INTERNATIONAL BIOHERBICIDE GROUP

IBG NEWS

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THE CHAIRMAN'S COMMENTS

Greetings to everyone!

July 2001 marks the tenth anniversary of the International Bioherbicide Group. As you will recall, the IBG was formed during the International Workshop on "Realizing the Potential of Bioherbicides," held February 11-13, 1992 at Orange, New South Wales, Australia. Since then, the group has maintained a forum, through this newsletter, to discuss and disseminate information concerning biological control of weeds by using plant pathogens. Sustained by an informal group of scientists who contribute news items and bring up discussion topics, the newsletter continues to serve as an important networking tool. The group has also managed to hold five bioherbicide workshops in different countries and in conjunction with major international conferences. It is heartening to realize that the group is providing a useful, vital link to promote the use of plant pathogens as weed control agents. I wish the group continued success.

I extend my special thanks to Maurizio Vurro for his tireless efforts to assemble, edit, and disseminate the newsletter. As always, I look forward to these newsletters with great interest and anticipation.

CHAIR
R. Charudattan (Charu)
1453 Fifield Hall, PO Box 110680, Plant Pathology Department, University of Florida, Gainesville, FL 32611-0680 USA Phone: 1-352-392-7240; Fax: 1-352-392-6532; e-mail: rc@gnv.ifas.ufl.edu

VICE CHAIR
Alan K. Watson
Department of Plant Science, Macdonald Campus of McGill University, 21,111 Lakeshore Road, Ste-Anne-de-Bellevue, QC H9X 3V9 CANADA Phone: 1-514-398-7851 Ext. 7858; Fax: 1-514-398-7897; e-mail: watson@macdonald.mcgill.ca
OR

APPA Division, International Rice Research Institute, MCPO Box 3127, 1271 Makati City, PHILIPPINES
Phone: 63-2-845-0563; Fax: 63-2-891-1292; e-mail: a.watson@cgiar.org

NEWSLETTER EDITOR
Maurizio Vurro
Istituto Tossine e Micotossine da Parassiti vegetali - C.N.R. - Viale Einaudi, 51 - 70125 - Bari - ITALY
Phone: +39.0805486037 - Fax: +39.0805486063 - e-mail: ma.vurro@area.ba.cnr.it

MEETINGS

N.A.T.O. Advanced Research Workshop
Enhancing biocontrol agents and handling risks

June 9-15, 2001, Hotel Villa Pitiana, Florence, Italy
Co-Directors: Maurizio Vurro & Jonathan Gressel

The increased relevance given to life and food quality, and environment preservation must be balanced with the needs to produce enough food to sustain humanity. Food production without pest (weed, disease and insect) control is impossible. The environmental considerations as well as the evolution of pesticide resistances requires that chemical pest control be augmented and significantly supplanted by other procedures. Despite many promising results obtained using biological control agents, their utility is still quite limited, replacing only a small part of the market of chemical products. This is due to many reasons, such as difficulties to find new and suitable agents, technological limits in application, storage and formulation of organisms, environmental restrictions of application, costs of registering, producing and delivering biopesticides. The major reason though is that very few agents have the efficacy of traditional chemicals, yet typically costing more than chemical pesticides. The advent of biotechnologies and molecular biology has opened new perspectives in using microrganisms in biocontrol, by allowing improvements in the properties of biocontrol agents and their delivery. The potential new risks associated with the introduction of new genes and organisms must be adequately accessed, which is a recurring motif throughout the proposed program. It is only if these issues are fully and transparently addressed can the necessary lines of communication be opened to the public.

The major reason for the meeting of minds in the NATO workshop was to exchange views and results on how a shift in balance towards efficient pest control can be efficiently achieved - safely. The major means explored is to improve the virulence of the biocontrol organisms such that a larger part of the target population is decimated, more quickly with a lower inoculum of biocontrol agents. The biocontrol agent can be cultivate industrially and applied like a pesticide, and will not become extinct. The above requires a multidisciplinary approach including molecular biology and genetics, physiologists and biochemistry of both the agent and the target, as well as epidemiology and ecology. The culture and delivery systems are then of utmost importance, as an enhanced agent must reach its target. All this must be performed safely to the applicator, to other crops and beneficial organisms and to the consumer. It must be ensured as well as possible that the biocontrol agents do not attack beneficial organisms, do not spread off target, and are not residual in the environment for longer than needed.

The workshop had a subject-oriented structure to permit the meeting of the major exponents in the scientific community working with different biological agents (fungi, bacteria, virus, nematodes, insects), on different targets (pathogens, insects, weeds). This multi-disciplinary group has backgrounds in the different aspects (molecular biology, formulation, genetics, risk assessment, new technology, biochemistry, physiology)

needed to design biocontrol agents with improved efficacy. 43 scientists from 18 countries attended the Workshop.

The workshop has had 4 major themes:

1. Risks from enhanced biocontrol agents and their mitigation. *(Chair: Jonathan Gressel)*
2. Needs for enhanced biocontrol agents and strategies for enhancement. *(Chairs: Maurizio Vurro and David Sands)*
3. Technology of enhancing biocontrol agents (chemistry, physiology, biochemistry, industrial technology). (*Chairs: Tariq Butt and Gary Harman)*
4. Genetics and molecular biology of enhancing biocontrol agents. (*Chairs: Ray St. Leger and Donald Nuss)*

The workshop has been organized to have in each day one main opening lecture related to the first of the mentioned themes, followed by lectures on one of the other three main sessions. In each session, lasting 1–1.5 day, several speakers have been invited to show progress in their field of interest, update results, and mainly to propose and discuss new approaches. Each talk was followed by a long discussion, whose most important aspects have been transcribed and will have trace in the book that will be published in autumn by IOS Press, The Netherlands. In fact, to speed up the process of editing and publishing the book, the lecturers have been required to come with ready manuscripts.
Several specific workshops have also been organized during the five-days-workshop:

- **Unmet needs for biocontrol agents** (Sands and Vurro)
- **Other technologies of enhancing biocontrol agents** (Butt and Harman)
- **Workshop - Finding the new genes** (St. Leger)
- **BioSafety Workshop** - Epilogue - what we have learnt that must be considered (Gressel, Kinderlerer, Tzotzos)
- **Marketing enhanced Biocontrol Agents** (Lidert, Johnson and Lisansky)

Space for short presentations was left to the other participants for self introduction of their research and why they attended. The workshop was the first meeting devoted to enrich biocontrol agents and generated considerable enthusiasm by all attendees in all sessions, and long discussions after each lecture. The discussions were recorded and will be part of the book. Many of the participants only knew a portion of the others due to the multi-disciplinary nature of the meeting, yet the group dynamics generated quickly interactions, and long lasting synergies are expected.

PEOPLE & PLACES

TOP

University of Florida, Gainesville

We are continuing our work on the following weed-bioherbicide systems: *Phomopsis amaranthicola-Amaranthus* spp.; *Dactylaria higginsii-Cyperus rotundus;* and fungal cocktails to control grasses. The Gainesville research team includes, R. ("Charu") Charudattan, Professor; Jim DeValerio and Mark Elliott, Senior Biologists; Angela Vincent, Camilla Yandoc, and Matt Pettersen, Graduate Assistants; and S. Chandramohan, Jugah Kadir, and Pablo Morales-Payan, Postdoctorates.

All three graduate students will graduate in the fall term and continue in doctoral programs (Angela and Matt) or undertake a postdoctoral position (Camilla). Dr. Jugah Kadir, Assistant Professor, Universiti Putra Malaysia,

Serdang, Selangor, is back in Gainesville on a sabbatical program from his university. Jugah did his Ph.D. work in Gainesville on the *Dactylaria*-purple nutsedge system. Dr. Pablo Morales-Payan, likewise, did his Ph.D. work in Gainesville on weed interference in vegetable crops. Dr. Chandramohan, who developed the fungal cocktail system for the control of several grasses, is continuing this work as a postdoc. Recently he and Charudattan received a U.S. patent (No. 6,235,347) titled, "Enhanced Bioherbicidal Control of Weeds Using Multiple Pathogens.

Submitted by R. Charudattan

Studies completed

Santosh Kumar Pandey, Department of Bioscience, R. D. University, Jabalpur - 482001(M.P.) India. E-mail: Pandeysk_2000@usa.net

Title of the work (Ph.D. 2000): *Mycoherbicidal potential of some fungal pathogens for the anagement of Lantana camara L.*

The *Lantana camara* L., a plant of tropical America was introduced in India at the beginning of 19th century as an ornamental and hedge plant. Its strong capacity to survive in a wide range of climatic and edaphic conditions and abundant vegetative growth makes it as a serious and hazardous weed in India which infested various forests, pastures, railway lines, roadsides etc. in almost all the states of the country. The present study was initiated with the objectives to evaluate the mycoherbicidal potential of indigenous fungal pathogens and to develop a suitable mycoherbicide for the management of this noxious weed. Extensive and periodical surveys were done to collect diseased plants parts of *Lantana* from the different parts of M.P.. More than 40 fungal isolates were recovered from the weed. Maximum LAD (leaf area damage) were caused by *A. alternata* (LC#20), *Phoma multirostrata* (LC# 31), *Fusarium xysporum* (LC#34), *Cochilobolus lunatus* (LC#35), *Pestalotiopsis guepinii* (LC#60), *Colletotrichum dematium* (LC#52), *Sclerotium rolfsii* (LC#58) and *Corynespora cassicola* (LC#61). These isolates were selected for further studies and their disease description and morphological characteristics were studied. The data of second screening on *Lantana* seedlings clearly revealed that maximum seedling mortality as well as leaf area damage was observed when plants were inoculated with *A. alternata* (LC#20). Looking to its potential and aggressiveness this pathogenic isolate of *A. alternata* (LC#20) was selected for further evaluation as a mycoherbicidal agent. Histopathologic observation of *A. alternata* (LC#20) on *L. camara* revealed that plant tissues have been successfully colonized within 12h and pathogen initiated syndrome formation and produced wide spread lesion and blight symptoms on the host leaves.

The requirement for optimum *in-vitro* growth, sporulation and virulence of the test fungus was defined. It was optimum at 28±1°C when grown on modified Richard's broth with a pH of 4.2. It was observed that not only the nature of carbon and nitrogen sources but also the proportions (C:N ratio of 1.75:1and 3.5:1) of these significantly influence the growth and virulence of the pathogen.

It was observed that the weed was highly susceptible till 2-4 leaved stage to all dosages of inoculum, after that there was a linear relation between the growth stages and inoculum concentrations. Pathogen showed maximum mortality of *L. camara* seedlings at 25°C and 30°C dew period temperature. Although, 6h dew duration was sufficient for the infection but longer dew period duration influenced increased mortality. Significant relation was also observed between relative humidity and mortality. Fungus caused mortality at very low RH i.e., 12%, which indicate its capability to survive and remain viable under low humidity level. With various

epidemiological factors, inoculum age also play an important role in the infection and viability of the pathogen. Maximum mortality was observed with 14 days old fungal cultures and a significant regression trend (R^2 = 1) was noted between inoculum age and mortality. The attempts were made to test the safety of the pathogen against non-target plants and revealed that the pathogen had no effect on other economically important plants associated with *L. camara*. However, fungus incited severe infection on *Xanthimum strumarium, Parthenium hysterophorus, Cassia tora, Hyptis suavaeolens, Ocimum sanctum* and different biotypes of *Lantana*.

Attempts were also made to test the compatibility of the pathogen with synthetic herbicides and other potential pathogenic isolates of *Lantana*. Screening test revealed that maximum mortality of *Lantana* seedling were caused by 2,4 DEE, Glyphosate and Paraquat. The selected herbicides when tested for *in-vitro* compatibility with test pathogen revealed that Glyphosate and Trifuralin induced maximum conidia germination at 0.5% and 0.1% concentration. Similarly pathogen was compatible with *Cochilobolus lunatus* (LC#35), and *Corynespora cassicola* (LC#61), which influenced synergistic effect. Various cheaper substrates with host stem and leaf extract were also screened for the mass production of the pathogen. Spores produced on *Lantana* leaf extract, stem extract and Dalia extract had strong potential to germinate as well as incited maximum disease severity. Selection of proper formulation is one of the major steps in the success of any microbial control programme. Different adjuvants were screened for their compatibility with *A. alternata* (LC#20). I*n-vivo* and *in-vitro* studies were performed which revealed that maximum seedling mortality observed when Glycerine was used as adjuvant followed by sunflower oil. The mycoherbicidal potential of the pathogen was tested in small-scale field trials which was carried out in the R.D. University Campus. Disease severity and mortality of the weed was maxium during monsoon season as compared to summer and winters.

Research Supervisor: Dr. A. K. Pandey, Sr. Lecturer.

a) The financial support of the Indian Council of Forestry Research and Education, Dehradun (India) is greatfully acknowledged.

b) Presently looking for PDF position in mycoherbicidal mangement of the weeds.

Dr. Christophe LAVERGNE - Environmental weed ecologist
Conservatoire Botanique National de Mascarin
Domaine des Colimaçons
F-97436 Saint-Leu - Ile de La Reunion (France)
Tel. : (0262) 24 92 27 - Fax: 0262 24 85 63
Mailto:clavergne@cbnm.org

" Invading strategies of an environmental weed in La Réunion Island, *Ligustrum robustum* subsp. *walkeri* (Oleaceae) ".

March 2000 - University of Réunion Island.

Abstract:

Ceylan privet, *Ligustrum robustum* subsp. walkeri (Oleaceae), has invaded all the indigenous forests in Mauritius island in less than 50 years, and has been introduced in La Réunion Island in the 60's. Now, privet represents a serious potential threat to the durability of the native forests of La Réunion. Ceylan privet is one of the rare examples in the world of alien woody species that are able to invade natural ecosystems initially little disturbed by human activities. Invasive species specialists are currently wondering about the mechanisms that contribute to biological invasions of natural environment. Searching the meaning factors of the success of

privet's invasion in old-growth forest in La Réunion was one of the purposes of this study. What's the invading strategy of that species? Do the native environments have a special sensitivity to the invader? Is it possible to predict how the Ceylan privet will spread over the island? What are the potential effects of the invasion upon the assortment, the structure and the running of the native ecosystems? Is it possible to control this alien invasive plant by ecological ways?
An experimental field work made it possible to study how the system invasive population-invaded environment is functioning. Monitoring of populations, experiments in laboratory and in nature were lead to underscore the intrinsic characteristics of the invasive plant, the interactions between species and the environmental factors associated with the invasion. Some measures were carried out in the native area of the plant (Sri Lanka), which lead to make some comparisons with the observations achieved in La Réunion.Privet species shows all the characteristics of the 'ideal' invader. Its ability to invade native ecosystems may be explained by a combination of specific biological advantages, and favourable biotic and abiotic environmental factors.
If we had known the biological achievements of *L. robustum* in its native area before, and if we had measured the vulnerability of La Réunion native ecosystems, we could have predicted the success of the invasion. Its introduction could have been avoided if we had applied the precautionary principle. Without a regional conservation programme, other species with the same invading hability might be introduced as well in the future, and might accelerate the degradation process of the last ecosystems of the Indian Ocean.

Key words: *Ligustrum robustum* subsp. *walkeri*, Oleaceae, alien invasive
plants, introduced species, biological invasions, native forest ecosystems, Réunion Island.

Cukurova University, Agricultural Faculty, Dept. of Plant Protection, Weed Science Working Group

Weed Science working group at Cukurova University studies on different research fields including weed biocontrol and allelopathy. Several researches are still in progress with the collaboration of national and international institutions.

- Allelopathy: Working since 1988. First project results are practically applied by the farmers in Turkey. *Raphanus sativus* (red radish) against *Sorghum halepense* (Johnson grass) control in cotton fields. Radish cultivation (half for harvest half for mixing into the soil) before cotton sowing succesfully controls johnson grass. Other projects on weed control using different preparation of various allelopathic plants, *Melia azedarach, Eucalyptus camaldulensis, Nerium oleander, Juglans regia*, *Thymus* spp., onion and garlic are going on. Currently one project is funded by Turkish Scientific Research Council entitled " Distrubution and Identification of *Orobanche* species, and their control with allelopathic plants in the East Mediterranean Region of Turkey".

- Biological Control Studies: Working since 1991. First PhD thesis on Fungal pathogens on common weeds of Cukurova with special emphasis on *Cynodon dactylon* and *Cichorium intybus* using *Dresclera cynodontis* and *Amerosporium concinnum* respectively was completed in 1997. Researches on biological control of important weed species such as *Acroptilon repens, Tamarix* spp. and *Centaurea solstitialis* are co-operative ongoing studies with the international organisations, CABI-Bioscience/Switzerland, EBCL/France, BBCA/Italy, USDA-ARS/USA.

Prof. Dr. F. Nezihi Uygur nuygur@mail.cu.edu.tr
Asst. Prof. Dr. Sibel Uygur suygur@mail.cu.edu.tr

Cukurova University, Agricultural Faculty, Dept. of Plant Protection, 01330 Adana/Turkey.

BIOHERBICIDE RESEARCH - STATUS REPORTS

This is by no means a complete account of all research projects on bioherbicides.

Bioherbicides a good alternative to conventional methods in integrated weed management for rice growers in Vietnam

Bruce Auld and Shane Hetherington
NSW Agriculture, Orange Agricultural Institute, Orange, NSW, Australia

Background

Developing effective and environmentally-safe methods for the control of grassy weeds in cereal production, especially rice, is important for a variety of reasons. Figures from the Mekong Delta suggest that weeds in rice reduce yield by about 34%. As rice provides about 35% of Vietnam's GDP, any increase in yield would be a considerable economic advantage. Control using chemical herbicides is difficult and expensive; in many countries, including Vietnam, much weeding is done manually as an alternative to herbicide use. In addition, the use of synthetic chemical herbicides may affect the production of fish and shrimp in rice paddies and may also contaminate the water collected for drinking. The development of herbicide resistance by the weed is another potential problem.

An alternative approach to controlling grassy weeds is the use of bioherbicides. These are applied like conventional herbicides but the active ingredient is a naturally- occurring microbe - usually a fungus. Such mycoherbicides are very specific, so there is less risk of damage to non- target plants. In addition, the bioherbicide does not persist in the environment beyond the target plant.

Prior to ACIAR project **CS2/94/002: Biological control of grassy weeds with fungi as bioherbicide,** there had been little work on bioherbicides in tropical countries. That project identified the major suitable fungal pathogens of the principal rice and wheat weeds. This ACIAR project **CS2/98/018: Bioherbicide development for cereals in integrated weed management** builds on this, by developing bioherbicides from the identified fungi. Project personnel aim to develop products which might be used by farmers to 'seed' agricultural by- products (such as rice husks or maize husks) to produce spores for on- farm use. This will provide a low-cost, easily accessible and sustainable source of bioherbicide for farmers.

Key collaborators in the project are from the National Institute of Plant Protection (Hanoi, Vietnam), Cuu- Long Rice Research Institute (Omon, Vietnam) and Cantho University (Cantho, Vietnam). The Australian project partners are from the Orange Agriculture Institute of the NSW Department of Agriculture. Project leaders are Dr Bruce Auld, NSW Agriculture; Dr Ha Minh Trung, National Institute of Plant Protection; Dr Duong Van Chin, Cuu Long Rice Research Institute; and Dr Pham Van Kim, Cantho University.

The Objectives of the project are to develop a biological herbicide for use against the most serious weeds of rice cultivation in the Mekong and Red River Deltas, and against the most serious weeds of wheat in Australia.

Achievements in Bioherbicide project

Achievements in the project to date have been encouraging.
The Cuu Long Research Institute (CLRRI) has conducted a weed survey which identified *Leptochloa chinensis* as a major weed of rice cultivation in the Mekong Delta; *Echinochloa crus- galli* was also important. A survey of pathogens of these weeds identified fungi that were effective in controlling both of these damaging weeds and screenhouse and other trials are continuing to confirm these findings.

Field experiments were conducted in June 2000 as a preliminary to a student's M.Sc. studies in collaboration with Cantho University. These experiments examined the effect of the three pathogens, alone and in combination on both weeds, and the subsequent effect on rice yield. Heavy weed infestations were found to reduce rice yield to 35- 40% of yields in hand- weeded control plots. Project leaders were pleased to note that all fungi tested as biological herbicides increased rice yields significantly.

The National Institute of Plant Protection (NIPP) has conducted preliminary field surveys, laboratory studies, screen house experiments and, since autumn 2000, some field studies.

An extensive survey of the Red River Delta was undertaken and *E. crus-galli* was identified as a principal weed of rice in the Delta. A survey of fungi associated with this weed identified 14 pathogenic genera. *Exserohilum monoceras* was chosen for further study following preliminary and host range testing. The same fungus has been independently suggested as the basis of a biological herbicide in Korea, Malaysia, Japan, China and the Philippines. A CIRAD sponsored group based at the International Rice Research Institute (IRRI, The Philippines) has provided valuable collaboration and has been visited by key Vietnamese personnel.

Ongoing laboratory studies at NIPP are optimising spore production of the fungi. Potato carrot agar supplemented by an *Echinchloa* extract has been found to be the best agar based media and darkness or near ultra- violet light promote sporulation. A two-phase spore production system adapted from IRRI has been found to be the best means of producing large numbers of spores for glasshouse and field experiments.

Screenhouse studies conducted by NIPP have found that *E. monoceras* (strain 85.1) is highly virulent against a range of *E. crus- galli* biotypes. All weeds are dead fifteen days after inoculation. The most widely grown cultivars of rice in southern and northern Vietnam are, however, totally resistant to *E. monoceras*. Studies have also revealed to researchers that weed seedlings are most susceptible at the 1 1/2 to 2 1/2 leaf stage.

In-field infection has been encouraging, but post-infection disease development has been disappointing. It is likely that lack of development is associated with this season's cooler temperatures. If this is the case, researchers are confident that no problem exists as rice cultivation and weed control are not normally practiced at this time of year. Controlled environment experiments are under way to examine the post-infection disease reaction to temperature.

In addition to the lab, screen house and field studies, several publications have been produced in association with this work. These include publications in Plant Protection Quarterly (1999), International Journal of Pest Management (2000) and Monthly Journal of science, Technology and Economic Management (2000) (*in Vietnamese*).

The researchers will also produce a weed identification book for Vietnam, which will be of use to extension workers, farmers and students.

Several presentations at international plant pathology, weed science and biological control conferences have also been given, in addition to poster presentations at consecutive Asia Pacific Weed Science Society conferences. The latter won awards for their quality.

The project has reached its half- way stage and is due to be completed in July 2002. It receives a boost with the arrival in early 2001 of Dr Brett Nietschke, a young weed scientist from Western Australia who will work on the project through AusAID's Australian Youth Ambassadors for development program. Brett will be based at NIPP in Hanoi.

Conidia of *Setosphaeria rostrata*, a promising fungus
for controlling *Leptochloa chinensis* in rice. It was isolated
and tested by scientists at the Cuulong Delta Rice Research Institute - Vietnam.
Courtesy of: Duong Van Chin duongvanchin@hcm.vnn.vn

Optimising biological control of a dominant weed in major crops

Piet Scheepens
Wageningen University and Research Centre (Wageningen UR)
Plant Research International
Department of Crop and Production Ecology
P.O.Box 16, NL-6700 AA Wageningen, The Netherlands
Email: p.c.scheepens@plant.wag-ur.nl

Under this title 6 European research institutions and a company carried out a 3 year EU-sponsored project. The general objective of the project was to develop an efficient microbial herbicide based on the fungus *Ascochyta caulina* for reliable field use against the weed fat hen (*Chenopodium album*) in a range of situations. The other participating institutions were the Universities of Aberdeen (UK) and Naples (IT), Long Ashton Research Station (UK), Institute of Toxins and Mycotoxins Research Bari (IT), Norwegian Plant Protection Institute Aas (NO) and the company Novartis (CH)

The work was based on earlier Dutch work. This focused on the potential use of *A. caulina* as a microbial herbicide to control *C. album* in crops. Unlike several pathogens, *A. caulina* can kill its host in appropriate conditions. It does not infect several crop species related to the weed host and seems to have a host range restricted to *Chenopodium* and *Atriplex* species.
Under field conditions, mortality of 70% of *C. album* plants, and a substantial growth reduction of surviving plants have been achieved by one application of *A. caulina*. The activity of the fungus is dependent on favourable weather conditions, especially humidity, immediately after application of the spores. In favourable conditions the fungus reduces the weed population sufficiently to prevent yield losses in maize, but not in sugar beet. It does not completely eliminate weed seed return to the soil.

The aspects that were investigated and the some results are presented below.

Identifying virulent new isolates of *A. caulina*
Around 250 new isolates from all over Europe were tested for virulence under environmental conditions that are favourable for the fungus, and a combination of spore density and plant stage that did not kill the Dutch reference isolate. Several isolates were as virulent as the reference or slightly better. Only very few, most of them from Norway, killed the test plants.

Activity profiles of new isolates
It was already known that spore density, high air humidity or a dew period after inoculation and plant development stage are crucial factors in the onset of disease and effects on the weed.
The influence of these and some other factors on disease development on *C. album* was investigated for 5 new isolates of *A. caulina*. Spore densities below 10^8 per m^2 of any of the tested isolates hardly reduced growth. The older the plants the less was the damage to the plants caused by all isolates.
Dew (>> 6 hours) was the single most important requirement for all isolates. Leaf nitrogen concentration was not a limiting factor under concentrations likely to be found in *C. album* on crop land.

Fungal attributes of *A. caulina* that contribute to activity
No significant correlation was found between the activity of cellulase or á-amylase and virulence of 6 *A. caulina* isolates.
Three phytotoxins were identified in the culture filtrate of *A. caulina* cultures: a sugar derivate of an unusual bis-amino acid named ascaulitoxin, its aglucon, and . 4-amino-proline. Large variation in concentration of the three toxins were found in 23 isolates of *A. caulina*. There was not an obvious correlation of toxin concentration and virulence of the respective isolate. Only little information is available of the individual toxins, but many experiments were done with the purified mixture of the three toxins. In the presence of a suitable wetting agent they caused leaf necroses on a wider range of plant species then *A. caulina* itself. The toxins showed an added or synergistic effect in mixtures with *A. caulina* spores or a low concentration of two herbicides.

Increasing performance and reliability
Spraying parameters for practical conditions were established with a track sprayer.
An invert emulsion was shown to enable infection in the absence of dew, but due to methodical difficulties in preparing and spraying this formulation of *A. caulina* spores this line of research was not followed any further.

A number of (mainly) polysaccharides in formulations were tested for their ability to reduce dew dependency of *A. caulina*. The dew dependency could be decreased by a few hours in this way but the reliability was still insufficient as shown in (semi-)field experiments.

Several herbicides were not toxic to A. caulina spores. Some of them added to the effect of the fungus or were even synergistic if added at low doses.

Integration of biological control in cropping systems

The best available isolates of *A. caulina* and the best formulation were finally tested in maize, sugar beet and cabbage crops in several European countries. The control effects varied from none to moderate, which is less than with the reference isolate in some previous years, and there was no difference between the isolates.

Conclusion

In general the system *C. album – A. caulina* is still very useful as a model for biological control of a noxious weed. From a scientific point of view there are obtained very interesting results concerning factors that influence the efficacy of the fungus. For practical use and a subsequent commercialisation the results were not encouraging, however. We did not find a more virulent isolate or a strain which can overcome drier conditions, nor a good formulation which can counteract those bad environmental conditions.

Biotechnology and Biological Control Agency

WHO WE ARE

The Biotechnology and Biological Control Agency is a new international non-profit association centered in Rome, Italy. Our aim is to provide a service in eco-compatible applied entomology, dedicated to the study and development of new least toxic systems to control weeds and insect pests. Main research areas include classical biological control of weeds and biological control of insect pests, screening and formulation of natural bioinsecticides, evaluation of synthetic pheromones and other new lure traps, breeding and selection of hybrids of crop plants showing resistance to insect pests. Our approach is focused on explorations of origin areas of pests and related biocontrol organisms, evaluation of their specificity by laboratory and field host range tests.

Official BBCA cooperators are the USDA ARS European Biological Control Laboratory, Montpellier, France, the Russian Academy of Sciences, Zoological Institute of St. Petersburg, Russia, the Agricultural University of Çukurova, Adana, Turkey, the University of Erzurum, Turkey, and the Agricultural University of Nitra, Slovakia.

WHY CONTACT US

To receive our newsletter, which will keep you informed about our progress
To support us applying for a membership position
To apply for a grant, Master, or PhD positions in co-operation with USDA-ARS
To suggest new cooperation projects > To cooperate with in going projects
To form and inform about biological control and sustainable agriculture practices worldwide

BIOLOGICAL CONTROL OF WEEDS

Alien weeds become often a problem in pasture and crop lands in United States and other countries.
A biological control approach involves the individuation, the screening and the evaluation of organisms (insects, mites and pathogens) closely related to the target weed. In particular, the evaluation of the host range is based on laboratory and field bio- assays, in no-choice and choice conditions. Three years and one hundred plant species are the minimum requirements to evaluate a new biocontrol candidate agent.
BBCA target weeds:

Centaurea solstitialis, Salsola australis, Convolvolus arvensis, Eupborbia esula;
BBCA exploration areas:
Mediterrancan Basin and Central Asia (Italy, Austria, Hungary, Slovakia, Russia, Armenia, Turkey, Uzbekistan, Iran, Romania, Greece, Morocco, Tunisia);
Laboratory activities:
micropropagation and *in vitro* culture, genetic analysis, behavioral observations;
Field activities:
insect and weed population dynamics; field data; host range open field choice tests; association, competition and behavioral observations.

BIOPESTICIDES
Biopesticides are poisonous chemicals, present in a living organism holding a variety of functions which can be helpful against insects.
Biopesticides have several advantages: it is more difficult for insects to develop resistance, they are often very specific and they are also safer than synthetic pesticides.
BBCA is able to evaluate the effectiveness of a biopesticide by means of an own protocol.
Laboratory activities:
Assessment of the general physiological mode of action; extension of the studies to the effects on target organs;
Field activities:
Evaluation of the effects on wild pest populations using several research methods.

SEMIOCHEMICALS AND CHEMIORECEPTION
The BBCA research on semiochemicals and insect chemioreception is mainly focused on field and laboratory development, improvement and testing of sexual and food based attractants.
Another important research area involves the analysis of pest's chemical orientation towards the host plant through electroantennography and behavioral bioassays.
Target pests: *Ceratitis capítata, Bactrocera oleae, Phthorimaea operculella, Leptinotarsa decemlineata.*

PLANT RESISTANCE
Breeding plant for resistance to insect concerns germ- plasm evaluation and identification of source of resistance with bioassays of antibiosis and antixenosis, electroantennography and olfactometry on the target insect pest; cross or somatic fusion, backcross and selection and field trials.
Plant chemicals and/or morphological traits interfere with insect behavior and growth: chemical analysis, compounds isolation, microscope observations are our tools in the characterization and the evaluation of resistance.

MICROPROPAGATION AND *IN-VITRO* CULTURE
The BBCA research on weeds includes the use of micropropagation and *in-vitro* culture of plants.
Our challenges are:
Micropropagation makes possible host-range and compatibility tests even on endangered, rare, useful and native plant species belonging to the same Family/Genus of the target weed;
In-vitro culture of weed and crop callus allows mass rearing of phytophagous pests and candidate biocontrol agents.
HOW TO CONTACT US
e-mail: info@e-bbca.net
website: http://www.e-bbea.net

regular mail : BBCA onlus
Largo Santo Stefano 3
00061 Anguillara Sabazia (Rome), Italy

Canadian Forest Service, Pacific Forestry Centre, Victoria, BC, Canada.

Dr. Simon F. Shamoun (Research Scientist) & Dr. Susanne Vogelgsang (NSERC Visiting Scientist)
Project: Biological control of salal (*Gaultheria shallon* Pursh.)

Abstract
Salal (*Gaultheria shallon* Pursh.), a perennial ericaceous shrub, is a serious competitor with conifer seedlings in coastal British Columbia. Current control methods are ineffective due to salal's extensive root system and leathery leaves that reduce herbicide translocation. A 1999 survey of the mycobiota associated with salal found a potential biological control agent, *Valdensinia heterodoxa* Peyronel. This fungus infects through multicellular, star-shaped conidia that are forcibly discharged onto the leaf surface. One of the major research challenges was to obtain adequate inoculum and a suitable delivery technique. An agar-based medium that allowed for sufficient production of conidia was developed, and the effect of various abiotic factors on growth, sporulation, and conidia discharge of *V. heterodoxa* were determined. In addition, a solid substrate method was established to assess virulence on intact salal plants. Our studies demonstrate that *V. heterodoxa* causes substantial leaf damage under laboratory conditions. Future research will focus on virulence under field conditions, evaluating the pathogenesis process, isolate selection, and deducing the genetic diversity of the fungus and salal.

Bioherbicide for Weeds Control
Cuban Institute for Research on Sugar Cane by-Products, Havana, Cuba.

A research National Project related to the production of Pseudomonas phytotoxic metabolites against Cuban weeds, has been developed since the past year.
The main objective of the project is to produce a Biological product constitute by phytotoxic metabolites effective against the main Cuban weeds. We have been working in the production by liquid fermentation of phytotoxic metabolites from *Pseudomonas aeruginosa* PSS in a 5 L fermentor and testing the phytotoxic activity by lettuce test, with promising results, and its isolation . At present time we know that they are peptidics compounds. Further work in the optimization of fermentation conditions and characterization of the isolated fractions is in progress.

María Elena Díaz de Villegas villegas@icidca.edu.cu

IMPECCA: developing a mycoherbicide against water hyacinth

In the last IBG Newsletter, Dr Charudattan introduced an exciting new development called the International Mycoherbicide Programme for Eichhornia crassipes Control in Africa (IMPECCA). Here, we welcome the opportunity to summarise the programme and provide further details on how to find more information about the

programme and our progress to date. Water hyacinth has spread throughout Africa during the last century, expanding its range particularly within the last decade. There are many well documented examples of the catastrophic effects that this weed has on riparian environments, and on communities which depend on them for their livelihood. IMPECCA's goal is the sustainable management of water hyacinth in Africa, and to achieve this, it will provide technical assistance to national programmes and develop a mycoherbicide for water hyacinth control based on a pathogen with a pan-African distribution.

The mycoherbicide will need to meet international standards of performance, safety and ease of application. It could replace the use of broad-spectrum chemical herbicides, such as glyphosate and 2,4,D, which in the past have caused concerns about contamination to fish, and degradation of water quality. In addition, a mycoherbicide may be more compatible with the use of insect biological control agents such as Neochetina spp.

In February 2000, US$2.5 million funding was granted by Danish International Development Assistance (Danida) for a 2½year phase. It is anticipated that programme scientists will have identified, characterised, formulated and field tested fungal agents for use as mycoherbicides by the time it ends. Amongst the key outputs will be:

- The strengthening of technical capacity and linkages within African national programmes to undertake biological control of weeds;
- Extensive exploration for pathogens of water hyacinth already present in Africa;
- Morphological, molecular and chemotaxonomic identification of both fungal isolates and water hyacinth biotypes;
- Assessment for suitability as the basis for a mycoherbicide, based on pathogenicity, acceptable host specificity, low mammalian toxicity, capacity for mass production and formulation;
- Laboratory and field testing for efficacy and compatibility with other (especially biological) control options;
- Development of population dynamic and analytical models in order to interpret the effects of interventions.
- A water hyacinth management strategy will be developed, appropriate for local needs.

This international, multi-institutional, multi-disciplinary programme is led by CABI Bioscience through its centres in the UK and Kenya, in collaboration with five other institutions: the International Institute of Tropical Agriculture, Benin (IITA), the University of Mansoura, Egypt, the Plant Protection Research Institute, South Africa (PPRI), the Seed Services of the Department of Research and Specialist Services, Zimbabwe (DRSS), and the Danish Institute of Agricultural Sciences (DIAS). The project will build upon existing studies of formulating water hyacinth fungal pathogens into mycoherbicides which have been carried out in Egypt, South Africa and Zimbabwe, and expertise gained by CABI Bioscience and IITA during the development and commercialisation of mycoinsecticides.

Updates on the programme can be found in the Water Hyacinth News newsletter (r.bateman@cabi.org to subscribe) produced by IMPECCA in association with the International Organisation for Biological Control (IOBC). Copies of the newsletter can also be downloaded together with other information outputs relating to the development of a mycoherbicide from the IMPECCA website at http://www.impecca.net/

CLASSICAL BIOLOGICAL CONTROL OF WEEDS WITH PATHOGENS TOP

We have launched a web site on our project on the bridal creeper rust (a classical biological control agent). You may include the link in thenewsletter if you want. The site has been developed for the communities whoare releasing the rust across Australia. We have had very good feedbacks. It may be of interest to other researchers in biocontrol.

The site url is:

http://www.ento.csiro.au/research/weedmgmt/bridal_creeper/

Louise.Morin louisem@ento.csiro.au

Canker and dieback of Scotch broom caused by Diaporthe inaequalis has been causing increasing damage to broom over the past few years on Vancouver Island, British Columbia, Canada. So far, I have not found this canker in Washington, Oregon or California but have had limited opportunities to look. Since this disease is common in Europe and apparently not too prevalent in North America, it would be interesting to know if it occurs in Australia, New Zealand, Hawaii and other countries where broom has been introduced during the past two centuries.

Scotch broom was first introduced to Vancouver Island in the 1850's, supposedly from seed collected in Hawaii. The first Diaporthe canker collections by the Canadian Forest Service were made in Powell River on the adjacent mainland in 1968 and it was also collected in Victoria and Parksville on Vancouver Island in 1993 and 1998, respectively. These collections are deposited in the Pacific Forestry Centre Mycological Herbarium in Victoria BC."

Ron Wall, rrwall@home.com

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Patent

- US Provisional Patent – Shamoun, Simon F. and Vogelgsang, Susanne (# 50549-1): "Culture, formulation, and delivery technique of *Valdensinia heterodoxa* for use as a biological control agent of salal (*Gaultheria shallon*)"

- ATTC deposit of two fungi strains of waterhyacinth, natives from Mexico Cercospora piaropi Tharp PTA-983 Acremonium zonatum (Saw) W. Grams PTA-984 (Maricela Martínez Jiménez)

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