs deposited on cowpea pods and bruchid adults which emerged from seeds are much higher in the standard jars than in the ones which have received regular inputs of the parasitoid. A rate of reduction of 75 % is observed in the treated jar compared to the standard one.

These results clearly demonstrated that biological control of bruchids using the ectoparasitoid *D. basalis* adults in cowpea stores throughout the storage period is possible and limits the evolution of bruchid populations.

No Designated Session Theme

**EVALUATION OF THE EFFECT OF PREDATOR EXCLUSION ON
CASSAVA INFESTED WITH THE CASSAVA GREEN MITE
(*MONONYCHELLUS TANAJOA* BONDAR) IN THE
MIDDLE BELT REGION OF NIGERIA**

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A chemical exclusion method using Permethrin to assess the effectiveness of a predator *Thyphlodromalus aripo* (De Leon) utilized in the Biological control of the cassava green mite (CGM) *Mononychellus tanajoa* (Acari; Tetranychidae) was investigated. The results obtained in trials showed significant differences in green mite population and cassava root yields between fields where the predators were present and the fields where they were excluded by chemical spray. There were also differences in both shoot and stem weight however these were not significant.

No Designated Session Theme

**BIODIVERSITY OF TRICHOGRAMMATIDAE (HYMENOPTERA)
EGG PARASITOIDS AS BENEFICIAL BIOCONTROL AGENTS**

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The biodiversity of parasitic wasps (Hymenoptera: Trichogrammatidae), being parasitoids of eggs of 12 orders of insects and other arthropods (Arachnida), are discussed on the basis of original and literature data.

Trophic relations of parasitic Hymenoptera are recorded for 14 orders of insects: Coleoptera, Lepidoptera, Diptera, Heteroptera, Psocoptera, Odonata, Isoptera, Orthoptera, Neuroptera, Thysanoptera, Blattoptera, Hymenoptera, Megaloptera, Embioptera, and also with spiders (Arachnida), mites (Acari) and nematodes.

The 5 types of parasitism (in relation to development stage of host) recorded for parasitic Hymenoptera are egg-parasitism, larval, pupal, egg-pupal, and imaginal parasitism. The egg-parasitism is represented in several groups of Hymenoptera Parasitica: in superfamily Chalcidoidea - 7 families Trichogrammatidae, Mymaridae, Aphelinidae, Encyrtidae, Pteromalidae, Tetracampidae, Eulophidae; in superfamily Platygastroidea – families Scelionidae, Platygastriidae.

Features of egg-parasitism in Hymenoptera are: 1) polyembryony (development of many parasitoid larvae from one laid egg); 2) thelythoky (only females progeny from laid eggs); 3) arrenothoky (only males from laid eggs); 4) deiterothoky (males and females from laid eggs); 5) symbiosis with microorganisms (Wolbachia) (only females develop in the offspring).

The family Trichogrammatidae and Mymaridae are exclusively egg-parasitoids of other insects. The egg-parasitoids of family Trichogrammatidae have trophic relations with 12 orders of insects and egg-parasitoids of the family Mymaridae with 8 insect orders.

Importance of worldwide use of *Trichogramma* in biocontrol is recorded. *Trichogramma* is used on 22 agricultural and forestry crops including maize, wheat, cabbage, soya bean, cotton, rice, tomatoes, sugar beet, sugar cane, apples, plums, citrus, grape, pine-tree forests and others. Only 6 species are used in biocontrol as most important – *T. evanescens* Westw., *T. pintoi* Voegelé, *T. brassicae* Voeg., *T. nubilale* Ertle & Davis, *T. dendrolimi* Mats., *T. pretiosum* Riley.

The situation for application of *Trichogramma* in Ukraine is reviewed. Eleven (11) species of *Trichogramma* were recorded from Ukraine: *T. evanescens* Westw., *T. pintoi* Voegelé, *T. dendrolimi* Mats., *T. telengai* Sorok., *T. embryophagum* Hart., *T. elegantum* Sorok., *T. principium* Sug. et Sorok., *T. semblidis* Auriv., *T. piceum* Djurich, *T. bistrae* Kostad., *T. aurosum* Sug. et Sorok.. Only 3 species *Trichogramma* are used in Ukraine for mass rearing and application in plant protection: *T. pintoi*, *T. dendrolimi*, *T. embryophagum*.

In the European part of Russia 19 species of *Trichogramma* are recorded, in the Palaearctic region 52 species of *Trichogramma* were recorded. In Japan 7 species of *Trichogramma* are recorded at the present time: *T. papilionis*, *T. ostrinia*, *T. japonicum*, *T. chilonis*, *T. dendrolimi*, *T. pintoi*, *T. yawarae*. In Japan 24 genera and nearly 70 species of the family Trichogrammatidae are recorded at the present time. It is highly desirable to discover and describe new species of the genus *Trichogramma* and other genera of Trichogrammatidae in the nearest future.

No Designated Session Theme

**PARASITOIDS (HYMENOPTERA: EULOPHIDAE, APHELINIDAE)
OF THE CABBAGE WHITEFLY, *ALEYRODES PROLETELLA*
(HEMIPTERA: ALEYRODIDAE), ASSOCIATED WITH THE
GREATER CELANDINE (*CHELIDONIUM MAJUS*)**

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The whiteflies (Hemiptera: Aleyrodidae) are an economically important group of insects damaging wide variety of plants. They are more pesticide-resistant compared to other sucking pests because the nymphs are well-protected by the external cases of their puparia. The most perspective area of their control is therefore the use of natural enemies, in particular parasitoids that attack the immature stages. The cabbage whitefly, *Aleyrodes proletella* (Linné, 1758) has a comparatively wide spectrum of host plants, being, in particular, serious pest of *Brassica oleracea* in Europe (Ramsey, Ellis, 1996) and North Africa (Nebreda, 2005). We have studied the parasitoids of this whitefly on the host populations inhabiting the greater celandine, *Chelidonium majus*, in Ukraine. Three species of parasitoids were reared from puparia of *A. proletella* collected in nature. These are: one eulophid, *Euderomphale chelidonii* Erdös 1966, and two aphelinids, *Encarsia tricolor* Forster, 1878 and *Encarsia inaron* (Walker, 1839). No-choice tests were conducted for the parasitoids with the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood, 1856). Both *Encarsia* species were found to attack the greenhouse whitefly, whereas *E. chelidonii* avoided the puparia of this species. The behavior and preimaginal stages of *E. chelidonii* were studied in details. It is found that only some penetrations of the whitefly puparia by the parasitoid ovipositor result in egg laying, but oviposition punctures often are used for host haemolymph extraction; both sexes of the parasitoid feed on host fluids. The courtship repertoire of *E. chelidonii* is complex and differs from most other chalcidoids. The larvae of *E. chelidonii* differ significantly from the larvae of *Encarsia tricolor* and *E. inaron* in habitual appearance (swollen in *E. chelidonii* and slender in *Encarsia*-species) and tiny details of morphology (e.g. shape of mandibles).

No Designated Session Theme

SMALL TUMBLEWEED MUSTARD, *SISYMBRIUM LOESELII*, AS A TRI-TROPHIC MODEL SYSTEM: INTERACTIONS BETWEEN SEED-EATING WEEVILS (COLEOPTERA: CURCULIONIDAE) AND THEIR PARASITIDS (HYMENOPTERA: EULOPHIDAE)

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The small tumbleweed mustard, *Sisymbrium loeselii* L. (Capparales: Brassicaceae), is a plant of European origin that was accidentally introduced to the New World, and is now recorded in 31 states of the USA, and regarded as an invasive weed (Stubbendieck *et al.*, 1994). The weevil *Ceutorhynchus sisymbrii* (Dieckmann) (Coleoptera: Curculionidae) was described nearly fifty years ago and it was proposed that this species is monophagous on *Sisymbrium loeselii*. However, the biology of this weevil remained unknown. No host records were available also for the parasitic wasp *Entedon sylvestris* Szélnyi (Hymenoptera: Eulophidae), a representative of the genus *Entedon*, which comprises many parasitoids of Curculionidae. The original studies conducted by the author demonstrated that adults of *C. sisymbrii* feed on shoots, flowers and fresh seeds of *S. loeselii*. Females lay eggs in May-July, when the seeds are fully-grown, but still soft. The female makes an opening in the pod with her rostrum, eats the seed below the opening and then turns back and begins oviposition. The hatched larvae feed on the seeds and remain in pods until they are fully-grown. In late May - beginning of July, the females of the parasitoid *Entedon sylvestris* search along the pods of *S. loeselii* for weevil larvae. Once the parasitoid female has located a host, she walks back and forth several times and starts ovipositing. Females of *E. sylvestris* parasitized weevil larvae of various instars and the parasitoid larva remains within the body of the host weevil larva until the emergence of the latter from the dried host-plant pods. In the end of June – beginning of July the pods of *S. loeselii* split and the mature larvae of the host weevil (parasitized and non-parasitized) leave the host plant's pods, fall to the ground and quickly bury themselves. Soon thereafter they prepare an earthen cell, in which pupate. The larvae of the parasitoid, *E. sylvestris*, are in their second instar stage when the host larvae leave the pods of *S. loeselii*. The moult of the parasitoid larva into the last (third) instar, as well as pupation takes place within the host's body, underground. Adults of *E. sylvestris* as well as their hosts, *C. sisymbrii*, must therefore overcome a soil layer to emerge the following spring. These data may be used in the development of the biocontrol programs against the small tumbleweed mustard and other invasive weeds of Brassicaceae.

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DIVERSITY AND ECOLOGY OF NATURAL ENEMIES OF OLIVE FRUIT FLY, *BACTROCERA OLEAE*, IN SOUTH AFRICA

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The recent establishment in North America of olive fruit fly, *Bactrocera oleae* (Gmelin) (Diptera: Tephritidae), has renewed interest in classical biological control of this pest. Previous surveys conducted in Africa and Asia during the 20th century demonstrated a greater natural enemy diversity in southern and eastern Africa than in the Mediterranean region, but comprehensive evaluations were not conducted, and all attempted introductions were unsuccessful. To identify new natural enemies of olive fly for efficacy and specificity evaluation and possible importation into California, a new exploration program was planned by ARS EBCL. Surveys have been conducted in wild olives, *Olea europaea* subsp. *cuspidata* (Wall.ex G. Don), in both East and West Cape Provinces (Rep. South Africa) during the southern hemisphere fall seasons of 2001-2004, in the NE provinces (vicinity of Pretoria) in 2003, and also in Namibia in 2004. Through co-operators, surveys have also been conducted in Kenya and Pakistan.

Olive fly populations were consistently higher in West Cape than in East Cape Province, as were populations of their natural enemies. Several braconids (*Bracon*, *Psytalia* and *Utetes* spp.) were the most abundant parasitoids of the fly recovered in these surveys in southern Africa. The parasitoid fauna of southern Africa was similar to that found in Kenya (Copeland et al., 2004, Bishop Museum Bull. Entomol. v.12), but much richer than the diversity known from North Africa. Only one species of braconid, *Psytalia* c.f. *ponerophaga* (Sylvestri), has thus far been reared from olives in Pakistan. Numerous chalcidoids were also reared from wild olive in Africa, but they do not appear to be as abundant or widespread as the braconid species, and many are associated with seed chalcids attacking olives.

Although wild olive is widely distributed across southern and eastern Africa, rainfall patterns strongly influence the occurrence and abundance of fruit, and consequently the abundance of flies and their parasitoids. The dominant parasitoid species varied from region to region, and also varied from one year to the next. Braconid parasitoids were often present even at low densities of olive fly.

No Designated Session Theme

TOWARDS CLASSICAL BIOLOGICAL CONTROL OF LEEK MOTH

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The leek moth, *Acrolepiopsis assectella* (Zeller) (Lepidoptera: Acrolepiidae), is an invasive alien species of European origin that has become established in eastern Ontario and southwestern Quebec in Canada. The larvae of this pest mine the leaves of *Allium* plants, with the potential to reduce the marketability of the plants if not destroy them outright. Laboratory- and field-based research is currently being carried out to evaluate European leek moth parasitoids as potential agents for classical biological control in Canada. While records of parasitism on leek moth can be found in the literature, there has been little recent research activity covering the ecological relationships between this pest and its natural enemies. Moreover, very few of the earlier relevant papers provide a close look at the dynamics of leek moth parasitoid communities.

In 2004 and 2005, field surveys were carried out in commercial organic *Allium* crops to assess the severity of leek moth infestations and the occurrence of parasitism on leek moth in west-central Europe. Since biological control programmes against highly concealed pests tend to have reduced success compared to similar initiatives targeting species with more exposed developmental stages, the relative vulnerability of the leek moth's developmental phases was examined. To accomplish this, a complete life table study was executed whereby the mortality associated with each leek moth life stage was evaluated. Preliminary results show that leek moth populations in general are effectively regulated on leek and onion crops in west-central Europe. Leek moth mortality was higher in the egg and pupal stages than in each of the five larval instars. Another interesting observation was that leek moths suffer high mortality during the brief period between egg hatch and first penetration into the leaf tissue of the host plant. Parasitism was consistently low at the life table study sites. However, pupal

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mortality was significantly higher on exposed plants than on control (mesh-covered) plants, suggesting that this stage of leek moth development is most susceptible to natural enemy attack. Collectively, these results suggest that unless a highly specialised and efficient larval parasitoid is identified, the eggs or pupae of the leek moth might be the most effective life stages to target for classical biological control.

No Designated Session Theme

ABIOTIC AND BIOTIC FACTORS AFFECTING THE BIOLOGICAL CONTROL OF CEREAL STEMBORERS IN EAST AFRICA

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The exotic braconid larval parasitoid, *Cotesia flavipes* Cameron (Hymenoptera: Braconidae), is used as a biological control agent against the invasive crambid *Chilo partellus* (Swinhoe) (Lepidoptera: Crambidae), a serious pest of cereal crops in East and Southern Africa. It was hypothesized that the success of the parasitoid was determined by the composition of stemborer species, which might be acceptable but suitable or unsuitable, climate (temperature mainly), the availability and suitability of host larval stages at the time of release, quality of the host plant (nutritional status and age of the plant, and host plant species), which affects both the insect host and parasitoid, as well as the accessibility of the host inside the stem.

To predict the effect of different borer species compositions, found in the different agroecological zones in Kenya, on the performance of *C. flavipes* a two-host-two-parasitoid model was used. The model assumed both intra-specific competition of *C. partellus* and included the major indigenous parasitoid species, *Cotesia sesamiae* Cameron, the invasive and the primary indigenous pest species in a given agroecological zone. The model predicted that in areas where both host species were suitable to either parasitoid species, stemborer densities would be reduced to and controlled at low densities and *C. flavipes* would become the dominant parasitoid species. This, however, would increase the risk of extirpation of *C. sesamiae*. If the indigenous host species was unsuitable to *C. flavipes*, both parasitoid species could coexist but the indigenous host would not be reduced to low densities. Intra-specific competition of host species was vital for the coexistence of the four species; high competition coefficients of indigenous stemborer hosts enhanced the stability of the system if the host was unsuitable to *C. flavipes*, but it increased the risk of extirpation of *C. sesamiae* if the host was suitable. The model output was validated with real-field situations.

Temperature effect studies showed that development time of *C. flavipes* immatures significantly decreased with temperature and with host instar, while sex ratio changed from male to female-biased. Fourth larval instars were considerably more suitable than third instars. Parasitized larvae kept feeding at the same rate as unparasitized. Thus, *C. flavipes* had no direct effect on yield formation of the plant but rather through a long-term reduction of the pest over several generations.

Nitrogen fertilization applied to sorghum and maize significantly increased survivorship of *C. partellus*. Parasitism tended to decrease with age of the maize plant and it tended to be lower on sorghum than maize. Nitrogen had no effect on number of progeny and sex ratio of the parasitoid while egg load increased significantly with nitrogen level applied to maize. On sorghum, the effects were mostly not significant.

Host species – cultivated or wild – had a significant effect on the performance of both *C. partellus* and *C. flavipes*. The percentage of *C. partellus* larvae producing cocoons and egg load of *C. flavipes* progeny tended to be lower on wild than cultivated host plants.

No Designated Session Theme

CONTROL OF CHESTNUT WEEVILS WITH ENTOMOPATHOGENIC NEMATODES: FIRST EXPERIENCES

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Entomopathogenic nematodes were used to control the chestnut weevil, *Curculio elephas* Gyll. (Coleoptera: Curculionidae), under semi-field conditions in 2003 and 2004. PVC tubes (diameter 10cm; length 40cm) filled with sandy soil were buried in a chestnut orchard and artificially infected with pest larvae by applying nematodes at a rate of 2 Mio infective juveniles (IJ)/m². Mortality was 43, 56, 56, and 52% respectively, after applying *Steinernema carpocapsae* (Weiser), *S. feltiae* (Filipjev), *S. kraussei* (Steiner) and *Heterorhabditis megidis* Poinar, Jackson and Kleine, all (Nematoda: Rhabditidae). However, mortality was not significantly increased in nematode treatments compared to the untreated control. Most of the surviving pest larvae were found at a depth of 31-40cm in the soil columns, whereas increased numbers of dead weevils were in the upper soil layers. It is concluded that nematodes failed to follow and attack chestnut weevils at increased soil depths and that application timing and technique needs improvement for sufficient control of this pest with nematodes.

No Designated Session Theme

APPLICATION OF RESPONSE SURFACE METHODOLOGY FOR DEVELOPMENT AND OPTIMIZATION OF INSECT DIETS FOR MASS PRODUCTION OF PARASITOIDS

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Insect diet development can be difficult and time-consuming because of the large number of component permutations to test. One-factor at a time (OFAT) approaches are time, labor and cost prohibitive and inherently confounded. However, factorial mixture designs provide a powerful and relatively simple approach to de-convoluting effects and interactions of multiple diet components. Screening designs facilitate analysis of up to 24 components in a single experiment, while crossed, full D-Optimal mixture designs are capable of quantifying interactions/effects of 6 or more components across a temperature range. The response surfaces generated by this approach can be used to define numerous diet “optima” as functions of multiple responses, such as development time, male to female ratios, and/or days to adult emergence. Mixture response surface methodology, as it relates to insect diet formulations and component screening, will be discussed along with results and implications of a newly developed rearing medium for the pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae).

M. hirsutus was unknown in the Western Hemisphere (with the exception of Hawaii) prior to its discovery in the Caribbean in 1994. It has now spread to the Americas including California, Mexico, Central America, and Guyana, Venezuela, and Colombia in South America. *M. hirsutus* is a global pest of ornamentals, vegetables, grapes and various tree crops. It was discovered in Florida (USA) in 2002 and has been inadvertently distributed through ornamental commerce to several US states. The economic risk to U. S. agriculture has been estimated at \$750 million per year due mainly to its wide host range that includes over 125 plant species. The largest risk is to ornamental crops, followed by vegetables, citrus, grapes, and avocados. The U.S.D.A. has provided two encyrtid parasitoids, *Anagyrus kamali* Moursi and *Gyranusoidea indica* Schaffe, Alam & Agarwal (Hymenoptera: Encyrtidae) throughout the Western hemisphere and is currently conducting international surveys for additional parasitoids. For mass-producing parasitoids, *M. hirsutus* must be reared on fruits of various species of cucurbits, sprouted potatoes, or hibiscus plants. Of these, the preferred host has been Japanese pumpkin (*Cucurbita moschata* Duchesne (Curbitales: Cucurbitaceae) due to its ribbed rinds and characteristic warted surface, which provide large settling areas for mealybugs. However, seasonal shortages of produce, and difficulties in maintaining a continuous supply

of etiolated sprouts of potato, threaten production and increase costs. Work in our laboratory compared plant hosts and showed the potential of a pumpkin-based meridic diet for rearing *M. hirsutus*. Complete mealybug development was obtained on meridic diet as well as five plant substrates. There was a positive linear relationship between developmental rates of female *M. hirsutus* obtained on plant hosts and diet, and those of *A. kamali* females and males. Adult parasitoid emergence was approximately 20% on meridic diet compared with 60% on the best plant substrate. These results with a simple diet encouraged us to pursue a semi-defined artificial diet based on readily available materials. To simplify diet optimization, we applied response surface methods. Our results and the broader application of this approach will be discussed.

No Designated Session Theme

EGG PARASITOIDS FROM ARGENTINA, POTENTIAL CANDIDATES FOR THE BIOLOGICAL CONTROL OF GLASSY-WINGED SHARPSHOOTER IN THE UNITED STATES

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The glassy winged sharpshooter (GWSS), *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae), vector of the Pierce's disease, is a very serious pest problem in several crops, especially grapes in southern California. Because GWSS does not occur in South America, its biological control using South American natural enemies relies on the 'new-association' approach. Field surveys for eggs parasitoids were conducted in Argentina on *Tapajosa rubromarginata* (Signoret), a common and native sharpshooter. Eggs of *T. rubromarginata* were obtained in the laboratory and later exposed to parasitization in 92 selected sites in nine ecological regions of Argentina.

Additionally, laboratory studies on bionomics (development time, progeny sex ratio, adult longevity) and host range were conducted with the most promising candidates, *G. tuberculifemur* (Ogloblin), *G. annulicornis* (Ogloblin) and *Gonatocerus* sp. (Hymenoptera: Mymaridae). For the specificity experiments, eggs of 18 potential hosts were tested for each parasitoid: Delphacidae (3 spp.), Cicadellidae (12 spp. in 3 subfamilies and 5 tribes), Lepidoptera (1 sp.), Coleoptera (2 spp.).

From the 14,922 eggs exposed, 1,730 (11.6%) were parasitized at 53 sites (57.6%) and wasps of 14 different species in 5 genera were obtained: eight species of *Gonatocerus* and one of *Polynema* (Mymaridae), one species of *Paracentrobia*, 2 species of *Oligosita* and 2 of *Zagella* (Trichogrammatidae). Parasitoids emerged from all eco-regions (from rain forest to desert) except from Temperate Patagonian Forest. A richness latitudinal gradient was observed (Fig. 1).

Bionomic results for the 3 spp. of *Gonatocerus* tested are shown in Table 1.

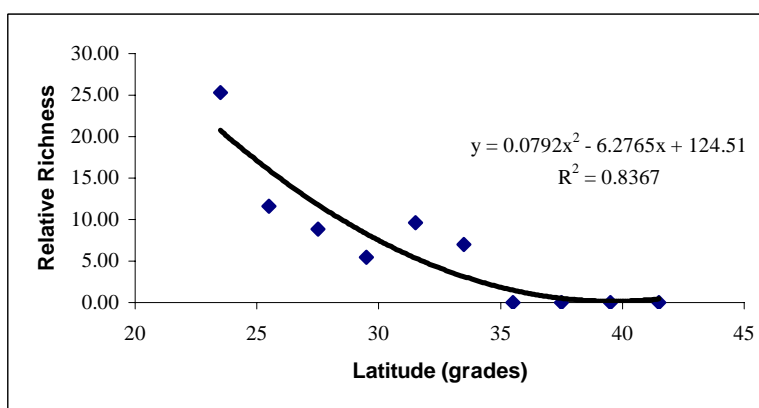


Figure 1. Relationship between species richness and latitude.

Table 1. Laboratory results on *T. rubromarginata* eggs exposure to 3 spp. of *Gonatocerus*. n.s. Not significant (*t* test), * significant (*t* test $P < 0.05$)

Species	Generations	Eggs			Longevity (days)		
		Exposed	Parasitized (%)	Sex ratio F/M	Development time (days)	Female	Male
<i>G. tuberculifemur</i>	7	2095	1500 (71.6)	2.1	12.6 ± 1.8	6.7 ± 3.9	n.s.
<i>G. annulicornis</i>	6	1277	969 (75.8)	1.2	10.9 ± 1.5	7.1 ± 4.0	4.7 ± 2.9*
<i>Gonatocerus</i> sp.	4	1503	907 (60.5)	1.4	11.0 ± 1.5	6.6 ± 4.3	4.3 ± 2.6*

Preliminary results of the host range studies indicated that the genus *Gonatocerus* is restricted to the tribe Proconiini, where GWSS belongs. The females of *G. tuberculifemur*, *G. annulicornis* and *Gonatocerus* sp. oviposited and the larvae developed to adults only in sharpshooters of that tribe. None of the other 14 species tested were attacked during the study. Since March 2001, these parasitoids have been successfully cultured using eggs of the factitious host, *H. coagulata*, at the quarantine facilities in USDA-APHIS Mission, TX, and Riverside, CA. Complementary studies on the biology and host range are being carried out.

No Designated Session Theme

ARTIFICIAL SELECTION ON FECUNDITY IN WINGLESS TWO-SPOT LADYBIRD BEETLES (*ADALIA BIPUNCTATA*)

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Wingless ladybirds in biocontrol. Predatory ladybird beetles are being used as control agents in augmentative biological control against aphids. Because adult ladybirds disperse by flight at low host density, larvae are commonly released. However, repeated releases are usually required making their use in biocontrol costly.

A ladybird stock that is not able to fly away would be likely to increase the efficiency of biocontrol and to decrease the costs. Wingless ladybirds might have additional benefits such as increased fecundity because energy is not invested in developing and maintaining the structures needed for flight. Such a trade-off has been found in other insects, e.g. crickets.

Fecundity of wingless *A. bipunctata*. A wingless individual of the two-spot ladybird beetle, *Adalia bipunctata* (L.) (Coleoptera: Coccinellidae), was collected in the wild and used to establish a pure-breeding laboratory stock. Pilot experiments showed that in segregating families, wingless *A. bipunctata* were larger and heavier than their winged siblings, indicating a trade-off. Although insect size is often positively correlated with fecundity, in contrast to this, the wingless individuals in our stock show reduced fecundity. However, if there is potential for evolution of a trade-off between fecundity and the ability to fly, artificial selection on the wingless stock might be able to increase fecundity, perhaps eventually to the extent of exceeding that of wildtypes.

Artificial selection on high early fecundity. We have, therefore, selected for high early fecundity in wingless *A. bipunctata*. Two lines were set up simultaneously, consisting of 120 single wingless pairs in each line in each generation. Pairs were kept together for 12 days and eggs collected several times. The number of larvae that hatched from these eggs was recorded, and the top 30 families then selected as parents for the next generation.

Here we present the results over the first seven generations of selection.

No Designated Session Theme

**SEASONAL OCCURRENCE OF THE *APHIDIUS ROSAE* HALIDAY
(HYMENOPTERA: BRACONIDAE) PARASITOID OF
MACROSIPHUM ROSAE L. (HOMOPTERA: APHIDIDAE) IN
ISFAHAN, IRAN**

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Aphidius rosae is the most important parasitoid of rose aphid, *Macrosiphum rosae*, in many parts of the world. This parasitoid is specialized on *M. rosae*. The seasonal occurrence of *A. rosae* in rose gardens in Isfahan, Iran, was studied from September 2003 to October 2004. Weekly sampling included 10-15 cm of terminal portion of 10 shoots. This parasitoid has two activity periods in a year. Initially spring activity started at the end of March and continued to early June, when the temperature increased and density of the *M. rosae* on rose decreased. The other activity period in autumn started from the end of October to early December. Thereafter, the cold weather did not allow the parasitoid to be active. The number of parasitized aphids gradually increased during spring and peaked in early May. In autumn, parasitoid peaked on 10 November. Range of percentage parasitism in the field in spring and autumn was from 2.75% to 11.69% and from 0.84% to 2.72%, respectively.

Percentage parasitism in spring when the number of parasitoids was high, was estimated for apterous adults and late nymphal stages (3 and 4) of aphids under laboratory conditions. The maximum parasitism was 27% and 29% for apterous adults and late instars, respectively. This information, together with seasonal changes of the aphid, can be used for determining the best date for applying classical biological control techniques on *M. rosae*.

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INFLUENCE OF TWO ENDOPARASITIC WASPS, *HYPOSOTER DIDYMATOR* (HYMENOPTERA: ICHNEUMONIDAE) AND *CHELONUS INANITUS* (HYMENOPTERA: BRACONIDAE), ON THE GROWTH AND FOOD CONSUMPTION OF THE HOST LARVA *SPODOPTERA LITTORALIS* (LEPIDOPTERA: NOCTUIDAE)

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The most obvious effect of parasitization is a reduction on numbers of the next host generation, but there can be also a direct effect on the population of the host, because parasitized pest larvae by different biological agents, usually modify their feeding behaviour reducing food consumption, giving as result, lesser damage on host plants before finally dying. *Hyposoter didymator* (Thunberg) and *Chelonus inanitus* (Linnaeus) are frequently found in Spain parasitizing larvae and eggs, respectively, of the important noctuid pest *Spodoptera littoralis* (Boisduval). In this study, we have compared effects of parasitism by these species, on development, growth and food consumption of the host larvae. Parasitized larvae always consumed significantly less artificial diet than non parasitized ones, and they were unable to finish the development, dying on the fifth larval instar. When *C. inanitus* parasitized *S. littoralis* eggs, host larval consumption was reduced very soon, since emerged larvae moulted into the second instar two days after eclosion. In contrast, *H. didymator*, which parasitized third instar *S. littoralis* larvae, only reduced their food consumption four days after parasitization. However, overall feeding reduction on host larvae achieved by the latter was higher than that caused by the former species *C. inanitus*. Final body weight of host larvae parasitized by *H. didymator* and *C. inanitus* was 6.7 and 13.0%, respectively, of the maximum weight of a healthy 6th instar host larvae. Based on these results, both parasitoids had a direct effect on *S. littoralis* populations, reducing enormously larval food consumption, but lesser damage on host plants can be expected from *H. didymator* parasitized larvae because this species seems to be more effective.

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BIOLOGICAL CONTROL OF THE GLASSY-WINGED SHARPSHOOTER IN CALIFORNIA - ANATOMY OF A STATE RUN PROGRAM

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The glassy-winged sharpshooter, *Homalodisca coagulata* (Say) (Homoptera: Cicadellidae) became established in California in the late 1980s and has now become naturalized throughout most of southern California. The insect's size, polyphagy, and longevity all contribute to its ability to vector a complex of plant diseases caused by the of xylem-limited bacteria *Xylella fastidiosa*. The threat to grape production due to an increase in the spread of Pierce's disease has led to the initiation of a State-run program to suppress populations of the glassy-winged sharpshooter in California

Initial evaluation of important mortality factors for the glassy-winged sharpshooter in California and in its native range identified the importance of a complex of mymarid egg parasitoids. Over one dozen agents were recovered from their native range and screened for target and non-target effects under quarantine conditions. Three species of mymarid, *Gonatocerus ashmeadi* Girault, *G. morrilli* Howard and *G. triguttatus* Girault, were selected for release as biological control agents against the glassy-winged sharpshooter in California in late 2000. One further species, *G. fasciatus*, was added in 2002.

On release from quarantine, the agents were moved to two biological control agent production facilities in California for production, release, and evaluation. Production research included the selection of plants and rearing conditions to optimize glassy-winged sharpshooter growth and egg production for parasitoid colonies. Release work included appraisal of release sites in urban, riparian, and agricultural settings. Evaluation of all release sites was carried out prior, during, and after releases to assess efficacy of both native species of egg parasitoids and introduced biological control agents.

Over one million agents have now been released in an area that covers over 50,000 square miles. Releases of single or combinations of agents have been made into multiple sites and have led to over 140 recoveries. molecular approaches are being developed to assist us in the rapid identification of parasitized eggs and in discriminating between native and exotic individuals of the same species.

The glassy-winged biological control program is one of the largest biological control efforts in existence and is the product of much theoretical research invested in biological

control. A large part of the responsibility of the biological control facilities is the support of research into to new control strategies and agents. The collaboration between all levels of government (county, state and federal), university, farmers, and industry is remarkable and is being seen as a model on which to base future biological control strategies in California. One facility is currently being purchased and will be dedicated to the development of biological control strategies of current and future pests in California. It is anticipated that the state-operated facilities will provide a bridge between researchers into biological control and the nation's biological control industry, 50% of which is based in California.

No Designated Session Theme

BIOLOGICAL CONTROL IN THE WALNUT-FRUIT FORESTS OF KYRGYZSTAN

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In Kyrgyzstan, the forest covers an area of about 864,900 ha, which represents only 4,32% of the territory. All forests are very valuable and highly protected. Management is focused mainly on protection and includes primarily sanitary activities, whereas industrial silviculture is prohibited. Despite the small areas involved, forests play an important role in the development of rural economies and in the improvement of environmental conditions. For example, forests provide important fruit crops such as walnut and pistachio.

However the sanitary condition of the forests causes concern. Among the main factors affecting resources in walnut forests are pest insects and diseases. Consequently, forest pro-

tection against pest insects is an essential part of the activity in the State Forest Service of Kyrgyzstan.

Walnut forests constantly suffer from pest insects and diseases, among which the main ones are: *Lymantria dispar* L. (Lepidoptera: Lymantriidae), *Erannis defoliaria* Clerck (Lepidoptera: Geometridae), *Malacosoma parallela* Staudinger (Lepidoptera: Lasiocampidae), *Sphaerocanium prunastri* Fonsc. (Homoptera: Coccidae), *Caliroa cerasi (limancina)* L. One of the most serious forest protection problems in Kyrgyzstan has been the outbreak of gypsy moth (*Lymantria dispar*) in walnut forests that started in 1970. Defoliation by the gypsy moth has strongly affected pistachio, walnut and apple harvests. The gypsy moth is one of the most important forest insect pest species in Central Asia. Larvae of this moth defoliate large areas of the walnut-fruit forest stands annually, not only in Kyrgyzstan (Romanenko, 1984; Ashimov, 1989; Orozumbekov et al., 2003).

The wide use of chemical insecticides against forest pests during the past 30-35 years has had a detrimental effect on the forest biocenose. Chemical insecticides are cheap, highly efficient but not environmentally friendly and non-selective. NPV (Nuclear Polyhedrosis Virus) has been used against gypsy moth since 1984 in Kyrgyzstan. Bt preparations are effective against gypsy moth but have not been evaluated yet in the mountain conditions of Kyrgyzstan. Investigating natural enemies in these populations should help to detect species that might play an important role in the natural control of the pest. Modern biological control methods in mountain forests would be practically harmless for the environment and could be applied not only in Kyrgyzstan but also in all Central Asian Republics where the use of chemical insecticides has been limited.

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TWO NEW LARVAL ECTOPARASITIC MITES (ACARI: ERYTHRAEIDAE) ON SUNN PEST FROM IRAN

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Sunn pest *Eurygaster integriceps* Puton (Hemiptera: Scutelleridae) is the most important pest of cereals in Iran especially in the Fars Province. During 2000-2002 studies were carried out to identify mites associated with sunn pest from Iran in the Fars region. A total of five species were recorded and identified. Of these two species of genera *Charletonia* Oudemans and *Leptus* Latreille are new species and ectoparasites of *E. integriceps*. Our literature survey sug-

gests that this might be the first *Charletonia* described in the world. Some of the key taxonomic characteristics distinguishing these red mites are noted. Sites of attachment on the sunn pest are the dorsal of the abdomen and legs. The idiosoma are oval in outline. The dorsal scutum is longer than wide and oblong with rounded angles. The anterior border is smoothly rounded, while lateral borders are less convex. The posterior border is almost straight, with sensilla bases protruding through the margin. Sensillary setae have small setules in distal halves. In the gnathosoma: chelicerae-bases are rounded; the squatpyriform is without special markings. The palpal tibial claw is small and terminally split, and the tines subequal. Field results showed that the adult of sunn pest was a suitable host for *Charletonia* and *Leptus*. These mites are suitable candidates for the biological control of agricultural pests, whereas the classification of these mites is still problematic.

No Designated Session Theme

**REPRODUCTIVE AND DEVELOPMENTAL BIOLOGY OF
GONATOCERUS ASHMEADI (HYMENOPTERA: MYMARIDAE),
AN EGG PARASITOID OF HOMALODISCA COAGULATA
(HEMIPTERA: CICADELLIDAE)**

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Gonatocerus ashmeadi Girault (Hymenoptera: Mymaridae) is a common natural enemy associated with the insect pest *Homalodisca coagulata* (Say) (Hemiptera: Cicadellidae) in its home range of southeastern USA and northeastern Mexico. It is a self-introduced resident of southwestern USA and most likely came into the area in parasitized eggs and has since established a local association with *H. coagulata* and the native congeneric *H. liturata*.

Investigating a biological control agent's environmental requirements for reproductive and developmental biology with that of the host can allow for an enhanced understanding of the potential impact the natural enemy will have on its target.

The developmental and reproductive biology of *G. ashmeadi* was studied in the laboratory at five different temperatures (15, 20, 25, 30, and 33°C) and collected data were used to create life tables for this parasitoid. Mean adult longevity was significantly different between temperatures ($P < 0.001$) and was greatest at 15°C and declined as the temperature increased. The rates for oviposition that led to successful reproduction of offspring were highest at 30°C

and were significantly different between temperatures ($P < 0.001$). At 30°C approximately 46% of host eggs presented to parasitoids produced viable offspring. This rate decreased with temperature to approximately 3% at 15°C. Higher temperatures similarly lowered the production of viable offspring with 10% of GWSS eggs producing viable offspring at 33°C.

The number of offspring produced by individual parasitoids over their lifetime was greatest at 25°C and fell as temperature either increased or decreased ($P < 0.001$). There were no statistically significant differences in offspring sex ratios between the temperatures. Survivorship of *G. ashmeadi* decreased as temperature increased and declined at a similar rate to female offspring production.

The lower threshold temperature for egg to adult development was determined by linear regression and was calculated as approximately 8°C. The Modified Logan Model indicated that the upper developmental threshold was approximately 36°C and the fastest rate of development was at 30°C. A total of 137 degree days above the minimum temperature threshold were required for successful development of *G. ashmeadi* from egg to adult.

Statistically significant differences were calculated across the five temperatures among five demographic growth parameters that were analyzed using the jackknifed values generated from the l_m^x life tables. Mean net reproductive rate ($P < 0.001$), intrinsic rate of increase ($P < 0.001$) and finite rate of increase ($P < 0.001$) were significantly higher for *G. ashmeadi* reared at a constant 25°C, 30°C and 33°C respectively. Population doubling times were significantly ($P < 0.001$) fastest when parasitoids were reared at 30°C. Mean generation time was significantly lower at 33°C ($P < 0.001$).

No Designated Session Theme

CAN MUSTARD SEED MEAL INCREASE THE ABUNDANCE OF *ALEOCHARA* SPECIES IN CANOLA?

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Delia radicum L. (Diptera: Anthomyiidae), the cabbage root maggot, is a species of European origin, and is a pest of vegetable brassicas and of canola in Canada. Three species of *Aleochara* (*A. bilineata* (Gyllenhal.), *A. bipustulata* (L.) and *A. verna* (Say) (Coleoptera: Staphylinidae)) have been identified as playing important roles in reducing the populations of *Delia radicum* (L.) in many brassica crops. *Aleochara bilineata* and *A. verna* are present in both North America and Europe. However, *A. bipustulata* is not present in North America and so may be a potential candidate for use as a biological control agent against *D. radicum* in Canadian canola. If risk assessment tests reveal *A. bipustulata* is safe to release in the Canadian prairies then a method of manipulating and maintaining the populations of *A. bipustulata* would be beneficial. In southern Sweden the addition of white mustard seed meal as a mulch was shown to attract and maintain *Aleochara* spp. to suppress *D. radicum* populations in vegetable brassicas (1992; IOBC/WPRS Bulletin XV (4): 171-175).

Our study examined whether attraction of *Aleochara* species to mustard seed meal would occur in canola, what species of *Aleochara* are attracted, and investigated the nature of the attraction mechanism. In field plots of summer canola in Switzerland, *Delia radicum* and *Aleochara* populations were compared between plots with and without mustard seed meal mulch. Parasitism levels and adult activity of *A. bipustulata* were consistently higher in plots treated with mustard seed than in untreated plots. However, levels of parasitism and adult activity of *Aleochara bilineata* were not influenced by the mustard seed meal treatment. Analysis of volatiles emitted by mustard seed meal, using GCMS, revealed seven major peaks, of which the highest was for limonene. Experiments in a Y-tube olfactometer were conducted to examine the attractiveness of mustard meal and associated volatiles to *A. bipustulata* and *A.*

bilineata. *Aleochara bipustulata* exhibited preference for volatiles from mustard seed meal when compared to clean air and to air passed over soil; whether the meal was wet or dry did not influence these responses. *Aleochara bipustulata* showed a tendency to choose the olfactometer arm with limonene over that with clean air, but in initial experiments this was not significant. *Aleochara bilineata* showed no attraction to mustard meal or associated volatiles in the olfactometer.

No Designated Session Theme

BIOLOGICAL CONTROL OF THE PINK HIBISCUS MEALYBUG, *MACONELLYCOCCUS HIRSUTUS* (GREEN), IN IMPERIAL VALLEY, CALIFORNIA, U.S.A.

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A cooperative biological control project against the pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green) (Homoptera: Pseudococcidae), infestation in the low-desert region of California, USA was initiated in the fall of 1999. At that time, the average mealybug density on mulberry trees was 256 second instar nymphs to adult mealybugs per branch terminal. Subsequently, several strains of two encyrtid parasitoid species (*Anagyrus kamali* Moursi and *Gyranusoidea indica* Shafee, Alam & Agarwal) (Hymenoptera: Encyrtidae) were mass reared and released. In 2000, over 400,000 parasitoids of the two species were released at 400 locations. Population densities of mealybug and percent parasitism were monitored at a number of mulberry tree and carob tree urban home sites for four consecutive years. The primary sample method consisted of sampling branch terminals and counting the number of second instar nymphs to adult mealybug life stages. The population density of *M. hirsutus* within the first year was reduced by approximately 95%. Over the entire four-year period of the project, the average regional population density of the mealybug exhibited a continued decline. *Anagyrus kamali* was the predominant parasitoid, often parasitizing in excess of 50% of the mid to late stage *M. hirsutus* in the first two years following the parasitoid's release. Although

Gyranusoidea indica was rarely found from spring through early fall, it did represent 40% of the parasitoid species composition during winter, based on data collected from corrugated cardboard bands wrapped around limbs near the trunk of each tree sampled. Hyperparasitism of *Anagyrus kamali* by resident species (*Marietta* sp. & *Chartocerus* sp.) was frequently over 35% during 2000. However, hyperparasitism was considerably lower during each successive year, coincident with declining densities of both mealybug and the primary parasitoid host. Field collections of two non-target species of mealybugs common in Imperial Valley demonstrated that they are not being utilized as alternate non-target hosts by the newly introduced parasitoids.

No Designated Session Theme

**STUDY OF POPULATION PARAMETERS OF SUNN PEST,
EURYGASTER INTEGRICEPS, EGG PARASITIDS *TRISSOLCUS
GRANDIS* AND *T. SEMISTRIATUS* (HYMENOPTERA:
SCELIONIDAE)**

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Sunn pest, *Eurygaster integriceps* Put. (Heteroptera: Scutelleridae), is the most important insect pest of wheat and barley in Iran. Yield losses from damage are estimated at 20-30% in barley and 50-90% in wheat if this pest is not controlled chemically. This pest is attacked by a number of generalist predators as well as more specialized parasites, the most important of which are hymenopteran egg parasitoids belonging to the genus *Trissolcus*. It is estimated that egg parasitoids reduce *E. integriceps* populations by ~23% each year in Iran. Life table parameters of *Trissolcus grandis* Thomp. and *T. semistriatus* Nees were determined under laboratory conditions. The second-generation (F₂) of parasitoids was used in all experiments. Twenty-five randomly chosen young female adults (<24 hr old) were used for life table studies of the parasitoids. Daily Schedules of mortality and fecundity were integrated into a life table format (Carey 1993) and used to calculate net reproductive rate (R_0), mean generation time (T), and intrinsic rate of increase (r_m).

Life table parameters of sunn pest *Eurygaster integriceps* egg parasitoids *Trissolcus grandis* and *T. semistriatus*

Parasitoid species	Longevity (day)	Proportion of males: m/(f+m)	Net reproductive rate (R0)	Intrinsic rate of increase (rm)	Generation Time (T)	Doubling Time (DT)
<i>T. grandis</i>	38.6±5.4	0.37±0.08	118.37	0.298	15.81	2.32
<i>T. semistriatus</i>	36.3±2.8	0.33±0.06	104.99	0.297	15.67	2.33

No Designated Session Theme

STRAWBERRY ROOT BEETLE BIOLOGY AND CONTROL MEASURES USING ENTOMOPATHOGENIC NEMATODES

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Strawberry root beetle, *Mimela schneideri* Ohaus (Coleoptera: Scarabaeidae) has been a invasive white grub extensively destroying roots of strawberry growing the northern region of Thailand. The 2.5 mm long, oval creamy white eggs were laid in soil during April-May, hatching within 10-24 days. The first, second larval instars obviously having creamy white body and brown head capsule, the third showing yellow skin were found about 5-20 cm under soil surface. Late third instar moved further to the depth of 30 cm or more to pupate in the formed earthen cells. Root damage to Strawberry caused mostly by the 2nd and 3rd instars occurred from May to October. The pupal period was between November and April. Adult emergence began in the last week of April and followed by the mating activities which happened 1-2 hours after sunset. The elongate and oval body of adults showed pale pea green with a very faint golden luster.

The efficacy of three species of entomopathogenic nematodes *Steinernema carpocapsae* (Weiser), *S. glaseri* (Steiner) and *S. riobrave* (Cabanillas, Poinar & Raulston) (Rhabditida: Steinernematidae) for controlling the strawberry white grubs was evaluated in the field at Doi Angkang, Research Station of Royal Project Foundation, Chiangmai province. The experimental design was RCB with 4 treatments and 4 replications. The concentrations of the three species of nematodes were applied at the rate of 1×10^6 IJs/m², in comparison with chlorpyrifos

(Lorsban 40% EC). The result showed that *S. glaseri* was significantly more efficient than *S. carpocapsae* and *S. riobrave*. The percent of dead strawberry plants occurred by the white grub - damaged roots in the treatment of spraying *S. glaseri* was 3.8% as low as in the chlorpyrifos treatment whereas the treatment of spraying *S. carpocapsae* and *S. riobrave* were 6.3% and 6.5% respectively (Table 1).

Table 1. Percent of strawberry plants having the white grubs - damaged root after spraying three species of nematodes and chemical.

Treatments	Percent damage of strawberry plants with their roots attacked by white grubs
<i>S. carpocapsae</i>	6.3 b*
<i>S. glaseri</i>	3.8 a
<i>S. riobrave</i>	6.5 b
chlorpyrifos	3.7 a
CV. (%) = 70	

* In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

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REARING OF *TRIALEURODES VAPORARIORUM* AND *ENCARSIA FORMOSA* ON TISSUE OF SQUASH FRUIT

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The use of artificial diets for rearing insect is a phenomenon that has been developed in order to aid the fields of physiology, ecology, genetics and insect control techniques. The greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) (Hemiptera: Aleyrodidae), is one of the well-known pests affecting several greenhouse-grown crops in Iran. The diversity of natural foods of these whiteflies is very large and larval instars, as well as adults, feed on the phloem sap of hundreds of species of plants. Three principle requirements in the formulation of a diet are: that it stimulates the insect to feed on an unfamiliar food; it must possess all the essential nutrients in balanced preparations needed of normal growth, development and reproduction; it must be free from microbial contamination.

Surfaces of tissue layers of squash fruit were disinfected and then females of *T. vaporariorum* released to oviposit on these layers for four hours and then removed. The layers were then kept in 21 C° to complete immature stages. After 20 days that host population is suitable for parasitism. *Encarsia formosa* Gahan (Hymenoptera: Aphelinidae) was then released for 24 hours and removed. Parasitized pupas were kept in 21 C° to complete all immature stages. All eggs completed immature stages successfully and adults emerged.

No Designated Session Theme

A ROLE OF *PEDIOBIUS SAULIUS* (WLK.) (HYMENOPTERA: EULOPHIDAE) IN THE PARASITOID COMPLEX OF THE HORSE CHESTNUT LEAFMINER, *CAMERARIA OHRIDELLA* DESCHKA & DIMIC (LEPIDOPTERA: GRACILLARIIDAE)

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Pediobius saulius (Wlk.) (Hymenoptera: Eulophidae) is a wide spread in Europe parasitoid on leafminers. Investigations on natural enemies of *Cameraria ohridella* Deschka & Dimic (Lepidoptera: Gracillariidae) conducted during last ten years showed that *P. saulius* was the dominant in the parasitoid complex of the moth mainly in Balkans. Study on biology and impact of *P. saulius* was conducted during the period 2001-2003.

Parasitoid biology. It was established that the parasitoid could be reared in laboratory conditions. The longevity of *P. saulius* depends on its diet. Females start their searching behavior 6-7 days after emergence. *P. saulius* has a synovigeny. Host feeding on prepupae and pupae was observed. *P. saulius* is a primary endoparasitoid on prepupae and pupae of the host. It is a koinobiont.

Impact. The information is given on the relative abundance of *P. saulius* of each moth generation during three seasons with different infestation level. The phenology of *P. saulius* is compared with that of *C. ohridella*. The impact of *P. saulius* on the population dynamics of *C. ohridella* in natural and urban stands of horse chestnut in Bulgaria is discussed.

 No Designated Session Theme

**TWO DIFFERENT SPECIES UNDER ONE NAME:
MORPHOLOGICAL, BIOLOGICAL, AND MOLECULAR
COMPARISON BETWEEN THE DIFFERENT GEOGRAPHICAL
POPULATIONS OF *ANAGYRUS PSEUDOCOCCI*, A WELL-
KNOWN AGENT USED IN BIOLOGICAL CONTROL OF CITRUS
AND VINE MEALYBUGS**

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Anagyrus pseudococci (Girault) (Hymenoptera: Encyrtidae) is a well-known primary parasitoid of *Planococcus* spp. (Hemiptera: Sternorrhyncha: Pseudococcidae) and has been used for biocontrol purposes in many countries. Over the course of a classical biocontrol project against the vine mealybug, *Planococcus ficus* (Signoret) (Hemiptera: Pseudococcidae), in California, USA, cultures of *A. pseudococci* of different origin were established at the University of California, Riverside quarantine laboratory, from the populations in Argentina, Israel, Italy (Sicily), Spain, Turkmenistan, and USA (Coachella Valley, California). Morphological and molecular studies as well as cross-breeding experiments revealed that the Argentine population (first funicle segment of the female antenna half black, half white) is also reproductively and genetically isolated from all other populations tested (first funicle segment of the female antenna entirely black), which cross-bred freely between each other and are genetically very similar. The type series of *A. pseudococci* (from Sicily) is morphologically identical to the population from Argentina; the latter apparently represents the real *A. pseudococci* and could have been unintentionally introduced there with the grape seedlings from Italy. The other, better known Mediterranean form (first funicle segment of the female antenna entirely black, previously introduced in California), thus belongs to a different, currently unnamed species which may coexist in Sicily with the real *A. pseudococci*.

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FILM DOCUMENTATION OF THE BEHAVIOUR OF PREDATORS OF THE TWO-SPOTTED SPIDER MITE *TETRANYCHUS URTICAE*

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This video film (15 minutes duration) shows at first how *Tetranychus urticae* Koch (Acarina: Tetranychidae) females remove food from epidermal cells of bean plants. It then focuses on the precopulatory behaviour of males that wait on resting female deutonymphs and combat male competitors by means of their protruded chelicerae. Upon ecdysis, females are immediately inseminated. Details of egg deposition and egg hatching are shown accelerated. The life cycle from egg hatching to adults is briefly documented. The introduction closes with sequences of mite dispersal on firm webbing strands and the formation of massive aggregations when food sources are depleted.

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Sequences of the foraging behaviour of *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae), the first predator presented, show how females locate prey and ingest food from eggs and captured mobile stages. A short sequence of the complex mating behaviour precedes a longer sequence, which demonstrates in detail how a female deposits its large egg. Further documentations of the life cycle include egg hatching, ecdysis processes and the predatory behaviour of proto- and deutonymphs. The foraging and feeding activities of another predatory mite, *Amblyseius californicus* McGregor (Acari: Phytoseiidae), are briefly demonstrated.

The tiny ladybird beetle *Stethorus punctillum* Weise (Coleoptera: Coccinellidae) has recently gained special attention as it also effectively controls the carmine spider mite *Tetranychus cinnabarinus* (Boisduval) (Acarina: Tetranychidae), against which *P. persimilis* is apparently not as effective as against *T. urticae*. Sequences of the predatory behaviour of *S. punctillum* show how adults and larvae feed upon eggs and mobile stages of both *Tetranychus* species. Adult beetles hunt for prey and then devour it completely, whereas repeated regurgitation is required for larvae to feed on captured mobile stages. *Tetranychus* eggs are sucked out within a few seconds by adult and older larval instars. Short sequences of the life cycle show the following features: mating behaviour, egg deposition, hatching from eggs, ecdysis, pupation and adult emergence from the pupa.

The larvae of the predatory gall midge *Feltiella acarisuga* (Vallot) (Diptera: Cecidomyiidae) usually stay immobile within dense *T. urticae* populations, where they wait for prey that is seized by sudden head movements. Sticky excretions glue the legs to the tiny

mouthparts, by means of which paralyzing substances are obviously injected before the contents of the prey are sucked out by a continuous and very rapid pharyngeal pumping action. Locomotion is required for predation upon eggs that are emptied within a short time by older larval instars. Newly hatched larvae require several hours to consume eggs completely. Older *F. acarisuga* larvae are well adapted to move on spider mite webs, where they show their typical ambush behaviour. Sequences of the life cycle document pupation and the emergence of a male midge from the pupa.

The film terminates with sequences of the predatory behaviour of the bug *Macrolophus caliginosus* (Wagner) (Heteroptera: Miridae), which is well adapted to forage in mite web-bings, although frequent grooming is then necessary. Preferred food sources are eggs, which are located accidentally by random proboscis probings and are then emptied within a few seconds by adult bugs.

No Designated Session Theme

INDUCED PLANT RESISTANCE AND INSECT FEEDING BEHAVIOR: A COMPARISON BETWEEN A CHEWING AND A PIERCING-SUCKING INSECT

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Laboratory experiments were done to compare the effects of plant growth promoting rhizobacteria (PGPR), an elicitor of induced systemic resistance (ISR), and Actigard[®], an elicitor of systemic acquired resistance (SAR), and combinations on feeding behavior of cucumber beetles (Coleoptera: Chrysomelidae) and aphids (Hemiptera). Results of cage experiments with cucumber indicated that cucumber beetle feeding was significantly greater on Actigard[®]-treated plants than on untreated control plants. However, the combination of PGPR plus Actigard[®] resulted in significantly lower feeding damage than occurred with Actigard[®] alone; suggesting that simultaneous induction of ISR by PGPR mitigated the stimulatory effect of Actigard[®]. Additional experiments to compare PGPR and Actigard[®] treatment effects on aphid feeding behavior were conducted using electronic monitoring of insect feeding (EMIF) techniques. Results of the EMIF experiments indicated that aphids feeding on cucumber plants where SAR was elicited by Actigard[®] exhibited an increased number of stylet probes and spent less time in phloem than aphids on untreated plants. As in the cucumber experiments, simultaneous induction of ISR by PGPR (in the combination treatments) mitigated this effect.

No Designated Session Theme

BIOLOGICAL CONTROL OF CASSAVA GREEN MITE IN THE HIGHLANDS OF CAMEROON - TESTING THE LIMITS OF A NEOTROPICAL PREDATOR

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Typhlodromalus aripo (DeLeon) (Acari: Phytoseiidae) is a neotropical predator which was first introduced into Africa in 1993 for the control of cassava green mite *Mononychellus tanajoa* (Bondar) (Acari: Tetranychidae). At present, *T. aripo* is established in 20 countries of sub-Saharan Africa. But the predator has been slow in colonizing and establishing in mid-altitude and highland regions (>1100 m asl) of the higher latitudes (> 4°N), which are climatically less favourable areas with cooler temperatures and more pronounced dry seasons. In our field release study, we tested a strain adapted to the mid-altitudes (Minas Gerais-Brazil) and compared it to the earlier released lowland strain (Piritiba-Brazil), in both the mid-altitude and the lowland ecology. Seeing that *T. aripo* prefers some cassava cultivars over others, we introduced two varieties which were new to the area, and compared their ability to host the predators to the local varieties. The predators were released in planted cassava fields in various habitats such as mid-altitude and low-altitude climates, savannah hill slopes, compound surroundings and riparian forests. The fields were monitored monthly, over two cassava cropping cycles (16 months per cycle).

We found that, unlike in other regions of sub-Saharan Africa, *M. tanajoa* populations peaked at the end of the dry season (or shortly after the beginning of the wet season) and not shortly after the onset of the dry season. At the same time, predator abundance dropped to very low levels in the dry season, and recovered only four to eight weeks after the beginning

of the rainy season. Despite the asynchrony with its prey, *T. aripo* was able to persist in both lower and higher altitudes for more than one year, most probably because of its ability to develop on alternative food (e.g., grass pollens and extrafoliar exudates). The great challenge for the predator's long term persistence in the mid-altitudes, however, was crop harvest: *T. aripo* did not spread to neighbouring fields, and wasn't able to persist beyond one cropping cycle. This is different to the lower altitudes where *T. aripo* spread and persisted for more than one cropping cycle. No differences were found between the two strains, regardless of altitude. Humid and fertile habitats such as fields near compounds promote vigorous plant growth and higher plant turgidity which facilitate the persistence of *T. aripo* longer into the dry season. Further, the more extreme the climatic conditions (e.g. in savannah hills in the mid-altitudes), the more important were host plants preferred by *T. aripo* (featuring hairy apices) for its survival. With the help of hairy varieties, and over-seasoning pools in fertile habitats nearby, it might be possible to establish *T. aripo* in these difficult environments, which are typical for cassava growing areas. But effectiveness of *T. aripo* to control cassava green mite in mid-altitude areas of higher latitudes is doubtful, because of its asynchronous cycle with the pest mite, and because of its low dispersal capacity in those environments.