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General News

Diaphorencyrtus aligarhensis Released in California for Biocontrol of Asian Citrus Psyllid

Asian citrus psyllid (ACP), *Diaphorina citri*, the vector of a bacterium that causes the lethal citrus disease huanglongbing (HLB), was first discovered infesting citrus in urban areas in southern California in 2008. The first citrus tree infected with HLB was detected in Los Angeles County in March 2010. Large-scale pesticide application programmes by the California Department of Food and Agriculture (CDFA) targeting ACP in urban areas failed to eradicate or contain it. The pest is now found extensively throughout southern California and ACP detections are becoming more frequent in the San Joaquin Valley where 85% of California's citrus is grown, and it has been recorded as far north as San Jose near San Francisco. The California citrus industry is worth approximately US$3 billion per annum and 85% of production is eaten as fresh fruit. Yet informal surveys have suggested that more citrus is grown in backyard gardens than in all the combined commercial production areas of California!

The ACP–HLB combination has been devastating to the Florida citrus industry. ACP was first detected there in 1998, HLB was discovered in 2005, and some estimates suggest that more than 75% of Florida citrus is infected with HLB-causing bacteria. Economic analyses of the ACP–HLB situation in Florida suggest that by 2012 the citrus industry had incurred $1.7 billion in losses.[1] This situation in Florida could be portentous and is therefore very troubling to California citrus growers.

Because there are limited control options available for residential areas, and pesticide use in backyard gardens is relatively low, there was an opportunity to develop a classical biological control programme for ACP in California. Foreign exploration for natural enemies of ACP was conducted in Pakistan's Punjab province from September 2010 to April 2013, with colleagues at the University of Agriculture in Faisalabad collaborating with University of California (UC) Riverside scientists. Punjab was selected for natural enemy prospecting because it is thought to be part of the native range of ACP and its climate has a 70% match with major citrus production areas in California.

In December 2011, the eulophid parasitoid *Tamarixia radiata* was released in southern California for ACP biocontrol following extensive safety testing in quarantine at UC Riverside.[2] Subsequent mass production and release efforts by UC Riverside and later the CDFA led to more than 1,000,000 *Tamarixia* being released in urban areas. This parasitoid has a 75% female sex ratio, has established, and is spreading in California. Life table studies across replicated field sites are underway to document the impact this natural enemy is having on ACP populations.

During foreign exploration efforts in Pakistan, an encyrtid parasitoid *Diaphorencyrtus aligarhensis* was also discovered attacking ACP nymphs. A total of 1023 *Diaphorencyrtus* were brought to the quarantine facility at UC Riverside.[3]

Diaphorencyrtus *Safety Testing*

Host-range testing of *Diaphorencyrtus* on seven non-target species (native psyllid species and psyllids used as weed biocontrol agents) took almost 18 months to complete, but demonstrated that *Diaphorencyrtus* has a very strong preference for ACP nymphs. It was concluded it posed little environmental risk.[4] Results from no-choice and choice tests were used to prepare an Environment Assessment Report that was submitted to the US Department of Agriculture – Animal and Plant Health Inspection Service (USDA-APHIS) for review on 1 November 2013. After a lengthy review process, USDA-APHIS issued a Finding of No Significant Impact (FONSI) on 26 October 2014. The official release permit issued on 24 November 2014 meant *Diaphorencyrtus* could be moved out of quarantine for release and use in California as a biocontrol agent of ACP. The first official release of this parasitoid occurred on 16 December 2014 at the UC Riverside Biocontrol Grove with the liberation of 556 parasitoids.

Diaphorencyrtus *Biology*

While *Tamarixia* prefers to parasitize fourth and fifth instar ACP nymphs, *Diaphorencyrtus* parasitizes second–fourth instars, but second and third instars seem to be preferred.[5] Regardless of stage that is parasitized, ACP nymphs continue to feed, develop, and moult to the fifth and final instar before becoming mummies.[5] The sex ratio of the Pakistani *Diaphorencyrtus* is ~50% female. Adult *Diaphorencyrtus* emerge from an exit hole at the posterior end of the ACP mummy, while *Tamarixia* chews an exit hole in the anterior or head region to emerge. The placement of exit holes is an excellent field diagnostic tool for determining which natural enemy species was responsible for ACP mortality. Both *Diaphorencyrtus* and *Tamarixia* females obtain protein for maturing eggs by host feeding on ACP nymphs, and the trauma of being fed upon is sufficient to kill ACP nymphs.

Diaphorencyrtus *Release Plan*

Releases of *Diaphorencyrtus* are planned either for areas where *Tamarixia* has not been released or in selected areas near *Tamarixia* release sites where surveys indicate little or no activity associated with this parasitoid. The reason for this strategy is to minimize competition between *Diaphorencyrtus* and *Tamarixia* so as to give *Diaphorencyrtus* the best possible chance to establish. Past experience suggests that establishing more than one natural enemy of a citrus pest in California can increase the chances of successful biological control. Perhaps one of the best recognized cases is control of the cottony cushion scale, *Icerya purchasi*, with the predatory beetle *Rodolia cardinalis* and the parasitic fly *Cryptochaetum iceryae*. Surveys indicate that the beetle provides control in arid desert interior regions, while the fly dominates in cooler coastal areas where citrus is grown.[6]

An additional factor that needs consideration is the number and frequency of releases needed to establish *Diaphorencyrtus* in California. Invasion biology can offer some help with this – a general rule of thumb indicates that multiple small releases or several large releases can increase the likelihood of establishment greatly. For example, large numbers of natural enemies released (i.e. >30,000) have resulted in natural enemy establishment approximately 80% of the time; releases of fewer than 5000 individuals had colonization rates of 10%, and intermediate release numbers had a 40% establishment rate. Release frequencies of more than 20 attempts of more than 800 individuals were more likely to establish, and establishment likelihood was low when fewer than 10 releases were made of fewer than 800 individuals.[7] To increase the likelihood of establishing *Diaphorencyrtus*, these generalizations suggest that significant effort should be invested in making multiple releases of relatively large numbers of natural enemies.

What Can We Realistically Expect from Diaphorencyrtus*?*

Diaphorencyrtus populations sourced from Taiwan, Vietnam and China (these are all-female colonies) have failed to establish in Florida despite multiple release efforts involving more than 11,000 parasitoids.[8] Reasons for this are unknown, but could be due to heavy pesticide use against ACP, lack of synchrony between releases and ACP life stages suitable for parasitism, competition from *Tamarixia*, and possible predation of parasitized nymphs.[8] Other factors may include low genetic diversity (owing to the all-female lines) and too little investment put into release and establishment efforts (i.e. failure to reach the >30,000 benchmark suggested by Bierne[7]?).

In many countries *Tamarixia* and *Diaphorencyrtus* coexist (e.g. Vietnam, China and Taiwan) and in Pakistan the results of ~2.5 years of surveys in kinnow mandarin and sweet orange suggest that average year-round parasitism of ACP nymphs by *Diaphorencyrtus* is ~20% while *Tamarixia* accounts for ~30% parasitism each year.[9] In Saudi Arabia, *Diaphorencyrtus* may be the only parasitoid species attacking ACP nymphs infesting Mexican limes with maximum parasitism rates of 64–71% being recorded.[10]

It is impossible to predict what level of ACP suppression *Diaphorencyrtus* is likely to provide in California. It is anticipated that if *Diaphorencyrtus* establishes it will complement the activity of *Tamarixia* thereby increasing overall ACP mortality. It is possible that it may have ecoclimatic preferences different to that of *Tamarixia* which may allow it to provide control in areas where *Tamarixia* is not effective. The only way to determine these potential outcomes is through a multi-year research programme that tracks the establishment, spread and impact of *Diaphorencyrtus* on ACP in urban and commercial citrus production areas in California.

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[1]Hodges, A.W. and Spreen, T.H. (2012) Economic impacts of citrus greening (HLB) in Florida, 2006/07–2010/11. EDIS document FE903, Food and Resource Economics Department, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida, Gainesville, Florida. Web: http://edis.ifas.ufl.edu

[2]Hoddle, M.S. and Pandey. R. (2014) Host range testing of *Tamarixia radiata* (Hymenoptera: Eulophidae) sourced from the Punjab of Pakistan for classical biological control of *Diaphorina citri* (Hemiptera: Liviidae: Euphyllurinae: Diaphorinini) in California. *Journal of Economic Entomology* 107, 125–136.

[3]Hoddle, M.S., Hoddle, C.D., Triapitsyn, S.V., Khan, S.Z. and Arif, M.J. (2014) How many primary parasitoid species attack nymphs of *Diaphorina citri* (Hemiptera: Liviidae) in Punjab, Pakistan? *Florida Entomologist* 97, 1825–1828.

[4]Bistline-East, A., Pandey, R., Kececi, M. and Hoddle, M.S. (2015) Host range testing of *Diaphorencyrtus aligarhensis* (Hymenoptera: Encyrtidae) for use in classical biological control of *Diaphorina citri* (Hemiptera: Liviidae) in California. *Journal of Economic Entomology*. (In press)

[5]Rohrig, E., Shirk, P.D., Hall, D.G. and Stansly, P.A. (2011) Larval development of *Diaphorencyrtus aligarhensis* (Hymenoptera: Encyrtidae), an endoparasitoid of *Diaphorina citri* (Hemiptera: Psyllidae). *Annals of the Entomological Society of America* 104, 50–58.

[6]Quezada, J.R. and DeBach, P. (1973) Bioecological and population studies of the cottony cushionscale, *Icerya purchasi* Mask., and its natural enemies, *Rodolia cardinalis* Mul. and *Cryptochaetum iceryae* Will. in southern California. *Hilgardia* 41, 631–688.

[7]Beirne, B.P. (1985) Avoidable obstacles to colonization in classical biological control of insects. *Canadian Journal of Zoology* 63, 743–747.

[8]Rohrig, E., Hall, D.G., Qureshi, J.A. and Stansly, P.A. (2012) Field release in Florida of *Diaphorencyrtus aligarhensis* (Hymenoptera: Encyrtidae) an endoparasitoid of *Diaphorina citri* (Homoptera: Psyllidae) sourced from mainland China. *Florida Entomologist* 95, 479–481.

[9]Khan, S.Z., Arif, M.J., Hoddle, C.D. and Hoddle, M.S. (2014) Phenology of Asian citrus psyllid (Hemi-

ptera: Psyllidae) and associated parasitoids on two species of *Citrus*, kinnow mandarin and sweet orange, in Punjab Pakistan. *Environmental Entomologist* 43, 1143–1156.

[10]Al-Ghamdi, K.M.S. and Faragalla, A. (2000) Parasitism of *Diaphorina citri* Kuwayama (Homoptera: Psyllidae) by *Diaphorencyrtus aligarhensis* (Hymenoptera: Encyrtidae) on Mexican lime trees, western Saudi Arabia. *Bio-Science Research Bulletin* 16, 99–105.

Fungal Pathogens as the 'Fer-de-Lance' for Miconia Biocontrol in the Pacific

The story of *Miconia calvescens* (miconia) in the Pacific is the familiar tale of a 'botanical marvel' turned invasive weed. The most spectacular invasion has been in French Polynesia: within 50 years of its first introduction to Tahiti in 1937, miconia came to dominate 70% of the island's forests, including its native species-rich montane cloud forests. Its large, overlapping leaves (they can be >1 m long) form a closed canopy that greatly reduces light in the understorey, shading out other plants and allowing miconia, which does not have a high light requirement, to form dense, monotypic stands. It was estimated to threaten 70–100 of Tahiti's native species including 40–50 endemics. Miconia also became invasive following later introductions to Hawaii (1960s), New Caledonia (1970s) and Queensland in Australia, and it remains a threat to many Pacific islands. Concern with biodiversity loss has been the main driver behind efforts to tackle the invasive plant. A fungal biocontrol agent, *Colletotrichum gloeosporioides* f. sp. *miconiae*, released in Hawaii and French Polynesia, has had well-documented impact in Tahiti. Progress with research in Brazil on another damaging pathogen, *Coccodiella miconiae*, raises the prospect of a second fungal biocontrol agent becoming available.

Classical biological control initiatives against miconia in the Pacific were spearheaded by the Hawaii Department of Agriculture (HDOA), initially in the person of their exploratory entomologist Robert Burkhart who conducted surveys in Brazil, Costa Rica and Trinidad in the early 1990s. He found a rich diversity of insects feeding on the plant and some damaging pathogens, particularly in Costa Rica, and shipped them to Hawaii for further study. His work has been built on since, and promising insects from South and Latin America are being evaluated in Costa Rica and Hawaii (this is outside the scope of the current article). Work on pathogens was picked up and pioneered by Robert Barreto (Universidade Federal de Viçosa, Brazil), in cooperation with the University of Hawaii and HDOA. He isolated *Colletotrichum gloeosporioides* f. sp. *miconiae* from miconia and showed it to be the causal agent of anthracnose disease associated with premature defoliation of miconia in Brazil. The pathogen also gave promising results against plants and seedlings in containment. Rigorous host-specificity testing conducted at the HDOA quarantine facilities to assess its safety for introduction into the Pacific involved testing species in the same order (Myrtales) and included Tahitian native and endemic melastomes (no melastomes are native to Hawaii although it has other invasive melastomes). Results indicated that this strain of *C. gloeosporioides* is highly specific to *M. calvescens*. The pathogen was released in 1997, first in Hawaii following approval by the US Department of Agriculture, the US Department of the Interior and the HDOA Plant Quarantine Branch, and then in Tahiti using material shipped from HDOA and with approval of the French Polynesian government.

The pathogen established in both countries and impact was observed. In Tahiti, the biocontrol agent inflicted 4–34% defoliation of miconia trees and substantial (<30%) mortality of young seedlings (less than 50 cm tall) 4–6 years after release. Long-term post-release monitoring has shown that the increased light penetration into the canopy afforded by miconia defoliation favours recruitment of native species and has allowed partial restoration of rainforest, with increased native and endemic species richness and plant cover; specific studies of two endemic species (one tree, one subshrub) confirmed that biological control has contributed to their recovery.

As anticipated with such a formidable weed, the *Colletotrichum* biocontrol agent does not provide complete control of miconia. In particular, in Tahiti it is less effective in lower altitude (higher temperature/lower rainfall) forest. While scientists are investigating potential insect agents in addition to pathogens for Hawaii, all insect species prioritized so far feed on other melastomes and are not likely to be sufficiently specific for French Polynesia with its native melastome species. Barreto's team is working on a number of pathogens, including a nematode, but the breakthrough with *Coccodiella miconiae* may provide the best current prospect for French Polynesia. Despite often intensive hyperparasitism in its area of origin, *C. miconiae* inflicts severe, frequently debilitating 'yellow pustule' disease on *M. calvescens*, and is found year-round, which indicates tolerance of varied environmental conditions. It also attacks younger leaves than *Colletotrichum gloeosporioides* f. sp. *miconiae*, so could be a useful complementary agent. The breakthrough, after more than ten years' research, allows the team to produce viable, infective inoculum of *Coccodiella miconiae*. They have been able to confirm the pathogen's life cycle and its status as the causative agent of yellow pustule disease, and can begin studying its potential for biological control of miconia. They caution that the fungus seems to have very high specificity, with perhaps specific strains of *C. miconiae* attacking specific biotypes of *M. calvescens*, so an initial step will be checking that they have the best isolate for the miconia biotypes in the Pacific.

Scientists and managers in French Polynesia face another hurdle in gaining public approval for a second fungal pathogen, despite the visible success of the first – perhaps even because of it. Community acceptance was gained in Tahiti through a campaign that explained the good science and careful safety testing underpinning the application to release *Colletotrichum gloeosporioides* f. sp. *miconiae*. Now, though, some diseases and new pathogens, including *Phytophthora* and other *Colletotrichum* strains, appearing on cultivated fruit trees in Tahiti are being informally attributed to the introduced fungal agent; an agent that was introduced in a transparent manner and whose impact is clear to all, while the causal agents of the emerging diseases are likely to be accidentally introduced and as yet unidentified microorganisms. The lessons about the safety of biological control based on good science seems to have been forgotten because biosecurity and what it means in practice are not fully implemented and understood – this is a gap that needs to be addressed.

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Main Sources/Further Reading

Alves, J.L., Santos-Seixas, C.D., Rodrigues, F.A. and Barreto, R.W. (2014) Production of viable and infective inoculum of *Coccodiella miconiae* (Ascomycota: Phyllachoraceae) for the biological control of *Miconia calvescens*. *Biological Control* 79, 16–23.

Killgore, E.M., Sugiyama, L.S. and Barreto, R. (1998) Prospective biological control of *Miconia calvescens* in Hawai'i with a non-indigenous fungus *Colletotrichum gloeosporioides* (Penz.) Sacc. f. sp. *miconiae*. In: Meyer, J.Y. and Smith, C.W. (eds) *Proceedings of the First Regional Conference on Miconia Control*, Tahiti, French Polynesia, 26–29 August 1997. Gouvernement de Polynésie Française/University of Hawaii at Manoa/Centre ORSTOM de Tahiti, pp. 65–71.

Meyer, J.Y., Taputuarai, R. and Killgore, E. (2008) Dissemination and impacts of the fungal pathogen, *Colletotrichum gloeosporioides* f. sp. *miconiae*, on the invasive alien tree, *Miconia calvescens*, in Tahiti (South Pacific). In: Julien, M.H., Sforza, R., Bon, M.C., *et al.* (eds) (2008) *Proceedings of the XII International Symposium on Biological Control of Weeds*, La Grande Motte, France, 22–27 April 2007. CABI, Wallingford, UK, pp. 594–600.

Meyer, J.Y., Duplouy, A. and Taputuarai, R. (2008) Dynamique des populations de l'arbre endémique *Myrsine longifolia* (Myrsinacées) dans les forêts de Tahiti (Polynésie Française) envahies par *Miconia calvescens* (Mélastomatacées) après introduction d'un champignon pathogène de lutte biologique: premières investigations. *Revue Ecologie (Terre Vie)* 62, 17–33.

Meyer, J.Y. and Fourdrigniez, M. (2011) Conservation benefits of biological control: the recovery of a threatened plant subsequent to the introduction of a pathogen to contain an invasive tree species. *Biological Conservation* 144, 106–113.

Meyer, J.Y., Fourdrigniez, M. and Taputuarai, R. (2012) Restoring habitat for native and endemic plants through the introduction of a fungal pathogen to control the alien invasive tree *Miconia calvescens* in the island of Tahiti. *BioControl* 57, 191–198.

Meyer, J.Y. (2013) "Of miconia and men": the story of a scientifically and socially successful biological control program in Tahiti, French Polynesia. In: Wu, Y., Johnson, T., Sing, S., *et al.* (eds) (2013) *Proceedings of the XIII International Symposium on Biological Control of Weeds*, Waikoloa, Hawaii, 11–16 September 2011. USDA Forest Service, Forest Health Technology Enterprise Team, Morgantown, West Virginia.

In Memory of Peter Harris (1930–2014)

An icon in classical weed biological control was lost in August of 2014 with the passing of Dr Peter Harris at the age of 83. Highly respected at home and internationally as a pioneering leader in his science, Peter will be remembered for laying the foundations for and then greatly contributing to Canada's reputable weed biological control programme. For future generations, he also has left an enduring legacy with the successful mitigation of some of North America's most invasive rangeland weeds.

Born and raised in England, Peter came to Canada initially in 1950 to obtain his BSc (1955) at the University of British Columbia (UBC, Vancouver, Canada), and later chose the country as his new home. It was during his undergraduate degree at UBC that Peter's interest in entomology was ignited through the study of forest pest insects, but it also was when he met his wife, Irene (married 1957) and rose to some prominence as an award-winning track and field athlete. After completing his BSc, Peter returned to England to obtain a PhD in entomology at the University of London (1958) studying the European pine shoot moth, while still maintaining collaborations with forest entomologists in Canada. This sustained Canadian connection likely helped him get hired in 1959 into a biological control research position with Agriculture Canada, which was the beginning of a highly productive career that would span 36 years. While with Agriculture Canada, Peter first worked at the 'Research Institute' in Belleville, Ontario until 1972, then at the Research Station in Regina, Saskatchewan until 1992, and lastly, at the Agriculture and Agri-Food Canada (AAFC) – Lethbridge Research Centre (Alberta) until he officially retired in 1995. Being one to never sit physically or mentally idle, Peter continued to work as an Emeritus Scientist at Lethbridge until early 2014.

Peter was innovative, a 'big picture' thinker, tenacious and politically astute, which helped greatly in the formative years of the Canadian biological control programme. He persistently sought out, engaged and locally recruited those who could help him achieve his unwavering goal of providing agricultural producers and land managers with a reliable, economical and environmentally safe weed control option. Very early in his career, Peter also cultivated close collaborative ties with entomologist colleagues at CABI in Switzerland, which has been the contracted source of Canada's weed biological control agents for over 60 years. Working in close partnership with CABI legends, Dr Helmut Zwölfer and subsequently Dr Dieter Schroeder, Peter set Canada's priorities for the overseas surveys, selection and host-specificity testing of mostly insect agents for a number of damaging rangeland weeds. During his career, Peter released and field assessed 36 insect and one nematode species, of which 70% established in Canada (a high percentage for weed biological control globally) and about a third of these have had measureable impact on their target weed hosts. He had excellent instincts in predicting which agents would be successful based on a broad knowledge of entomology and botany and keen observational skills. He typically was engrossed in developing mechanistic hypotheses on why an agent either succeeded or failed, and could easily bridge disciplines to arrive at novel explanations for observed field patterns (e.g. his application of soil microbiology with a colleague to investigate the role of plant mycorrhizae in weed biological control). Some of Peter's acclaimed successes include the biological control of diffuse and spotted knapweeds in British Columbia using a suite of 11 established insects, and of leafy spurge in Canada's prairies using a complex of root-feeding flea beetles (*Aphthona* spp.). He also is recognized for using biological control to produce major reductions of the pasture weeds nodding thistle and tansy ragwort, such that herbicides were no longer needed in many areas, thereby accruing savings for affected livestock producers. Many of the successful agents introduced into Canada by Peter were subsequently approved for use by the USA against the same weeds.

Among Peter's many contributions in classical weed biological control were those that advanced the science globally through immediate and sustained uptake by researchers. Together with colleagues, he played a major role in the development of currently used host-specificity testing protocols. Peter was ahead of his time in advocating consideration of both crops and native plant species of concern when delineating the host range of candidate biocontrol agents during pre-release testing. He also encouraged a process of independent scientific review of petitions for agent release, thereby reducing conflicts of interest for the researchers that produce and submit the petitions to regulators for decision. Other contributions of note were his development of an easy-to-use, standardized scoring method for assessment and comparisons of agent impact in the field, and being the first to implement an economic analysis of the costs and benefits of weed biological control that could help in its promotion to potential project funders.

Peter also was a tireless advocate and educator when it came to encouraging general acceptance and adoption of weed biological control in Canada by governments, agricultural industry and the public. He readily shared and spread his biological control successes by directly engaging Canadian provinces and other stakeholders in 'hands-on' extension events (e.g. farmer field days for the collection and redistribution of leafy spurge beetles), thus engendering an understanding and sense of ownership of the projects and the biological control insects. As a result, many a field person became a loyal convert to biological control, especially after witnessing what it could accomplish. When project funding declined, Peter created the concept of 'weed biological control consortia', which brought Canadian and American stakeholders together to jointly fund the overseas exploration and testing of new agents for weeds of common interest.

Among his many accomplishments and awards for his career contributions, a few stand out as particularly noteworthy. These include: published papers in both *Science* and *Nature* in 1969 reporting on how mosquitoes can benefit from feeding on insect haemolymph; awarded the Commemorative Medal for the 125th Anniversary of Canadian Confederation (1994); given an award of recognition by the Canadian Forum for Biological Control (1996); awarded the Entomological Society of Canada's Gold Medal for "outstanding entomological contributions in Canada" (1997); inducted as a member of the highly prestigious Order of Canada (1997); and formally recognized for his career contributions by his biological control peers at the X International Symposium on the Biological Control of Weeds (1999).

Peter inspired a whole generation of weed biological control researchers and practitioners with his boundless energy and dedication to learning more about the art and science of weed biological control. For those of us who had the privilege of knowing him, he always will be remembered as an impressive, likeable man, who challenged and expanded our thinking and encouraged us in our careers. He will be greatly missed.

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Adapted by the author from her article in Bulletin of the Entomological Society of Canada *46(4), December 2014.*

Dr S.P. Singh (1941–2014)

Prominent biocontrol specialist Dr Surinder Pal Singh passed away on 1 December 2014 at the age of 73. The growth and popularity of modern-day biological control in India is inextricably linked to Dr Singh's career and era.

Born in the agriculturally inclined state of Punjab on 11 August 1941 into a family of three brothers and a sister, his interest in agriculture was but natural. After completing his early education in his hometown of Faridkot, he obtained a bachelor's degree in agriculture from the College of Agriculture, Ludhiana, in 1961. His entomological journey began when he enrolled for a master's in entomology in the same college. His penchant for research led him to leave the Punjab Agriculture Department to become a research assistant in the entomology department of his alma mater in 1963. He began his career in the Indian Council of Agricultural Research (ICAR) in

1967, with a posting at the Central Potato Research Station in Patna, and served the organization till his retirement.

It was during his doctoral programme in Russia that he mastered biological control. An Indian government scholarship from the Ministry of Education helped him obtain a PhD from the Kuban Agricultural Institute, Krasnodar, for his research in bioecology and biocontrol of the cotton bollworm. His fluency in the Russian language was also the result of his stay there from 1968–1973.

Upon his return from Russia, he was placed at the Central Potato Research Institute in Shimla as a Pool Officer. Then, in 1974, he was appointed as Scientist in agricultural entomology at the Central Horticultural Experiment Station in Chettalli, Karnataka, which he later headed for three years from 1980. Working on citrus, a major crop in the Kodagu region, he quantified the role of natural enemies in the suppression of several citrus pests. One of his major contributions was evolving production-and-release technology for the mealybug predator *Cryptolaemus montrouzieri* and its successful transfer to farmers under the Lab-to-Land programme during 1978–1983. He was also successful in mass rearing *Pareuchaetes pseudoinsulata*, an introduced biocontrol agent for the invasive weed *Chromolaena odorata*.

The turning point in his career came in 1984 when he took the reins of the All-India Coordinated Research Project (AICRP) on Biological Control of Crop Pests and Weeds. As the Project Coordinator of the AICRP operating out of the Indian Institute of Horticultural Research (IIHR) in Bangalore (now renamed Bengaluru), he was in full control of countrywide biological control research programmes. In 1988, he was elevated to Head, Biological Control Centre, which was located at the erstwhile Commonwealth Institute of Biological Control (CIBC) Indian Station in the same city.

Appreciating the research progress, ICAR upgraded the Centre to the Project Directorate of Biological Control (PDBC) and made Dr Singh the Project Director in November 1993. Within five years of its inception, PDBC bagged the coveted Best Institution Award of ICAR for 1998, which spoke volumes about his leadership.

As chief of the AICRP, he demarcated applied and basic research. While field-level applied research was assigned to the research centres spread all over the country, basic research was taken care of by PDBC. His favoured thrust areas were biology and ecology of natural enemies, rearing and mass production techniques, development of stress-tolerant strains, tritrophic interactions, and augmentative and classical biological control. In later years, he promoted research on microbials, and introduced new and emerging subjects including weed pathology, plant disease antagonists, fungal and bacterial pathogens of nematodes, entomopathogenic nematodes, and fungal pathogens of phytophagous mites. He was instrumental in taking up UK Department for International Development-funded collaborative projects with CABI on pathogens for classical biocontrol of *Parthenium hysterophorus* and *Mikania micrantha*.

Though predators and parasitoids were his strengths initially, in the last few years of his tenure at PDBC he used to stress that pathogens would gain in importance. He encouraged young pathologists, including this author, often saying "the future belongs to pathogens". He saw potential in pathogens (and plant disease antagonists) because of the ease of mass multiplication and their suitability for formulation like chemical pesticides. He felt deeply that only through commercialization could biocontrol products be taken to small and marginal farmers, who incur high costs through unscientific and indiscriminate pesticide application. His championing of commercialization of biocontrol agents resulted in the development and sale of technology for the first ever endosulfan-tolerant strain of *Trichogramma chilonis* in India, named 'Endogram'. Under his leadership, PDBC played a significant role in generating baseline data for the *cry1Ac* gene of *Bacillus thuringiensis*, used in transgenic cotton by Monsanto-MAHYCO in India.

Among farmers and scientists, Dr Singh's name was often synonymous with biocontrol-based technologies for the management of mealybugs, scales, psyllids and lepidopteran pests of maize, rice, sugarcane, tomato, cabbage, cotton and other crops.

His strong belief in a biocontrol solution for practically any invasive pest was evident when he arranged for the introduction of *Curinus coeruleus* from Mexico to suppress the accidentally introduced psyllid *Heteropsylla cubana*, thus saving the subabul (*Leucaena leucocephala*) crop. When the eriophyid mite *Aceria guerreronis* started threatening the coconut plantations in peninsular India, the first control measure he thought about was use of *Hirsutella thompsonii*.

When he was at the helm of PDBC, the World Bank-funded National Agricultural Technology Project recognized PDBC as a Team of Excellence for strengthening human resources in biological control.

The well-travelled Dr Singh published his scientific research extensively and regularly presented and published status papers and reviews. During his time, PDBC was known for its quality publications and books such as *Technology for Production of Natural Enemies* and *Fifteen Years of AICRP on Biological Control*, which are still popular today. He was the President of the Society for Biocontrol Advancement and was on the editorial boards of several scientific journals, including *Biocontrol News and Information*.

Dr Singh retired from ICAR–PDBC in February 2002 with a sense of fulfilment, though he time and again remarked that there was a lot to be completed. Post retirement, he was signed up by the United Nations Food and Agriculture Organization for consultancy on coconut integrated pest management (IPM) in the Asia–Pacific region. Based in Jakarta, Indonesia, he contributed immensely to sustainable pest management of coconut crops across the region.

He was active even after returning from Indonesia, operating from the beautiful city of Chandigarh in his native state. He provided consultancy in biocontrol and IPM and acted as an expert on panels and scientific review teams.

He is survived by his wife Zenya and son Tajinder Singh. The legacy of biocontrol knowledge and practice that he has left is incalculable, as is his impact on those he worked with:

Dr Abraham Verghese, Director, ICAR–NBAIR: "Dr Singh was a pioneer in the use of *Cryptolaemus montrouzieri* in the management of mealybugs on citrus and other crops. He was a man of few words but proved himself more with his actions than words. The great infrastructure that he developed is a proof of his actions."

Dr Chandish R. Ballal, Principal Scientist & Head, Division of Insect Ecology, ICAR–NBAIR: "Dr S.P. Singh, the founder Director of PDBC (now NBAIR), was an 'Institution' by himself. He wanted the Institute to be a home for the scientists, who could work with total dedication, but without any stress or pressure. He created in each scientist an acumen to work with confidence in specific areas of research, making us experts in identified fields and thus gave each one of us an identity. I remember Dr Singh with a lot of respect and reverence for what he has contributed to the science of biological control and also to encourage biocontrol researchers whole-heartedly."

Dr T.M. Manjunath, Former Director, Monsanto Research Centre & Former Vice President, Bio-Control Research Laboratories, Bengaluru: "In Dr S.P. Singh's demise […] we truly lost a committed scientist who always championed the cause of biological control and IPM. He possessed exemplary leadership qualities."

Dr S.S. Hussaini, Principal Scientist (Retd), PDBC: "[His] contributions for the cause of biological control in the country shall ever be remembered. […] He was responsible for the rise of this strategy as a valuable asset to sustainable agriculture and a reliable part of value based IPM. Dr Singh provided the much needed leadership at very crucial junctures and transformed country's image by his ingenuity and hard work. Many youngsters will emulate his example of commitment, sincerity, hard work. [He] was a mentor par excellence, always supportive yet disciplined, […] qualities he possessed which we and the younger generations need to emulate."

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Dr S.P. Singh: Stalwart Supporter of CABI and Biological Control

Dr Singh's contributions to CABI during his dedicated and selfless career were many and panoramic. He was a major part of our initial expansion across India and his contributions to our organization will not be forgotten. He was a remarkable soul and will be sadly missed by all of us who knew and worked with him – a fair and industrious worker and a man respected by colleagues, associates and competitors alike. He always played an active role in team building and social interaction with CABI.

CABI retained close links and collaboration with PDBC under Dr Singh, after ICAR took responsibility for the former CIBC Indian Station. CABI staff worked with Dr Singh and his team on a number of important projects. One in particular was the long and detailed process of demonstrating to the Government of India the importance of host-specific plant pathogens for the biological control of invasive plants; needless to say, Dr Singh was at the centre of steering the way through the process, bringing all the key stakeholders into the discussions to reach a successful outcome.

Dr Singh was also a long-standing, active and resourceful member of the editorial advisory board for this journal, ensuring that it kept the wider biocontrol world up to date about the activities of PDBC in particular, and what was happening in biological control in India and regionally. He remained a proactive board member after he retired when a lesser figure might have felt the need to retrench. He was always a person one could turn to for advice and his name clicking into the email inbox was a source of pleasure.

CABI salutes Dr Singh for the difference he made in the lives of many people, on both a professional and personal front.

By: Ravi Khetarpal and Sean T. Murphy, CABI.

Microbial Agents for Ambrosia Beetle Control in Florida

Entomopathogenic fungi may have a key role in controlling vectors of an exotic disease that attacks native Lauraceae in the southeastern USA and is threatening Florida's avocado industry. The results of studies conducted by scientists from the University of Florida's Tropical Research and Education Center in Homestead and Indian River Research and Education Center in Fort Pierce, and the US Department of Agriculture, Agricultural Research Service, Crop Bioprotection Research Unit in Peoria, indicate that a commercial strain of *Beauveria bassiana* shows particular promise.[1]

Although drought was initially suspected when redbay (*Persea borbonia*) trees began dying in Georgia and South Carolina in 2003, closer inspection of the trees revealed vascular wilt symptoms and beetle entrance holes.[2] A fungus was isolated from discoloured sapwood plus three scolytine beetles: two native species and a Southeast Asian species often associated with Lauraceae in its home range. *Xyleborus glabratus* or redbay ambrosia beetle (RAB), as it became known, had only been reported once before from the USA, from Port Wentworth in the Savannah area of Georgia in 2002.

In 2008, a previously undescribed fungus, *Raffaelea lauricola*, was proved to be the cause of the disease, which was named laurel wilt to indicate the range of susceptible species, so far restricted to plants in the family Lauraceae.[2] The fungus was also shown to be a symbiont of RAB. Like other ambrosia beetles, RAB harbours fungi in specialized mycangia, cuticular pockets near the mandibles. As the beetle constructs its galleries in host trees and lays eggs, fungal spores are exuded from the mycangia, colonize the walls of the beetle galleries, and digest plant tissues to provide food for the beetle larvae. In the case of *R. lauricola*, the fungus also moves systemically through the xylem vessels and causes vascular wilt. Little was known about *X. glabratus* in its native Asian habitat. However, the beetle was recently found in Japan harbouring *R. lauricola*[3] and laurel wilt was detected affecting avocados in Myanmar (R. Ploetz, pers. comm.). It is now confirmed that the fungal pathogen has an Asian origin.[3]

Laurel wilt spread rapidly in the southeastern USA, causing extensive mortality of redbay and swamp bay (*P. palustris*) and wilt disease in other native Lauraceae. It soon became clear that it was also lethal in avocado. The threat to Florida's avocado industry, the state's second largest fruit crop after citrus, became critical when the disease was recorded in northern Florida in 2005. In 2010, RAB was recovered during routine trapping in Miami-Dade County in southeastern Florida where 95% of the state's avocados are grown, and in 2012 the first case of laurel wilt in commercial avocado was detected. Current measures to reduce beetle populations and limit disease spread involve felling, chipping and burning infested trees, then spraying adjacent trees with insecticide. Some 6000 (1%) of Florida's commercial avocado trees have already been destroyed owing to the disease, and additional control measures are urgently needed.

Researchers tested three commercial strains of entomopathogenic fungi: *Isaria fumosorosea* Ifr 3581 and PFR blastospore powder formulations, and *B. bassiana* GHA emulsifiable conidia suspension.[1] Survival of RAB collected from redbay trees and either dipped in fungal solutions or allowed to walk on fungus-treated wood was 3–5 days, and shortest with *B. bassiana*. *Beauveria bassiana*-dipped beetles also had the most viable spores attached to their bodies, possibly because the oil-in-water formulation with emulsifiers and hydrophobic aerial conidia meant the spores were better able to adhere to the hydrophobic insect cuticle than hydrophilic *I. fumosorosea* blastospores in aqueous formulations. Treatment did not affect beetle boring activity, but they died after initiating tunnelling activity. As RAB does not start egg-laying until about 7 days after initiating boring, these studies, which used a low product dose compared with typical field application rates, indicate that biopesticides could prevent beetles from laying eggs. The authors stressed that field trials are needed to verify these lab results and suggest focusing on *B. bassiana*.

Although RAB is the key vector of laurel wilt in the southeastern USA and is easily detected in infected native Lauraceae, it is seldom found in avocado orchards in Florida. Other 'resident' (exotic and native) species of ambrosia beetles have become vectors of the pathogen. Trapping in orchards revealed a complex of species in several genera[4], and some of these have been shown to transmit the disease to avocado[5]. Scientists have also been conducting experiments with some of these species.[6] After conducting similar tests using *I. fumosorosea* PFR, *B. bassiana* GHA and *Metarhizium brunneum* strain F52 against *Xylosandrus crassiusculus* and *Xyleborus volvulus*, these authors concluded that all three products were pathogenic to the beetles, with *B. bassiana* again the most effective overall. They found from field experiments that *B. bassiana* and *M. anisopliae* persisted up to 21 days after application to avocado wood and advocated field testing the *B. bassiana* product; field trials are now underway.

The Asian ambrosia beetle and the fungal pathogen were almost certainly introduced together. The most likely – almost certain – pathway is untreated or inadequately treated wood packaging material used in international trade. Although guidance is given by the International Plant Protection Convention in ISPM (International Standards in Phytosanitary Measures) 15, *Regulation of Wood Packaging Material in International Trade*, wood packaging is an all too common pathway by which invasive species are introduced. The emerald ash borer (*Agrilus planipennis*) and Asian longhorn beetle (*Anoplophora glabripennis*) are two others that have entered North America by this route in recent decades. Until and unless this pathway can be regulated more effectively, more alien pests can be expected to arrive.

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[1]Carrillo, D., Dunlap, C.A., Avery, P.B., Navarrete, J., Duncan, R.E., Jackson, M.A., Behle, R.W., Cave, R.D., Crane, J., Rooney, A.P. and Peña, J.E. (2015) Entomopathogenic fungi as biological control agents for the vector of the laurel wilt disease, the redbay ambrosia beetle, *Xyleborus glabratus* (Coleoptera: Curculionidae). *Biological Control* 81, 44–50.

[2]Fraedrich, S.W., Harrington, T.C., Rabaglia, R.J., Ulyshen, M.D., Mayfield, A.E., III, Hanula, J.L., Eickwort, J.M. and Miller, D.R. (2008) A fungal symbiont of the redbay ambrosia beetle causes a lethal wilt in redbay and other Lauraceae in the southeastern United States. *Plant Disease* 92, 215–224.

[3]Harrington, T.C., Yun, H.Y., Lu, S.S., Goto, H., Aghayeva, D.N. and Fraedrich, S.W. (2011) Isolations from the redbay ambrosia beetle, *Xyleborus glabratus*, confirm that the laurel wilt pathogen, *Raffaelea lauricola*, originated in Asia. *Mycologia* 103, 1028–1036.

[4]Peña, J., Narvaez, T., Carrillo, D. and Duncan, R.E. (2014) Ambrosia beetles associated with laurel wilt-affected avocado orchards in south Florida. Annual Meeting of the Entomological Society of America, 16–19 November 2014, Portland, Oregon.

[5]Carrillo, D., Duncan, R.E., Ploetz, J., Campbell, A., Ploetz, R. and Peña, J.E. (2014) Lateral transfer of a phytopathogenic symbiont among native and exotic ambrosia beetles. *Plant Pathology* 63, 54–62.

[6]Avery, P.B., Bojorque F., V., Carrillo, D. and Cave, R.D. (2014) Biotic parameter assessments of candidate entomopathogenic fungi evaluated for ambrosia beetle pest management in avocado production. Annual Meeting of the Entomological Society of America, 16–19 November 2014, Portland, Oregon.

Canadian Biofungicide for Fusarium Head Blight of Wheat

Fusarium head blight (FHB) is a fungal disease of small-grain cereals that has become of increasing international importance in recent decades. It reduces grain yield and quality, and infected kernels are contaminated with the mycotoxin deoxynivalenol (DON), which is harmful to livestock and a safety concern in the human food chain, thus limiting uses of the grain and reducing its monetary value. FHB is now the most serious wheat disease in North America. Although it was first recorded in the USA in the late nineteenth century and in Canada in the 1920s, the first major outbreak in eastern Canada occurred only in 1980 and in Manitoba in 1993. Since then it has spread westwards through the Prairie Provinces with frequent epidemics occurring in both eastern and western Canada in years of wet weather coinciding with the flowering stage of wheat. Worldwide FHB is caused by a number of species, but the major species in North America is *Gibberella zeae* (anamorph: *Fusarium graminearum*).

Resistant cultivars are an important bastion against plant diseases, but complex disease genetics have prevented progress towards cultivars with complete resistance to FHB. Management practices include crop rotation and ploughing in residues (because they harbour the disease – see below), but these measures alone have not prevented epidemics. There is therefore reliance on fungicides, but they do not provide consistent control. And as well as imposing additional costs and pre-harvest intervals on the farmer, there is public concern about the environmental impact of pesticides, and residues in the human food chain. A number of antagonistic microorganisms have been evaluated for control of FHB across the USA and two products, TrigoCor 1448 (*Bacillus amyloliquefaciens*) and OH 182.9 (the yeast *Cryptococcus flavescens*) are being pursued as commercial biocontrol agents for use in integrated pest management. Now Canadian scientists have a contender: CLO-1, based on the fungus *Clonostachys rosea* strain ACM941, which has shown particular promise as a residue treatment.

The product CLO-1 is an experimental wettable powder formulation of *C. rosea* strain ACM941; this strain was isolated from a pea plant in 1994 by Allan Xue and patented by Agriculture and Agri-Food Canada (AAFC) in 1999. It was identified during assays of a range of potential biocontrol agents as having strong antagonistic activity against soil- and seed-borne pathogens of cereal crops, including FHB. It was also found to be a good plant colonizer, a property that proved key to its development as a biocontrol agent. It showed activity as a foliar spray, but its activity in suppressing *G. zeae* in crop residues really caught scientists' attention because of how this might be exploited against FHB. Although *G. zeae* produces asexual conidia as well as sexual ascospores, and they are equally capable of causing disease, FHB is a monocyclic disease (with just one cycle each growing season). *Gibberella zeae* perithecia fruiting bodies that form in crop residues produce ascospores, and these provide the initial source of disease inoculum, which is also thought to be primarily responsible for generating epidemics when weather conditions are favourable. A treatment that suppresses ascospore production should contribute to successful disease management by reducing infection pressure and therefore the danger of outbreaks.

A series of greenhouse experiments and field trials between 2005 and 2010 indicated that, although it was less effective than the fungicide terbuconazole at reducing disease when applied as a foliar spray, *C. rosea* ACM941 – lately known as CLO-1 – was at least as effective as terbuconazole at reducing the pathogen perithecia production when applied to crop residues. Experiments showed that the impact of CLO-1 was achieved whether it was applied before or after the substrate was infected with FHB, and in field trials applied in either autumn or spring. CLO-1 applied to wheat residues in the field in spring delayed the appearance of perithecia by 7–10 days and reduced quantities of ascospores, with production dampened so that the characteristic production peak was not seen – results comparable to those achieved with terbuconazole. CLO-1 showed good activity in maize and soybean residues as well as wheat, indicating that it could be applied effectively to residues of preceding crops in a rotation to provide pre-planting protection of a wheat crop. In wheat, the biocontrol agent was most effective in combination with a moderately resistant cultivar – the best currently available. There was considerable variation between years, however, and this was tentatively ascribed to differences in weather or perhaps prevalent FHB strains between years, something the scientists are keen to investigate further. Laboratory and field trials in 2009–2010 demonstrated that concentrations of above 10^6 cfu/ml of CLO-1 provided consistent and significant effects, with FHB suppression generally equivalent to that achieved with terbuconazole.

In August 2014, AAFC signed a ten-year licensing agreement with Adjuvants Plus Inc. (Kingston, Ontario) to develop the technology and gain regulatory approval, with a provisional two-year timescale for bringing it to market. CLO-1 is seen as providing an additional measure for an integrated FHB management strategy that will reduce initial inoculum load and the risks of epidemics, and it should have an additive effect in tandem with the most resistant wheat cultivars currently available, and thus reduce the need for fungicide applications against FHB.

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Main sources: Xue, A.G., Voldeng, H.D., Savard, M.E., Fedak, G., Tian, X. and Hsiang, T. (2009) Biological control of fusarium head blight of wheat with *Clonostachys rosea* strain ACM941. *Canadian Journal of Plant Pathology* 31, 169–179.

Xue, A.G., Chen, Y.H., Sant'anna, S.M.R., Voldeng, H.D., Fedak, G., Savard, M.E., Längle, T., Zhang, J.X. and Harman, G.E. (2014) Efficacy of CLO-1 biofungicide in suppressing perithecial production by *Gibberella zeae* on crop residues. *Canadian Journal of Plant Pathology* 36, 161–169.

Xue, A.G., Chen Y.H., Voldeng, H.D., Fedak, G., Savard, M.E., Längle, T., Zhang J.X. and Harman, G.E. (2014) Concentration and cultivar effects on efficacy of CLO-1 biofungicide in controlling fusarium head blight of wheat. *Biological Control* 73, 2–7.

Role for Biological Control in Reducing Greenhouse Gases: Rabbits in Australia

Australia is committed to making at least a 5% reduction in greenhouse gas emissions by 2020. Biosequestration, or the capture and storage of atmospheric carbon, is one of the key pathways to achieve this. A report published in 2014[1] argues that control of feral invasive mammals (rabbits, goats, pigs camels) that facilitates restoration of native vegetation could prove a more economic and sustainable method of increasing carbon sequestration than tree planting, and it would also generate other benefits for agriculture, communities and the environment. The report suggests that it might take only a small increase in tree density in Australia's existing, rabbit-damaged woodlands to allow emission reduction targets to be met and explores how feasible this is to achieve. The report's authors use economic and ecological studies on the impact of major rabbit control events, including biological control by the myxoma virus and rabbit haemorrhagic disease virus (RHDV), to illustrate their case.

Rabbits are widespread over some 70% of Australia. Their devastating impact on vegetation is well-documented, but their impact on carbon storage and the magnitude of benefits that could be derived from their control have not yet been quantified. Intuitively they can be anticipated to be enormous: rabbit grazing destroys vegetation, especially forbs and perennials, on a massive scale, both reducing carbon input into the soil and increasing carbon loss through erosion. Selective grazing by rabbits has been shown to have ecosystem-changing impacts, for example in the mulga woodlands that cover 143 million hectares of inland Australia. Mulga (*Acacia aneura*) and other arid-zone acacias of this ecosystem are long-lived, slow-growing species with very dense wood whose mature trees can store large amounts of carbon, and much of this remains locked up long after trees die. Yet as few as one rabbit per hectare has been shown to destroy all acacia seedlings and halt recruitment. Conversely, studies have revealed dramatic increases in native woody and perennial species cover after rabbit control in rangelands and conservation areas as well in the semi-arid interior. In mulga woodland, acacias and other native species flourished for the first time in 100 years after rabbits were reduced 95% during the 1990s by RHDV. But across Australia, some populations of rabbit are developing resistance to the introduced strain of the virus and are increasing.

The report's authors say there is a critical need to understand the relationship between rabbits and carbon storage in order to understand how their control could help Australia meet greenhouse gas reduction targets. They propose a dual strategy of predictive modelling and field studies measuring carbon sequestration with manipulated rabbit populations.

They also tackle the realities of achieving rabbit control, noting that very substantial and persistent reductions in rabbit populations will be needed for carbon benefits to be realized. While conventional measures such as warren ripping are economically feasible in some rangeland areas, only biological control is realistic in the vast semi-arid inland areas. Whether biological control can achieve sufficient control is not clear, although a major cooperative research programme (RHD-BOOST) has been funded to research new and effective virus strains for Australia. With few previous studies on the impact of invasive mammals on carbon storage, this report is intended to "stimulate informed consideration of the potential for invasive herbivore control to contribute to Australia's carbon pollution reduction targets."

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[1]Bengsen, A and Cox, T (2014) *The Role of Rabbit and Other Invasive Herbivore Control in Reducing Australia's Greenhouse Gas Emissions*. PestSmart Toolkit publication, Invasive Animals Cooperative Research Centre, Canberra, Australia.
Web: www.feral.org.au/invasive-herbivore-control-and-greenhouse-gas-emissions/

Neonicotinoid Moves Sluggishly but Surely through a Food Chain

When neonicotinoid insecticides were first introduced, some scientists believed that as seed treatments they would be safe for non-target species because the active ingredient would be absorbed systemically into the growing plant, where only herbivorous (= pest) insects would ingest it. Neonicotinoid seed treatments are now widely used to protect field crops from soil-dwelling pests but mounting evidence suggests they may, after all, have non-target impacts on beneficial species, notably bees, and some studies have queried their impact on soil communities. Now a study from the USA has shown that a non-susceptible pest was able to transfer a neonicotinoid to predatory soil arthropods, diminishing biological control to the extent that yields were reduced.[1] The authors say this pathway needs to be considered in risk assessment and stewardship.

The study looked at the impact of thiamethoxam seed coating of soybean on pest slugs and their predatory beetles. Molluscs are serious pests in many crops but tend to be overlooked. Laboratory studies indicated that slugs (*Deroceras reticulatum*) readily attacked soybean seedlings whether or not thiamethoxam had been applied to the seeds, and their growth and survival were unaffected by seed treatment. The carabid beetle *Chlaenius tricolor* also readily attacked the slugs, but slugs fed on soybeans grown from treated seed induced neurotoxic symptoms in over 60% of beetles.

Slug damage was intense where the researchers conducted field experiments in 2012, with *D. reticulatum* the dominant species at experimental sites. Pitfall trap catches and slug counts indicated that seed treatment reduced the activity–density of potential slug predators (85% carabid and staphylinid beetles) by 31% during the early growing season, and increased the activity–density of slugs. Slugs continued to be more prevalent in seed-treated soybeans throughout the season even though predator activity–density recovered.

Residue analysis indicated that levels of thiamethoxam and its metabolites declined exponentially through the food chain but were still found in slugs and beetles (and earthworms). Dietary toxicity of neonicotinoids in beetles is not well characterized, but the high potency of these compounds suggests that levels were high enough in slugs in the early stages of crop growth (~13 and ~6 ng per slug at the cotyledon and two-leaf stages respectively) to harm their beetle predators.

Because of this previously unsuspected pathway, neonicotinoid seed treatment may be detrimental when and where slugs are an important pest: the outcome of thiamethoxam soybean seed treatment in this trial was a 19% reduction in established plant numbers and a 5% reduction in final yield.

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[1]Douglas, M.R., Rohr, J.R. and Tooker, J.F. (2014) Neonicotinoid insecticide travels through a soil food chain, disrupting biological control of non-target pests and decreasing soya bean yield. *Journal of Applied Ecology*. DOI: 10.1111/1365-2664.12372.

Apple Scab Antagonist Protects the Canopy

Apple scab caused by the fungus *Venturia inaequalis* has been described as the economically most destructive disease in world apple production. It is especially prevalent in humid, cool conditions. It attacks leaves, flowers and fruit, defoliating trees and reducing yield and quality of the fruit, potentially making it unmarketable. Disease control, currently heavily dependent on fungicides, could be improved by the antagonist *Cladosporium cladosporioides* H39, a biological control agent being developed in Europe as a result of EU-funded research in the REPCO and CO-FREE projects, both aiming to reduce the use of copper-based fungicides in organic farming.

Venturia inaequalis overwinters in fallen leaf litter where saprophytic colonization gives rise to pseudothecia which produce ascospores. These airborne spores are released during spring and produce a primary season of infection by infecting young leaves and fruitlets on apple trees. Driven by this primary infection, summer epidemics arise from conidia produced on leaves, with rain splash aiding conidial dispersal and subsequent infection of new leaves and fruit, meaning they can become infected throughout the growing season. There is little prospect for resistant cultivars at present (owing to breakdown of the main resistance gene and consumer preferences) and control relies on multiple fungicide applications. Timing of treatments is critical as failure to control scab in spring cannot usually be redressed later in the season. Despite alternating use of different active ingredients, fungicide resistance is an increasing problem, exacerbated by EU restrictions on fungicide use and residues – and there is of course growing demand for fruit produced without pesticides.

What other options are there? Preventative measures to reduce the initial inoculum by removal of leaf litter have a use in orchard systems. In addition, long-running biological control research efforts have focused on antagonists of the overwintering fungus, but prospective agents for applying to the canopy have been rare.

Cladosporium cladosporioides H39, isolated from a sporulating colony on an apple leaf in a Dutch orchard, was identified to have potential for application in the canopy during previous research, in which fungi were screened for potential to reduce conidial infection of apple seedlings. Since then, two years' trials have been conducted with different apple cultivars under a range of orchard conditions in Germany, Hungary and Poland. The results provide a first demonstration that stand-alone applications of *C. cladosporioides* H39 can reduce apple scab in leaves and fruit, and in both organic and conventional orchard systems. Reductions comparable to those achieved with fungicides were recorded for both calendar sprays (every seven days in appropriate weather conditions) during the primary and summer seasons, and sprays applied one day after a predicted scab infection event. This latter finding was explored further in a separate field trial in the Netherlands, which found that a single spray before or up to several days after an infection event could protect apple trees from scab development. Efficacy with *C. cladosporioides* H39 treatment was comparable to that achieved with the common fungicide schedules (using copper oxychloride, calcium polysulfide, dithianon or dodine), reaching 42–98% for leaf scab and 41–94% for fruit scab, depending on weather conditions and scab incidence.

The scientists involved in the trials say the results are a first step towards the use of *C. cladosporioides* H39 in an integrated scab control strategy and reducing dependency on chemical fungicides, although further understanding of the biology of the antagonist will improve its application as a scab control measure.

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[1]Köhl, J., Scheer, C., Holb, I.J., Masny, S. and Molhoe, W. (2014) Towards an integrated use of biological control by *Cladosporium cladosporioides* H39 in apple scab (*Venturia inaequalis*) management. *Plant Disease* 'First Look'.
Web: http://dx.doi.org/10.1094/PDIS-08-14-0836-RE

New Weed Biocontrol Catalogue Online

The fifth edition of *Biological Control of Weeds: a World Catalogue of Agents and Their Target Weeds*[1] is now available online:
Web: www.ibiocontrol.org/catalog/

The print version is available soon and can also be downloaded as a PDF (current to end 2012) from the above site. Funding for both versions of the catalogue was provided by the US Forest Service (Forest Health Technology Enterprise Team). Work was conducted by MIA Consulting, the University of Georgia, the Queensland Department of Agriculture, Fisheries and Forestry, the University of Idaho and CABI, in collaboration with weed biocontrol scientists around the world. Particular gratitude is expressed in this edition to Mic Julien, who of course collated and wrote the first edition back in 1982 and has been the lynchpin ever since. He was closely involved in the most recent updating in retirement. It's a nice touch that the PDF version of this edition includes "JulienCatalogue" in the file name. Well done everyone, it's a joy to use.

Any changes or information for future updates, please send to Rachel Winston (rachel@getmia.net).

[1]Winston, R.L., Schwarzländer, M., Hinz, H.L., Day, M.D., Cock M.J.W. and Julien, M.H. (eds) (2014) *Biological Control of Weeds: a World Catalogue of Agents and Their Target Weeds*, 5th edn. FHTET-2014-04. USDA Forest Service, Forest Health Technology Enterprise Team, Morgantown, West Virginia. 838 pp.

XIV ISBCW Proceedings Published

The Proceedings of the acclaimed XIV International Symposium for the Biological Control of Weeds have been published and can be downloaded from the conference website.

Impson, F.A.C., Kleinjan, C.A. and Hoffmann, J.H. (eds) (2014) *Proceedings of the XIV International Symposium on Biological Control of Weeds*, Kruger National Park, South Africa, 2–7 March 2014. University of Cape Town, South Africa.
Web: www.isbcw2014.uct.ac.za/