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# In Vitro Dry Matter Disappearance, Crude Protein Concentration, and Leaf Percentage of Erect Glandular-Haired Medicago Populations

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# In Vitro Dry Matter Disappearance, Crude Protein Concentration, and Leaf Percentage of Erect Glandular-Haired *Medicago* Populations<sup>1</sup>

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## ABSTRACT

Available resistance in alfalfa (*Medicago sativa* L.) cultivars is currently inadequate to control the alfalfa weevil, *Hypera postica* (Gyllenha), or potato leafhopper, *Empoasca fabae* (Harris), the two most injurious arthropod pests of alfalfa in North America. Resistance to both insects has been documented in other *Medicago* species having erect, glandular hairs. These hairs have been transferred to alfalfa. A field trial in 1985 was to determine the effects of erect, glandular hairs and their exudates on forage quality of several perennial *Medicagos*. Glandular and eglandular plant populations were selected from each of the diploids, *M. prostrata* Jacq. and *M. glandulosa* David., and tetraploids, *M. glutinosa* Bieb., *M. sativa* (MS4n) × *M. glutinosa* (KS160) and MS4n × *M. prostrata* (KS159). Eglandular *M. sativa* cv. Riley and diploid *M. sativa* ssp. *caerulea* (Less ex Ledeb.) Schmal. were included as controls. In vitro dry matter disappearance and CP concentration were

determined for four harvests in 1985 and one in 1986. Leaves were separated from stems to obtain leaf percentage for three harvests in 1985 and one in 1986. The presence of erect, glandular hairs on the wild *Medicago* species or hybrids did not significantly affect dry matter disappearance, CP, or leaf percentage. Glandular-haired entries were similar in forage quality to alfalfa. Data from this study indicate that plant breeders may utilize erect, glandular hairs to improve pest resistance in alfalfa without affecting forage dry matter disappearance, CP, or leaf percentage.

## INTRODUCTION

Alfalfa (*Medicago sativa* L.) is the premier forage crop for dairy production in temperate and semiarid climates. The potential for high yields of nutritious feed has resulted in the production of alfalfa on approximately 11,000,000 ha in the United States (2). The two most destructive arthropod pests of alfalfa in North America are the alfalfa weevil, *Hypera postica* (Gyllenhal), and the potato leafhopper, *Empoasca fabae* (Harris), which limit forage yield and quality by phytophagy (weevil) (1) or plugging of phloem cells and hindering carbohydrate translocation (leafhopper) (18). Although some alfalfa cultivars have moderate tolerance to both pests, none are resistant (1, 16).

Most perennial species within the genus *Medicago* contain trichomes (plant hairs) of

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two major types, simple and glandular (8). Glandular trichomes contain exudate-secreting cells, whereas simple hairs do not. Glandular hairs vary in stature from procumbent to erect and have single or multicellular stalks (12). Glandular exudates from some plant genera have antimicrobial or antifungal activities (10) or induce photosensitivity in sheep (11). Triebe, et al. (26) determined that exudates from *Medicago scutellata* (L.) Mill., an annual, wild relative of alfalfa, do not present a toxicological problem for forage-consuming livestock. Burns (6) reviewed the physical characteristics of plants, such as trichomes, cuticular waxes, and histological classes, which can impart negative influences on ruminant forage crop utilization.

Some glandular-haired *Medicago* species effectively controlled the alfalfa weevil (7) and potato leafhopper (4) under laboratory conditions. There is little information, however, regarding digestibility of these wild species. Our study was undertaken to determine if the presence of erect, glandular hairs and their exudates affect in vitro dry matter disappearance (IVDMD), CP, or leaf percentage (LF) of diverse perennial, *Medicago* populations. We also compared forage quality of the wild, erect, glandular-haired and hybrid populations to that of cultivated alfalfa to determine if the high nutritive value of currently available alfalfa cultivars would be compromised by the incorporation of erect, glandular hairs.

#### MATERIALS AND METHODS

Populations of plants with (+) and without (–) erect glandular hairs were selected from each of the perennial diploids, *M. prostrata* and KS94, and the tetraploids, KS108, KS159, and KS160. Erect, glandular-haired plants of KS94 and KS108 were selected from populations of the sixth and fifth cycles, respectively, of recurrent phenotypic selection for increased density of erect, glandular hairs. Essentially eglandular plants were selected from the first cycles of selection for erect, glandular hairs. KS94GH6 (22), *M. glandulosa* (perennial diploid), and KS108GH5, *M. glutinosa* (Plant Introduction 346919, perennial tetraploid) (21) have been previously described in detail. KS159 is an *M. sativa* × *M. prostrata* tetraploid hybrid backcrossed three times to *M. sativa* pop-

ulations. KS160 is an *M. sativa* × *M. glutinosa* hybrid backcrossed three generations to *M. sativa* populations. Selected (–) and (+) populations were from first and fourth cycles of selection for erect, glandular hairs, respectively. Eglandular diploid *M. sativa* subsp. *caerulea* (Plant Introduction 172984) and tetraploid *M. sativa* cv. Riley (20) were included as controls.

Seeds of all populations were scarified, treated with a commercial *Rhizobium* inoculant (Inoculant for Alfalfa and Clovers, Farmland Industries, Kansas City, MO), and planted in flats in a greenhouse. Seedlings selected for (+) or (–) were transplanted to peat pots and later transplanted into a field nursery on May 19 to 21, 1985. Plants were spaced 30 and 45 cm within and between rows, respectively. The experimental design was a randomized complete block with four replications of 20 plants per replicate. Diseases and insects were controlled with maneb (manganese ethylene bis-dithiocarbamate) and carbofuran (2, 3-dihydro-2, 2-dimethyl-7-benzofuranylmethylcarbamate), respectively, to eliminate confounding effects on forage quality (27). Plots were irrigated due to low rainfall twice in July and once in August 1986.

Tetraploid populations were harvested four times in 1985, but diploids were harvested only three times because growth was insufficient for an October harvest. All populations were harvested once in May 1986. Freshly harvested materials were taken immediately to the laboratory.

#### Scanning Electron Microscopy

Fresh, greenhouse-grown materials were used for scanning electron microscopy. Sections of stems or petioles were mounted on aluminum stubs with Pella silver paste (Ted Pella, Inc., P.O. Box 510, Tustin, CA) and observed with an Etec Autoscan (Perkin Elmer Electron Beam Div., Hayward, CA) scanning electron microscope, operating at an accelerating voltage of 2.5kV. Polaroid PN 55 film (Polaroid Corp., Cambridge, MA) was used for photomicrographs.

#### Leaf Percentage

Leaf percentages were determined for the August, September, and October 1985 harvests, and the May 1986 harvest. Twenty-five stems,

with three to four buds per stem, were randomly selected from each treatment replication. After being rinsed gently with water to remove soil, leaves were separated from stems. Leaflets and petioles composed the leaf fraction, whereas the stem fraction included primary stems, axillary branches, and stipules. Buds were discarded. Samples were oven dried at 60°C and then weighed. The LF was calculated as leaf DM/(leaf DM + stem DM).

#### **In Vitro Dry Matter Disappearance**

Plant materials with three to four buds per stem were selected from each treatment replication, and all subsequent analyses were run only on this phenological stage. *Medicago prostrata* materials used for analyses were at full-bloom stage of maturity because of their indeterminate flowering. October-harvested materials used for laboratory analyses were at the one bud per stem stage of maturity. Samples were oven-dried at 60°C and ground to pass a 1 mm screen with a Wiley Mill (Arthur A. Thomas Co., Philadelphia, PA). A modified Tilley and Terry technique (25) was used to determine IVDMD. Inoculated substrates were centrifuged in incubation chambers after 3, 6, 12, 24, and 48 h of fermentation, and supernatant was discarded. A 24-h acid-pepsin digestion followed each fermentation period. Samples were run in duplicate. The IVDMD of field replications within and among harvests were determined separately. Ruminant fluid was collected from a ruminally cannulated steer fed a diet of 2.27 kg alfalfa hay and .91 kg mixed concentrate (corn 49.15%, grain sorghum 49.15%, dicalcium phosphate 1.0%, trace minerals .5%, and vitamins A, D, and E .2%) twice daily. Ruminant fluid was collected approximately 9 h postfeeding and strained through four layers of cheese cloth.

#### **Crude Protein Concentration**

Nitrogen was determined colorimetrically following a  $\text{H}_2\text{SO}_4\text{:H}_2\text{O}_2$  digestion (24). The CP concentration was calculated as percent N  $\times$  6.25.

#### **Statistical Analysis**

Analysis of variance procedures, followed by protected Least Significant Difference tests for mean separation, were used to analyze data from each IVDMD time period, CP, and LF (17). Arcsine-square root transformations were

made on all percentage data (19). Because conclusions were similar for transformed and original data, we present the original data.

## **RESULTS**

#### **Scanning Electron Microscopy**

Simple and procumbent-glandular hairs (Figures 1, 2, 3, and 4) were present on all the test entries. These two types of hairs are present in varying densities on the alfalfa cultivars currently being used for hay and pasture. In addition, erect glandular hairs (Figures 5 and 6) were present on the (+) entries of MP, KS94, KS108, KS159, and KS160. These erect glandular hairs provide self-defense against insects. High magnifications (Figures 2, 4, 6) illustrate the morphological differences among these trichomes.

#### **Leaf Percentage**

Populations differed significantly within each of the four harvests for which leaves and stems were separated. However, the populations  $\times$  harvest interactions were significant, so the data are presented for each cutting (Table 1). In general, the LF of above ground DM was lower in May than during August, September, and October. This was especially true for the diploids, MP (+) and (−), KS94 (+) and (−), and MS2n (Table 1). Most differences between (+) and (−) entries for each populations were nonsignificant in each of the four cuttings. Only six of 18 comparisons between corresponding (+) and (−) entries were significantly different, with (+) and (−) each being favored three times. None of the tetraploid (+) entries had significantly lower LF than Riley. The diploid entries, MS2n, KS94, and MP, had lower LF than Riley for the May harvest.

#### **In Vitro Dry Matter Disappearance**

Populations differing only for (+) or (−) seldom differed for any digestion period for harvests during the warm periods of growth (July and August) (Tables 2 and 3). In all cases in which differences were significant, IVDMD was higher for (+) than for (−) populations. July-harvested KS159 (+) had higher IVDMD than Riley at all periods except 3-h fermentation. The wild diploid MS2n was generally lower in IVDMD than MS4n.

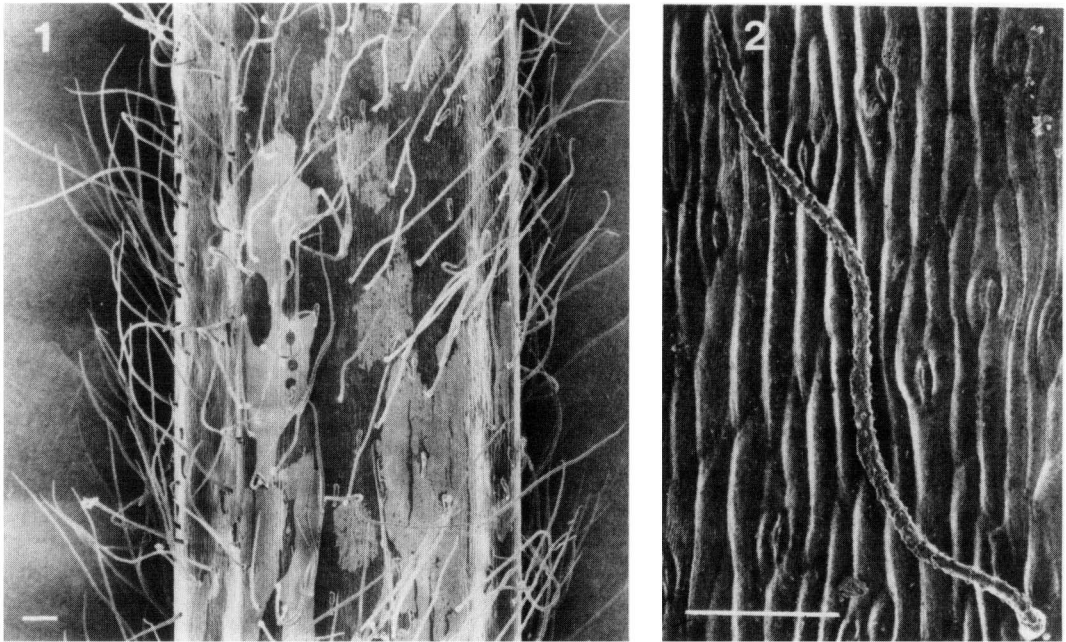


Figure 1. Simple hairs on alfalfa.  $\times 40$ ; bar = 100  $\mu\text{m}$ .

Figure 2. Simple hair on alfalfa.  $\times 200$ ; bar = 100  $\mu\text{m}$ .

TABLE 1. Leaf percentage of above-ground dry matter from four harvests in 1985 and 1986 of 12 *Medicago* populations.

Entry <sup>1</sup>	Aug 1985	Sep 1985	Oct 1985	May 1986
	(% leaf of DM)			
MP(+)	60.7 <sup>a</sup>	54.6 <sup>bcd</sup>	...	41.8 <sup>fg</sup>
MP(-)	59.2 <sup>ab</sup>	55.7 <sup>abc</sup>	...	46.8 <sup>de</sup>
KS94(+)	53.0 <sup>d</sup>	58.1 <sup>a</sup>	...	44.1 <sup>ef</sup>
KS94(-)	56.9 <sup>bc</sup>	51.3 <sup>ef</sup>	...	43.4 <sup>ef</sup>
MS2n	48.5 <sup>e</sup>	50.4 <sup>f</sup>	...	39.0 <sup>g</sup>
Riley	55.3 <sup>cd</sup>	52.8 <sup>cdef</sup>	58.8 <sup>b</sup>	48.9 <sup>cd</sup>
KS108(+)	58.0 <sup>abc</sup>	56.3 <sup>ab</sup>	60.5 <sup>b</sup>	53.3 <sup>ab</sup>
KS108(-)	57.2 <sup>abc</sup>	52.6 <sup>def</sup>	59.6 <sup>b</sup>	51.1 <sup>bc</sup>
KS159(+)	57.8 <sup>abc</sup>	52.4 <sup>def</sup>	60.4 <sup>b</sup>	49.0 <sup>cd</sup>
KS159(-)	59.1 <sup>ab</sup>	54.1 <sup>bcde</sup>	60.6 <sup>b</sup>	48.8 <sup>cd</sup>
KS160(+)	55.0 <sup>cd</sup>	55.3 <sup>abcd</sup>	59.0 <sup>b</sup>	55.2 <sup>a</sup>
KS160(-)	55.5 <sup>cd</sup>	56.0 <sup>ab</sup>	63.6 <sup>a</sup>	50.0 <sup>bcd</sup>
Mean	56.3	54.1	60.4	47.6
SE	2.4	2.1	1.3	2.6

a,b,c,d,e,f,g Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup>MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* ssp. *caerulea*, Riley = *M. sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa*  $\times$  *M. prostrata*, KS160 = *M. sativa*  $\times$  *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

<sup>2</sup>Diploid populations were not harvested in October.

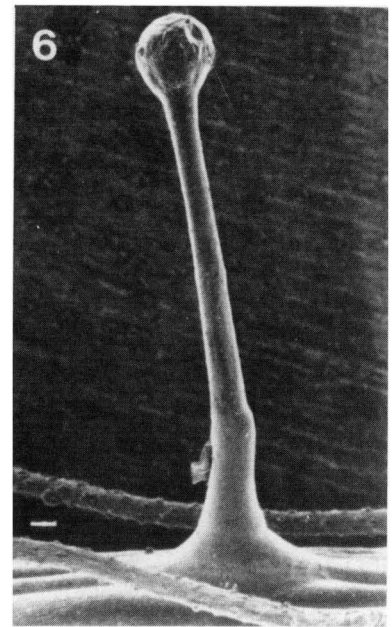
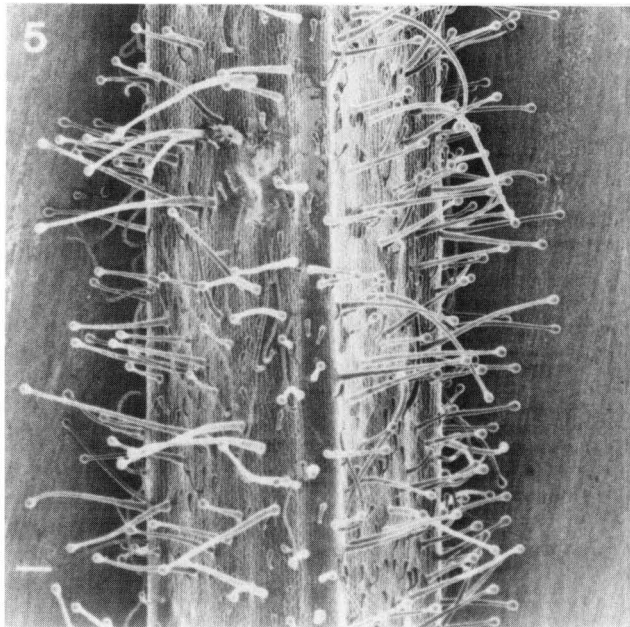
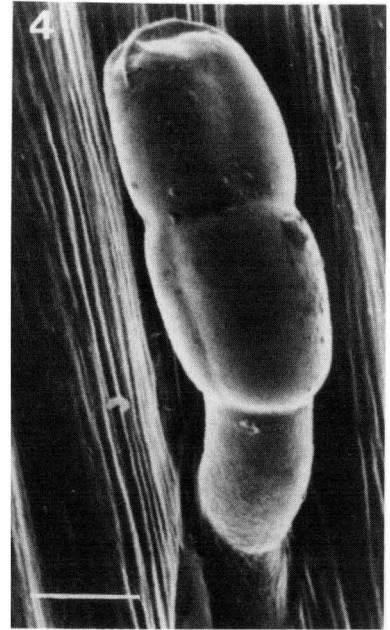
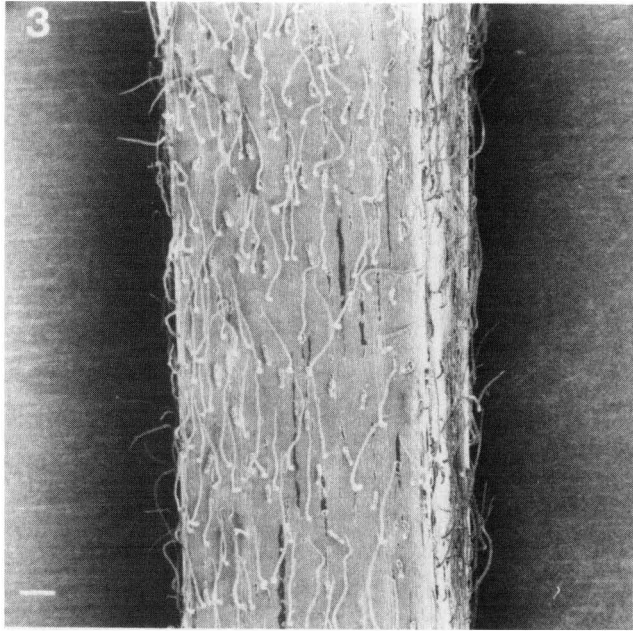


Figure 3. Procumbent glandular and simple hairs on alfalfa.  $\times 40$ ; bar = 100  $\mu\text{m}$ .

Figure 4. Procumbent glandular hair on alfalfa.  $\times 1400$ ; bar = 10  $\mu\text{m}$ .

Figure 5. Erect glandular hairs on *Medicago glandulosa*  $\times 40$ ; bar = 100  $\mu\text{m}$ .

Figure 6. Erect glandular hair on *M. glandulosa*  $\times 300$ ; bar = 10  $\mu\text{m}$ .

TABLE 2. In vitro dry matter disappearance of 12 *Medicago* populations at five times, July 1985 harvest.

Entry <sup>1</sup>	3 h	6 h	12 h	24 h	48 h
	(% in vitro dry matter disappearance)				
MP(+)	45.3 <sup>a</sup>	52.3 <sup>a</sup>	59.7 <sup>ab</sup>	63.5 <sup>ab</sup>	68.8 <sup>ab</sup>
MP(-)	43.3 <sup>ab</sup>	51.0 <sup>abc</sup>	58.5 <sup>bcd</sup>	62.2 <sup>bcd</sup>	68.0 <sup>abcd</sup>
KS94(+)	40.4 <sup>c</sup>	48.7 <sup>de</sup>	57.3 <sup>bcd</sup>	61.1 <sup>d</sup>	66.2 <sup>ef</sup>
KS94(-)	41.8 <sup>bc</sup>	50.0 <sup>cde</sup>	56.2 <sup>d</sup>	61.0 <sup>d</sup>	65.2 <sup>f</sup>
MS2n	42.0 <sup>bc</sup>	48.4 <sup>e</sup>	57.1 <sup>cd</sup>	61.4 <sup>cd</sup>	66.9 <sup>cde</sup>
Riley	40.1 <sup>c</sup>	49.8 <sup>cde</sup>	58.5 <sup>bcd</sup>	63.0 <sup>abc</sup>	66.6 <sup>def</sup>
KS108(+)	41.4 <sup>bc</sup>	50.0 <sup>cde</sup>	58.8 <sup>bc</sup>	63.4 <sup>ab</sup>	68.3 <sup>abc</sup>
KS108(-)	42.0 <sup>bc</sup>	51.3 <sup>abc</sup>	58.9 <sup>bc</sup>	63.9 <sup>ab</sup>	67.8 <sup>bcde</sup>
KS159(+)	43.2 <sup>ab</sup>	51.9 <sup>ab</sup>	61.5 <sup>a</sup>	64.6 <sup>a</sup>	69.5 <sup>a</sup>
KS159(-)	41.6 <sup>bc</sup>	50.0 <sup>cde</sup>	58.0 <sup>bcd</sup>	62.6 <sup>bcd</sup>	67.1 <sup>bcde</sup>
KS160(+)	41.9 <sup>bc</sup>	49.5 <sup>cde</sup>	58.4 <sup>bcd</sup>	63.3 <sup>ab</sup>	67.5 <sup>bcde</sup>
KS160(-)	40.0 <sup>c</sup>	50.4 <sup>bcd</sup>	58.3 <sup>bcd</sup>	63.7 <sup>ab</sup>	66.8 <sup>cdef</sup>
Mean	41.9	50.2	58.5	62.8	67.4
SE	1.7	1.3	1.6	1.1	1.1

a,b,c,d,e,f Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup>MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* ssp. *caerulea*, Riley = *M. sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

TABLE 3. In vitro dry matter disappearance of 12 *Medicago* populations at five times, August 1985 harvest.

Entry <sup>1</sup>	3 h	6 h*	12 h	24 h	48 h
	(% in vitro dry matter disappearance)				
MP(+)	41.6 <sup>a</sup>	47.1 <sup>a</sup>	58.7 <sup>ab</sup>	62.8 <sup>bcd</sup>	67.8 <sup>ab</sup>
MP(-)	40.7 <sup>ab</sup>	46.0 <sup>a</sup>	56.7 <sup>bcd</sup>	62.1 <sup>d</sup>	66.6 <sup>bc</sup>
KS94(+)	38.4 <sup>cd</sup>	44.7 <sup>ab</sup>	56.3 <sup>cd</sup>	62.3 <sup>cd</sup>	66.3 <sup>bc</sup>
KS94(-)	39.0 <sup>bcd</sup>	44.8 <sup>ab</sup>	56.5 <sup>cd</sup>	62.6 <sup>bcd</sup>	66.5 <sup>bc</sup>
MS2n	37.0 <sup>cd</sup>	42.3 <sup>b</sup>	54.9 <sup>d</sup>	60.3 <sup>e</sup>	65.4 <sup>c</sup>
Riley	38.5 <sup>cd</sup>	45.8 <sup>a</sup>	57.8 <sup>abc</sup>	63.4 <sup>abcd</sup>	67.2 <sup>ab</sup>
KS108(+)	41.1 <sup>a</sup>	46.9 <sup>a</sup>	59.0 <sup>a</sup>	65.0 <sup>a</sup>	68.7 <sup>a</sup>
KS108(-)	40.7 <sup>ab</sup>	45.4 <sup>a</sup>	58.3 <sup>abc</sup>	63.5 <sup>abcd</sup>	66.9 <sup>bc</sup>
KS159(+)	39.8 <sup>abc</sup>	46.2 <sup>a</sup>	58.6 <sup>ab</sup>	64.1 <sup>ab</sup>	67.3 <sup>ab</sup>
KS159(-)	40.8 <sup>ab</sup>	46.6 <sup>a</sup>	58.7 <sup>ab</sup>	63.8 <sup>abc</sup>	67.4 <sup>ab</sup>
KS160(+)	39.0 <sup>bcd</sup>	46.1 <sup>a</sup>	57.8 <sup>abc</sup>	63.0 <sup>bcd</sup>	66.9 <sup>bc</sup>
KS160(-)	38.0 <sup>cd</sup>	45.3 <sup>a</sup>	57.7 <sup>bc</sup>	63.2 <sup>bcd</sup>	66.7 <sup>bc</sup>
Mean	39.6	45.6	57.6	63.0	67.0
SE	1.4	1.8	1.4	1.1	1.1

a,b,c,d,e Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup>MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* ssp. *caerulea*, Riley = *M. sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

\* Significant at  $P = .056$ .

Numerous differences between corresponding (+) and (–) populations were found for the September harvest (Table 4). The wild (+) populations of KS94 and KS108 had higher IVDMD than their corresponding (–) populations at all fermentation times, whereas the hybrid (+) populations of KS159 and KS160 generally had lower IVDMD than their corresponding (–) populations for most fermentation times. The (+) populations of the wild diploids MP and KS94 had higher IVDMD than Riley at most time periods, and eglandular MS2n generally tended higher than Riley but the differences were not significant. There were no (+) populations that had significantly lower IVDMD than Riley at any period.

The presence of erect, glandular hairs on KS108 or hybrid populations, KS159 and KS160, did not influence IVDMD of materials harvested during cooler October weather (Table 5). IVDMD of (+) was higher than that of (–) when differences existed between them. No (+) entries were significantly lower in IVDMD than Riley for the October harvest.

For the May harvest, presence of erect, glandular hairs did not greatly influence IVDMD (Table 6). No differences occurred between corresponding (+) and (–) populations, except for KS108, for which the (+) entry had significantly higher IVDMD for nearly all periods. The (+) diploid entries, MP and KS94, and eglandular MS2n had lower IVDMD than Riley in the few cases when mean separations were significant.

#### Crude Protein Concentration

The population × harvest interaction for CP was significant, as were differences among entries when data were analyzed within harvests (Table 7). The presence of erect, glandular hairs usually did not affect the CP content of forage, but when significant differences occurred between corresponding (+) and (–) entries, the (+) populations generally had higher CP. Results of mean separations were similar when (+) entries were compared to Riley. Although few differences occurred, the majority were in favor of (+) entries (Table 7).

TABLE 4. In vitro dry matter disappearance of 12 *Medicago* populations at five times, September 1986 harvest.

Entry <sup>1</sup>	3 h	6 h	12 h	24 h	48 h
	(% in vitro dry matter disappearance)				
MP(+)	42.2ab	50.3ab	59.3b	64.2ab	68.7a
MP(–)	40.2cd	49.0abcd	58.5bc	63.4bc	69.3a
KS94(+)	42.6a	50.5a	60.9a	65.5a	68.9a
KS94(–)	38.3d	47.2cdef	57.0def	61.9cdefg	65.6cd
MS2n	40.2cd	48.2cde	57.6cde	63.2bcd	67.0b
Riley	41.0abc	47.5cdef	56.4efg	61.5efg	65.3cd
KS108(+)	42.7a	48.2bcde	58.4bcd	63.5bc	66.4bc
KS108(–)	40.1cd	45.0g	55.4gh	61.6defg	64.5de
KS159(+)	40.5bc	46.9efg	55.4fgh	60.8fg	64.5de
KS159(–)	42.3ab	49.1abc	58.2bcd	62.9de	66.3bc
KS160(+)	39.8cd	45.7fg	54.4h	60.5g	63.8e
KS160(–)	40.4bc	46.9defg	56.1efg	62.4cdef	65.2cd
Mean	40.9	47.9	57.3	62.6	66.3
SE	1.2	1.2	0.9	1.0	0.8

a,b,c,d,e,f,g,h Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup>MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* ssp. *caerulea*, Riley = *M. sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (–) = absence of erect glandular hairs.



TABLE 5. In vitro dry matter disappearance of seven *Medicago* populations at five times, October 1985 harvest.

Entry <sup>1</sup>	3 h	6 h	12 h	24 h	48 h
	(% in vitro dry matter disappearance)				
Riley	46.3 <sup>bc</sup>	54.8 <sup>bc</sup>	63.1 <sup>abc</sup>	69.4 <sup>abc</sup>	72.5 <sup>ab</sup>
KS108(+)	47.4 <sup>ab</sup>	55.1 <sup>abc</sup>	64.2 <sup>a</sup>	69.9 <sup>ab</sup>	73.0 <sup>a</sup>
KS108(-)	48.2 <sup>ab</sup>	57.0 <sup>a</sup>	64.6 <sup>ab</sup>	70.4 <sup>a</sup>	73.4 <sup>a</sup>
KS159(+)	48.7 <sup>a</sup>	56.0 <sup>ab</sup>	63.4 <sup>abc</sup>	69.7 <sup>ab</sup>	72.2 <sup>ab</sup>
KS159(-)	46.6 <sup>ab</sup>	53.9 <sup>c</sup>	62.7 <sup>bc</sup>	68.6 <sup>bc</sup>	71.8 <sup>ab</sup>
KS160(+)	46.3 <sup>bc</sup>	54.2 <sup>bc</sup>	62.4 <sup>c</sup>	68.0 <sup>cd</sup>	71.0 <sup>bc</sup>
KS160(-)	44.2 <sup>c</sup>	50.7 <sup>d</sup>	60.1 <sup>d</sup>	66.8 <sup>d</sup>	70.0 <sup>c</sup>
Mean	46.8	54.5	62.9	69.0	72.0
SE	1.5	1.3	1.2	1.1	1.1

a,b,c,d Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup> Riley = *Medicago sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

### DISCUSSION

Several investigators have demonstrated that morphological characteristics of plants affect their nutritive value (6). For example, simple hairs on fresh leaf sections of pearl millet

[*Pennisetum americanum* (L.) K. Schum.] increased IVDMD but, when ground samples were analyzed, digestibilities of haired and hairless lines were equal (9). The authors hypothesized that the trichomeless line had

TABLE 6. In vitro dry matter disappearance of 12 *Medicago* populations at five times, May 1986 harvest.

Entry <sup>1</sup>	3 h	6 h	12 h	24 h	48 h
	(% in vitro dry matter disappearance)				
MP(+)	37.0 <sup>bc</sup>	42.7 <sup>def</sup>	52.2 <sup>de</sup>	59.5 <sup>f</sup>	63.0 <sup>e</sup>
MP(-)	36.3 <sup>cd</sup>	43.1 <sup>de</sup>	52.6 <sup>de</sup>	59.6 <sup>f</sup>	62.9 <sup>e</sup>
KS94(+)	35.2 <sup>cd</sup>	40.8 <sup>f</sup>	52.1 <sup>de</sup>	61.0 <sup>ef</sup>	65.5 <sup>cd</sup>
KS94(-)	34.3 <sup>d</sup>	40.9 <sup>f</sup>	51.7 <sup>e</sup>	60.8 <sup>ef</sup>	65.6 <sup>cd</sup>
MS2n	35.2 <sup>cd</sup>	41.8 <sup>ef</sup>	52.4 <sup>de</sup>	62.3 <sup>bcd</sup>	66.0 <sup>bcd</sup>
Riley	36.5 <sup>c</sup>	44.5 <sup>bcd</sup>	54.1 <sup>cd</sup>	62.6 <sup>bcd</sup>	66.1 <sup>bcd</sup>
KS108(+)	40.5 <sup>a</sup>	46.7 <sup>a</sup>	57.7 <sup>a</sup>	65.6 <sup>a</sup>	69.4 <sup>a</sup>
KS108(-)	38.8 <sup>ab</sup>	44.1 <sup>bcd</sup>	55.0 <sup>bc</sup>	63.1 <sup>bcd</sup>	67.2 <sup>bc</sup>
KS159(+)	36.2 <sup>cd</sup>	42.0 <sup>ef</sup>	52.3 <sup>de</sup>	61.2 <sup>def</sup>	65.2 <sup>d</sup>
KS159(-)	37.0 <sup>bc</sup>	43.3 <sup>cde</sup>	53.3 <sup>cde</sup>	61.9 <sup>cde</sup>	65.6 <sup>cd</sup>
KS160(+)	39.4 <sup>a</sup>	45.9 <sup>ab</sup>	56.7 <sup>ab</sup>	64.1 <sup>ab</sup>	67.6 <sup>ab</sup>
KS160(-)	39.3 <sup>a</sup>	45.3 <sup>abc</sup>	55.4 <sup>bc</sup>	63.7 <sup>abc</sup>	67.0 <sup>bcd</sup>
Mean	37.1	43.4	53.8	62.1	65.9
SE	1.6	1.4	1.5	1.5	1.3

a,b,c,d,e,f Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup> MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* subsp. *caerulea*, Riley = *Medicago sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

TABLE 7. Crude protein concentrations of 12 *Medicago* populations from five harvests.

Entry <sup>1</sup>	July 1985	Aug 1985	Sept 1985	Oct 1985	May 1986
	(% CP of DM)				
MP(+)	21.6 <sup>ab</sup>	23.2 <sup>a</sup>	19.2 <sup>de</sup>	...	18.6 <sup>bcde</sup>
MP(-)	20.0 <sup>cd</sup>	21.7 <sup>ab</sup>	17.5 <sup>f</sup>	...	18.5 <sup>cde</sup>
KS94(+)	18.7 <sup>d</sup>	18.9 <sup>c</sup>	19.2 <sup>de</sup>	...	17.8 <sup>e</sup>
KS94(-)	18.7 <sup>d</sup>	20.7 <sup>b</sup>	18.1 <sup>ef</sup>	...	18.4 <sup>de</sup>
MS2n	19.8 <sup>cd</sup>	19.3 <sup>c</sup>	20.9 <sup>bc</sup>	...	20.0 <sup>ab</sup>
Riley	20.1 <sup>bcd</sup>	22.0 <sup>ab</sup>	20.0 <sup>cd</sup>	26.6 <sup>abc</sup>	18.6 <sup>bcde</sup>
KS108(+)	20.2 <sup>bcd</sup>	23.3 <sup>a</sup>	21.9 <sup>a</sup>	28.1 <sup>a</sup>	18.9 <sup>bcde</sup>
KS108(-)	19.8 <sup>cd</sup>	21.5 <sup>ab</sup>	20.1 <sup>cd</sup>	27.4 <sup>ab</sup>	20.0 <sup>abc</sup>
KS159(+)	22.9 <sup>a</sup>	22.0 <sup>ab</sup>	19.9 <sup>cd</sup>	26.2 <sup>bc</sup>	20.6 <sup>a</sup>
KS159(-)	20.4 <sup>bc</sup>	22.0 <sup>ab</sup>	22.6 <sup>a</sup>	22.9 <sup>d</sup>	19.5 <sup>abcd</sup>
KS160(+)	19.9 <sup>cd</sup>	22.0 <sup>ab</sup>	22.0 <sup>ab</sup>	26.6 <sup>abc</sup>	20.6 <sup>a</sup>
KS160(-)	21.0 <sup>bc</sup>	21.7 <sup>ab</sup>	20.6 <sup>bcd</sup>	25.5 <sup>c</sup>	20.5 <sup>a</sup>
Mean	20.3	21.5	20.2	26.2	19.3
SE	1.1	1.2	1.1	1.1	1.0

a,b,c,d,e,f Means within columns with different superscripts differ ( $P < .05$ ).

<sup>1</sup> MP = *Medicago prostrata*, KS94 = *M. glandulosa*, MS2n = *M. sativa* ssp. *caerulea*, Riley = *M. sativa*, KS108 = *M. glutinosa*, KS159 = *M. sativa* × *M. prostrata*, KS160 = *M. sativa* × *M. glutinosa*; (+) = presence of erect glandular hairs, (-) = absence of erect glandular hairs.

<sup>2</sup> Diploids were not harvested in October.

increased surface wax, which reduced microbial entry and delayed degradation of the cell walls. Our study indicated that rate of or total digestibility of forage from *Medicago* species was not affected by erect, glandular hairs. However, palatability studies are needed to determine acceptance of glandular-haired alfalfa by animals. Physical characteristics of forages may favor in vitro digestion but depress voluntary or preferential intake by animals. When animals were given a choice, trichomed millet was less preferred than glabrous types (9).

Anatomical features of forage crops also may alter dietary intake and digestion. Structural components may alter accessibility of cells to microbial attack. Suk-sayretrup (23) found higher lignification of vascular bundles and interfascicular areas in stems of glandular-haired *M. prostrata* than in alfalfa (*M. sativa*). This was corroborated by Brewer et al. (5), who suggested that this highly lignified cylinder surrounding the pith prevented oviposition by the potato leafhopper, providing an effective resistance mechanism. We

found that fiber content and IVDMD of stems and leaves were similar for glandular and eglandular plants selected from each of *M. prostrata*, *M. glandulosa*, and *M. glutinosa* (14, 15). Thus, although content of lignin may be similar, its structural organization may be different in alfalfa and the erect glandular-haired wild species. Apparently, lignin content does not affect rate of digestion in alfalfa (3) or corn (13), but total digestibility decreases as lignin content increases.

Excessive losses in yield and quality of alfalfa forage caused by alfalfa weevil or potato leafhopper are well-documented. Our trials were conducted in the absence of both pests. If host-plant resistance, conferred by the presence of erect, glandular hairs, is satisfactory to control these pests, yield and quality of alfalfa forage could be protected without additional costs to producers.

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