IXth INTERNATIONAL BIOHERBICIDE GROUP WORKSHOP

ORLANDO, FLORIDA, USA
February 8, 2009

Supported by a conference grant from the United States Department of Agriculture, National Research Initiative

Also sponsored by:

novozymes®
Scotts
SePRO

Rethink Tomorrow
Evaluation of *Alternaria alternata* as a potential biocontrol agent for field bindweed (*Convolvulus arvensis*)

Ehsanallah Zeydali, Natural Research Center of Lorestan, Iran; Alireza Koocheki, Ferdowsi Univ. of Mashhad, Iran; Nader Azadbazht, Natural Research Center of Lorestan, Iran and Reza Ghorbani*, Ferdowsi Univ. of Mashhad, Iran. * Email: ghorbani43@gmail.com

**Abstract**

Field bindweed can be found in a wide range of habitats and causes substantial yield reduction in many crops. *Alternaria* spp. are airborne molds that were proposed to have potential for the biological control of weeds. Results of the present study showed that *Alternaria alternata* isolate A2 developed the most infection among different spore concentrations (10^4, 10^5, 10^6 and 10^7 spores in 1 ml distilled water) the treatment of 10^7 spores in 1 ml at 4 leaf stage caused most weed control of field bindweed. The maximum disease development was observed with application of the fungus of *A. alternata* at dew periods of 24 and 48 hour, however, plant damage was also observed with a length of 6 hours dew period. These experiments have confirmed the potential of *A. alternata* as a mycoherbicide under specific environmental conditions.

**Key words:** Biological control, Bioherbicide, Dew period, Saturation humidity, Spore concentration.

**Introduction**

Field bindweed (*Convolvulus arvensis*) is an important perinial weed of agricultural crops world-wide that can be found in a wide range of habitats and causes substantial yield reduction and causes problems in harvest in many crop. Biological control agents, especially plant pathogenic fungi, have been shown to have control potential for specific problem weeds. However, under field conditions control activity of mycoherbicides is often variable. Detailed knowledge about the environmental conditions required for disease development and weed mortality is an important prerequisite for the development of mycoherbicides and their use in integrated weed control strategies (Ghorbani et al, 2005). Susceptibility of plants to a given dose of pathogen inoculum may vary with weed growth stage. In some plant pathogen combinations, the hosts are susceptible only in seedling stages, and older plants become resistant (Agrios, 1997), although the pattern varies depending on pathogen host systems. Kadir and charudatton (2000) reported which *Cyperus rotundus* in 4-6 leaf stage is more sensitive toward older plant (8 leaf stage), to fungus *Dactilaria higginsii*. Ghorbani et al (2000) found that the best stage for biological control of *Amaranthus retroflexus* with fungus *Alternaria alternata* observed at 2-4 leaf growth stage. Abundant, prolonged, or repeated high moisture, whether in the form of rain, dew, or high humidity, is the dominant factor in the development of most epidemics caused by fungi, bacteria and nematodes (Greaves et al. 1999). The aim of this study was to investigate the effects of field bindweed developmental stage, spore concentration, temperature and humidity or dew period on the activity of different *Alternaria alternata* to assess their potential for biological control.

**Material and Methods**

A series of studies were carried out in Faculty of agriculture, Ferdowsi University of Mashhad, and Natural Research Center of Lorestan, Iran, in order to evaluate *Alternaria alternata* collected from Mashhad, against field bindweed. The weed seeds were planted in the pots with 10cm diameter and 15cm height contains sandy loam soil. In the first experiment, three plants per pot at different growth stages (cotyledons, 4, 6 and 9-11 leaf-stages) were sprayed with 10^7 spores per ml of *Alternaria alternata* strain A2. In the
second experiment, three *Convolvulus arvensis* plants per pot were sprayed with a suspension containing different *A. alternata* spore concentrations of $10^4$, $10^5$, $10^6$ and $10^7$ spores in 1ml distilled water. Control plants were sprayed by distilled water. To create a relative humidity of over 90%, treated plants were immediately covered with plastic bags for 48 h. Pots were then placed in a greenhouse with 60 to 65 %relative humidity. Three plants per pot were scored for disease development 3 and 10 days after spraying using the following scoring system: 0 = no symptoms, 1 = 1 to 25% necrosis of total leaf area, 2 = 26 to 50% necrosis of total leaf area, 3 = 51 to 75% necrosis of total leaf area, 4 = 76 to 99% necrosis of total leaf area, 5 = %100 necrosis of total leaf area (plant death). After the second assessment, plants were cut at the soil surface and their aboveground fresh weight was determined. Plant materials were dried in an oven (70 C, 48 h) and dry weight was recorded. For dew requirement experiments, *C. arvensis* plants at 2-4 leaf stage were sprayed with suspensions of $10^7$ spores per ml of *A. alternata*. Treated plants were covered with plastic bags to maintain %90 humidity and incubated for 6, 12, 24 or 48 hours in growth cabinets.

Results and discussion
Results showed that *Alternaria alternata* isolate A2 had a higher effects than the fungus *Fusarium* sp.. The effect of fungi was significantly different (p < %5) between plant in different growth stages and both fungi were more effective at 2-4 leaf-stages (Fig. 1).

Fig. 1. Interaction effects of plant growth stage and fungi isolates of *Alternaria* sp. and *Fusarium* sp. On area under the curve of disease development (AUCDD) in field bindweed. Different letters indicating differences between means based on Dunkan test (%5).

The maximum disease development were observed with application of the fungus of *A. alternata* at dew periods of 24 and 48 hour, however, plant damage was also observed with a length of 6 hours dew period (Figs 2 and 3).
Fig. 2- Effect of dew period length on disease development caused by *A. alternata* in field bindweed. Different letters indicating differences between means based on Duncan test (%5).

Fig. 3- Effect of dew period length on the area under the curve of disease development (AUCDD) OF *Alternaria alternata* in field bindweed of 4 leaf stage.

In conclusion, for maximum activity of *A. alternata* as a mycoherbicide for *C. arvensis*, the inoculum should be applied (a) at a rate of 10⁷ spores ml, (b) to plants at the two- to four-leaf growth stages, and (c) when environmental conditions are favorable for high humidity to occur on the foliage of *C. convolvulus*, for at least 12 hours after inoculation. According to the above findings, there are two major limitations for using *A. alternata* as a mycoherbicide.
Acknowledgment
The authors gratefully acknowledge the Faculty of Agriculture, Ferdowsi University of Mashhad, Iran for financial supports.

References