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# Efficacy of Biological and Chemical Herbicides on Non-Native Buckthorn during Three Seasonal Periods

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**ABSTRACT:** The invasive-exotic *Rhamnus cathartica* has been growing in parks and natural areas of North America for over 100 years where it has replaced native vegetation. Chemical herbicides have limited success on *R. cathartica* and often require follow-up applications. This multiyear study is the first to investigate the efficacy of *Chondrostereum purpureum*, the active agent in Chontrol Peat Paste (CPP), as a biological herbicide for *R. cathartica*. The objective of this study was to determine the efficacy of CPP and Roundup on *R. cathartica* trees by comparing re-growth/mortality rates of mechanically wounded trees treated with either herbicide. *Rhamnus cathartica* trees were girdled or cut and received either CPP or Roundup applications in late-fall (LF), early-summer (ES), and late-summer (LS) at Assiniboine Park in Winnipeg, Canada. All trees were evaluated for mean re-growth, number, and condition of basal sprouts during spring following each application. It was expected that trees treated with CPP would show less re-growth than those that were solely mechanically wounded (controls). In LF, the most effective mechanical/herbicide combination for reducing overall stem re-growth was found to be the cut treatment followed by Roundup. In ES, however, the most effective treatment combination for suppressing re-growth was the CPP application to girdled trees as conditions were optimal for inoculation of trees. These results will allow herbicides to be effectively applied over a longer duration of the season and have implications for the development of future management protocol for *R. cathartica* in urban parks and natural areas.

*Index terms:* Assiniboine Park, biological herbicide, *Chondrostereum purpureum*, integrated pest management, *Rhamnus cathartica*

## INTRODUCTION

*Rhamnus cathartica* L. (European buckthorn) is an undesirable invasive, exotic plant species in North America with a high fecundity and prolific growth rate (Knight et al. 2007). The distribution of *R. cathartica* within the City of Winnipeg coincides with historical urban development where the species was used as hedgerows in parks and neighborhoods. Displacement of native plants occurs since *R. cathartica* possesses a longer growing season and overtakes understory species through vigorous growth (Boudreau and Wilson 1992).

An understanding of *R. cathartica* biology is crucial to recognize its weaknesses and for making informed management and restoration decisions. Mechanical extraction of *R. cathartica* has proved to be very time consuming and labor intensive when numerous *R. cathartica* are located amongst desirable vegetation. Roundup has been determined to be among the best chemical herbicides in reducing the threat of *R. cathartica* in north-eastern Illinois (Pergams and Norton 2006). Previous *R. cathartica* control measures in Assiniboine Park have included removing the trunk near the soil level and applying herbicide. However, chemical herbicides often involve repeat applications on re-sprouting shoots and may have environmental consequences possibly affecting the public perception of

natural areas.

*Chondrostereum purpureum* (Pers.) Pouzar strain PFC2139 is a naturally occurring fungal plant pathogen sold under the product label, Chontrol Peat Paste (CPP), intended for the reduction of stem density of broad-leaved plants following brush cutting (Mycologic 2009). *Chondrostereum purpureum* has no geographic or host specialization with tree mortality occurring only if infected trees are already heavily stressed or wounded (Becker et al. 2005). Differing rates of *C. purpureum* infection have been attributed to host susceptibility. Four months post-treatment, *Alnus sinuata* (Regel) Rydb. (sitka alder) and *Populus tremuloides* Michx. (trembling aspen) were shown to have infection frequencies of 90% and 40%, respectively (Becker et al. 1999), suggesting a species-specific effect. To date, no research has studied the efficacy of CPP as a herbicide for *R. cathartica* control. Furthermore, previous studies examining herbicide efficacy have largely focused testing on cut stumps while little attention has been paid to girdled trees or to trials spanning multiple seasonal periods.

The objective of this study is to determine the efficacy of CPP as a herbicide for *R. cathartica* trees by comparing re-growth/mortality rates of trees treated with no herbicide, CPP, or Roundup replicated

over three distinct seasonal periods. It is expected that *C. purpureum* will colonize the stems of *R. cathartica* and inhibit re-growth through decay. Measures of success would include: (1) higher mortality, and (2) fewer number of re-sprouts from stems inoculated with *C. purpureum* compared to stems where no herbicide was applied (controls).

## METHODS

### Mechanical and Herbicide Treatments

The study area includes three parcels of *R. cathartica* infested forest (80% – 90%) inter-dispersed with *Quercus macrocarpa* Michx. (bur oak) within Assiniboine Park, Winnipeg, Manitoba, Canada. Research notifications for application of CPP in the study areas were obtained from the Pest Management Regulatory Agency, Health Canada, for 2010 – 2011.

Timing of herbicide application is a crucial consideration when adopting a new approach to eliminating an invasive exotic species. From 2010 – 2011, herbicide trials were conducted during three distinct periods throughout the *R. cathartica* growing season: November, June, and September. These herbicide trials will be herein referred to as late-fall (LF), early-summer (ES), and late-summer (LS), respectively. A total of 364 treated *R. cathartica* trees were divided over these three seasonal periods. For each seasonal period, 20 trees were allotted for each of the four treatment combinations (mechanical treatment x herbicide application). The four combinations in this study were: girdled trees with

either CPP or Roundup applications, and cut trees with either CPP or Roundup applications. Undiluted Roundup Concentrate Grass and Weed Control with 143 g/L of glyphosate present as isopropylamine salt (Monsanto Canada 2008) was used in this study. Twenty girdled and 20 cut stems with no herbicide applied were also included to act as controls in each period.

All stems were girdled/cut at 30 cm above ground level and were > 3.5 cm in diameter at this height. To girdle trees, a Barkblaster was used to peel off a 5-cm section of bark/cambium around the stem. Immediately after the girdled/cut, a thin layer of herbicide was painted directly on to fresh stems. Each tree was identified by a unique number referring to its characteristics and randomly selected treatment. The tree identification number, mechanical/herbicide treatment, diameter at 30 cm, % stem mortality prior to treatment (proportion of cambial dieback of each stem), and number of sprouts present (if any) were recorded at the time of treatment.

### Evaluation of Treated Trees

The study area was re-visited to evaluate any re-growth present on each tree during the spring following each herbicide application. The efficacy of treatments was evaluated on a 0 – 4 re-growth rating system: 0 indicating no re-growth to 4 indicating re-growth on the entire stem (Pergams and Norton 2006). In addition, the number of basal sprouts present, if any, along with their health based on a 1 – 10 rating (10 indicating green, vigorous growth) was also recorded. The number of sprouts noted before treatment was

subtracted from that counted during the follow-up evaluation to exclude pre-existing sprouts. Whenever possible, multi-stemmed *R. cathartica* trees were avoided so that additional stems would not bias the investigation.

### Statistical Analysis

A MANOVA (multivariate analysis of variance) was employed to determine whether herbicide type, mechanical treatment, stem diameter, and % stem mortality prior to treatment had any effect on re-growth of stems or basal sprouts. Subsequent one-way ANOVAs were also conducted to explore significance within treatment combinations. All statistical analyses were performed using SPSS v.11.0.1 for Windows (SPSS 2001).

## RESULTS

### General Statistics

Mean stem diameter at 30 cm was shown to be 6.6 cm (min: 3.8, max: 13.4; n = 337). A total of 337 *R. cathartica* trees were recovered from the original 364 included in this multiyear study (Table 1). Twenty-seven *R. cathartica* trees were mistakenly harvested or injured by arborists before evaluations could be conducted reflecting the challenges of conducting research within parks.

Re-sprouting in partially dead *R. cathartica* stems (% stem mortality prior to treatment) included in this study did not significantly differ from stems that were completely alive the following year. Moreover, the

**Table 1. Final sample size of herbicide/mechanical treatment combinations applied to *R. cathartica* trees throughout late-fall 2010, early-summer 2011, and late-summer 2011. Assiniboine Park, Winnipeg, Canada.**

Treatment	Girdled Stems			Cut Stems		
	Late Fall	Early Summer	Late Summer	Late Fall	Early Summer	Late Summer
No Herbicide	18	16	20	19	20	19
Chontrol Peat Paste	21	19	20	18	20	18
Roundup	15	19	20	16	19	20

mechanical treatment and stem diameter effects were not significant during any seasonal period. An exception was in LF where the stem diameter effect achieved significance (F-ratio: 1.728,  $p < 0.05$ ,  $n = 107$ ) but lacked replication in the higher diameter classes.

### Late-Fall (November) herbicide application

The MANOVA indicated that the herbicide effect was significant (F-ratio: 5.977,  $p < 0.001$ ,  $n = 107$ ). An ANOVA subsequently conducted on herbicide with exclusively girdled trees was significant (re-growth rating: F-ratio: 8.780,  $p < 0.005$ ,  $n = 54$ ; # basal sprouts: F-ratio: 5.621,  $p < 0.05$ ,  $n = 54$ ; condition basal sprouts: F-ratio: 14.058,  $p < 0.001$ ,  $n = 54$ ) as was that conducted on herbicide with exclusively cut trees (re-growth rating: F-ratio: 38.869,  $p < 0.001$ ,  $n = 53$ ; # basal sprouts: F-ratio: 16.644,  $p < 0.001$ ,  $n = 53$ ; condition basal sprouts: F-ratio: 43.384,  $p < 0.001$ ,  $n = 53$ ; see Figure 1). In LF, suppression of all re-growth variables was best shown for cut *R. cathartica* trees treated with Roundup (Figure 1). This mechanical/herbicide combination resulted in zero re-growth for 14/16 stems (88% mortality) while a high level of re-growth was observed for both girdled/cut trees with no herbicide and those treated with CPP at the time of evaluation.

### Early-Summer (June) herbicide application

The MANOVA indicated that the herbicide effect was significant (F = 3.639,  $p < 0.01$ ,  $n = 113$ ). An ANOVA subsequently conducted on herbicide with exclusively girdled trees was significant (re-growth rating: F = 19.823,  $p < 0.001$ ,  $n = 54$ ; # basal sprouts: F = 8.240,  $p < 0.01$ ,  $n = 54$ ; condition basal sprouts: F = 20.648,  $p < 0.001$ ,  $n = 54$ ; Figure 2). Compared to girdled trees, the application of herbicides to cut trees was not effective in reducing re-growth. The ANOVA conducted on herbicide with exclusively cut trees was not significant for re-growth rating (F = 2.084,  $p = 0.134$ ,  $n = 59$ ) or # basal sprouts (F = 0.138,  $p = 0.872$ ,  $n = 59$ ) but was significant for condition basal sprouts (F = 3.805,  $p < 0.05$ ,  $n = 59$ ; Figure 2).

In ES, girdled trees followed by application of CPP was shown to yield the least re-growth (Figure 2) with 17/19 stems in this category showing zero re-growth (90% mortality). However, no significant difference in re-growth was observed between girdled trees with no herbicide applied and those treated with Roundup.

### Late-Summer (September) herbicide application

The MANOVA indicated that both the herbicide effect (F = 2.794,  $p < 0.05$ ,  $n = 117$ ) and the mechanical treatment-stem diameter interaction (F = 1.697,  $p < 0.05$ ,  $n = 117$ ) were significant. An ANOVA subsequently conducted on herbicide with exclusively girdled trees was significant for re-growth rating (F = 4.556,  $p < 0.05$ ,  $n = 60$ ) but not significant for # basal sprouts (F = 0.281,  $p = 0.756$ ,  $n = 60$ ) or condition basal sprouts (F = 2.093,  $p = 0.133$ ,  $n = 60$ ). In addition, the ANOVA conducted on herbicide with exclusively cut trees was significant (re-growth rating: F = 4.417,  $p < 0.05$ ,  $n = 57$ ; condition basal sprouts: F = 8.225,  $p < 0.01$ ,  $n = 57$ ) with the exception of # basal sprouts (F = 1.182,  $p = 0.314$ ,  $n = 57$ ). In LS, there was no significant difference between CPP and Roundup among any of the re-growth variables (Figure 3). Instead, significant differences were a result of trees with no herbicide applied showing the highest level of re-growth.

## DISCUSSION

### Efficacy of glyphosate

Many noxious weeds are successfully controlled by chemical herbicides. Pergams and Norton (2006) tested five common chemical herbicides in October – November and found Roundup to be the most effective for *R. cathartica*. In our study, Roundup was only shown to be effective during LF in reducing re-growth but not in ES or LS. *Rhamnus cathartica* trees are still growing in ES or LS; whereas, the general downward storage of nutrients during LF could assist Roundup in reaching the root system. Chemical herbicides applied to *R. cathartica* will usually not affect non-target plants as they will have entered dormancy (Boudreau and Wilson 1992). In LF, Roundup consistently held

the lowest mean re-growth and sprout rating relative to either CPP or the no herbicide groups.

Interestingly, less re-growth was shown in cut trees with Roundup applied than for the same herbicide applied to girdled trees. However, Pergams and Norton (2006) noted that Roundup yielded a consistently low level of re-growth regardless of the mechanical intervention applied beforehand. These contrasting results could be related to differences in chemical absorption from environmental conditions/stand differences. The transport of glyphosate, the active chemical in Roundup, depends on the amount initially introduced into the plant (Monsanto Canada 2008). Transport of Roundup generally slows after 4 hours from application and stops after 48 hours (Monsanto Canada 2008). In LF, it is hypothesized that Roundup may be better absorbed into the plant from a horizontal surface (cut stem), which could provide a longer contact period with the herbicide compared to a vertical surface, as on a girdled stem, where the herbicide may be more susceptible to runoff.

### Efficacy of *Chondrostereum purpureum*

*Chondrostereum purpureum* has been found to reduce the number, vigor and re-growth of cut hardwood stems in Canada (Wall 1990). This is the first study to date that has investigated the efficacy of *C. purpureum* on *R. cathartica* as a mycoherbicide. The biological herbicide, CPP, was most effective during ES but was seemingly ineffective during LF and LS. In LF and LS, re-growth variables for trees treated with CPP were not significantly different than those of trees where no herbicide was applied.

Applications during these seasonal periods may not have been applied early enough for *C. purpureum* to successfully establish in the Canadian Prairies. However, weather conditions also influence the establishment and virulence of *C. purpureum* (Becker et al. 2005). During LF applications, freezing night-time temperatures were likely too cold for CPP inoculation of *R. cathartica* stems. Inversely, temperatures exceeding 30 °C during LS applications may have been too hot. The mean maximum temperature during successful field applications

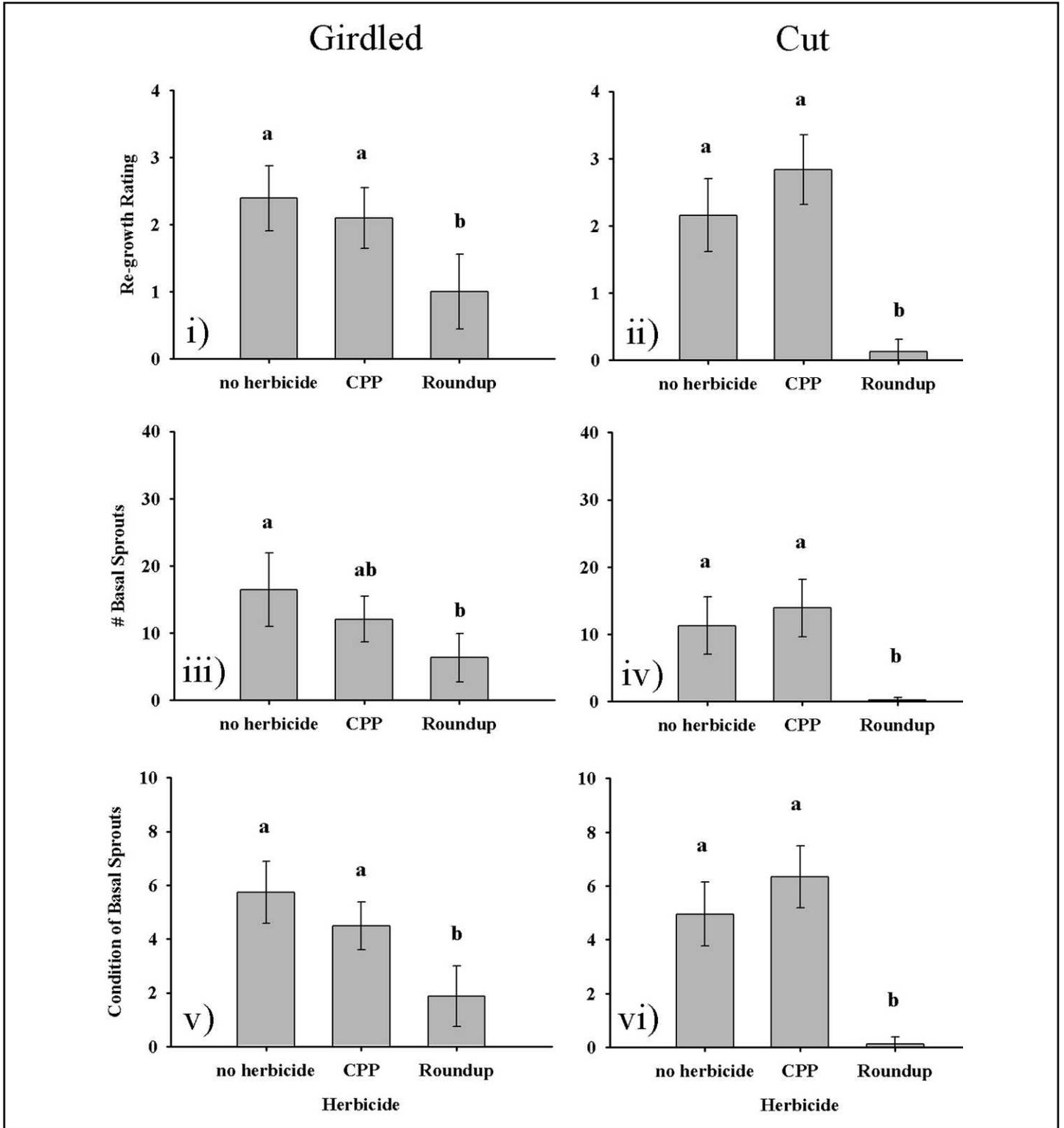


Figure 1. Late-fall 2010: mean re-growth rating, # basal sprouts, and condition of basal sprouts present in each herbicide category (no herbicide, *Chondrostereum purpureum* (CPP) and Roundup) for girdled (left column; i, iii, v) and cut trees (right column; ii, iv, vi). The 95% confidence intervals are shown for each mean value (n = 107). Factors that are not significantly different at  $p < 0.05$  from each other will share the same lower case letter denoted above. Assiniboine Park, Winnipeg, Canada.

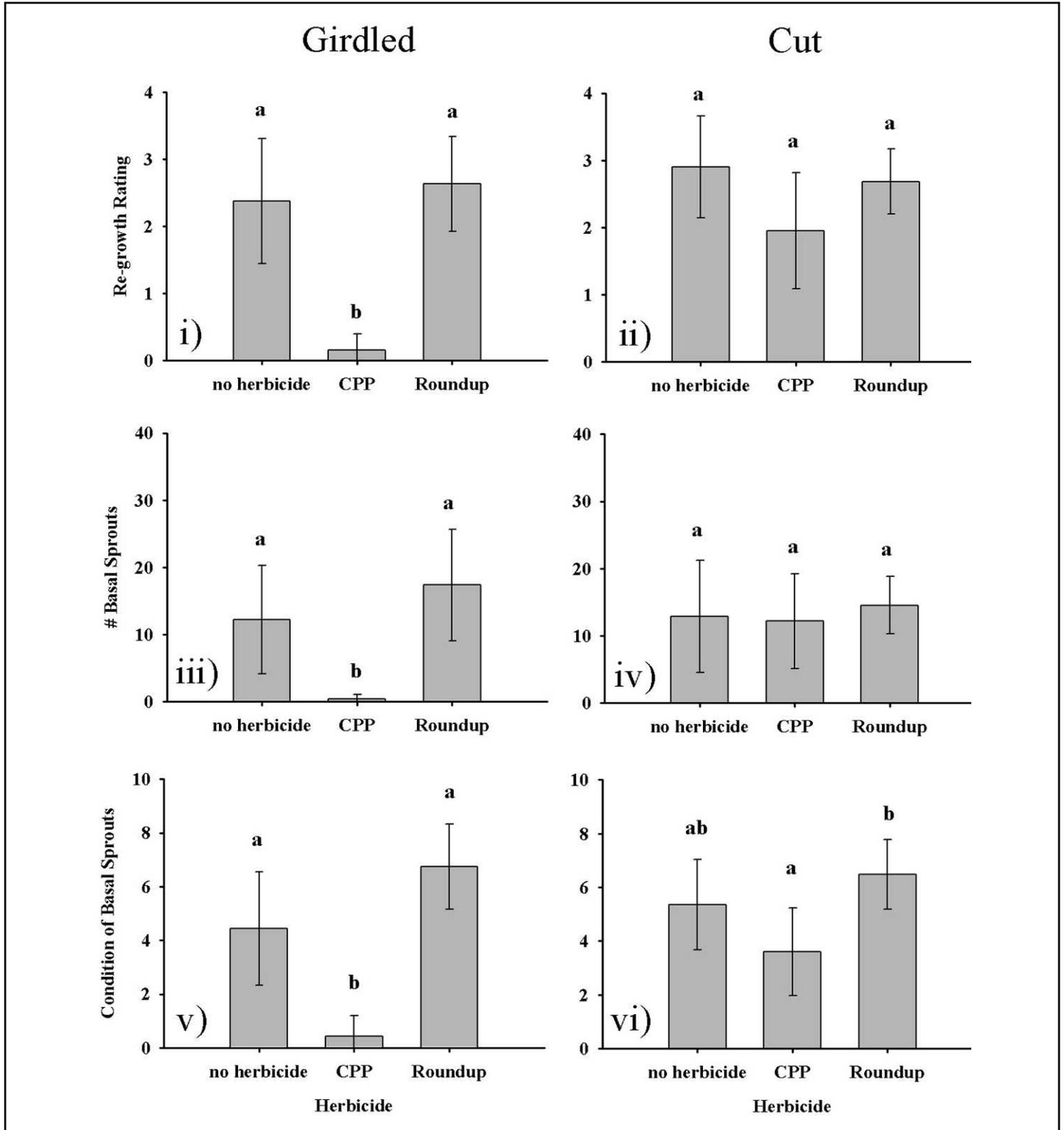


Figure 2. Early-summer 2011: mean re-growth rating, # basal sprouts, and condition of basal sprouts present in each herbicide category (no herbicide, *Chondrostereum purpureum* (CPP) and Roundup) for girdled (left column; i, iii, v) and cut trees (right column; ii, iv, vi). The 95% confidence intervals are shown for each mean value (n = 113). Factors that are not significantly different at  $p < 0.05$  from each other will share the same lower case letter denoted above. Assiniboine Park, Winnipeg, Canada.

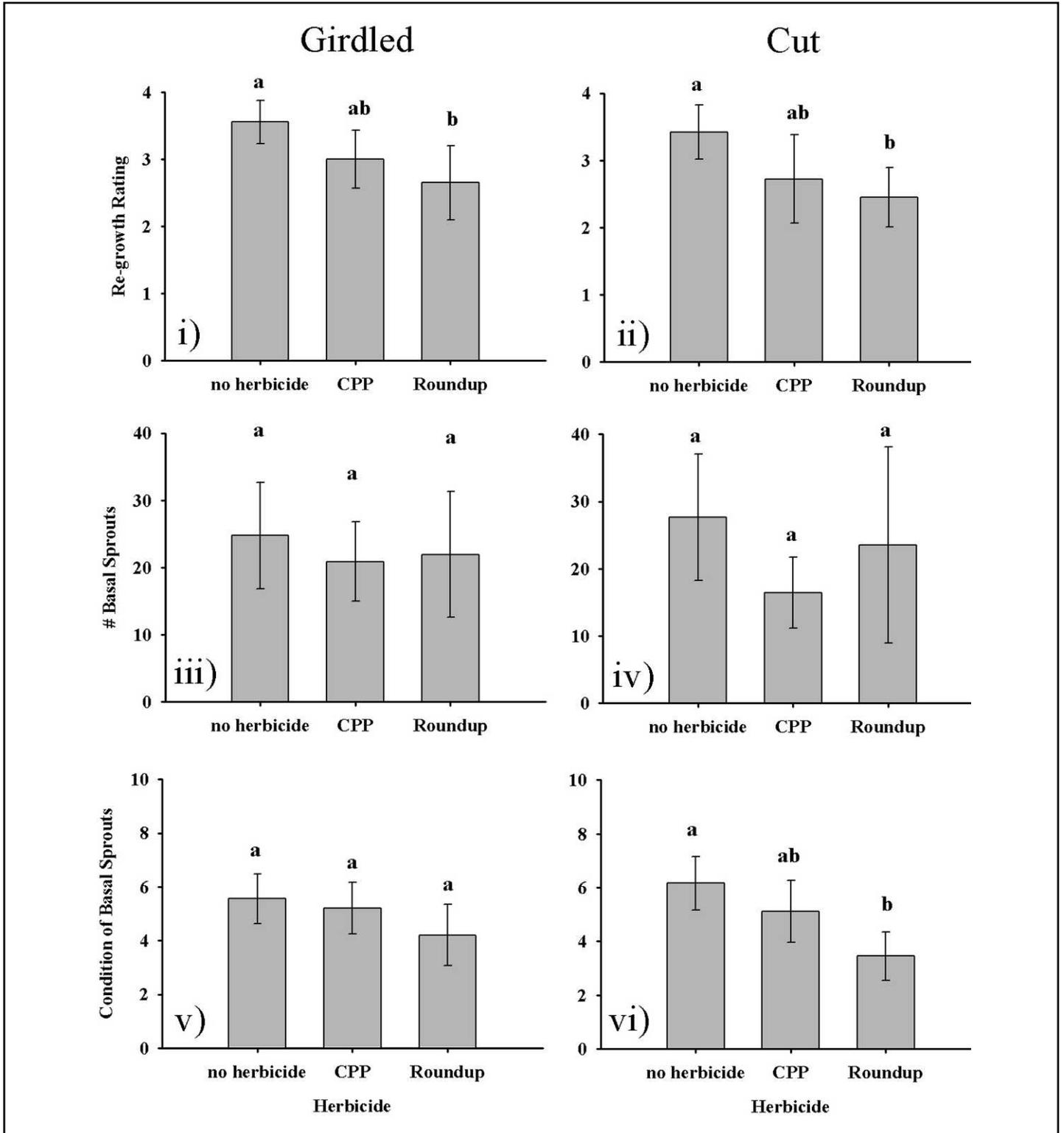


Figure 3. Late-summer 2011: mean re-growth rating, # basal sprouts, and condition of basal sprouts present in each herbicide category (no herbicide, *Chondrostereum purpureum* (CPP) and Roundup) for girdled (left column; i, iii, v) and cut trees (right column; ii, iv, vi). The 95% confidence intervals are shown for each mean value (n = 117). Factors that are not significantly different at  $p < 0.05$  from each other will share the same lower case letter denoted above. Assiniboine Park, Winnipeg, Canada.

of CPP was 27 °C (Becker et al. 2005) and 24 °C during ES in this study. Warm temperatures (i.e., 15 °C – 25 °C) and a relatively long period for establishment prior to freezing temperatures are likely advantageous for *C. purpureum* survival and successful control of re-growth.

The measures of success for CPP were met for applications conducted in ES where CPP was applied to girdled trees resulting in 90% mortality. In contrast, cut stems with CPP applied showed a similar level of re-growth to stems with no herbicide treatment. Fungal inoculation and growth may be more favorable on girdled trees than on the more exposed surfaces of cut trees whose canopies have been completely removed. Girdled CPP-applied trees could also have a greater efficacy than cut CPP applied trees since girdling creates a greater surface area for the fungus to initially establish itself. Holmer and Stenlid (1993) studied competition among wood decay fungi by growing them on various areas of wood discs. Their findings suggest that larger mycelial size resulted in greater competitive ability and colonizing success. Performance of *C. purpureum* on *R. cathartica* trees may, therefore, be related to the size of the wound where CPP is initially applied.

In this study, cut *R. cathartica* trees did not provide a suitable medium for *C. purpureum* to successfully inhibit re-growth one-year following treatment. It has been shown, however, that the mortality of trees inoculated with *C. purpureum* increased from 92% – 100% during the second year following treatment (Becker et al. 2005). It is possible that cut *R. cathartica* stems could require a longer *C. purpureum* colonization period before inhibition of re-growth is detected.

## CONCLUSION

As integrated pest management becomes more central in our focus to remove noxious weeds, so does the need for a greater number of alternative control measures. In this first study investigating the efficacy of *C. purpureum* on *R. cathartica*, the inoculum was shown to be as effective as

Roundup in the inhibition of re-growth and mortality of treated stems, albeit in different seasonal periods. The optimal seasonal window of herbicide efficacy was shown to be LF for Roundup and ES for CPP. Importantly, these results allow herbicides to be effectively applied to *R. cathartica* over a longer duration of the growing season. Moreover, the mechanical treatment/herbicide combination was crucial in herbicide efficacy. Cut trees followed by Roundup application resulted in 88% mortality while girdled trees followed by application of CPP resulted in 90% mortality of treated stems in LF and ES, respectively. Interestingly, CPP applied to cut trees was shown to be ineffective in reducing re-growth. Subsequent evaluations are required to determine whether *C. purpureum* requires a longer establishment period to effectively inhibit re-sprouts on cut trees and also whether the mortality of girdled trees increases further.

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