Effects of inoculation of high dry matter alfalfa silage on ensiling characteristics, ruminal nutrient degradability and dairy cow performance

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Abstract: The objective of this study was to determine the effects of a homolactic acid inoculant on ensiling characteristics and nutritive value of high dry matter (DM) alfalfa. The ensiling characteristics were determined by ensiling inoculated and untreated alfalfa haylage in mini-silos for 0, 2, 4, 8, 16 and 45 days. Two lactating cows fitted with ruminal fistulas were used to determine ruminal degradabilities of nutrient in inoculated and untreated alfalfa silage (45-day silage). Effects of feeding inoculated and untreated alfalfa silage on animal performance were determined using 28 lactating dairy cows fed total mixed diets (40% forage and 60% concentrate) with the forage portion consisting of inoculated or untreated alfalfa silage. The pH of the inoculated silage declined from 5.9 to 4.5 within 2 days of ensiling while the pH of the untreated silage did not drop below 5 until day 16 post-ensiling. At all ensiling times, inoculated alfalfa silage had lower pH than untreated alfalfa silage. The concentration of lactic acid was higher while that of water-soluble carbohydrates was lower for inoculated than untreated alfalfa silage at all ensiling times. Inoculation increased proteolysis as indicated by a reduction in true protein and neutral detergent insoluble protein and an increase in non-protein nitrogen. Ruminal degradability of DM, crude protein and neutral detergent fiber of alfalfa silage were not affected by inoculation. The average effective degradability of dry matter, crude protein and neutral detergent fiber was 66, 65 and 62%, respectively. Dairy cows fed inoculated alfalfa silage had DM intake (average 22 kg day−1) and milk yield (average 42 kg day−1) similar to cows fed untreated alfalfa silage. It was concluded that the inoculant used in this study improved the ensiling characteristics of alfalfa silage with no significant effects on dairy cow performance.

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Keywords: alfalfa silage; inoculation; proteolysis; dairy cows

INTRODUCTION
Silage inoculants are usually added to improve silage fermentation and to obtain silage with high nutritive value with minimum dry matter (DM) losses. Studies have shown that inoculants based on homolactic acid bacteria can improve silage fermentation characteristics. The improved fermentation was evident by a rapid decline in pH, high concentrations of lactic acid and low ammonia nitrogen levels.1–3 The rapid decline in pH is crucial in minimizing nutrient losses by reducing rate of proteolysis during early stages of ensiling.4 The activity of bacteria used in silage inoculants is largely dependent on moisture content of the ensiled forages.5 The term ‘water activity’ is used to describe more accurately the amount of moisture available for microbial growth during ensiling.5 Forages with high DM content (ie low water activity) ferment at slower rates than forages with low DM content because low water activity reduces microbial growth.2,3

Alfalfa is the main legume forage in eastern Canada and is usually wilted to DM higher than 30% to reduce the risk of clostridial fermentation.2 High DM alfalfa (54% DM) treated with a single strain of Lactobacillus plantarum had a lower pH and a higher lactic acid concentration than untreated alfalfa after 2 days of ensiling.2 However, effects of inoculants containing more than one strain of lactic acid bacteria on fermentation of high DM alfalfa have not been determined. Furthermore, information on the effects of such inoculants on animal performance is not available. The objectives of this study were to determine the effects of inoculation with multiple strains of Lactobacillus plantarum on proteolytic activities and in situ ruminal degradability of high DM alfalfa silage and to determine the effects

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of feeding inoculated high DM alfalfa silage on the performance of dairy cows.

**MATERIALS AND METHODS**

**Silage preparation**
A second-cut alfalfa (Medicago sativa) was harvested on 6 July 2002 at early bloom stage of maturity and wilted for 24 h. The wilted forage was chopped to a theoretical cut length of 0.95 cm using a flail forage harvester. Lactic acid bacteria inoculum (11H50 Sila-Bac; Pioneer Hi-Bred Inc, IA, USA) containing multiple stains of Lactobacillus plantarum was applied (liquid application) to portions of the chopped forages following the recommendations of the manufacturers to supply 10^5 CFU g^-1 of ensiled material. Alternate loads of the forage were either inoculated or left untreated at the silo blower as the herbage was being uploaded. The inoculated and untreated forages were ensiled in two separate, upright concrete towers-silos (capacity of 100 tons DM each) for 2 months.

**Effects of inoculation on ensiling characteristics**
Effect of inoculation on ensiling characteristics of alfalfa silage was determined using mini-silos. Representative herbage samples (1000 g) of inoculated and untreated alfalfa silage were collected manually using a pestle in triplicate, into mini-silos made of PVC tubing (7.6 cm diameter and 25 cm height; capacity 1 kg). The filled silos were sealed with plastic lids equipped with gas valves and stored at ambient temperature and allowed to ensile for 2, 4, 8, 16 and 45 days. Triplicate samples of fresh forage (0 day after ensiling) from each alfalfa silage treatment were also stored at −20°C for later analysis. Dry matter recoveries for the 45-day silages were estimated by weighing the mini-silo before and after the 45-day ensiling period.

**Laboratory chemical analyses**
After the designated ensiling time, the mini-silos were opened and the ensiled forage was mixed thoroughly. The fresh and the ensiled forages (25 g) were homogenized for 1 h in 250 ml of distilled water. The pH of the water extract was immediately determined using an Accumet pH meter (Denver Instrument Company, Mansfield, TX, USA). A portion of the extract (20 ml) was filtered through a Whatman 54 filter paper, acidified with 50 μl of 50% H₂SO₄ and frozen before further analysis. Lactic acid and WSC were determined following the procedure of Barker and Summerson and Dubois et al., respectively. Subsamples (500 g) of the fresh (0 day) and the ensiled forages (2, 4, 8, 16 and 45 days) were also dried in a forced-air oven at 55°C for 48 h and then ground through a 2-mm screen using a Wiley Mill and composited to obtain a single sample of each treatment. Quadruplicate samples, weighing approximately 5 g (air-dry basis), of each silage treatment were weighed into nylon bags (25 × 33 cm, 50-μm pore size, ANKOM Technology, Fairport, NY, USA). Duplicate bags from each silage treatment were incubated in the ventral sac of the rumen of the two cows for 3, 6, 12, 24, 48, 72 and 96 h. Following removal from the rumen, bags were washed in at tap water 20°C until the runoff water was clear. The 0-h washout was measured by washing duplicate bags containing samples of the two silage treatments. Washed bags were dried at 55°C for 48 h in a forced-air oven.

**In situ ruminal degradability**

The effects of inoculation on ruminal kinetic parameters and ruminal degradability of alfalfa silage were determined with the nylon bag technique. Two lactating Holstein cows fitted with flexible ruminal fistulas were used. The cows were fed ad libitum 50:50 forage: concentrate diet (DM basis). The mixed diet contained (DM basis) 17.6% CP, 32.6% NDF and 2.9% ether extract. Animals were fed in equal portions at 08:00 and 16:00 h and had free access to water.

Untreated and inoculated dried alfalfa silages obtained from the 45-day mini-silos were used. Subsamples (n = 3) of inoculated and untreated alfalfa silages were ground through a 2-mm screen using a Wiley Mill and composited to obtain a single sample of each treatment. Quadraplicate samples, weighing approximately 5 g (air-dry basis), of each silage treatment were weighed into nylon bags (25 × 33 cm, 50-μm pore size, ANKOM Technology, Fairport, NY, USA). Duplicate bags from each silage treatment were incubated in the ventral sac of the rumen of the two cows for 3, 6, 12, 24, 48, 72 and 96 h. Following removal from the rumen, bags were washed in at tap water 20°C until the runoff water was clear. The 0-h washout was measured by washing duplicate bags containing samples of the two silage treatments. Washed bags were dried at 55°C for 48 h in a forced-air oven.

Residues from the nylon bags at each incubation time were analyzed for moisture, CP and NDF as previously described. Ruminal disappearance at each incubation time was calculated from nutrient concentration in the original samples and the ruminal residues. Ruminal disappearance data were used to estimate DM, CP and NDF degradation parameters using the equation of Dhanoa:

\[
P = a + b \times \left(1 - e^{-ct(1-L)}\right)
\]

where \(P\) is ruminal disappearance at time \(t\) (%), \(a\) is the soluble fraction (%), \(b\) is the slowly degradable fraction (%), \(c\) is the rate at which the \(b\) fraction is degraded (% h⁻¹) and \(L\) is a discrete lag phase. Effective ruminal degradability (ED) of DM, CP and
NDF were estimated using the equation of Ørskov and McDonald:12

\[ ED = a + ((b \times c)/(c + k)) \]

where \( k \) is the estimated ruminal flow rate of 5.0% h\(^{-1}\).

**Dairy production study**

Untreated and inoculated alfalfa silage stored in the upright silos were used in this study. Two isonitrogenous and isocaloric diets were formulated to meet the requirements of dairy cows in early lactation\(^1\) (Table 1). The diets were formulated as total mixed diets with 40% forage and 60% concentrate. In both diets, untreated or inoculated alfalfa silage was the only source of forage. Alfalfa silage DM was determined weekly and diet formulations were adjusted accordingly to account for changes in DM levels. Diets were offered twice daily (08:00 and 16:00 h) and feed quantity was adjusted every 2 days to allow weigh-back of 5–10% of intake.

Twenty-eight lactating Holstein cows of mixed parities (16 multiparous cows, 614 kg \( \pm \) 47.7 and 12 primaparous cows, 544 kg \( \pm \) 29.4) were used. Cows were blocked by parity and milk yield at calving and randomly assigned to one of the two dietary treatments for 10-week period. Cows were housed in tie stalls and had continuous access to water. Cows were milked three times daily at 04:00, 12:00 and 18:00 h and milk yield was recorded at each milking. The quantities of feed offered and refusals were measured daily for each cow to determine daily feed intake. Milk yields were recorded at each milking and milk samples were collected once weekly from the three milkings for milk component analysis.

Samples of total mixed diets and silages were collected weekly, dried for 48 h at 55°C in a forced-air oven and pooled monthly. Samples were later ground through a 1-mm screen before chemical analysis. Ground samples of total mixed diets and alfalfa silages were analyzed for DM, ash, NDF, ADF and CP as previously described. Milk samples were analyzed for fat, protein, lactose, milk urea nitrogen and somatic cell count at the Dairy Herd Analysis Service (Programme d’analyse des troupeaux laitiers du Quebec) with an infrared system using an electric Milk-O-Scan 4000 (Foss-Food technology, Hillerød, Denmark) calibrated with reference standards determined by the Mojonnier and Kjeldahl methods.9 Fat, protein and lactose yield were also calculated by multiplying milk yield and the concentration of the corresponding parameters. Daily milk yield and daily DM intake were averaged every week and weekly values were obtained.

**Table 1.** Ingredients and chemical composition of diets in the dairy cow trial

<table>
<thead>
<tr>
<th>Alfalfa silage treatment</th>
<th>Untreated</th>
<th>Inoculated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diet ingredients (% of DM)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Untreated alfalfa haylage</td>
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<td>40.0</td>
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<tr>
<td>Inoculated alfalfa haylage</td>
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<td></td>
</tr>
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<td>High moisture corn</td>
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<td>45.6</td>
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<td>Soybean meal</td>
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<tr>
<td>Beet pulp</td>
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<td>2.0</td>
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<tr>
<td>Commercial dairy supplement(^a)</td>
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<td>3.0</td>
</tr>
<tr>
<td>Commercial fat supplement</td>
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<td>2.0</td>
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<tr>
<td>Sodium bicarbonate</td>
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<td>0.8</td>
</tr>
<tr>
<td>Mineral–vitamin premix(^b)</td>
<td>0.5</td>
<td>0.5</td>
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<tr>
<td>Chemical composition (% of DM)</td>
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<td>Dry matter</td>
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<tr>
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<tr>
<td>Acid detergent fibre</td>
<td>27.5</td>
<td>26.7</td>
</tr>
</tbody>
</table>

\(^a\) Contained (% DM basis): 50% CP, 4% crude fat, 5% crude fiber, 2.5% Ca, 1.5% P, 0.8% Mg, 0.5% S, 0.5% K. Supplied (per kg) 160 mg vitamin E, 39 500 IU vitamin A, 11 850 IU vitamin D\(_3\).

\(^b\) Contained (%) 10 Ca, 10 P, 7.8 Na, 8.0 Mg, 3.0 S. Supplied 45 mg I, 3600 mg Fe, 740 Cu, 2300 mg Mn, 2380 mg Zn, 10 mg Co, 500 mg F, 300 000 IU vitamin A, 80 000 IU vitamin D\(_3\), 800 IU vitamin E per kg.

\[ Y_{ijklm} = \mu + \text{week}_j + \text{diet}_i + \text{parity}_k + \text{block}_l + \text{diet} \times \text{week}_j + \text{diet} \times \text{parity}_k + \text{parity} \times \text{week}_j + \text{parity} \times \text{diet} \times \text{week}_j + e_{ijklm} \]

\( Y \): variable studied during the week ‘j’ (1,2...), diet ‘i’ (1 and 2) for the ‘m’th cow of parity ‘k’ and block ‘l’

\( \mu \): overall mean

week: effect of the jth week
diet: effect of the ith diet
parity: effect of the kth parity
block: effect of the lth block
e: random error of the ijklmth measure.
RESULTS AND DISCUSSION

Effects of inoculation on ensiling characteristics of alfalfa silage

The pH of the inoculated alfalfa silage declined faster ($p < 0.05$) than the pH of the untreated silage (Fig 1). The inoculated alfalfa silage reached a pH of 4.5 on day 2 post-ensiling while it took 45 days for the untreated alfalfa silage to reach a similar value. The pH of the inoculated alfalfa silage showed little changes between day 4 and day 45, but remained significantly lower ($p < 0.05$) than the pH of untreated alfalfa silage at all ensiling times. The pH of the untreated silage continued to decline up to day 45 post-ensiling. Lactic acid concentration was higher ($p < 0.05$) for inoculated than for untreated alfalfa silage at all ensiling times (Fig 1). Lactic acid content of inoculated alfalfa silage on day 2 post-ensiling was again similar to the value reached at day 45 for the untreated silage. For both silages, the lactic acid content continued to rise up to day 45 post-ensiling. Water-soluble carbohydrates was lower ($p < 0.05$) for inoculated than for control alfalfa silage at all ensiling times and decreased more rapidly ($p < 0.05$) during the first 2 days of ensiling for inoculated than for untreated silage (Fig 1).

Our results are in good agreement with other studies which showed higher lactic acid concentrations and lower pH for inoculated than untreated silages. Our results also support the findings of Whiter and Kung who found that inoculation could improve fermentation of high DM alfalfa silage. The authors studied the effects of inoculation with *Lactobacillus plantarum* MTD1 (either in a dry or liquid form) on fermentation of alfalfa wilted at 30 and 54% DM and found that in silages with 30% DM, both forms of inoculation resulted in silages with more lactic acid and a lower pH than untreated silages after 2 days of ensiling. In silages containing 54% DM, dry and liquid inoculation produced more rapid decrease in pH between day 4 and day 14 when compared with untreated silage, but the effect was greater when the inoculant was applied in water.

The slow fermentation of untreated alfalfa silage in our study is probably due to its high DM content. It has been suggested that, as the DM content of ensiled forages increases, the number of lactic acid producing bacteria decreases because low water activity restricts bacterial growth. Woolford attributed the high pH values in high DM silages to the increased osmotic pressure, which inhibits the growth of lactic acid bacteria. Low lactic acid bacterial population may explain the higher water soluble carbohydrate concentrations observed for untreated relative to inoculated alfalfa silage (Fig 1).

Effects of inoculation on protein fractions of alfalfa silage

Soluble protein and NPN increased ($p < 0.05$) rapidly from day 0 to day 4 (more than 53% increase in NPN and SCP) in the inoculated alfalfa silage with little changes in SCP and NPN between day 8 and day 45 post-ensiling (Fig 2). The untreated silage had a more gradual rise in NPN and SCP from day 0 to day 16 post-ensiling (42% increase in NPN and SCP) followed by a slow increase from day 16 to day 45 post-ensiling. Soluble protein and NPN content of inoculated alfalfa silage were higher ($p < 0.05$) than untreated alfalfa silage at all ensiling times (except for day 0). Changes in NPN and SCP during ensiling followed a common pattern. At any ensiling time, NPN constituted most of SCP for both inoculated and untreated silage (88.8 to 90.3%). Studies on the effects of inoculation on protein fractions of alfalfa silage are rather limited. In a similar study with high DM alfalfa (50% DM), Phillip et al reported a trend for higher

Figure 1. Effects of inoculation on pH, lactic acid and water-soluble carbohydrates of high dry matter alfalfa silage.
Effect of inoculation on alfalfa silage quality

Figure 2. Effects of inoculation on protein fractions of high dry matter alfalfa silage.

NPN for inoculated than for untreated alfalfa silages at several ensiling times with a significantly higher NPN for inoculated silage after 21 days of ensiling. Muck found that, of the soluble N fraction, only ammonia was reduced by inoculation. Petit and Flipot found no difference in SCP content between inoculated and untreated alfalfa silage.

Neutral detergent insoluble protein declined \((p < 0.05)\) at a decreasing rate from day 2 to day 45 post-ensiling for the inoculated silage while that of the untreated silage was unaffected by ensiling (Fig 2). Overall, the NDICP of the untreated silage was only reduced by 0.2% after 45 days of ensiling compared with a 4.2% decrease for the inoculated alfalfa silage. Reduction in NDICP reported in this study for inoculated alfalfa silage agrees with a similar drop reported for other legume forages.

For both silages, ADICP decreased \((p < 0.05)\) after 2 days of ensiling, reached a minimum between day 2 and day 4 and rose again between day 4 and day 8 post-ensiling (Fig 2). The ADCIP stabilized after 8 days of ensiling for the inoculated alfalfa. However, it continued to rise until day 45 post-ensiling for the untreated silage. The overall changes in ADICP at between day 0 and day 45 post-ensiling were relatively small (0.5 and 0.7% increase for the inoculated and the untreated alfalfa silage, respectively) and the low ADICP values suggest that neither silages overheated during ensiling.

True protein decreased \((p < 0.05)\) more rapidly for the inoculated than for the untreated alfalfa silage (Fig 2). Inoculated alfalfa silage lost more than 25% of its TP from day 0 to day 4 post-ensiling compared with less than 8.5% for the untreated silage. No further decline in TP was observed between day 8 and day 45 for the inoculated alfalfa silage and between day 16 and day 45 for the untreated alfalfa. At all ensiling times, inoculated alfalfa had a lower \((p < 0.05)\) TP value than...
untreated alfalfa silage. Differences in TP between inoculated and untreated alfalfa can be attributed mainly to differences in other protein fractions (ie SCP, NPN and NDICP).

One of the main objectives of silage inoculation is to increase the acidification rate which, in turn, is expected to limit protein degradation during early stages of fermentation. It is believed that plant enzymes, collectively known as proteases, are responsible for the breakdown of protein to NPN in the early stages of ensiling. The higher proteolytic activity reported in this study for inoculated relative to untreated alfalfa silage was rather unexpected since it is thought that the activity is plant proteases reduced when the pH drops below 5. Factors other than plant proteases can also affect proteolytic activities. Winters et al suggested that proteolysis could be mediated by both plant and microbial activities. They suggested that some heterolactic acid bacteria could have proteolytic activities.

Although inoculation increased rate of proteolysis, differences in SCP, NPN and TP between inoculated and untreated alfalfa at day 45 post-ensiling were small and therefore are not expected to be of great biological significance (Table 2). Our results also suggest that proteolysis was low in both silages. This is mainly due to the high DM content of the silages. Muck found a negative relationship between rate of proteolysis (measured as increase in NPN) and DM content of alfalfa silage. For alfalfa wilted to 50% DM (an initial NPN 25.6% of CP), NPN increased to 64.8% of CP after 60 days of ensiling. When alfalfa was wilted to 40% DM, the final NPN was 82.6% of CP.

Inoculation significantly improved (p < 0.05) DM recovery of alfalfa silage (Table 2). This is an agreement with Cai who found that inoculating silage with lactic acid bacteria reduced dry matter losses.

### Table 2. Chemical composition of untreated and inoculated alfalfa silage (dry matter basis)

| Alfalfa silage | Untreated | Inoculated | SEM
|----------------|-----------|------------|-----
| Dry matter (%) | 55.8      | 54.2       | 0.54 |
| Ash (%) | 9.3       | 9.3        | 0.09 |
| Neutral detergent fibre (%) | 44.0       | 44.0       | 0.47 |
| Acid detergent fibre (%) | 36.0       | 35.9       | 0.55 |
| Acid detergent lignin (%) | 6.6        | 6.7        | 0.07 |
| Crude protein (%) | 21.7       | 21.6       | 0.26 |
| Soluble protein (% of CP) | 53.4b      | 55.0a      | 0.25 |
| Non-protein nitrogen (% of CP) | 47.5b      | 49.7a      | 0.24 |
| Neutral detergent insoluble protein (% of CP) | 14.7a      | 10.5b      | 0.27 |
| Acid detergent insoluble protein (% of CP) | 7.0a       | 6.4b       | 0.14 |
| Dry matter recovery (%) | 92.4b      | 97.3a      | 0.31 |

a Means in the same row with different letters differ significantly (p < 0.05).
b Pooled standard error of the mean.

effects of inoculation on ruminal nutrient degradability

The results of the in situ study are presented in Table 3. Inoculation had no effects on ruminal DM kinetic parameters of the 45-day silages. The average values of soluble DM, slowly degradable DM, degradation rate of the slowly degradable DM and effective DM degradability were 39.4%, 38.7%, 11.6% h⁻¹ and 66.3%, respectively. In accordance with higher NPN content, soluble CP fraction was higher (p < 0.05) for inoculated than untreated alfalfa silage. Inoculation had no significant effect on the in situ slowly degradable CP fraction or on its rate of degradation. Effective ruminal CP degradability was also similar for both silages (average 84.3% of CP) suggesting that the difference in soluble CP fraction was not large enough to affect ruminal CP degradability. Ruminal kinetic parameters and effective degradability of NDF were similar for untreated and inoculated alfalfa silage. Our values for ruminal kinetic parameters and degradabilities are in good agreement with values previously reported for alfalfa silage.

Data on the effects of microbial inoculation on ruminal nutrient degradability of alfalfa silage are limited. In agreement with our findings, Hristov and McAllister found that inoculating barley silage with three different inoculants had no effects on ruminal degradability. However, other studies reported improved ruminal DM degradability of whole-crop barley or grass silage as a result of inoculation. Salawu treated pea/wheat bi-crop with two lactic acid bacteria inoculants and found a decrease in

### Table 3. Ruminal nutrient kinetic parameters and effective degradabilities of inoculated and untreated alfalfa silage

| Alfalfa silage | Untreated | Inoculated | SEM
|----------------|-----------|------------|-----
| Dry matter (DM) | 39.3      | 39.4       | 0.37 |
| Soluble (% of DM) | 39.0      | 38.3       | 0.53 |
| Degradation rate (% h⁻¹) | 11.3      | 11.9       | 0.79 |
| Lag time (h) | 0.2       | 0.1        | 0.06 |
| Effective degradability (% of DM) | 66.3      | 66.2       | 0.71 |
| Crude protein (CP) | 57.8b      | 59.4a      | 0.47 |
| Slowly degradable (% of CP) | 35.5      | 33.0       | 0.97 |
| Degradation rate (% h⁻¹) | 14.3      | 16.2       | 0.73 |
| Lag time (h) | 0.2       | 0.4        | 0.16 |
| Effective degradability (% of CP) | 84.0      | 84.6       | 0.25 |
| Neutral detergent fiber (NDF) | 2.0       | 1.8        | 0.10 |
| Soluble (% of NDF) | 40.5      | 40.3       | 0.40 |
| Slowly degradable (% of NDF) | 6.7       | 6.0        | 0.27 |
| Lag time (h) | 0.2       | 0.3        | 0.19 |
| Effective degradability (% of NDF) | 23.4      | 25.2       | 0.56 |

a Means in the same row with different letters differ significantly (p < 0.05).
b Pooled standard error of the mean.
soluble NDF, an increase in potentially degradable NDF and decrease in ruminal NDF degradability.

**Performance of dairy cows fed inoculated alfalfa silage**

Inoculation had no effect on DM intake of lactating dairy cows (20.2 kg day\(^{-1}\), Table 4). Our results are in accordance with several studies, which found no improvement in DM intake of lactating cows,\(^{30,31}\) lambs\(^{19}\) or heifers\(^{32}\) fed inoculated alfalfa silage. Our results are also in agreement to other studies that showed no effect of inoculated grass\(^{33}\) and barley\(^{27}\) silage on DM intake of beef cattle. However, other researchers have reported improved DM intake when inoculated grass\(^{34}\) and corn\(^{35}\) silages were fed to ruminants. The increased DM intake of inoculated silages has been attributed to improvement in fermentation parameters\(^{21}\) and/or enhancement in NDF digestibility.\(^{34,35}\) However, others provide no explanation.\(^{36,37}\) In our study the inoculant failed to have a significant effect on ruminal NDF degradability (Table 3).

Inoculation had no effects on total or 4% fat corrected milk yield (Table 4). Several studies have reported positive\(^{31,36}\) or no\(^{38}\) effects of silage inoculation on milk yield. Improvement in milk yield was for most part attributed to increased metabolizable energy intake, either because of increased DM intake or improved DM digestibility.\(^{39}\)

Milk composition was similar for cows fed untreated or inoculated alfalfa silage diet (Table 4). Our results agree with most studies that found no effect of inoculation on milk composition, whether inoculation improved\(^{31,39}\) or failed to enhance animal performance.\(^{30,31}\) However, in a few studies, inoculation with homolactic acid bacteria increased milk fat\(^{40}\) and milk protein\(^{30}\) percentages.

**CONCLUSIONS**

Treatment of high DM alfalfa with lactic acid bacteria inoculant based on multiple strains of *Lactobacillus plantarum* improved fermentation by reducing pH and increasing lactic acid production. However, inoculation increased proteolysis of alfalfa protein during early stages of ensiling. The improvement in fermentation characteristics as a result of inoculation was not reflected in ruminal *in situ* nutrient degradability. Furthermore, feeding inoculated relative to untreated alfalfa silage to dairy cows did not show any benefits in terms of DM intake or milk yield.

**REFERENCES**

C Rizk, AF Mustafa, LE Phillip


