

Final Report: CIPM Seed Money Grant Program

Project Title: Impacts of integrated pest management strategies for sustainable Canada thistle control.

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Proposal Abstract: Canada thistle is an aggressive perennial forb that is a serious pest in agronomic, range, pasture, and wild lands throughout the United States. This plant is very difficult to control due to extensive root energy reserves and its regenerative ability from adventitious buds on horizontal and vertical roots. Although previous studies have indicated biocontrol agents released for Canada thistle have not been highly effective when used alone, there is considerable interest in understanding how to best integrate biological control with other management strategies such as herbicides. Previous research on biological control of Canada thistle has found that the Canada thistle stem mining weevil (*Ceutorhynchus litura*) may negatively impact root energy reserves in the early and mid summer. However, the overall impact is limited in many areas due to the plants ability to compensate for early summer damage over the late summer and fall. The primary objective of this research is develop integrated management strategies that target Canada thistle directly following maximum stem weevil damage before the plants are able to compensate. The central hypothesis is that herbicide treatment timed directly following maximum biocontrol damage will result in improved control of Canada thistle.

Results: In 2004, above-average early spring temperatures resulted in earlier than normal emergence of *C. litura* from collection sites near Bozeman, Montana. This resulted in an asynchrony between *C. litura* emergence at the collection site and Canada thistle emergence at the Cheyenne research site. By the time Canada thistle rosettes were emerged in Cheyenne, there was a lack of available biocontrol agents for our study from the Montana supplier. Therefore, we delayed the initiation of the experiment to the spring of 2005. In the spring of 2005, agents were collected in early May near Bozeman and were released (~50 per plot) on May 13, 2005 at the Cheyenne site. This timing was still generally asynchronous as Canada thistle emergence was just beginning. However, we proceeded with the experiment. While *C. litura* are not considered extremely mobile, we did not feel it necessary to enclose each individual plot. However, this was a mistake as the bioagents unexpectedly dispersed over the entire study area. Our fall sampling indicated this as the bioagent damage was observed across the study site, infesting 42.8 % of the Canada thistle stems in each treatment. Analysis of variance indicated no significant differences in infested stems between any of the treatments ($p=0.94$). Additionally, mining length (measured length of the stem mine created by *C. litura*) averaged 70.5 mm and there were no differences between treatments ($p=0.88$). When non-infested stems were removed from the analysis, there were still no differences observed in mining length ($p=0.91$).

Clopyralid was applied in the late bolting stage (June 24, 2005). Just prior to application plant vegetative cover data was taken from each plot. There were 24 total species identified across the study area with the majority of the native species richness occurring as grasses and grasslike plants (Table 1). The majority of the forbs were exotic (Table 1). Clopyralid application had no effect on total, native, or exotic species richness by the fall plant vegetative sampling date ($p>0.65$ for all three). Additionally, Simpson's index of diversity was calculated for each treatment. However, there were no significant differences in diversity observed ($p=0.1467$). In terms of plant cover, Canada thistle cover was significantly reduced by clopyralid ($p<0.001$) as expected. However, total native grass cover was not significantly different ($p=0.11$) and averaged 69% between spring treated and untreated plots. Both native and exotic forb cover (other than Canada thistle) also did not respond either positively or negatively to the clopyralid application (Native forb cover: 5.1%, $p=0.45$; Exotic forb cover: 2.9%, $p=0.91$).

Due to the problems associated with the asynchronous emergence of the biocontrol agent and the Canada thistle and the subsequent dispersion of the bioagent across all of the plots, it is impossible to provide an answer for objective #1 (Determine the impacts of integrating the Canada thistle stem mining weevil and clopyralid treatment on Canada thistle shoot dynamics) from this study. However, infestation levels (42% of the stems) may have not been high enough anyway as it has been previously proposed that very high numbers of *C. litura* may be needed to provide any significant impact. In terms of objective #2 (Determine the impact of clopyralid application timing on non-target desirable vegetation), we interpret our data to conclude that clopyralid had neither a positive nor a negative impact on non-target desirable native grasses and forbs within the season of application. However, the caveat here is that there were unfortunately few forbs within our study site that are known to be sensitive to the rate of clopyralid used in this study (0.28 kg ae/ha). We do believe that the site we selected is representative of many true rangeland (unplowed) sites infested with Canada thistle across Wyoming. These are typically characterized by high Canada thistle cover with an understory mix of native and exotic grasses and few native forbs. In many of these areas, the nontarget risk of clopyralid should be low. In terms of Objective #3 (Determine the role of clopyralid application timing on the establishment and survival of the Canada thistle stem mining weevil), our data showed no negative impact on infestation levels (% of stems infested) or on mining length within infested stems. It appears that by the late bolting stage of Canada thistle, the weevils have completed almost all stem mining activity and have exited the stems. While earlier clopyralid applications (Canada thistle rosette stage) may be problematic, late bolting to early bud applications appear to be safe for optimizing *C. litura* feeding damage.

Long Term Goal and Continued Progress of Research: Given our difficulties in getting *C. litura* properly established to effectively test for impacts of integrating the bioagent with herbicides in the field, we developed a backup plan using a greenhouse approach to address objective #1. In late April, we planted single Canada thistle root segments (15 cm) in forty PVC columns (15 cm diameter by 38 cm high), filled with a greenhouse potting mix. New shoots emerged within 10 days and on May 13, we released three *C. litura* bioagents onto each of 20 randomly selected planted columns. To keep the bioagents on the correct columns, we caged all forty columns individually with a fine mesh screen for two weeks. At the end of two weeks, we removed the cages and aspirated off every living *C. litura* that could be detected. The plants were allowed to grow until the early bud stage at which time one of three herbicide treatments was applied with a chamber sprayer at 187 l/ha. These were clopyralid (0.28 kg ae/ha), glyphosate (2.5 kg ae/ha), or 2,4-D (2.2 kg ae/ha). Plants were then allowed to regrow for 90 days. After 90 days, the thistle shoots and roots were harvested and oven dried for 72 h at 60 C and weighed. Each herbicide/bioagent combination had five replicate columns and the columns were arranged in a completely randomized design in the greenhouse. Analysis of variance was used to test the effects of herbicide (clopyralid, glyphosate, or 2,4-D), bioagent (*C. litura* or none) and the interaction herbicide by bioagent on root dry weights, new shoot dry weights, and total shoot number. For root dry weights, both herbicide ($p < 0.001$) and *Ceutorhynchus litura* ($p < 0.001$) were highly significant but the interaction of herbicide by *C. litura* was not significant ($p = 0.85$) (Figure 1). Mean separations of the herbicide main effect indicated significant differences among treatments with clopyralid providing the greatest reduction in root dry weights (Table 2). Results were similar for both shoot weight and shoot number. While the interaction was not significant, these additional greenhouse data are promising in terms of future experiments to examine

potential additive and synergistic effects of combining biocontrol with herbicides for Canada thistle control.

Publications: Due to the problems associated with the field experiment, we have not currently generated any peer reviewed publications from this work. However, we will repeat the greenhouse study before proceeding to submit an article for publication.

Benefits of Seed Money: We have taken this idea (integrating biological control with herbicides) and applied it to another serious invasive plant Dalmatian toadflax. The Dalmatian toadflax project has been funded (2005-2007) by the Western Region IPM Competitive Grants Program for \$95,000. We will apply everything we have learned from the Canada thistle project as we go forward with this new research project. We are very grateful to CIPM for providing the seed money for the initial idea.

Table 1. Species list of all vascular plants identified in the plots.

Common Name	Scientific Name	Life form	Native/Exotic
Canada thistle	<i>Cirsium arvense</i>	Forb	E
Flixweed	<i>Descurania Sophia</i>	Forb	E
Dalmatian toadflax	<i>Linaria dalmatica</i>	Forb	E
Field pennycress	<i>Thlaspi arvense</i>	Forb	E
Buckhorn plantain	<i>Plantago coronopus</i>	Forb	E
Russian thistle	<i>Salsola iberica</i>	Forb	E
Houndstongue	<i>Cynoglossum officinale</i>	Forb	E
Scarlet globemallow	<i>Sphaeralcea coccinea</i>	Forb	N
Fringed sage	<i>Artemisia frigida</i>	Forb	N
Western yarrow	<i>Achillea millefolium</i>	Forb	N
Wild licorice	<i>Galium lanceolatum</i>	Forb	N
Kentucky bluegrass	<i>Poa pratensis</i>	Grass	E
Japanese brome	<i>Bromus japonicus</i>	Grass	E
Downy brome	<i>Bromus tectorum</i>	Grass	E
Western wheatgrass	<i>Pascopyrum smithii</i>	Grass	N
Slender wheatgrass	<i>Elymus trachycaulus</i>	Grass	N
Inland saltgrass	<i>Distichlis spicata</i>	Grass	N
Blue grama	<i>Bouteloua gracilis</i>	Grass	N
Ring muhly	<i>Muhlenbergia torreyi</i>	Grass	N
Needle and thread grass	<i>Hesperostipa comata</i>	Grass	N
Sand dropseed	<i>Sporobolus cryptandrus</i>	Grass	N
Baltic rush	<i>Juncus balticus</i>	Rush	N
Scouring rush	<i>Equisetum arvense</i>	Rush	N
Threadleaf sedge	<i>Carex filifolia</i>	Sedge	N

Table 2. Mean separation of herbicide main effects on Canada thistle root dry weights.

Herbicide	Transformed root dry weight (g)
None	2.25 a
2,4-D	1.73 ab
Glyphosate	1.57 b
Clopyralid	0.95 c

Figure 1. Impacts of integrating herbicides with the biological control agent, *Ceutorhynchus litura* on Canada thistle root dry weights (means + standard deviation).

