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Effects of soil-borne *Rhizoctonia solani* on canola seedlings after application of glyphosate herbicide

Glyphosate herbicide is a non-selective and broad-spectrum herbicide that has activity against both broad-leaved and grassy weeds. It binds rapidly to soil particles, and so exhibits negligible residual activity in soil because it is not available for absorption by plants. It can remain unbound in soils where the glyphosate binding sites on soil particles are occupied by phosphorus. Also, glyphosate is exuded from the roots of glyphosate susceptible plants into the rhizosphere, where it can affect soil microbial populations. Glyphosate has been shown to reduce the growth of some bacterial endophytic communities in plants and to reduce populations of some beneficial soil microorganisms. It has also been reported to increase the populations of some soil-borne fungal pathogens and the severity of the diseases that they cause.

Rhizoctonia is a genus of fungal pathogens that often becomes more prevalent after application of glyphosate. Pre-emergence application of glyphosate resulted in a rapid elevation in root rot severity caused by *Rhizoctonia* spp. on susceptible weeds and volunteer plants. Greater root rot severity was associated with an increase in inoculum concentration of the pathogen, which in turn was associated with a higher frequency of damping-off and root rot in the following crop. However, the impact of glyphosate application on *Rhizoctonia* spp may dissipate quickly.

Single *df* contrast	2008	2009
Plants m^{-2}		
Glyphosate applied	37	19
Control (no glyphosate)	33	16
Pr > F	0.01	0.01
Yield t ha^{-1}		
Glyphosate applied	2.8	1.5
Control (no glyphosate)	2.0	1.0
Pr > F	0.01	0.01

Table 1. Effect of pre-seeding application of glyphosate on seedling establishment and yield of canola in 2008 and 2009 in field trials inoculated with *Rhizoctonia solani* in the fall prior to planting, Edmonton AB.

Rhizoctonia solani is an important component of the seedling disease complex on canola (*Brassica napus* L.). Symptoms of rhizoctonia seedling blight on canola include reduced seed germination and seedling establishment caused by seed decay and pre- and post-emergence damping-off of seedlings.

In the present study, experiments were conducted under field and greenhouse conditions to assess the effect of pre-plant application of glyphosate on disease severity caused by *R. solani* on a glyphosate-resistant canola cultivar. A secondary objective was to determine if increasing the time between a pre-plant application of glyphosate and seeding of canola affected seedling blight severity and subsequent crop development under controlled conditions.

MATERIALS AND METHODS

Field Trial

The main plot factor was inoculation (treated versus control) and the subplot factor was application of glyphosate. The subplot treatments included: 1) application of glyphosate at 680 g ai ha^{-1} 10 days before seeding, 2) application of glyphosate approximately 10 days pre- and post-seeding at 440 g ai ha^{-1} per application 3) application of glyphosate at 680 g ai ha^{-1} 10 days after seeding, and 4) non-treated control.

The plot area was seeded on May 26, 2008 at a 1.5-cm depth using untreated seed of the glyphosate-resistant canola cv. 3465. The plots were harvested on October 10, 2008. The trial was repeated the following year.

Greenhouse Trial

Soil was collected from a field near the Crop Diversification Centre – North, Alberta Agriculture and Rural Development (AARD), Edmonton, Alberta, Canada. There were four glyphosate treatments: i) Pre-seeding glyphosate: the canola crop was seeded at either the 15-day pre-plant glyphosate interval or the 33-day pre-plant glyphosate application interval at 680 g ai ha^{-1} ii) Post-seeding (in-crop) glyphosate: glyphosate was applied at 680 g ai ha^{-1} at 14 days after crop seeding; iii) Pre- + Post-seeding glyphosate: the canola crop was seeded at either the 15-day pre-plant glyphosate interval or the 33-day pre-plant glyphosate application interval at 440 g ai ha^{-1} (double application field

This article is adapted from the research paper "Effects of soil-borne Rhizoctonia solani *on canola seedlings after application of glyphosate herbicide" by A. Rashid, S.F. Hwang, H.U. Ahmed, G.D. Turnbull, S.E. Strelkov, and B.D. Gossen published in the Canadian Journal of Plant Science, 2013, 93(1): 97-107.*

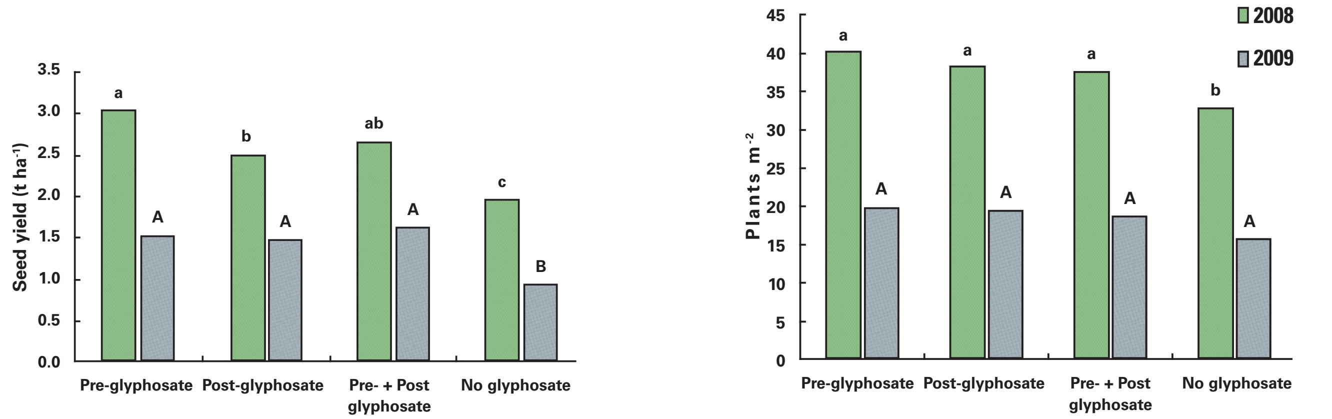

Fig. 1. Effect of timing of glyphosate treatment on seedling emergence and seed yield of canola in field trials at Edmonton, AB in 2008 and 2009. Bars within a year capped with the same letter do not differ based on single *df* contrast analysis at $P \leq 0.05$.

rate). The same rate of glyphosate was applied at 14 days after canola seeding (post-plant); and iv) Control: no glyphosate was applied and the cups were seeded only with canola. The experiments were repeated once under similar greenhouse conditions. The number of colonies of *Rhizoctonia* spp. per Petri dish was counted five days after plating and the number of colony forming units (cfu) per g of soil was calculated.

RESULTS

Field Trial

Application of glyphosate increased seedling emergence and subsequent seed yield of canola relative to the non-sprayed control in both years. (Table 1). Inoculation with *R. solani* reduced seedling emergence relative to the non-inoculated control in both years (data not shown). Inoculation also reduced seed yield in 2009, but the difference was not significant in 2008. Application of glyphosate increased seedling emergence over that of the non-sprayed control in 2008 (Fig. 1). There were no differences in emergence among the glyphosate treatments in either year, except that all were greater compared to the non-sprayed control in 2008. Similarly, application of glyphosate increased seed yield over that of the non-sprayed control in both years. There were no differences in yield among the glyphosate treatments in 2009, but yields in the glyphosate treatments were highest with pre-plant application, intermediate with the pre + post application, and lowest in the post-seeding application in 2008 (Fig. 1).

Greenhouse Trial

Establishment of canola 15 days after glyphosate application

Pre-seeding applications of glyphosate at either single (680 g a.i. ha^{-1}) or double (440 g a.i. ha^{-1}) application field rates reduced subsequent emergence of canola seedlings (emergence range = 19-53%) in the inoculated treatments relative to the non-sprayed controls (≥69%) (Table 2). In inoculated soil, pre-seeding glyphosate application increased the incidence of damping-off on canola seedlings (range 39-78%) compared to the non-sprayed controls (range 1-15%). As expected, no damping-off was observed in the non-inoculated controls. The concentration of CFUs of *R. solani* in the soil at the end of the trial was not consistently influenced by glyphosate application. The inoculum concentration treatments affected final CFUs, and there were no CFUs in the non-inoculated controls.

Establishment of canola 33 days after glyphosate application

Pre-seeding glyphosate did not affect emergence (range 83-96%) relative to the non-sprayed control (99% emergence) in the non-inoculated controls. Pre-seeding application of glyphosate did not affect the frequency of damping-off (range 0-9%) relative to treatments without pre-seeding glyphosate (range 0-5%) in the inoculated treatments, and there was no damping-off in the non-inoculated controls (Table 3). Pre-seeding glyphosate had no effect on plant height (range 14.0-16.5 mm) relative to treatments without pre-seeding glyphosate (range 14.0-19.0 mm) in the non-inoculated controls. Pre-seeding application of glyphosate did not affect the population of *R. solani* in the soil at the end of the experiment except that the number of CFUs was higher in treatments that received relatively higher initial inoculum concentrations at the outset of the experiment.

Comparison of response between seeding 15 and 33 days after glyphosate application

Contrast analysis across inoculum concentrations and glyphosate treatments showed that i) seedling emergence was higher, ii) the incidence of damping-off was reduced, and iii) plant biomass was larger (higher shoot and root weights), when canola was seeded 33 days after glyphosate application as opposed to 15 days after application (Fig. 2). Plant height was not significantly different between the two application intervals.

The population of *R. solani* in the soil, as determined by the number of CFUs at the termination of each greenhouse trial, was lower when the canola was seeded 33 days after glyphosate application compared to 15 days (Fig. 2). However the *R. solani* populations did not significantly differ between treatments that received pre-plant glyphosate compared to those without pre-seeding glyphosate on either application date.

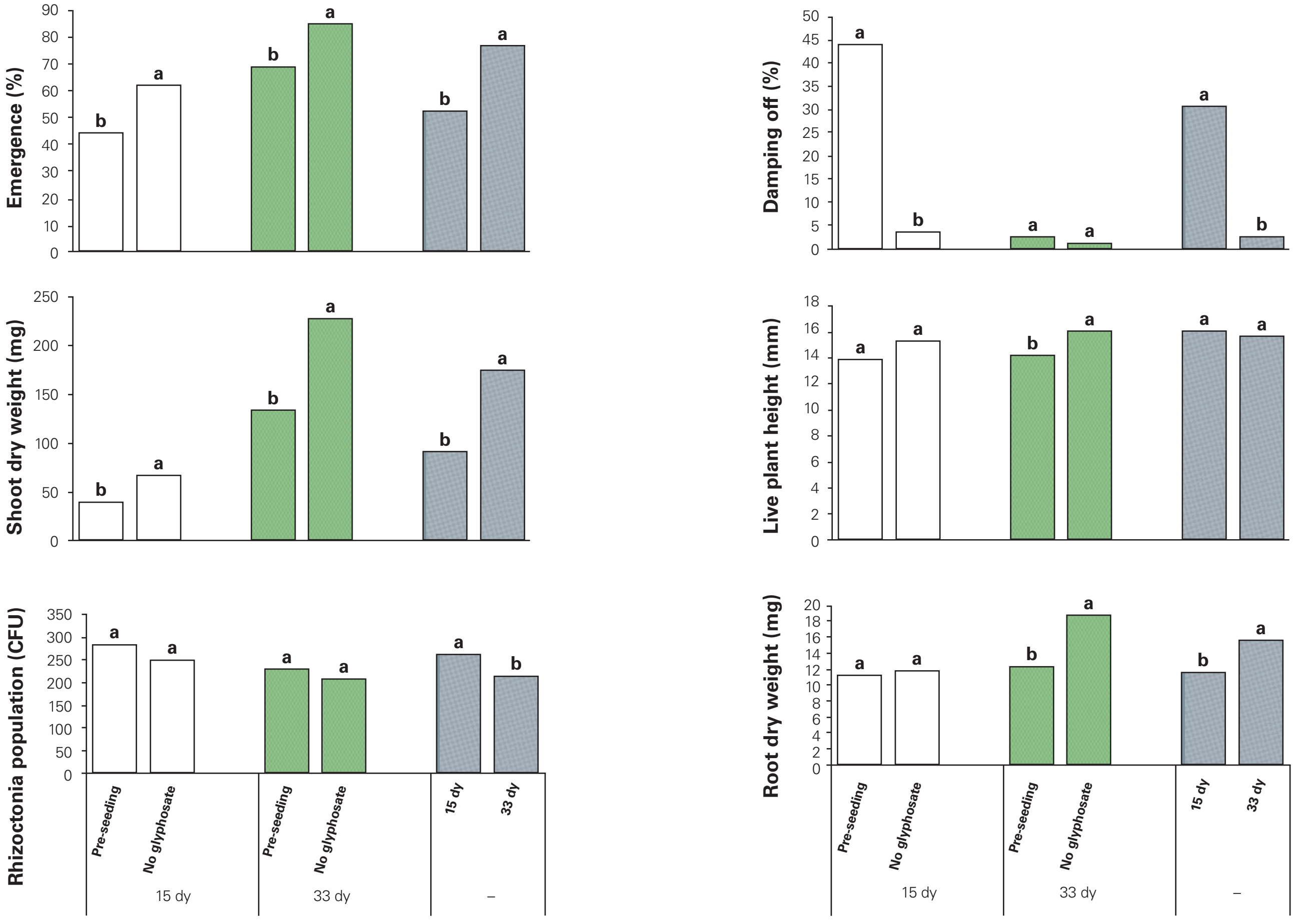

Fig. 2. Effect of timing of glyphosate application (15 versus 33 days before seeding) on seedling emergence and damping off, plant growth and biomass of canola grown in soils infested with *Rhizoctonia solani* under greenhouse conditions and on *R. solani* populations in the soil at the end of the experiment. Bars capped with the same letter within each interval date comparison do not differ at $P \leq 0.05$ based on pre-planned contrasts of treatments with pre-seeding glyphosate application vs. all other treatments that did not receive pre-seeding glyphosate application at 15 and 33 days before seeding. A contrast comparing an aggregate of all treatments at 15 vs. 33 days is included.

DISCUSSION

The results of a number of field studies indicate that application of glyphosate can pre-dispose the subsequent crop to soil-borne diseases. It has been suggested that when glyphosate acts on weeds and susceptible volunteer crop plants, the inherent defense responses of the affected plants against pathogens is compromised, and soil-borne pathogens grow rapidly on the dying roots As a result, pathogens multiply rapidly in the soil. Increasing pathogen populations in the soil can result in greater disease pressure on the subsequent crop In the current study, the impact of glyphosate application on the interaction between *R. solani* and canola seedlings was examined under field and greenhouse conditions.

Field experiments showed that glyphosate application, irrespective of timing and rate, increased plant establishment and the seed yield of a glyphosate-resistant canola cultivar in both years (Table 1, Fig. 1). Application of glyphosate effectively manages weeds and sensitive volunteer canola in the glyphosate-resistant crop, encouraging the establishment of the resistant crop by reducing interplant competition for nutrients and water. Prior inoculation with *R. solani* reduced seedling establishment in both years, and reduced seed yield in 2009 compared to the non-inoculated control (data not shown). These results indicate that as the population of *R. solani* rose with inoculum concentration, it adversely affected the emergence and the seed yield of glyphosate-resistant canola. Both of these results were expected. However, there was no interaction between glyphosate treatment and inoculation in either year, which indicates that glyphosate application did not result in a measurable change in disease pressure from *R. solani* in these trials.

To examine the relationship between application of glyphosate and subsequent seedling disease on canola in more detail, greenhouse experiments on the effects of pre-seeding interval on the incidence of seedling blight in canola were conducted using *R. solani* as the seedling blight pathogen and redroot pigweed as the target for the glyphosate application. Seedling blight, as determined by seedling emergence and damping-off of seedlings, was much more severe on plants seeded 15 days after glyphosate application than those seeded 33 days after application (Fig. 2). Adverse effects on root and shoot dry weight of the crop were also larger when canola was seeded 15 days after glyphosate application compared to 33 days after application (Fig. 2).

Together, these results suggest that glyphosate application temporarily makes glyphosate-tolerant canola more prone to infection by *R. solani*, but the effect declines quickly over time. A delay in seeding after glyphosate application may reduce disease pressure over time, but it is impractical to delay seeding for an extended period under Canadian conditions. Therefore, pre-plant applications of glyphosate should be accompanied by effective seed treatment fungicides and be preceded by a crop rotation that encourages a reduction of pathogen populations in the soil. The results of the current study are consistent with a recent report that suggests that seedling blight of barley caused by *Rhizoctonia* spp. can be reduced by increasing the time gap between pre-seeding application of glyphosate and crop seeding date

Although the number of CFUs of *R. solani* in the soil was higher in canola planted 15 days after glyphosate treatment compared to 33 days, pathogen populations did not show a significant response to pre-seeding glyphosate treatment within each seeding date. This suggests that the experimental conditions promoted the same rate of pathogen population decline over time regardless of glyphosate treatment.

In conclusion, the results of our study indicate that delaying the seeding of the crop after pre-seeding application of glyphosate can reduce the incidence of seedling blight caused by soil-borne *R. solani* in canola. While pre-seeding application of glyphosate increased the incidence of seedling blight under greenhouse conditions, it did not affect population levels of *R. solani* in the soil, nor did it affect the incidence of seedling blight in two years of field trials. These observations suggest that a precautionary delay in seeding after application of pre-seeding glyphosate, accompanied by appropriate crop rotation and fungicide seed treatments, may reduce the harmful effects of soil-borne pathogens on canola seedlings. Additional work is required to determine the nature of the interaction among glyphosate, soil-borne pathogens and susceptible weeds.

Editor's Note:
Tables 2 and 3 can be found online at www.caar.org/training/cca-examiner. Please review the table online in order to complete the exam.

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