

# Survey and Evaluation of Fungal Pathogens for Biological Control of Grass Weeds

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## ABSTRACT

Wild oats and green foxtail rank as the most abundant annual grass weeds in the Canadian prairies. Prolific seed production, life cycle similarities to cereal crops and the development of herbicide-resistance make these weeds major pests. Extensive surveys for microbial weed control agents (foliar and soilborne) were conducted from 1994 to 1996. In detached leaf bioassays, 36 foliar fungal agents from green foxtail and 24 from wild oats were pathogenic. Out of 70 soilborne fungi isolated from roots and crowns of wild oat plants, nine isolates reduced germination by 90% or greater in growth pouch bioassays. Soil application of the mycelium at a concentration of 0.8% (w/v) resulted in lower weed emergence over a 3-wk period. Adjuvants improved efficacy of three foliar agents for green foxtail control. Spray application method also had an impact on the efficacy of foliar biocontrol agents. Application with an airbrush provided superior control over a flat-fan nozzle, but with higher water volumes (1800 L/ha), a flat fan nozzle provided similar control to that exhibited with the airbrush. Development of suitable formulation and application methods will be important for preserving efficacy under more practical application conditions.

## INTRODUCTION

Wild oats (*Avena fatua* L.) and green foxtail (*Setaria viridis* [L.] Beauv.) rank as the two most abundant annual grass weeds in the Canadian prairies. Prolific seed production and the development of herbicide-resistance make these weeds economically important pests (Beckie and Morrison, 1993). Biological control with fungal pathogens provides another weed control strategy that can complement cultural and chemical control methods. This type of biological weed control, also known as bioherbicides, involves the application of high inoculum levels of a host-specific fungal pathogen over a weed-infested area (Boyetchko, 1997). Although the majority of bioherbicides investigated have been foliar fungal pathogens, some researchers have been exploring soilborne fungal pathogens (Charudattan, 1991).

The objectives of this research were i) to conduct surveys for soilborne and foliar fungal agents for biological weed control and ii) to evaluate their efficacy and possible formulations in controlled environment conditions.

## MATERIALS AND METHODS

### Surveys for soilborne and foliar pathogens

**Soilborne pathogens:** Over 70 agents isolated from roots of wild oats were evaluated in growth pouches. Mycelial suspensions (1 ml/seed, 2% w/v) were placed onto wild oat seeds (10 seeds/pouch) and incubated for 7 days (20°C, 16-hour photoperiod); germination was determined.

**Foliar pathogens:** In weed disease surveys conducted from 1994 to 1996, fungi were identified from green foxtail and wild oats. Using detached leaf bioassays, potential biocontrol agents were identified by fulfilling Koch's postulates.

### Efficacy and audication of soilborne and foliar pathogens

**Soilborne pathogens:** A mycelial suspension of agent **133** was added directly to wild oat seeds (1 mL/seed, 2% w/v) and as a soil inoculant to a depth of 5 cm with doses of 0, 0.08, 0.8, and 2.0% mycelium/cm<sup>3</sup> soil. Emergence was evaluated weekly up to 3 weeks.

**Foliar pathogens:** Efficacy of 3 formulations (water, non-ionic surfactant, adjuvant mix) applied at the 't-leaf-stage with an airbrush and flat fan nozzle (water volumes of 200,600, and 1800 L/ha) were evaluated for 3 agents of green foxtail. Disease severity was assessed after 7 days.

## RESULTS AND DISCUSSION

### Survey for soilborne and foliar pathogens

**Soilborne pathogens:** Nine of the 70 fungi evaluated in growth pouch bioassays reduced wild oat germination by greater than 90% (Figure 1). Direct application of a mycelial suspension can have significant deleterious effects on germination. Identification and host-range tests are underway to evaluate the potential of these nine pathogens.

**Foliar pathogens:** A total of 73 fungal isolates in 12 genera showed pathogenicity in detached leaf bioassays (Table 1). *Bipolaris* spp. and *Fusarium* spp. were recovered more frequently in green foxtail and *Drechslera avenacea* and *Cephalosporium* spp. were most common on wild oats. Pathogenecity in these bioassays does not guarantee that these agents will be suitable for biological control. Further evaluation is being conducted.

### Efficacy and application of soilborne and foliar pathogens

**Soilborne pathogens:** Agent 133 caused significant reductions in emergence when directly applied to wild oat seeds (Figure 2). Application of the fungus to soil at a concentration similar to the seed treatment (0.08% w/v) did not result in significant reductions in emergence when compared to controls. However, in dose response studies, increasing the concentration of mycelium from 0.08% to 0.8% and higher reduced wild oat emergence over a 3-wk period (Figure 3).

**Foliar pathogens:** The adjuvant mix, followed by the surfactant alone, improved efficacy of the 3 agents compared to the formulation controls (Figure 4). Application of all 3 agents with an airbrush resulted in almost 100% control of green foxtail (Figure 5). Spray application with a flat-fan nozzle at very high water volumes (1800 Wha) provided similar control to that exhibited with the airbrush. Lower water volumes with the flat-fan nozzle gave inferior control.

## SUMMARY

- Targeting both soilborne and foliar fungal pathogens will provide diversified strategies for biological control of grass weeds. Further research in host-range, efficacy, formulation, etc. are required.
- Evaluation of effective delivery systems and dose response that reflect practical inoculum levels will dictate the suitability of the fungal agents for biological control.
- Adjuvants improved efficacy of the foliar agents, but phytotoxicity and reduced selectivity have been observed. Research into novel formulations that do not exhibit these negative characteristics is required.

- Spray application method had an impact on the efficacy of the foliar biocontrol agents. The airbrush provided an effective and aggressive spray that bombarded the plant surface with water and air, thus predisposing the plant to injury and wounds of entry for the biocontrol agents. The flat-fan nozzle may be less effective due to reduced spray retention compared to the airbrush at practical volumes. Leaf surface characteristics which control wettability will play a role in determining the optimal formulation and application method.

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**Table 1.** Number of fungal isolates from green foxtail and wild oats showing pathogenicity in detached leaf bioassays in surveys conducted from 1994 - 96.

Weed	Fungus	Year			Total
		1994	1995	1996	
<b>Green Foxtail</b>	<i>Alternaria</i> sp.	2	1		113
	<i>Bipolaris</i> spp.	6	4	1	
	<i>Cephalosporium</i> spp.	2			
	<i>Colletotrichum</i> spp.	2	2		
	<i>Fusarium</i> spp.	9	6		
	<i>Phoma</i> spp.	1	2	2	
	<i>Pyricularia grisea</i>	2	5		
	Unidentified fungi				
<b>Wild Oats</b>	<i>Cephalosporium</i> spp.	5			5
	<i>Colletotrichum</i> spp.	1	1		2
	<i>Drechslera avenacea</i>		8		9
	<i>Fusarium</i> spp.		2		2
	<i>Verticillium</i> spp.		1		1
	Unidentified fungi		2		2

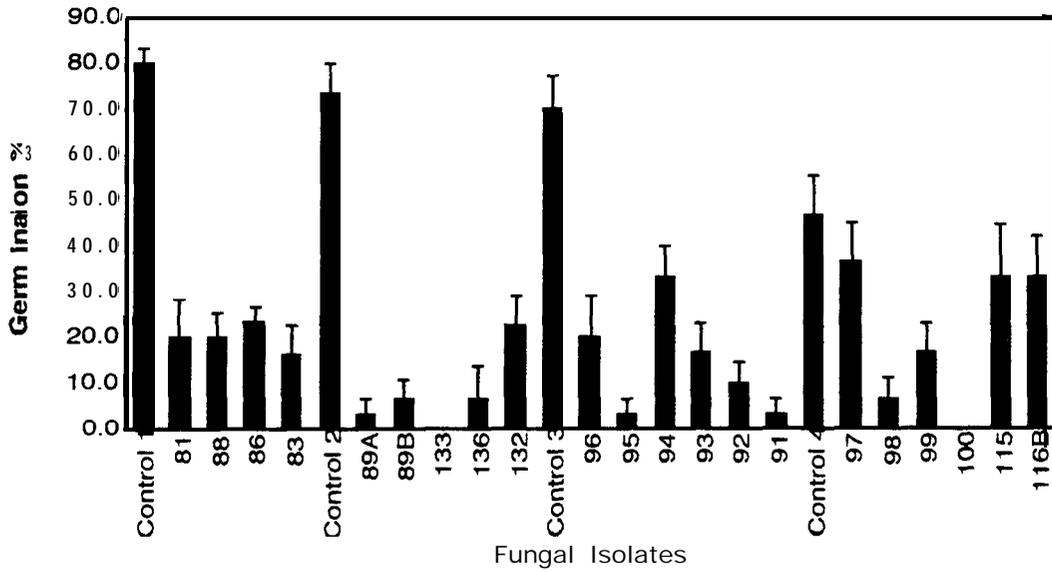


Figure 1. Effect of 21 fungal agents on wild oats germination in growth pouch bioassays. The suppressive activity of each agent was compared to uninoculated controls.

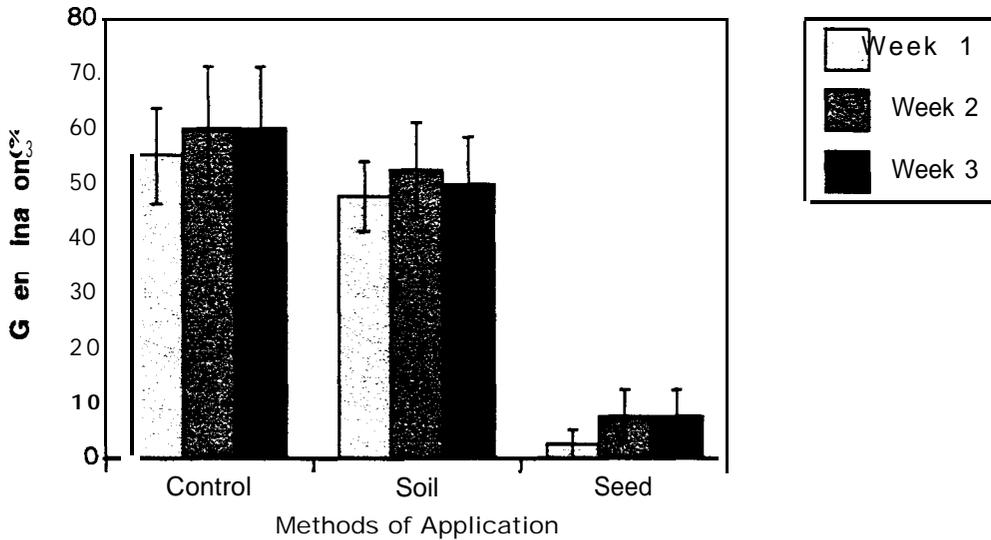
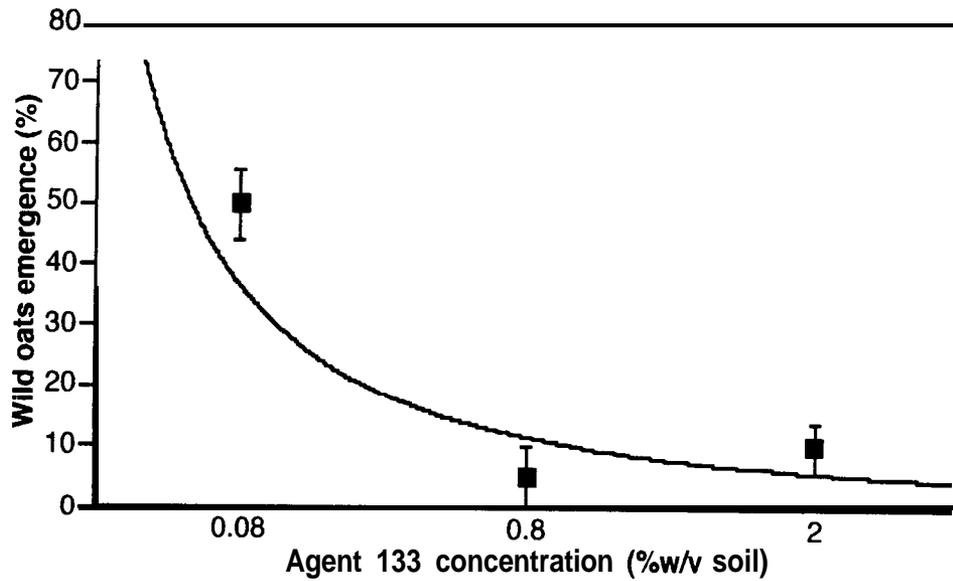
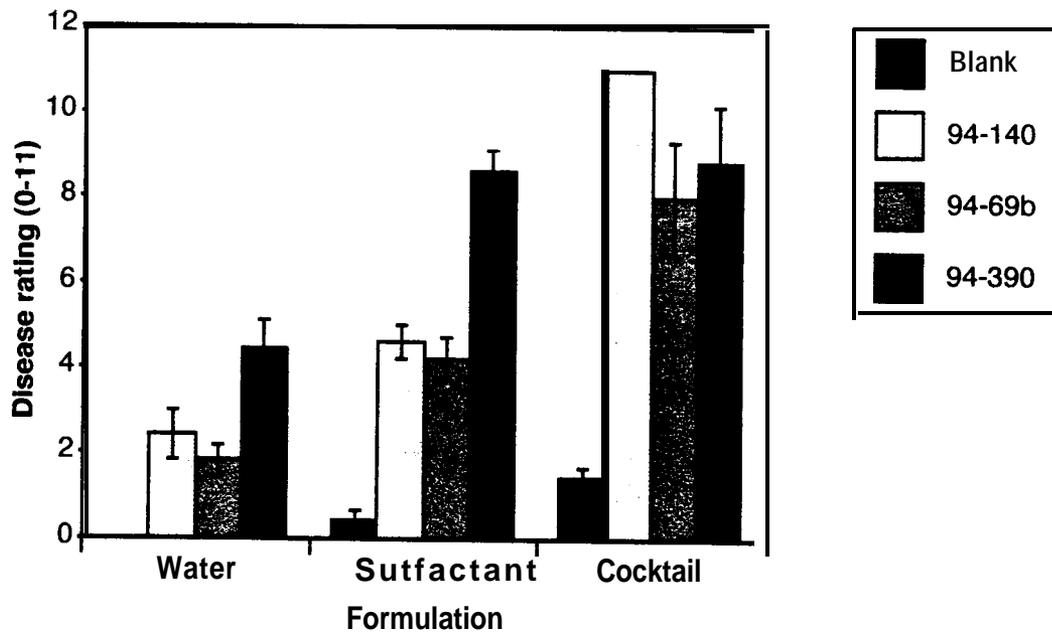


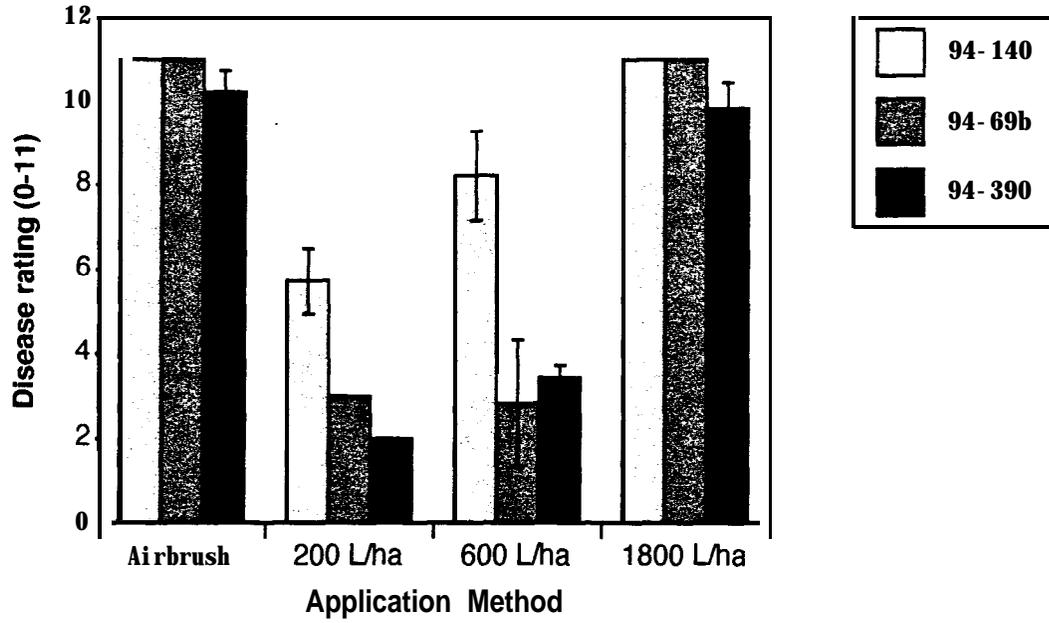
Figure 2. Effect of fungal agent 133 on emergence of wild oats. Treatments consisted of mycelium mixed with soil (0.08% w/v, mycelium/cm<sup>3</sup> soil) and 1 ml direct application (2% w/v) onto wild oats seeds. Controls consisted of water minus the fungal agent. Experiments consisted of 10 seeds/pot, 4 reps per treatment.



**Figure 3.** Typical dose response of fungal agent 133 (soil inoculations) on wild oats emergence after 1 week.



**Figure 4.** Effect of formulation (water, surfactant, adjuvant mix) on disease of green foxtail by 3 fungal pathogens. Disease ratings (0-11) according to Horsfall-Barrett. Surfactant contained Agral 90; adjuvant mix contained Intac (1% v/v), Silwet L-77 (0.15% v/v), dextrose (0.1% w/v), and gelatin (0.05% w/v). Formulation applied with XR8002 flat-fan nozzle, 275 kPa, 600 L/ha.



**Figure 5.** Effect of spray application method (airbrush vs. flat fan nozzle with different spray volumes) on control of green foxtail by 3 fungal pathogens. Disease ratings (0-11) according to Horsfall-Barrett. Formulation used was adjuvant mix.