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Sheep Preference for Perennial Glandular-haired and Eglandular Medicago Populations

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ABSTRACT

Erect glandular-haired *Medicago* species and hybrid populations have been developed that possess a high degree of resistance to the alfalfa weevil [*Hypera postica* (Gyllenhal)] and potato leafhopper [*Empoasca fabae* (Harris)]. Trials were conducted at the North Agronomy Farm, Manhattan, KS, in June and August 1986 to determine if erect glandular hairs on diverse perennial *Medicago* populations affect forage preference of sheep (*Ovis aries*). Glandular-haired (+) and eglandular (−) populations were selected from each of the following: diploids *M. prostrata* Jacq. and *M. glandulosa* David (KS94) and tetraploids *M. glutinosa* Bieb. (KS108), *M. sativa* L. × *M. prostrata* (KS159), and *M. sativa* × *M. glutinosa* (KS160). Eglandular *M. sativa* 'Riley' ($2n = 4x = 32$) and *M. sativa* spp. *caerulea* (Less. ex Ledeb.) Schmal. ($2n = 2x = 16$) (MS2*x*) were included as controls. Four pairs of Suffolk wethers were allocated randomly among four replicates of *Medicago* populations in a cafeteria grazing trial. Sheep bites were counted 1 of every 5 min that grazing occurred. Pre- and postgrazing plant heights were measured. Populations differing by (+) or (−) showed significant differences for five of the 10 comparisons for the two grazing periods. The KS108(+), KS159(+), and KS160(+) entries were preferred to their corresponding eglandular populations in the June period. The KS108(+) and KS159(−) entries were preferred to their corresponding (−) and (+) entries, respectively, in the August period. Riley was significantly preferred over MS2*x* in the August period. Tetraploid entries had significantly higher initial plant height and were generally more preferred than diploid entries for both periods. For these populations, the presence of glandular hairs and their exudates did not negatively affect forage preference by sheep.

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NUMEROUS physical characteristics of plants, including trichomes, reduce ruminant forage utilization (Burns, 1978). Hanna et al. (1974) reported that fresh pearl millet [*Pennisetum americanum* (L.) K. Schum.] leaves with simple hairs had higher in vitro dry matter disappearance (IVDMD) than did glabrous lines. When animals were given a choice, however, glabrous lines were preferred over lines with trichomes.

Although toxin-containing exudates from glandular hairs are found on many plant species, (Levin, 1973; Knox and Dodge, 1986), no toxins were detected in the exudate from *Medicago scutellata* (L.) Mill. (Triebe et al., 1981). Populations of perennial glandular-haired

Medicago species and hybrids have been developed that possess a high degree of resistance to the alfalfa weevil and potato leafhopper. Cultivars also have been developed from several annual *Medicago* species (Heyn, 1963) that possess erect glandular hairs (Jackson and Jacobs, 1985), but animal preference relative to presence or absence of glandular hairs apparently has not been studied. Based upon laboratory estimates, Lenssen et al. (1988a, b, and c) reported that forage quality of diverse *Medicago* species and hybrid populations was unaffected by the presence or absence of erect glandular hairs and their exudates. Alfalfa breeders have expressed concerns that incorporating erect glandular hairs into alfalfa populations might affect animal preference or intake.

The primary goals of our study were to determine: (i) whether erect glandular hairs and their exudates affect forage preference of sheep and (ii) possible sheep preferences among wild, perennial *Medicago* species, hybrid populations, and cultivated alfalfa.

MATERIALS AND METHODS

Populations of plants with (+) and without (−) erect glandular hairs were selected from perennial diploids, *M. prostrata* Jacq. and KS94, and tetraploids, KS108, KS159, and KS160. The populations KS94GH6 (*M. glandulosa* David; perennial diploid) (Sorensen et al., 1986), and KS108GH5, (*M. glutinosa* Bieb.; PI 346919, perennial tetraploid) (Sorensen et al., 1985) have been previously described in detail. Erect glandular-haired plants of KS94 and KS108 were selected from populations of the sixth and fifth cycles, respectively, of recurrent phenotypic selection for higher frequency of those hairs. Eglandular plants representing KS94 and KS108 were selected from the first cycles of selection for erect glandular hairs. KS159 is an *M. sativa* L. × *M. prostrata* tetraploid hybrid backcrossed three times to *M. sativa* populations. KS160 is an *M. sativa* × *M. glutinosa* hybrid backcrossed three times to *M. sativa* populations. Selected (−) and (+) populations representing KS159 and KS160 were from first and fourth cycles, respectively, of selection for erect glandular hairs. Eglandular tetraploid *M. sativa* 'Riley' (Sorensen et al., 1978) and diploid *M. sativa* subsp. *caerulea* (PI 172984) were included as controls.

Seeds of all populations were scarified, treated with a commercial *Rhizobium* inoculant, and planted in flats in a greenhouse. Seedlings selected for (+) or (−) were transplanted to peat pots and later transplanted into a field nursery on 19 to 21 May 1985. Plants were spaced 30 cm within rows, which were 45 cm apart. Plots were arranged in a randomized complete block design with four replicates of 20 plants per *Medicago* entry. The experimental site was in an area mapped as Wymore silty clay loam, 1 to 4% slopes (Wymore series: fine, montmorillonitic, mesic Aquic Argiudolls) on the North Agronomy Farm, Manhattan, KS. Diseases and insects were controlled to eliminate possible confounding effects on forage quality (Willis et al., 1969). Irrigation water was supplied as needed to prevent moisture stress. Pre- and postgrazing in situ plant heights were measured. Prior to grazing, percent bloom was determined for each plant population.

Suffolk lamb birth dates were from 22 Jan. 1986 to 10 Feb. 1986. Lambs and ewes were allowed access to smooth bromegrass (*Bromus inermis* Leyss.) pasture. Wethers were weaned on 11 April 1986 and housed in pens without access to fresh pasture until the June grazing period commenced. Sheep were fed 1.8 kg head^{-1} d^{-1} of mixed concentrate and ad libitum alfalfa hay (cultivar unknown) after weaning. Seven days prior to the start of grazing, wethers were switched to ad libitum prairie hay and 0.9 kg head^{-1} d^{-1} of mixed concentrate. Poloxalene was mixed with the concentrate to prevent possible occurrence of frothy bloat. Concentrate was added to the prairie hay diet to prevent confounding preference with physiological stress and subsequent hunger from deficient dietary crude protein concentration (CP) prior to grazing (Elliott and Topps, 1963). Previous research has shown that these *Medicago* populations contain sufficient CP to preclude low-protein stress (Lenssen et al., 1988c).

The first grazing period was 9 to 18 June 1986. Plants were not grazed on 10 and 11 June because of rainfall and muddy conditions. Grazing dates for the second period were 16 to 26 August. Grazing did not occur on 19, 21, and 25 August. Average weights of wethers were 42.6 and 54.0 kg for June and August, respectively.

Wethers were randomly paired to encourage normal behavior (Marsden and Wood-Gush, 1986) and pairs were confounded with plant replicates. Approximately 20 plants from each of the 12 populations (240 total plants per replicate) were available to each pair of sheep in a cafeteria grazing trial. Steel panels were used to confine sheep within each grazing area. Shade and drinking water were provided. Pairs were allowed to graze a designated replicate for approximately 8 h per day for 2 d during the June and August grazing periods. Animals were removed from the grazing area at night.

Sheep preference was quantified by counting the number of bites per *Medicago* population for each wether for 1 of every 5 min that grazing occurred. Two observers were present to facilitate bite count data collection.

Analysis of variance procedures were used on pre- and postgrazing plant height data. Actual and percentage height changes were calculated and analyzed in the same manner. Mean separations were tested with Fisher's Least Significant Difference at the 0.05 level of probability (Snedecor and Cochran, 1980).

Bite data were divided into three time periods, the first day (A), and morning (B) and afternoon (C) of the second day to measure differential utilization of populations across time. Data were tested with Hartley's Homogeneity of Variance test (Cochran and Cox, 1957) within each time period. Using the three time periods, four weighted models were used to describe utilization of populations across time. The weighted models were: RW1)$1/2A + 1/3B + 1/6C$; RW2)$1/3A + 1/3B + 1/3C$; RW3)$1/6A + 1/3B + 1/2C$; and 4)$1/4A + 1/2B + 1/4C$. Populations were evaluated by ranking within each of the four models, and the assigned rank values were tested by Friedman's Nonparametric Two-way Analysis of Variance Test (Lehmann, 1975). Means were separated with a two-tailed Friedman's Test at the 0.05 level of probability (Conover, 1980). Correlation coefficients among rankings and plant parameters were calculated.

RESULTS AND DISCUSSION

Conclusions rarely differed among the four weighted models compared for the June and August harvests (Table 1). Correlation coefficients between ranking from the four models were positive and highly significant (Table 2). We selected Model RW1, with the highest weight on bites taken the first day, to describe the utilization of the populations under grazing.

Wide differences for forage preference by sheep were noted among the 12 *Medicago* entries during both

Table 1. Summed ranks for four weighted models to estimate sheep preference for 12 *Medicago* populations, June and August 1986.

	June				August			
Entry†	RW1‡	RW2	RW3	RW4	RW1	RW2	RW3	RW4
Pros(+)	6§	6	9	6	7	7	6	7
Pros(−)	7	5	5	5	5	5	6	5
KS94(+)	23	21	15	17	20	18	20	17
KS94(−)	21	19	15	20	17	15	17	16
MS2*x*	30	32	27	31	26	25	22	23
Riley	36	40	40	38	41	40	35	37
KS108(+)	40	39	41	42	40	39	37	38
KS108(−)	30	33	38	32	22	27	33	29
KS159(+)	40	40	40	39	37	39	41	40
KS159(−)	23	24	26	24	43	43	41	43
KS160(+)	33	30	34	34	29	26	28	30
KS160(−)	24	22	21	23	25	28	26	27
$t_{0.975}$¶	7	7	6	6	6	5	6	6

† Pros = *M. prostrata*, KS94 = *M. glandulosa*, MS2*x* = *M. sativa* subsp. *caerulea* (diploid), Riley = *M. sativa* (tetraploid), KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*, (+) = presence of erect, glandular hairs, (−) = absence of erect, glandular hairs.
‡ RW1 = $1/2A + 1/3B + 1/6C$; RW2 = $(A + B + C)/3$; RW3 = $1/6A + 1/3B + 1/2C$; RW4 = $1/4A + 1/2B + 1/4C$; A = number of bites counted during first day, B = number of bites counted during morning of second day, and C = number of bites counted during afternoon of second day.
§ 1 = least preferred; 48 = most preferred.
¶ Least significant difference for multiple comparisons among summed ranks for $P = 0.05$.

grazing periods. The KS108(+), KS159(+), and KS160(+) populations were significantly preferred over their corresponding (−) counterparts for June (Table 1). The KS108(+) and KS159(−) entries were significantly preferred to their corresponding (−) and (+) populations, respectively, in the August period. No significant differences occurred between the other corresponding (+) and (−) entries during either grazing period when summed ranks from Model RW1 were tested.

Sheep preferences were not significantly different for the selected tetraploid glandular-haired populations and the alfalfa cultivar Riley for the June period (Table 1). However, Riley was significantly preferred over KS160(+) in the August period. The diploids, *M. prostrata* and KS94, were generally less preferred than the tetraploid entries for both grazing periods.

Pre- and postgrazing in situ plant heights (cm), and percentage reduction in height and bloom differed significantly among entries for both grazing periods (Table 3). Corresponding (+) and (−) entries did not differ significantly for pregrazing height in June or August, except for KS108, where (+) was significantly shorter than (−) at both times. Sheep preference and pregrazing plant height correlations were positive and significant (Table 4). The alfalfa cultivar Riley was the tallest entry prior to grazing in June and August. Tetraploid entries were significantly taller than diploid entries.

Postgrazing plant heights were similar among corresponding (+) and (−) populations except for KS108, for which the (−) population was taller than the (+) (Table 3). Tetraploid entries were significantly taller than diploid entries postgrazing, except for KS108(+) in June. Because of stripping of leaves, postgrazing tetraploid plants consisted primarily of stems, whereas the diploid entries, KS94 and MS2x, were more uniformly grazed. Previous research showed that stems of the diploids frequently had higher *in vitro* dry matter disappearance and lower fiber concentrations than did stems of the tetraploids (Lenssen et al., 1988a,b), which could have reduced selectivity by sheep. Conversely, the diploids had smaller leaflets and lower leaf

Table 2. Correlation coefficients for four models to estimate sheep preference for 12 *Medicago* populations, June and August 1986.

	Model†		
Model	RW2	RW3	RW4
	June		
RW1	0.95**	0.81**	0.94**
RW2		0.88**	0.97**
RW3			0.89**
	August		
RW1	0.94**	0.83**	0.90**
RW2		0.94**	0.98**
RW3			0.96**

** Significantly different from zero at P < 0.01 (N = 48).
† RW1 = $1/2A + 1/3B + 1/6C$; RW2 = $(A + B + C)/3$; RW3 = $1/6A + 1/3B + 1/2C$; RW4 = $1/4A + 1/2B + 1/4C$; A = number of bites counted during first day, B = number of bites counted during morning of second day, and C = number of bites counted during afternoon of second day.

Table 3. Pre- and postgrazing plant heights, reduction in height during grazing, and percentage pregrazing bloom stage for 12 perennial *Medicago* populations, June and August 1986.

Population†	June				August			
	Plant height		Reduction in plant height	Bloom stage	Plant height		Reduction in plant height	Bloom stage
	Pre-grazing	Post-grazing			Pre-grazing	Post-grazing		
	mm		%		mm		%	
Pros(+)	129	128	0	100	164	139	15	100
Pros(−)	127	141	−11	100	170	132	20	100
KS94(+)	185	162	12	38	251	138	44	45
KS94(−)	170	162	4	33	231	143	36	38
MS2x	173	124	28	35	183	106	39	3
Riley	479	345	27	28	553	368	33	20
KS108(+)	330	217	36	23	406	222	45	25
KS108(−)	400	306	24	18	503	322	36	26
KS159(+)	432	275	36	13	500	299	41	19
KS159(−)	432	353	18	13	520	317	38	16
KS160(+)	438	339	24	23	524	313	41	21
KS160(−)	458	394	14	28	513	293	42	16
LSD (0.05)	31	79	21	10	45	65	18	10

† Pros = *M. prostrata*, KS94 = *M. glandulosa*, MS2x = *M. sativa* subsp. *caerulea* (diploid), Riley = *M. sativa* (tetraploid), KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*, (+) = presence of erect glandular hairs, (−) = absence of erect glandular hairs.

Table 4. Correlation coefficients among weighted ranks of sheep bites and pre- and postgrazing plant heights, reduction in plant height, and percentage bloom stage of 12 *Medicago* populations, June and August 1986.

	Plant height		Percentage bloom stage	Reduction in plant height
	Pregrazing	Postgrazing		
	June			
Sheep-bite ranking	0.52**	0.18	−0.61**†	−0.82**
	August			
Sheep-bite ranking	0.67**	0.54**	−0.65**	−0.37**

** Significantly different from $P < 0.01$ (N = 48).
† N = 36 for June.

percentages than the tetraploids during warm growing conditions, which could have impeded selection of leaves without ingestion of stems. Correlation coefficients between sheep-bite rankings and postgrazing plant heights were low and positive for June and significant and positive in August (Table 4).

No significant differences in percentage change of plant heights occurred between corresponding (+) and (−) entries. None of the tetraploid entries differed significantly in actual or percentage height change from Riley alfalfa (Table 3).

Since percentage bloom did not differ significantly between corresponding (+) and (−) populations for June or August grazing periods, these comparisons were not affected by maturity differences. The *M. prostrata* and KS94(+) entries were physiologically more mature than tetraploid entries for both grazing periods. The *M. prostrata* flowered 7 to 10 d postharvest during warm conditions, compared to 28 to 30 d for the other entries. The cultivated alfalfa, Riley, was more mature than KS159 (+) and (−) and KS108(−) during the June trial. No differences in maturity were found among tetraploid entries for the August period. The percent bloom stage was negatively and significantly correlated with sheep bite rankings (Table 4).

Arnold (1960) presented evidence that sheep, when pastured on alfalfa, graze a horizontal plane and select within a vertical plane. He reported that, as alfalfa matured and more forage became available, sheep exhibited more selectivity for leaves. In our trials, differences in maturity and pregrazing plant height may have confounded sheep preference among these *Medicago* species. Not all the variation in rankings, however, was explained by correlations with plant height or maturity.

Sheep bite data and actual and percentage changes in plant heights following grazing indicated that the presence or absence of erect glandular hairs on diverse, perennial *Medicago* species and hybrids can affect forage preference by sheep. Since four of the five comparisons in which (+) and (−) differed were in favor of (+), glandular hairs and their exudates may have a positive effect on forage preference by sheep.

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