

General News

TEAMwork against Leafy Spurge

Biological control programmes for leafy spurge (*Euphorbia esula* complex) in North America have made significant progress since *BNI*'s last report on the problematic Eurasian perennial [*BNI* 18(3), 65N (September 1997)].

Although 15 biological control agents for leafy spurge have been released in the USA since 1963, two in particular are well established throughout the continental 'spurge belt' in the northern Great Plains (North and South Dakota, Montana, Wyoming and the Prairie Provinces of Canada). The two European flea beetles – *Aphthona nigricutis* and *Aphthona lacertosa*, first released in the USA in 1988 and 1994, respectively – have established large populations at numerous locations across a wide geographic area, and are now providing significant levels of leafy spurge control. Perhaps more importantly, this build-up of numbers and sites is allowing for easy collection and redistribution of flea beetles, which further promotes the concept of biological control.

The past 2 years have been especially exciting for those involved with leafy spurge management. Although reductions in spurge densities due to flea beetle activity have been documented at many locations for several years, never before have there been so many dramatic reductions spread across such a wide area. At some sites, extremely heavy infestations were completely eliminated. Biocontrol worked so well in some places, in fact, that collection efforts were hampered: people returned to sites that had historically produced large numbers of flea beetles to discover that both the spurge and flea beetles were gone. This phenomenon can be expected to become more common as flea beetle populations increase and more sites are established, and it emphasizes the need to educate people about properly managing their biocontrol resource.

A major development in leafy spurge management occurred in 1997, when the US Department of Agriculture (USDA) Agricultural Research Service joined forces with a sister agency, the USDA Animal & Plant Health Inspection Service, to create 'TEAM Leafy Spurge' (TLS).

This biologically based Integrated Pest Management (IPM) programme focuses on researching and demonstrating effective, affordable and ecologically sustainable leafy spurge management techniques. The cooperative programme stresses teamwork, resulting in a vast network of partnerships between the two USDA agencies, land grant universities and numerous other local, state and federal entities. Fields of study include (but are not limited to) classical biological control, multi-species grazing, herbicide use, cultural controls (such as tillage, reseeding and burning), the integration of various control tools, socio-economic impacts, the application of GPS/GIS and hyperspectral imaging technologies to weed management, and foreign exploration to investigate new leafy spurge biocontrol agents.

Data collected by TLS programme participants in the past 3 years are especially promising, and suggest that widespread control of leafy spurge with biologically based IPM strategies is no longer a question of 'if' but 'when'. This is a brief summary of some TLS research efforts:

Biocontrol

Flea beetle establishment has improved dramatically the past 3-5 years, primarily because people now know more about using biocontrol and have access to large numbers of insects. A few years ago, releases of 100-500 flea beetles were common, with roughly one-third of all releases successfully establishing a population; releases of 3000 flea beetles are now considered the minimum, and establishment success has more than doubled. In an effort to quantify flea beetle establishment, population expansion and the resultant impact on leafy spurge, TLS established 264 'inventory and assessment' sites in 1998. Each site was inventoried – i.e. extensive data regarding soil type, moisture, topography, species composition, etc. were collected – prior to being seeded with 6000 *Aphthona* flea beetles (3000 *A. lacertosa* and 3000 *A. nigricutis*). Data collected from these sites in the summer of 2000 document an establishment success rate of 85-95%, a seven-fold increase in flea beetle numbers and spurge canopy cover reductions in the range 40-95%. In addition, grass production and species rich-

ness (diversity) increased by averages of 47% and 27%, respectively.

Multi-Species Grazing

TLS is demonstrating that a combination of multi-species grazing and biocontrol works well as an effective spurge management tool. The concept is simple: use sheep to graze heavy, dense patches of spurge, thus giving flea beetles increased opportunities to establish populations in the resultant thinner stands of spurge. For this demonstration, TLS selected a site that originally featured an extremely dense, widespread spurge infestation (more than 50% of the total demonstration area was infested with spurge exceeding 200 stems/m²). In just 3 years, the combination of multi-species grazing and biocontrol has reduced spurge densities by 31-50%; native vegetation and desirable grasses are responding by reestablishing in areas formerly occupied by spurge. Based on previous research, significant reductions are expected to occur in the 4th and 5th years (i.e. 2001 and 2002) of the demonstration. The demonstration clearly shows the economic and environmental advantages offered by combining the two biologically based IPM strategies.

Herbicides

Herbicide use is declining in areas where biologically based IPM programmes have been implemented. Ward County, located in northwestern North Dakota, provides an excellent example. Ten years ago, weed managers there invested considerable resources (both fiscal and personnel) into spraying 8000 of the 12,000 acres [3250/4850 ha] infested with spurge within the county. Last year, after just 3 years of aggressive biocontrol efforts, the county had 9500 acres [3850 ha] of spurge (a 20% reduction in total acreage) and used herbicides on just 400 acres [160 ha] (a 95% reduction). Similar examples can be expected from across the region as weed managers learn how to use biological control as a stand-alone tool and in combination with other tools.

In addition to creating cooperative partnerships and funding various research and demonstration projects, TEAM Leafy Spurge also stresses information and education. Although a variety of tools (press releases, magazine articles, pamphlets,

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CDs, etc.) are used to increase awareness of the problem and potential solutions, TLS has discovered that low tech, grass-roots efforts such as field day events are extremely valuable in regard to teaching people about biocontrol and IPM.

The programme's information and education efforts will be highlighted at 'Spurgefest II', set for June 2001. The 3-day event will include a leafy spurge symposium, tours of TLS research and demonstration sites, and demonstrations of flea beetle collection and redistribution techniques. Its precursor, 'Spurgefest '99', was wildly successful, capturing a considerable amount of media interest across the northern Great Plains and drawing participants from across the USA and Canada.

For additional information about TEAM Leafy Spurge or Spurgefest II, see the TLS website at:

<http://www.team.ars.usda.gov>

or email: teamls@sidney.ars.usda.gov

By: Steve Merritt, TEAM Leafy Spurge technology transfer specialist, USDA-ARS Northern Plains Agricultural Research Laboratory, 1500 N. Central Ave., Sidney, MT 59270, USA

Email: smerritt@sidney.ars.usda.gov

Fax: +1 406 433 5038

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Weevils' Success against Canadian Rangeland Weeds

Progress on projects involving the classical biocontrol of two rangeland weeds, Dalmatian toadflax (*Linaria dalmatica*) and houndstongue (*Cynoglossum officinale*), was significant in 2000. Both projects involve success with relatively recent introductions of European insects, which were screened by the CABI Bioscience Centre in Switzerland with funding from Canadian and US sponsors.

Dalmatian Toadflax

This perennial with the bright yellow, snapdragon-shaped flowers is an invasive weed of grasslands, open forests, roadsides and waste areas of western North America. Originally introduced as an ornamental from eastern Europe in the 1900s, Dalmatian toadflax has since become a serious problem in Canada, particularly in the southern interior of British Columbia (BC) and contiguous areas of southwest Alberta, where it currently infests thousands of hectares of range and forest lands. An extensive root system and strong, early spring growth allows Dalmatian toadflax to out-compete surrounding rangeland vegetation.

The weed also is a prolific seed producer, thus contributing to its continued spread and invasion into new areas. Cattle and wildlife generally avoid grazing on Dalmatian toadflax. Cattlemen in BC have listed the weed as their third priority for control after the knapweeds (*Centaurea* spp.) and houndstongue, because of the loss in grazing potential of toadflax infested lands.

The options for control of Dalmatian toadflax are few. Chemical treatment is uneconomical, difficult because of the plant's deep roots and waxy leaves, and potentially environmentally damaging when applied to large weed stands on grasslands. Furthermore, habitats where toadflax often grows, such as coarse-textured soils or near water, restrict the use of effective herbicides (e.g. picloram). Mechanical control (i.e. hand-pulling or mowing) also has not proven feasible. Biological control is considered the best long-term control option.

A European stem-boring weevil, *Mecinus janthinus*, first released in 1991 in Canada against Dalmatian toadflax, has established well and has produced major attack on the weed in southern BC. Monitoring of several 5- to 6-year-old release sites in 2000 revealed 100% attack of toadflax shoots for at least 50 m around the original release points. In some instances, this level of attack was evident for several hundred metres. Most of the damage is attributed to spring feeding on shoot tips by mass-emerging adults, thus causing significant stunting and a complete loss of flowering on reproductive shoots. The insect's ability to cause such impressive damage to toadflax was not predicted from earlier European studies. Although it has been too soon to detect actual reductions in toadflax density at most sites, we continue to monitor permanent plots to document any changes in vegetation.

Houndstongue

Houndstongue is a biennial or short-lived perennial weed of mountainous rangelands in northwestern North America. Originally from Eurasia, the weed is thought to have been accidentally introduced in the 1800s and has since spread considerably. The weed thrives particularly in forest openings created through logging or mining activities, sometimes forming dense monocultures in these habitats.

In British Columbia houndstongue is a major concern to cattlemen because it hinders establishment of forage on new pastures and its barbed seeds or 'burrs' attach to cattle, causing irritation, potential reductions in auction price of animals, and a negative impact on the rancher's reputation. Houndstongue also is highly toxic to live-

stock, possessing pyrrolizidine alkaloid levels which are much higher than those found in another toxic range weed, tansy ragwort (*Senecio jacobaea*). Normally, livestock avoid feeding on green houndstongue, but problems may arise once the plant dies back in late summer or fall, or if it happens to get into hay.

As with Dalmatian toadflax, options for houndstongue control are limited, hence, a biocontrol programme was initiated in 1988 with European exploration for potential agents. The root weevil, *Mogulones cruciger*, was the first agent to be released in Canada after 9 years of host specificity testing. Since the initial releases in 1997, the weevil has established at all sites so far (i.e. 67 by the end of 1999) and already is showing evidence of reducing houndstongue density at the oldest sites. Monitoring of target and any potential nontarget impact will be of upcoming emphasis in our programme.

By: Rose De Clerck-Floate, Agriculture and Agri-Food Canada, Lethbridge Research Centre, 5403 - 1 Avenue South, P.O. Box 3000, Lethbridge, Alberta, Canada, T1J 4B1
Email: Floate@em.agr.ca

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Azolla Biocontrol in South Africa

Azolla filiculoides or red water fern is a free floating aquatic fern native to South America. It was first recorded in South Africa in 1948. For many years the fern was confined to small streams and farm dams in the Colesburg area in the centre of the country. However, the lack of natural enemies and the presence of enriched waters have contributed to its inevitable spread by man, waterfowl and floods to many sites around the country. By 1998 the weed had been recorded at some 176 sites.

The increasing abundance of *A. filiculoides* in conservation, agricultural, recreational and suburban areas over the last 10 years is cause for great concern. Among the major consequences of the dense mats (5-30 cm thick) of the weed on still and slow-moving water bodies in South Africa are: reduced quality of drinking water caused by bad odour, colour and turbidity; increase in waterborne, water-based and water-related diseases; increased siltation of rivers and dams; reduced water surface for recreation (fishing, swimming and water-skiing) and water transport; deterioration of aquatic biodiversity; clogging of irrigation pumps; drowning of livestock; and reduced water flow in irrigation canals.

In South Africa, no herbicides are registered for use on red water fern, and this, in conjunction with the fact that its rapid rate of increase renders manual and mechanical control ineffective, suggested that biological control was the most suitable option for this weed. The frond-feeding weevil *Stenopelmus rufinasus* was collected from *Azolla caroliniana* in Florida (USA) and imported into quarantine in South Africa in late 1995. Following host specificity testing, the weevil was released in December 1997. The first release, of 900 weevils, was made on a one-hectare dam in a bird sanctuary in Pretoria, which was 100% covered by the weed. By February 1998 (2 months later) the red water fern mat had collapsed, and some 30,000 weevils were reared from one 2 m² sample of decaying material.

To date the weevils have been released (usually in batches of 100 adults) at some 110 sites throughout South Africa. The information that we have on these sites is that the weevil has been responsible for clearing 72 of them completely. For the remaining 38 sites, either the weed has been washed away during flooding, or we have not revisited them, or it is too early to tell. All of the sites that have cleared have done so in less than one year. In addition to this, the weevils have migrated to other sites, sometimes up to 80 km away from the release site. We are uncertain if the weevils have been transported on weed by waterfowl, or if there has been short distance hop dispersal onto other dams of the weed, or if it is as a result of long-range dispersal by the adults. At around 40% of the sites the weed has returned up to 2 years after the initial clearing. Interestingly the weevils have located 90% of these and the weed is under control.

A thorough cost benefit analysis of this project has been completed and is in the process of being published; the preliminary benefit to cost ratio is 1200:1.

There are several interesting aspects to this project. Firstly, this appears to be a new association: although *A. filiculoides* has been recorded as native to the southwestern USA, this weevil is associated with *A. caroliniana* in Florida. Secondly the speed with which weevils were able to control even the largest mats in the most eutrophied waters. Thirdly, the weevils have been able to locate red water fern mats up to 18 months after the original mats had collapsed. It remains to be seen if the current level of control will be sustained over the long-term.

Contact: Andrew McConnachie,
Ecophysiological Studies Research Programme (ESRP),

Animal, Plant & Environmental Sciences,
University of the Witwatersrand,
Private Bag 3, Wits 2050, South Africa
Email: andrewm@gecko.biol.wits.ac.za

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Taste of Its Own Medicine?

Search for *Rhinocyllus conicus* references in recent literature, and you are likely to come up with a host of publications discussing the nontarget effects of this introduced weevil on native thistles in North America. However, down in New Zealand, it is a different story, for there the weevil has itself become the subject of nontarget attack.

The braconid *Microctonus aethiopoidea*s was introduced to New Zealand in 1982 as a biocontrol agent for the weevil *Sitona discoides* on lucerne, and has been useful in reducing damage in this important crop. Now, however, it has been found attacking *R. conicus* in some agricultural environments. *Rhinocyllus conicus* was released in New Zealand as a biocontrol agent for nodding thistle (*Carduus nutans*), and in conjunction with other agents often provides excellent control. The results of preliminary studies suggest that *M. aethiopoidea*s is likely to have limited impact on *R. conicus* in the field, but this is not the first instance of this parasitoid exhibiting a catholic taste for hosts in New Zealand. It has previously been recorded from three other weevil species in the field here (*Nonotus albicans* and two *Irenimus* spp.) with parasitism rates of 40% or more.

The nontarget effects recorded with *Microctonus aethiopoidea*s contrast with another species of the genus, *M. hyperodae*, introduced more recently for control of another weevil pest of lucerne, *Listronotus bonariensis*. This second parasitoid has so far proved to be restricted to its target host. The difference reflects at least in part the increase in awareness of environmental risk that has occurred in recent years: greater rigour in testing procedures meant that the narrow host range of *M. hyperodae* was predicted with a good degree of reliability.

Sources: Watch out, *Microctonus* is about. Lincoln, New Zealand; Manaaki Whenua – Landcare Research. What's new in biological control of weeds, No. 16 (November 2000), p.8.

Internet: <http://www.landcare.cri.nz>
Lynch, L.D.; Thomas, M.B. (2000) Nontarget effects in the biocontrol of insects with insects, nematodes and microbial agents: the evidence. *BNI* 21(4), 117N-130N.

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Florida Fast Food

Our 'Florida Stripper' [*BNI* 21(3), 57N (September 2000)], the melaleuca weevil *Oxyops vitiosa*, has developed a taste for an artificial diet. This does not mean it will be invading hamburger bars, but it does make it much easier to rear. Ultimately, the success of a biocontrol programme relies on both having appropriate agents, and being able to mass rear and release them. This rearing technique includes both a diet and a pupation medium. Developed by USDA scientists at the ARS Invasive Plant Research Laboratory in Fort Lauderdale, it gives a real boost to the programme for biocontrol of *Melaleuca quinquenervia* in Florida.

The rearing technique has two key features: first, it contains the right balance of nutrients (including sucrose, glucose, maize starch, vitamins and minerals) and feeding stimulants (*M. quinquenervia* leaf extracts) to support the complete development of the insect from egg to adult; second, a substrate is described that provides a relatively high rate of pupation. Although larval development requires nothing more than the right nutrients, the pupae are more choosy. The substrate that works best included a mixture of sand and water with an absorbent material such as crushed florist's foam or peat moss; such a mixture retains enough moisture and allows enough air exchange for successful pupal development. Artificially reared weevils have already been released at sites infested with melaleuca in south Florida and appear to be performing well.

Contact: Gregory S. Wheeler, ARS Invasive Plant Research laboratory, Ft. Lauderdale, FL 33314, USA
Email: wheelerg@saa.ars.usda.gov
Fax: +1 954 476 9169

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Mist Flower Flagging?

Mist flower (*Ageratina riparia*) spreads at a phenomenal rate, and in New Zealand the public has been engaged to help to monitor this. From the results of a questionnaire, researchers at Landcare Research have drawn up a map of its current distribution and, by identifying the environmental conditions in which it grows, have also drawn up a 'worst-case' map showing the areas it could potentially invade. Presently most common in the north of North Island, it could spread to other parts of North Island and the top of South Island. However, with the help of two exotic biocontrol agents with a track record of success in Hawaii, its spread may soon be slowed if not halted.

The white smut *Entyloma ageratinae* was the first agent to be released against mist flower in New Zealand [*BNI* 20(4), 107N (December 1999)], and now an insect agent has also been approved for release by the Environmental Risk Management Authority. A shipment of the gall fly *Procecidochares alani* has been imported from Hawaii, and releases are expected to begin this summer. It is hoped the insect will complement the striking success of the pathogen.

The smut, which was released in November 1998, has been spreading even faster than its mist flower host. It has been recorded up to 56 kilometres from release sites in the north of North Island. In the Waitakere Ranges, the spread has been so effective that by December 1999 no uninfected areas could be found for studies into long-term vegetation changes. Even more unexpectedly, an intrepid pathologist who ventured out to Great Barrier Island to release the smut found it had beaten her there. Although it may have arrived on the wind (the nearest release site is on Waiheke Island, 77 km away), Jane Fröhlich suggests it is equally likely to have been taken there by an unwitting human carrier.

There are also encouraging signs that the fungus is causing significant damage to the weed. At the nine sites where the fungus was released, 30-50% of the mature leaves last summer were destroyed in the first wave of attack, and 30-90% of the regrowth that season met a similar fate. Monitoring will continue this summer. However, there are signs that the defoliation at release sites is already allowing the recovery of some native plants, including orchids and ferns, that had been choked out by mist flower.

Sources: History sometimes repeats. Lincoln, New Zealand; Manaaki Whenua – Landcare Research. Patua te otaota – Weed clippings. Biological control of weeds annual review 1999/2000, p. 3.

Drawing up enemy lines. Lincoln, New Zealand; Manaaki Whenua – Landcare Research. What's new in biological control of weeds, No. 16 (November 2000), pp.5-6. Internet: <http://www.landcare.cri.nz>

Contact: Jane Fröhlich, Landcare Research, Private Bag 92170, Mt Albert, Auckland, New Zealand
Email: FrohlichJ@landcare.cri.nz
Fax: +64 9 849 7093

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Rust for Bridal Creeper

Bridal creeper (*Asparagus asparagoides*) is one of Australia's most damaging and persistent environmental weeds, and a search

for effective control is well underway [*BNI* 20(4), 108N-109N (December 1999)]. In the last 2 years two defoliating biocontrol agents have been released against it by researchers from CSIRO (Commonwealth Scientific and Industrial Research Organisation) and the Cooperative Research Centre (CRC) for Weed Management Systems.

The leafhopper *Zygina* sp. was released in May 1999 across southern Australia. It successfully overwintered at the majority of sites. At two research sites the insect has spread 300 m from the release site after 18 months. Schools and community groups are involved in rearing and release of the insect. It has now been released at over 200 sites, and is beginning to impact on the weed at many sites, where the onset of senescence is advanced by several weeks. Studies are underway to monitor reserve accumulation in the tubers.

Leafhopper releases were followed in 2000 by releases of the rust *Puccinia myrsiphylli*. There are high hopes for this pathogen, as it causes considerable damage to the creeper in its native South Africa. It was released at 50 sites across all states of southern Australia from mid July until the plants began to senesce with the onset of the hot summer weather in November. The rust established well at most sites, and the epidemic developed steadily until the onset of senescence. Spread, though, was slow from the artificially inoculated release areas. At one site, the rust spread about 30 m within 4 months of release, while at another site along the side of the road it spread as far as 100 m. It caused premature defoliation of the cladodes at the release points at these two sites. The slowness of spread may be related to the microclimate, which is characterized by very little wind movement because the weed tends to grow below other shrubs. However, teliospores (sexual spores) have been produced in abundance. This autumn (February/March) researchers will be able to see if and how the rust reappears at the release sites, and field trials have been set up to monitor the spread and epidemic development of the rust over the next 2 years.

Contacts: Louise Morin, CRC for Weed Management Systems, CSIRO Entomology, GPO Box 1700, Canberra, ACT 2601, Australia
Email: louisem@ento.csiro.au
Fax: +61 2 6246 4362

Tim Woodburn, CRC for Weed Management Systems, CSIRO Entomology, Western Australian Laboratory, Private Bag 5, PO Wembley, Western Australia 6913, Australia

Email: t.woodburn@ccmar.csiro.au
Fax: +61 8 93336646

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Mimosa invisa: a Growing Menace in South India

Mimosa invisa, a noxious fast growing invasive weed of South American origin has recently emerged as a new threat to natural forests, forest plantations and agricultural systems in Kerala State in southwest India. The weed is an annual but wherever water is available year-round it can also grow as a biennial. *Mimosa invisa* has a scrambling stem bearing four or five rows of sharp prickles. Leaves are small, bipinnate and fluorescent green in colour. The inflorescence is a condensed spike (capitate) which is pinkish in colour. The weed produces a large number of seeds which have a long period of viability. It is heliophytic in adaptation and cannot grow well under closed canopy. *Mimosa invisa* is moderately drought resistant. The fact that it can invade and cover the ground completely, competing with other plants, smothering herbaceous growth implies habitat degradation and loss of biodiversity. *Mimosa invisa* growing areas are impenetrable because of the characteristic thick growth and the stem being armed with sharp prickles. It is known to be toxic to cattle.

A preliminary survey conducted in Kerala indicated that *M. invisa* is widespread in the central and southern parts of the State. Of the 52 sites surveyed in seven districts in the State, over 50% were heavily infested. In central Kerala, natural moist deciduous forests are heavily infested. In evergreen and semi-evergreen forests infestation is seen only on the fringes where canopy is partially or fully open. Among forest plantations, teak is seriously affected. The weed grows luxuriantly on roadsides and fallow lands. *Mimosa invisa* is found to overgrow and smother *Mikania micrantha* in most parts of Kerala. It appears that at the present rate of growth and spread, *M. invisa* may even exceed *Mikania micrantha* in posing serious threat to natural forests, forest plantations and agricultural systems in Kerala. The potential combined impact of these two weeds cannot be over-estimated.

Mechanical control of *M. invisa* by manual weeding is difficult and labour intensive. Moreover, the weed can sprout vigorously from the cut base soon after the onset of monsoon. An attempt to control it using 2,4 D and dinitrobutyl phenol (denoseb) in Brazil was not very successful. According to Rachel McFadyen, biological control using insect enemies is showing success in

Queensland (Australia), and attempts to control this weed through biological means are highly warranted in India.

By: K. V. Sankaran,
Kerala Forest Research Institute,
Peechi – 680 653, Thrissur Dist.,
Kerala, India
Email: sankaran @kfri.org
Fax: +91 487 282249

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DNA Fingerprinting: Pointing the Way

Finding biocontrol agents, whether for old or new pests, may superficially seem a straightforward business, but in practice it is very complex. Factors that make it so include (a) incomplete taxonomy of both pests and natural enemies; (b) wide native distribution of pests; and (c) limited information on associations between natural enemies and the target and its related species. Unravelling which are the best natural enemies to introduce, and how many species are needed is a time-consuming and therefore costly process, yet projects are generally severely limited by both. Projects also have a habit of coming to an untimely end: once the agents are out, it can be hard to persuade funding agencies that their progress needs to be monitored.

Molecular biologists have developed a range of techniques, based on multiplying up small variations in DNA to detectable levels, to allow them to distinguish between species, and between populations of the same species. An account of some of these techniques is given in a previous news item¹, and more are being developed. They form a powerful battery of investigative tools with a lot to contribute if applied in a targeted intervention way in combination with other biocontrol research methods. Here we look at what kind of information DNA analysis can provide (drawing particularly on work with whiteflies). We see how this can inform decision-making in biocontrol projects. At this time, we are on a learning curve, and the usefulness and limitations of DNA techniques are still becoming apparent. However as they begin to be more widely understood, they will be used more often and more effectively.

Taxonomic Tool

In the last ten years, the whitefly *Bemisia tabaci* has become a serious worldwide pest, and one of the foremost whitefly vectors in field and glasshouse crops. It is a vector that it poses the most severe threat, as it takes very few individuals to infect and devastate a field with viral disease (many of which are little studied as yet). A

particularly polyphagous, fecund and hardy population of *B. tabaci* was identified in the USA in the early 1990s, which proved also to be a vector for a new arsenal of viruses. It spread rapidly through the New World, and to Europe, Australia, Southeast Asia and the Pacific. It was designated *B. tabaci* biotype-B based on esterase profiles (while the *B. tabaci* population native to the southwestern USA and northwestern Mexico was designated biotype-A). In 1994, biotype-B was given species status as *B. argentifolii* on the basis of allozyme and RAPD-PCR (random amplified polymorphic DNA – polymerase chain reaction) analysis, and viral transmission, morphological and mating studies. However, the status of the biotype/species has remained contentious, as indeed has that of *B. tabaci* as a whole. Many studies of many kinds have indicated wide variation in numerous parameters between different *B. tabaci* populations, and possibly more than would be expected within many species, yet it has not been possible to establish a reliable phylogeny. Recent DNA analyses (PCR amplification and sequencing of two mitochondrial markers (portions of the 16S ribosomal subunit and cytochrome oxidase I genes, COI) and one of the ribosomal internal transcribed spacers (ITS1)) for *B. tabaci* from different locations around the world have provided the first molecular evidence of important genetic divergence between geographically isolated populations^{2,3}. The authors argue that it is more realistic to consider *B. tabaci* as a whole as a species complex, for any alternative would require the description of a separate species for each unique population.

The taxonomy of whitefly parasitoids provides another challenge. Some of the problems were outlined by Andrew Polaszek in an earlier news article⁴. A number of teams are now using DNA techniques to clarify the taxonomy of some groups. RAPD-PCR markers were first developed to help sort out the large number of foreign collections made as part of an interagency whitefly programme (involving the US Department of Agriculture, Agricultural Research Service (USDA-ARS) and Animal and Plant Health Inspection Service (USDA-APHIS), state agencies and university researchers). The USDA-APHIS team based at Mission, Texas has conducted studies using classical taxonomy backed up by RAPD-PCR analysis of *Encarsia* and *Eretmocerus* parasitoids from around the world^{5,6}. Work is also being conducted by USDA-ARS researchers at Fargo, North Dakota, who are using satellite DNA sequences as species-specific markers. Sat-

ellite DNA contains highly repetitive non-coding sequences that accumulate mutations quickly as compared with other parts of the genome. This has the potential to give better information about gene flow between populations and to distinguish strains in rapidly evolving systems.

Conveniently for this morphologically indistinct group, RAPD banding patterns of *Eretmocerus* and *Encarsia* species differ. These unique patterns were found, by comparing them with minute species differences in antenna morphology, to correspond directly to distinct species within the two genera. In this instance, RAPD is a useful quarantine tool for making preliminary separations of geographic populations or strains of *Eretmocerus*, and possibly new species. However, there is not always agreement between traditional taxonomic and DNA techniques. RAPD analysis at the Mission laboratory consistently distinguished between several geographically isolated collections of *Encarsia sophia* (= *E. transvena*) that could not be distinguished by morphological methods.

DNA sequencing (of the D2 expansion region of the 28S rDNA gene) of *Encarsia* species has been carried out by Chris Babcock & John Heraty (University of California at Riverside, USA). Sequencing (of the ITS 1 and 2, mitochondrial COI and D2 and D3 regions) of *Encarsia* and *Eretmocerus* parasitoids of *B. tabaci* and *Trialeurodes vaporariorum* in Australia has been carried out by Paul De Barro & Felice Driver (CSIRO Entomology, Australia). The aim of these studies is to successfully characterize several species and species-groups of these parasitoids, as well as contribute to phylogeny reconstruction. Their studies have shown a strong relationship between morphological characters and molecular phylogenetic structure. This work is continuing at Imperial College, UK (Shahab Manzari, Andrew Polaszek, Robert Belshaw & Donald Quicke), focusing especially on the *Encarsia inaron* species group. As with the Mission work, sequence data have already shown the presence of undescribed species in the *inaron*-group which are morphologically extremely difficult to distinguish.

Contacts: Paul De Barro, Project Leader,
Whitefly Research, CSIRO Entomology,
120 Meiers Road, Indooroopilly, Qld 4068,
Australia
Email: Paul.DeBarro@brs.ento.csiro.au
Fax: +61 7 3214 2885

John A. Goolsby,
ARS-Australian Biological Control Lab,
120 Meiers Rd., Qld 4068, Australia
Email: john.goolsby@brs.ento.csiro.au

or Don Vacek & Matt Ciomperlik, APHIS, Mission Plant Protection Center, P.O. Box 2140 Mission, TX, USA 78573, USA
Email: Don.C.Vacek@usda.gov

Andrew Polaszek, Unit of Parasitoid Systematics, Dept of Biology, Imperial College at Silwood Park, Ascot, Berkshire SL5 7PY, UK
Email: a.polaszek@ic.ac.uk
Fax: +44 207 594339

Forensic Biogeography

DNA analysis has potential for helping in one of the great conundrums of biogeography and classical biocontrol – where species originated and how they have moved, naturally or otherwise, around the globe. By analysing the genetic variation in populations of a pest in its indigenous range and comparing them to samples from its introduced range, it is often possible to pinpoint where the introduced material originated. For an introduced pest with a very large home range, this is probably a good place to start the search for a biocontrol agent, as classical theory tells us that this is where effective co-evolved natural enemies are most likely to be found.

The area of origin of the target pest is not always easy to pinpoint, particularly for pandemic species. For example, cypress aphid (*Cinara cupressivora*) was introduced to Africa in the late 1980s, with immediate and devastating impact on plantation and smallholder trees throughout the continent. Classical biological control was deemed to have the best chance of providing an effective long-term solution, but there was a catch. Cypress trees are grown all over the world, in subtropical and temperate climates, and exactly where the pest in Africa had come from was a mystery. To add to the confusion, the identity of the species in Africa was unclear (indeed, originally thought to be *Cinara cupressi*, it was later described as a new species). Beginning in 1992, CABI and collaborating scientists from many countries searched exhaustively (and exhaustedly) on cypress trees throughout North and Central America, and from the Atlantic coast of western Europe, through Central Europe, the Mediterranean, North Africa and the Middle East to Pakistan. Eventually, the probable source was tracked down to Syria, to cypress trees nestling below the ruins of castles built during the Crusades many hundreds of years ago, which proved to be the centre of origin of *C. cupressivora* (although it is now also found in the southern Mediterranean region). Doubtless, the castle walls have sheltered many a

wary traveller in their time, but probably never before a footsore exploratory entomologist. For the search had taken 5 years, and during this time, although the biological control efforts had continued in tandem with the searches, they were hampered by the uncertainties. DNA analysis would not have obviated the need for all the exploration, nor for climate matching that directed the searches, nor for the morphometric studies that finally clarified the taxonomy. But by using DNA techniques in combination with conventional ones, not only might taxonomic uncertainties be resolved more easily, but promising areas to focus on for natural enemy exploration may be pinpointed more quickly.

Taking up the *Bemisia tabaci* story again, we can see how. Although, based on species and natural enemy diversity, the centre of origin of *B. tabaci* was believed to be either the Indian subcontinent or the Middle East/Africa, there was no indication where biotype-B had sprung from. Host plant affiliations suggested perhaps an origin somewhere in the Eastern Hemisphere, and this was supported by preliminary DNA sequence analysis (of the mitochondrial 16S ribosomal gene). Recent studies using DNA techniques have made a significant contribution to unravelling the convoluted biogeographical relationships within the species and of its parasitoids. In two of these^{2,3}, putting ITS1, 16S and COI profiles into analytical models gave consistent results that distinguished New and Old World populations, and separated Old World populations into a number of groups, including Africa; Sahel and Sahel-like regions; Australia; and several distinct clades in Asia. Biotype-B was found to fit with the ‘Sahel’ group, and this provided the first definitive molecular evidence to support the hypothesis that biotype-B is an introduction into the New World from the Old World. It further suggested an area of origin in the eastern Mediterranean/North Africa. This fitted with features of its ecology, and with observations that only populations from the ‘Sahel’ group produced the phytotoxic symptoms associated with biotype-B in the New World. Further, the molecular data supports the hypothesis that the unique physiological changes induced through the feeding of biotype-B is a recently evolved trait.

In another study⁵, DNA analysis and classical taxonomy were demonstrated to be a powerful combination. They helped to establish the identities and predict the putative centres of diversity of both *B. tabaci* hosts and their parasitoids, using material from southern Spain, Thailand and Texas (a target site for the biocontrol programme).

Morphological analysis indicated the Spanish whiteflies to be a non-biotype-B, and those from Texas to be biotype-B, while those from Thailand were identified as ‘*B. tabaci* species complex’. Molecular classification (based on sequencing of the mitochondrial COI gene) also suggested that the Spanish material was a distinct local variant. However, comparison with similar analyses of other *B. tabaci* populations indicated that it fitted into a clade containing reference B-biotypes from several locations, including Texas. Molecular identification of the Thailand whiteflies placed them in a Far East-Southeast Asian clade. The *Eretmocerus* data from the Australian study³ suggests that the species of effective *Eretmocerus* species from Spain, Pakistan, the Middle East and Australia are all very closely related. These results can be used in combination with other criteria such as climate matching to make decisions on priorities for surveys and testing of potential agents.

Diadegma Dilemma

The area of origin of natural enemies may be far from apparent. It is not uncommon for a natural enemy to appear serendipitously, having been introduced to a new area along with the pest. The diamondback moth (DBM), *Plutella xylostella*, is the most important pest of crucifers worldwide. It is notorious for developing pesticide resistance and, therefore, biological control has been widely and successfully deployed in highland growing areas in Asia. The most important parasitoid is an introduced ichneumonid wasp of European origin, *Diadegma semiclausum*. This parasitoid is now the target of a new biocontrol attempt in East Africa.

Earlier efforts by the GTZ/ICIPE (Gesellschaft für Technische Zusammenarbeit/International Centre for Insect Physiology and Ecology) IPM Horticulture Project in cooperation with a number of national research organizations in eastern and southern Africa – and collections by others – have consistently yielded an apparently indigenous parasitoid identified as *D. semiclausum*. However, parasitization rates were very much lower (<15%) than the ones reported from Asia. Owing to this, and as there is no record of an introduction to this region of Africa, doubt always persisted about the correctness of the identification. Now, molecular taxonomic methods are being employed to solve the problem. The African material has recently been declared identical with *Diadegma mollipla*, a potato tuber moth (*Phthorimaea operculella*) parasitoid⁷. Preliminary molecular work confirms this separation from *D. semiclausum*: material collected from two popu-

lations in Kenya and one each from Ethiopia and Tanzania can be distinguished from the *D. semiclausum* introduced to Taiwan by their mitochondrial DNA.

Molecular taxonomy will also be used to help keep track of the establishment and spread of *D. semiclausum* and its impact on the local species once an introduction has been made. For this purpose, a simple and cheap yet safe method will need to be developed as thousands of samples from all over the region will have to be screened. [Progress in this field with other pest species is described below.] Finally, as the taxonomy of the genus *Diadegma* is so notoriously difficult, the project will also attempt to make a contribution towards the molecular classification of species.

Bernhard Löhner and Barbara Wagener are requesting fresh material from anywhere in the world for inclusion in this study.

Contact: Bernhard Löhner
or Barbara Wagener,
International Centre of Insect Physiology
and Ecology, P.O. Box 30772,
Nairobi, Kenya
Email: blohr@icipe.org
or bwagener@icipe.org

Qualified Match-Making

It is important not to allow the results of DNA analysis alone dictate the path of a biocontrol project. AFLP (amplified fragment length polymorphism) is proving a particularly useful technique to distinguish between populations of the same species, and give a measure of the relatedness of them. It was used to analyse the relatedness of weed populations in a CABI Bioscience/Kerala Forest Research Institute (KFRI) project (funded by the UK Department for International Development, DFID) to develop a biocontrol strategy for *Mikania micrantha* (mile-a-minute) in India. *Mikania micrantha* is a Neotropical weed with an indigenous range extending from Mexico to Paraguay. In India, it is an invasive weed of tropical moist forest regions. It is a serious pest of tree crop/agroforestry systems in the Western Ghats and is spreading through Kerala towards Karnataka. It is also having a severe impact on tea production in Assam in northeastern India. AFLP analysis indicated the Indian weed to be of Central American origin. Surveys for pathogens revealed only mildly pathogenic species to be present in India, while 29 species were collected in Brazil, Mexico, Trinidad and Costa Rica, and four were considered to have potential as biocontrol agents. Of these, the rust *Puccinia spegazzinii* was identified as most promising, yet laboratory host testing of 11 strains indicated one from Trinidad to be the most

pathogenic to most populations of the weed from India. This illustrates the importance of using DNA analysis in combination with other techniques, in this case with host testing. The results indicated that the most pathogenic isolate is a new association, information that would not have been apparent if either technique had been used in isolation.

Contact: Sean Murphy, CABI Bioscience
UK Centre, Silwood Park, Buckhurst Road,
Ascot, SL5 7TA, UK
Email: s.murphy@cabi.org
Fax: +44 1491 829123

Needless to say, the *Bemisia* story has something to contribute here. The molecular analysis of *Bemisia* hosts described above was supplemented with morphological and RAPD-PCR analyses of parasitoids from these host populations. The new knowledge gained about the biogeographical relationships of the host meant that resources could be focused on the Spanish material as likely to provide the most co-evolved parasitoid strains⁵. In particular, attention was focused on a strain of *Eretmocerus mundus*. However, although this strain of *E. mundus* established successfully and seems to be spreading, the most successful introduced parasitoid in the Lower Rio Grande Valley, Texas appears to be *E. hayati* from Pakistan. This parasitoid attacks *B. tabaci* from the Asia group of populations, which are quite distinct from those found in the Middle East. In this latter case, climate matching appears to have been more important than co-evolution.

Contact: John Goolsby (details above)
or Kim Alan Hoelmer,
USDA, ARS,
European Biological Control Laboratory,
Campus International de Baillarguet,
CS 90013, Montferrier-sur-Lez, 34988 St.
Gely du Fesc Cedex, France
Email: khoelmer@ars-ebcl.org
Fax: +33 499 62 30 49

Too fine a focus for exploration for natural enemies may be undesirable for other reasons. The specificity requirements of the recipient country (which may be affected by the pest status of the target organism) may dictate otherwise. For example, if the target site is an island with no other indigenous members of the same genus, or the target pest is causing catastrophic damage, a less than host-specific natural enemy might be acceptable to its authorities, and a wider search area may be appropriate. The former scenario is illustrated by a current CABI Bioscience biocontrol programme against the invasive privet species *Ligustrum robustum* ssp. *walkeri* in La Réunion. Although molecular taxonomy confirmed

the area of origin of the subspecies as Sri Lanka, surveys have also been carried out in India, Vietnam and China on other *Ligustrum* species, since La Réunion has no desirable congeners and only two other native members of the same family. This approach should maximize the chances of finding suitable biocontrol agents in this instance.

Contact: Richard Milne,
Department of Plant Sciences,
University of St Andrews,
St. Andrews, Fife KY16 9TH, UK
Email: rim@st-andrews.ac.uk

Less Testing

The *Mikania* project illustrates a decision-making and potentially time-saving use of the AFLP results. Although the Trinidadian *Puccinia spegazzinii* isolate was highly pathogenic to most samples (indeed, more so than to the host from which it was isolated), some Assam populations proved resistant. These were also separated by AFLP analysis, and all proved susceptible to a Mexican isolate of another pathogen, *Dietelia portoricensis*. Traditionally, the only way of testing for promising host – natural enemy associations is to conduct independent matching of potential natural enemies for the entire invasive population. Given the practical and financial constraints of most biocontrol programmes, such comprehensive testing was and is seldom done, with the result was that aberrant populations were and are missed.

However, using AFLP analysis, it is now possible to identify 'representative' populations and 'hot spots' of genetic variation. The outcome of this is that natural enemy testing can be done on a more representative selection of the pest in its introduced range, and, depending on the genetic homogeneity of the introduced material, the extent of the testing necessary may be reduced. In the case of *Mikania*, material from Assam shows variation which testing protocols need to take into account, but that from the Western Ghats is more homogenous.

Rationalizing culturing and host testing was seen as a priority by the US whitefly programme. This programme was unique in the availability of so many species of natural enemies from intensive directed foreign exploration, combined with extensive rearing facilities. Exploration for natural enemies was carried out by staff of the USDA-ARS European Biological Control Laboratory in Montpellier (France) together with other collaborating scientists, at sites around the world selected on the basis of climate matching with target areas in the USA. USDA-ARS scientists at Mission, Texas developed a unique quarantine

protocol, integrating DNA (RAPD-PCR) analysis and morphologically-based systematics, to ensure "the maximum amount of species diversity with a minimum amount of duplication in cultures"⁶. This allowed them to assess a vast array of candidate parasitoid populations/biotypes on a large scale simultaneously.

Foreign collections were categorized in quarantine by plant type, collection site and macrotaxonomic characters of both hosts and parasitoids. Only parasitoids of 'the *Bemisia tabaci* complex' were accepted. *Eretmocerus* and *Encarsia* were separated on the basis of pupal and adult female characters. Individuals from each accession were then characterized using RAPD-PCR (primers CO4 and A10). Typically, material was characterized by both methods within three days, and unique accessions were set up in pure cultures on local *B. tabaci*, while duplicates of existing cultures were combined with them to increase their genetic diversity. USDA-ARS scientists used this process to allow them to evaluate multiple species on a large-scale. First, fecundity of candidate agents was assessed on hosts on selected crop plants. Promising parasitoids were reared and cage released onto the same crops to measure parasitism under field conditions. All species approved for release were tested by field release in an establishment evaluation. Then the 'sentinel plant' technique was used to test whether populations became widely established. In total, 38 exotic and two native parasitoids were evaluated⁸.

Establishing Results

The role of DNA techniques in biocontrol programmes is not limited to the pre-release phase. Probes are now being developed to make post-release monitoring of natural enemies more effective. However, morphological analyses continue to provide the framework within which the more expensive and time-consuming molecular methods can be targeted.

DNA analysis has been used in the *Bemisia* programme to help evaluate the success of the released agents and conclusively identify field recoveries of cryptic species/populations in Texas, desert valleys of Arizona and California, and the San Joaquin Valley, California. For example, species-specific probes developed from satellite DNA were used to identify *Eretmocerus* species from material collected post-release⁷. However, the cost of RAPD analysis is the limiting factor, and to overcome this and time limitations, research has been initiated to develop specific DNA probes from the satellite DNA for development into a 'squash blot' kit. Such refinements are not always

necessary. In an Australian study, molecular methods were used to help provide a structure within which the morphological systematist can operate. On the basis of this, they developed a user-friendly morphological key to *Encarsia* species that removes the need for any molecular analysis.

DNA techniques can also be used to obtain reliable information on the dynamics of parasitoid populations in the field. The Mission laboratory conducted RAPD-PCR analysis on parasitoid material recovered from uninoculated plots after agent release in Imperial Valley, California. The most promising agents in this desert climate appeared to be *Eretmocerus emiratus*, *E. hayati*, *E. mundus* and an *Eretmocerus* species from Ethiopia, while *Encarsia transvena* from Pakistan also looked promising. Morphological characters were used to determine that exotic *Eretmocerus* were in the majority (80%) at a time of normally peak native population levels. *Encarsia* populations also peaked, and DNA analysis of recoveries was used to show that all *E. transvena* recovered in the plots were of the newly released Pakistani strain, replacing the Spanish strain of the same species, which was not capable of such population increase during the summer months in this region.

Keeping Track

Biocontrol of some other key crop pests in the USA is receiving a helping hand from DNA fingerprinting. Cornell University scientists, in collaboration with ARS researchers at the Beneficial Insects Introduction Research Laboratory in Newark, Delaware have modified an AFLP method for identifying parasitoid larvae within their pest host⁹. This method can even detect and identify early stages of parasitoid larvae which are difficult to distinguish by conventional means. A similar procedure was developed and used for blackflies (*Simulium* spp.) infected with *Onchocerca* in the 1980s, and this was an important development for assessing the prevalence of river blindness.

At a stroke, this does away with some lengthy and painstaking monitoring procedures. Once adult parasitoids have been reared in the laboratory and identified to species (a process which can take months), DNA probes can be developed. Then there is no longer any need to go through the lengthy 'rearing out' of parasitoids before they can be identified. Estimating percentage parasitism no longer requires the skilled dissection of large numbers of field-collected hosts. However, it is important to note that the DNA evaluations also require special equipment, skill, and time; the resources of a given laboratory (DNA

machinery vs. growth chamber availability) would help determine the most practical method for that situation.

So far, the new method identifies two introduced European parasitoids: *Peristenus digoneutis*, which attacks the tarnished plant bug, *Lygus lineolaris*, a pest of many crops, and *P. conradi*, which attacks the alfalfa plant bug, *Adelphocoris lineolatus*, and it can distinguish them from the native *P. pallipes*. The Newark laboratory introduced both European species, and they have shown that the wasps are permanently established and are spreading in the north-eastern USA. The bugs are pests of crops grown for seed, vegetables, fruits, cotton and seedling trees throughout the USA, and annually cause tens of millions of dollars in losses and control costs. Research is continuing to determine whether other related parasite species can also be identified with the new technique. DNA probes do not invariably supply all the answers, but provide another tool in the taxonomic kit. In this project, determining the phylogenetic relationships of the various species of *Peristenus* is one of the continuing objectives. In cases where clear morphological differences between species exist, their relationships have been confirmed by DNA information. More work on the most closely related species is needed to advance further toward the ultimate goal of phylogenetic reconstruction, a process requiring appropriate quantities of fresh material of all species in the genus, and the time to perform the analyses.

Contact: William H. Day,
ARS Beneficial Insects Introduction
Research Laboratory,
Newark, DE19713-3814, USA
Email: wday@biir.ars.usda.gov
Fax: +1 302 737 6780
Kelley Tilmon,
Entomology Department,
Cornell University, Ithaca,
NY 14853 USA
Email: kjt6@cornell.edu
Fax: +1 607 255 0939

The technique is not limited to insects, for ARS scientists elsewhere have developed similar methods to monitor different strains of weed pathogens following their release into the environment as biocontrol agents. Methods are emerging for detecting and identifying several isolates of *Myrothecium verucaria*, a soil fungus that kills morning glories, which plague sugarcane and other crops. In field studies, spraying redroot- and smallflower-morning glories with an oil-based carrier containing *Myrothecium* spores proved as lethal to these weeds as the herbicide atrazine.

The DNA fingerprinting technique will help biocontrol scientists keep close tabs on the spore growth and spread, host range and effectiveness of different strains of biocontrol pathogens such as *Myrothecium* following release. In this way, it can give genetic evidence linking a specific microbial release to a specific disease seen in target weeds. It also reveals the spread of biocontrol microbes and demonstrates their effectiveness in reducing invasive weed populations.

Contact: Jan Suszkiw, USDA – ARS, Information Staff,
5601 Sunnyside Avenue, Mailstop 5129, Beltsville, MD20705, USA
Email: jsuszkiw@ars.usda.gov
Fax: +1 301 504 1641
Douglas G. Luster,
USDA – ARS Foreign Disease – Weed Science Research Unit,
1301 Ditto Ave, Ft. Detrick, MD 21702-5023, USA
Email: luster@ncifcrf.gov
Fax: +1 301 619 2880

Strategic Designs

Programmes in future may be able to use information from DNA analysis in formulating biocontrol and IPM management strategies. One project that intends to capitalize on this is aimed at developing an integrated strategy for management of the coconut mite, *Aceria guerreronis*. This pest, which is of undetermined origin, causes significant problems for coconut growers around the world; crop losses of more than 30% have been reported in the Caribbean. It has long been a problem in the Americas, from where it was first recorded, and more recently from Africa (while coconuts are probably indigenous to Melanesia). The mite is unlikely to have been transported on coconut as it is not found on the mature nut, which is both the natural mode of dispersal and the form transported by humans. It is therefore assumed that the mite's original host belongs to the indigenous flora of the Americas. However, outbreaks in Asia (Sri Lanka) have only been reported recently, and it is unclear whether these have followed a recent introduction, or indicate the breakdown of natural population regulation; outbreaks tend to occur in pockets separated from each other by long distances. This issue could be addressed by comparing DNA samples from populations in the Caribbean and Africa with multiple samples from Sri Lanka. If the origin of the mite in Sri Lanka can be ascertained, an IPM strategy can be designed specifically either to deal with a newly introduced pest, or to identify and correct whatever has caused the upsurge.

Contact: Dave Moore, CABI Bioscience UK Centre, Bakeham Lane, Egham, TW20 9TY
Email: d.moore@cabi.org
Fax: +44 1491 829100

Although there are too many strategic applications of DNA analysis to cover here, we include one that has particular significance for disease quarantine and management. In Canada, probes are being developed (US patent #5792611) to identify tree root rots and scleroderis canker in the absence of symptoms. One problem with such diseases is that they may remain latent and asymptomatic for long periods, during which time they are hard to detect but can still spread. Often, by the time symptoms are visible, it is too late to manage the disease. The DNA probes will have a two-fold application: first, to ensure the health of nursery seedlings before planting, which will help to prevent the spread of established diseases; second, to monitor imported stock for diseases, which will be a useful quarantine tool, enabling forest pathogens to be detected and diagnosed at the point of entry and thus prevent their introduction.

Contact: Richard Hamelin, Natural Resources Canada, Laurentian Forestry Center,
1055 rue du P.E.P.S.,
Ste-Foy, Quebec, Canada G1V 4C7
Email: hamelin@cfl.forestry.ca
Fax: +1 418 648 5849

Picking Winners

Finally, in a world where funding is hard to come by, being able to convince a potential donor that you have a success story waiting to happen could be what clinches the deal. Molecular techniques can sometimes identify in advance certain situations where classical biocontrol is likely to be successful. Where there is little variation within a population, host-specific natural enemies are less likely to be challenged by resistant varieties and therefore stand more chance of succeeding. For example, plant cytologists at Leicester University proved that all the invasive Japanese knotweed (*Fallopia japonica*) in the UK (and probably USA and Europe) appears to be same male-sterile clone, which would make it extremely susceptible to biological control – as well as providing the press with opportunities for headlines such as ‘Largest female on earth set to swamp Britain’!

Contact: Richard Shaw, CABI UK Centre, Silwood Park, Buckhurst Road, Ascot, SL5 7TA, UK
Email: r.shaw@cabi.org
Fax: +44 1491 829123

To return for a final time to the USDA *Bemisia* biocontrol programme, what makes it particularly striking is that DNA techniques were used at all stages of the programme, to inform decisions, to increase efficiency, and to contribute to the scientific basis. As a result of introductions of *Eretmocerus mundus* and *E. hayati*, sentinel plant sampling indicated a dramatic increase in the numbers of introduced *Eretmocerus* spp. Before releases of these, native *E. tejanus* formed more than 95% of recoveries, yet within three months, exotic populations had risen to 85% of the *Eretmocerus* spp. recovered. Evaluations of their impact on *B. tabaci* populations are now underway, and are expected to confirm that they are making a significant contribution to biocontrol⁶. The authors sum up “We hope these results will encourage other biological control programs to develop predictive methods and test their predictions in field settings. The information gathered... might further the theoretical aspects of our science and in turn increase the likelihood of success in future biological control programs”.

Sources

- ¹Reid, A.; Murphy, S. (1999) Biogeographic fine-tuning helps weed biocontrol. *BNI* **20(2)**, 51N-54N.
- ²Frohlich, D.R.; Torres-Jerez, I.; Bedford, I.D.; Markham, P.G.; Brown, J.K. (1999) A phylogeographical analysis of the *Bemisia tabaci* species complex based on mitochondrial markers. *Molecular Ecology* **8**, 1683-1691.
- ³De Barro, P.J.; Driver, F.; Trueman, J.W.H.; Curran, J. (2000) Phylogenetic relationship of world populations of *Bemisia tabaci* (Gennadius) using ribosomal ITS1. *Molecular Phylogenetics and Evolution* **16**, 29-36.
- ⁴Polaszek, A. (1999) Identification of whitefly parasitoids: some advice. *BNI* **20(4)**, 117N-119N.
- ⁵Kirk, A.A.; Lacey, L.A.; Brown, J.K.; Ciomperlik, M.A.; Goolsby, J.A.; Vacek, D.C.; Wendel, L.E.; Napompeth, B. (2000) Variation in the *Bemisia tabaci* s.l. species complex (Hemiptera: Aleyrodidae) and its natural enemies leading to successful biological control of *Bemisia* biotype B in the USA. *Bulletin of Entomological Research* **90**, 317-327.
- ⁶Goolsby, J.A.; Ciomperlik, M.A.; Kirk, A.A.; Jones, W.A.; Legaspi, B.C., Jr.; Legaspi, J.C.; Ruiz, R.A.; Vacek, D.C.; Wendel, L.E. (2000) Predictive and empirical evaluation for parasitoids of *Bemisia tabaci* (biotype ‘B’) based on morphological and molecular systematics. *In*: Austin,

A; Dowton, M. (eds) Hymenoptera: evolution, biodiversity and biological control. Canberra, Australia; CSIRO Publishing, pp. 347-358.

⁷Azidah, A.A.; Fitton, M.G.; Quicke, D.L.J. (2000) Identification of *Diadegma* species (Hymenoptera: Ichneumonidae, Campopleginae) attacking the diamond-back moth, *Plutella xylostella* (Lepidop-

tera: Plutellidae). *Bulletin of Entomological Research* **90**, 375-389.

⁸Goolsby, J.A.; Ciomperlik, M.A.; Legaspi, B.C., Jr.; Legaspi, J.C.; Wendel, L.E. (1998) Laboratory and field evaluation of exotic parasitoids of *Bemisia tabaci* (biotype 'B') in the Lower Rio Grande Valley of Texas. *Biological Control* **12**, 27-135.

⁹Tilmon, K.J.; Danforth, B.N.; Day, W.H.; Hoffmann, M.P. (2000) Determining parasitoid species composition in a host population: a molecular approach. *Annals of the Entomological Society of America* **93**, 640-647.

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IPM Systems

This section traditionally deals with IPM techniques compatible with biocontrol, particularly in agriculture. As a diversion this quarter, we consider the community health sector, and how action at community level is bringing about reductions in pesticide use for head lice control.

Bringing Head Lice Control up to Scratch

Head lice (*Pediculus capitis*) are not a danger to health; they neither serve as vectors nor do they generally cause direct harm or disease. Yet their management is a headache for public health workers worldwide. Here, we look at why they are perceived as such a problem, reasons for the limits to the success of current control measures, and how the limits are being addressed.

Head lice are wingless, parasitic insects adapted to living solely on the head hair of their human hosts. The claws on the ends of their legs are adapted for grasping hair shafts. They feed exclusively on blood, taking several blood meals a day, and die of dehydration and starvation within 2 days if removed from the head. Head lice do not jump, fly or swim but they can move swiftly along hair shafts. Transfer to a new host is almost exclusively by direct contact with infested hair. Nymphal stages tend to stay on the head where they hatch, while adults (particularly males) tend to migrate where possible to maximize out-crossing.

Why Scalps Crawl

Head lice are rather less contagious than the common cold and far less serious in terms of health. Yet although revulsion to them may be groundless it has been the basis for most management strategies. It is particularly in schools, where reports of waves of infestations are commonplace, that most control efforts are focused – often needlessly and unsuccessfully in the view of Richard Pollack (Harvard School of Public Health, USA). Control measures fail, he

argues, because they are designed to appease the public rather than deal with the problem, and instead they tend to burden the child. He points to exclusion from school of allegedly infested children and mass screening as pointless measures. He argues that quarantining has not been shown to be effective (by the time head lice are detected the child may have had them for a month or more), and that more than half of the children identified as infested may be misdiagnosed.

Exclusion of children from school because they have 'nits' is a widespread practice in the USA, although no distinction is made between live eggs and inviable/hatched eggs. Such exclusion policies are discouraged by the American Academy of Pediatrics and the (US) National Association of School Nurses. Pollack argues that most reported outbreaks of head lice in schools are no such thing: most 'infestations' he examines show no evidence of current (or past) infestation, normal debris on the hair or scalp frequently being mistaken for lice or eggs. In schools reporting more than 50% prevalence amongst its pupils, Pollack found actual prevalence rarely exceeded 1%, and sometimes he identified not a single case. A study of specimens associated with diagnosis of infestation submitted by health care workers and lay people found that less than 50% were head lice stages. Ian Burgess (Cambridge Medical Entomology Centre, UK) says that diagnosis of real infections has been consistently poor, and often based on paranoia and misunderstanding. He cites a survey of school nurse managers attending the human louse control management courses in the UK in 1982. It found that 50% of the health professionals interviewed (all of whom had been involved in screening for head lice in schools) had never seen a live louse, and relied on the presence of nits to confirm an infestation. A report by the 'Stafford Group' for the Consultants in Communicable Disease Control (CCDCs) in the UK

criticized both high-input screening and heavy-handed exclusion policies. They argued that the former has no measurable impact on infestation levels (although there are as yet no reliable data for these). They also pointed out that exclusion policies do little to reduce spread in schools, and only serve to increase the stigma of head lice infestations.

The UK health authorities used to cover the cost of routine screening of schoolchildren for head lice, but discontinued this practice in the early 1980s declaring it to be largely ineffective. Failure to establish an alternative management strategy, though, has led parents in the UK to blame a perceived increase in head lice incidence in schools on the withdrawal of school nurse inspections. Ian Burgess notes that in most districts most children had been screened less than once a year, if that. The little data collected after inspections stopped suggested that prevalence dropped, if anything, after screening ceased! Although Belgian health workers have demonstrated that inspection based on finding nits gives a high level of false positives, they and the UK nonprofit organization Community Hygiene Concern (CHC) both found that visual inspection also misses a large number of light, but contagious cases. In the absence of firm current data the Stafford Group suggest that prevalence has not increased, only public awareness – or perhaps over-awareness?

Conditions Right for Combing

In Richard Pollack's view, there is no conceivable need to apply insecticides for head lice within schools (and neither are such ancillary measures as cutting hair, bagging clothes and not using shared protective sports equipment justified; indeed, the latter carries far greater risk than a head louse infestation.) However, many are uncomfortable with the notion of parasites they can actually see or feel crawling about on their heads or their children's, so even if control cannot be justified on health

grounds, there is still a demand for it. In some cases, too, infestations are so chronic and severe that treatment is justified on health grounds. But what or when to treat is not obvious to the non-expert. Many health authorities and other extension services in the USA, the UK and elsewhere disseminate information by various means (leaflets, videos, websites, etc.), but quality is variable and sometimes only increases confusion. Clear guidance is needed.

The first step is finding out whether or not someone is infested. There are strong advocates for inspecting dry and wet hair, and the methods may be appropriate in different situations. Both, though, rely on combing with a fine-toothed comb to locate any lice or nits. Recent advances in this technique have been due to the work of CHC, who in 1988 set up a community development programme, Bug Busting, to help parents of schoolchildren in the UK acquire head louse detection skills. They had already established the value of combing wet hair with a plastic fine-toothed comb for this purpose. Wet detection combing is now the most widely-recommended method in the UK for confirming presence or absence of active head lice. The CHC protocol comprises methodical combing of washed, still-wet, well-conditioned hair. The moisture temporarily immobilizes lice, which can be comfortably combed out and then inspected. Emphasis is also placed on tracing infested contacts. CHC have conducted a series of case studies that have demonstrated the value of wet Bug Busting to detect low-level infestation, and thus recommend it for parents anxious to know if a child is infested. However, Ian Burgess argues that dry combing is equally accurate and is routinely practised by the Cambridge Medical Entomology Centre. The method has been used in several trials. He points out it is more practical (particularly where large numbers of heads are examined) because it is quicker and less messy.

For either method, generally no indication is given of how long should be spent on combing, and without adequate guidance, detection combing by inexpert hands can lead to both false negatives and false positives. Richard Pollack advises using the presence of live lice or viable eggs as the sole basis to confirm an infestation, but notes that recognition of eggs as viable requires suitable magnification and some training. Moving objects maybe also erroneously identified as head lice. CHC suggest that if parents are in doubt, 'combing' can be taken to a professional for identification, but say that most parents, given the right information and combs, can identify them without this recourse.

In the USA, a different detection policy has been developed for determining whether an infestation is active or a treatment is working. The America Head Lice Information Center (AHLIC) educate health professionals and parents that eggs are firmly attached at an angle to one side of the hair shaft, and cannot be removed by blowing or flicking. Once nits have been confirmed to be present, an active infestation is diagnosed by a two-step process: all eggs are removed, then the hair is inspected to see if new ones appear.

No Certain Solutions

The question of quite how to kill the head lice presents the next dilemma. The conventional approach is a synthetic insecticidal application or series of applications. A variety of products is registered in different countries, commonly pyrethroids and malathion but others include carbaryl or lindane. There are two problems with such treatments. The first is the growing antipathy of the public to synthetic pesticide use, with children a particular concern; the second is the perception that the insecticide treatments 'don't work'. Together these have led to disenchantment. There are reports of side-effects for all the synthetic insecticides currently registered, although proponents argue that the doses used in head lice products are so low as to be reasonably safe when used properly.

There is evidence from a number of countries for the development of resistance to the most commonly used pesticides, the pyrethroids and malathion, and Ian Burgess says that places they have surveyed in the UK in the past 6 years have double or even treble resistance. Resistance is likely to increase in incidence and intensity with time, but how often do treatments really fail? One common misconception at least partly to blame for perceived failure is the expectation that an insecticide treatment provides an instant fix. This is reinforced by labels on many products proclaiming that they kill both lice and eggs in a single application, which most do not, and certainly not reliably. Eggs are resistant to many of the registered products. A second application (recommended in the UK by the Stafford Group to be made on day 8) is intended to mop up the lice that hatch after day 1. Some of the products provide a limited residual effect, designed to kill hatching nymphs. Egg survival may be the first sign of resistance, but it is unclear how far widespread treatment failure is due to this. However, the residual action of current products, so hatching nymphs are exposed to lessening levels of insecticide, is likely to enhance resistance development.

Less contentious is the adverse impact of poor application of products, which is often based on poor understanding of the life cycle of the head louse. Instructions may not be followed precisely. Too little may be used (products are perceived as expensive), and/or the treatment may not be followed up where users believe that all the head lice have been killed by the first treatment. Another 'wave' of head lice appearing some weeks post-treatment is often blamed on a 'new' infestation (acquired from another person) rather than the more likely explanation of a resurgence from surviving eggs and lice, or misdiagnosis (with only dead or hatched eggs present). Joan Sawyer says that AHLIC call it 'the myth of reinfestation'. Misunderstandings about insecticide action are also reflected in attempts to apply prophylactic treatments.

When synthetic insecticides appear to fail, people may turn to other treatments, and increasingly to 'alternative' remedies in the misguided belief that because they are 'natural' they are inevitably safe. Yet amongst various herbal remedies widely touted are ingredients such as rosemary oil, which is known to induce abortion. Even products generally considered safe for some uses on humans, such as tea tree oil and lavender, may be applied at doses well in excess of those recognised as safe doses on children's heads. Research on essential oils suggests that they act as contact nerve poisons in the same way as synthetic insecticides. The efficacy of both synthetic and natural insecticides relies in part on their lipid soluble characteristics, as this facilitates their entry into the body (through either insect cuticle or human skin). Children are more sensitive than adults to the toxic effects of pesticides because they have incompletely-developed detoxification mechanisms and are growing rapidly. The active ingredients in most natural remedies are monoterpenes, which as a class are widespread in plants and are also found in some insects, and there is every reason to suppose that they will also be subject to resistance development in time. For example, there has been interest in coconut oil in the USA, and it may have efficacy as a treatment against head lice populations there. However, this effect may not be sustainable, and may not be mirrored in countries where coconut oil is commonly present in hair products already. The use of sublethal levels of monoterpene compounds (as are likely to be present, for example, in tea tree oil shampoo) will enhance resistance development.

The lack of certainty about the efficacy and toxic effects of synthetic and natural insecticide treatments is creating a vacuum, but the challenge is to come up with an effective alternative.

Look to Our Roots

Grooming and 'nit picking' are practised by our ape relatives, and were once part of human hygiene, a routine task undertaken by older siblings or parents (and still remembered by some of us, although not necessarily fondly). Perhaps it is a casualty of modern life, perhaps the advent of insecticide treatments led to its demise, but since 1995 its metamorphosis into Bug Busting as a remedy has occurred. CHC developed wet detection combing into a treatment, which depends on systematic removal of head lice to break the life cycle. The Bug Busting method follows a prescribed protocol, twice weekly for two weeks. It is based on three tenets:

- 'coated' lice move more slowly and so are more easily removed by combing
- removing lice before they mature prevents them spreading and reproducing
- co-ordinated community action helps decrease the rate of reinfestation

CHC produced a Bug Buster kit containing full instructions and the necessary comb, which was piloted in 1995 and commercialized in 1996. An improved model with a modified comb became available early in 1999. Currently, the London School of Hygiene and Tropical Medicine (LSHTM) is conducting a randomized controlled trial comparing the effectiveness of the Bug Buster kit with insecticidal medication.

A number of organizations in the USA, including the National Pediculosis Association (NPA) and the American Head Lice Information Center (AHLIC) promote combing together with other measures for head lice control. However, the emphasis US head lice policies placed on children being nit (rather than active lice) free means that mechanical methods to resolve infestation have taken a different path from those in the UK, with an emphasis on nit as well as active lice removal. AHLIC have produced a book and a video, 'Head Lice to Dead Lice', which uses humour to get across the message. The programme is based on a five-step battle plan, and includes the optional use of (a pyrethroid) insecticide. Instructions are given on the correct technique for smothering hair with olive oil (which is left on overnight under a rubber cap to smother the lice by blocking the spiracles), the correct days and method for combing to remove nits and lice, and advice on nit picking on dry hair.

A modified 'no nit' policy was included in a programme encouraging combing as a method of head lice removal that was developed by the extension service of the University of Nebraska at Lincoln. Barbara

Ogg says that a major problem with head lice in the local community was exacerbated by health professionals giving different recommendations on control to parents, who became confused. A head lice task force to formulate a policy was formed that included health professionals (school nurses, public health nurses, university educators), the state medical entomologist and child care programme administrators (from the health department). The policy was announced before the beginning of the school year in autumn 1999. Parents were urged to check their children's hair before school started and school nurses inspected children during the first week of term, and again mid-term and after the Christmas break (January 2000). School nurses agreed that they would not recommend products or treatment methods not proven to be effective against head lice; this was a major achievement because there were many advocates for mayonnaise and other home remedies. The University of Nebraska Cooperative Extension provided a videotape and fact sheets that were sent home with children.

The focus of the recommendations promotes combing with a long-toothed metal comb over all other activities as the 'alternative' method of control to remove nits. Where time is limited, people are encouraged to nit comb above all else. Barbara Ogg says that this really was a community collaboration and worked well over a trial period. Head lice have not been eliminated, but (and arguably more importantly) the school system is now seen to have a more proactive policy and to be taking the necessary steps to minimize the problem. An acceptable level of success was being maintained one year on: in the year 1999-2000, head lice cases had dropped to 2000 from a pre-programme level of 6700 (presumably based on the presence or absence of nits). This programme set itself realistic goals, and undertook active education at the community level to achieve these.

Testing Time

Advocates consider that bug busting offers an affordable head louse remedy, which is re-usable, does not require insecticides, clears lice that are resistant to insecticides, and promotes self reliance in head louse control. Detractors have some reservations about details of current protocols, but more significantly consider that the bug busting method is simply too time-consuming and demanding in commitment to be of value at a community level. They also point to social constraints: some children's hair is not readily combed, and bug busting can cause pain to them. Some parents are unable or unwilling to comply with the rec-

ommended protocol. Others may become over-keen on bug busting and family conflict can ensue. All these are issues which proponents of the method recognise and have sought to deal with. CHC have specifically developed methods for combing different hair types and instructions for these are included in their Bug Busting kit. Joan Sawyer of AHLIC argues that far from resenting the time that combing takes, parents and health professionals are increasingly aware of the even greater cost in time and money of relying on poor information and ineffective or dangerous pesticides.

The success of a bug busting programme may depend on how effectively the message is disseminated through the community and, in the long term, on the degree to which the momentum can be maintained once the initial wave of enthusiasm has subsided. Recognizing this, CHC resource designated Bug Busting days on 31 January and 31 October each year in the UK. They say that success relies to a large extent on the understanding by schools of their role in generating coordinated, informed effort followed by a top-up each Bug Busting day. Getting the message across would be helped by including head lice in the primary/first school science curriculum (in the UK, the national science curriculum at this level currently does not include insects with incomplete metamorphosis – a no-nits policy with a difference).

Although some good short-term results have been reported, success levels vary. How far, too, is initial success simply a reflection of the focus on controlling head lice rather than the method employed? Until recently, there have been few attempts to quantify the success of the method in comparison with other (insecticide) treatments. The mechanisms of long-term sustainability of the CHC programme are currently undergoing evaluation in addition to being included in the 3-year full-scale evaluation of head lice treatments by the London School of Hygiene and Tropical Medicine begun in 1999. Other studies comparing insecticide treatments and bug busting in the UK have recently published results. These varied in design; for example, in using professionals to apply treatments, or working through parents, and the results are far from clear cut. However, a trial in Bristol which looked at insecticide resistance found 87% and 64% failure for permethrin (pyrethroid) and malathion, respectively. A randomized controlled trial in North Wales, which compared the efficacy of two malathion treatments with CHC Bug Busting (using the 1996 comb) found 78% and 38% success, respectively. Arguments over the trials and their inter-

pretation have focused not only on trial designs and interpretation of results, but also on whether it is justifiable to recommend a head lice management strategy based on either method. However, CHC are encouraged by the North Wales results, which showed 38% success using the more laborious 1996 comb without initial demonstration, or the back-up they provide. They point out that skill at Bug Busting grows with familiarity, while lice resistance to insecticides increases with continued use. A knowledge of Bug Busting, they argue, is empowering to a community, while it does not benefit from dependence on failing insecticides.

Teasing Out the Snags

What, then, are the current constraints to bug busting and what are the keys to making it work? Areas to consider include: (a) communicating the message; (b) length of treatment; (c) hair lubricants and how to use them; and (d) combs and how to use them.

According to Ian Burgess, Bug Busting is a well-known and often-tried method, which can and should work. When it fails, it does so for a number of reasons, probably most importantly, he says, because many users do not understand why they are doing what they are doing. They follow the method, as they understand it, and are surprised and disappointed if it fails. Flawed information from unreliable sources and word-of-mouth are at least partly to blame, but in order to get the message across it may be over-simplified. On the other hand, CHC maintain that more than 80% of people they deal with are able to understand and follow their instructions, and achieve good results. Clear consistent information is crucial to this, and technical backstopping, as provided by their Bug Busting days, is important for reinforcing the message and contributes to the sustainability of the approach.

Most protocols (including CHC's) involve combing twice-weekly for 2 weeks; a total of four or at most five treatments. AHLIC is rare in advising a longer treatment period of 3 weeks, with six combings over this period. The more common 2-week period to remove live lice makes the method seem manageable, and CHC and AHLIC concur that this is an essential consideration. It is not much point developing a protocol that is too onerous for people even to attempt; but equally, advocate a protocol that frequently fails and you quickly lose credibility. Consideration of the head louse life cycle indicates that two weeks does not allow much of a margin for error: miss a few lice, and you will soon be back to square one. AHLIC say that in devising their strategy they were determined to create a protocol

that covered all possible errors, worked consistently for people unskilled at nit-picking, and did not overtax the family. The criterion for success that they used to evaluate the protocol during the research phase was that the entire family should be louse and nit free 10 days after completion of the programme. The CHC protocol includes a follow-up Bug Busting 4 days after the last treatment; if live lice are found, treatments are repeated. The UK Department of Health advises a wet combing check 3-5 days after using an insecticidal product. But even if a protocol seems foolproof, it won't be: there will be a few who will not follow it strictly for whatever reason, and a few will make mistakes. An important message to get across, especially in societies that expect sure-fire solutions, is that bug busting is not a guaranteed fix, but it is a skill which improves with practice. AHLIC's experience is that people are willing to admit mistakes and try again.

There are some robust disagreements about hair lubricants and their use. Olive oil is favoured by Ian Burgess and Joan Sawyer, as it is fairly easily emulsifiable (and therefore washed out afterwards), non-irritant and emollient (and so can be used on sufferers with eczema and psoriasis). Burgess notes that it is available in the UK as a pharmaceutical preparation. Sawyer describes it as "cheap, healthy and healing, with the only side-effect of gorgeous hair"! However, hair conditioner has the edge as a familiar and attractive product to use, which CHC say makes it more immediately acceptable. They suggest that even the most stout-hearted parent might be put off by the thought of trying to control and comb small slippery children dripping oil all over their soft furnishings. Although there are disagreements over which is the more effective for allowing lice/nits to be extracted, and also over safety (and lack of testing for this use), all parties report good success rates.

Protocols rarely suggest how long combing should take either for detection or for treatment, although the University of Nebraska-Lincoln programme stresses that nit combing should take precedence over all other anti-lice activities. Burgess suggests that detection combing may take 30 minutes, treatment combing several hours. Given normal family pressures, this time is likely to be hard to find, and it is understandable that bug busting programmes do not set out draconian regimes that will probably be ignored, if not put people off altogether. However, if combing is not performed adequately, the method is bound to fail. Successful combing is a technique which needs to be demonstrated, and this is addressed in information products pro-

duced by the programmes discussed in this article. CHC point out that the time for bug busting depends on hair type and length but agree that poor explanation of how to comb by some information sources contributes to failure, which is then used to discredit bug busting as a whole. CHC has produced a demonstration video, recommended by many UK health authorities, to introduce Bug Busting. In this and printed resources, they demonstrate clearly how to go about detection and treatment combing for different hair types and lengths.

Finally there is disagreement over the optimal comb design, and this is perpetrated in many leaflets, websites and other information products which do not clearly distinguish between detection and treatment combs, or lice and nit combs. CHC raised funding to research the issue. It built on the results of field tests made in the first half of the 20th century to select the precise slant on the leading edge of the new plastic Bug Buster comb that most effectively removed even newly hatched lice close to the scalp. Burgess agrees that a blunt-tipped plastic comb with appropriately and evenly spaced teeth works best for active lice removal. Both CHC and Burgess say that the use of inappropriate combs is a widespread reason for failure. In the USA, metal toothed nit combs are more popular, and these work particularly well for removing nits. Apparent discrepancies in advice may well be a reflection of the UK focus on breaking the lice cycle by removing hatched lice but not worrying about nits, and the US focus on removing nits as well as lice so children can return to school.

Putting Heads Together

In conclusion, IPM of head lice may be struggling a little at the moment, but there is an urgent need for it to succeed. Ian Burgess argues that failure in head lice management begins at the primary level: stakeholders from health professionals at all levels through public and community workers of all description to affected individuals and families would rather not know about head lice, or have to deal with them. In addition, conflicting and confusing advice on how to deal with head lice is widespread. Barbara Ogg says that the success of the University of Nebraska-Lincoln programme was rooted in forming a task force so all 'experts' spoke with one voice, a view firmly supported by John Simpson, chairman of the 'Stafford Group'. Clear, consistent high-quality information based, as CHC emphasize, on sound science and practical approaches, is crucial. Parents need to be aware of the likely reasons if control fails, so they can consider how to revise their practice to optimize the chances

of success the next time. If this option is not available, they are more likely to abandon bug busting for some other remedy.

The clear message coming through is that combing for head lice management can and does work under a variety of protocols, and this is very positive. Its sustainability at the community level is being assessed. However, the approach taken by agricultural IPM of integrating a variety of approaches to help overcome the shortcomings of individual components has limited application currently. Whether new non-pesticidal methods will become available remains to be seen, but other techniques would be useful. Ian Burgess observes that potential methods in the pipeline based on a non-neurotoxic physical approach could fill one niche.

Contacts/information:

American Head Lice Information Center
Email: sawyermac@aol.com
Website: <http://www.headliceinfo.com>

Ian Burgess, Medical Entomology Centre,
Cambridge Road, Fulbourn,
Cambridge CB1 5EL, UK
Email: ifburgess.medentcent@tesco.net
Fax: +44 1223 882690

Community Hygiene Concern (CHC),
160 Inderwick Road, London N8 9JT, UK
Email: bugbusters2K@yahoo.co.uk
Fax: +44 20 8292 7208
Website: <http://www.nits.net/bugbusting>

The National Pediculosis Association
Website: <http://www.headlice.org/>

Richard J. Pollack,
Laboratory of Public Health Entomology,

Dept. Immunology & Infectious Diseases,
Harvard School of Public Health,
665 Huntington Avenue, Boston,
MA 02115-6021, USA

Email: rpollack@hsph.harvard.edu
Website: <http://www.hsph.harvard.edu/headlice.html>

University of Nebraska-Lincoln
Website: <http://www.ianr.unl.edu/ianr/lanco/nviro/pest/lice.htm>

John Simpson,
Public Health Laboratory Service,
Communicable Disease Surveillance Centre
South East, Lower Ground Floor,
40 Eastbourne Terrace,
London W2 3QR, UK
Fax: +44 207 725 3460

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Announcements

Are you producing a newsletter, holding a meeting, running an organization or rearing a natural enemy that you want other biocontrol workers to know about? Send us the details and we will announce it in BNI.

First International Symposium on Biological Control of Arthropods

The first international symposium dedicated exclusively to biocontrol of arthropods, will be held on 17-21 September 2001 in Honolulu, Hawaii. It will bring together biological control practitioners from around the world to discuss international issues relating to the use of parasites and predators against pest mites and insects. Biological control of arthropods, while in increasing demand, faces a series of challenges that, unless addressed, will lead to a substantial reduction in such work. Better communication is needed among scientists working in this area. Until now there has been no meeting that regularly brings this group together in an atmosphere conducive to focused exchange of information. The Symposium on Biological Control of Arthropods has as its goal to provide for such meetings. Symposia will be held every 4 years, using the same style as the highly successful meetings held by weed biocontrol workers over the last 40 years.

The meeting will be limited to 250 people, so that the group is small enough to meet as a whole body. The organizers hope to attract largely if not exclusively people actually conducting programmes of arthropod biological control. The focus on applied pest

control projects is meant to differentiate this meeting from more basic research on natural enemy biology, such as is presented in the Entomophagous Insects Workshop. Focus will also be limited to the work on the use of parasitoids and predators, to the exclusion of pathogens. This is largely to permit the assembly of a smaller group.

A full day will be given to address pertinent aspects of each of the following major topics:

1. *Classical biological control*: Key issues in the future expanded use of classical biological control; Better methods for colonization, evaluation and monitoring of new natural enemies; Applications of molecular methods to the processes of classical biological control; Modelling and theory as tools to clarify causes of success or failure of biocontrol projects.

2. *Augmentative biological control*: Successes in augmentative biological control; Economics of production and use of reared natural enemies (including storage and shipping); Post-release dispersal, distribution, and impact of augmented natural enemies in field crops; Survey of actual and potential use in outdoor crops.

3. *Conservation of natural enemies in IPM systems*: Nectar feeding by parasitoids; Alternative hosts and habitat refuges for parasitoids; Effects on natural enemies of using *Bt* crops in IPM systems; Pesticide effects on natural enemies.

4. *International examples* of recent, important projects of classical biological control,

and monitoring for effects of biocontrol agents on nontarget organisms.

International focus is vital, and regional coordinators have been identified who will promote awareness of the meeting among biological control workers in specific geographic areas.

Regional contacts:

Europe: Ulli Kuhlmann
(U.Kuhlmann@cabi-bioscience.ch)
North America (including Mexico):
Mark Hoddle (Mark.Hoddle@ucr.edu)
South and Central America:
Elizabeth De Nardo
(edenardo@yahoo.com)
China: Dr Da-Wei Huang
(huangdw@panda.ioz.ac.cn)
Australia, New Zealand and Oceania:
Don Sands (Don.Sands@brs.ento.csiro.au)
Middle East: Moshe Coll
(coll@agri.huji.ac.il)
Africa: Peter Neuenschwander
(P.Neuenschwander@cgiar.org)

Volunteers are being sought as regional coordinators for Japan, Southeast Asia and Southern Asia (contact Roy Van Driesche, details below).

Further information:

Roy Van Driesche,
Dept. Entomology,
Univ. of Massachusetts,
Amherst, MA 01003, USA
Email: vandries@fnr.umass.edu
Website: <http://www.isbca.ucr.edu/>

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Biocontrol Meeting Focuses on Education

An international symposium 'The Practice of Biological Control: Importation and Management of Natural Enemies and Agents' will be held on 2-5 August 2001 at Montana State University in Bozeman, USA. The symposium is intended for practitioners in all disciplines of biological control, and is being sponsored by: the International Organization for Biological Control, Nearctic Regional Section (IOBC-NRS), the Experiment Station Committee on Organization and Policy (ESCOPE-BCWG), and the National Biological Control Institute (UDSD-APHIS-PPQ-CPHST).

The science and application of biological control are the focus of greater public appreciation and scrutiny than ever before. To develop and implement biological control programmes, practitioners must be able to present the activities and goals of their discipline to diverse audiences, and hence the focus of this symposium is on education: what we have learned from the past century of biological control efforts, and how we can apply this knowledge. It will emphasize: a renewed focus on why biological control should be of major consideration in pest control; development of effective education programmes about biological control that target diverse audiences; a new set of biological control case histories that illustrate important successes; and a discussion of the issues that promote and challenge the practice of biological control.

Keynote sessions will cover: the need for biological control; challenges to biological control; success in biological control; approaches to biological control of invasive species; conservation of natural enemies and antagonists; augmentation of natural enemies and antagonists; size, accountability and coordination of biological control programmes. Panel discussions will cover: challenges to biological control; systematics and biological control; approaches and methods used in applied biological control; and coordinating biological control activities. Posters may be on any biological control topic.

Contact: Tim Kring,
Univ. of Arkansas-Entomology,
Cralley-Warren Research Laboratories,
2601 N. Young Avenue, Fayetteville,
AR 72704, USA
Email: tkring@comp.uark.edu
Fax: +1 501 575 3348
Website: http://opal.msu.montana.edu/conf_services/biocontrol/index.htm

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Aphelinid and Trichogrammatid Meeting

The systematics and biology of Aphelinidae and Trichogrammatidae will be the subject of a symposium to be held at the University of California, Riverside, USA on 18-19 June 2001. The symposium will bring together more than 20 of the world's leading systematists, behaviorists and biological control specialists from China, India, England, Europe, Canada and the USA. The meeting will promote an exchange of ideas across disciplines, stimulate greater interaction among participants and, perhaps most importantly, provide a single venue for training students from various disciplines interested in these parasitic wasps.

Parasitizing scale insects, whiteflies, aphids, leafhoppers, Lepidoptera, and several other groups of insects, for biocontrol purposes these wasps rank among the top ten most important taxa. While trichogrammatids are important for augmentative control programmes, aphelinids are primarily used for classical biological control. The taxonomic diversity and relationships of both groups are poorly understood. Trichogrammatidae are represented by 75 genera and 675 valid species, and Aphelinidae have 38 genera and more than 975 valid species. However, little is known about species of either family in most habitats. Knowledge is especially poor in tropical regions, where, it has been suggested, microparasitic wasps attacking early or cryptic life stages may be the dominant fauna. Both families exhibit peculiarities in behaviour associated with host choice, competition and sex ratio distortion, which have made them model organisms for numerous studies.

Contact: Phyllis Crabtree,
Department of Entomology,
University of California, CA 92521, USA
Email: phyllis.crabtree@ucr.edu
Fax: +1 909 787 3086

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Fifth International Conference of Hymenopterists

The 5th International Conference of Hymenopterists will be held in Beijing, China from 22-26 July 2002. Nominations are sought for plenary speakers, and also ideas and organizers for symposia and specialist discussion groups.

Contact: Chao-dong ZHU,
Institute of Zoology,
Chinese Academy of Sciences,
Beijing, Haidian,
Zhongguancun Road 19#, P. R. China

Email: sea@pandaoz.ac.cn
Fax: +86 10 62565689
Website: <http://www.ioz.ac.cn/zcd/>

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African Entomology Congress

The 13th Entomological Congress, organized by the Entomological Society of Southern Africa in association with the University of Natal, will be held in Pietermaritzburg, KwaZulu-Natal, South Africa on 2-5 July 2001. This will provide a forum for exchange of information and ideas relevant to entomologists of all persuasions and with particular emphasis on the needs of Africa. Symposia will be held on a number of topics including: biodiversity and insect conservation; biotechnology, insects and plants; entomology and sustainable development; forensic entomology; Hymenoptera; and insect pathology. The following workshops are also planned: Final workshop of the Southern African Stem Borer Management Project; Insect rearing; Permits and legislation for collection of invertebrates; and Spatial data and the African entomologist.

Contact: Professor Denis J. Brothers,
School of Botany and Zoology, and Centre
for Environment & Development,
University of Natal, Pietermaritzburg,
Private Bag X01, Scottsville,
3209 South Africa
Email: brothers@nu.ac.za
Fax: +27 0 33 260 5105

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Fungal and Bacterial Plant Pathogens

The seventh workshop of the IOBC/WPRS Working Group 'Biological Control of Fungal and Bacterial Plant Pathogens' will be held at Kusadasi, Turkey, in May 2002, organized by the Ege University at Izmir. The meeting will focus on the 'Influence of abiotic and biotic factors on biocontrol agents'. Factors emphasized will include: microclimate, soil/substrate/crop/fertilization, chemicals, saprophytes/nontarget microorganisms, and mesofauna, their effects on and interaction with the population dynamics/survival of antagonists, on their biocontrol activity and on the economics of biocontrol.

Contact: Yigal Elad,
Dept. of Plant Pathology, ARO,
The Volcani Center, Bet Dagan,
50250 Israel
Email: elady@netvision.net.il
Fax +972 3 9683688 / 9683543 / 9604180 / 9604180

Website: <http://www.agri.gov.il/Depts/IOBCPP/IOBCPP.html>

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Weed Biocontrol Publications

The USDA Forest Service has produced two books on weed biological control that are now available to interested individuals.

Book 1: 'Weed biocontrol: extended abstracts from the 1997 Interagency Noxious Weed Symposium' (58 pp.). This covers the following aspects of weed biological control: History of weed biological control (articles by J.R. Coulson and L.A. Andres); Regulation (articles by R.E. Pizel, G.P. Markin and S. Stenquist); Safety (articles by Q. Paynter & J.L. Littlefield, J.K. Balciunas,

and P.B. McEvoy); Implementation (articles by A. McClay, R.W. Hansen, E.M. Coombs *et al.*, B. Villegas), and Monitoring (articles by M.J. Pitcairn and D.A. Pyke). Websites for weed biological control are also provided.

Book 2: This 95 pp. publication comprises the proceedings of the session: 'Host specificity testing of exotic arthropod biological control agents – the biological basis for improvement in safety', which was held during the X International Symposium on Biological Control of Weeds, Bozeman, Montana, USA, July 1999 (*eds*: R. G. Van Driesche, T. Heard, A. McClay & R. Reardon). It covers: (1) Concepts of insect-host plant selection behaviour and application to host specificity testing (T.A. Heard); (2) Physiological issues in host range expansion

(D.W. Tallamy); (3) The influence of time dependent processes on the outcome of bioassays (T.M. Withers *et al.*); (4) Evolution of host range in herbivorous insects (D.J. Futuyma); (5) Host specificity testing – why it is done and how it can be improved (R. Dekker van Klinken); (6) Evaluating host ranges – rationale, methodology and interpretation (D.P.A. Sands & R.G. Van Driesche); (6) Host specificity assessments – case studies (U. Kuhlmann *et al.*).

Contact: Roy Van Driesche, University of Massachusetts at Amherst, USA
Email: vandries@fnr.umass.edu
or Richard Reardon, USDA Forest Service
Email: rreardon@fs.fed.us

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Conference Reports

IOBC Water Hyacinth Meeting

The second Working Group Meeting for the Biological and Integrated Control of Water Hyacinth held every 2 years under the auspices of the International Organization for Biological and Integrated Control of Noxious Animals and Plants (IOBC) took place in Beijing, China on 9-12 October 2000. This meeting brought together 31 delegates from 11 countries with the common purpose of identifying suitable biological and integrated control solutions for water hyacinth.

The meeting took the form of a series of oral presentations, which included three keynote presentations covering the biological control of water hyacinth using insects, the biological control of water hyacinth using pathogens both as classical biological control agents and as mycoherbicides, and an overview of the current status of research on the weed in China. In addition, there was a series of reviews from different countries around the world, several papers dealt with the need for additional biological control agents, both insects and pathogens, and there were several papers dealing with aspects of the integrated control of the weed. These papers will be published by ACIAR, in a proceedings, which is expected to be available in the first half of 2001.

One of the roles of this working group is to identify further research needs on water

hyacinth. From the presentations and discussions the following ideas emerged for further investigation.

- Investigate the impact of cold climates on the success of biological control.
- Use plant competition studies between water hyacinth and other aquatic plants as an indication of how effective particular agents are.
- Investigate the compatibility of the different control options that could be used in integrated management.
- Select suitable locations and undertake integrated management of water hyacinth where biological control is the base technique.
- Identify and conduct surveys in areas where detailed surveys for additional natural enemies (both insects and pathogens) have not been conducted in the region of origin of water hyacinth.
- Research the interaction between insect natural enemies and pathogen natural enemies.
- Make a thorough investigation into the development of mycoherbicide for water hyacinth.

The workshop closed with a general meeting of the working group (the participants). During the meeting the following mission statement was developed for the working group:

“The mission of the IOBC Working Group for the Biological and Integrated Control of Water Hyacinth is to promote better management of water hyacinth through:

- facilitation of interactions,
- dissemination of information, and
- identification of research needs.

This will be achieved by:

- holding a meeting every 2-3 years,
- publishing the meeting proceedings, a water hyacinth newsletter and maintaining website,
- supporting activities that contribute to better management of water hyacinth.”

The next meeting will be held in Uganda on the shores of Lake Victoria in early August 2002.

By: Martin Hill, PPRI, South Africa

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Fifth Chromolaena Workshop

The Fifth International Workshop on Biological Control and Management of *Chromolaena odorata* was held at the Umhlanga Protea Hotel near Durban, South Africa, on 23-25 October 2000, with a post-workshop fieldtrip to northern KwaZulu-Natal province from 26-28 October. It was organized by the Plant Protection Research Institute of the Agricultural Research Council of South Africa (ARC-PPRI) in association

with the International Organization for Biological Control of Noxious Animals and Plants (IOBC), the provincial conservation service (KZN Wildlife) and Dr R.N. Muniappan of the University of Guam. This is the second time the workshop has been held in Africa (the first being the Third Workshop, held in Abidjan in 1993).

It was attended by 20 international delegates from 12 countries (India, the Philippines, Indonesia, Papua New Guinea, Guam (USA), Australia, Malaysia, Ghana, Congo-Brazzaville, Cameroon, Benin, Swaziland) and 30 South African delegates. These delegates included agriculturalists, water conservationists, biologists and nature conservationists. Major sponsors of delegates included the Australian Centre for International Agricultural Research (ACIAR) (for Southeast Asia) and the Technical Centre for Agricultural and Rural Co-operation (CTA) based in the Netherlands (for West and Central Africa).

The Workshop was opened by the Minister of Agriculture and Environmental Affairs for KwaZulu-Natal Province, Mr Narend Singh. The keynote address was delivered by the Chief Executive of the World Wide Fund for Nature (South Africa), Dr Ian Macdonald, and was entitled "*Chromolaena* at the cutting edge, or, Why the battle to control *Chromolaena* is a flagship battle for the new millennium". Dr Macdonald outlined his belief that, since chromolaena has all the characteristics of one of the world's worst invasive plants, if it can be beaten, any invasive can be beaten. As an invasive of tropical, largely developing countries, the successful struggle against chromolaena is a good indicator that we can get to grips with many of the underlying problems affecting conservation in the tropics, which is the repository for much of the world's biodiversity. Dr Macdonald's optimism for such success came from the fact that the chromolaena workshops are being held on a regular basis and that they are bringing together on-the-ground workers from the developing world; by the progress being made on biocontrol, a sustainable rather than symptomatic form of control; and by the global groundswell of awareness and action on invasive species.

Thirty-three oral and two poster presentations were made. Presentations were grouped into seven sessions with the following general topics:

- country and regional reports (13 papers),
- taxonomy, ecology and impacts of chromolaena (5 papers),
- impacts and management of chromolaena (4 papers),

- biological control of chromolaena (10 papers).

A mid-workshop tour on 24 October took delegates to the ARC-PPRI laboratories at Cedara to view the insects in quarantine presently being cultured and tested as biocontrol candidates on chromolaena and the South American invasive *Solanum mauritianum* (bugweed).

The workshop ended with an afternoon of:

1. Summarizing the workshop sessions.
2. Drawing up a list of biocontrol agents established in, available from and wanted by the various countries, and funding possibilities for regional biocontrol programmes.
3. The proposal and discussion of recommendations emanating from the workshop.

This information has been published in the *Chromolaena odorata* Newsletter No. 14.

The post-workshop fieldtrip afforded delegates the opportunity to see chromolaena in the field and its effect on renowned conservation areas such as the St Lucia estuary (a World Heritage Site) and the Hluhluwe-Umfolozi Reserve, at the historical centre of the effort to conserve both black and white rhinoceros. Control efforts on chromolaena and other invasive plants, undertaken by KZN Wildlife and the Department of Water Affairs's Working-for-Water Programme, were also outlined on the trip.

Two of the highlights of the workshop were:

1. The progress that has been made since the Fourth Workshop (India, 1996) on chromolaena biocontrol research and implementation. This is largely due to the proactive role taken by ACIAR in Southeast Asia, and the South African and Ghanaian programmes. The tephritid stem-galling fly, *Procecidochares connexa*, is currently being distributed widely in Southeast Asia, where it is establishing readily and causing significant damage. The defoliating arctiid moth, *Pareuchaetes pseudoinsulata*, which has had an inconsistent history in terms of its success as a biocontrol agent around the world, is spreading and causing significant damage in both Ghana and Sumatra (Indonesia). The nymphalid *Actinote* sp., whose larvae defoliate chromolaena, was rejected as an agent for South Africa because it fed on indigenous *Mikania* species, but has been released in Sumatra, as *Mikania* is invasive there. Its establishment has yet to be confirmed. A number of promising new

candidate agents are being tested in South Africa. These include the leaf-mining agromyzid fly, *Calycomyza eupatorivora*, and the stem-boring weevil, *Lixus aemulus*, both of which have been tested for host specificity to near-completion, with favourable results. The stem tip-galling weevil *Conotrachelus reticulatus* and the root-boring flea-beetle *Longitarsus horni* are also undergoing testing in South Africa. The ACIAR programme has also resulted in the establishment of centres of expertise in most Southeast Asian countries, for the testing and distribution of agents on chromolaena.

2. The progress made on unblocking the biocontrol programme on chromolaena in West and Central Africa. Due to perceived conflicts of interest over the role of chromolaena in agriculture here, outside of Ghana little progress has been made over the past decade on biocontrol of chromolaena. Discussion between parties representing these conflicting interests at the Fifth Workshop resulted in a commitment to host a meeting within the next year bringing together all stakeholders in West and Central Africa.

In addition, several delegates who had not visited South Africa previously and thus not seen the southern African form of chromolaena were struck by its different growth habit, morphology and ecology. That this form of chromolaena has a different biology (e.g. susceptibility to fire) to that invading other areas of the world was also evident from research presented at the workshop.

Papers for the Proceedings are currently being collated for refereeing. The Proceedings will be published during the course of 2001.

By: Costas Zachariades, PPRI, South Africa

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IOBC Fungal and Bacterial Plant Pathogens Workshops

A workshop of the IOBC/WPRS Working Group 'Biological Control of Fungal and Bacterial Plant Pathogens', held in Taormina, Sicily in September 2000, focused on the biocontrol of foliar pathogens. Organized and chaired by Yigal Elad (Bet Dagan, Israel) in cooperation with EFPP (the European Foundation of Plant Pathology), it was attended by some 200 people, and was devoted to the interaction of biocontrol agents with foliar plant patho-

gens, and to biological control and its mechanisms. Presentations included: an introduction to the subject and field use of a biocontrol preparation; induced resistance to control leaf pathogens by microbial inoculants that are applied to the root system; strategies and application of biological control for a disease of stone fruits; and how it is possible to cope with variability and inconsistency of biocontrol.

The sixth workshop of the Working Group, held on 30 November-3 December 2000, focused on 'Biocontrol agents, modes of action and their interaction with other means of control'. The meeting, organized by Enrique Monte (Salamanca, Spain) and Yigal Elad, with local and international

cooperation, was attended by 108 people from 33 countries. Presentations and discussions focused on the use of biocontrol agents against soilborne and foliar pathogens of all kind of plants either in the open field or in greenhouses and at post harvest stages, in the roots, stems, leaves, flowers, fruits and wood. It dealt with the use and implementation of biocontrol, various modes of action, the nature and use of genes that originate from biocontrol agents, involvement of mycorrhizae and improvements of biocontrol activity. Modes of action that were dealt with can be categorized as competition including space and nutrient exclusion, parasitism including phages that are hosted by bacteria, antibiotics, different modes of induced resistance,

restraining the pathogenicity enzymes of the pathogen and its antigens, and arresting the production of multiplication propagules of the pathogens. It was noted that in many systems multiple modes of action are involved. Several commercial biocontrol agents were presented. The improvement of efficacy, integration of biocontrol agents among themselves and with other means of disease control were discussed with respect to the improved implementation of biocontrol and the reduction of variability in the performance of biocontrol agents.

By: Yigal Elad, Bet Dagan, Israel

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