

The effect of cattle grazing on the growth and miserotoxin content of Columbia milkvetch

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Abstract

The growth and miserotoxin content of Columbia milkvetch (*Astragalus miser* Dougl. var. *serotinus* (Gray) Barneby) were examined following grazing of early growth by cows at a grassland site in southern British Columbia. Grazing behavior and forage consumption of cows were observed. Growth of Columbia milkvetch was determined by measuring the freeze-dried weight of each plant and miserotoxin levels were determined by a rapid screening method. Cows had a tendency to either avoid Columbia milkvetch or to consume it incidentally with other forage so long as there was adequate grass available. As grass became scarce the use of Columbia milkvetch increased. After being grazed, the rate of growth and the toxicity of Columbia milkvetch were substantially reduced. In comparison to ungrazed plants, the aboveground biomass of grazed plants was reduced by more than 50% and the average miserotoxin content per plant was reduced by more than 75% during a 6-week period of regrowth. While early grazing may reduce the potential hazard of Columbia milkvetch to livestock, the plant is not a preferred species and may not be consumed by cattle until other forage becomes scarce. Heavy grazing intensity may, in turn, result in low vigor of bunchgrasses and a deterioration of range condition which may result in more weeds in the plant community. Clearly these aspects of management require further study.

Key Words: *Astragalus miser*; toxicity; growth; biomass; diets; selection

Columbia milkvetch (*Astragalus miser* Dougl. var. *serotinus* (Gray) Barneby), also known as timber milkvetch, and about 50 other species of *Astragalus* (Fabaceae) contain toxic glycosides of 3-nitro-1-propanol (Williams and Gomez-Sósa 1986). Miserotoxin, 3-nitro-1-propyl β -D-glucopyranoside, is the poisonous glycoside in 10 species and varieties of *Astragalus* that occur on rangelands in western North America (Stermitz and Yost 1978). In

ruminants, miserotoxin is rapidly hydrolyzed by microbial enzymes of the rumen and the liberated nitroalcohol is rapidly absorbed and converted to 3-nitropropionic acid, an oxidation that is probably catalyzed by hepatic alcohol dehydrogenase (Majak et al. 1984, Pass et al. 1985). The nitroacid is the lethal metabolite and it is a potent inhibitor of mitochondrial enzymes essential to respiration (Gustine and Moyer 1983, Gould et al. 1985).

In an earlier study, we determined the effect of clipping on the growth and miserotoxin content of Columbia milkvetch in southern British Columbia (Majak et al. 1988). Growth was determined by measuring the freeze-dried weight of individual plants and the miserotoxin content of each plant was determined by a rapid screening procedure that permitted the analysis of a large number of plants. In response to early clipping in the spring, the biomass and toxicity of Columbia milkvetch were reduced by about 50% in both years (1984 and 1986) of the study. In practical terms, these results indicated that early grazing of Columbia milkvetch could reduce the potential hazard of the poisonous plant significantly. If cattle grazed the plant at early stages of growth when the biomass was small and the amount of toxin per plant was low, then the subsequent availability and toxicity of Columbia milkvetch would be reduced. This could be especially beneficial in a "second pass" situation where livestock forage on regrowth. The present study was conducted to verify the results of the earlier clipping trial. Biomass and miserotoxin content were again determined but this time in response to the effects of actual grazing and not mechanical clipping, which may not necessarily simulate the animals' feeding habits.

Materials and Methods

The experiment was conducted in a fenced paddock (37 \times 44 m) at the rough fescue (*Festuca scabrella* Torr.) grassland site near Kamloops, B.C., which was described previously (Majak et al. 1988).

Plant community composition was estimated by establishing 3, 36-m long transects across the paddock. Cover values using a modified loop technique (10-cm loop) were taken at 1-m intervals

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Manuscript accepted 13 February 1989.

along the transects. All plants rooted within the loop were recorded. This technique had been tested against the point frame method and found to give comparable results in this community (D. Quinton unpublished data). Biomass production was estimated by clipping 6 randomly located 1-m plots to ground level.

In the grazing trial 3 Hereford cows (3 years old) and their calves grazed the paddock from noon 6 May to noon 12 May 1987. The cattle were put in an adjacent holding pen to minimize trampling on the study site. Water was available at all times. Observations on grazing behavior were made at 0600, 1200, and 2000 h using the bite count technique (Reppert 1960). Labor constraints of other responsibilities dictated that all observations were not made during all time periods on all days. Each cow was followed closely by 1 of 3 observers who recorded a minimum of 700 bites per grazing time. The average period of observation was 54 min and the cattle were observed for a total time of 11 h. Bites of bunch grasses and the larger forbs were recorded by species. When cows were grazing a mixture of species among the larger plants it was impossible to identify intake by individual species so mixtures were recorded.

To measure the effects of grazing on Columbia milkvetch, the size (basal area \times height) of 120 plants within the paddock ("grazed" plants) and 140 plants adjacent to the paddock ("ungrazed" plants) was determined before the grazing trial started. Grazed and ungrazed plants were arranged by volume from smallest to greatest and then sorted, according to size, into 20 blocks of 6 plants each in the case of grazed plants and 7 plants each in the case of ungrazed plants. One plant from each grazed block was randomly assigned to each of the 6 grazed groups within the paddock. Similarly, one plant from each ungrazed block was randomly assigned to each of the 7 ungrazed groups adjacent to the paddock. This ensured an even distribution of biomass in all sample groups. To provide base data one ungrazed group adjacent to the paddock was harvested when grazing began. All grazed plants were measured following grazing to estimate the degree of use. After the grazing period ended, 1 ungrazed and 1 grazed group of 20 plants (excised at the base just above the crown) was collected each week for 6 weeks from 12 May to 23 June. Data were collected in a design of 2 treatments and 6 harvest dates (Table 1).

Table 1. Experimental design for collecting groups of grazed (C) and ungrazed (U) Columbia milkvetch plants after the grazing period which ended on 12 May 1987. Each group contained 20 plants.

Regrowth period (week)	Sampling Date						
	12 May	19 May	26 May	2 June	9 June	16 June	23 June
0	U	U	U	U	U	U	U
1		G					
2			G				
3				G			
4					G		
5						G	
6							G

Freshly collected plants were freeze-dried and extracted individually. Miserotoxin concentrations were estimated by the rapid, colorimetric procedure described previously (Majak et al. 1988). Fifty plant extracts were also analyzed by high pressure liquid chromatography (HPLC) to check the accuracy of the colorimetric method. Extracts were treated with 10% $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ (1 volume) and 0.5 N NaOH (0.5 volume) to precipitate protein prior to HPLC. Duplicate HPLC determinations were facilitated with a Varian 9090 auto sampler. The HPLC column was cleaned with a methanol gradient between injections of plant extracts (Muir and Majak 1984). The differences in miserotoxin values obtained by the 2 methods were calculated and the accuracy of the colorimetric

method was estimated from the standard deviation (SD) of the differences.

The effects of grazing and regrowth time on plant weight (g of dry matter), toxin concentration (percent miserotoxin), and the total amount of toxin per plant were examined by analysis of variance. The amount of toxin per plant was transformed to logs for analysis. Linear and quadratic curves were fitted using polynomial regression (Freund and Littell 1981).

Results and Discussion

Eighty-seven percent of the total production of 242 g/m² (dry matter) was grasses (Table 2). *Festuca scabrella*, occurring in less

Table 2. Average production of rough fescue grassland paddock near Kamloops, B.C. during a grazing trial in May 1987.

Species	Percentage of total production	Dry matter g/m ² \pm SE
<i>Festuca scabrella</i>	54.2	131.2 \pm 39.9
Grass and grass-like plants ¹	24.8	60.0 \pm 19.5
<i>Agropyron spicatum</i>	8.3	20.1 \pm 15.1
<i>Astragalus miser</i>	7.5	18.2 \pm 4.4
Forbs ²	5.2	12.6 \pm 1.0

¹Grass and grass-like plants may contain species of *Poa*, *Carex*, *Juncus*, *Koeleria* and *Stipa*.

²Forbs may contain species of *Achillea*, *Erigeron*, *Ranunculus*, *Calochortus*, *Tragopogon*, *Delphinium*, *Aster*, *Antennaria*, *Taraxacum*, *Cerastium* and *Saxifraga*.

than 20% of the cover loops (Table 3), accounted for more than one-half the total biomass production. Columbia milkvetch, occurring in 44% of the cover plots, accounted for 59% of the total production (30.4 g/m²) of forbs.

Table 3. Cover classes of forage inside 37 \times 44 m paddock for grazing of Columbia milkvetch in southern B.C.

Percent cover	Species
80–100%	None
60– 80%	<i>Poa pratensis</i>
40– 60%	<i>Astragalus miser</i> , <i>Achillea millefolium</i>
20– 40%	<i>Antennaria</i> , <i>Koeleria</i> , <i>Montia</i> / <i>Ranunculus</i>
1– 20%	<i>Agropyron spicatum</i> , <i>Festuca scabrella</i> , <i>Koeleria</i> , <i>Cerastium</i> , <i>Fritillaria</i> , <i>Tragopogon</i> , <i>Collinsia</i> , <i>Microsteris</i> , <i>Taraxacum</i> , <i>Erigeron</i> , <i>Juncus</i> , <i>Delphinium</i> , <i>Calochortus</i> , <i>Balsamorhiza</i> , <i>Aster</i> , <i>Spartina</i> , <i>Stipa</i> , <i>Poa sandbergii</i> , <i>Lithospermum</i> , <i>Polygonum</i> , <i>Comandra</i> , <i>Crepis</i>

Preliminary observations of cow diets (Table 4) showed that cows initially consumed the *Festuca* and *Poa* grass/forb mix. Cows made several passes over the paddock regrazing plants that

Table 4. Diets of 3 cows grazing Columbia milkvetch paddock during 6–12 May 1987 in southern B.C.

Day	% of total bites				
	<i>Agropyron spicatum</i>	<i>Festuca scabrella</i>	Grass/forb mix ¹	Grass/forb/-vetch mix ¹	<i>Astragalus miser</i>
1	1.9	49.7	47.4	0.4	0.7
2	1.2	21.2	45.0	30.6	2.1
3	1.7	13.7	72.7	10.1	1.8
4	1.6	43.7	24.7	28.4	1.7
5	4.2	24.0	62.2	2.3	7.3
6	0.9	17.0	77.9	0.0	4.5
Average	1.8	22.7	60.4	12.2	2.9

¹Mixes may contain species of *Poa*, *Carex*, *Juncus*, *Koeleria*, *Stipa*. Forbs, including vetch made up less than 10% of composition.

had been initially selected. After 2 days of grazing, most *Festuca* plants had been grazed with some plants showing severe use. At this time most of the palatable forage in the paddock had some degree of usage. Most Columbia milkvetch consumed early in the study was taken incidentally in conjunction with other forages by cow #2 (Table 5), which grazed mainly the grass/forb/vetch component of the community in a nonselective manner. Columbia

Table 5. Consumption of Columbia milkvetch by individual cows in a rough fescue paddock in B.C. during 6–12 May 1987.

		% of total bites		
		Cow 1	Cow 2	Cow 3
Day:				
1	12 NOON	0.7	0.6	0.0
	8 PM		3.2	
2	6 AM		8.7	0.2
	12 NOON	0.0	0.9	
	8 PM		1.6	0.4
3	6 AM		2.5	0.2
	12 NOON		3.8	0.5
4	6 AM	1.1	2.2	0.4
	8 PM		2.0	
5	6 AM	2.3	2.0	
	8 PM			9.6
6	6 AM	1.8	8.1	

milkvetch plus other forbs constituted less than 10% of the composition of diet mixes. Cows #1 and #3 were observed to mouth vetch, reject it and then avoid it in grazing. However, as grasses and other forage became scarce, more Columbia milkvetch was consumed (Tables 4 and 5). Cows #2 and #3 were observed to be actively seeking Columbia milkvetch on days 5 and 6, reaching through the fence to graze plants on the perimeter of the paddock. The average miserotoxin concentration at the time of grazing was about 6%. At this level it would take 1.2 kg dry weight of plant material to acutely poison a cow (Williams et al. 1967, James et al. 1980). No visual effects of poisoning were present during this study. Usage of the grazed milkvetch plants (Table 6) was relatively uniform. Ninety-

Table 6. Average height of 120 marked plants of Columbia milkvetch following heavy grazing of a 37 × 44 m paddock in southern B.C.

Grazing intensity	No. of plants	Height (cm)
All stems grazed	44	2.2
Few stems ungrazed	65	2.6
Two heights	2	7.0 & 2.8
Topped	5	6.0
Ungrazed	1	10.0
Manure covered	3	

one percent were grazed to an average height between 2 and 3 cm, 2% were partially grazed to 2 heights, 4% were topped and 3% were not grazed.

In agreement with our previous study there was a good correlation between the analytical HPLC procedure and the rapid screening method for miserotoxin determination ($r=0.99$ $n=50$). In 1987, the average difference (\bar{x}) between the quick method and the HPLC procedure was 0.69% miserotoxin and the SD of the difference was 0.39. In the earlier studies, the comparable values were $\bar{x} = 0.16$ (SD=0.55) for 1986 and $\bar{x} = 0.54$ (SD=0.77) for 1984. Thus the dispersion about the mean was the smallest in 1987 but the largest in 1984 when the protein precipitation step was not used. The use of the auto-sampler in 1987 was especially advantageous since it greatly improved the efficiency and reproducibility of the HPLC

procedure and this may have improved the correlation between the 2 methods.

In 1987 the rate of growth of the milkvetch plants at the grassland site (Fig. 1) closely resembled the pattern that was observed in 1986 (Majak et al. 1988). This could be partly attributed to the

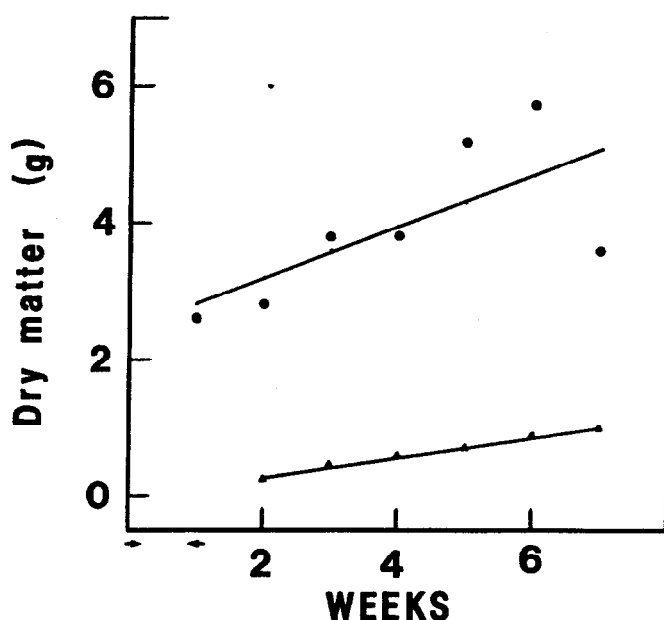


Fig. 1. Effects of grazing on the average above ground biomass (dry matter) of Columbia milkvetch plants during 1987. Ungrazed plants are indicated with circles and grazed plants with triangles. Arrows indicate the grazing period. SE (dry matter) = 0.56.

average temperatures for May and June, which were close to normal in both of those years unlike 1984, which had the coldest May on record (Atmospheric Environment Service 1984, 1986, 1987). The phenological developments of the plants in 1986 and 1987 were also similar as the plants progressed from the early bud stage of growth and into the bloom and pod stages during the 6-week study periods.

The analysis of variance revealed the same trends reported previously in the clipping study (Majak et al. 1988). The synthesis and accumulation of miserotoxin followed a pattern similar to the change in the biomass with the significantly higher amount of the toxin (Fig. 2) being present in the largest plants (Fig. 1). The effects of grazing on the dry matter biomass of Columbia milkvetch (Fig. 1) were identical to the effects of clipping (Majak et al. 1988). The growth curves for the clipped plants at the grassland site in 1986 and the grazed plants at the same site in 1987 were superimposable. Both treatments resulted in a marked diminution of Columbia milkvetch dry matter (Fig. 1) and a correspondingly significant decrease in the amount of available toxin per plant (Fig. 2). These combined results verify our previous hypothesis that early grazing can significantly reduce the potential hazard of Columbia milkvetch to livestock.

When the miserotoxin content was expressed as a percentage of the dry matter, ungrazed plants showed a typical decline in toxin concentration with advancing stages of growth (Table 7). It should be emphasized, however, that this decrease in concentration was overshadowed by the larger increase in biomass and the concomitant greater availability of the toxin (Fig. 1 and 2).

Miserotoxin concentrations were also determined in grazed plants where the ungrazed portion of the plant was usually combined with the regrowth material to make a sample of sufficient size for chemical analysis. The miserotoxin concentrations in the total

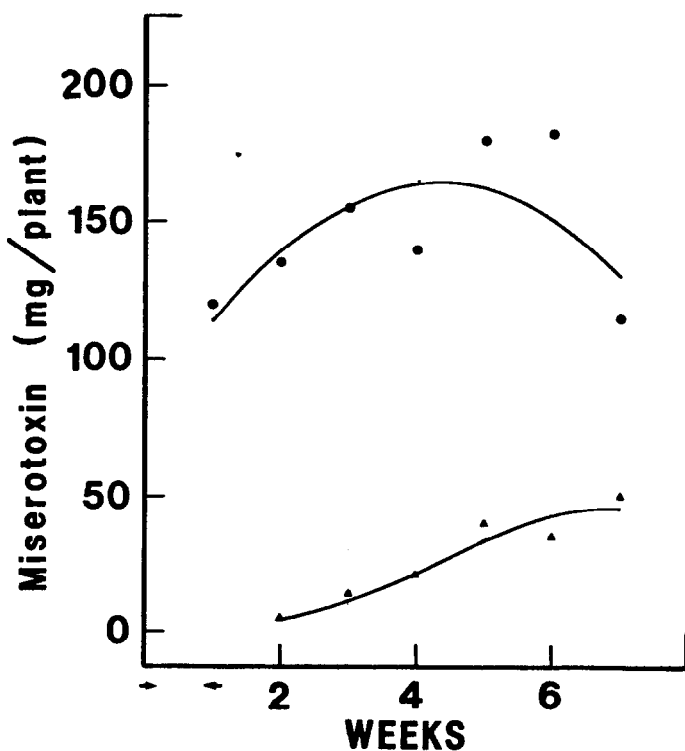


Fig. 2. Effects of grazing on the average amount of miserotoxin in Columbia milkvetch plants during 1987. Ungrazed plants are indicated with circles and grazed plants with triangles. Arrows indicate the grazing period. SE (miserotoxin) = 0.20 (\bar{x}).

biomass of the grazed plants showed an increase during the study period (Table 7). This increase could be attributed to the increasing proportion of the regrowth biomass and its higher level of the toxin (Table 7). In addition, this trend would be enhanced by the decreasing biomass of the ungrazed portion due to senescence and its lower level of the toxin (Table 7). The miserotoxin levels in the

Table 7. Average miserotoxin concentrations (%) in grazed and ungrazed Columbia milkvetch plants after the grazing trial at the grassland site in southern B.C., in 1987, data expressed on a dry matter basis.

Date	Ungrazed plants ¹	Grazed plants		Total biomass ¹
		Regrowth biomass ²	Ungrazed biomass ²	
		(% miserotoxin)		
12 May	5.89	—	—	—
19 May	5.85	—	—	3.40
26 May	4.96	6.03 (5)	4.50 (4)	3.93
2 June	5.23	7.43 (9)	3.70 (9)	4.54
9 June	4.83	7.49 (14)	4.03 (12)	7.34
16 June	4.31	7.63 (6)	3.53 (4)	7.04
23 June	3.86	6.55 (12)	2.65 (7)	6.09

¹Each value is the mean of 20 determinations. SE = 0.30.

²Number of determinations are indicated in brackets.

ungrazed biomass appear to be lower than in the controls, but whether this is a physiological response to the effects of grazing is not known. Higher concentrations of miserotoxin in regrowth tissue were also reported earlier (Majak et al. 1988) but the consequences of this should not be misconstrued because the amount of toxin available for consumption is substantially reduced by the effects of grazing (Fig. 2).

Preliminary grazing studies indicate that Columbia milkvetch is neither a highly preferred species nor is it selectively grazed by cattle under good range conditions. Cattle, however, can be forced to eat Columbia milkvetch when other forages are scarce. Caution should be exercised in early grazing systems or in grazing that utilizes the regrowth of milkvetch since anecdotal evidence in southern B.C. indicates that under certain conditions cattle may become addicted to the plant. Caution may also be given to preventing overuse of preferred species in an effort to force cattle to graze Columbia milkvetch. Bluebunch wheatgrass and rough fescue in particular are in a stage of accelerated growth with minimum annual root reserves (McIlvanie 1942) and thus they are at the most vulnerable stage of growth to be defoliated by grazing. Overuse may result in reduced vigor of grasses, deteriorating range condition, and even more weeds in the plant community. Clearly, using early grazing as a tool to manage rangeland infested with Columbia milkvetch warrants further study.

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