Proceedings of the IX International Symposium on Biological Control of Weeds, pp.133-136. V.C. Moran and J.H. Hoffmann (eds). 19-26 January 1996, Stellenbosch, South Africa. University of Cape Town.

# Non-indigenous 'exploiter' plant pathogens as potential biocontrol agents

ERIC J. COTHER

*NSW Agriculture, Agricultural Research and Veterinary Centre, Forest Rd, Orange 2800, Australia*

**Abstract.** Inundative biocontrol with fungal plant-pathogens has been based on the use of indigenous pathogens. Endemic pathogens and their hosts may have developed a degree of homeostasis which may lessen their successful use in biological control, in which case, new associations between the biocontrol agent and the target plant may have more likelihood of success. The approach of Hokkanen and his idea of 'exploiter' species provides an added perspective for bioherbicide research. Effective biocontrol agents are seldom to be found among the 'average' exploiters of the target weed. More destructive agents are most likely to be found among the non co-evolved 'exploiter' species from outside the native area of the target plant. In Australia, a naturally occurring fungus, *Rhynchosporium alismatis,* is being studied as a potential mycoherbicide for control of two broad-leafed monocotyledenous weeds in rice. The weeds are suppressed but not killed by the fungus. The host range of *R. alismatis* extends to other genera in the family Alismataceae and spray-inoculation of seedlings may cause significant reductions in biomass production. Where indigenous Alismataceae species are important rice weeds with few endemic diseases, the concept of introduced inundative agents (under quarantine protocols) is a fertile area for research. *Rhynchosporium alismatis* and some Alismataceae species represent new 'exploiter'/target associations. Some field efficacy has been demonstrated with this pathogen in Indonesia but more strain selection, especially for temperature tolerance, is required.

## Introduction

Mycoherbicide research is now an established branch of plant pathology although the term 'mycoherbicide' is an unfortunate one. It sets expectations in the public mind (weed mortality) whilst concurrently creating mistrust (herbicide/residues) in their use (Winder and Sharmoun 1991). There is a need to adopt a more appropriate term to describe this control tactic. Biological control agents have long been regarded as 'alternative' control agents and the development of mycoherbicides has been conditional upon their desired properties being similar to those of chemical herbicides (Yang and TeBeest 1993). Although high target-specificity is seen by industry as a significant achievement for chemical herbicides (McMinn and Thomas 1991), the high level of host specificity observed in some fungal plant-pathogens is simultaneously seen as a strong argument for their use as mycoherbicides (Hasan and Ayers 1990) and as an economic hindrance to their development (Templeton and Heiny 1989; Greaves and McQueen 1990).

Each plant host/pathogen relationship is unique. Most plant pathogens do not cause rapid or complete death of the plants they infect unless environmental conditions stress the plants and, or, favour substantially increased frequency of infection and disease severity. Although complete kill of the target weed population is the theoretical aim of inundative biological control, it is rarely achieved and the continued use of this criterion will drastically reduce the number of candidate pathogens available for development as mycoherbicides.

The present tactic in classical biocontrol is to search the area of origin of the weed for diseases that are usually absent in the areas where the weed is now a problem. The approach in inundative biocontrol with fungal plant-pathogens has been based on the use of indigenous pathogens. They are exempt from quarantine regulations, have co-evolved with native plants and weed hosts, and selection for host specificity and adaptation to regional climatic conditions has already taken place (Charudattan 1988). Endemic disease is a result of homeostasis between host and pathogen (Charudattan 1988) and an endemic pathogen may thus be assisted to initiate epidemic disease by inundative application of inoculum.

## New associations

A particular fungus, however, has its own history of evolution with the target species and, if it has been abundant over a wide range of the host for a long evolutionary time period, then there is a greater possibility of co-evolution of some form of defence in the host species (Dennill and Hokkanen 1990). These researchers argued that specialized agents and their hosts may develop a degree of homeostasis which may hamper their successful use in biological control. They further argued that new associations between biocontrol agent and target plant have more likelihood of success. Although these arguments are derived from insect associations, they are equally valid for new host/plant-pathogen associations (Hokkanen 1985) and provide a different perspective for bioherbicide research.

A plant pathogenic fungus, *Rhynchosporium alismatis* (Oudem.) J.J. Davis, isolated in Australia from *Alisma lanceolatum* With. and *A. plantago-aquatica* L. (Cother *et al.* 1994) is currently under investigation as a potential mycoherbicide for two weed species (*A. lanceolatum* and *Damasonium minus* (R. Br.) Buch.) in the Alismataceae (Cother and Gilbert 1994b). Several species in the family Alismataceae grow in rice crops in many parts of the world, and they range from being considered serious weeds to being regarded as minor components of the rice weed-flora. For example, *Alisma canaliculatum* A. Br. and Bouche is a common paddy weed in Japan (Holm *et al.* 1979) and another *Alisma* species, *A. plantago-aquatica* has a wide distribution in most continents, other than Central and South America (Häfliger *et al.* 1982). It is a particularly serious weed of wild rice (*Zizania palustris* L.) in North America (Ransom and Oelke 1982). *Sagittaria guyanensis* H.B.K. occurs throughout Central and South America, and parts of Africa (Häfliger *et al.* 1982). It grows in South East Asia but is only a problem in Malaysia (Waterhouse 1993). *Sagittaria trifolia* L. has a similar distribution in S.E. Asia with the status of a weed in the Philippines and Vietnam. *Sagittaria pygmaea* Miq. is limited in its distribution to S.E. Asia, China, Japan and Korea where it is a serious rice weed (Anon. 1990; Park *et al.* 1993).

Where indigenous Alismataceae species are important rice weeds and there are few endemic diseases, then the approach of Hokkanen (1985) with his idea of 'exploiter' species is a fertile area for research. Effective biocontrol agents are seldom to be found among the 'average' exploiters of the target weed. More destructive agents are most likely to be found among the non co-evolved 'exploiter' species from outside the native area of the target weed (Hokkanen 1985, 1989). This is illustrated with the example of *R. alismatis* in southern Australia (Cother and Gilbert 1994a). The pathogen has co-evolved with *A. lanceolatum* and *A. plantago-aquatica* for many decades. Leaf necrosis is only observed in both species when plants are maturing. However, inoculation of seedlings with *R. alismatis* causes stunting in *A. lanceolatum,* but to a lesser extent in *A. plantago-aquatica*, which is an 'old world' host of this fungus, the first record being almost 120 years ago. A more pronounced stunting effect is observed in *S. guyanensis*, a species which does not occur in Australia, when inoculated at the seedling stage under glasshouse conditions (Cother and Gilbert 1994b). *Sagittaria guyanensis* has not previously been reported as a host of this pathogen. Some field efficacy on *S. guyanensis* has been demonstrated with this pathogen in Indonesia but more strain selection, especially for temperature tolerance, is required. Thus *R. alismatis* and *S. guyanensis* represent a new 'exploiter'/target association.

The identification process for potentially new bioherbicide agents could imitate that suggested by Hokkanen and Pimentel (1984): (i) identification of pathogens from herbarium surveys of disease occurrence on plants in the same genus or family elsewhere in the world (a fungus with a wide host-range need not be excluded as it may have use in a selective niche for weed control where the other potential non-target hosts are absent); (ii) obtain pathogens from a climatic region similar to that in which the target weed is growing; and (iii) select pathogens with a high degree of host specificity.

## Host-range testing and risks associated with the 'exploiter' approach

Efficacy studies with new fungi, or exotic strains of indigenous fungi, on the target weed and potential hosts, should include application of inoculm to plants at various growth stages and the results should be assessed not only on weed mortality, but also on plant growth-rates, biomass production and the plant's reproductive potential. Additionally, testing should include several isolates from different hosts and, or, geographic locations, for comparative assessment of variation within the pathogen and a greater chance of selecting the most virulent isolate from a heterogeneous population of the fungus. Host specificity should be considered a continuum of specificities within a pathogen population which may consist of sub-populations adapted to a smaller number of hosts (Weidemann and TeBeest 1990). Host-range studies conducted with these 'new' potential pathogens should approach the natural situation as much as is feasibly possible (Cother 1975) for the plant, pathogen and the environment (Watson 1985) and challenge taxa that represent the full range of plant genetic-diversity within the area of intended use (Weidemann and TeBeest 1990). Moreover, because of the capacity of a facultative pathogen to adapt to new hosts, the pathogen must be avirulent on useful plants, not just weakly virulent (Leonard 1982). If the weed species are not closely related to the indigenous flora, a less specific pathogen with a wider host-range may be more acceptable than would be the case if the problem weed was closely related to beneficial, or environmentally important flora. On the other hand, any risk or damage to non-target species must be evaluated relative to the risk to the environment and to society posed by the weed remaining uncontrolled, or being managed by large-scale application of chemical herbicides.

There are perceptions that technological 'constraints' hinder bioherbicide development (Auld and Morin 1995) and there are counter arguments that the only long-term constraints are attitudinal in origin (Cother 1996). Nevertheless, the efficacy of mycoherbicides will be greatly improved if the inundatively-applied inoculum is packaged with various adjuvants (eg., nutrient sources, wetting agents, u.v. protectants) to improve infection by providing artificial dew periods, etc. These requirements may act in favour of acceptance of the introduced pathogen in that its 'escape' beyond the target locality into the wider environment may be constrained by their absence. Host-range assessments should take into account that natural dispersal of inoculum from subsequently-diseased plants may not pose a threat to other plant species because of the absence of these specifically-enhanced requirements for infection. This would be particularly so if the requirements for infection were fundamentally a function of the biology of the pathogen rather than the biology of the plant. To be a risk to non-target plants, the introduced pathogen needs to establish and function temporally and spatially within an ecosystem. This may not occur if the human-assisted infection conditions are absent. Bioherbicide candidates should be viewed solely from the perspective of plant pathology: each host/pathogen relationship is unique and individual constraints peculiar to an association should not be used to support broad opposition should one candidate fungus prove unsuitable for wider use.

There will be plant pathologists who feel uneasy about the risks associated with this approach to the acquisition of new potential inundative agents. No matter how many families and genera are artificially challenged by the fungus, it is impossible to test adequately all potential plant encounters in every environment/inoculum combination to be certain that all future hazards are identified. The occurrence of unpredictable jumps to previously unknown hosts remains a risk (Walker 1983). Provided that properly considered testing is based on the precautionary principles discussed by Cother (1975), Leonard (1982) and Weidemann and TeBeest (1990), this approach holds no greater threats than those already accepted in the wider biocontrol disciplines (Wilson 1969). More than 700 introduced insects have been released in various countries for classical biocontrol (Julien, 1992) through set protocols for quarantine that have been in place for years. Similar procedures exist for plant pathogens (usually obligate parasites). Moreover, release of appropriately screened 'new' candidates should pose no greater threat than that envisaged for genetically-modified organisms (Charudattan 1990). In South-East Asia, national and regional centres such as ASEAN PLANTI already exist and have a unified approach to the introduction of biological control agents, to the dissemination of information and to personnel training (Sastroutomo 1992). In addition, FAO has recently established a code of conduct (Labrada-Romero 1995) for importation and release of exotic biocontrol agents. Such approaches will facilitate an expanded approach to biocontrol with access to a wider array of possible weed control pathogens.

## Acknowledgements

The helpful suggestions made on an earlier draft of this manuscript by Alex Nikandrow and Ross Gilbert are gratefully acknowledged.

## References

Anon. (1990) *Farmland weeds in China - a collection of coloured illustrative plates.* W. Zhirong (ed.). Agricultural Publishing House, Beijing.

Auld B.A. and Morin L. (1995) Constraints in the development of bioherbicides. *Weed Technology*, 9: 638-652.

Charudattan R. (1988) Inundative control of weeds with indigenous fungal pathogens. In: *Fungi in Biological Control Systems*, pp. 86-124. M.N. Burge (ed.). Manchester University Press, Manchester.

Charudattan R. (1990) Release of fungi: large scale use of fungi as biological weed control agents. In: *Risk assessment in agricultural biotechnology: Proceedings of the International Conference*, pp. 70-84. August 1988. Davis, California. Division of Agricultural and Natural Resources, University of California.

Cother E.J. (1975) *Phytophthora drechsleri*: pathogenicity testing and determination of effective host range. *Australian Journal of Botany*, 23: 87-94.

Cother E.J. (1996) Challenging current perceptions of biological control; A pathologist's perspective of the future of bioherbicides in weed management in Asian rice production. In: *Herbicide Use in Asian Rice Agriculture: Ecosystem Effects and Alternatives.* R.L. Naylor (ed.). International Rice Research Institute, Los Banos. (in press).

Cother E.J. and Gilbert R.L. (1994a) Efficacy of a potential mycoherbicide for control of *Alisma lanceolatum* and *Damasonium minus* in Australian rice crops. *Australian Journal of Experimental Agriculture,* 34: 1043-1050.

Cother E.J. and Gilbert R.L. (1994b) Pathogenicity of *Rhynchosporium alismatis* and its potential as a mycoherbicide on several weed species in the Alismataceae. *Australian Journal of Experimental Agriculture*, 34: 1039-1042.

Cother E.J., Gilbert R.L. and Pollock D.C. (1994). First record of *Rhynchosporium alismatis* on *Alisma lanceolatum* and *Damosonium minus. Australasian Plant Pathology*, 23: 46-49.

Dennill G.B. and Hokkanen H.M.T. (1990) Homeostasis and success in biological control of weeds - a question of balance. *Agriculture, Ecosystems and Environment*, 33: 1-10.

Greaves M.P. and MacQueen M.D. (1990) The use of mycoherbicides in the field. In: *The Exploitation of Micro-organisms in Applied Biology. Aspects of Applied Biology* 24, pp. 163-168. Association of Applied Biologists, Wellesbourne.

Häfliger E., Kähn U., Hämet-Ahti L., Cook C.D.K., Faden R. and Speta F. (1982) *Monocot Weeds 3.* Documenta Ciba-Geigy, Basel.

Hasan S. and Ayers P.G. (1990) The control of weeds through fungi: principles and prospects. *New Phytologist*, 115: 201-222.

Hokkanen H. (1985) Exploiter-victim relationships of major plant diseases: implications for biological weed control. *Agriculture, Ecosystems and Environment*, 14: 63-76.

Hokkanen H.M.T. (1989) Choosing an effective biocontrol agent - an evolutionary perspective. *Acta Entomologica Fennica*, 53: 19-24.

Hokkanen H. and Pimentel D. (1984) New approach for selecting biological control agents. *Canadian Entomologist*, 116: 1109-1121.

Holm L., Pancho J.V., Herberger J.P. and Pluckett D.L. (1979) *A geographical atlas of world weeds.* John Wiley, New York.

Julien M.H. (1992) *Biological control of weeds: a world catalogue of agents and their target weeds. Third edition.* CABI, Wallingford, UK.

Labrada-Romero R. (1995) Code of conduct for the import and release of exotic biocontrol agents. In: *Abstracts of papers and posters, Workshop on Biological Control of Weeds,* p. 6. 8-10 February, 1995, Montpellier, France. European Weed Research Society.

Leonard K.J. (1982) The benefits and potential hazards of genetic heterogeneity in plant pathogens. In: *Biological Control of Weeds with Plant Pathogens* pp. 99-112. R. Charudattan and H.L. Walker (eds). Wiley, New York.

McMinn A.L. and Thomas B. (1991) The impact of community regulations on the future development of new compounds: an industry view. In: *Proceedings of the Brighton Crop Protection Conference, Weeds,* pp. 563-570. Vol. 2. 1991.

Park K.H., Ku Y.C. and Oh Y.J. (1993) Changes of weed communities in lowland rice fields in Korea. In: *Proceedings of the 10th Australian and 14th Asian Pacific Weed Science Societies,* pp. 349-352. 6-10 September 1993 Brisbane, Australia. Weed Society of Queensland, Brisbane.

Ransom J.K. and Oelke A. (1982) Common waterplantain (*Alisma triviale*) interference with wild rice (*Zizania palustris*). *Weed Science*, 30: 10-14.

Sastroutomo S.S. (1992) The role of ASEAN PLANTI in the enhancement of biocontrol activities in the region. In: *Biological Control and Integrated Management of Paddy and Aquatic Weeds in Asia,* pp. 409-423. National Agricultural Research Centre, Tsukuba.

Templeton G.E. and Heiny D.K. (1989) Improvement of fungi to enhance mycoherbicide potential. In: *Biotechnology of Fungi for Improving Plant Growth,* pp. 127-151. J.M. Whipps and R.D. Lumsden (eds). Cambridge University Press, Cambridge.

Walker J. (1983) Possible problems in the use of plant pathogenic fungi for biological control of weeds in Australia. In: *Proceedings of the Fourth International Congress of Plant Pathology,* p. 14. 17-24 August 1983. Melbourne Australia. Rowprint Services, Melbourne.

Waterhouse B.F. (1993) *The Major Arthropod Pests and Weeds of Agriculture in S.E. Asia: Distribution, Importance and Origin.* Monograph No. 21. ACIAR, Canberra.

Watson A.K. (1985) Host specificity of plant pathogens in biological weed control. In: *Proceedings of the VI International Symposium on Biological Control of Weeds,* pp. 577-586. E.S. Delfosse (ed.). 19-25 August 1984, Vancouver, Canada. Agriculture Canada, Ottowa.

Weidemann G.J. and TeBeest D.O. (1990) Biology of host range testing for biocontrol of weeds. *Weed Technology,* 4: 465-470.

Wilson C.L. (1969) Use of plant pathogens in weed control. *Annual Review of Plant Pathology,* 7: 411-434.

Winder R.S. and Shamoun S.F. (1991) Terminology in microbial control of weeds. *Biological Control*, 1: 339.

Yang X.B. and TeBeest D.O. (1993) Epidemiological mechanisms of mycoherbicide effectiveness. *Phytopathology,* 83: 891-893.