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BIOLOGICAL CONTROL OF CANADA THISTLE *(CIRSIUM ARVENSE)* IN SOUTH DAKOTA

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ABSTRACT

Canada thistle, *Cirsium arvense L.,* is an aggressive invasive species that is found in the northern United States and not easily controlled by chemical and mechanical methods. Integrating biological control agents with other methods may improve control. This study investigated the effects of Canada thistle gall fly (*Urophora cardui* Diptera: Tephritidae); Canada thistle stem or crown weevil (*Ceutorhynchus litura* Coleoptera: Curculionidae); Canada thistle bud weevil (*Larinus planus* Coleoptera: Curculionidae); and Canada thistle tortoise beetle (*Cassida rubiginosa* Coleoptera: Chysomelidae) on Canada thistle at three SD sites 1997 and 1998, about three years after initial release. Active stem weevil larvae were found in all release sites early in the season (May, June). Bud weevils were found in a limited number of plants in July. Galls and gall fly larvae were found late in the season. Defoliation from the tortoise beetle was not noted at the one site of release. Total nonstructural carbohydrate (TNC) contents of roots were 50% lower in roots from release sites compared to TNC levels in roots from control sites at two locations in 1998. Since Canada thistle infestations rely on roots for survival, reduction of root TNC may be an important mechanism to reduce patch vigor, and ultimately, Canada thistle's importance in the SD landscape.

Keywords

Canada thistle gall fly, *Urophora cardui,* Canada thistle stem weevil, *Ceutorhynchus litura,* Canada thistle bud weevil, *Larinus planus,* Canada thistle tortoise beetle, *Cassida rubiginosa*

INTRODUCTION

Canada thistle, *Cirsium arvense L.,* is a perennial weed that colonizes disturbed and undisturbed sites with rapid growth occurring through production of multiple shoots from lateral roots. Chemical and mechanical methods used to control Canada thistle have been moderately successful. Integrating other management techniques with these more traditional control methods may improve Canada thistle control. Nichols (1993) reported that integrated use of biological and chemical control reduced Canada thistle infestations by 90% in 10 yrs in eastern Colorado.

Biological control of weeds is the use of living organisms (insects, pathogens, bacteria, and other organisms) for control of a certain weed. The weeds chosen for biological control are usually non-native in origin and one that has natural predators in it's native home range. Before introducing the predator into the new environment, it must be screened to ensure that it will not attack desirable plants in the new area. In order to be successful, the control agent must establish in the new environment and then attack the weed with enough vigor to do harm to the weed. Therefore, the control agent must already be adapted or adaptive to the new environment in order to be successful. Success in a biocontrol project is defined as: 1) having the control agent continuing to perpetuate itself in the new environment; and 2) reducing the weed population to levels below an economic threshold. Complete eradication of weed will not occur because the control agent will not eat itself out of "house and home".

There are several positive effects of using biological control agents for Canada thistle management. These positive effects include: 1) biocontrol agents that are approved for release are selective and will not move to economically important crops; 2) the agents can infest plants in inaccessible areas; 3) once established in the habitat, the control agents are self-perpetuating and may migrate to other Canada thistle infestations; and 4) once established, the method is cost effective, less expensive, and less labor intensive than chemical and mechanical methods. The ultimate goal of Canada thistle biocontrol is to reduce plant vigor and its dominance in the landscape.

Several Canada thistle biological control agents have been approved for release in SD by USDA-APHIS-PPQ within the past decade. The first insect, Canada thistle gall fly (*Urophora cardui* Diptera: Tephritidae), was released in 1993 at Custer State Park (Helbig, USDA-APHIS-PPQ, Pierre, SD personnel communication). Other biological control agents approved for release are: the Canada thistle stem or crown weevil (*Ceutorhynchus litura* Coleoptera: Curculionidae) and the Canada thistle bud weevil (*Larinus planus* Coleoptera: Curculionidae), released in 1994; and the Canada thistle tortoise beetle (*Cassida rubiginosa* Coleoptera: Chysomelidae), released in 1995. Release sites are now located from the Black Hills to eastern SD with numbers released at one time ranging from 100 to 20,000. In order to assess the impact of the insect on Canada thistle the biology of the insect must be understood. Each one of these insects attacks the Canada thistle plant in a unique way.

Canada thistle gall fly *(Urophora cardui)*

This insect was the first biological control agent to be released on Canada thistle in SD in 1993. About 51,000 units of the Canada thistle gall fly have been introduced into SD (Helbig, personnel communication). Most of these insects were released in 1994 (40,000 units) with lesser amounts in 1995 (7,500 units) and 1993 (3,000 units).

The adult thistle gall fly lays eggs in the stem of the Canada thistle plant. As the larvae develop, a marble to walnut size gall forms in the stem during the summer. The gall becomes a nutrient sink, reducing the plant's vigor. As the vigor decreases, the Canada thistle plant becomes less competitive and less able to resist pathogens and other insect infestations. In addition, plants with galls often produce fewer flowers and seeds. Physiological stress from stem gall formation was greatest in young Canada thistle plants (Forsyth, 1985) and during the growth phase of gall development (Lalonde, 1985). In addition, gall formation on the main shoot results in stunted plants and reduced flowering (Forsyth and Watson, 1985).

The adult gall fly has a distinctive black W-shaped markings on its wings. This fly does best in dense stands of Canada thistle, where water is available. It seems to prefer semi-shaded areas to full sun. Areas subject to disturbance (mowing, grazing, chemical treatment) are not conducive to gall fly survival. This species also has difficulty establishing in areas with cold, open winters and hot, dry summers (Rees et al., 1995).

Canada thistle stem weevil *(Ceutorhynchus litura)*

About 37,000 units of the Canada thistle stem weevil have been released in SD since 1994 according to USDA-APHIS-PPQ records. The Canada thistle stem weevil is a native insect of Europe. Adults feed on stem and leaf tissue and can be found on foliage starting in August and overwinter in soil. The adult weevil has a pronounced white T-shaped marking on the back.

Adult feeding is not a major source of damage. Eggs are laid in shoots of plant that are less than 5 cm tall. Developing larvae eat the vascular cambium of the stem tissue during spring and early summer which causes mining damage. Once the larvae exit the stem, other organisms (nematodes, pathogens) enter the stem causing secondary damage, which is the damage that causes decline of plant infestations. Stem weevils effectively reduce overwinter survival of Canada thistle, as well as reduce shoot production in the spring (Rees, 1990).

Rotting of root tissues reduces shoot production the following spring. Favorable habitats include dry areas where Canada thistle is in high densities and undisturbed by grazing, mowing, or flooding.

Canada thistle bud weevil *(Larinus planus)*

About 25,000 Canada thistle bud weevils have been released in SD since 1994. Originally, this insect was accidentally introduced into the northwestern states of the U.S. but has shown some promise in Canada thistle control. The larval stage is the most destructive stage of the weevils lifecycle, although high numbers of adults can also cause plant damage on the upper leaves and buds. The adults lay eggs in unopened buds. As larvae develop, they feed on the bud tissues and developing seed, reducing seed production. Unfortunately, most plants regrow from vegetative shoots that are not attacked by this insect. The effectiveness of this insect is questioned because it only attacks terminal buds, leaving late developing buds uninfested. Augmentation with different biocontrol agents has been recommended.

Tortoise beetles *(Cassida rubiginosa)*

About 10,000 tortoise beetles have been introduced into SD since 1995. Not much work has been done on the Canada thistle tortoise beetle. Even though both adults and larvae feed on plant leaves, the adult is more destructive than earlier insect stages (Ward and Pienkowski, 1978). If this insect is in high enough densities, constant defoliation will reduce plant vigor (Forsyth, 1985; Ang et al., 1994).

Unlike chemical control, where injury can be seen within weeks after application, injury from biological agents may not be observed for months or years after release. To determine if the control agents are reducing Canada thistle vigor, assessment is needed. This study examined the effect of Canada thistle biocontrol agents on Canada thistle growth and root carbohydrate content at three release sites in eastern SD. Biocontrol agents had been released from 1 to 5 years prior to the beginning of the study.

MATERIALS AND METHODS

Experiment sites and insects

This experiment was conducted at three release sites in eastern SD (Table 1). Site 1 was located near Clear Lake, SD. Two locations near the Clear Lake release site were chosen as control sites where biocontrol agents had not been released, and a third location had the Canada thistle stem weevil released. Site 2 was located at Lake Louise State Park near Miller, SD. A control area where

Table 1. Canada thistle biocontrol species introduced to study sites in South Dakota.

SITE	SPECIES	YEAR RELEASED
Deuel County		
Clear Lake	Canada thistle stem weevil *(Ceutorhynchus litura)*	1995
Hand County		
Lake Louise	Canada thistle stem weevil *(Ceutorhynchus litura)*	1995, 1996
	Canada thistle bud weevil *(Larinus planus)*	1995,1996
	Thistle stem gall fly *(Urophora cardui)*	1995
	Tortoise beetle *(Cassida rubiginosa)*	1996
Charles Mix County		
Lake Andes NWR	Canada thistle stem weevil *(Ceutorhynchus litura)*	since 1994
Chytka	Canada thistle bud weevil *(Larinus planus)*	since 1994
	Thistle stem gall fly *(Urophora cardui)*	

no insects were released was chosen and a second area was chosen where four bioagents, the Canada thistle stem weevil, thistle stem gall fly, Canada thistle bud weevil, and tortoise beetle had been released. Site 3 was located near Lake Andes, SD. A control location was chosen and 2 release locations were monitored. One release location was near Lake Andes in the Lake Andes National Wildlife Refugee (LANWR) and the second was on a private landowner's acreage (Chytka). The biocontrol agents released at these sites were the Canada thistle stem weevil, thistle stem gall fly, and bud weevil.

Sample collection

About 30 Canada thistle stem samples were collected from each site three to four times between May and October in 1997 and 1998. When stem samples were collected stem height, leaf condition, stem-mining damage, number of galls, and number of larvae present in stem or gall or both were determined. Stem mining was quantified by splitting the stem in half and measuring the length of the mining tunnel. Five plant roots were selected from each site and analyzed for total nonstructural carbohydrate (TNC) content.

Carbohydrates analysis

Canada thistle root samples were cleaned, air dried, cut into thin slices, and dried at 70°C. The dry sample was ground with mortar and pestle to pass through a 40-mesh screen.

The total nonstructural carbohydrate content (TNC) in 0.1 g of root material was removed using a modified Weinmann method: the samples were digested by boiling with water for 5 min., cooled to room temperature, pH buffer and mylase enzyme solutions were added, and incubated at 40°C for 24 h. The solution was filtered through a Whatman #5 filter paper, and analyzed using Shaeffer-Somogyl copper-iodometric titration (Smith, 1981).

RESULTS AND DISCUSSION

Clear Lake

Canada thistle plant height. Canada thistle plants at the control site were taller than plants at the release site in May and September 1997 (Table 2). Height differences were not observed in July 1997 (Table 2). However, Canada thistle plants at the control site were shorter than plants at the release site in June and July, 1998 (Table 2). Height differences between control and release sites most likely are due to environmental effects. The release site was located on a toeslope, close to Clear Lake, where it was cool and moist compared to the control area that was in a summit position. Insect damage may also have contributed to the plant height with injured plants elongating to a greater extent than the noninjured plants in the control area.

Canada thistle stem damage. At Clear Lake, stem damage (defined as length of stem that was mined by insect larvae) was first observed in May,

Table 2. Canada thistle stem weevil damage at Clear Lake in 1997 and 1998. Data reported in the table include the number and percentage of damaged plants, the length of the stem damage and the percentage of stem that was damaged and the number of stem weevil larvae found in all plants and the average number of larvae per plant, and the total nonstructural carbohydrate content of roots from the control and release sites. The control site did not have any damage from any bioagent in 1997. The control site had one stem larvae and one gall and gall fly larvae on the plants in August 1998. Two bud larvae were also found in the flower head of one Canada thistle plant from the control site in August.

	NUMBER OF PLANTS COLLECTED		THISTLE HEIGHT			STEM WEEVIL DAMAGE						TOTAL NONSTRUCTURAL CARBOHYDRATES		
						Damaged plants		Stem damage		Larvae				
Date	Control site (no.)	Release site (no.)	Control site (cm)	Release site (cm)	Pr > t	(no.)	(%)	(cm)	(%)	(total)	(no./plant)	Control site (%)	Release site (%)	Pr > t
1997														
May	10	43	11.9	8.5	0.006	20	47	na	na	17	3	na	na	
June	0	30	na	68.7	na	14	47	4.7	7	1	3	na	na	
July	30	30	87.9	86.7	0.45	8	27	4.4	5	0	0	2.8	4.0	>0.20
Sept	30	31	64.8	53.5	0.0001	0	0	0	0	0	0	3.3	3.6	>0.20
1998														
May	na	19	na	6.2	na	0	0	0	0	0	0	3.7	1.9	0.06
June	30	30	38.8	48.3	0.0001	7	23	10	20	4	1	3.8	1.7	0.06
July	30	30	55.2	87.9	0.0001	16	53	8	10	0	0	5.1	2.3	0.001
Aug	31	30	73.9	87.6	0.56	14	47	12	13	0	0	4.6	1.8	0.002
Oct	na	28	na	53.9	na	23	82	9	17	0	0	na	7.3	na

1997. Forty-seven percent of the collected stems from the release site had mining damage (Table 2). Stem damage was not observed until June in 1998 and then only 23% of the collected stems had mining damage (Table 2). By August 1998, 82% of the Canada thistle stems had mining damage whereas in September 1997, no collected stems had damage.

Mining damage averaged 4.5 cm of the stem length in 1997 which was 7% of the stem in June but only 5% in July due to stem elongation. In 1998, about 9.5 cm of stem was mined accounting for 10 to 20% of the stem length. These data indicate that Canada thistle stem weevils were active at the release site although the larvae were not always observed.

Canada thistle stem weevil larvae were found 40% of the stems with an average density of 3 larvae/stem in May 1997. In June 1997, the incidence of larvae was 3% with 3 larvae/stem. In 1998, larvae were found only in June in 13% of the stems at a density of 1 larvae/stem. Although fewer larvae were found in 1998 than in 1997, stem mining incidence and mining length were greater in 1998.

Observing larvae in the plant in May and June would be consistent with the reported life cycle of the Canada thistle stem weevil. The Canada thistle stem weevil lays its eggs in March and April, with larval feeding occurring in the stem through late June (Rees et al., 1995). The larvae exit the stem and pupate in the soil with adults emerging in August. The adults overwinter in soil and become active again in March of the following year.

Canada thistle root carbohydrate content. TNC contents of thistle roots were similar among all samples collected in 1997 (Table 2). In 1998, root TNC content at the control site averaged about 4.2 mg TNC/g of root and averaged about 1.9 mg TNC/g of root at the release site, a 60% reduction in TNC in roots at the release site throughout most of the growing season. In October, TNC content of the roots in the release site increased by 4-fold.

This dramatic increase in TNC root content has been reported for Canada thistle (McAllister and Haderlie 1985) and leafy spurge (Lym and Messersmith 1987). TNC helps the root to overwinter and aids in promoting spring shoot growth. It was unfortunate that Canada thistle in the control site was not collected so that a late-season comparison between insect-stressed and non-stressed plants could be made. However, the root TNC content in May 1998 was 1.5 times lower in the release site compared the control site. Biocontrol agents may be adversely affecting overall root health. In the long run, this type of effect should reduce stand vigor, since the main mechanism of Canada thistle spread and sustainability is due to buds on lateral roots.

Lake Louise

Canada thistle plant height. Canada thistle plants were taller at the release site in June and September 1997 and June and October 1998 when compared to plants at the control site during the same months (Table 3). Insect damage may have contributed to differences in plant height with injured stems elongating to a greater extent at the release site than at the control site.

Table 3. Canada thistle stem damage at Lake Louise in 1997 and 1998. Data reported in the table include the number and percentage of damaged plants, the length of the stem damage and the percentage of stem that was damaged and the number of stem weevil larvae found in all plants and the average number of larvae per plant, and the total nonstructural carbohydrate content of roots from the control and release sites. Plants in the control site did not have any damage from any bioagent in 1997. In 1998, plants in the control site had damage from the stem weevil in July, August, September, and October. The stem weevil was found in July and August with about 4 larvae found in 4 plants. In August, 13 bud larvae eggs were found in the flowers of Canada thistle.

	NUMBER OF PLANTS COLLECTED		THISTLE HEIGHT			STEM WEEVIL DAMAGE						TOTAL NONSTRUCTURAL CARBOHYDRATES		
						Damaged plants		Stem damage		Larvae				
Date	Control site (no.)	Release site (no.)	Control site (cm)	Release site (cm)	Pr > t	(no.)	(%)	(cm)	(%)	(total)	(no./plant)	Control site (%)	Release site (%)	Pr > t
1997														
June	30	31	104	128	0.001	29	94	18	10	3*	3*	2.4	2.5	>0.5
Aug	30	40	133	131	0.54	29	73	10	8	9**	3**	na	3.7	na
Sept	23	40	81	141	0.001	26	65	11	8	2**	2**	4.3	na	na
1998														
May	30	30	16	13	0.005	20	67	1.3	10	14*	3*	3.7	3.0	0.48
June	30	54	49	62	0.0001	44	81	16	26	29*	3*	6.5	1.7	0.002
July	30	30	149	117	0.0001	25	83	10	9	1*	3*	2.5	1.9	0.39
Aug	31	30	140	143	0.34	20	67	8	6	1**	1**	4.7	2.7	0.02
Oct	15	29	29	105	0.001	18	62	23	22	0	0	7.5	1.1	0.001

* Stem weevil larvae.

**Galls found on stem and gall fly larvae found in galls.

Canada thistle stem damage. Stem damage was observed at the first sampling date (June, 1997 and May, 1998) and throughout the rest of the season (Table 3). The percentage of stems with mining damage ranged from 65% to 94% in 1997 and from 62% to 83% in 1998. Only 3 stem weevil larvae were found in 1 stem in June, 1997. In 1998, stem weevil larvae were found in 48% of the stems in May and 60% of the stems in June, averaging 3 larvae per stem. In August, more than 65% of the stems had mining damage, although no stem weevil larvae were present.

The length of mined stem varied between years. In 1997, the average mined length was about 13 cm. In 1998, the mined length ranged from about 1 cm (May) to 23 cm (October) which was 5 to 25% of the entire stem length. These data indicate that thistle stem weevils were active in the release site.

In 1998, 60% of the sampled stems from the control site had mining damage in October and stem weevil larvae were found in 5 stems. These data indicate that stem weevils were present at the control site. In our study, we sampled control areas prior to sampling release sites and the three locations were sampled on different days. Therefore, the outmigration of the insect may have occurred by natural movement (wind, flight, etc.) rather than our movement between sites.

Galls and gall fly larvae were found on stems in August and September 1997, and August 1998 although not in high abundance (Table 3). The appearance of galls and gall fly larvae indicate that the gall fly was active at the Lake Louise release site.

Bud damage and bud weevils were observed on one plant at Lake Louise in July 1998. Egg cases were found in buds of 13 more plants indicating that 43% of the sampled plants were infested. The greatest number of egg cases that were found in one bud was five. Bud weevil larvae were found in four plants with the highest density of 2 larvae/bud. The fact that buds were infested is encouraging. Destroying Canada thistle seed, while not reducing established Canada thistle infestations, would reduce the possibility of newly seeded infestations.

No tortoise beetles or evidence of damage from tortoise beetles were observed. This damage would have been characterized as defoliation of the Canada thistle plant.

Canada thistle carbohydrate content. TNC content of the Canada thistle roots was lower in roots from the release site compared to the content observed in the control site throughout 1998, although no differences were observed in June 1997. TNC contents in roots from the release site averaged about 2.1 mg TNC/g of root during 1998, whereas TNC in control roots averaged about 5 mg/g of root. In October, TNC content of control roots increased about 160% compared to TNC in August, whereas TNC in roots from the release site decreased by 60% during that period. In the long run, this type of TNC reduction should reduce stand vigor.

Lake Andes

Canada thistle plant height. When compared to control plants, plants were taller at LANWR in May and June 1997 and at Chytka in June and September 1997 (Table 4). In 1998, Canada thistle plants at Chytka were taller than control plants in May, June, and July whereas Canada thistle plants at LANWR were shorter in May and taller in July than Canada thistle plants from the control area.

Canada thistle stem damage. Stem damage was first observed in June at LANWR, and in May at Chytka in 1997 (Table 4a). At LANWR, 28% of the stems collected in June 1997 had damage, whereas in September only 3% of the stems were damaged. At Chytka in 1997, the percentage of stems mined ranged from 15% to 53%, with the lowest percentage in September, and highest percentage in July. In 1998, some stem damage was observed in May and June samples (3% of plants sampled), but by October 75% of the collected plants had mining damage. The length of stem mined at LANWR ranged from 1 to 21 cm in 1997, and 0.5 to 10.5 cm in 1998, respectively. At Chytka, stem damage ranged from 6 to 15 cm in 1997, and 1 to 22 cm in 1998, respectively. This damage occurred in 4% to 27% of the entire stem length.

Stem weevil larvae were only found at Chytka in May 1997. At LANWR in 1998, larvae were found in one stem in both May and June, with a density of 5 and 1 larvae/stem, respectively. At Chytka, stem weevil larvae were found in 4 stems in June and 3 stems in July, with average densities of 2 and 1 larvae/stem, respectively.

Canada thistle gall damage was observed in both 1997 and 1998 samples from both locations. At LANWR, galls were present on 7 stems with an average larval density of 6 larvae/gall in July 1997, and on 12 stems averaging 5 larvae/gall in September 1997. Galls were present on 2 stems in July 1998 although no larvae were found, and on 3 stems with about 5 larvae/gall in September 1998. At Chytka, one stem had a gall in July and September 1997, with 4 larvae/gall and 1 larvae/gall, respectively. Galls were found on 7 stems collected in August 1998 with an average density of 3 larvae/gall.

Bud damage was observed on six plants at Chytka in July 1998. The average number of larvae found per bud was 16.

Stem mining was not observed in stems from the control site in 1997. However in 1998, mining was observed in 4% of stems collected from May through August. Larvae were found in two stems at a density of 1 larvae/stem in August. In October, 54% of the sampled plants from the control site had some mining damage that ran for about 30% of the entire stem length. Galls were also found on control stems in September 1997 (3 plants with 6 larvae/gall) and August 1998 (3 plants with 4 larvae/gall).

These observations indicate that Canada thistle stem weevil, Canada thistle gall fly, and Canada thistle bud weevil were active in the release sites. There was some movement from release site(s) to the control area.

Canada thistle carbohydrate content. TNC contents of the Canada thistle roots from both release sites were similar compared to TNC in roots from the control (Table 4b). Although stem mining was evident in a number of plants, this damage did not affect the root health. Since most of the TNC in-

Table 4a. Canada thistle stem damage at Lake Andes site in 1998. Data reported in the table include the number and percentage of damaged plants, the length of the stem damage and the percentage of stem that was damaged and the number of stem weevil larvae found in all plants and the average number of larvae per plant, and the total nonstructural carbohydrate content of roots from the control and release sites. Plants in the control site had damage from the gall fly in September of 1997 with three plants having galls and larvae. In 1998, plants from the control site had damage from the stem weevil and gall fly in August, 1998 with two plants having galls and larvae. Stem weevil damage was also observed in the control site in October, 1998 with 13 plants having damage, although no larvae were observed.

	NUMBER OF PLANTS COLLECTED		THISTLE HEIGHT				LANWR RELEASE SITE DAMAGE						CHYTKA RELEASE SITE DAMAGE					
							Damaged plants		Stem damage		Larvae		Damaged plants		Stem damage		Larvae	
Date	LANWR site (no.)	Chytka site (no.)	Control site (cm)	LANWR site (cm)	Chytka site (cm)	Pr > t	(no.)	(%)	(cm)	(%)	(total)	(no./plant)	(no.)	(%)	(cm)	(%)	(total)	(no./plant)
1997																		
May	14	14	5.4	7	7	0.1	0	0	0	0	0	0	4	29	na	na	4*	3
June	32	21	59	121	127	0.001	9	28	21	17	0	0	8	38	15	12	0	0
July	31	30	na	142	139	na	4	13	1	<1	7**	6**	16	53	8	6	1**	4**
Sep.	34	27	104	101	139	0.001	1	3	0	0	12**	5**	4	15	6	50	1**	1**
1998																		
May	30	29	12	8	24	0.001	1	3	0.5	6	1*	5*	2	7	1	4	0	0
June	30	30	53	58	79	0.05	1	3	6.5	11	1*	1*	5	17	13	16	4*	2*
July	30	28	92	113	134	0.001	0	0	0	0	2**	2	7	25	9	7	3*	1*
Aug.	30	31	96	120	138	0.20	0	0	0	0	3**	5**	2	6	22	17	7**	3**
Oct.	40	16	50	39	38	0.04	10	25	10.5	27	0	0	12	75	2	5	0	0

*Stem weevil larvae.
**Galls found on stem and gall fly larvae found in galls.

Table 4b. Total nonstructural carbohydrate control of roots from Lake Andes sites in 1997 and 1998. No differences from the control site were found in either release site.

	1997			1998				
Date	Control	LAWNR (%)	Chytka	Control (%)	LAWNR (%)	p>t	Chytka (%)	p>t
May	NA	NA	NA	3.5	2.8	0.41	2.8	0.41
June	2.6	2.2	2.7	1.9	2.0	0.90	2.0	0.90
July	NA	NA	NA	1.5	2.3	0.29	2.7	0.15
Aug.	NA	NA	NA	3.1	3.8	0.37	1.9	0.15
Oct.	NA	2.4	2.3	5.9	6.0	0.95	7.1	0.36

formation came from roots collected in 1998 and there was evidence of stem mining at the control site, there may have been no differences to detect.

SUMMARY

Stem mining by the stem weevil was the most observed damage. Stem mining often was not observed until June but was prevalent in late season sampling dates. Stem mining was not observed unless the Canada thistle stem was split down the center, so that tunneling could be seen. Stem weevil larvae were found in the release sites early in the season (May, June), and occasionally in the control sites late in the season. Bud weevils also were found in a limited number of plants from Lake Louise and Lake Andes. In most cases, damage was not observed until July and the bud had to be split to find egg cases or larvae feeding on immature seed and/or flower receptacle parts. Galls and gall fly larvae were found on stems late in the season. Defoliation from the tortoise beetle was not noted at Lake Louise even though this insect had been released there.

Plant height was not an indicator to determine if biological agents were active. Peschken and Wilkinson (1981) reported that mining activity did not affect thistle height. However, galls on the main stem have been reported to reduce stem height compared with plants that had galls on the side shoots or plants with uninfested shoots (Peschken and Finnamore, 1982; Forsyth, 1985).

Total nonstructural carbohydrate (TNC) content of the root was generally lower in roots from release sites compared to roots from control sites at Clear Lake and Lake Louise in 1998. The test for TNC, although complicated, may be a very good method to determine if the biocontrol agents are affecting the ability of Canada thistle to survive. High root TNC content has been linked to increased winter hardiness of roots. If TNC of the roots of an infested plant is low, then the plant may not survive the harsh SD winters. Since Canada thistle infestations rely on root survival to perpetuate or increase patch size or both, the reduction of root TNC may be an important mechanism to reduce patch vigor, and ultimately, Canada thistle's importance in the SD landscape.

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