

Soilborne fungi associated with root galls of *Lepidium draba* caused by *Ceutorhynchus* spp.

Anthony J. Caesar

USDA/ARS Pest Management Research Unit, 1500 N. Central Ave., Sidney MT 59270

Abstract

Isolation of fungi from insect-damaged roots of *Lepidium draba* in Switzerland, Hungary and Austria revealed that this species was often infected with one or more soilborne fungi. Plants with evident stunting and/or chlorosis and reddening of leaves nearly always exhibited root damage by one or more insects. In Switzerland, roots of *L. draba* damaged by one or more unknown insects were infected with *Rhizoctonia solani* and *Fusarium* spp. Surveys for *L. draba* plants with characteristic chlorosis and reddening in Hungary and Austria in 2004 and 2005 showed that such plants had root galls typical of weevils of *Ceutorhynchus* spp. The galls were often decayed at exit points of the insects and adjacent root tissue was consistently infected with *Rhizoctonia solani*,

Fusarium and *Pythium* spp. A similar complex of soilborne fungi had been found in association with insect damage to roots of leafy spurge and spotted and diffuse knapweed and comparative virulence tests of such fungi showed that greater virulence was associated with isolates of this origin. The prevalence of such associations, especially in yet another instance involving a highly invasive herbaceous perennial weed may indicate that such highly destructive insect/pathogen combinations are a previously overlooked key to biological control success. A concept for utilizing a multitrophic approach to screening for new biocontrol agents of exotic, invasive herbaceous perennial weeds of rangelands will be discussed.



Figure 3. Field on the outskirts of Salt Lake City, Utah (left), undergoing rapid invasion by *Lepidium draba*, compared to a typical stand of *L. draba* in Europe (right). *L. draba* populations in the U.S. have no root-feeding insects; furthermore, none of ca 40 isolates of *Fusarium* spp. from U. S. stands tested on *L. draba* has shown pathogenicity to white top. Natural biological control of European populations of *Lepidium draba*, as exhibited by the comparative density of *L. draba* in foreign settings, is apparently multitrophic in nature.

Introduction

Lepidium draba is a deep-rooted, highly invasive perennial weed of exotic origin, capable of aggressively and rapidly expanding infestations (Figure 3.) Such infestations occur in riparian areas, rangelands and other natural areas and cause multiple negative biological and economic effects.

Biological control is often the sole viable option for the control of invasive plants. However, increasing regulatory and public attention to possible nontarget effects (Pearson and Callaway, 2004) has led to a reconsideration of how biocontrol programs are conducted and a consensus that the previous practices have resulted in what has been termed a "lottery approach" (Balciunas, 2004). Thus, this work was conducted in a manner that considered this consensus and past and recent findings by the author that indicate the value of combined searches for the multitrophic effects that are essential to biocontrol of such weeds as *L. draba*. Highly regarded candidate insects for biological control of *L. draba* are *Ceutorhynchus* spp. which have shown a high degree of host specificity. As this new consensus and recent critical findings indicate, the potential of *Ceutorhynchus* will not be based on its narrow host range alone, but on a demonstrated potential for a high degree of impact (Caesar, 2000, Balciunas 2004) and thus a reduced hazard level of nontarget effects (Pearson and Callaway, 2003). It has been concluded by the author that these concerns are best addressed with indications that the mode of biocontrol action by the insect is synergism with plant pathogens (Caesar, 2005). Thus, investigations are in progress to assess the prevalence of such interactions in the native range of *L. draba* and *Ceutorhynchus* spp.

Results

Galls characteristic of *Ceutorhynchus* spp. on *Lepidium draba* (Figure 1) were found to be infected with soilborne fungi. The set of species found in the galls and tissue adjacent to the galls included *Rhizoctonia* spp., *Fusarium oxysporum* and *F. solani* and pythiaceus fungi. A similar complex of soilborne fungi associated with insect damage has been found to infect leafy spurge in association with root damage caused by larvae of the insects *Aphthona*, *Chamaesphacia* or *Oberea* spp. and have been identified and extensively characterized. A similar synergistic syndrome has been found for spotted and diffuse knapweeds.

In each instance root tissue adjacent to the galls was cultured on a range of media (Acidified potato dextrose agar, Ko and Hora medium, Nash and Snyder medium and corn meal agar with pimarinic-ampicillin-rifampicin), one or more of *Rhizoctonia* spp., *Fusarium* spp. and pythiaceus fungi (as yet unidentified) were isolated. The table at right above indicates that there is typically a complex of soilborne fungi associated with insect damage to roots of *L. draba*. Figure 4 shows diseases of *L. draba* caused by *Rhizoctonia* (A), *Fusarium* (B) and *Phoma* spp. (C) in association with insect damage. Similar complexes have been found to occur with insect damage to roots of leafy spurge and spotted and diffuse knapweed. In each of these cases *Rhizoctonia* was the most aggressive invader of root tissue (Figure 2).

Sample name	Location	Insect presence	Symptoms	Fungi isolated
98f-3	Saillon-Fully, Switzerland	+	Roots with vascular necrosis	<i>Rhizoctonia</i> spp.
99f-3, 4	Scanno,	<i>Ceutorhynchus</i> spp.	root galls	<i>Rhizoctonia</i> , <i>Fusarium</i> spp.
99f-7, 9	Conthey, Switzerland	+	Chlorosis and stunting, root lesions and stem cankers	<i>Rhizoctonia</i> spp.
00f-4	Saillon-Fully, Switzerland	+	Roots with vascular necrosis	<i>Rhizoctonia</i> spp., <i>Fusarium</i> spp.
00f-5	Conthey, Switzerland	—	Dead top-growth, vascular streaking	<i>Fusarium oxysporum</i>
04f-2	Mezokovezd, Hungary	+	Insect damaged roots	<i>Fusarium</i> spp.
04f-5	Balmazjuvaros' Hungary	+	Chlorosis, root damage	<i>Fusarium oxysporum</i>
05f-5	Besenyotelek, Hungary	<i>Ceutorhynchus</i> spp.	root galls	<i>Rhizoctonia</i> , <i>Fusarium</i> spp. pythiaceus fungi
05f-7	Poroszlo, Hungary	<i>Ceutorhynchus</i> spp.	root galls	<i>Rhizoctonia</i> , <i>Fusarium</i> spp. pythiaceus fungi
05f-12	Theresienfeld, Austria	<i>Ceutorhynchus</i> spp.	root galls	<i>Rhizoctonia</i> , <i>Fusarium</i> spp. pythiaceus fungi



Figure 1. The chlorosis and reddening of foliage of *L. draba* related to the occurrence of galls typical of those caused by *Ceutorhynchus* spp. and infection of the roots with soilborne fungi as a consequence.

Figure 2. Vascular tissue of a root of *L. draba* with a central area of internal necrosis from which *Rhizoctonia* spp. and *Fusarium* spp. were isolated, typifying such roots when they were damaged by insect feeding.



Conclusions

These findings indicate the potential of *Ceutorhynchus* spp. for the successful biocontrol of *L. draba*, given the evident association of this and other root-attacking insects with infection of roots by species of soilborne fungi, which have been shown by the author to be highly pathogenic in previous studies (Caesar, 2005), especially the isolates from insect-damaged root tissues. These findings indicate that the collection of *Ceutorhynchus* spp. by locating plants of *L. draba* with galls is facilitated by the simultaneous occurrence of chlorosis and reddening which is often accompanied by significant stunting. Additionally, such plants were predominantly found to be infected with soilborne fungi. This case illustrates that integrating the exploration for new insects as candidate biocontrol agents and the isolation of likely synergistic partners is readily done. Future programs would benefit from combined searches for both insects and associated synergistic fungi. Prerelease co-assessment of insects and their apparent synergists has the potential to realize an emphasis on the highest impact as the necessary central focus of biological control of invasive weeds (Caesar, 2000; Pearson and Callaway, 2004) of *L. draba*, emulate the success of biological control of leafy spurge through the of direct synergistic interaction between insects and plant pathogenic root infecting fungi, a mode of action recognized both in the biocontrol of leafy spurge (Lym and Carlson, 2002) by non plant pathologists, and increasingly regarded of general importance in weed biocontrol (Bacher et al., 2002; Sing et al., 2005).

The mechanism through which natural control in the native range of this species is exerted presents a preview of how success upon release and establishment of *Ceutorhynchus* is achievable. It is likely that the soilborne fungi described here are key effectors of the impact necessary to reduce stand density of *L. draba* as was shown in the case of leafy spurge (Caesar, 2003) where it was shown that soilborne fungi were more than twice as likely to be responsible for mortality of leafy spurge compared to insects, using comparative risk survival analysis. The supplementation with plant pathogens along with releases of insect with a capacity for synergisms may be necessary for effective biological control. Such a program is planned for Colorado to "jump-start" biocontrol synergisms at established, but yet to be effective, release sites of *Aphthona* spp. Prerelease testing of *Ceutorhynchus* spp. in combination with the fungi described herein is planned in the quarantine greenhouse facility at Montana State University, Bozeman.

Literature Cited

- Bacher, S., Friedli, J., Schar, I. 2002. Developing in diseased host plants increases survival and fecundity in a stem-boring weevil. *Entomologia Experimentalis et Applicata* 103: 191–195.
- Balciunas, J.K., 2004. Are mono-specific agents necessarily safe? The need for pre-release assessment of probable impact of candidate biocontrol agents, with some examples. In: Cullen, J.M., Briese, D.T., Kriticos, D.J., Lonsdale, W.M., Morin, L. Scott, J.K. (Eds.), Proceedings of the XI International Symposium on Biological Control of Weeds, CSIRO, Canberra, pp. 252–257.
- Caesar, A.J., 2000. Insect-pathogen synergisms are the foundation of weed biocontrol. In: Spencer, N.R. (Ed.), Proceedings of the X International Symposium on Biological Control of Weeds, 4–14, July 1999, Montana State University, Bozeman, Montana, USA, pp. 793–798.
- Caesar, A.J., 2003. Synergistic interaction of soilborne plant pathogens and root-attacking insects in classical biological control of an exotic rangeland weed. *Biol. Control* 28: 144–153.
- Caesar, A.J., 2005. Melding ecology, classical weed biocontrol, and plant microbial ecology can inform improved practices in controlling invasive plant species. *Biological Control* 35: 240–246.
- Fumanal B., Martin, J. F., Sobhian, R., Blanchet, A., Bon, M. C. 2004. Host range of *Ceutorhynchus assimilis* (Coleoptera : Curculionidae), a candidate for biological control of *Lepidium draba* (Brassicaceae) in the USA. *Biological Control* 30: 598–607.
- Lym, R. G., Carlson, R. B., 2002. Effect of Leafy Spurge (*Euphorbia esula*) Genotype on Feeding Damage and Reproduction of *Aphthona* spp.: Implications for Biological Weed Control *Biological Control* 23: 127–133.
- Pearson, D.E., Callaway, R.M., 2003. Indirect effects of host-specific biological control agents. *Trends Ecol. Evol.* 18: 456–461.
- Sing, S. E., Peterson, R. K. D., Weaver, D. K., Hansen, R. W., Markin, G. P. 2005. A retrospective analysis of known and potential risks associated with exotic toadflax-feeding insects. *Biological Control* 35: 276–287.

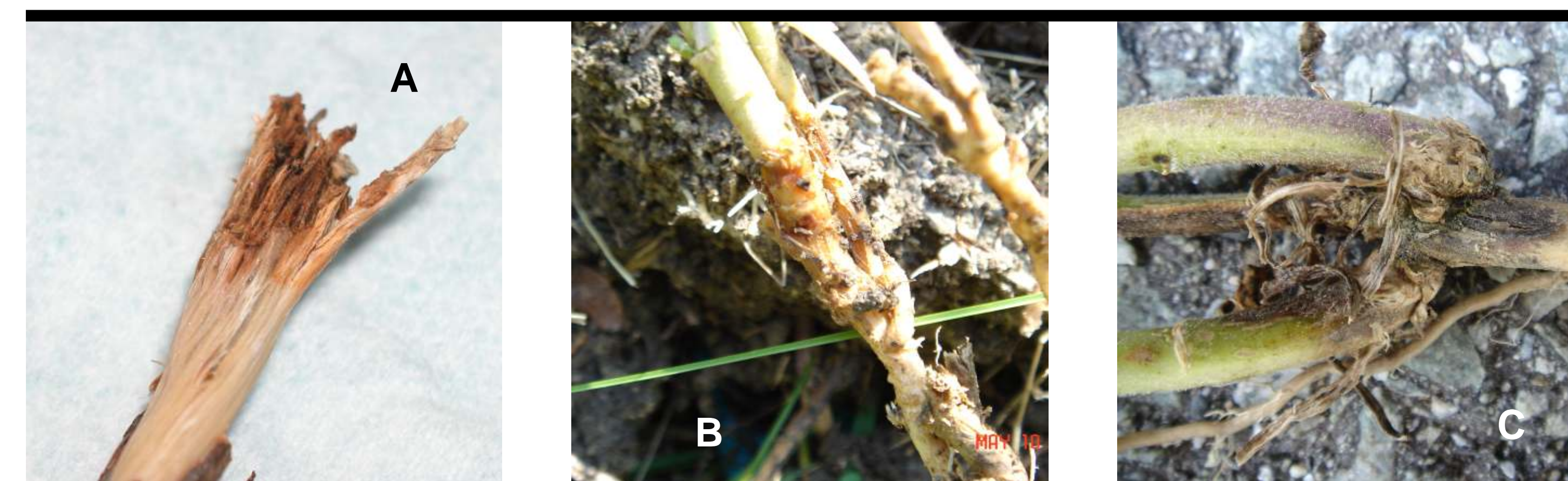


Figure 4. Root and crown diseases of *L. draba* associated with insect damage through the tunneling (A, showing discolored and infected root tissue distal to the insects-damaged tissue) and mining (B, C) of root cortexes of *L. draba* by insects.